# Effects of ascorbic acid supplementation on broiler chickens stocked at two different densities in a humid tropical environment

**Adeyemi, O.A.*, Adedoyin, E.O., Olaleye, O.O., Njoku, P.C. and Sanwo, K.A.**
College of Animal Science and Livestock Production, Federal University of Agriculture, Abeokuta, Nigeria.
*Corresponding author: olajideadeyemi@gmail.com

## Abstract

The quest to meet up with the high demand for poultry products has made farmers adopt new means of rearing birds without incurring more cost. One of these husbandry practices is by varying the stocking density. Increasing stocking density may however result in additional stress which may reduce production efficiency. Arising from this, a completely randomized design experiment was carried out to determine the effects of ascorbic acid (AA) supplementation on performance and carcass characteristics of broiler chickens stocked at two different stocking densities. A total of 150 day-old Marshall broilers were brooded for 2wk. and randomly allotted into 4 treatment groups of 3 replicates each: Treatment 1 optimal stocking at 7birds/$m^2$, Treatment 2 double stocking at 14 birds/$m^2$and no AA supplementation, Treatment 3 double stocking at 14 birds/$m^2$and 150% AA supplementation and Treatment 4 double stocking at 14 birds/$m^2$ and 300% AA supplementation. Feed intake, live weight, feed:gain ratio and livability differed ($p<0.05$) among treatment groups. Total and daily feed intake of birds on optimal stocking density (Treatment 1) and those of birds on double optimal stocking density with 300mg AA/litre of water were similar (3823.00 vs 3794.12 g/bird and 91.02 vs 90.34g/bird, $p>0.05$). Highest weight gain amongst birds on double stocking density was obtained from birds with 300mg AA supplementation/litre of water (1391.82 compared with 1161.48 and 1308.33 g/bird for Treatments 2 and 3, respectively. Dressed weight, dressing percentage, breast cut, drumstick, wing and liver weights as percentage of live weight were influenced ($p<0.05$) by stocking density and AA supplementation. Gross Margin analysis indicated that increasing the stocking density resulted in % gross margin changes of -56.73, +50.73 and 80.47% for birds produced on double stocking density without AA, double stocking density and 150mg AA/litre of drinking water and double stocking density and 300mg AA/litre of drinking water, respectively compared to the optimal stocking density. The findings of this study conclude that double stocking density of broilers at 14birds/$m^2$and 300mg AA supplementation/litre of water can be adopted in the study location to improve production efficiency per unit space.

**Keywords:** Broilers, stocking density, ascorbic acid supplementation, feed intake, carcass.

## Introduction

Chicken producers are interested in increasing the number of birds per unit area of floor space and reducing housing and labour cost per bird (Ayoola *et al*., 2014). Stocking density has thus become a veritable economic tool of consideration due to the intensification of industrial poultry production, which resulted in decreased performance per bird housed and too small profit margin (Carey *et al*., 1995). Stocking density can be defined as the number of birds per unit space or space per bird or mass per unit of space (Thaxton *et al*., 2006). Stocking density of broiler chickens contributes either negatively or positively to their performance as uncontrolled increases in number of birds per unit space may lead to overcrowding. Farmers often increase stocking density with the aim of increasing profit without necessary increase in size of facilities. In many cases, producers have to settle for slightly reduced performance to achieve a satisfactory economic return. The ultimate goal of poultry producers worldwide is to maximize body weight of chicken produced per square metre of space while preventing production losses due to overcrowdings to achieve a satisfactory economic return (Abudabos, 2013).

High stocking densities always result in the build-up of heat and may consequently lead to heat stress. Several methods are available to alleviate the effects of high environmental temperature and increased stocking density on performance of poultry which include the use of ventilators, fans and foggers on poultry farms (DEFRA, 2005). Feed and feeding manipulations (Gonzalez-Esquerra and Leeson, 2006), vitamin supplementation (Curca *et al.,* 2004) and mineral supplementation (Teeter *et al.*, 1985) are among the other means of reducing the negative effects of high stocking densities. It is often expensive to cool animal buildings hence methods aimed at reducing body temperature are focused mostly on dietary manipulation.

Ascobic acid (AA) also known as Vitamin C produces anti-stress hormones which help to combat the effect of stress caused on animal body. AA is a water soluble anti-oxidant that is normally produced in adequate quantity by metabolism in birds when not stressed. Butcher and Miles (2003) explained that its use as an anti-stress agent is often considered during periods of heat-stress since AA plays a major role in the biosynthesis of corticosterone, a hormone that enhances energy supply during stress.

It is known that high stocking densities pose stress on birds. Therefore, as a way of improving performance, ascorbic acid could be administered to birds stocked at double the optimal stocking density in a humid tropical environment to evaluate the effects of such supplementation on growth performance and carcass attributes of broilers. A study was therefore conducted to determine the effects of ascorbic acid supplementation on performance and carcass characteristics of broiler chickens stocked at two different stocking densities.

## Materials and Methods

### *Experimental location*

The experiment was carried out at the poultry unit of the Directorate of University Farms (DUFARM), Federal University of Agriculture, Abeokuta, Nigeria. The location lies within the rainforest vegetation zone of South West Nigeria with mean annual rainfall of 1100 mm and temperature of 34.7 $^0$C and relative humidity of 82%. It is in the region 70 m above sea level on latitude 7$^0$5' to 7$^0$8'N and longitude 3$^0$11.2'E (Federal University of Agriculture, Abeokuta, Meteorological Station).

### *Experimental birds and management*

One hundred and fifty day-old broiler chicks were purchased from a reputable commercial hatchery in Abeokuta, Ogun State, Nigeria and floor brooded together for 2 wk. Out of the original number purchased, 147 2-wk old chicks were randomly allotted into 5 treatment combinations of stocking densities: optimal (7 birds/m$^2$) and double (14 birds/m$^2$) after balancing their live weight and ascorbic acid (AA) supplementation in drinking water. The optimal and double stocking densities translated into $0.1m^2$/bird and $0.05m^2$/bird, respectively. The optimal stocking density was as recommended by Atteh (2004). The four treatment groups were:

1. Stocking Density of 0.1 $m^2$/bird (Positive Control)
2. Stocking Density of 0.05 $m^2$/bird (Negative Control)
3. Stocking Density of 0.05 $m^2$/bird + 150 mg AA/litre of drinking water
4. Stocking Density of 0.05 $m^2$/bird + 300 mg AA/litre of drinking water

Each treatment was replicated thrice with appropriate number of birds per replicate in the respective stocking densities.

The experiment lasted for 42 d. The birds were fed broiler starter and finisher diets containing 3000 Kcal/kg ME and 23% CP; and 3200 kcal/kg ME and 19% CP, respectively, *ad libitum* in the course of the experiment. Water was supplied ad lib. Routine vaccinations and other medications were administered.

The study protocol was approved and conducted in accordance with the Animal Ethics Committee guidelines of Federal University of Agriculture, Abeokuta, Nigeria (FUNAAB, 2013). The indices of performance measured were feed intake, weight gain, livability while feed conversion was calculated.

*Nutrient digestibility study*

At the third wk of the trial, a nutrient digestibility trial was carried out. Because of the nature of the experiment and in order to easily maintain the stocking density, an elevated, screen meshed floor platform was placed in the replicate pens for collection of droppings. The birds were acclimatized for 3 d prior to the commencement of excreta collection. Sample collection was done for 4 d, excreta (devoid of feed particles, shed feathers and other contaminants) were collected on aluminum foil papers placed under the raised platform on a daily basis using the total collection method as described by Cullison (1982) and dried in a Gallenkhamp® drying cabinet at 60 $^oC$ for 12 h. Feed samples and ground excreta samples were used for proximate analysis of nutrients. Daily feed intake during the 4-d collection period was also measured.

*Carcass evaluation*

At 42 d of the experiment, 3 birds per replicate were selected on the basis of the closeness of their live weight to the average replicate weight, starved overnight and killed by cutting the throat, bled and defeathered. The head, crop and shank were removed and the carcass eviscerated for calculation of dressing percentage of the different parts: thigh, drumstick and breast-cut. In addition, organs including the heart, liver and gizzard were excised out and weighed.

*Cost implications of treatments*

Cost implications of treatments, sales and gross margins were calculated as itemized below:

Total Input Cost / bird
= Cost of 3-wk old bird + Cost of feed/bird + Cost of AA supplementation + Cost of labour/bird + Cost of space/ bird + Cost of medication/bird

Total Input Cost/treatment
= Total Input Cost/bird x Number of birds stocked per treatment.

Total Sales/Treatment
= Selling price/kg x Av. live weight (kg) x Number of live birds/treatment.

Margin
= Total Sales/treatment - Total Input Cost/ treatment

Chemical and analytical procedure

*Proximate analysis*

The proximate analyses of the experimental diets and droppings were carried out using the method of AOAC (1995).

*Statistical analysis*

Data collected were subject to a one-way analysis of variance as outlined in Statistical Analysis Software (SAS, 2000) package. Significant (P<0.05) different means among variables were separated using New Duncan's Multiple Range Test as contained in SAS (2000) package.

## Results and Discussion

Feed intake, live weight, feed: gain and livability of birds were found to differ (p<0.05) between optimal stocking density and double stocking density (Treatments 1 and 2, Table 1). The addition of AA in drinking water (Treatments 3 and 4) however appeared to result in an improvement in these indices of performance at double stocking density compared to Treatment 2 (double stocking density without AA addition).

Table 1. Growth performance and cost margin evaluation of broiler chickens supplemented with ascorbic acid in drinking water at optimal and double stocking densities

| | Treatment[1] | | | | **SEM** |
|---|---|---|---|---|---|
| | Optimal SD | Double SD | Double SD +150mg AA/L | Double SD + 300mgAA/L | |
| Initial weight (g) | 234.80 | 235.20 | 235.00 | 235.00 | 0.51 |
| Final weight (g) | 1,833.33[a] | 1,396.68[d] | 1,543.33[c] | 1,626.82[b] | 2.88 |
| Weight gain (g) | 1,598.53[a] | 1,161.48[d] | 1,308.33[c] | 1,391.82[b] | 20.35 |
| Average daily weight gain (g) | 38.06[a] | 27.65[c] | 31.15[bc] | 33.14[b] | 0.89 |
| Total feed intake (g) | 3,823.00[a] | 3,503.45[c] | 3,682.17[b] | 3,794.12[a] | 55.78 |
| Average daily feed intake (g) | 91.02[a] | 83.41[c] | 87.67[b] | 90.34[a] | 2.34 |
| Feed: Gain ratio | 2.39[c] | 3.02[a] | 2.81[ab] | 2.73[b] | 0.72 |
| Livability % | 95.24[a] | 92.86[b] | 95.23[a] | 95.23[a] | 0.31 |

| Parameter | Treatment[1] | | | | |
|---|---|---|---|---|---|
| | Optimal SD | Double SD | Double SD +150mg AA/L | Double SD + 300mgA A/L | SEM |
| Cost Implications[2] | | | | | |
| Cost of 2-wk old chicks | 304.50 | 304.50 | 304.50 | 304.50 | - |
| Total cost of 2-wkold chicks/treatment | 6384.00 | 12768.00 | 12768.00 | 12768.00 | - |
| Total cost of feed intake/treatment | 6101.51 | 11183.40 | 11753.40 | 12110.60 | - |
| Cost of feed intake/bird | 290.55 | 266.27 | 279.84 | 288.35 | - |
| Cost of AA supplementation / bird | 0.00 | 0.00 | 15.48 | 30.95 | - |
| Cost of labour/replicate pen | 1333.33 | 1333.33 | 1333.33 | 1333.33 | - |
| Cost of labour/bird | 190.48 | 95.24 | 95.24 | 95.24 | - |
| Cost of space/replicate pen | 500.00 | 500.00 | 500.00 | 500.00 | - |
| Cost of space/bird | 71.43 | 35.71 | 35.71 | 35.71 | - |
| Cost of .medication /bird | 18.50 | 18.50 | 18.50 | 18.50 | - |
| Total input cost /bird | 874.98 | 719.22 | 748.27 | 772.25 | - |
| Total Input Cost/ treatment | 18374.58 | 30207.24 | 31427.34 | 32434.5 | - |
| Sales | | | | | |
| No. of live birds/treatment | 20 | 39 | 40 | 40 | - |
| Av. live weight (kg) | 1.83 | 1.39 | 1.54 | 1.62 | - |
| Total sales/treatment[3] | 21228.00 | 31441.80 | 35728.0 | 37584.00 | - |
| Margin /kg live weight | 2853.42 | 1234.56 | 4300.66 | 5149.50 | - |
| % Change in Gross Margin | - | -56.73 | +50.72 | +80.47 | |

[abcd]Means in the same row followed by different superscript (s) are significantly different ($p<0.05$).
SEM: Standard Error Mean
[1] Optimal and double stocking density (SD)
[2] in ₦ 1USD = ₦ 195.25 at the time of the study.
[3] at selling price of ₦580.00 per kg live weight

The total feed intake of broilers on optimal stocking density was similar ($p>0.05$) in feed intake of birds on double optimal stocking density supplemented with 300mg AA/litre of water. The least feed intake was however recorded for broilers on double optimal stocking density without AA supplementation. A similar trend was recorded for average daily feed intake. The trend of feed intake reported in this study agrees with previous findings that Vitamin C supplementation improved feed consumption (Branton *et al*., 2004; Saha *et al*., 2000; Takahashi *et al*., 1991;Tanveer *et al*., 2005;Vathana *et al*., 2002). These researchers showed that supplementation of Vitamin C in drinking water or feed increased feed intake. However, the result was contrary to the reports of Blaha and Kreosna (1997), Jaffar and Blaha (1996) and Taweli and Kassab (1990) who reported that feed intake of broilers was not affected by the supplementation of Vitamin C. It appears that supplementation of Vitamin C in drinking water is useful in bringing improvement in feed intake at a higher stocking density.

Doubling the stocking density resulted in reduced weight gain ($p<0.05$). In this study a reduction in average daily weight gain of 27.35% was obtained as the stocking density was doubled. Ravindra *et al*. (2006) and Turkyilaz (2008) reported that the decreased individual body weights with increasing stocking density were not significantly influenced by stocking density. The findings of this study however confirm earlier studies (Beloor *et al.*, 2010; Galobart and Moran, 2005; Skrbic *et al*., 2009), that stocking density had a negative effect on body weight gain. The trend of lower rate of growth and lower values in other parameters of broiler performance raised under high stocking densities presents significant indicators of compromised welfare. Dozier *et al.*(2006)explained that reduction of final body mass could be related to decrease of food consumption because of inaccessibility to feeding space in conditions of high stocking density.

Iyasere *et al.* (2012) posited that chickens that were raised under higher stocking densities were posed to heat sensitivity because of increased litter temperature and limited ventilation around the birds. Apart from this direct effect, stocking density was known to indirectly influence the creation of micro climate in the facility and forming of other environment factors (Skrbic *et al*., 2009).

Working on **i**nfluence of stocking density on welfare indices of broilers, Abudabos *et al*. (2013) reported that broilers stocked at high stocking density experienced pronounced elevations of their body temperatures as well as head, body and shank surface temperatures over the controlled broilers stocked at low density. Broilers on high stocking density had an impediment for dissipation of their body heat and consequently increased their body heat content (Brake and Yahav, 2012; Yahav and Giloh, 2010).

Addition of AA in the drinking water at 150 and 300mg/litre however led to 12.66 and 19.85% improvement, respectively, in average daily weight gain of birds subjected to double stocking density. This result agrees with those of Jaffar and Blaha (1996), Lohakare *et al*. (2005), Pirompud *et al*. (2005), Rezaei *et al*. (2004), Tanveer *et al*. (2005) and Vathana *et al.* (2002). These earlier workers observed that addition of Vitamin C at 300-1500 ppm in drinking water or feed improved body weight. In the hot climate of Nigeria, Vitamin C supplementation also was reported to improve body weight of broiler chickens (Njoku, 1984, 1986). Bonsembinate *et al*. (2002) and Doan (2000) however concluded that addition of Vitamin C in drinking water or feed did not alter growth of broilers. Beneficial effects of Vitamin C supplementation would be most expressed under high ambient temperatures.

The efficiency of treatments measured in terms of feed: gain ratio showed that the birds on optimum stocking density performed best out of the lot, while birds stocked at double stocking density was the least efficient requiring 3.02 kg of feed to achieve a kg of weight in flesh representing an additional 26.36% more feed to produce a kg of flesh compared to the control treatment. Many reasons have been adduced for weight reduction arising from higher stocking densities, Puron *et al*. (1995) explained that higher stocking density resulted in decreased feed intake since access to feeders would be limited as well as the increased competition among birds to get to the feeder. The addition of AA in drinking water at 150 and 300mg/litre however improved this index of efficiency by 6.95 and 9.60% in birds stocked at double stocking density. The results also demonstrated a better FCR with increasing AA supplementation. This finding is in agreement with the findings of Blaha and Kreosna (1997), Mckee and Harrison (1995), Mirsnamsollahi *et al.* (2003), Nagra *et al.* (2005), Tanveer *et al.* (2005), Soleiman and Zulkifli (2010) and Vathana *et al.* (2002) who also detected an improvement in FCR of broilers as a result of Vitamin C supplementation during stress. Vitamin C is associated with the conversion of body proteins and fat into energy for production and survival through increased corticosterone secretion (Bain, 1996; Marshall and Hughes, 1980). Ascorbic acid (Vitamin C) enhances secretion of corticosterone and thus could be a useful stress management strategy.

From the performance indices measured in this study, increasing the stocking density appeared to reduce some of the indices. However a deeper observation showed that more birds were actually produced when double optimal stocking density was applied. Doubling the number of birds stocked per unit space resulted in a reduction in input cost per bird. This is expected as economy of scale comes into play. At double stocking density gross margin changed by -56.73, +50.73 and +80.47% for birds produced on double stocking density without AA, double stocking density and150 mg AA/litre of drinking water and double stocking density and300 mg AA/litre of drinking water respectively when the gross margin of birds produced on optimal stocking density is taken as the baseline.

Table 2. Effects of ascorbic acid supplementation on ether extract and crude protein retention of broilers raised on optimal and double stocking densities

| Parameter | Treatment[1] | | | | |
|---|---|---|---|---|---|
| | Optimal SD | Double SD | Double SD + 150 mg AA/l | Double SD + 300 mgAA/l | SEM |
| Ether extract | 68.65 [b] | 61.88 [c] | 68.40 [b] | 73.61[a] | 2.21 |
| Crude protein | 71.89 [a] | 63.34 [b] | 70.92 [a] | 71.37 [a] | 1.74 |

[abc]Means in the same row followed by different superscript (s) are significantly different (p<0.05).
SEM: Standard Error Mean
[1] Optimal and double stocking density (SD)

Subjecting birds to double stocking density resulted in a 9.86% reduction in ether extract digestibility, however the addition of 150mg AA/litre of drinking water at double stocking density gave ether extract digestibility that was not different (p>0.05) from that of birds on optimal stocking density (Table 2). Allowing birds access to 300mg AA/litre of drinking water however resulted in a 7.23% improvement (p<0.05) in ether extract digestibility over that of birds on optimal stocking density. This observation that vitamin C reduced fat deposition might explain the reported inverse relationship between adiposity and vitamin C status . With respect to protein retention birds subject to higher stocking density with AA supplementation had similar digestibility values (p>0.05) as the birds stocked at optimal density. Birds at double optimal stocking density without AA supplementation elicited lower (p<0.05) protein digestibility. Sahin and Kucuk (2001) found increased digestibility of nutrients after dietary vitamin C supplementation in Japanese quails reared under heat stress (34°C), while Sahin *et al.* (2002) reported similar observation for laying hens.

A similar observation of reduction in nutrient digestibility with increasing stocking density was made by Zhang *et al.*(2013) in pigs. They explained that significant reduction in dry matter and nitrogen digestibility with increasing stocking density was a result of the pigs in larger groups experiencing higher levels of social stress, which negatively affected nutrient digestibility.

The digestibility of nutrients measured was significantly higher in AA supplemented group compared to unsupplemented group. Broilers of high stocking density rate created an impediment for dissipation of their body heat and consequently increased their body heat content (Abudabos *et al*.,2013). According to Tatli-Seven (2008), bioavailability of nutrients was affected by heat stress. Vitamin C alleviated the negative effects of heat stress on apparent nutrient digestibility. Tatli-Seven (2008) explained further that vitamin C improved iron assimilation by reduction of $Fe^{3+}$ to $Fe^{2+}$, which was better assimilated by the intestine, and thereby vitamin C improved resistance to infections. Locally, oxidative lesions leading to conformational modification of proteins could induce pancreatic enzyme inhibition and/or dietary protein resistance to digestion. Consequently, the presence of antioxidants (vitamin E/or C) could partially interfere with oxidative protein denaturation and would improve digestibility of nutrients.

Table 3. Retail cuts and relative organ weights of broiler chickens supplemented with ascorbic acid in drinking water at optimal and double stocking densities

| Parameter | Treatments | | | | SEM |
|---|---|---|---|---|---|
| | Optimal SD | Double SD | Double SD +150 mgAA/l | Double SD + 300 mgAA/l | |
| Live weight (g) | 1825.06 | 1355.50 | 1540.10 | 1620.15 | 21.05 |
| Dressed weight (g) | 1379.02[a] | 915.65[d] | 1085.50[c] | 1205.35[b] | 18.88 |
| Dressing (%) | 75.56[a] | 67.55[c] | 70.48[bc] | 74.39[a] | 1.35 |
| Prime cuts(% live weight) | | | | | |
| Breast | 38.06[a] | 27.65[c] | 31.15[bc] | 33.14[b] | 0.89 |
| Drumstick | 14.44[a] | 13.79[b] | 14.34[a] | 14.41[a] | 0.34 |
| Thigh | 15.68 | 14.45 | 14.84 | 15.29 | 0.78 |
| Wing | 14.02[a] | 12.82[c] | 13.01[b] | 13.59[ab] | 0.72 |
| Relative organ weights(% live weigh | | | | | |
| Liver | 2.16[b] | 3.07[a] | 2.87[ab] | 2.72[ab] | 0.14 |
| Gizzard | 2.21 | 2.13 | 2.18 | 1.99 | 0.06 |
| Heart | 0.44 | 0.50 | 0.53 | 0.49 | 0.21 |

[abc]Means in the same row followed by different superscript (s) are significantly different (P<0.05).
SEM: Standard Error Mean
[1] Optimal and double stocking density (SD)

Dressed weight, dressing percentage and relative breast cut weights were significantly depressed as the stocking density was doubled from Treatment 1 to 2 (Table 3). Earlier report of Dozier *et al.* (2005, 2006) showed that carcass weight decreased as stocking density increased. However, Tong *et al.* (2012) and Nogueira *et al.* (2013) observed no such development. The positive effects of ascorbic acid supplementation in the present study was seen in the improved weights of these retail cuts in birds stocked at double the optimal stocking density as evidenced in the improved dressing percentage, breast and wing cuts of broilers. Some earlier reports (Mbajiorgu *et al.,* 2007; Ali *et al*., 2010), showed that supplementation of ascorbic acid through feed or water improved dressing yield and breast cut percentage. Stocking density influenced ($p<0.05$) percentage of liver but had no effect on gizzard and heart. Buijis *et al.* (2009) reported that stocking density had no influence on internal organ weights of broilers while Puvadolpirod and Thaxton (2000) reported that liver weight increased in periods of stress.

## Conclusion

The results of the study suggest that supplementation of AA at high stocking density was beneficial to broilers since the indices of performance were shown to have improved. Dressing percentages and cut up parts were also positively affected by AA supplementation at double the optimal stocking density. While the summary of performance of individual birds appears to favour the optimal stocking density, the total production of live or dressed meat per $m^2$ of floor space decreased which resulted in higher revenue. The beneficial effects of AA supplementation lead to improvement in gross margin and at the same time show to welfare groups the extent to which efforts are being made to ameliorate the negative effect that may affect the birds' comfort.

**References**

Abudabos, A.M., Samara, E.M., Hussein, E.O.S., Al-Ghadi, M.Q and Al-Atiyat, R.M., 2013. Impacts of stocking density on thee performance and welfare of broiler chickens. *Ital. J. Anim. Sci.* 12(11):66-71.

Ali, M.T., Howlider, M.A.R., Azad, A.K. and Rahman, M.S., 2010. Vitamin C and electrolyte supplementation to support growth and meat yield of broilers in a hot humid environment. *J. Bangladesh Agric. Univ.* 8(1): 57–60.

AOAC. 1995. Official Methods of Analysis. 16th edition. Association of Official Analytical Chemists. Washington D.C.

Atteh, J.O.,2004. Theory and Practice of Poultry Production (ISBN 978-187-115-6). Adlek Printers, Ilorin,Nigeria.

Ayoola, A.A., Adeyemi,O.A., Eruvbetine, E., Egbeyale, L.T., Sogunle, O.M. and Ekunseitan, D.A., 2014. Response of male quails (*Cortunixcoturnix japonica*) to different stocking densities and wing clipping. Nigerian J. Anim. Sci. 16(1):39-50.

Bains, B.S. 1996. The role of Vitamin C in stress management. *World Poultry* 12(4): 38-41.

Beloor, J., Kang, H.K., Kim, Y.J., Subramani, V.K., Jang, I.S., Sohn, S.H. and Moon, Y.S., 2010. The effect of stocking density on stress related genes and telomeric length in broiler chickens. *Asian Aust. J. Anim. Sci*. 23:437-443.

Bláha, J. and Kroesna, K. 1997. Effect of vitamin and electrolyte supplements on broilers' performance, slaughter value and chemical composition of meat during the heat stress. *Universitäs Agriculturäe Praga J.*, 30: 103-113.

Bonsembiante, M., Chiericato, G.M. and Rizzi, C. 2002. Influence of different sodium bicarbonate dietary contents on the metabolicprofile, productive performance and meat quality of broilers. *Rivista-di-Avicoltura*, 71(1): 33-42.

Brake, J. and Yahav, S. 2012. Responses of poultry to environmental challenges. In: R.J. Collier and J.L. Collier (eds.) Environmental Physiology of Livestock. John Wiley & Sons Publ., West Sussex, UK, pp 309-335.

Branton, S.L., Reece, F.N. and Deaton, J.W. 1985. Use of ammonium chloride and sodium bicarbonate in acute heat exposure of broilers. *Poultry Sci.* 65: 1659-1663.

Buijis, S., Keeling, L., Rettenbacher, S., van Poucke, E. and Tuyttens, F.A.M., 2009. Stocking density effects on broiler welfare: Identifying sensitive ranges for different indicators. *Poultry Sci.* 88: 1536-1543.

Butcher, G.D. and Miles, R. 2003. Heat stress management in broilers. IFAS Extension.University of Florida. USA VM65: 1-3.

Cullison, A.E. 1982. Feeds and Feeding. 3rd edition. Reston Publishing Co, Virginia.

Curca, D., Andronie, V., Andronie, I.C. and Pop, A. 2004. The influence of feed supplementation with ascorbic acid and sodium ascorbate on broilers under thermal stress. Book of Abstract XXII World Poultry Congress of WPSA, Istanbul, Turkey. Pp 290.

DEFRA, 2005. Heat stress in poultry: Solving the problem. Department of Environment, Food and Rural Affairs. Defra publication. pp 1-10.

Doan, B.H. 2000. Effect of different levels of dietary calcium and supplemental Vitamin C on growth, survivability, leg abnormalities, total ash in the tibia, serum calcium and phosphorous in 0-4 week-old chicks under tropical conditions, Livestock Research for Rural Development, CIPAV Publication, Colombia,12(1).

Dozier, W.A., Thaxton, J.P., Branton, S.L., Morgan, G.W., Miles, D.M., Roush, W.B., Lott, B.D. andVizzier-Thaxton, Y. 2005. Stocking density on growth performance and processing yields of heavy broilers. *Poultry Sci.*84:1332-1338.

Dozier, W.A., Thaxton, J.P., Purswell, J.L., Olanrewaju, H.A., Branton, S.L. and Roush,W.B. 2006. Stocking density effects on male broilers grown to 1.8 kilograms of body mass. *Poultry Sci.* 85:344-351.

FUNAAB. 2013. Policy on research of the Federal University of Agriculture. Abeokuta. Nigeria. http://www.unaab.edu.ng (04/08/2013).

Galobart, J. and Moran, E.T. Jr. 2005. Influence of stocking density and feed pellet quality on heat stressed broilers from 6 to 8 weeks of age. *Int. J. Poult. Sci*. 4: 55-59.

Gonzalez-Esquerra, R. and Leeson, S. 2006. Physiological and metabolic responses of broiler to heat: Implications for protein and amino acid nutrition.*World Poult. Sci. J.* 62(2):282-295.

Iyasere, O.S., Daramola, J.O., Bemji, M.N., Adeleye, O.O. , Sobayo, R.A., Iyaser, E. and Onagbesan, O.M. 2012. Effects of stocking density and air velocity on behaviour and performance of Anak broiler chickens in South-Western Nigeria . *Int. J. Appl. Anim. Sci.* 1(2):52-56.

Jaffar, G.H. and Bláha, J. 1996. Effect of ascorbic acid supplementation in drinking water on growth rate, feed consumption and feed efficiency of broiler chickens maintained under acute heat stress conditions. *Universitas Agriculturae Praga Press*, 41(1): 485-490.

Lohakare, J.D., Ryu, M.H., HahnT.W., Lee, J. K. and. Chae, B. J. 1995. Effects of supplemental ascorbic acid on the performance and immunity of commercial broilers. *J. Appl. Poult. Res.* 14:10–19.

Marshall, P.T. and Hughes, G.M. 1980. Physiology of Mammals and Other Vertebrates, Chapter 11: The Endocrine System, Pp 277-300, Cambridge University Press, USA.

Mbajiorgu, C.A., Ngambi, J.W. and Norris, D. 2007. Effect of time of initiation of feeding after hatching and influence of dietary ascorbic acid supplementation on productivity, mortality and carcass characteristics of Ross 308 broiler chickens in South Africa. *Int. J. Poult. Sci*, 6(8): 583-591.

McKee, J.S. and Harrison, P.C. 1995. Effect of supplemental ascorbic acid on the performance of broiler chickens exposed to multiple concurrent stressors, *Poultry Sci.* 74: 1772-1785.

Mirshamsollahi, A., Moghaddam, G., Adl, K.N. and Janmohamadi, H. 2003. The effects of electrolyte supplement in drinking water on broiler performance under heat stress condition. *Agric. Sci. Tabriz*, 12(4): 36-45.

Nagra, S.S., Sodhi, S.,Yashpal Singh and Diwedi, P. N. 2005. Effect of Vitamin C supplementation on performance of broiler chicks during heat stress. *Proceeding of XXIII IPSACO N*, 2nd - 4th Feb: 181.

Njoku, P.C. 1984. The effect of ascorbic acid supplementation on broiler performance in a tropical environment. *Poultry Sci.* 63: 156 (Abstract).

Nogueira, W.C.L, Velasquez, P.A.T., Furlan, R.L. and Macari, M. 2013. Effect of dietary energy and stocking density on the performance and sensible heat loss of broilers reared under tropical winter conditions. *Braz. J. Poult. Sci.* 15(1): 53-58.

Pirompud, P., Attamangkune, S., Bunchasak, C. and Promboon, A. 2005. Effect of acid base balance in feed to broilers reared under tropical conditions on performance and carcass traits. *Proceedings of 43rd Kasetsart University Annual Conference*, Thailand, 1-4 February.Pg 254-261.

Puron, D., Santamaria, R., Segura, J.C. and Alamilla, J.L. 1995. Broiler performance at different stocking densities. *J. Appl. Poultry Res.* 4:55-60.

Puvadolpirod, S. and Thaxton, J.P. 2000. Model of physiological stress in chickens: 1. Response parameters. *Poultry Sci.* 79:363-369.

Ravindran, V., Thomas, D.V., Thomas, D.G. and Morel, P.C.H. 2006. Performance and welfare of broilers as affected by stocking density and zinc bacitracin supplementation. *J. Anim. Sci.* 77: 110-116.

Rezaei, M., Moghaddam, H.N., Reza, J.P. and Classen, H.L. 2004. Effect of dietary electrolyte balance on the performance and body composition of broiler chicks. *Agric. Sci. Tech*. 18(2): 69-78.

Saha, S., Chowshury, S.D. and Ahmed, K.D. 2009. Productive performance and cost-effectiveness of using additional vitamins in commercial broiler diets. *Proceeding of 6th International Poultry Show and Seminar*, World Poultry Science Association, Bangladesh.

Sahin, K. and Kucuk,O. 2001. Effects of vitamin C and vitamin E on performance, digestion of nutrients and carcass characteristics of Japanese quails reared under chronic heat stress ($34^0$ C). *J. Anim. Physiol. Anim. Nutr.* (Berlin) 85(11-12):335-41.

Sahin, K., Sahin, N. and Kucuk, O. 2002. Effects of dietary chromium and ascorbic acid supplementation on digestion of nutrients, serum antioxidant status, and mineral concentrations in laying hens reared at a low ambient temperature. *Biological Trace Element Research*, 87(1-3):113-124.

SAS Institute. 2000. User’s Guide Statistics. Cary; NC, USA.

Škrbic, Z., Pavlovski, Z. and Lukic, M. 2009. Stocking density–factor of production performance, quality and broiler welfare. *Biotechnology in Animal Husbandry*. 25(5-6):359-372.

Skrbic, Z., Pavlovski Z., Lukic, M., Peric, L. and Milosevic, N. 2009. The effect of stocking density on certain broiler welfare parameters. *Biotechnology in Animal Husbandry*. 25: 11-21.

Soleiman, A.F. and Zulkifli, I. 2010. Effect of high ambient temperature on blood parameters in red jungle fowl, village fowl and broiler chickens. *J. Anim. Vet. Adv*. 9:1201-1207.

Takahashi, K., Akiba, Y. and Horiguchi, M. 1991. Effects of supplemental ascorbic acid on performance, organ weight and plasma cholesterol concentration in broiler treated with propylthiouracil. *Brit. Poult. Sci.* 32 (3): 545-554.

Tanveer, A., Sarwar, M., Mahr, U.N., and Ahsan, U.H. 2005. Influence of varying sources of dietary electrolytes on the performance on broiler reared in a high temperature environment. *Anim. Feed Sci. Tech*. 120(3/4): 277-298.

Tatli-Seven, P. 2008. The effects of dietary Turkish propolis and Vitamin C on performance, digestibility, egg production and egg quality in laying hens under different environmental temperatures. *Asian-Aust. J. Anim. Sci*. 21(8): 1164-1170.

TaweliA.L and Kassab, A. 1990. Effect of dietary vitamin C on ascites in broiler chicks. *Int. J. Vietnamese Nutritional Research*, 60 (4): 366-371.

Teeter, R.G. 1994. Optimizing production of heat stressed broilers. *Poult. Dig.,* 5: 10-16.

Teeter, R.G., Smith, M.O., Owens, F.N., Arp, S.C., Sangiah, S. and Breazile, J.E. 1985. Chronic heat stress and relative humidity: Occurrence and treatment in broiler chicks. *Poult. Sci.* 64:1060-1064.

Thaxton, J.P., Dozier, W.A., Branton, S.L., Morgan, G.W., Miles, D.W., Roush, W.B., Lott, B.D. and Vizzier-Thaxton, Y. 2006. Stocking density and physiological adaptive responses of broilers. *Poultry Sci*. 85:819-824.

Tong, H.B., Lu, J., Zou, J.M., Wang, Q. and Shi, S.R. 2012 Effects of stocking density on growth performance, carcass yield, and immune status of a local chicken breed. *Poult. Sci*. 91:667-673.

Turkyılmaz, M.K. 2008. The effect of stocking density on stress reaction in broiler chickens during summer. *Turk. J. Vet. Anim. Sci*., 32: 31-36.

Vathana, S., Kang, K, Loan, C.P, Thinggaard, G, Kabasa, J.D and Meulen, U. 2002. Effect of Vitamin C supplementation on performance of broiler chickens in Cambodia. In Proceedings Conference on International Agricultural Research for Development Deutscher Tropentag Witzenhausen, Germany. October 9-11, 2002.

Yahav, S. and Giloh, M. 2010. The use of body temperature as an indicator for the chicken physiological status and as a measure to control poultry houses in order to achieve maximal genetic potential. Proc. 45th Nat. Annual Conf. Israeli Poultry Science Association, Jerusalem, Israel.

Zhang, Z.F., Li, J., Park, J.C. and Kim, I.H. 2013. Effect of vitamin levels and different stocking densities on performance, nutrient digestibility, and blood characteristics of growing pigs. *Asian-Australasian J. Anim. Sci.* 26(2): 241–246.