

# Impacts of Heat Stress on Some Performance Parameters of Broiler Chicken Reared Under Different Stocking Densities

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## Abstract

The current study was designed to investigate the impacts of heat stress (HS) on performance parameters of broiler chicken reared at different stocking densities, also study assessed the effects of anti-stress (vitamin) supplementation in the mitigation of different levels of stress. A total of 720, 7 days old Cobb ® chicks were randomly distributed into 18 groups, (two replicates within each group). The experiment with a factorial arrangement of treatments (3x3x2), 3 levels of stocking densities (RSD: 10 chicks/m<sup>2</sup>, MSD: 14 chicks/m<sup>2</sup>, and HSD: 18 chicks/m<sup>2</sup>), 2 levels of vitamin supplementation (0 mg/l and combination of 250 mg/l ascorbic acid plus 0.5 ml/l Vit E + Se) and three temperatures (Thermoneutral temperature (TN), sudden chronic heat stress exposure (CHS) and gradual chronic heat stress exposure). Broilers were kept either under thermoneutral conditions (24±1°C) during the whole life period or slowly introduced to CHS from the 7<sup>th</sup> to 21<sup>st</sup> d of age and kept at high temperature thereafter and the third chamber had chicks that were exposed to CHS (32±2 °C for 8 h/day) during the period from the 21<sup>st</sup>: 42<sup>nd</sup> day of age. Chicks were reared on a deep litter system and had free access to feed and water. Performance parameters (FI, BW, BWG, and FCR) were determined on the 42<sup>nd</sup> day of age. The results showed HSD had adverse effects on the growth performance of broilers reared under thermoneutral or CHS conditions where the differences between densities were significant (P<0.05) under TN and sudden CHS conditions and insignificant in case of gradual CHS exposure conditions for most of the performance parameters. Vitamins supplementation had improved growth performance (BW and FCR) of broilers kept under MSD or HSD and exposed to thermoneutral or sudden CHS conditions as compared to corresponding not supplemented birds. While it was effective in combating the adverse effects of gradual HS exposure in RSD and MSD kept broilers only. In addition, there was no significant difference between RSD not supplemented broilers and MSD-supplemented birds under TN conditions; concluding that broilers can be stocked at MSD under thermoneutral conditions if they were supplemented with vitamins.

## KEYWORDS

Broilers, Heat Stress, Stocking densities

## INTRODUCTION

The contribution of broiler chicken to meat production has significantly expanded over the previous few decades, primarily due to better genetic and management techniques (Kryeziu *et al.*, 2018). Even though all parts of production have improved, environmental issues continue to have a significant impact on the production of chicken (Mahmoud and Yaseen, 2005). A variety of environmental stressors, such as vaccination, feed withdrawal, heat stress, high stocking density, disease challenges, poor sanitation, and improper management, are constantly present in modern broiler production systems, endangering the productive performance, health status, and well-being of birds (Goo *et al.*, 2019).

According to Lara and Rostagno (2013), heat stress, combined with rising global temperatures and current poultry genotypes' decreased heat tolerance, is one of the most significant stressors affecting the poultry business globally (IPCC, 2007). The poultry sector is thought to incur annual costs of millions of dollars (St-Pierre *et al.*, 2003). Because they lack sweat glands and

are covered in feathers, poultry has been demonstrated to be vulnerable to heat stress (Nicol, 2011). This is because they have a restricted ability to control heat loss by evaporation (Hirakawa *et al.*, 2020). Heat stress is brought on by an imbalance between the quantity of heat energy produced and the net amount of energy flowing from the body to its surroundings (Lara and Rostagno, 2013). When environmental temperature surpasses 30°C in the raising area, heat stress negatively impacts chicken health and performance by lowering feed intake, feed efficiency, nutrient utilization, and feed conversion ratio (Sahin *et al.*, 2003). Heat stress can even result in death (De Basilio and Picard, 2002).

In addition to HS, stocking density (SD) is regarded as one of the most crucial environmental elements affecting the production of the chicken business (Adeyemo *et al.*, 2016). According to Kryeziu *et al.* (2018), SD is typically reported as the body mass (kg) or several birds being raised in each area (m<sup>2</sup>). According to EU regulation 43/2077, SD can vary depending on the country and husbandry system (Mahrose *et al.*, 2019), although the most common stocking densities recommended are 33:42 kg living weight/m<sup>2</sup> kg (at least in Europe) and 30 kg live weight/m<sup>2</sup> in

warm climates (Qaid *et al.*, 2016).

In the modern poultry industry, broilers are typically raised at high stocking densities to increase the amount of meat produced per unit area, lower production costs, and achieve satisfactory financial returns (Li *et al.*, 2019); however, these conditions have a negative impact on broiler health, welfare, and performance. These issues could be brought on by restricted availability to feed and water, strange behavior, and poor air and floor quality (Estevez, 2007). Furthermore, excessive SD might cause a mild case of HS by raising the temperature in chickens' microenvironments and lowering body heat dissipation (Cengiz *et al.*, 2015).

Therefore, several methods, such as genetics (Gowe and Fairfull, 2008), housing, the use of ventilators and foggers on poultry farms (DEFRA, 2005), thermal conditioning Yahav and McMurtry (2001), as well as feeding and nutrition, have been suggested to mitigate the negative effects of high environmental temperature and increased stocking density on poultry performance (Chand *et al.*, 2016; Daghir, 2008).

According to Sahin *et al.* (2004), one of the most popular and effective strategies to reduce the harmful effects of high environmental temperature on chickens is through dietary changes that include antioxidant vitamins and minerals (Das *et al.*, 2011). While ascorbic acid (vitamin C), often known as the anti-stress hormone, is at the top of the list of antioxidant vitamins that help to resist the effects of stress on animal bodies. It is associated with a lower plasma level of corticosterone in stressed poultry (Mahmoud *et al.*, 2004). Because of its capacity to combat free radicals and lessen lipid peroxidation in both plasma and skeletal muscles, vitamin E (-tocopherol), another biological antioxidant, helps to improve growth, physiological, and immunological parameters in broilers (Selim *et al.*, 2013). Additionally, the trace element selenium is crucial for the immunological system of chickens. It is a crucial part of the antioxidant enzyme glutathione peroxidase (GPx), which is crucial in the fight against disease (Kidd, 2004). As a result, it indirectly contributes to the body's defense against oxidative stress (Inbaraj *et al.*, 2016).

Therefore, the current study was designed to investigate the effects of chronic heat stress on the performance parameters of broiler chicken reared under different stocking densities in comparison with thermoneutral-reared chicken and to evaluate the effect of the addition of vitamins as a method of stress alleviation.

## MATERIALS AND METHODS

### Study period and location

All procedures and experiments were performed in accordance with the Ethics of the Committee of Local Experimental Animal Care and were approved by the Nutrition and Veterinary Clinical Nutrition Institutional Committee, Faculty of Veterinary Medicine, Damanhour University, Egypt (DMU2018-0045).

All feasible measures were taken to reduce animal suffering. The current study, which lasted six weeks from September to October 2019, was conducted at the Poultry Unit of the Faculty of Veterinary Medicine at Damanhur University in Egypt.

### Experimental chicks and bird husbandry

Al Shrouk Company provided 800 Cobb® chicks that were one day old and unsexed in total. At arrival, the chicks were all weighed. To test maternal immunity, about fifteen chicks were raised until the third day of life and then slaughtered. The remaining chicks were subsequently raised in the same room (using a deep litter system) until the seventh day of life, which is the

period of adaption.

A total of 720 chicks were weighed on their seventh day of life and chosen to serve as the experimental unit, with the other chicks being culled. Three rooms with a total of 36 floor pens each holding 12 chicks were randomly chosen to house the 36 chicks. Pens had wire mesh in the middle and were constructed of solid wood with 1.6 m<sup>2</sup>, 1.42 m<sup>2</sup>, and 1.33 m<sup>2</sup> floor area. Chicks were reared on a cement floor covered in a 5 cm deep layer of rice hulls from the 1<sup>st</sup> to the 42<sup>nd</sup> day of age and fresh litter was supplied as required by good management practice.

Using manual plastic feeders and drinkers, which were distributed as one bell drinker and one feed hopper in the same spot per each enclosure, food and water were made available to animals as they needed it. The birds were fed a well-balanced commercial poultry diet that had been designed to satisfy the NRC (1994) specifications for broiler chicks. Birds got three intra-ocular doses of the Newcastle disease vaccine: Hitchner IB plus HIPRAGUMBORO at seven days old, ND LaSota MLV plus Gumboro D78 at fourteen days old, and BAL - ND LaSota at twenty-one days old.

With the help of small electric fans for air circulation and gas, and heaters set 50 cm above the floor, the temperature of the rooms was managed. Exhaust fans and two holes of 16 cm diameter were provided for ventilation where the location of the fans ensured air movement through the whole room and cooling for birds (Hassan *et al.*, 2007).

Using a mercury thermometer, daily maximum, and minimum temperatures were recorded inside and outside the shed during the experiment (TFA Dostmann GmbH & Co., KG, D-97B77, Germany). At chick height, the temperature within the shed was recorded. Using a hair hygrometer, relative humidity (RH%) was monitored twice daily at 9:00 and 18:00. (HYGRO, made in Germany) (Tuerkyilmaz, 2008).

### Experimental design

Chicks were distributed into 18 experimental groups as represented in Table 1, where the experiment was designed to investigate the impacts of stocking density, heat treatment, and stress alleviation additive within two replicates for each treatment. Chicks at the 7<sup>th</sup> day old were assigned randomly to one of three rooms until the end of the experimental period (Thermo neutral room, gradually chronic heat stress exposure, and sudden chronic heat stress adjusted room). Chicks in each room were housed in the floor pens present into three stocking density levels with two replicates per each: Recommended stocking density (RSD), medium stocking density (MSD), and high stocking density (HSD) which were represented as 10 chicks/m<sup>2</sup>, 14 chicks/m<sup>2</sup>, according to the guidelines published by Barnett *et al.* (2008) and 18 chicks/m<sup>2</sup>; respectively; where chicks were kept under these densities from the 7<sup>th</sup>: 42<sup>nd</sup> day of age. This distribution provided a floor space of 1000 cm<sup>2</sup>/bird in RSD, 714.285 cm<sup>2</sup>/bird in MSD, and 555.55 cm<sup>2</sup>/bird in HSD. The available feeder space per bird for each stocking density was 7.07 cm/bird in RSD, 5.657 cm/bird in MSD, and 4.714 cm/bird in HSD; respectively; according to the equation described by Madilindi *et al.* (2018) as following: Feeder space /bird = circumference of the feeder (cm) /the number of birds in each pen.

The corresponding drinker space was 5.693 cm/ bird in RSD, 4.555 cm/bird in MSD, and 3.795 cm/bird in HSD; respectively; according to the same previous equation. Allocation of birds was based on minimizing variation in initial weights between replicate pens. The cubical space per bird was adjusted regularly in case of mortality occurrence by isolation with a wire net to make sure

that the density treatment was not affected (Tong *et al.*, 2012).

#### Thermal stress protocol

Birds were allocated into three adjacent chambers with a different temperature protocols. Generally, Chicks in the first chamber were raised as thermo-neutral groups where they were exposed to a room temperature of  $33 \pm 1^\circ\text{C}$  for the first 3 days of life that decreased by  $3^\circ\text{C}$  weekly ( $0.5^\circ\text{C}$  per day) until a temperature of  $24 \pm 1^\circ\text{C}$  was reached at the end of the third week and maintained at that level till the end of the trial.

While chicks in the second chamber were maintained at a room temperature of  $33 \pm 1^\circ\text{C}$  for the first 3 days of age then decreased by  $1^\circ\text{C}$  weekly until a temperature of  $30^\circ\text{C}$  was reached by the end of the third week then they were subjected to the chronic heat stress protocol afterward. Finally, chicks in the third chamber were raised as the thermo-neutral groups of the first room till the 21<sup>st</sup> day of age then they followed the chronic heat stress protocol later (Gonzalez-Esquerria and Leeson, 2005).

At the age of 21<sup>st</sup> day, chicks of the second and third climatic rooms were subjected to a chronic heat stress protocol scheme by exposure to the environmental temperature of  $32 \pm 2^\circ\text{C}$  for 8 h (from 10.00:18.00 h) and  $22 \pm 2^\circ\text{C}$  (from 18.00:10.00 h) during the period of 21<sup>st</sup>: 42<sup>nd</sup> day of age (Moeini *et al.*, 2012).

#### Stress alleviation additive

Chicks, with different densities, at the three chambers were classified into: treated and non-treated groups according to the type of water they received at the 7<sup>th</sup> to the 42<sup>nd</sup> day of age as follows:

##### Non-treated groups

Chicks received fresh drinking water alone during the whole period of the experiment.

##### Treated groups

Chicks received drinking water supplied with 250 mg/l ascorbic acid (CM 003 VTN ASCORBIC ACID, Vitamin C 100%, and Batch NO.76019, Producer& supplier: Vita Trace Nutrition Ltd, P.O. Box 23886, Nicosia 1687, CYPRUS. Imported Company: Dakahlia Poultry Co., Eng. Mohmoud Al Anani & Co., Nasr City, Cairo. Egypt) plus 0.5 ml/l Vit E + Se (Farma Selenium 10%; Batch No:

FS 1513, manufactured by: Kanzy Farmatech Inc., Canada) during the 7<sup>th</sup> to the 42<sup>nd</sup> day of age according to Jena *et al.*, (2013).

#### Broilers performance parameters evaluation

Performance traits including feed intake, body weight, body weight gain, feed conversion ratio, and mortality rate were calculated for each age interval and cumulatively from day 7<sup>th</sup> to 42<sup>nd</sup> of age. These performance parameters were corrected according to mortality as follows:

##### Feed intake (FI)

Feed consumption was recorded on a pen basis at weekly intervals by weighing a known amount of feed for each pen (replicate) at the beginning of each week and the residual feed was collected and weighed at the end of each week replicate using an electronic balance. The FI per pen was calculated from the difference between the amount of feed added every week and the feed residues of each pen. Then FI per each group was recorded as the average of FI of its two replicates.

##### Body weight (BW)

All chicks were individually weighed on arrival to determine the initial weight and thereafter body weight (g) was determined weekly for up to 6 weeks using a movable weighing scale. From the individual weights, the mean weight of all the groups was calculated separately. The weight recorded was with no previous fasting period.

##### Body weight gain (BWG)

Body Weight Gain (BWG) was assessed on a pen basis at weekly intervals throughout the experimental period. Weekly body gain was obtained by calculating the difference between body weights of each two successive weeks according to Brady (1968).

##### Feed Conversion Ratio (FCR)

The pen feed conversion ratio was calculated by dividing total feed intake by weight gain for each week interval according to Fritz *et al.* (1969). Total FCR was calculated as an average of the FCR of the 2 replicates per each experimental group. The FCR was

Table 1. Experimental design implemented in this study.

Experimental groups	Thermal Protocol					
	Thermoneutral conditions		Sudden chronic heat stress exposure		Gradual chronic heat stress exposure	
	Description	No. of birds / m <sup>2</sup>	Description	No. of birds /m <sup>2</sup>	Description	No. of birds / m <sup>2</sup>
G <sub>1</sub>	RSD – not supplemented birds	10 birds	RSD – not supplemented birds	10 birds	RSD – not supplemented birds	10 birds
G <sub>2</sub>	RSD – supplemented birds	10 birds	RSD – supplemented birds	10 birds	RSD – supplemented birds	10 birds
G <sub>3</sub>	MSD – not supplemented birds	14 birds	MSD – not supplemented birds	14 birds	MSD – not supplemented birds	14 birds
G <sub>4</sub>	MSD - supplemented birds	14 birds	MSD - supplemented birds	14 birds	MSD - supplemented birds	14 birds
G <sub>5</sub>	HSD – not supplemented birds	18 birds	HSD – not supplemented birds	18 birds	HSD – not supplemented birds	18 birds
G <sub>6</sub>	HSD - supplemented birds	18 birds	HSD - supplemented birds	18 birds	HSD - supplemented birds	18 birds

\* RSD: Recommended stocking density, MSD: Medium stocking density; HSD: High stocking density.

calculated after adjusting for the body weight (BW) of dead birds.

Statistical analysis

The data were statistically analyzed using the GLM procedure for one-way analysis of variance (SAS, 2002) to investigate the significant difference between different treatments.

RESULTS AND DISCUSSION

The results achieved in Table 2, showed that performance parameters were significantly decreased by increasing SD in thermoneutral reared broilers where RSD recorded the highest mean value of FI, BW, BWG, and FCR (4600.618 g/bird, 2694.38±73.15 g/bird, 2546.25±70.31 g/bird and 1.83±0.05; respectively in non-supplemented groups and 4844.73 g/bird, 2693.75±99.35 g/bird, 2545.94±95.85 g/bird and 1.95±0.09; respectively in non-supplemented groups) as compared to corresponding HSD reared birds. These results complied with results recorded by Tong et al. (2012) and Heidari and Toghyani (2018), who found that FI, BW, and BWG of broilers significantly declined when SD was increased.

The negative impacts of HSD on broilers' performance can

be attributed to inadequate floor space which results in poor microenvironmental conditions inside the poultry house, competition for feed and water, increased litter moisture condition, and elevated ammonia levels due to degradation of uric acid by microorganisms (Jayalakshmi et al., 2009). On the other hand, FI results were contrasted to the findings of Thomas et al. (2004) and Ravindran et al. (2006), who found that the FI of birds grown at different densities was similar (P ≤ 0.05). Also, BW results interfered with the results recorded by Adeyemo et al. (2016), who observed that birds reared at HSD (14 birds /m<sup>2</sup>) had numerically higher final BW (2262.80 g) than birds placed at LSD (10 birds/ m<sup>2</sup>). While BWG results disagreed with Ravindran et al. (2006) who found that live WG of birds reared at densities of (10, 15, and 20 birds per m<sup>2</sup>) were similar concluding that SD did not influence performance parameters.

FCR results coincided with Gupta et al. (2017) findings ' which recorded that FCR was significantly (P<0.05) higher (1.73 and 1.76; respectively) in SD of 12 and 8 birds/m<sup>2</sup> as compared to 16 birds/m<sup>2</sup> SD (1.52) and the result recorded by Kryeziu et al. (2018) which found that FCR was improved by HSD treatment (1.66) as compared to other densities (1.64 in MSD and 1.67 in LSD) although the difference among different groups was insignificant. A possible explanation is that birds of the LSD group had more

Table 2. Performance parameters of broiler chicken reared under different stocking densities at the thermoneutral condition.

Treated groups	Performance parameters			
	FI / g	LBW / g	BWG / g	FCR
Group (1): Thermoneutral RSD not supplemented with additive	4600.62	2694.38±73.15 <sup>a</sup>	2546.25±70.31 <sup>a</sup>	1.83±0.05 <sup>abc</sup>
Group (2): Thermoneutral MSD not supplemented with additive	4181.27	2480±62.16 <sup>bc</sup>	2340±59.26 <sup>bc</sup>	1.81±0.06 <sup>abc</sup>
Group (3): Thermoneutral HSD not supplemented with additive	3594.70	2301.46±36.59 <sup>c</sup>	2166.25±34.48 <sup>c</sup>	1.67±0.03 <sup>cd</sup>
Group (4): Thermoneutral RSD supplemented with additive	4844.73	2693.75±99.35 <sup>a</sup>	2545.94±95.85 <sup>a</sup>	1.95±0.09 <sup>a</sup>
Group (5): Thermoneutral MSD supplemented with additive	4169.1	2548.89±69.59 <sup>ab</sup>	2408.06±66.96 <sup>ab</sup>	1.75±0.05 <sup>bc</sup>
Group (6): Thermoneutral HSD supplemented with additive	3555.26	2410.43±60.6 <sup>bc</sup>	2274.35±58.25 <sup>bc</sup>	1.59±0.04 <sup>d</sup>

RSD: Recommended stocking density; MSD: Medium stocking density; HSD: High stocking density. Means within the same column carry different superscripts are significantly different.

Table 3. Performance parameters of broiler chicken reared under different stocking densities exposed to sudden chronic heat stress conditions.

Treated groups	Performance parameters			
	FI / g	LBW / g	BWG / g	FCR
Group (1): Sudden CHS RSD not supplemented with additive	4347.35	2577.67±60.24 <sup>a</sup>	2429.33±57.87 <sup>a</sup>	1.8±0.04 <sup>ab</sup>
Group (2): Sudden CHS MSD not supplemented with additive	3990.53	2307.5±51.54 <sup>b</sup>	2171.25±48.61 <sup>b</sup>	1.86±0.04 <sup>ab</sup>
Group (3): Sudden CHS HSD not supplemented with additive	3651.19	2223.75±70.76 <sup>b</sup>	2085.5±68.71 <sup>b</sup>	1.79±0.06 <sup>ab</sup>
Group (4): Sudden CHS RSD supplemented with additive	4556.67	2535.67±47.7 <sup>a</sup>	2392.67±45.7 <sup>a</sup>	1.91±0.04 <sup>a</sup>
Group (5): Sudden CHS MSD supplemented with additive	3953.54	2310.5±47.06 <sup>b</sup>	2167±44.76 <sup>b</sup>	1.84±0.04 <sup>ab</sup>
Group (6): Sudden CHS HSD supplemented with additive	3647.74	2301.96±47.53 <sup>b</sup>	2162.61±44.9 <sup>b</sup>	1.7±0.04 <sup>b</sup>

RSD: Recommended stocking density; MSD: Medium stocking density; HSD: High stocking density. Means within the same column carry different superscripts are significantly different.

Table 4. Performance parameters of broiler chicken reared under different stocking densities exposed gradually to chronic heat stress conditions.

Treated groups	Performance parameters			
	FI / g	LBW / g	BWG / g	FCR
Group (1): Gradual CHS RSD not supplemented with additive	5059.42	2332.92±54.79 <sup>a</sup>	2185.83±52.14 <sup>a</sup>	2.33±0.06 <sup>a</sup>
Group (2): Gradual CHS MSD not supplemented with additive	3709.09	2236.84±64.2 <sup>a</sup>	2090.79±62.26 <sup>a</sup>	1.82±0.05 <sup>bc</sup>
Group (3): Gradual CHS HSD not supplemented with additive	3675.93	2207.61±59.04 <sup>a</sup>	2068.48±57.03 <sup>a</sup>	1.81±0.05 <sup>c</sup>
Group (4): Gradual CHS RSD supplemented with additive	5035.58	2336.67±69.01 <sup>a</sup>	2186.33±66.88 <sup>a</sup>	2.34±0.08 <sup>a</sup>
Group (5): Gradual CHS MSD supplemented with additive	4020.37	2236.84±46.33 <sup>a</sup>	2090±44.61 <sup>a</sup>	1.94±0.04 <sup>b</sup>
Group (6): Gradual CHS HSD supplemented with additive	3539.41	2160.42±39.47 <sup>a</sup>	2024.17±37.56 <sup>a</sup>	1.76±0.03 <sup>c</sup>

RSD: Recommended stocking density; MSD: Medium stocking density; HSD: High stocking density. Means within the same column carry different superscripts are significantly different.



available feeding and moving space, and although consumed more feed, they did not convert it effectively into tissues due to energy losses (Kryeziu *et al.*, 2018). On the other hand, FCR results were not in line with Palizdar *et al.* (2017) and Elkolaly *et al.*, (2019), who found that FCR was adversely affected by increasing SD.

The results recorded in Tables 3 and 4, showed that performance parameters were decreased by increasing SD under chronic heat stress conditions where the difference between the experimental groups was significant in sudden CHS exposure and insignificant under gradual CHS exposure conditions. Whereas, RSD reared broilers recorded the highest mean value of FI, BW, and BWG (4347.353 g/bird, 2577.67±60.24 g/bird and 2429.33±57.87 g/bird; respectively, in non-supplemented groups and 4556.665 g/bird FI, 2535.67±47.7 g/bird BW and 2392.67±45.7 g/bird BWG; respectively in vitamin supplemented birds) in sudden CHS exposure and (5059.42 g/bird, 2332.92±54.79 g/bird, and 2185.83±52.14 g/bird; respectively in non-supplemented groups and 5035.58 g/bird FI, 2336.67±69.01 g/bird BW and 2186.33±66.88 g/bird BWG; respectively in vitamin supplemented birds) in gradual CHS exposure as compared to corresponding HSD housed birds.

Results of FI, BW, and BWG recorded in Tables 3 and 4, agreed with the general view that performance parameters were significantly decreased in birds raised under HSD and HS conditions (Uzum and Toplu, 2013; Chegini *et al.*, 2019; Gholami *et al.*, 2020). On the other hand, FI results were in contrast with Rabie *et al.* (2013) findings that found that FI was not significantly affected by different SD (9.04 and 11.3 birds/m<sup>2</sup>) in broilers reared during the summer season which may be referred to the lower and convergence of stocking densities applied in their study so its influence was not clear on FI. On the other hand, BW and BWG results were not in line with the results recorded by Villagra *et al.* (2010), who found that there was no difference in BW of birds housed at 15 birds/m<sup>2</sup> (1386±17) and BW of those found at 20 birds/m<sup>2</sup> (1379±17) under cyclic HS (32°C for 4h/day).

There are many explanations for poor growth performance in heat-stressed chickens especially under HSD rearing conditions, the most important factor responsible for decreasing performance parameters can be referred to as the reduction of voluntary feed intake of stressed birds as a mechanism to decrease heat increment (Sohail *et al.*, 2012) which consequently adversely influences the absorption of amino acids and other essential nutrients required for growth.

According to FCR results recorded in Tables 3 and 4, it was declared that there were significant differences ( $P < 0.05$ ) in total FCR among different experimental groups housed at different densities under sudden or gradual CHS exposure conditions. Where it was observed that the broilers group reared at HSD had the lowest FCR (1.79±0.06 in the non-supplemented group and 1.7±0.04 in supplemented birds) under sudden CHS exposure conditions and (1.81±0.05 in non-supplemented group and 1.76±0.03 in supplemented birds) under gradual HSD exposure conditions as compared to corresponding RSD housed birds.

In comparing the results of Tables 2 and 3, with the results of Table 1, in the point related to FCR, it was noticed that HS-exposed groups had worse FCR as compared to thermoneutral reared groups under different SD which come following the general viewpoint reported by Quinteiro-Filho *et al.* (2012); Lara and Rostagno (2013); Olfati *et al.* (2018) and He *et al.* (2020), who assured that HS adversely affected FCR of exposed birds. On the other hand, FCR results recorded in Tables 2 and 3 were not in harmony with results obtained by Rabie *et al.* (2013) and Goo *et al.* (2019), who found that high SD had significantly decreased ( $p$

< 0.05) feed efficiency (483 g/bird in 9 birds/m<sup>2</sup> and 486 g/bird in 9 birds/m<sup>2</sup>) of heat-stressed broilers.

These results are difficult to explain because of limited information on the interactive effects of HS and SD on poultry. However, it is speculated that stress responses may be synergistic between HS and high SD (Goo *et al.*, 2019).

Dietary supplementation of birds with vitamin C, vitamin E, or a combination of these two antioxidant compounds was recommended in various studies in attenuating the deleterious impacts of HS, especially on poultry performance (Attia *et al.*, 2017). By analyzing the impacts of the addition of vitamin supplementation on performance parameters, it was observed that there were no significant differences in growth performance parameters (BW, BWG, or FCR) between supplemented and corresponding not supplemented birds reared at different stocking densities under thermoneutral or CHS conditions which came following the results obtained by El-Gogary *et al.* (2015) who recorded that dietary supplementation with VE at different levels insignificantly affected live BW, BWG, FI, and FCR.

Although of the significant difference in performance parameters between supplemented and non-supplemented groups, FI, BW, BWG, and FCR were improved with vitamin supplementation in MSD and HSD-housed birds under thermoneutral conditions. These results came in line with Desoky (2018), who observed that dietary supplementation of vitamin E improved the broiler performance under intensive birds' density (12 birds/m<sup>2</sup>) as compared to the density of 10 birds/m<sup>2</sup>. Also, there was no significant difference between RSD not supplemented broilers and MSD-supplemented birds; concluding that broilers can be stocked at MSD under thermoneutral conditions if they were supplemented with vitamins which comes parallel to the finding recorded by Adebiji *et al.* (2011).

Concerning CHS-exposed groups, there were no significant differences in growth performance parameters (BW, BWG, or FCR) between supplemented and corresponding not supplemented birds although BW and FCR were improved in MSD and HSD-supplemented birds suddenly exposed to CHS. In gradual CHS conditions, it was found that BW; BWG, and FCR were improved in RSD-supplemented birds; FI and FCR only were higher in MSD-supplemented broilers while the performance parameters were not increased by vitamin supplementation in HSD as compared to the corresponding not supplemented groups; concluding that vitamin supplementation was effective in combating adverse effects of gradual HS exposure in RSD and MSD kept broilers. These results agree with Sahin *et al.* (2003), who demonstrated that a combination of vitamin C and vitamin E was able to ameliorate the effect of HS and overstocking where it provided the greatest performance in Japanese quails reared under HS (34°C) and Tawfeek *et al.* (2014) who showed that the heat stressed broilers supplemented vitamin C, vitamin E, and minerals had significantly improved weight gain, FCR, and decreased FI.

Moreover, it was noticed that performance parameters were improved in MSD supplemented with vitamins (FI and FCR were lower while BW was higher) as compared to control RSD not supplemented birds under gradual CHS conditions; concluding that broilers can be stocked at MSD if they were supplemented with vitamins. This agreed with Khattak *et al.* (2012) who registered that performance of broilers subjected to HS and supplemented with vitamin E at 300 mg kg<sup>-1</sup> diet was improved as compared to the control group (35 mg kg<sup>-1</sup> vitamin E). However, these results were not in agreement with Lopes *et al.* (2015), who claimed that levels of zinc and vitamin E above the commonly used do not improve the growth performance of broilers kept under high temperatures and reared on reused litter.

## CONCLUSION

This study's results revealed that HSD had adverse effects on the growth performance of broilers reared under thermoneutral or CHS conditions where the differences between densities were significant ( $P < 0.05$ ) under TN and sudden CHS conditions and insignificant in case of gradual CHS exposure conditions for most of the performance parameters. There were no significant differences in growth performance parameters (BW, BWG, or FCR) between supplemented and corresponding not supplemented birds reared at different stocking densities under thermoneutral or CHS conditions, although vitamins supplementation had improved growth performance (BW and FCR) of broilers kept under MSD or HSD and exposed to thermoneutral or sudden CHS conditions as compared to corresponding not supplemented birds. Vitamin supplementation was effective in combating the adverse effects of gradual CHS exposure in RSD and MSD-kept broilers only. In addition, there was no significant difference between RSD not supplemented broilers and MSD-supplemented birds under TN conditions; concluding that broilers can be stocked at MSD under thermoneutral conditions if they were supplemented with vitamins.

## CONFLICT OF INTEREST

The authors declare that they do not have conflict of interest.

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