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## **Modulatory Effects of Dietary Betaine and Ascorbic Acid on Some Production Parameters of Sexually Mature Japanese quails during the Dry Season in Derived Savannah Ecological zone of Nigeria**

*Ifeanyichukwu Chukwuemeka Egbuniwe*  
*Chukwuka Nwocha Uchendu*  
*Ikechukwu Reginald Obidike*

### **ABSTRACT**

*This study examines the modulatory effects of dietary betaine and/or ascorbic acid modulate some production parameters in sexually mature Japanese quails (*Coturnix coturnix japonica*) during the dry season. A total of 388 quails, 2 week-old, were randomly allotted into 4 groups and 3 replicate per group (32 per replicate). Group I: given basal diet; Group II, basal diet + 200 mg/kg diet ascorbic acid (AA); Group III – basal diet + 2 g betaine/kg diet, and Group IV – basal diet + 200 mg AA + 2 g betaine/kg. Dry-bulb temperature (DBT) was measured thrice daily with hygrometer; for relative humidity (RH) – hygrometric tables; temperature–humidity index (THI) was derived. Feed conversion efficiency to egg mass and per dozen eggs (FCE, FEPP) were derived. The DBT, RH and THI were predominantly outside thermoneutral zone. Betaine increased ( $P < 0.05$ ) AVDFI, AVDWC, live weight; but decrease FCE and FEPP. Dietary AA increased ( $P < 0.05$ ) AVDFI; AVDWC, live weight, FCE and FEPP, but decreased AVDFI. Betaine + AA increased ( $P < 0.05$ ) AVDWC; live weight but lowered FCE and FEPP. Betaine and/or AA enhances AVDFI, AVDWC, live weight and feed conversion to egg mass and per dozen production of sexually mature Japanese quails during dry season.*

**Keywords:** Ascorbic acid, betaine, dry season, quails

### **INTRODUCTION**

Heat stress results in decreases in economic parameters and may consequently contribute to financial losses recorded by farmers who rear quails for meat and eggs (Kalafova *et al.*, 2017). Mora *et al.* (2017) reported that heat stress results from a combination of high ambient temperature (AT) and high relative humidity

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*\*Ifeanyichukwu Chukwuemeka Egbuniwe (Ph.D), is a Lecturer in the Department of Veterinary Physiology and Biochemistry, University of Benin, Benin City, Edo State, Nigeria. Chukwuka Nwocha Uchendu and Ikechukwu Reginald Obidike are Lecturers in the Department of Veterinary Physiology and Biochemistry, University of Nigeria, Nsukka, Enugu State, Nigeria. Email: ifeanyi@egbuniwe@gmail.com.*



(RH). Temperature–humidity index (THI) is a reliable indicator of thermal stress for the combined effects of AT and RH in Japanese quails (El–Tarabany, 2016). Betaine supplementation is one agents used in reliving the adverse effects of heat stress in poultry birds (Park and Kim, 2017). Ascorbic acid has been utilized for its antioxidant properties to mitigate the negative effects of heat stress in laying hens (Attia *et al.*, 2016). Egg production indicators determine productivity and profitability of poultry (and quail) production (Rasul *et al.*, 2019). Feed consumptions accounts for 70% cost of production in the poultry industry. Poultry birds require plenty of cool and clean water to ensure proper physiological activities, maintain production and thermoregulation, especially when they are reared under thermally stressful environment conditions (Ambazamkandi *et al.*, 2015). The right live weight must be attained for young sexually mature quails and poultry birds to commence laying (Al-Tikriti, 2018). The aim of this study is to examine the modulatory effects of dietary betaine and/or ascorbic acid on some production parameters (like intakes of feed and water, live weight, and efficiency of feed conversion to egg mass and a dozen eggs produced) in sexually mature Japanese quails during the dry season.

## MATERIALS AND METHOD

**Experimental Site:** The experiment was carried out at the animal house of Department of Parasitology and Entomology, Faculty of Medicine, University of Nigeria, Nsukka. The experimental site is located in the Derived Savannah Ecological zone of Nigeria (Uguru, Baiyeri and Aba, 2011) and has an annual rainfall range of 986 – 2098 mm (Momoh, Gambo and Dim, 2010). This study was conducted from January – March 2019.

**Animal and Management Conditions:** A total of 388 female Japanese quails (*Coturnix coturnix japonica*) weighing between 26 – 28g were purchased commercially at 2 week-old from National Veterinary Research Institute (NVRI), Vom, Nigeria, and used for the study. Quails were acclimatized for 7 days and subsequently, assigned to 4 groups by complete randomized design. Each group consisting of 97 quails, were subdivided into 3 replicates (33 birds per replicate). Quails in each replicate were housed in separate cages measuring 0.91 m × 0.76 m × 0.91 m. These cages were confined within a windowed poultry pen. The birds were subjected to at least 17 hours lighting, required for optimum growth, development and production for Japanese quails (Elkomy, Taha, Basha, Abo-Samaha and Sharaf, 2019). Quails were reared under the natural high ambient temperature, high relative humidity prevailing during the dry season in the zone.



Experimental groups are: Group I, quails given basal (control) diet; Group II, basal diet + 200 mg/kg diet ascorbic acid (Kempex Holland BV, Volkel, The Netherlands); Group III – basal diet + 2 g betaine/kg diet (Sigma-Aldrich, St. Louis, Missouri, USA), and Group IV – basal diet + 200 mg ascorbic acid + 2 g betaine/kg of diet. The birds were fed commercial starter (22 % CP; 3100 kcal/kg of metabolizable energy/kg of feed) at 2 – 4 week-old; grower mash (15.50 % CP; 2550 kcal/kg ME) at 4 – 7 week-old and layer mash (16.80 %; 2680 kcal/kg ME) at 7 – 10 week-old. Dietary treatment commenced at age of 3 week-old, lasted for 56 days, at 10 week-old. Quails were given access to water and feed *ad-libitum* throughout the study period. The ingredients and chemical composition of the basal diets supplied the quails at different growth phase are shown in Table 1. Experimental procedures followed in the present study was in accordance to the guideline outlined by the Guide for the Care and Use of Laboratory Animals (Committee on Care and Use of Laboratory Animals, 1996).

**Environmental conditions:** Environmental conditions prevailing during the study period from January – March 2019 were recorded daily at 08:00 h, 13:00 h and 17:00 h. Ambient temperature (AT) was measured using Mason's type dry- and wet-bulb hygrometer (Zeal, London, England). Relative humidity (RH) was obtained using hygrometric tables for computation of relative humidity (Zeal, London, England). Temperature-humidity index (THI), which is an index of thermal comfort, was derived using the formula hitherto described by Zuluovich and DeShazer (1990), but modified by El-Tarabany (2016). Temperature-humidity index (THI) =  $0.6T_{db} + 0.4T_{wb}$ ; where  $T_{db}$  = dry-bulb temperature, (°C), and  $T_{wb}$  = wet-bulb temperature (°C).

**Production parameters:** Production parameters were evaluated at the age of 3 – 10 week-old, as average daily feed intake (AVDFI), average daily water consumption (AVDWC) and weekly live weight. Average daily feed intake and water consumed were measured by net feed consumed by the flock divided by the number of quails in each group (Hassan H., Mohamed, Youssef and Hassan, 2010). Weekly live weight of 15 quails per group were individually measured using digital scale to the nearest 0.1 g (Asia Technoweigh India, Haryana, India). Eggs were collected from each replicate in every group once daily from onset of lay at 7 week-old, till 10 week-old. The study focused on growth and early mature phases of the birds. All eggs laid in each replicate were weighed using digital milligram scale (WAOAW, Lexington, Kentucky), to the nearest 0.001g. Total number of eggs laid in each replicate was recorded. Egg mass was obtained as average egg weight multiplied by the number of egg laid daily, in each replicate. Feed-to-egg mass ratio was determined as the ratio between the daily feed



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consumption and the daily egg mass in each group, during the entire study period, as described by Abouelezz, Sayed and Abdelnabi (2019). Feed efficiency per dozen eggs produced was obtained as feed-to-egg (per dozen eggs) = (multiply kilogram of feed consumed by 12) and divided by total eggs produced (Dahiya, Berwal, Sihag and Patil, 2016).

**Statistical Analysis:** Data obtained were expressed as mean  $\pm$  standard error of mean (mean  $\pm$  SEM). Values were subjected to analysis of variance (ANOVA), and followed by Tukey's *post hoc* test. Values of  $P < 0.05$  were considered significant (Snedecor and Cochran, 1994). Graphpad 6.0 for windows (San Diego, California, USA) software was used for the analysis. Complete Random experimental design was used for the study.

## RESULTS AND DISCUSSION

**Environmental conditions:** Environmental conditions recorded during the study period is shown in Figure 1 a and b. The result of the present study revealed that mean values of DBT, WBT, RH and THI obtained were  $31.4 \pm 0.3$  pC,  $24.2 \pm 0.3$  pC,  $73.6 \pm 1.0\%$  and  $83.4 \pm 0.4$ , respectively. The minimum and maximum DBT values were  $25.0 - 37.0$  pC; RH,  $48.0 - 92.0\%$  and THI,  $69.8 - 91.0$ , respectively. These findings of the present study demonstrate that DBT and THI values observed during the study period varied widely and were positively skewed. Similarly, RH values were high and varied over a wide range. The results indicated that fluctuations of DBT, RH and THI values were wide and predominantly outside the thermo-neutral zone for quails ( $23.8 \pm 0.7^\circ\text{C}$ ,  $58.5 \pm 5.7\%$  and  $76 - 80$ , respectively, for DBT, RH and THI – El-Tarabany, 2016). Furthermore, wide fluctuations in environmental conditions such as DBT are known to result in heat stress in poultry birds (Sinkalu *et al.*, 2015).

The findings of this study agrees with those of Mehaisen *et al.* (2017), who showed that DBT values above  $30^\circ\text{C}$  are considered thermally stressful to Japanese quails. The finding of the present study agrees with those of Minka and Ayo (2013 a,b), who showed that environmental condition (such as ambient temperature and relative humidity) prevailing during the hot-dry season in the Northern Guinea Savannah zone – recording DBT and RH values of  $32.5 \pm 1.4^\circ\text{C}$ ,  $66.9 \pm 5.7\%$  – were stressful to quails. Thermally stressful conditions are detrimental to health and welfare of quails (El-Tarabany, 2016). The environmentally stressful conditions prevailing during the dry season could increase thermal load in the quails, impair their capacity to dissipate excess heat to the environment by thermoregulatory mechanisms, and result in excessive



generation of free radicals and consequently, oxidative stress (Sahin, N., Orhan, Tuzcu, Sahin and Kucuk, 2008).

**Production parameters:** Average daily feed intake (AVDFI) of quails on weekly basis is shown in Figure 2. The results revealed that AVDFI at 7 week-old in birds fed diets with either AA or betaine + AA was lower ( $P < 0.05$ ) when compared to either control or betaine diets fed to quails. However, quails fed with either AA or betaine consumed more feeds ( $P < 0.05$ ) than the control group at 5 and 8 week – old. Average daily feed intake did not differ significantly ( $P > 0.05$ ) at 4, 6, 9 and 10 week – old. Quails fed diet with either betaine, AA or betaine + AA drank more water ( $P < 0.05$ ) than the control birds throughout the growing periods of (4, 5, 6, 7, 8, 9 and 10 week–old) (Figure 3). Figure 4 shows variations in live weight of quails during the study period on weekly basis. Live weight of quails fed diets incorporated with either betaine, AA or betaine + AA were heavier at 8, 9 and 10 week – old when compared with the control group.

However, live weight did not differ ( $P > 0.05$ ) amongst the experimental groups during growth phase (3, 4, 5 and 6 week-old). Feed–to–egg ratio in each group during the entire study period are reported in Figures 5 and 6. The lowest feed-to-egg mass ratio of 4.85 % was obtained in quails given diets with betaine. This was closely followed by 5.69 % recorded in the group fed diets containing betaine + AA. The feed –to-egg mass ratio in group fed diets incorporated with either betaine or betaine + AA were 1.04 % and 0.2 % lower than value of 5.89 % obtained in the control birds. The highest feed-to-egg mass ratio (6.39 %) was recorded in quails fed diets with AA, and this was 0.5 % higher than values recorded in the control group (5.89 %). Similarly, quails given diet containing betaine recorded the lowest feed efficiency per dozen eggs produced with 0.57 %, closely followed by 0.66 % in betaine + AA administered quails. The values obtained in quails fed diets supplemented with either betaine or betaine + AA are 0.1 % and 0.01 % lower than those in the control (0.67 %). The birds fed diets with AA recorded feed efficiency per dozen eggs of 0.76 %, which is 0.09 % more than control group (0.67 %).

The result revealed that the production parameters such as AVDFI, AVDWC and live weight increased in quails fed betaine and/or AA diets at different ages of their growth. The finding suggests that betaine and/or AA enhanced average daily feed and water intake, as well as live weight of quails reared during the dry season. The results agree with those of Ratriyanto and Prastowo (2019), who showed that betaine and floor space influenced productive and reproductive performances in Japanese quails, by improving nutrient utilization in quails reared under the tropical environment. Although, heat stress



conditions (such as high ambient temperature and high relative humidity) are known to result in decline of feed intake and live weight in poultry birds (Rasul *et al.*, 2019), the results suggest that dietary betaine and/or AA improves AVDFI, AVDWC and live weight. Enhanced AVDFI will improve nutrient utilization for physiological and productive activities in quails during the dry season. The result revealed that betaine and/or AA significantly ( $P < 0.05$ ) increased water consumption in quails during the dry season. The finding suggests that there was increased demand for water by quails fed betaine and/or AA diets. The obtained result disagrees with the report of Egbuniwe, Ayo, Kawu and Mohammed (2018), who showed that betaine could lower the quantity of water consumed by broiler chickens during hot–dry season. The discrepancy in the findings of the present study with those of Egbuniwe *et al.* (2018) may be due to species differences in levels of activity. Levels of activity could increase water consumption as demonstrated by Westerterp (2017). However, species differences on activity were not investigated in the study.

The results of study reveal that dietary supplementation with betaine and/or AA enhances conversion of feed to egg production, evidenced by lower values of FCR and FEPD in quails reared during the dry season. It implies that quails administered betaine, either alone or in combination with AA, require less quantity of feed to produce heavier and one dozen eggs. The findings of the study also agree with those of Ratriyanto and Prastowo (2019) who demonstrated that betaine and floor space on improved productive and reproductive performance in Japanese quails reared under tropical conditions. The osmolyte function of betaine ensures osmotic balance in intestinal epithelium (Ratriyanto, Indreswari and Nuhriawangsa, 2017), thus, enhancing nutrient utilization in poultry birds, particularly Japanese quails (Ratriyanto and Prastowo, 2019).

Furthermore, betaine functions as a methyl donor and plays a role in protein synthesis through DNA epigenesis (Ratriyanto, Indreswari, Nuhriawangsa and Haryanti, 2015). This implies that the combined functions of betaine as an osmolyte and methyl donor may be responsible for the improved laying analysis in Japanese quails during the dry season, especially at early laying phase of egg production. The findings is of particular importance because poultry birds (including Japanese quails) lay fewer and smaller sized eggs at the commencement of egg production (Chimezie, Fayeye, Toye and Ayorinde, 2017), especially under thermally stressful environmental conditions. The results suggest that diets containing betaine and fed to sexually mature Japanese quails may increase the production of more acceptable eggs in terms of heavier and more number of eggs produced and profitability to quail farmers.



## CONCLUSION

The study was conducted to examine the modulatory effects of dietary betaine and/or ascorbic acid on some production parameters (like intakes of feed and water, live weight and efficiency of feed conversion to egg mass and a dozen eggs produced) in sexually mature Japanese quails during the dry season. A total of 388 female Japanese quails (*Coturnix coturnix japonica*) weighing between 26 – 28g were purchased commercially at 2 week-old from National Veterinary Research Institute (NVRI), Vom, Nigeria, and used for the study. Environmental conditions prevailing during the study period from January – March 2019 were recorded daily. Production parameters were evaluated at the age of 3 – 10 week-old, as average daily feed intake (AVDFI), average daily water consumption (AVDWC) and weekly live weight. Dietary betaine and/or AA modulate average daily consumptions of feed and water, live weight, and feed conversion to egg mass and a dozen egg production of Japanese quails reared during the dry season.

**Table 1:** Composition and proximate analysis of diets fed quails

Ingredients (%)	Starter mash	Grower mash	Layer mash
Maize	30.00	0.00	20.00
Sweet potato meal	30.00	60.00	40.00
Blood meal	5.00	5.00	5.00
Groundnut cake	29.70	29.70	29.70
Wheat offal	1.00	1.00	1.00
Bone meal	3.25	3.25	3.25
dl-Methionine	0.25	0.25	0.25
Lysine	0.25	0.25	0.25
Vitamin premix	0.30	0.30	0.30
Salt	0.25	0.25	0.25

### Proximate analysis

Crude protein (%)	22.00	15.50	16.80
Metabolizable energy (Kcal/kg)	3000.00	2550.00	2680.00
Fat (%)	5.10	3.60	3.60
Crude fibre (%)	4.30	4.60	4.20
Calcium (%)	1.20	1.10	4.20
Available phosphorus (%)	0.45	0.40	0.50
Methionine (%)	0.56	0.37	0.45
Lysine (%)	1.30	0.75	0.85

Vitamin premix supplied per kg diet: retinol: 10,000IU, cholecalciferol: 2,000IU, alpha-tocopherol: 51 mg, phyloquinone: 2.34 mg, riboflavin: 5.5 mg, calcium pantothenate: 10 mg, niacin: 25 mg, chlorine chloride: 250 mg, folic acid: 1 mg, manganese: 56 mg, zinc: 50 mg, copper: 10 mg, iron: 20 mg, cobalt: 1.25 mg.



Fig. 1a: Dry- and wet- bulb temperature prevailing inside the poultry pen during the study period

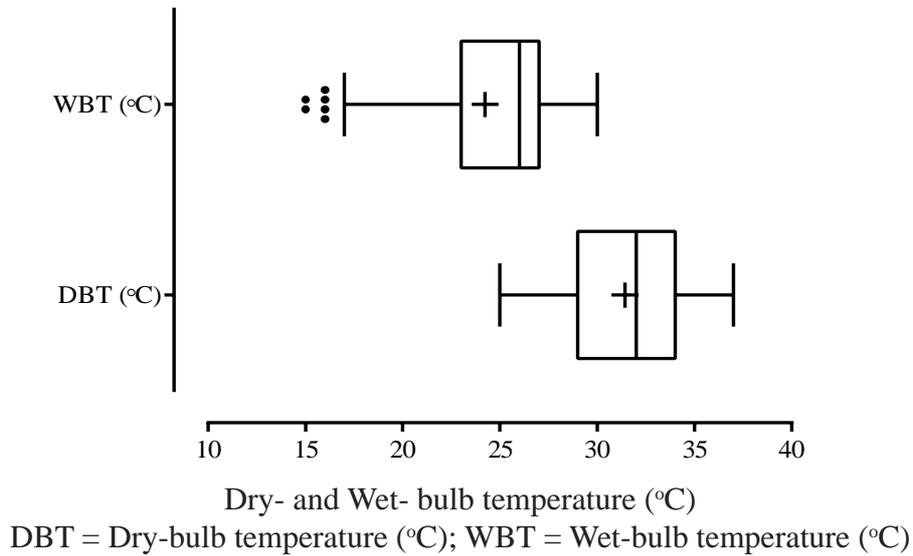
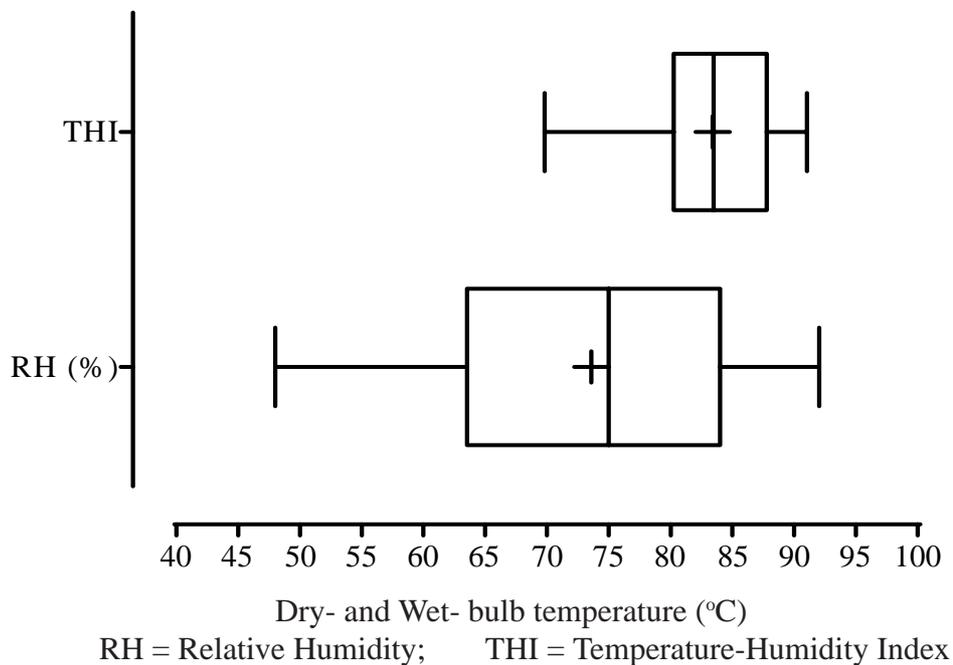
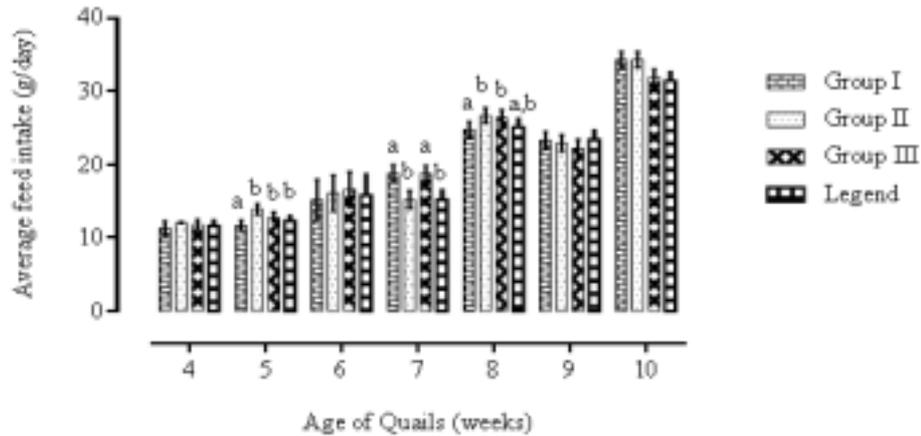


Fig. 1b: Relative humidity and temperature-humidity index prevailing inside the poultry pen during the study period

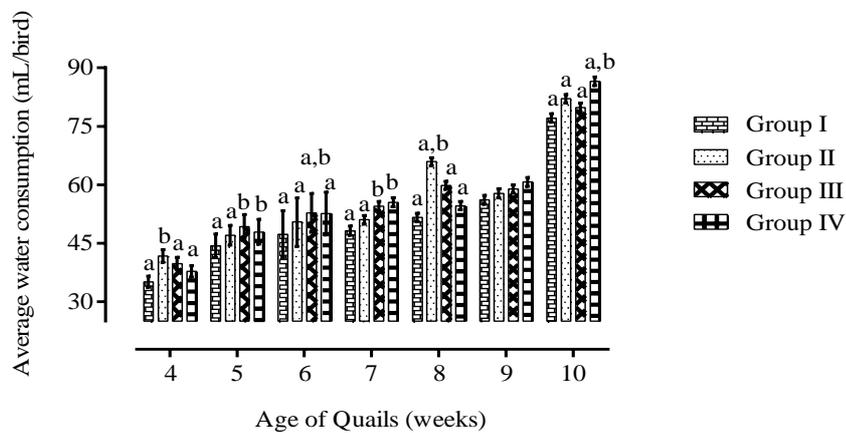


**Figure 2:** Average daily feed intake of quails on weekly basis (n=97)



<sup>ab</sup> = Mean values with different superscripts are significantly ( $P < 0.05$ ) different. Group I = Control; Group II = Asorbic acid administration; Group III = Betaine administration; Group IV = Co-administration of betaine and asorbic acid

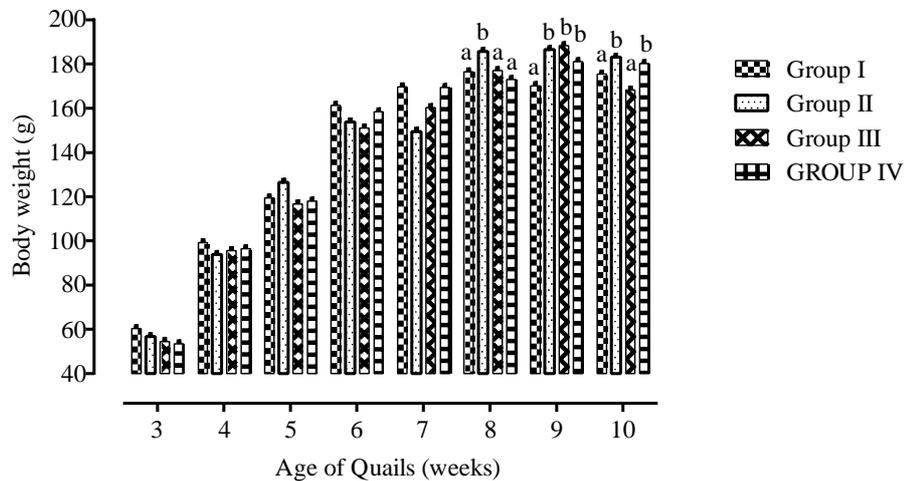
**Figure 3:** Average daily water consumption of quails on weekly basis (n=84)



<sup>a,b</sup> = Mean values with different superscripts are significantly ( $P < 0.05$ ) different. Group I = Control; Group II = Asorbic acid administration; Group III = Betaine administration; Group IV = Co-administration of betaine and asorbic acid.

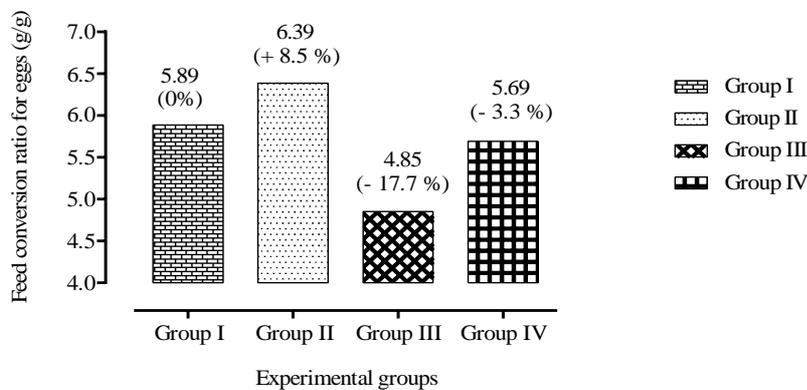


**Figure 4:** Body weight of quails on weekly basis (n=15)



<sup>a,b</sup> = Mean values with different superscripts are significantly ( $P < 0.05$ ) different. Group I = Control; Group II = Ascorbic acid administration; Group III = Betaine administration; Group IV = Co-administration of betaine and ascorbic acid.

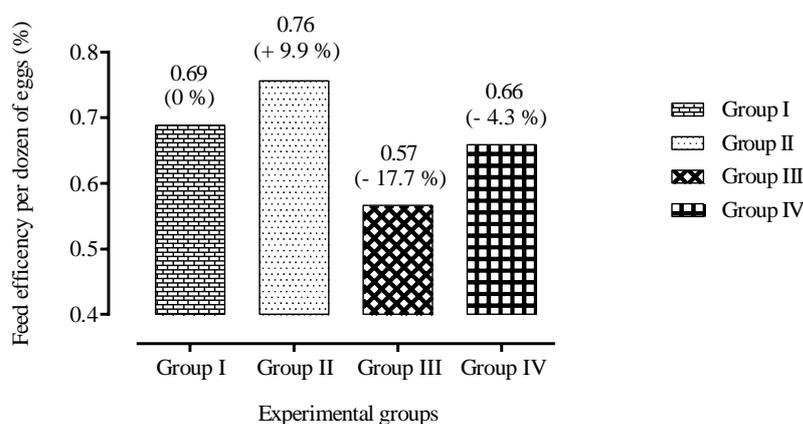
**Figure 5:** Overall feed conversion ratio for eggs produced by quails on weekly basis (n=97)



<sup>a,b</sup> = Mean values with different superscripts are significantly ( $P < 0.05$ ) different. Group I = Control; Group II = Ascorbic acid administration; Group III = Betaine administration; Group IV = Co-administration of betaine and ascorbic acid. Minus (-) and plus (+) signs indicate that values of feed-to-egg mass ratio in the treatment groups are greater or less compared to that obtained in the control group, respectively. Control served as the basis for comparison.



Figure 6: Overall feed efficiency per dozen of eggs laid by quails on weekly basis (n=97)



<sup>a,b</sup> = Mean values with different superscripts are significantly ( $P < 0.05$ ) different. Group I = Control; Group II = Ascorbic acid administration; Group III = Betaine administration; Group IV = Co-administration of betaine and ascorbic acid. Minus (-) and plus (+) signs indicate that values of feed-to-dozen egg ratio in the treatment groups are greater or less compared to that obtained in the control group, respectively. Control served as the basis for comparison.

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