





# **Original** Article

# **Production and Physiological Parameters of Broiler Chickens Administered Chilled Drinking Water under High Ambient Temperature During Finisher Period**

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## **ABSTRACT**

High environmental temperature is of great concern in production of broiler chickens in the tropics, and this experiment was conducted to evaluate the ameliorative effect of chilled drinking water on the production and physiological parameters of broiler chickens reared in the tropics. This 56-day experiment contained 3 treatments ( $T_0$ ,  $T_1$ , and T<sub>2</sub>), replicated 3 times to contain 16 birds per replicate. T<sub>0</sub> (control) was offered non-chilled drinking water without increasing ambient temperature; T<sub>1</sub> was offered non-chilled drinking water with an increased ambient temperature of 30°C between 11:30 – 15:30 GMT, daily; T<sub>2</sub> was offered chilled drinking water (8-10 °C) with the increased ambient temperature of 30°C between 11:30 – 15:30 GMT, daily. The total body weight gain of the birds was similar (P>0.05) in  $T_0$  and  $T_2$ , which were significantly (P<0.05) higher than that of  $T_1$ . Feed intake was significantly (P<0.05) higher in  $T_0$  than in  $T_1$  and  $T_2$ , which were similar (P>0.05), while feed conversion ratio (FCR) was significantly (P<0.05) higher in  $T_1$  than in  $T_0$  and  $T_2$ , which were similar (P>0.05). Water intake was significantly (P<0.05) higher in  $T_2$  than in  $T_1$ ,

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which was significantly (P<0.05) higher than the water intake of  $T_0$  chickens. Hemoglobin (Hb) concentration and red blood cell (RBC) count were similar (P>0.05) between  $T_0$  and  $T_2$ , which were significantly (P<0.05) higher than values for  $T_1$ . Packed cell volume (PCV), white blood cell (WBC), lymphocyte and platelet counts, and blood coagulation time were similar (P>0.01) between  $T_0$  and  $T_2$ , which were significantly (P<0.01) higher than those of  $T_1$ . The serum globulin of  $T_0$  chickens was similar (P>0.05) to that of  $T_2$ , but significantly (P<0.05) higher than the serum globulin of  $T_1$  chickens, which was similar (P>0.05) to that of  $T_2$ . Serum sodium (Na+) and potassium (K+) ions were significantly (P<0.05) higher in  $T_0$  than in  $T_1$  and T<sub>2</sub>, which were similar (P>0.05), while serum hydrogen carbonate ion (HCO<sup>3-</sup>) was significantly (P<0.05) higher in  $T_2$  than in  $T_1$  which was similar (P>0.05) to that of  $T_0$ , and that of T<sub>0</sub> similar to that of T<sub>2</sub>. Respiratory rate and cloacal temperature were significantly (P  $\leq 0.01$ ) different among the treatments (T<sub>1></sub>T<sub>2></sub>T<sub>0</sub>), while body temperature was significantly (P<0.05) higher in  $T_1$  than in  $T_0$  and  $T_2$ , which were similar (P>0.05). Therefore, administering chilled drinking water (8-10 °C) to broiler chickens during periods of high ambient temperature can ameliorate heat stress through improved FCR, weight gain, stabilization of blood parameters and constituents, and reduction of high respiratory rate, body, and cloacal temperatures. Keywords: Blood, Broiler chickens, Electrolytes, Growth Performance, Respiratory rate, Temperature.

#### Introduction

Broiler chickens (*Gallus gallus domesticus*) are domesticated fowls reared mainly for meat production (Garrigus, 2007). They are a hybrid of the egg-laying chicken, both being sub-species of the red jungle fowl (*Gallus gallus*), which is a tropical member of the family, *Phasianidae*, and can freely interbreed with populations of red jungle fowl (Liu, 2006). Chickens belong to a class of animals that regulate body temperature within a narrow range, provided the ambient temperature is maintained within the comfort zone (Genc, 2005).

High ambient temperatures accompanied by high humidity which characterizes the tropics and sub-tropic areas of the world have been major problems affecting the performance and physiological attributes of chickens (Attia *et al.*, 2016). The high ambient temperature usually results in heat stress (Lucas and Marcos, 2013), which has been reported to affect the respiratory rate, feed intake, and blood parameters (Allan *et al.*, 2000). Heat stress is responsible for major incidences of morbidity and mortality among broilers (Attia *et al.*, 2009), and constitutes a major impediment to broiler bird production in tropical environments. Broilers are probably prone to heat stress because they lack sweat glands, and have low respiratory evaporation, coupled with their high metabolic rate (Deeb and Cahaner, 2002; Olanrewaju *et al.*, 2010),

thereby limiting their ability to maintain normal temperature during heat stress (Bollengier-lee *et al.*, 1999; Narongsak, 2004).

Although birds may pant and increase water intake in their attempt to reduce heat stress, several approaches have been adopted to curb incidences of heat stress in broiler bird production, occasioned by high environmental temperatures in the tropics (Lin et al., 2006; Abioja et al., 2011; Pradeepta et al., 2015). Administration of cold water and vitamin C to broiler birds during high environmental temperature in Southwest Nigeria significantly increased the relative weight of spleen, weekly and total weight gains, while 500 mg of Vitamin C significantly increased the relative weights of their breast meat when compared with birds not receiving vitamin C (Abioja et al., 2011). Poultry houses have been designed and constructed to reduce heat stress through proper insulation and ventilation, while acclimatization and feeding strategies were also not left out among the techniques to curb heat stress in poultry production (Pradeepta et al., 2015). Genetic strategies geared toward selection for heat-tolerant breeds are also an option for the prevention of heat stress in broilers (Lin et al., 2006). This experiment was conducted to evaluate the ameliorative effect of chilled drinking water on the production and physiological parameters of broiler chickens reared in the tropics.

## **MATERIALS AND METHODS**

This 56-day study was carried out at the Federal University of Technology Owerri, Teaching and research farm, located between latitude  $4^{\circ}-4'-6^{\circ}-3'N$  and longitude  $6^{\circ}-15'-8^{\circ}-15'E$  in the Southeast agro-ecological zone of Nigeria. The mean annual rainfall, temperature, and humidity of the area are 250mm, 26.5-27.5°C, and 70-80 %, respectively (Iwuji *et al.*, 2017).

One hundred and forty-four (144) One-day-old broiler birds of Abor acres breed were used in a completely randomized design (CRD), containing three treatments (T<sub>0</sub>, T<sub>1</sub>, and T<sub>2</sub>), and replicated thrice to contain sixteen birds per replicate. The birds were brooded for three weeks with fresh clean water and a standard broiler chick diet containing 22 % CP and 2800 kcal ME/kg offered ad libitum. Routine management, vaccination, and medication practices as described by Oluyemi and Roberts (2007) were carried out during the experiment. On the  $22^{nd}$  day, the birds were transferred to the experimental pens where they were allowed to acclimatize for one week. On the  $28^{th}$  day, the birds were divided into three groups, on a weight equalization basis, and randomly allotted to the treatments. Birds on Treatment zero (T<sub>0</sub>) served as control and were offered ordinary non-chilled fresh water under the natural subsisting ambient temperature (without heat application to increase the ambient temperature). Birds on Treatment one (T<sub>1</sub>) were administered ordinary fresh water, and heat was supplied to increase the ambient temperature of the pens, at the level of the birds, to  $30^{\circ}$ C between the hours of 11:30 - 15:30 GMT, daily, for four weeks. Birds on Treatment two (T<sub>2</sub>) were administered chilled drinking water, at 8-10°C, and heat was supplied to increase the ambient temperature of the pen, at the level of the birds, to 30°C between the hours of 11:30 - 15:30 GMT, daily, for four weeks.

The chilled water at 8-10 °C was achieved through the use of ice blocks in the drinkers, monitored with the aid of a digital thermometer. The ambient temperature of  $30^{\circ}$ C in T<sub>1</sub> and T<sub>2</sub> were maintained through the use of heating stoves. The ambient temperature and humidity of the pens were monitored and recorded with the aid of wet and dry bulb thermometers. All the broiler finisher chickens were offered the same standard diet, containing 20 % CP and 3200 kcal ME/kg, from the 28<sup>th</sup> day till the end of the experiment (56<sup>th</sup> day).

## **Data Collection**

## **Growth Performance**

Data were collected on initial body weight, feed intake, weight gain, feed conversion ratio, and final body weight of the bird's following procedures stated by Iwuji *et al.* (2019), while water intake of the birds was calculated daily as the difference between the volume offered (including volume of ice) and the volume remaining. No mortality was recorded during the experimental period.

## **Blood Parameters**

At the end of the experimental period, six birds per treatment (three birds per pen) were randomly selected, and blood collection was done during slaughter, following the procedure outlined by Thaxton, *et al.* (2009). Blood samples for hematological analysis were collected into bottles containing ethylene diamine tetra acetic acid (EDTA) (anticoagulant), while blood samples for serum biochemical analysis were collected into sterile bottles containing no anticoagulant. Hematological analysis was done using Sysmex KX-21N<sup>TM</sup> Automated Hematology Analyzer (Sysmex Europe GmbH), while biochemical analysis was carried out using the Semi-automated clinical chemistry analyzer, Microlab 300 (Vital Scientific, India); and serum electrolytes determined following procedures described by Wanda *et al.*, (2013).

## **Respiratory Rate, Body, and Cloacal Temperatures**

Respiratory rate, body, and cloacal temperatures were measured three times a day at 7.00 - 8.00h, 13.00 - 14.00h, and 17.00 - 18.00h GMT on two randomly selected birds from each pen. Respiratory rate was determined by counting the number of panting breaths for 15 seconds and multiplying by 4 to get the number of panting breaths per minute (respiratory rate). Body and cloacal temperatures were measured under the wings and at the cloaca respectively, using a digital thermometer.

Data generated from this study (growth performance, hematology, serum biochemistry and electrolytes, respiratory rate, body, and cloacal temperatures) were subjected to analysis of variance (ANOVA), using SAS (2010). Significantly (P < 0.05) different means were separated using the LSD test.

## RESULTS

## **Growth Performance**

The growth performance of the experimental birds is presented in Table 1. The average final body weight and total body weight gain of the birds were similar (P>0.05) between birds on  $T_0$  and  $T_2$  which were significantly (P<0.05) higher than those of birds on  $T_1$ . Total feed intake was significantly (P<0.05) higher in  $T_0$  than in  $T_1$  and  $T_2$  birds which were similar (P>0.05). The feed conversion ratio of the birds on  $T_1$  was significantly (P<0.05) higher than those of the birds on  $T_0$  and  $T_2$ , which were similar (P>0.05). Total water intake was significantly (P<0.05) different among the experimental groups in the order of  $T_{2>}T_{1>}T_0$ .

 Table 1: Growth performance of broiler birds drinking chilled water under high ambient temperature

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Parameters (g)	T <sub>0</sub>	T <sub>1</sub>	$T_2$	P-value	
Av. Initial body weight	$1143.20{\pm}1.05$	1146.41±1.22	$1141.22 \pm 0.98$	0.071	
Av. Final body weight	$3090.11 {\pm} 0.93^{a}$	$2752.30{\pm}0.48^{b}$	$3031.19{\pm}0.77^{a}$	0.024	
Av. Total body weight gain	1946.91ª	1505.89 <sup>b</sup>	1889.97ª	0.011	
Av. Total feed intake	4123.02ª	3894.34 <sup>b</sup>	3991.33 <sup>b</sup>	0.032	
Feed conversion ratio (g feed/g gain)	2.12 <sup>b</sup>	2.59ª	2.11 <sup>b</sup>	0.041	
Av. Total water intake (ml)	7628.59°	9835.85 <sup>b</sup>	9981.81ª	0.012	
Mortality (%)	-	-	-	-	

<sup>abc</sup> Means within a row with different superscripts are significantly (P<0.05) different.

## **Blood Parameters**

The hematological parameters of the experimental broiler birds are presented in Table 2. Hemoglobin (Hb) concentration and red blood cell (RBC) count, were similar (P>0.05) between T<sub>0</sub> and T<sub>2</sub>, which were significantly (P<0.05) higher than values obtained for T<sub>1</sub>, while packed cell volume (PCV), white blood cell (WBC), lymphocyte and platelet counts, and blood coagulation time were similar (P>0.01) between T<sub>0</sub> and T<sub>2</sub>, which were significantly (P<0.01) higher than those of T<sub>1</sub>. Heterophil count and heterophil/lymphocyte ratio of the birds were significantly (P<0.05) higher in T<sub>1</sub> (3.01 ×10<sup>3</sup>/mm<sup>3</sup> and 0.49) than in T<sub>0</sub> (4.14×10<sup>3</sup>/mm<sup>3</sup> and 0.25) and T<sub>2</sub> (3.15×10<sup>3</sup>/mm<sup>3</sup> and 0.28) birds which were similar (P>0.05) between T<sub>1</sub> and T<sub>2</sub> (138.89 and 15.36 fl) which were significantly (P<0.05) higher than MCV of birds on T<sub>0</sub> (127.34 fl). Mean corpuscular hemoglobin (MCH) was significantly (P<0.05) higher in T<sub>1</sub> (45.76 pg) than in T<sub>0</sub> (36.43 pg); but similar (P>0.05) between T<sub>0</sub> and T<sub>2</sub> (40.53 pg), and between T<sub>1</sub> and T<sub>2</sub>. Mean corpuscular hemoglobin concentration

(MCHC) was significantly (P<0.05) higher in  $T_1$  (32.95 g/dl) than in  $T_0$  (28.61 g/dl) and  $T_2$  (29.94 g/dl) which were similar (P>0.05).

Table 2. Hematological parameters of the experimental imister broner enterens				
Parameters	To	<b>T</b> 1	<b>T</b> 2	<b>P-value</b>
Hb (mg/dl)	$10.82{\pm}0.39^{a}$	9.06±0.21 <sup>b</sup>	$10.66 \pm 0.38^{a}$	0.034
RBC ( $x10^{6}/mm^{3}$ )	$2.97{\pm}0.20^{a}$	$1.98{\pm}0.15^{b}$	$2.63{\pm}0.10^{a}$	0.022
PCV (%)	$37.82{\pm}0.48^{a}$	$27.50 \pm 0.34^{b}$	35.60±0.41ª	0.001
MCV (fl)	$127.34 \pm 2.16^{b}$	$138.89 \pm 2.10^{a}$	135.36±2.13 <sup>a</sup>	0.045
MCH (pg)	$36.43 \pm 0.42^{b}$	45.76±0.33 <sup>a</sup>	$40.53 \pm 0.41^{ab}$	0.041
MCHC (g/dl)	$28.61 \pm 0.31^{b}$	$32.95{\pm}0.45^{a}$	$29.94{\pm}0.29^{b}$	0.035
WBC $(x10^{3}/mm^{3})$	16.11±0.21ª	$13.17 \pm 0.22^{b}$	$15.47 \pm 0.29^{a}$	0.002
Lymphocytes (x10 <sup>3</sup> /mm <sup>3</sup> )	$12.06 \pm 0.44^{a}$	$8.41 \pm 0.29^{b}$	11.35±0.23 <sup>a</sup>	0.010
Monocytes $(x10^3/mm^3)$	$0.38 \pm 0.04$	$0.27 \pm 0.02$	$0.35 \pm 0.06$	0.065
Heterophils $(x10^3/mm^3)$	$3.01 \pm 0.09^{b}$	$4.14{\pm}0.10^{a}$	$3.15 {\pm} 0.08^{b}$	0.014
H/L ratio	$0.25 \pm 0.02^{b}$	$0.49{\pm}0.05^{a}$	$0.28 {\pm} 0.03^{b}$	0.048
Eosinophils (x10 <sup>3</sup> /mm <sup>3</sup> )	$0.34{\pm}0.03$	$0.24{\pm}0.03$	$0.33 \pm 0.04$	0.078
Basophils (x10 <sup>3</sup> /mm <sup>3</sup> )	$0.17 \pm 0.02$	$0.12 \pm 0.01$	$0.16\pm0.01$	0.094
Platelets $(x10^3/mm^3)$	$22.04{\pm}0.12^{a}$	$18.14{\pm}0.14^{b}$	21.61±0.21 <sup>a</sup>	0.002
Blood coagulation time (Secs)	74.31±2.23 <sup>b</sup>	$87.96 \pm 3.04^{a}$	75.11±3.12 <sup>b</sup>	0.001

Table 2: Hematological parameters of the experimental finisher broiler chickens

 $^{ab}$  Means on the same row with different superscripts are significantly different. H/L = heterophil/lymphocyte ratio.

Serum biochemical analysis of the experimental birds (Table 3) recorded higher (P<0.05) values of total protein (TP) and globulin (Glb) in T<sub>0</sub> (3.88 and 1.68 g/dl) than in T<sub>1</sub> (3.01 and 1.03 g/dl) birds, but TP and Glb values of the birds were similar (P>0.05) between T<sub>0</sub> and T<sub>2</sub> (3.42 and 1.42 g/dl), and between T<sub>1</sub> and T<sub>2</sub>.

 Table 3: Serum biochemical parameters of the experimental finisher broiler

chickens					
Parameters	T <sub>0</sub>	<b>T</b> <sub>1</sub>	<b>T</b> <sub>2</sub>	<b>P-value</b>	
TP (g/dl)	$3.88{\pm}0.49^{a}$	$3.01 \pm 0.12^{b}$	$3.42{\pm}0.34^{ab}$	0.049	
Albumin (g/dl)	$2.20\pm0.11$	$1.98 \pm 0.08$	$2.00 \pm 0.06$	0.084	
Globulin (g/dl)	$1.68{\pm}0.10^{a}$	$1.03{\pm}0.08^{b}$	$1.42{\pm}0.14^{ab}$	0.047	
Uric acid (g/dl)	$3.61 \pm 0.44$	$3.74 \pm 0.41$	$3.62 \pm 0.32$	0.062	
Creatinine (mmol/L)	$1.33 \pm 0.14$	$1.26\pm0.11$	$1.28 \pm 0.18$	0.073	
ALT (IU/L)	$102.04{\pm}1.88$	$104.51 \pm 1.24$	$100.21 \pm 2.00$	0.067	
AST (IU/L)	$23.06 \pm 1.06$	$26.64 \pm 0.84$	$24.55 \pm 0.91$	0.059	

<sup>ab</sup> Means on the same row with different superscripts are significantly (P<0.05) different.

The results of the serum electrolyte analysis of the experimental birds are presented in Table 4. Higher (P<0.05) values of sodium and potassium ions (Na<sup>+</sup> and K<sup>+</sup>) were recorded in T<sub>0</sub> (1.34 and 3.42 mmol/L) than in T<sub>1</sub> (1.02 and 2.88 mmol/L) and T<sub>2</sub> (1.11 and 2.98 mmol/L), which were similar (P>0.05). Serum hydrogen carbonate ion

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(HCO<sup>3-</sup>) was similar (P>0.05) between  $T_0$  (21.25 mmol/L) and  $T_1$  (20.19 mmol/L), and between  $T_0$  and  $T_2$  (22.10 mmol/L), but higher (P<0.05) in  $T_2$  than in  $T_1$ .

Parameters (mmol/L)	To	$T_1$	Τ2	P-value	
$Na^+$	$1.34{\pm}0.22^{a}$	$1.02{\pm}0.14^{b}$	$1.11 \pm 0.12^{b}$	0.048	
$Ca^+$	9.86±0.41	$9.44 \pm 0.24$	9.61±0.32	0.082	
$\mathbf{K}^+$	$3.42{\pm}0.13^{a}$	$2.88 \pm 0.25$ <sup>b</sup>	$2.98{\pm}0.18^{b}$	0.043	
HCO <sup>3-</sup>	$21.25 \pm 0.16^{ab}$	$20.19 \pm 0.20^{b}$	$22.10 \pm 0.44^{a}$	0.045	
Cl-	132.40±1.22	132.22±1.39	$133.02{\pm}1.48$	0.191	

<sup>ab</sup> Means on the same row with different superscripts are significantly (P<0.05) different.

## **Respiratory Rate, Body, and Cloacal Temperatures**

Table 5 presents the respiratory rate, as well as the body and cloacal temperatures of the experimental birds. The respiratory rate and cloacal temperature of the birds on  $T_1$  were significantly (P<0.01) higher than those of  $T_2$ , which were higher (P<0.01) than those of  $T_0$ . The body temperature of the birds was significantly (P<0.05) higher in  $T_1$  than in  $T_0$  and  $T_2$ , which were similar (P>0.05).

 Table 5: Respiratory rate, body, and cloacal temperatures of broiler birds drinking chilled water under high ambient temperature

	0	1		
Parameters	To	<b>T</b> 1	T <sub>2</sub>	P-value
Respiratory rate (breaths/minute)	69.33±2.92°	$91.33{\pm}2.76^{a}$	$88.67 \pm 2.22^{b}$	0.002
Body temperature (°C)	$40.77 \pm 0.17^{b}$	$41.84{\pm}0.13^{a}$	$40.97 \pm 0.15^{b}$	0.023
Cloacal temperature (°C)	41.24±0.16°	42.52±0.11 <sup>a</sup>	$41.71 \pm 0.13^{b}$	0.010
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<sup>abc</sup> Means within a row with different superscripts are significantly different.

## DISCUSSION

The similar values recorded in  $T_0$  (control) and  $T_2$  birds for their final body weight, total body weight gain, and feed conversion ratio (FCR) are probably due to the ability of the chilled drinking water administered to the birds on  $T_2$  to ameliorate the effect of high ambient temperature, which has a declining effect on the performance of broiler chickens (Joseph *et al.*, 2012). Administration of chilled drinking water did not ameliorate the reduction in feed intake occasioned by high ambient temperature, but it caused a significant increase in the total water intake of the birds. It is then observed that the temperature of the chilled drinking water in addition to the ambient temperature also affected the water intake of the birds. Administering chilled drinking water to birds on  $T_2$  under high ambient temperature may have made more energy available for digestion, which otherwise would have been used for heat loss; and could be the reason for the significant increase recorded in the total weight gain and better FCR of  $T_2$  than  $T_1$  birds.

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Heat stress occasioned by high environmental temperature is a factor militating against the performance of broiler chickens (Altan *et al.*, 2000), and has also been implicated in the reduction of total serum protein and erythropoietin, thereby hindering or disrupting erythropoietic process for the production of blood cells (Altan *et al.*, 2000). The significant reduction in the red blood cell (RBC) count of the birds receiving heat treatment without chilled drinking water ( $T_1$ ) can be attributed to the effect of high ambient temperature. Since hemoglobin (Hb) is a component of the RBC, and packed cell volume (PCV) is majorly made up of the RBC, it then justifies the same effect as in RBC, observed in the Hb concentration and percentage PCV, recorded for  $T_1$  broiler chickens. The similar Hb, RBC, and PCV values recorded between heat-treated broiler chickens administered chilled drinking water ( $T_2$ ) and non-heat-treated broiler chickens administered ordinary water ( $T_0$  or control) indicates that chilled drinking water (8-10 °C) completely mitigated the effect of high ambient temperature on the Hb, RBC, and PCV of the broiler chickens.

The higher mean corpuscular volume (MCV) of  $T_1$  birds could be due to physiological mechanism(s) to meet energy requirements in the respiration muscles to support heavy muscle contraction in heat evaporation of heat-stressed broilers (Mushawwir *et al.*, 2018). This increase in the size of the corpuscles may have resulted from the adverse effect of heat on the hemoglobin (Hb), which is the oxygen-carrying component of the corpuscles (Fransdon *et al.*, 2009). It is expected that birds with more RBCs should also have more Hb, but since the reverse is the case in this study, and the mean corpuscular hemoglobin (MCH) and mean corpuscular hemoglobin concentration (MCHC) were significantly higher in  $T_1$ , it could be that the RBC and Hb of birds on  $T_1$  had inflammatory responses to high ambient temperature, resulting in accommodation of more moisture and oxygen aimed at mitigating the effect of high temperature, and also meet the oxygen requirement of the tissue cells. This scenario will add more weight to the Hb of the birds, as evident in the significantly higher MCH and MCHC of  $T_1$  birds.

Reduced white blood cell (WBC) and lymphocyte counts have been reported in poultry subjected to heat stress, and this is a result of the interruption of the process of blood cell formation by high ambient temperature (Altan *et al.*, 2000). The higher heterophil count and heterophil/lymphocyte ratio of  $T_1$  birds indicate stress (Altan *et al.*, 2000; Soleimani and Zulkifli, 2010), which was ameliorated by the administration of chilled drinking water in  $T_2$  birds. However, similar values of basophil counts recorded between the treatments indicate that the  $T_1$  birds did not suffer acute heat stress (Altan *et al.*, 2000).

High ambient temperature reduced the platelet count of  $T_1$  birds and this effect was ameliorated by chilled drinking water in  $T_2$  birds. The reduction in the platelet counts of  $T_1$  birds could also be due to the interruption of the process of blood cell formation in the bone marrow by high ambient temperature (Altan *et al.*, 2000). The

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significantly higher coagulation time recorded in  $T_1$  (heat treatment without chilled drinking water) birds may not necessarily be due to low platelet count but could be a result of disintegration, dissociation, or distortion of thrombocytic components by heat, thereby reducing their efficiency to form a clot or coagulate, which implies that in a situation of injury, the birds with higher coagulation/clotting time will lose more blood (Frandson *et al.*, 2009).

Administration of chilled drinking water to  $T_2$  birds ameliorated the adverse effect of high ambient temperature on serum globulin recorded in  $T_1$  birds. Since serum albumin was not significantly different between the treatments, it implies that the significant difference between the total protein (TP) of  $T_0$  and  $T_1$  birds was due to that, recorded in their serum globulin levels (Table 3). High ambient temperature reduces growth performance in broiler chickens as a result of decreased feed intake, growth rate, and feed utilization (Tang *et al.*, 2013). Reduced feed intake and utilization in  $T_1$  birds, occasioned by a relatively higher ambient temperature may have resulted in the low serum globulin levels of the birds. The low serum globulin level and total white blood cell (WBC) count of  $T_1$  birds could be an indication of a compromised immune system of the birds resulting from high ambient temperature; since WBC and globulin are involved in immune response (Frandson *et al.*, 2009).

Heat treatment has been reported to reduce serum Na and K concentrations of Red Jungle fowl, Village fowl, and broiler chickens (Soleimani and Zulkifli, 2010). In the present study, heat treatment of  $T_1$  and  $T_2$  birds resulted in a significant reduction of their serum Na<sup>+</sup> and K<sup>+</sup> concentrations. The physiological mechanism of the effect of heat treatment on the serum electrolytes of birds is not well understood, as there have been records of conflicting results (Soleimani and Zulkifli, 2010). However, a rise in blood dilution has been considered as the reason for a general lowering of the electrolyte concentration of the birds after heat treatment (Soleimani and Zulkifli, 2010). Administration of chilled drinking water to  $T_2$  birds probably eliminated the reduction effect of heat treatment on HCO<sup>3-</sup> as recorded in T<sub>1</sub>. During high ambient temperatures, the rate of respiration increases in birds, and carbonate losses have been recorded during respiration (Ahnad and Sarwar, 2006).

The high ambient temperature increased the respiratory rate of birds on  $T_1$ , while the administration of chilled drinking water stabilized the respiratory rate of birds on  $T_2$  to be similar to that of birds on the control treatment ( $T_0$ ). Birds engage in evaporative heat loss during high ambient temperatures through panting and increased respiratory rate (Narongsak, 2004; Ahnad and Sarwar, 2006). Body and cloacal temperatures of the birds on  $T_2$  (administered chilled drinking water) were significantly reduced (similar to control), then those of  $T_1$ , indicating a better thermo-regulatory mechanism, and reduction of the incidence of heat stress on  $T_2$  birds than those of  $T_1$ .

## CONCLUSION

Although ambient temperature levels recorded in this study did not induce acute stress in the experimental birds, the elevation of the ambient temperature of the  $T_1$  pens induced heat stress, leading to increased panting (Narongsak, 2004; Ahnad and Sarwar, 2006), and adversely affected weight gain, feed utilization and blood parameters of the birds, as indicated in the significant decrease of hemoglobin (Hb) concentration, red blood cells (RBC) count, packed cell volume (PCV), white blood cells (WBC), platelet counts, and the increase in their blood coagulation time. Furthermore, increased respiratory rate, as well as cloacal temperature was evident in the heat-treated birds, without chilled drinking water. However, these adverse effects observed in birds on T<sub>1</sub> were prevented or ameliorated by the administration of chilled drinking water to the birds on T<sub>2</sub>. Considering the body weight gain (BWG), feed conversion ratio (FCR), white blood cell (WBC) count, blood coagulation time, and serum globulin level results of the experimental birds, it is evident that heat stress is capable of negatively affecting feed utilization, cellular immunity, clotting ability and immunoglobulin levels of broiler birds, but can be ameliorated by chilled drinking water. Therefore, it will not be erroneous to say that chilled drinking water at 8-10 °C is a veritable option for combating heat stress in broiler bird production.

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## **CONFLICT OF INTERESTS**

The authors have no relevant financial or non-financial interests to disclose.

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