Influence of Stocking Density on Bone Development and Carcass Characteristics of Family Chickens Reared up to 18 Weeks of Age Under Intensive System

Master of Science (MSc) in Animal Science

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INFLUENCE OF STOCKING DENSITY ON BONE DEVELOPMENT AND CARCASS CHARACTERISTICS OF FAMILY CHICKENS REARED UP TO 18 WEEKS OF AGE UNDER INTENSIVE MANAGEMENT SYSTEM

by

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DECLARATION

I declare that the dissertation hereby submitted by me for the Master of Science (Animal Science) degree at Botswana College of Agriculture, University of Botswana, is my own independent work and has not previously been submitted by me at another university/faculty for the award of any other degree or diploma.

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ABSTRACT

The influence of stocking density on bone development and carcass characteristics of family chickens was investigated. A total of 232 unsexed day-old family chicks were used in a completely randomized design. Birds were randomly assigned to four stocking densities, *i.e.*, D1 (10 birds/m^2), D2 (13 birds/m^2), D3 (16 birds/m^2) and D4 (19 birds/m^2) in the first phase (0-6 weeks). Each treatment was replicated four times. Two birds were slaughtered at 6, 12 and 18 weeks of age from each replicate to evaluate bone length, bone width, bone weight, bone chemical composition (ash weight, Ca%, P%, Mg%) and carcass characteristics. In the second phase (*i.e.*, from 7 to 12 weeks of age) the stocking densities were 8 birds/m^2 (D1), 11 birds/m^2 (D2), 14 birds/m^2 (D3) and 17 birds/m^2 and 6 birds/m^2 (D1), 9 birds/m^2 (D2), 12 birds/m^2 (D3) and 15 birds/m^2 (D4) in the final phase, from 13 to 18 weeks of age. General Linear Model (GLM) procedure of Statistical Analysis System was used to estimate the differences between treatment means for different stocking densities. Stocking density in all the three phases did not have a significant ($p>0.05$) effect on bone dimensions. Tibia length, width and weight, and humerus length, width and weight in the first phase ranged from 75.71 to 78.41 (± 17.31) mm, 4.80 to 5.25 (± 4.21) mm, 4.25 to 4.63 (± 2.77) g, 54.02 to 55.52 (± 1.71) mm, 5.10 to 5.56 (± 1.69) mm and 2.06 to 2.63 (± 1.34) g, respectively. In the second phase, the length, width and weight of both tibia and humerus ranges, following the same order as in the first phase, were 125.82 to 131.33 (± 43.43) mm, 7.88 to 8.46 (± 12.53) mm, 13.69 to 15.06 (± 11.04) g, 81.18 to 85.61 (± 4.89) mm, 7.88 to 8.25 (± 12.53) and 5.69 to 6.63 (± 17.31) g. In the final phase, following the same order as in the first phase the length, width and weight of both tibia and humerus ranged from 143.28 to 149.67 (± 71.54) mm, 9.39 to 10.11 (± 18.56) mm, 18.31 to 20.69 (± 20.42) g, 88.91 to 93.14 (± 6.49) mm, 9.01 to 9.61 (± 8.26) mm and 8.34 to 8.75 (± 3.70) g. In all the three phases stocking density did not have significant ($p>0.05$) effect on bone chemical composition. The levels of ash ranged from 0.79 to 0.86 (± 0.07 g), 2.83 to 3.29 (± 0.16 g) and 4.66 to 5.16 (± 0.27 g) in the first, second and final phases respectively. In the first phase Ca, P and Mg ranged from 32.68 to 32.94 (± 0.20%), 24.23 to 24.35 (± 0.21%) and 0.82 to 0.90 (± 0.02%), respectively. The levels of Ca, P and Mg in the second phase ranged from 32.68 to 33.25 (± 0.27%), 23.41 to 23.86 (± 0.21%) and 0.78 to 0.83 (± 0.03%), respectively. In the final phase Ca ranged from 34.19 to 34.31 (± 0.29%), P from 24.52 to 24.76 (± 0.11%) and Mg from

0.76 to 0.81 (± 0.02%). Stocking density in all the three phases did not have a significant (p>0.05) effect on carcass weight and primal cuts (*i.e.*, breast weight, back weight, drumstick weight, thigh weight and wing weight) of family chickens. The carcass weight, breast weight, back weight, drumstick weight, thigh weight and wing weight in the first phase ranged from 257.31 to 276.13 (± 17.31) g, 58.75 to 64.38 (± 4.21) g, 35.88 to 38.50 (± 2.77) g, 18.19 to 20.63 (± 1.71) g, 20.38 to 22.25 (± 1.69) g and 18.94 to 20.38 (± 1.34) g, respectively. In the second phase, the carcass weight and primal cuts weights ranges, following the same order as in the first phase, were 909.00 to 969.13 (± 43.43) g, 197.38 to 216.13 (± 12.53) g, 120.00 to 150.00 (± 11.04) g, 66.63 to 75.00 (± 4.89) g, 70.50 to 77.88 (± 17.31) g and 54.13 to 60.44 (± 17.31) g. In the final phase, following the order as in the first phase the carcass weight and primal cuts weights ranged from 1565.63 to 1719.75 (± 71.54) g, 379.75 to 390.25 (± 18.56) g, 221.38 to 260.13 (± 20.42) g, 119.00 to 135.31 (± 6.49) g, 135.88 to 155.38 (± 8.26) g and 91.13 to 102.25 (± 3.70) g. Therefore, it can be concluded that stocking density had no influence on bone development and carcass characteristics of family chickens raised up to 18 weeks of age under intensive system. It appeared that family chickens could be raised at a density of 15 birds/m^2in winter without any detrimental effect on bone development and related parameters. Further studies should be done on the use of identical densities throughout the research period to avoid disturbing the control which will make blocking by age possible.

Keywords: Bone development, carcass characteristics, family chickens, intensive system, stocking density.

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CONTENTS

LIST OF TABLES

ACRONYMS AND ABBREVIATIONS

BCA	Botswana College of Agriculture
BW	Body weight
BWG	Body weight gain
Ca	Calcium
cm	centimetre
CRD	Completely randomized design
FCR	Feed conversion ratio
GLM	General Linear Model
P	Phosphorus
Mg	Magnesium
g	gram
kg	kilogramme
L	litre
m^2	metre squared
max	Maximum
min	Minimum
ml	millilitre
SAS	Statistical Analysis System
°C	degrees Celsius
m^2	meter squared

CHAPTER 1

GENERAL INTRODUCTION

1.1 Background

Of all livestock reared in Botswana, poultry (mainly chickens) is the most widespread. Almost every family owns chickens, which provide a valuable source of family protein and additional income (Moreki, 2010). The terms indigenous, native, village, family, free range, traditional or scavenging chickens are often used interchangeably (Guèye, 1998; Kitalyi, 1998; Badubi *et al.*, 2006; Moreki, 2006; Kgwatalala *et al.*, 2013). The population of family chickens is estimated to be approximately 1.1 million in Botswana (Central Statistics Office, 2010). Family chickens are mainly owned and cared for by women (98%) and are kept in flock sizes ranging between 15 and 20 (Badubi *et al.*, 2006). Likewise, Badhaso (2012) in Ethiopia found the average flock size of indigenous chickens to range from 7 to 10 in each household. Generally, family chickens are of small body size, having slow growth rate with different colours of plumage, and of dual purpose type with variable body conformation and physical characteristics. Body weight is also variable indicating that family chickens lack uniformity in growth (Moreki, 2006).

Poultry production systems of tropical regions are mainly based on the scavenging indigenous chickens found in virtually all villages and households in the rural area. Approximately 80% of the chicken populations in Africa are reared in these systems (Guèye, 1998). With the exception of urban areas in northern and Southern Africa, most poultry production in Africa is undertaken through an extensive system at village or family level based on the scavenging domestic fowl (*Gallus domestics)* (Dwinger *et al.*, 2003).

Most family poultry farmers in Botswana do not house their chickens. As a result, chickens roost on tree tops and sometimes on any raised item in the homestead such as piles of bricks/blocks, old vehicles, bush fences, walls, under roof overhangs or on top of the huts, thus exposing themselves to the risks of predation, climatic hazards and theft (Badubi *et al.*, 2006; Moreki, 2010).

1.2 Stocking Density and Bird Performance

In the past, the number of birds in a given area was recognized as the only method of expressing stocking density. However, many producers in the poultry industry now express stocking density as mass per unit of space (Shanawany, 1988; Bilgili and Hess, 1995; Puron *et al.*, 1995; Feddes *et al.*, 2002). This takes into account the fact that large birds require more floor space per bird. This expression of stocking density is calculated based on body mass in kilogrammes per metre squared (kg/m^2) (Thaxton *et al.*, 2006). The advantage of using bird weight per unit area is that the standards are consistent despite the target weight (Abudabos *et al.*, 2013). Factors to consider when determining stocking density include but are not limited to bird size, feeder space, drinker space, house dimensions, bird welfare, nutrition, breed, performance and economic return (Yardimici and Kenar, 2008).

Stocking density is considered as one of the most important environmental factors because of its established effect on growth rate of broiler chickens. According to Sekeroglu *et al.* (2011), the stocking densities in broilers vary widely by countries, husbandry systems and final body weight. In many countries, stocking density in broiler production is not regulated, but top limiting values are determined which don't exceed 35 kg/m^2(Elwinger, 1995).

Commercial poultry producers are often tempted to increase the number of breeding stock per pen as a way of reducing housing, equipment, and labour costs per pen. However, literature indicates that high stocking densities can have a deleterious effect on the economics and welfare of poultry production (Mtileni *et al.*, 2007). Ferrante *et al.* (2006) reported that very high densities may impair the birds' welfare directly through physical restriction of movement.

A previous study by Dozier III *et al.* (2005) indicated that increasing the stocking density from 30 to 45 kg of body weight/m^2 of floor space influenced body weight gain and feed consumption, but meat yields were not significantly altered. Beg *et al.* (2011) found that broilers under lower and higher stocking densities showed no significant difference in different carcass parts. On the other hand, Škrbić *et al.* (2011) observed that broilers reared in lower stocking density had significantly better carcass conformation and higher breast yield.

Bone is a complex tissue that is continuously undergoing changes throughout an animal's life due to the processes of bone formation and bone resorption. It is a specialized connective tissue composed of intercellular calcified material, the bone matrix (which is approximately 70% mineral, 20% organic matter and 10% water), and three major cell types; osteoblasts, osteocytes and osteoclasts. Osteoblasts secrete 'Type 1' collagen and noncollagenous proteins, while osteocytes maintain mature bone and osteoclasts resorb bone by acidification (Junqueira and Carneiro, 1983; Rath *et al.*, 2000; Klein and Enders, 2010).

There are two processes of bone development: intramembranous ossification and endochondral ossification. Endochondral ossification includes the activities responsible for the formation of bones that support weight, and also for the elongation of most of the skeletal mass during growth. In this process, hyaline cartilage is deposited in the shape of the required bone and is subsequently transformed into bone by mineralization. On the other hand, intramembranous ossification results in the ultimate shape of a limited number of bones formed, which are not performed by cartilage (Ham, 1969; Almeida Paz *et al.*, 2005). According to Almeida Paz *et al.* (2006), the skeletal growth rate of broiler is very fast between 22 to 70 days of age, and 80% of the mature size is reached at 56 days of age and at approximately 12 weeks of age, 95% of bone growth potential is achieved.

1.3 Justification

Stocking density is considered as one of the most important environmental factors because of the established effect on growth rate of chickens. There was little information on how stocking density influences bone development and carcass characteristics in family chickens. Therefore, a study was undertaken to investigate the influence of stocking density on growth parameters and carcass traits of family chickens up to 18 weeks of age.

1.4 Study Objectives

Experiment 1: Influence of stocking density on bone development of family chickens reared up to 18 weeks of age under intensive system

The overall objective of the study was to determine the influence of stocking density on bone development of family chickens subjected to different stocking densities under intensive system up to 18 weeks of age.

1.4.1. Specific objectives

To measure bone dimensions of tibiae and humeri (*e.g.*, bone length, bone width and bone weight) of family chickens.

To evaluate the chemical composition (ash, Ca, P, Mg) of tibiae of family chickens.

The hypothesis tested was:

H_O: Different stocking densities have no effect on bone development of family chickens raised under the same management system.

H_A: Different stocking densities have a significant effect on bone development of family chickens raised under the same management system.

Experiment 2: Influence of stocking density on carcass characteristics of family chickens reared up to 18 weeks of age under intensive system

The overall objective of the study was to determine the influence of stocking density on carcass characteristics of family chickens subjected to different stocking densities under intensive system up to 18 weeks of age.

1.4.2. Specific objective

To measure carcass traits (carcass weight, dressing percentage, breast, back, drumstick, thigh, wing and giblets) of family chickens subjected to different stocking densities under intensive system up to 18 weeks of age.

The hypothesis tested was:

H_O: Different stocking densities have no effect on carcass characteristics of family chickens raised under the same management system.

H_A: Different stocking densities have a significant effect on carcass characteristics of family chickens raised under the same management system.

This thesis is presented in the form of two separate articles, augmented by a general introduction, literature review and conclusion in an effort to create a single unit. Although care has been taken to avoid unnecessary repetition, some repetition has been inevitable.

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CHAPTER 2

LITERATURE REVIEW

2.1 Introduction

Several studies have illustrated the effects of stocking density on growth performance of chickens (Puron *et al.*, 1995; Dozier III *et al.*, 2005). According to Jayalakashmi (2009), poor production performance of broilers and meat quality have been attributed to inadequate floor space as a result of poor micro environmental conditions inside the poultry house and competition for the feed and water. In broilers, high densities have been associated with a decline in body weight, feed consumption and feed conversion ratio (FCR), increased tibia curvature and mortality (Proudfoot *et al.*, 1979; Shanawany, 1988; Cravener *et al.*, 1992; Bilgili and Hess, 1995; Feddes *et al.*, 2002; Dozier III *et al.*, 2006; Buijs *et al.*, 2012). Some studies show large benefits in reducing stocking density while others show little or no differences (Beg *et al.*, 2011). Different stocking densities are used, depending on the country and production system (Buijs *et al.*, 2009).

2.2 Growth Performance

2.2.1 Body weight, body weight gain and final live body weight

A previous study by Škrbić *et al.* (2007) showed that Cobb 500 and Arbor Acres broilers had significantly greater body weights (BW) at lower housing density. In another study, Mtileni *et al.* (2007) found that birds at stocking density of 15 per pen were about 183 g heavier than those at stocking density of 20 per pen. Moreover, Kalita *et al.* (2004) reported an increase in BW of broilers reared at lower stocking density. A more recent study by Seven*et al.* (2013) reported a significantly decreased BW of quails at high stocking density. Lewis *et al.* (1997) reported that broilers at high stocking density had significantly lower BW than broilers at lower stocking density. Similar observations were made by Olympio *et al.* (1982), Bilgili and Hess (1995), Puron *et al.* (1995), Dozier III *et al.* (2005; 2006) and Tong *et al.* (2012). The study by Carey (1987) found that at higher stocking density body weight of pullets was significantly reduced. Increased bird density results in a linear reduction in BW of broilers (Proudfoot *et al.* 1979; Shanawany, 1988). On the other hand, Yakubu *et al.* (2010) found that the effect of placement density on final BW did not follow a linear trend. Recently, Beloor *et al.* (2010) observed that increased BW in low density stocked broilers compared to broilers stocked at high density did not show significant difference. Several studies have shown that stocking density does not affect BW in broilers (Turkyilmaz, 2008; Ventura *et al.*, 2010; Angelovičová *et al.*, 2012; Houshmand *et al.*, 2012; Huo and Na-Lampang, 2012).

The study by Sekeroglu *et al.* (2011) showed that live weight gain of broilers raised at low stocking density was higher than those at highest stocking density group. Similar observations were made by Kalita *et al.* (2004) and Iyasere *et al.* (2012). A previous study by Sahin *et al.* (2007) found that live body weight gain was adversely affected at high stocking density in layers. Recently, Seven *et al.* (2013) observed a significant BWG decrease at high stocking density in quails. Dozier III *et al.* (2005) and Abudabos *et al.* (2013) reported a reduction in cumulative body weight gain (BWG) in broilers as stocking density increased. On the other hand, Thomas *et al.* (2004) observed no influence of stocking density on live weight gain of broilers raised at densities of 10, 15 and 20 birds/m^2. Likewise, Ravindran *et al.* (2006) and Houshmand *et al.* (2012) found no effect of stocking density on BWG in broilers.

Previous studies by Sorensen *et al.* (2000) and Ratsaka *et al.* (2012) observed that broilers housed at high stocking density had lower live weight than those housed at low density. Bolton *et al.* (1972) reported that at 10 weeks old a decrease in space allowance from 0.093 to 0.047 m^2/bird was accompanied by reduced final live-weight. The average live weight of broilers under stocking density of 12 birds/m^2 was significantly higher compared to other density groups (8, 10, and 14 birds/m^2 (Beg *et al.*, 2011). Onbasilar *et al.* (2008)andFeddes *et al.* (2002) observed that BW of broilers decreases with increased stocking density. However, Jayalakshmi *et al.* (2009) observed that lower stocking density of 8 bird/m^2 and 12 bird/m^2 did not show significant difference in final live weight. Similar observation was made by Lee and Moss (1995) in pullets.

2.2.2 Feed conversion ratio

Lewis *et al.* (1997), Hassanein, (2009) and Houshmand *et al.* (2012) reported a significant improvement of FCR in broilers at lower stocking density compared to high stocking density. On the other hand, Dozier III *et al.* (2006) and Seven*et al.* (2013) found that FCR was adversely affected by increasing stocking densities in broilers and quails, respectively. Similarly, Bilgili and Hess (1995), Mtileni *et al.* (2007) and Beg *et al.* (2011) observed a significant effect of high stocking density on FCR in broilers. However, several studies reported no influence of stocking density on FCR in broilers (Puro *et al.*, 1995; Imaeda, 2000; El-Deek and Al-Harthi, 2004; Dhaliwal and Nagra, 2006; Zhao *et al.*, 2009; Ventura *et al.*, 2010; Abudabos *et al.*, 2013; Tayeb *et al.*, 2011; Sekeroglu *et al.*, 2011; Iyasere *et al.*, 2012; Lallo *et al.*, 2013).

2.2.3 Feed intake

The influence of stocking density and air velocity on behaviour and performance of broilers was evaluated in a 28-days trial and it was observed that increasing stocking density reduced feed intake but enhanced feeding behaviour of the broilers (Iyasere *et al.*, 2012). The increase in feeding behaviour at higher stocking density can be related to social hierarchy. Feed intake reduced with an increase in stocking density at 5 and 6 weeks of age. Similarly, El-Deek and Al-Harthi (2004) reported that chicks reared under stocking density of 18 chicks/m^2 consumed significantly less feed than those kept at 10 chicks/m^2. Likewise, Shanawany (1988) found that average feed intake over the whole experimental period declined linearly with densities above 20 birds/m^2. Puron *et al.* (1995) also reported 3.5% reductions in cumulative feed intake of 49 day birds in response to increased stocking density from 10 to 12 birds/m^2. Dozier III *et al.* (2006) found that increased stocking densities negatively impacted on feed intake in male broilers raised to 1.8 kg. In a study by Abudabos *et al.* (2013), feed intake was reduced by 15.6% as the stocking density increased from 0.037 m^2/bird to 0.030 m^2/bird. Moreover, Carey (1987) and Rios *et al.* (2009) observed a significantly reduced feed intake at high stocking density in pullets. Feed intake was significantly affected at high stocking density in quails (Seven *et al.*, 2013). Similar findings were reported by Onbasilar *et al.* (2008) and Hassanein (2009) in broilers. On the other hand, Dhaliwal and Nagra (2006) and Ravindran *et al.* (2006) found no significant effect of stocking density on feed intake in Japanese quails and broilers, respectively.

2.2.4 Mortality

Higher mortality (43%) for broilers housed at 18 birds/m^2 compared to 33% for birds housed at 12 birds/m^2 was observed by Imaeda (2000). Similarly, Tayeb *et al.* (2011) observed high mortality at a stocking density of 13.36 birds/m^2 compared to stocking densities of 8.66 and

10.41 birds/m^2. A previous study by Dhaliwal and Nagra (2006) reported increased mortality with an increase in stocking density of Japanese quails above 125 birds/m^2 in summer and above 143 birds/m^2 in winter. Moreover, Rios *et al* (2009) reported an increase in mortality due to high stocking density in pullets. However, no significant differences were observed in mortality rate of broilers and pullets due to stocking density (Cravener *et al.*, 1992; Lee and Moss, 1995; Puron *et al.*, 1995; Onbasilar and Aksoy, 2005; Dozier III *et al.*, 2006; Beg *et al.*, 2011; Abudabos *et al.*, 2013; Lallo *et al.*, 2013).

2.3 Bone Development

Bone deposition is regulated primarily by parathyroid hormone, which is secreted in response to low serum calcium levels (Klein and Enders, 2010). Vitamin D3 is one of the most important nutritional factors crucial to Ca and P absorption and proper skeletal development. It is generally added to diets in the form of cholecalciferol; however, in order to carry out its physiological function, it must be hydroxylated in a two-step process: in the liver (to 25-OH-D3) and in the kidneys (to 1.25-OH-D3) (Świątkiewicz and Arczewska-Wlosek, 2012).

A previous study by Ventura *et al.* (2010) reported that broilers reared at a density of 18 birds/m^2 had significantly longer tibiae (84.61 mm) than those reared at a lower density of 8 birds/ m^2 (84.38 mm). Recently, Škrbić *et al.* (2011) found that broilers reared in lower stocking density (12 bird/m^2) improved tibia quality. A more recent study by Buijs *et al.* (2012) found that increased stocking density in broilers resulted in a shorter tibiae, however, tibia weight remained unaffected. On the contrary, Simsek *et al.* (2011) and Oleviera *et al.* (2012) showed that the length of both tibia and humerus and the width of tibia were not affected by stocking density except for humerus width at stocking densities of 10 and 16 birds/m^2. Similarly, Ventura *et al.* (2010) reported no influence of stocking density on tibia width in broilers. Bone ash of broilers was not affected by stocking density (Tablante *et al.*, 2003).

2.4 Carcass Characteristics

Feddes *et al.* (2002) reported decreased carcass weight with increased stocking density. A similar observation was made by Dozier III *et al.* (2005; 2006). The carcass weight of chickens on 0.08 m^2/chicken stocking density for the pen system was significantly smaller than carcass weight of chickens from stocking densities of 0.06 m^2/chicken and 0.05 m^2/chicken by 165.2 g and 186.6

g, respectively (Ratsaka *et al.*, 2012). However, Cravener *et al.* (1992) found that birds housed at 0.07, 0.09, and 0.11 m^2/bird had similar 7-week carcass weights, all significantly higher than birds housed at 0.05 m^2/bird. Jayalakshmi *et al.* (2009) recorded significantly higher eviscerated carcass weight in 0.075 m^2/ bird density, followed by 0.06 m^2, 0.09 m^2 and 0.045 m^2/bird density.Similarly, Nahashon *et al.* (2009) observed higher carcass yields in French guinea fowl broilers raised in floor densities of 13.6 and 12 birds/m^2 than those raised in floor densities of 15.6 and 10.7 birds/m^2. On the other hand, Yakubu *et al.* (2010) found that stocking density did not affect carcass yield. A previous study by Thomas *et al.* (2004) showed that stocking density had no influence on carcass characteristics of broilers grown at densities of 10, 15 and 20 birds per m^2.

In a study by Škrbić *et al.* (2011), it was observed that broilers reared in lower stocking density (12 bird/m^2) had significantly higher yield of breast compared to those with a stocking density of 16 birds/m^2. Moreover, Toplu and Fidan (2008) reported a significant effect of stocking density on breast weight of Japanese quail. Likewise, Osman (1993) observed a decrease in breast weight of ducks with increasing stocking density. Several studies reported no influence of stocking density on breast yield of broilers and rock partridges (Feddes *et al.*, 2002;Moreira *et al.*, 2004; Dozier III *et al.*, 2005; Esen *et al.*, 2006; Ravindran *et al.*, 2006;Yakubu *et al.*, 2010; Sekeroglu *et al.*, 2011; Simsek *et al.*, 2011; Zuowei *et al.*, 2011). A similar observation was made by Dhaliwal and Nagra (2006) on Japanese quails.

Recently, Beg *et al.* (2011) reported that lower stocking density produced higher dressing percentage in broilers. Similarly, Jayalakshmi *et al.* (2009) reported significantly higher dressed weight in 0.075 m^2 stocking density followed by 0.060 m^2, 0.045 m^2 and 0.090 m^2/ bird density groups. Moreover, Dhaliwal and Nagra (2006) found that dressing percentage increased with decreased stocking density in Japanese quails. El-Deek and Al-Harthi (2004) found that dressing percentage decreased due to increasing stocking density in broiler chicks.

The study by Tong *et al.* (2012) found that the thigh yield of local chickens was significantly affected in the medium-density group (35 birds/m^2) compared to those of the low (25 birds/m^2) and high (45 birds/m^2) densities. Recently, Škrbić *et al.* (2011) observed that broilers reared in lower stocking density (12 bird/m^2) had significantly high yield of thigh and drumstick compared to those at a stocking density of 16 birds/m^2. A similar observation was made by Dhaliwal and Nagra (2006) in Japanese quails. Several studies have shown that stocking density does not affect thigh and drumstick yield in broilers and rock partridges (Lewis *et al.*, 1997; Mizubuti *et al.*, 2000; Moreira *et al.*, 2004; El-Deek and Al-Harthi, 2004; Dozier III *et al.*, 2005; Esen *et al.*, 2006; Jayalakshmi *et al.*, 2009; Yakubu *et al.*, 2010; Beg *et al.*, 2011; Sekeroglu *et al.*, 2011; Simsek *et al.*, 2011; Zuowei *et al.*, 2011; Tong *et al.*, 2012).

The study by Yakubu *et al.* (2010) reported that housing density has a significant effect on wing weight of broilers. However, Lewis *et al.* (1997), Moreira *et al.* (2004), El-Deek and Al-Harthi (2004), Jayalakshmi *et al.* (2009), Sekeroglu *et al.* (2011), Beg *et al.* (2011), Simsek *et al.* (2011) and Tong *et al.* (2012) found that wing weights of broilers were unaffected by stocking density. Likewise, Nahashon *et al.* (2009) observed no significant difference in mean wing weights of French guinea fowl broilers due to stocking density. Similar observations were made by Osman (1993) and Esen *et al.* (2006) in ducks and rock partridges, respectively.

Broilers reared at high stocking density had high weight percentage of heart that those raised at low stocking density. Because of worse litter at high stocking density due to overcrowding the broilers might be having rapid respiration and therefore higher heart percentage (Onbasilar *et al.*, 2008). Similarly, Toplu and Fidan (2008) observed a significant effect of stocking density on heart weight of Japanese quail. A previous study by Simitzis *et al.* (2012) found that lower stocking density had a significant effect on the weight of the broiler's liver. In contrast,Nicol *et al.* (2006) reported that birds housed at 7 and 9 birds/m^2 had no effect on the weight of the liver, suggesting that the birds were not chronically stressed. Nahashon *et al.* (2009) observed that the weight of the gizzard of French guinea fowl broilers was not affected by stocking density. Recently, Beg *et al.* (2011) observed that different stocking densities had no effect on average giblet weight percentage in broiler chickens. Also, the study by El-Deek and Al-Harthi (2004) found no significant effect of stocking density on the weight of internal organs, showing that internal organs of broilers developed normally under different densities during rearing. Likewise, Esen *et al.* (2006) observed that the weight of the gizzard, liver and heart of rock partridges was not affected by stocking density.

2.5 Conclusion

Several studies have been conducted to investigate the effect of stocking density in broilers with diverse genetic lines. Majority of these studies reached variable conclusions. Some studies show large benefits in reducing stocking density, while others have documented little or no differences. There is no documented work on how stocking density affects the productivity of family chickens. The absence of stocking density standards for family chickens has forced farmers to rely on personal experience in determining the space allowances. Therefore, it is necessary to investigate the influence of stocking density on bone development and carcass characteristics of family chickens, and establish more precise stocking density standards for these chickens to ensure their effective production.

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CHAPTER 3

INFLUENCE OF STOCKING DENSITY ON BONE DEVELOPMENT OF FAMILYCHICKENS REARED UP TO 18 WEEKS OF AGE UNDER INTENSIVE MANAGEMENT SYSTEM

3.1 Introduction

In poultry management, housing is a method involving the allotment of a definite floor space to a bird to provide a comfortable environment for satisfactory performance. Stocking density is a housing variable that can affect chickens' development. Bone development is part of animal growth and the growth of the skeleton determines the size and proportions of the body (Martini *et al.*, 2000; Yakubu *et al.*, 2010; Buijs *et al.*, 2012). Bone is a dynamic tissue influenced by physiological, nutritional, and physical factors such as mechanical stress and physical activities (Rath *et al.*, 2000). The deposition of bone is regulated primarily by parathyroid hormone, which is secreted in response to low serum calcium levels (Klein and Enders, 2010).

Several studies have been conducted to study the effect of stocking density on broiler production. According to Hall (2001), increased stocking density can negatively influence skeletal development of broilers, as shown by an increase in leg culls, which may be due to a decrease in activity as density increases. A study by Škrbić *et al.* (2009) observed that providing more floor space per chicken influenced the level of physical activity, development and firmness of the skeleton, especially legs. Physical activity of broilers influenced cross section of the cortex and as a result improved their mechanical characteristics by better supply with blood of epiphysis of long bones and sufficient mineralization. Recently, Škrbić *et al.* (2011) observed that more physical activity of broilers in lower stocking density improved the parameters of tibia quality.

The term "family poultry" was defined as small-scale poultry keeping by households using family labour and locally available feed resources (Sonaiya and Swan, 2004).Family poultry production systems of tropical regions are mainly based on family chickens found in nearly all villages and households in rural areas (Guèye, 1998)

There is little information on how stocking density influenced bone development in family chickens. The absence of stocking density standards for family chicken may have forced farmers to rely on personal experience in determining the space allowances and this may have affected productivity. Therefore, a study was undertaken to investigate the influence of stocking density on bone development of family chickens reared up to 18 weeks of age under intensive system.

3.2 Materials and Methods

3.2.1 Study location

The experiment was carried out at the Guinea Fowl Unit of the Botswana College of Agriculture (BCA), Sebele for a period of 18 weeks. The site is at an altitude of 994 m above sea level and the coordinates are latitude 24° 33' S and longitude 24° 54' E (Aganga and Omphile, 2000). The experiment started in April and ended in August 2013. During the study period, environmental temperature averaged 21 °C and ranged from 5 to 21 °C.

3.2.2 Experimental Design

A completely randomized design (CRD) with four treatments was used in the experiment. Each treatment was replicated four times. The four treatment levels were D1 (10 birds/m^2), D2 (13 birds/m^2), D3 (16 birds/m^2) and D4 (19 birds/m^2) in the first phase (0 to 6 weeks). The experimental birds were distributed randomly among the four stocking densities. Two birds were slaughtered at 6, 12 and 18 weeks of age from each replicate. In the second phase (7 to 12 weeks) the stocking densities were 8 birds/m^2 (D1), 11 birds/m^2 (D2), 14 birds/m^2 (D3) and 17 birds/m^2 and 6 birds/m^2 (D1), 9 birds/m^2 (D2), 12 birds/m^2 (D3) and 15 birds/m^2 (D4) in the final phase (13 to 18 weeks).

3.2.3 Animal management.

A total of 232 unsexed day-old family chicks were obtained from a farmer in Gaborone north and reared in a deep litter system. Initial body weights of the birds were determined by weighing 10% of the birds prior to allocation to four stocking densities. Birds were individually identified using wing bands. The chicks were housed under deep litter management system in an open-sided shed. The size of each pen was one metre squared (m^2). All pens were bedded with wood-shavings and equipped with one tube feeder and a 10 L waterer. Birds were raised under artificial light for the first two weeks of acclimatization to the experimental diets prior to collection of data and later under natural light throughout the study period. Feeds and water were provided *ad libitum* throughout the experimental period. Birds in each replicate were group fed. Chickens in each pen were weighed weekly.

3.2.4 Experimental diets

Birds were fed a commercial broiler starter crumbled diet for the first 6 weeks, pelleted broiler grower diet (7 to 12 weeks) and pelleted broiler finisher diet (13 to 18 weeks). Commercial broiler diets were sourced from some retail shops in Gaborone.

Table 3.1: Chemical composition of experimental diets fed to family chickens from 0 to 18 weeks of age

Chemical Composition	Feed type and age of birds		
	Broiler starter crumbles (0-6 weeks)	Broiler grower pellets (7-12 weeks)	Broiler finisher pellets (13-18 weeks)
	Amount in g/kg	Amount in g/kg	Amount in g/kg
Protein (min)	200.0	180.0	160
Moisture (max)	120.0	120.0	120.0
Fibre (max)	50.0	60.0	70.0
Calcium (min)	8.0	7.0	6.0
Calcium (max)	12.0	12.0	12.0
Fat (min)	25.0	25.0	25.0
Phosphorus (min)	6.0	5.5	5.0
Total lysine (min)	12.0	10.0	9.0

Source: OPTI Feeds Botswana (Pty) Ltd, 2014.

3.2.5 Data collection

At 6, 12 and 18 weeks of age, two birds from each replicate were sacrificed to determine bone dimensions and chemical composition. After slaughter carcasses were packed, identified, and chilled to 0 °C overnight in a cold room and the bones removed 24 hours *post mortem*. The tibiae (right and left) and the right humerus from each of the birds were removed and defleshed without boiling. Thereafter, bones were individually weighed using an electronic balance with a precision of 0.001 g, (Sartorius AG Germany, TE 313S model) and their widths and lengths determined by using an electronic calliper with an accuracy of 0.001 cm, (Starrett® 798 B 12"/300 mm model). The left tibiae were used for bone chemical composition analysis (ash, Ca, P, Mg). Bone samples were oven-dried in porcelain crucibles at 105 °C for 48 hours and weighed (Liu *et al.*, 2003). Thereafter, bone samples were ashed in a muffle furnace at 550 °C for 8 hours. Approximately 1 g of ash samples was dissolved in 10 ml of 3M hydrochloric acid and boiled for 10 minutes. The samples were allowed to cool and filtered into a 100 ml volumetric flask. Thereafter, the volume was topped up to 100 ml with distilled water and later analyzed for minerals (AOAC, 1996).

3.2.6 Statistical analysis

General Linear Model (GLM) Procedures of Statistical Analysis System (SAS Institute, 2009) version 9.2.1 was used to analyse the data according to the following statistical model:

$Y_{ij} = \mu + \tau_i + \varepsilon_{ij}$.

Where: Y_{ij}= response variables (bone width, bone length, bone weight, and bone chemical composition).

μ = general mean effect. τ_i =i^{th} stocking densities effects on family chickens' growth.

Where i= 1,2,3,4. Where 1= 10 birds/m^2, 2=13 birds/m^2, 3=16 birds/m^2, 19 birds/m^2.

ε_{ij}= random error

Least significant difference comparisons were made between treatment means using paired t-test. Statistical significance was established at P≤0.05.

3.3 Results and Discussion

3.3.1 Bone dimensions

3.3.1.1 Bone length

Stocking density had no significant (p>0.05) effect on tibia and humerus length of family chickens (Tables 3.2 to 3.4). However, the longest tibia (78.41 mm) and humerus length (55.52 mm) was found in the stocking densities of 19 bird/m^2 at week 6, 14 bird/m^2 (131.33 mm;85.61 mm) at week 12 and 9 bird/m^2 (149.67 mm; 93.14 mm) at week 18, respectively. These results are consistent with those obtained by Olevicra *et al.* (2012) who found that the length of both tibiae and humeri were not affected by stocking density of 10 and 16 birds/m^2. A more recent study by Buijs *et al.* (2012) found that increased stocking density (15.5, 18.5 and 21.8 birds/m^2) in broilers resulted in shorter tibiae. The variations in the results for the two studies may be due to the differences in genotype. In this study family chickens of slow growing genotype were used, whereas Buijs *et al.* (2012) used broilers of fast growing genotype. This suggests that as broilers increase in body weight, the tibiae increase in width to support the muscle mass thus forcing them to curve. Tibial length increased by 65% and 13.8% between 6 and 12 weeks and 12 and 18 weeks, respectively. On the other hand, humerus length increased by 52% and 8% between 6 and 12 weeks and 12 and 18 weeks, respectively. This indicates that bone development and growth in family chickens were most pronounced during the first 12 weeks, a similar observation was made by Moreki *et al.* (2011) who found that tibia and humerus length increased by 46% and 36% in broiler breeder pullets between 6 and 12 weeks, respectively. According to Ross Breeders (2006) the skeletal size of broiler breeder is fixed at 12 weeks of age. Rath *et al.* (2000) stated that the increment in bone length is correlated with an increase in the content of hydroxylysylpridinoline and lysylpyridinoline, the collagen crosslinks.

Table 3.2: Least square means for bone dimensions of family chickens at 6 weeks of age reared under intensive system

Treatments	Parameters					
	Tibia			Humerus		
Bird density/m^2	Length(mm)	Width (mm)	Weight(g)	Length(mm)	Width(mm)	Weight(g)
10	77.22^a	5.25^a	4.63^a	55.07^a	5.56^a	2.50^a
13	75.71^a	4.80^a	4.31^a	54.02^a	5.10^a	2.63^a
16	78.02^a	5.10^a	4.50^a	54.91^a	5.40^a	2.31^a
19	78.41^a	4.86^a	4.25^a	55.52^a	5.30^a	2.06^a
SE	1.74	0.21	0.48	1.13	0.15	0.21
P-value	0.7103	0.4103	0.9425	0.8211	0.2377	0.2865
CV	4.5045	8.2050	21.8815	4.1286	5.6659	17.3232

Means within the same column within a parameter not having similar superscripts are significantly different ($P<0.05$).

SE = Standard Error, CV = Coefficient of variation.

Table 3.3: Least square means for bone dimensions of family chickens at 12 weeks of age reared under intensive system

Treatments	Parameters					
	Tibia			Humerus		
Bird density/m^2	Length(mm)	Width (mm)	Weight(g)	Length(mm)	Width(mm)	Weight(g)
8	125.82^a	7.88^a	13.81^a	83.54^a	7.96^a	5.69^a
11	126.03^a	7.89^a	13.69^a	81.18^a	7.88^a	5.75^a
14	131.33^a	8.41^a	14.75^a	85.61^a	8.25^a	6.44^a
17	127.79^a	8.46^a	15.06^a	83.48^a	8.21^a	6.63^a
SE	2.16	0.31	0.88	1.58	0.13	0.45
P-value	0.2939	0.3983	0.6258	0.3141	0.1876	0.3773
CV	3.3858	7.5774	12.2578	3.7764	3.3032	14.6456

Means within the same column within a parameter not having similar superscripts are significantly different ($P<0.05$).
SE = Standard Error, CV = Coefficient of variation.

3.3.1.2 Bone width

No significant influence of stocking density on tibial and humerus width of family chickens could be detected (Tables 3.2 to 3.4). The greatest tibial and humerus width was found in the stocking densities of 10 bird/m^2 (5.25 mm; 5.56 mm) at week 6, 17 bird/m^2 and 14 birds/m^2 (8.46 mm for tibia;8.25 mm for humerus) at week 12 and 9 bird/m^2 (10.11 mm; 9.61 mm) at week 18, respectively. The present results are consistent with those of Simsek *et al.* (2011) who found that the width of tibia was not affected by stocking densities of 22.5, 18.75, 15, 11.25, 7.5 broilers/m^2 in broilers. Similarly, Ventura *et al.* (2010) reported no influence of stocking density (8 birds/m^2, 13 birds/m^2 and 18 birds/m^2) on tibial width in broilers. In contrast to the present results, Oleviera *et al.* (2012) observed that the width of humerus in Ross 308 and Hybro PG broilers was affected by stocking density (10 and 16 birds/m^2) at 42 days of age. The differences in the results of the current study and that of Oleviera *et al.* (2012)may be attributable to differences in birds' genotype. Several studies have observed that hybrid broilers raised intensively grow rapidly and as they approach market age and weight, their bodies take up most of the allotted space, leaving no room to perform simple exercises which may lead to a decrease in bone mass of the wings (Lewis *et al.*,1997; Jones, 2010). In this study, tibial width increased by 63.1% and 18.6% between 6 and 12 weeks and 12 and 18 weeks, respectively, whereas humerus width increased by 51.2% and 16.0% between 6 and 12 weeks and 12 and 18 weeks, respectively. This suggests that in response to increased body weight, bones of the family chickens increased in bone width.

Table 3.4: Least square means for bone dimensions of family chickens at 18 weeks of age reared under intensive system

Treatments	Parameters					
	Tibia			Humerus		
Bird density/m^2	Length(mm)	Width(mm)	Weight(g)	Length(mm)	Width(mm)	Weight(g)
6	143.30^a	9.39^a	18.31^a	90.05^a	9.40^a	8.38^a
9	149.67^a	10.11^a	20.69^a	93.14^a	9.61^a	8.75^a
12	143.28^a	9.68^a	19.19^a	88.91^a	9.44^a	8.44^a
15	145.07^a	9.53^a	18.81^a	88.93^a	9.01^a	8.44^a
SE	2.39	0.38	1.11	1.34	0.21	0.49
P-value	0.2421	0.5895	0.4925	0.1376	0.2876	0.9478
CV	3.2834	7.9160	11.5248	2.9597	4.5447	11.5950

Means within the same column within a parameter not having similar superscripts are significantly different ($P<0.05$),

SE = Standard Error,CV = Coefficient of variation.

3.3.1.3 Bone weight

The weight of tibia and humerus was not significantly influenced by stocking density (Tables 3.2 to 3.4). However, the highest tibial weight observed in week 6 was 4.63 g (10 birds/m^2), 15.06 g (17 birds/m^2) in week 12 and 20.69 g (9 birds/m^2) in week 18. The highest humerus weight observed in week 6 was 2.63 g (13 birds/m^2), 6.63 g (17 birds/m^2) in week 12 and 8.75 g (9 birds/m^2) in week 18. In agreement with the current findings on tibia, Buijs *et al.* (2012) found that stocking density had no effect on tibia weight in broilers. Similarly, Oleviera *et al.* (2012) observed that weight of tibia and humerus in broilers was not affected by stocking density at 42 days of age.

3.3.2 Bone chemical composition

Stocking density had no significant effect on chemical composition of bones (Tables 3.5 to 3.7). The highest bone ash weight was found in the stocking densities of 16 bird/m^2 (0.86 g) at week 6, 17 bird/m^2 (3.29 g) at week 12 and 9 bird/m^2 (5.16 g) at week 18.These results are consistent with those obtained by Tablante *et al.* (2003) who found that bone ash of broilers was not affected by stocking densities of 10, 15 and 20 birds/m^2. Although the mean weight of ash did not differ significantly among birds reared at different stocking densities, it increased with age. Similar observation was made by Moreki *et al.* (2011) who reported a significant increase in bone ash with age in broiler breeders up to 18 weeks of age. In the present study, the highest levels of Ca, P and Mg were observed at 16 birds/m^2 in week 6 (32.94 %, 24.35%, 0.90%), 14 birds/m^2 in week 12 (33.28%, 23.86%, 0.83%) and 12 birds/m^2 in week 18 (34.31%, 24.76%, 0.81%), respectively. The levels of Ca, P and Mg decreased from 6 to 12 weeks of age, and from week 12 to 18 weeks of age only Ca and P contents increased, whereas Mg content continued to decline. The decline of Ca and P from 6 to 12 weeks of age could be due to the birds increased demand for nutrients for increased muscle mass.

Table 3.5: Least square means for bone chemical composition of family chickens at 6 weeks of age reared under intensive system

Treatments	Variables			
Bird density/m^2	Ash (g)	Ca (%)	P (%)	Mg (%)
10	0.84^a	32.68^a	24.23^a	0.82^a
13	0.79^a	32.83^a	24.25^a	0.83^a
16	0.86^a	32.94^a	24.35^a	0.90^a
19	0.80^a	32.84^a	24.27^a	0.85^a
SE	0.07	0.20	0.21	0.02
P-value	0.8236	0.8363	0.9760	0.7086
CV	16.1889	1.2104	1.6986	3.4858

Means within the same column within a parameter not having similar superscripts are significantly different ($P<0.05$), SE = Standard Error, CV = Coefficient of variation.

Table 3.6: Least square means for bone chemical composition of family chickens at 12 weeks of age reared under intensive system

Treatments	Variables			
Bird density/m^2	Ash (g)	Ca (%)	P (%)	Mg (%)
8	2.83^a	32.90^a	23.50^a	0.78^a
11	2.89^a	32.90^a	23.65^a	0.78^a
14	3.04^a	33.28^a	23.86^a	0.83^a
17	3.29^a	32.68^a	23.41^a	0.82^a
SE	0.16	0.27	0.21	0.03
P-value	0.2383	0.5097	0.4803	0.3244
CV	10.7187	1.6686	1.7801	6.3518

Means within the same column within a parameter not having similar superscripts are significantly different $P<0.05$), SE = Standard Error, CV = Coefficient of variation.

Table 3.7: Least square means for bone chemical composition of family chickens at 18 weeks of age reared under intensive system

Treatments	Variables			
Bird density/m²	Ash (g)	Ca (%)	P (%)	Mg (%)
6	4.66ᵃ	34.19ᵃ	24.52ᵃ	0.76ᵃ
9	5.16ᵃ	34.23ᵃ	24.55ᵃ	0.79ᵃ
12	4.81ᵃ	34.31ᵃ	24.76ᵃ	0.81ᵃ
15	4.72ᵃ	34.25ᵃ	24.57ᵃ	0.79ᵃ
SE	0.27	0.29	0.11	0.02
P-value	0.5812	0.9933	0.4586	0.2904
CV	11.17686	1.6891	0.9188	4.4908

Means within the same column within a parameter not having similar superscripts are significantly different ($P<0.05$).SE = Standard Error, CV = Coefficient of variation.

3.4 Conclusion

Bone dimensions and bone chemical composition were not influenced by stocking density. Therefore, it can be concluded that stocking density had no influence on bone development in family chickens raised under intensive system up to 18 weeks of age. It appeared that family chickens could be raised at a density of 15 birds/m²in winter without any detrimental effect on bone development and related parameters. Further studies should be done on the use of identical densities throughout the research period to avoid disturbing the control which will make blocking by age possible.

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CHAPTER 4

INFLUENCE OF STOCKING DENSITY ON CARCASS CHARACTERISTICS OF FAMILY CHICKENS REARED UP TO 18 WEEKS OF AGE UNDER INTENSIVE MANAGEMENT SYSTEM

4.1 Introduction

Family poultry production systems of tropical regions are mainly based on family chickens found in nearly all villages and households in rural areas (Guèye, 1998). The term "family poultry" is defined as small-scale poultry keeping by households using family labour and locally available feed resources (Sonaiya and Swan, 2004).Family chickens are usually kept in places of varying sizes in households with some chickens being crowded while others are extensively spaced (Gabanakgosi *et al.*, 2014).

In broiler production, stocking density is a very important environmental factor which directly and indirectly influences and determines the growth performance of chickens. The influences are associated with physical restricted movement which is shown on development of locomotive apparatus, forms of broiler behavior, quality of air and litter, conditions of legs, effect on incidence of diseases and body feathering (lesions, blisters and dermatitis) (Škrbić *et al.*, 2009).The ultimate goal of poultry producers globally is to maximize kilogrammes of chicken produced per square metre of space while preventing production losses due to overcrowding to achieve a satisfactory economic return (Abudabos *et al.*, 2013). According to Thaxton *et al.* (2006), stocking density is currently expressed as a mass per unit space rather than numbers of birds being reared in a given area. Several studies have shown that rearing broilers in lower stocking density provides more intensive growth and higher absolute yield of processed carcass, better body development, *i.e.*, carcass conformation which represents the basis for development of musculature and higher shares of carcass parts which contain more meat, especially breast (Škrbić *et al.*, 2008; 2009).

There is little information on how stocking density influences carcass characteristics in family chickens. Therefore, a study was undertaken to investigate the influence of stocking density on carcass characteristics of family chickens reared up to 18 weeks of age under intensive system.

4.2 Materials and Methods

4.2.1 Study location

The experiment was carried out at the Guinea Fowl Unit of the Botswana College of Agriculture (BCA), Sebele for a period of 18 weeks. The site is at an altitude of 994 m above sea level and the coordinates are latitude 24° 33' S and longitude 24° 54' E (Aganga and Omphile, 2000). The experiment started in April and ended in August 2013. During the study period, environmental temperature averaged 21°C and ranged from 5 to 21°C.

4.2.2 Experimental design

A completely randomized design (CRD) with four treatments was used in the experiment. Each treatment was replicated four times. The four treatment levels were D1 (10 birds/m^2), D2 (13 birds/m^2), D3 (16 birds/m^2) and D4 (19 birds/m^2) in the first phase (0 to 6 weeks). The experimental birds were distributed randomly among the four stocking densities. Two birds were slaughtered at 6, 12 and 18 weeks of age from each replicate. In the second phase (7 to 12 weeks) the stocking densitieswere8 birds/m^2 (D1), 11 birds/m^2 (D2), 14 birds/m^2 (D3) and 17 birds/m^2 and 6 birds/m^2 (D1), 9 birds/m^2 (D2), 12 birds/m^2 (D3) and 15 birds/m^2 (D4) in the final phase (13 to 18 weeks).

4.2.3 Animal management

A total of 232 unsexed day-old family chicks were obtained from a farmer in Gaborone north and reared in a deep litter system. Initial body weights of the birds were determined by weighing 10% of the birds prior to allocation to four stocking densities. Birds were individually identified using wing bands. The chicks were housed under deep litter management system in an open-sided shed. The size of each pen was one metre squared (m^2). All pens were bedded with wood-shavings and equipped with one tube feeder and a 10 litre waterer. Birds were raised under artificial light for the first two weeks of acclimatization to the experimental diets prior to collection of data and later under natural light throughout the study period. Feeds and water were provided *ad libitum* throughout the experimental period. Birds in each replicate were group fed. Chickens in each pen were weighed weekly.

4.2.4 Experimental diets

Birds were fed a commercial broiler starter crumbled diet for the first 6 weeks, pelleted broiler grower diet (7 to 12 weeks) and pelleted broiler finisher diet (13 to 18 weeks). Commercial broiler diets were sourced from some retail shops in Gaborone.

Table 4.1: Chemical composition of experimental diets fed from 0 to 18 weeks of age

Chemical Composition	Feed type and age of birds		
	Broiler starter crumbles (0-6 weeks)	Broiler grower pellets (7-12 weeks)	Broiler finisher pellets (13-18 weeks)
	Amount in g/kg	Amount in g/kg	Amount in g/kg
Protein (min)	200.0	180.0	160
Moisture (max)	120.0	120.0	120.0
Fibre (max)	50.0	60.0	70.0
Calcium (min)	8.0	7.0	6.0
Calcium (max)	12.0	12.0	12.0
Fat (min)	25.0	25.0	25.0
Phosphorus (min)	6.0	5.5	5.0
Total lysine (min)	12.0	10.0	9.0

Source: OPTI Feeds Botswana (Pty) Ltd, 2014.

4.2.5 Data collection

At 6, 12 and 18 weeks of age, two birds from each replicate were sacrificed to estimate dressing percentage and weight of breasts, thighs, drumsticks, backs, necks, wings, livers, gizzards, hearts, and intestines. All birds to be slaughtered were fasted overnight and weighed the following day. After slaughter the heads, shanks, hearts, livers, intestines and gizzards were removed and individually weighed. Thereafter, the eviscerated carcasses were weighed. The weight of breast, thigh, drumstick, back, wing, head, shank and neck were also determined. The following formulae by Beg *et al.* (2011) were used to calculate carcass weight and dressing percentage:

Carcass weight = Live weight - (blood + feathers + head + shank+ digestive system)

$$\text{Dressing \%} = \frac{\text{Carcass weight}}{\text{Live weight}} \times 100$$

Giblet weight = weight of liver + heart + gizzard + neck

4.2.6 Statistical analysis

General Linear Model (GLM) procedure of Statistical Analysis System (SAS Institute, 2009) version 9.2.1 was used to analyse the data:

$Y_{ij} = \mu + \tau_i + \varepsilon_{ij}$.

Where: Y_{ij}= response variables (carcass weight, dressing percentage, breast weight, back weight, drumstick weight, thigh weight and wing weight).

μ = general mean effect. τ_i =i^{th} stocking densities effects on family chickens' growth.

Where i= 1,2,3,4. Where 1= 10 birds/m^2, 2=13 birds/m^2, 3=16 birds/m^2, 19 birds/m^2.

ε_{ij}= random error

Treatment means separation was by paired t-test and statistical significance was established at $P \leq 0.05$.

4.3 Results and Discussion

4.3.1 Carcass weight

Carcass weight was not significantly ($p>0.05$) influenced by stocking density (Tables 4.2 to 4.4). The highest carcass weight recorded in week 6 was 276.13 g (10 birds/m^2), 969.13 g (17 birds/m^2) in week 12 and 1719.75 g (9 birds/m^2) in week 18. These results are consistent with those obtained by Gabanakgosi *et al.* (2014) who found that stocking density did not have any effect on carcass weight of family chickens at 6, 12 and 18 weeks of age. The similarities may be brought by the fact that they were raised under similar conditions in which their metabolizable energy was used to maintain uniform internal body temperature. Conversely, Dozier III *et al.* (2006) reported decreased carcass weight with increased stocking density of 45 kg of body weight per m^2. The differences in the results in the two studies may be due to differences in genotypes used. In this study, family chickens of slow growing genotype were used, whereas

Dozier III *et al.* (2006) used broilers of fast growing genotype and the environmental conditions were controlled.

4.3.2 Dressing percentage

There was no significant difference in dressing percentage among the stocking densities (Tables 4.2 to 4.4). The highest dressing percentage recorded in week 6 was 56.83% (16 birds/m^2), 64.08% (17 birds/m^2) in week 12 and 68.98% (12 birds/m^2) in week 18. In contrast to the present results, Beg *et al.* (2011) reported that lower stocking densities (8 birds/m^2; 10 birds/m^2) produced higher dressing percentages in 6 weeks old broilers. The difference in the results of the current study and that of Beg *et al.* (2011) may be attributable to the differences in environmental conditions. In this study, family chickens were raised in winter where greater portion of their nutrient intake was used to generate heat in order to maintain their thermal balance thus adversely affecting dressing percentage, whereas Beg *et al.* (2011) raised them in summer where average temperature and humidity at bird level were 31.9°C and 78% respectively. Moreover, Dhaliwal and Nagra (2006) found that dressing percentage increased with decreased stocking density (125 birds/m^2) in Japanese quails. The differences in the results of the current study and that of Dhaliwal and Nagra (2006) may be attributable to differences in bird species used.

4.3.3 Breast weight

Stocking density had no significant effect on breast weight (Tables 4.2 to 4.4).The highest breast weight observed in week 6 was 64.38 g (16 birds/m^2), 216.13 g (17 birds/m^2) in week 12 and 390.25 g (15 birds/m^2) in week 18. Similar observations were made by Feddes *et al.* (2002), Moreira *et al.* (2004), Ravindran *et al.* (2006), Sekeroglu *et al.* (2011) and Zuowei *et al.* (2011) in broilers. In contrast to the present results, Škrbić *et al.* (2011) observed that broilers reared at lower stocking density (12 birds/m^2) had significantly higher yield of breast compared to those with a stocking density of 16 birds/m^2. The variations in the results for the two studies may be due to the differences in their genotype. In this study, family chickens of slow growing genotype were used whereas Škrbić *et al.* (2011) used broilers of fast growing genotype. The growth of breast muscle in broilers of the fast growing genotype is rapid such that provision of wider space helps them to reach their full potential. Similarly, Osman (1993) observed a decrease in breast weight of ducks with increasing stocking density (16 birds/m^2). The differences in the results of the current study and that of Osman (1993) may be due to differences in bird species used.

Table 4.2: Least square means for carcass traits of family chickens at 6 weeks of age reared under intensive system

Treatments	Weight of carcass parts (g)								
Bird density/m^2	Carcass	Dressing(%)	Breast	Back	Drumstick	Thigh	Wing	Giblet	shank
10	276.13^a	56.63^a	62.75^a	38.50^a	19.94^a	21.88^a	20.38^a	70.88^a	9.63^a
13	259.81^a	55.85^a	61.63^a	37.38^a	18.19^a	20.81^a	18.94^a	62.75^a	8.94^a
16	274.75^a	56.83^a	64.38^a	36.13^a	20.63^a	22.25^a	20.31^a	64.88^a	9.88^a
19	257.31^a	55.20^a	58.75^a	35.88^a	18.81^a	20.38^a	18.94^a	65.00^a	9.19^a
SE	17.31	0.89	4.21	2.77	1.71	1.69	1.34	3.71	1.22
P-value	0.8098	0.5643	0.8126	0.8996	0.7491	0.8456	0.7764	0.4763	0.9471
CV	12.9642	3.1558	13.5930	14.9706	17.6670	15.8738	13.6067	11.2591	36.5492

Means within the same column within a parameter not having similar superscripts are significantly different ($P<0.05$), SE = Standard Error, CV = Coefficient of variation.

Table 4.3: Least square means for carcass traits of family chickens at 12 weeks of age reared under intensive system

Treatments	Weight of carcass parts (g)								
Bird density/m^2	Carcass	Dressing(%)	Breast	Back	Drumstick	Thigh	Wing	Giblet	Shank
8	943.50^a	59.13^a	203.00^a	135.50^a	67.25^a	70.50^a	57.00^a	175.88^a	28.86^a
11	909.00^a	55.55^a	197.38^a	128.13^a	66.63^a	71.69^a	54.13^a	175.88^a	26.94^a
14	959.50^a	62.33^a	207.00^a	120.00^a	70.56^a	73.25^a	58.81^a	171.25^a	33.38^a
17	969.13^a	64.08^a	216.13^a	150.00^a	75.00^a	77.88^a	60.44^a	143.25^a	30.19^a
SE	43.43	3.19	12.53	11.04	4.89	5.19	2.97	13.58	2.30
P-value	0.7766	0.2943	0.7578	0.3078	0.6171	0.7633	0.5019	0.3089	0.2684
CV	9.1895	10.5798	12.1727	16.5118	13.9878	14.1469	10.3030	16.3066	21.8240

Means within the same column within a parameter not having similar superscripts are significantly different ($P<0.05$), SE = Standard Error, CV = Coefficient of variation.

Table 4.4: Least square means for carcass traits of family chickens at 18 weeks of age reared under intensive system

Treatments	Weight of carcass parts (g)								
Bird density/m²	Carcass	Dressing(%)	Breast	Back	Drumstick	Thigh	Wing	Giblet	Shank
6	1565.63ᵃ	67.50ᵃ	387.88ᵃ	226.38ᵃ	119.00ᵃ	135.88ᵃ	91.13ᵃ	264.38ᵃ	39.50ᵃ
9	1719.75ᵃ	67.60ᵃ	390.13ᵃ	248.63ᵃ	135.31ᵃ	155.38ᵃ	100.31ᵃ	294.13ᵃ	46.00ᵃ
12	1681.50ᵃ	68.98ᵃ	379.75ᵃ	260.13ᵃ	132.25ᵃ	146.75ᵃ	102.25ᵃ	291.63ᵃ	43.63ᵃ
15	1597.06ᵃ	67.80ᵃ	390.25ᵃ	221.38ᵃ	125.25ᵃ	137.31ᵃ	95.50ᵃ	285.25ᵃ	42.19ᵃ
SE	71.54	1.09	18.56	20.42	6.49	8.26	3.70	14.01	2.47
P-value	0.4235	0.7614	0.9742	0.5140	0.3295	0.3486	0.1962	0.4565	0.3246
CV	8.7186	3.2090	9.5938	17.0763	10.1419	11.4894	7.6041	9.8729	16.3316

Means within the same column within a parameter not having similar superscripts are significantly different ($P<0.05$), SE = Standard Error, CV = Coefficient of variation.

4.3.4 Back weight

Stocking density had no significant effect on back weight (Tables 4.2 to 4.4) of family chickens. The highest back weight was found in stocking densities of 10 birds/m² (38.50 g) at week 6, 17 birds/m² (150.00 g) at week 12 and 12 birds/m² (260.13 g) at week 18. These results are in agreement with Thomas *et al.* (2004) who found no influence of stocking density on carcass characteristics of broilers grown at densities of 10, 15 and 20 birds/m².

4.3.5 Drumstick weight

It is evident from Tables 4.2 to 4.4 that drumstick weight of family chickens was not significantly affected by stocking density. However, the highest drumstick weight was found in the stocking densities of 16 birds/m² (20.63 g) at week 6, 17 birds/m² (75.00 g) at week 12 and 9 birds/m² (135.31 g) at week 18. In agreement with the current results, El-Deek and Al-Harthi (2004) found that stocking density does not affect drumstick yield in broilers. The results of the present study are not in-line with Škrbić *et al.* (2011) who observed that at 42 days old broilers reared at a stocking density of 12 bird/m² had significantly high drumstick weight compared to those at a stocking density of 16 birds/m². The differences in the results of the current study and that of Škrbić *et al.* (2011) may be attributable to differences in birds' genotype. In this study family chickens of slow growing genotype were used, whereas Škrbić *et al.* (2011) used broilers of fast growing genotype and the environmental conditions were controlled.

4.3.6 Thigh weight

No significant difference in thigh weight was found among different stocking densities (Tables 4.2 to 4.4). The highest thigh weight observed in week 6 was 22.25 g (16 birds/m^2), 77.88 g (17 birds/m^2) in week 12 and 155.38 g (9 birds/m^2) in week 18. The present results are in consonance with those of Lewis *et al.* (1997), Mizubuti *et al.* (2000), Jayalakshmi *et al.* (2009), Beg *et al.* (2011) and Simsek *et al.* (2011) who found no influence of stocking density on the weight of the thigh of broilers. However, Tong *et al.* (2012) found that the thigh yield of local chickens at 8 weeks old was significantly affected at stocking density of 35 birds/m^2 compared to 25 birds/m^2 and 45 birds/m^2. Environmental factors might have contributed to variations in the results of this study. The local chickens in the study of Tong *et al.* (2012) were raised in an environmentally controlled house compared to naturally ventilated house in this study.

4.3.7 Wing weight

The wing weight of family chickens was not affected by stocking density. The highest wing weight was recorded at stocking density of 10 birds/m^2 (20.38 g) at week 6, 17 birds/m^2 (60.44 g) at week 12 and 12 birds/m^2 (102.25 g) at week 18. Similar observations were made by Moreira *et al.* (2004), El-Deek and Al-Harthi (2004), Jayalakshmi *et al.* (2009), Sekeroglu *et al.* (2011) and Simsek *et al.* (2011) in broilers. Likewise, Nahashon *et al.* (2009) observed no significant difference in mean wing weights of French guinea fowl broilers due to stocking density. However, Yakubu *et al.* (2010) reported that stocking densities of 8.3 birds/m^2, 11.1 birds/m^2 and 14.3 birds/m^2 had significant effect on wing weight of broilers. The difference in the results of the current study and that of Yakubu *et al.* (2010) may be attributable to the differences in breeds of chickens used. In this study, family chickens of slow growing genotype were used, whereas Yakubu *et al.* (2010) used hybrid broilers of fast growing genotype. Intensively raised hybrid broilers grow rapidly and as they approach market age and weight, their bodies take up most of the allotted space, leaving no room to perform simple exercises needed for muscular development (Whitehead, 2008; Jones, 2010), and this may have an effect on wing development since they are late maturing traits (Nsoso *et al.*,2006).

4.4 Conclusion

Carcass characteristics of family chickens were not influenced by stocking density. However, carcass weight was highest at 10 birds/m^{2}in the first phase, 17 birds/m^2 in the second phase and 9 birds/m^2 in the third phase. Therefore, it appears that raising family chickens at 10 birds/m^2 to market age of 18 weeks during winter has no effect on growth parameters.

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Influence of Stocking Density on Bone Development in Family Chickens Reared up to 18 Weeks of Age Under Intensive System

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Abstract: This study investigated the influence of stocking density on bone development of family chickens up to 18 weeks of age. A total of 232 unsexed day-old family chicks were used in a completely randomized design. Birds were randomly assigned to four stocking densities, i.e., D1 (10 birds/m^2), D2 (13 birds/m^2), D3 (16 birds/m^2) and D4 (19 birds/m^2) in the first phase (0-6 weeks). Each treatment was replicated four times. Two birds were slaughtered at 6, 12 and 18 weeks of age from each replicate to evaluate bone length, bone width, bone weight and bone chemical composition (ash weight, Ca, P and Mg). In the second phase (7 to 12 weeks) the stocking densities were 8 birds/m^2 (D1), 11 birds/m^2 (D2), 14 birds/m^2 (D3) and 17 birds/m^2 and 6 birds/m^2 (D1), 9 birds/m^2 (D2), 12 birds/m^2 (D3) and 15 birds/m^2 (D4) in the final phase (13 to 18 weeks). General Linear Model (GLM) procedure of Statistical Analysis System was used to estimate the differences between treatment means for different stocking densities. Stocking density in all the three phases did not have a significant ($p>0.05$) effect on bone dimensions and chemical composition. It is therefore concluded that stocking density had no influence on bone development of family chickens raised up to 18 weeks of age under intensive system probably due to slaughtering that occurred at six weekly intervals. It appeared that family chickens could be raised at a density of 15 birds/m^2 in winter without any detrimental effect on bone development and related parameters. Further studies should be done using identical densities throughout the research period to avoid disturbing the control which will make blocking by age possible.

Key words: Bone development, family chickens, intensive system, stocking density

INTRODUCTION

Stocking density is a housing variable that can affect chickens' development. Bone development is part of animal growth and the growth of the skeleton determines the size and proportions of the body (Yakubu *et al.*, 2010; Buijs *et al.*, 2012). Bone is a dynamic tissue that is influenced by physiological, nutritional and physical factors such as mechanical stress and physical activities (Rath *et al.*, 2000). The deposition of bone is regulated primarily by parathyroid hormone, which is secreted in response to low serum calcium levels (Klein and Enders, 2010).

Several studies have been conducted to study the effect of stocking density on broiler production. According to Hall (2001), increased stocking density can negatively influence skeletal development of broilers, as shown by an increase in leg culls, which may be due to a decrease in activity as density increases. Skrbic *et al.* (2009) observed that providing more floor space per chicken influenced the level of physical activity, development and firmness of the skeleton, especially legs. The author noted that physical activity of broilers influenced cross section of the cortex and as a result improved their mechanical characteristics by better supply with blood of epiphysis of long bones and sufficient mineralization. In another study, Skrbic *et al.* (2011) observed that more physical activity of broilers in lower stocking density improved the parameters of tibia quality.

According to Sonaiya and Swan (2005), the term "family poultry" is defined as small-scale poultry rearing by households using family labour and locally available feed resources. Family poultry production systems of tropical regions are mainly based on family chickens found in nearly all villages and households in rural areas (Gueye, 1998). Generally, feeding, health control and housing are inadequate in family chicken rearing.

There is little information on how stocking density influences bone development in family chickens. The absence of stocking density standards for family chickens forces farmers to rely on personal experience in determining the space allowances and this may affect their productivity. Therefore, a study was undertaken to investigate the influence of stocking density on bone development of family chickens reared up to 18 weeks of age under intensive system.

MATERIALS AND METHODS

Study site: The experiment was carried out at the Guinea Fowl Unit of the Botswana College of Agriculture (BCA), Sebele for a period of 18 weeks. The site is at an

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altitude of 994 m above sea level and the coordinates are latitude 24°33' S and longitude 24°54' E (Aganga and Omphile, 2000). The experiment ran from April to August 2013. During the study period, environmental temperature averaged 21°C and ranged from 5 to 21°C.

Experimental design: A completely randomized design (CRD) with four treatments was used in the experiment. Each treatment was replicated four times. The four treatment levels were D1 (10 birds/m²), D2 (13 birds/m²), D3 (16 birds/m²) and D4 (19 birds/m²) in the first phase (i.e., 0 to 6 weeks). The experimental birds were distributed randomly among the four stocking densities. Two birds were slaughtered at 6, 12 and 18 weeks of age from each replicate. In the second phase (i.e., 7 to 12 weeks) the stocking densities were 8 birds/m² (D1), 11 birds/m² (D2), 14 birds/m² (D3) and 17 birds/m² and 6 birds/m² (D1), 9 birds/m² (D2), 12 birds/m² (D3) and 15 birds/m² (D4) in the final phase (i.e., 13 to 18 weeks).

Animal management: A total of 232 unsexed day-old family chicks were obtained from a farmer in Gaborone north and reared in a deep litter system. Initial body weights of the birds were determined by weighing 10% of the birds prior to allocation to four stocking densities. Birds were individually identified using wing bands. Chicks were housed under deep litter management system in an open-sided shed. The size of each pen was one metre squared (m²). All pens were bedded with wood-shavings and equipped with one tube feeder and a 10 L waterer. Birds were raised under artificial light for the first two weeks to acclimatize birds to the experimental diets prior to collection of data and thereafter natural light throughout the study period. Feeds and water were provided *ad libitum* throughout the study period. Birds in each replicate were group fed. Chickens in each pen were individually weighed on a weekly basis.

Experimental diets: Birds were fed a commercial broiler starter crumbled diet up to 6 weeks of age, pelleted broiler grower diet from 7 to 12 weeks and pelleted broiler finisher diet from 13 to 18 weeks. Commercial broiler diets were purchased from some retail shops in Gaborone.

Data collection: At 6, 12 and 18 weeks of age, two birds from each replicate were sacrificed to determine bone dimensions and chemical composition. After slaughter carcasses were placed in plastic bags, identified and chilled to 0°C overnight in a cold room and bones removed 24 h post mortem. The right and left tibiae and the right humerus from each of the birds were removed and defleshed without boiling. Thereafter, bones were individually weighed using an electronic balance with a precision of 0.001 g, (Sartorius AG Germany, TE 313S model) and their widths and lengths determined using an electronic calliper with an accuracy of 0.001 cm, (Starrett® 798 B 12"/300 mm model). The left tibiae were used for bone chemical composition analysis (ash, Ca, P, Mg) and the right tibiae for bone dimensions. Bone samples were oven-dried in porcelain crucibles at 105°C for 48 h and weighed (Liu *et al.*, 2003). Thereafter, bone samples were ashed in a muffle furnace at 550°C for 8 h. Approximately 1 g of ash samples was dissolved in 10 mL of 3M hydrochloric acid and boiled for 10 min. The samples were then allowed to cool and filtered into a 100 mL volumetric flask. Thereafter, the volume was topped up to 100 mL with distilled water and later analyzed for minerals according to AOAC (1996).

Statistical analysis: General Linear Model (GLM) Procedures of Statistical Analysis System (SAS Institute, 2009) version 9.2.1 was used to analyze the data according to the following statistical model:

$$Y_i = \mu + J_i + g_i$$

where, Y_i: Response variables (bone width, bone length, bone weight and bone chemical composition). μ: General mean effect. J_i: i^{th} stocking densities effects on family chickens' growth. Where, i = 1, 2, 3, 4. Where 1 = 10 birds/m², 2 = 13 birds/m², 3 = 16 birds/m², 19 birds/m². g_i = random error.

Least significant difference comparisons were made between treatment means using paired t-test and statistical significance was established at $p \leq 0.05$.

RESULTS AND DISCUSSION

Bone dimensions: Stocking density had no significant ($p > 0.05$) influence on tibia and humerus length of family chickens (Tables 2 to 4). However, the longest tibia (78.41 mm) and humerus length (55.52 mm) was found in the stocking densities of 19 bird/m² at week 6, 14 bird/m² (131.33 and 85.61 mm) at week 12 and 9 bird/m² (149.67 and 93.14 mm) at week 18, respectively. These

Table 1: Chemical composition of experimental diets fed to family chickens from 0 to 18 weeks of age

	Feed type and age of birds		
Chemical composition	Broiler starter crumbles (0-6 wks)	Broiler grower pellets (7-12 wks)	Broiler finisher pellets (13-18 wks)
	Amount in g/kg		
Protein (min)	200.0	180.0	160
Moisture (max)	120.0	120.0	120.0
Fibre (max)	50.0	60.0	70.0
Calcium (min)	8.0	7.0	6.0
Calcium (max)	12.0	12.0	12.0
Fat (min)	25.0	25.0	25.0
Phosphorus (min)	6.0	5.5	5.0
Total lysine (min)	12.0	10.0	9.0

Source: OPTI Feeds Botswana (Pty) Ltd. 2014

Table 2: Least square means for bone dimensions of family chickens at 6 weeks of age reared under intensive system

Treatment	Tibia			Humerus		
Bird density/m^2	Length (mm)	Width (mm)	Weight (g)	Length (mm)	Width (mm)	Weight (g)
10	77.22^a	5.25^a	4.63^a	55.07^a	5.56^a	2.50^a
13	75.71^a	4.80^a	4.31^a	54.02^a	5.10^a	2.63^a
16	78.02^a	5.10^a	4.50^a	54.91^a	5.40^a	2.31^a
19	78.41^a	4.86^a	4.25^a	55.52^a	5.30^a	2.06^a
SE	1.74	0.21	0.48	1.13	0.15	0.21
p-value	0.7103	0.4103	0.9425	0.8211	0.2377	0.2865
CV	4.5045	8.2050	21.8815	4.1286	5.6659	17.3232

Means within the same column within a parameter not having similar superscripts are significantly different ($p<0.05$)
SE: Standard Error, CV: Coefficient of variation

Table 3: Least square means for bone dimensions of family chickens at 12 weeks of age reared under intensive system

Treatments	Tibia			Humerus		
Bird density/m^2	Length (mm)	Width (mm)	Weight (g)	Length (mm)	Width (mm)	Weight (g)
8	125.82^a	7.88^a	13.81^a	83.54^a	7.96^a	5.69^a
11	126.03^a	7.89^a	13.69^a	81.18^a	7.88^a	5.75^a
14	131.33^a	8.41^a	14.75^a	85.61^a	8.25^a	6.44^a
17	127.79^a	8.46^a	15.06^a	83.48^a	8.21^a	6.63^a
SE	2.16	0.31	0.88	1.58	0.13	0.45
p-value	0.2939	0.3983	0.6258	0.3141	0.1876	0.3773
CV	3.3858	7.5774	12.2578	3.7764	3.3032	14.6456

Means within the same column within a parameter not having similar superscripts are significantly different ($p<0.05$)
SE: Standard Error, CV: Coefficient of variation

Table 4: Least square means for bone dimensions of family chickens at 18 weeks of age reared under intensive system

Treatments	Tibia			Humerus		
Bird density/m^2	Length (mm)	Width (mm)	Weight (g)	Length (mm)	Width (mm)	Weight (g)
6	143.30^a	9.39^a	18.31^a	90.05^a	9.40^a	8.38^a
9	149.67^a	10.11^a	20.69^a	93.14^a	9.61^a	8.75^a
12	143.28^a	9.68^a	19.19^a	88.91^a	9.44^a	8.44^a
15	145.07^a	9.53^a	18.81^a	88.93^a	9.01^a	8.44^a
SE	2.39	0.38	1.11	1.34	0.21	0.49
p-value	0.2421	0.5895	0.4925	0.1376	0.2876	0.9478
CV	3.2834	7.9160	11.5248	2.9597	4.5447	11.5950

Means within the same column within a parameter not having similar superscripts are significantly different ($p<0.05$)
SE: Standard Error, CV: Coefficient of variation

results are consistent with those obtained by Oliveira *et al.* (2012) who found that the lengths of both tibiae and humeri were not affected by stocking density of 10 and 16 birds/m^2. In disagreement with current results, Buijs *et al.* (2012) found that increased stocking density (15.5, 18.5 and 21.8 birds/m^2) in broilers resulted in shorter tibiae. The variations in the results for the two studies may be due to the differences in genotype. In this study family chickens of slow growing genotype were used compared to broilers of fast growing genotype in the study of Buijs *et al.* (2012). This suggests that as broilers increase in body weight, the tibiae increase in width to support the muscle mass thus forcing them to curve. In this study, the length of tibia increased by 65 and 13.8% between 6 and 12 weeks and 12 and 18 weeks, respectively. On the other hand, humerus length increased by 52 and 8% between 6 and 12 weeks and 12 and 18 weeks, respectively. This indicates that bone development and growth in family chickens were most pronounced during the first 12 weeks, a similar observation was made by Moreki *et al.* (2011) who found that tibia and humerus length increased by 46 and 36% in broiler breeder pullets between 6 and 12 weeks, respectively. According to Ross Breeders (2006) the skeletal size of broiler breeder is fixed at 12 weeks of age. Rath *et al.* (2000) stated that the increment in bone length is correlated with an increase in the content of hydroxylysylpyridinoline and lysylpyridinoline, the collagen crosslinks.

No significant influence of stocking density on tibial and humerus width of family chickens could be detected (Tables 2 to 4). However, the greatest tibial and humerus width was found in the stocking densities of 10 bird/m^2 (5.25 and 5.56 mm) at week 6, 17 bird/m^2 and 14 birds/m^2 (8.46 mm for tibia; 8.25 mm for humerus) at week 12 and 9 bird/m^2 (10.11 mm; 9.61 mm) at week 18, respectively. Similarly, Simsek *et al.* (2011) found that the width of tibia was not affected by stocking densities of 22.5, 18.75, 15, 11.25, 7.5 broilers/m^2. Ventura *et al.* (2010) also reported no influence of stocking density (8 birds/m^2, 13 birds/m^2 and 18 birds/m^2) on width of tibia in broilers. In disagreement with the present

Table 5: Least square means for bone chemical composition of family chickens at 6 weeks of age reared under intensive system

Treatments	Variables			
Bird density/m^2	Ash (g)	Ca (%)	P (%)	Mg (%)
10	0.84^a	32.68^a	24.23^a	0.82^a
13	0.79^a	32.83^a	24.25^a	0.83^a
16	0.86^a	32.94^a	24.35^a	0.90^a
19	0.80^a	32.84^a	24.27^a	0.85^a
SE	0.07	0.20	0.21	0.02
p-value	0.8236	0.8363	0.9760	0.7086
CV	16.1889	1.2104	1.6986	3.4858

Means within the same column within a parameter not having similar superscripts are significantly different ($p<0.05$)

SE: Standard Error, CV: Coefficient of variation

Table 6: Least square means for bone chemical composition of family chickens at 12 weeks of age reared under intensive system

Treatments	Variables			
Bird density/m^2	Ash (g)	Ca (%)	P (%)	Mg (%)
8	2.83^a	32.90^a	23.50^a	0.78^a
11	2.89^a	32.90^a	23.65^a	0.78^a
14	3.04^a	33.28^a	23.86^a	0.83^a
17	3.29^a	32.68^a	23.41^a	0.82^a
SE	0.16	0.27	0.21	0.03
p-value	0.2383	0.5097	0.4803	0.3244
CV	10.7187	1.6686	1.7801	6.3518

Means within the same column within a parameter not having similar superscripts are significantly different $p<0.05$

SE: Standard Error, CV: Coefficient of variation

Table 7: Least square means for bone chemical composition of family chickens at 18 weeks of age reared under intensive system

Treatments	Variables			
Bird density/m^2	Ash (g)	Ca (%)	P (%)	Mg (%)
6	4.66^a	34.19^a	24.52^a	0.76^a
9	5.16^a	34.23^a	24.55^a	0.79^a
12	4.81^a	34.31^a	24.76^a	0.81^a
15	4.72^a	34.25^a	24.57^a	0.79^a
SE	0.27	0.29	0.11	0.02
p-value	0.5812	0.9933	0.4586	0.2904
CV	11.17626	1.6891	0.9168	4.4908

Means within the same column within a parameter not having similar superscripts are significantly different ($p<0.05$)

SE: Standard Error, CV: Coefficient of variation

results, Oliveira *et al.* (2012) observed that the width of humerus in Ross 308 and Hybro PG broilers was affected by stocking density of 10 and 16 birds/m^2 at 42 days of age. The differences in the results of the current study and that of Oliveira *et al.* (2012) may be due to differences in birds' genotype. Several studies have observed that hybrid broilers raised intensively grow rapidly and that as they approach market age and weight, their bodies take up most of the allotted space, leaving no room to perform simple exercises which may lead to a decrease in bone mass of the wings (Lewis *et al.*, 1997; Jones, 2010). In this study, width of tibia increased by 63.1 and 18.6% between 6 and 12 weeks and 12 and 18 weeks, respectively, whereas humerus width increased by 51.2 and 16.0% between 6 and 12 weeks and 12 and 18 weeks, respectively. This suggests that in response to increased body weight, bones of the family chickens increased in bone width.

The weight of tibia and humerus was not significantly ($p>0.05$) influenced by stocking density (Table 2 to 4). In agreement with the current findings on tibia, Buijs *et al.* (2012) found that stocking density had no effect on tibia weight in broilers. Similarly, Oliveira *et al.* (2012) observed that weight of tibia and humerus in broilers was not affected by stocking density at 42 days of age.

Bone chemical composition: Stocking density had no significant ($p>0.05$) effect on chemical composition of bones (Table 5 to 7). The highest bone ash weight was found in the stocking densities of 16 bird/m^2 (0.86 g) at week 6, 17 bird/m^2 (3.29 g) at week 12 and 9 bird/m^2 (5.16 g) at week 18. In agreement with current results, Tablante *et al.* (2003) found that bone ash of broilers was not affected by stocking densities of 10, 15 and 20 birds/m^2. Although the mean weight of ash did not differ significantly among birds reared at different stocking densities bone weight increased with age. Similar observation was made by Moreki *et al.* (2011) who reported a significant increase in bone ash with age in broiler breeders up to 18 weeks of age. In the present study, the highest levels of Ca, P and Mg were observed at 16 birds/m^2 in week 6 (32.94, 24.35 and 0.90%), 14 birds/m^2 in week 12 (33.28, 23.86 and 0.83%) and 12 birds/m^2 in week 18 (34.31, 24.76 and 0.81%), respectively. The levels of Ca, P and Mg decreased from 6 to 12 weeks of age and from 12 to 18 weeks only Ca and P contents increased, whereas Mg content continued to decline. The decline of Ca and P from 6 to 12 weeks of age could be due to the birds increased demand for nutrients for increased muscle mass.

Conclusion: Bone dimensions and bone chemical composition were not influenced by stocking density. Therefore, it can be concluded that stocking density had no influence on bone development probably because of slaughtering that occurred at 6, 12 and 18 weeks of age. Further studies should be done using identical densities throughout the research period to avoid disturbing the control which will make blocking by age possible.

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