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Research Article
Interactive Effects of Light-Sources, Photoperiod and Strain on Blood Physiological Variables of Broilers Grown to Heavy Weight

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Abstract

Objective: Effects of light sources, photoperiods and strains on blood physiological variables of broilers grown to heavy weights (>3 kg) were investigated in 2 trials. **Materials and Methods:** The experimental design was a 4×2×2 factorial consisting of 4 light sources [incandescent (ICD, standard), compact fluorescent (CFL), neutral LED (Neutral-LED) and cool poultry specific LED (Cool-PS-LED)], 2 photoperiods (Regular/intermittent [2L:2D] and Short [8L:16D] and 2 strains (A, B). In each trial, 480 (240 males/240 females) 1-d-old chicks of each strain from different commercial hatcheries were equally and randomly distributed into 16 environmental-control rooms (30 males +30 females/room) at 50% RH. Each room was randomly assigned one of 16 treatments from 1-56 days of age. Birds were provided similar diets. Feed and water were provided *ad libitum*. Venous blood samples were collected on d 14, 28, 42 and 56 of age and analyzed immediately. **Results:** Light sources had significant ($p<0.05$) effects on BW, electrolytes, pCO_2, angap, T_3 and T_4 in comparison with birds reared under ICD. Short photoperiod significantly ($p<0.05$) reduced BW, pH, pO_2, SaO_2, electrolytes, Osmo and T_3, along with significantly ($p<0.05$) increased pCO_2, Hb, Hct and McHc compared with regular intermittent photoperiod. Acid-base regulation during the short photoperiod exposure had not deteriorated despite higher pCO_2 that consequently decreased blood pH due to a respiratory acidosis. Also, Strain B had significantly ($p<0.05$) increased BW, pCO_2, HCO_3-, McHc, electrolytes, angap, Osmo and T_3, along with significantly ($p<0.05$) reduced pH level, pO_2, SaO_2, Hb, Hct and T_4 in comparison with Strain A. All these changes were within broilers normal acid-base homeostasis ranges. Plasma corticosterone and blood glucose concentrations were not affected by treatments, indicating an absence of physiological stress. **Conclusion:** The results indicating that the 3 light sources evaluated along with a regular/intermittent photoperiod in commercial poultry facilities would reduce energy costs and optimize production without compromising the welfare of broilers grown to heavy weights.

Key words: Light-source, photoperiod, strains, acid-base-balance, broiler

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Data Availability: All relevant data are within the paper and its supporting information files.

INTRODUCTION

The poultry industry is making rapid progress in maximizing the efficiency of broiler growth. Advances include improved genetics and nutrition along with changes in environmental management, resulting in optimizing the genetic potential of broiler growth. In order to develop a profitability program useful to most broiler growers, environmental (air, temperature, humidity, light) recommendations to optimize profitability and welfare of broiler production are important. Light management is one of the important factors affecting broiler production and welfare[1-3]. Incandescent (ICD) bulbs have been phased out in favor of more energy-efficient lighting alternatives in poultry houses. Energy efficient lighting upgrades are a major capital investment for reducing electrical energy inputs and maximizing outputs of a poultry company.

Consumer demand for breast meat has driven a shift in market composition towards increased market weights. Over the past five decades, consumption of poultry meat has increased dramatically, which is expected to continue in the future due to the relative price-competitiveness compared to other meat products[4-6]. In addition, poultry meat production globally will further grow by 2.4% per year over the next 20 years and the total production in 2030 will be around 160 million tons that will have a share of 39% in total meat production[7]. However, as the demand for animal protein products increases, increasing poultry production and production efficiencies will be critical to the continued viability of the U.S. poultry and livestock industries.

Modern broiler strains are selected for a number of characteristics including growth rate and meat quality and these selection criteria have been adapted through the decades according to market demand. They are genetically selected for a high daily growth rate thereby resulting in fast growth with a low feed conversion. However, the genetic potentiality of poultry may not be utilized fully due to environmental constraints. Information on genotype×environmental interaction is lacking on the environmental (air, temperature, humidity, light) needs of broilers grown to heavy weights (≥3.0 kg). Concern among the American public regarding animal well-being in food production systems continues to grow and increased production efficiency is perceived to negatively impact the well-being of the animal. Environmental management strategies must be developed that increase efficiency without detrimental effects on the welfare of poultry and livestock. In the United States, market share of commercial broilers is dominated by birds originating from two strains (A and B) that are regularly used in the poultry industry worldwide. The first strain A is a late-developing strain, which has been marketed as a high meat yielding bird with a superior feed conversion efficiency rate and maximized breeder performance high yield. The second strain B is early-developing, which is a unique product genetically developed to provide the best live production efficiency with the highest breast meat yielding at the least cost and best feed efficiency. The rate of development differs among strains and lighting program needs may be strain specific for these two strains in order to optimize growth performance. Substantial unbiased scientific information is limited on comparison of these strains A and B on the effect of differing photoperiods in combination with the new LED light-sources currently used by the poultry industry on blood physiological indices, since establishing proper welfare practices are central to international trade negotiations of meat products.

Hematological analyses in combination with other biochemical methods, provide a thorough evaluation of the health status of individuals and they are the indicators of internal organ health and systemic homeostasis. Plasma or serum biochemical analyses provide information about internal organs, electrolytes and metabolic parameters, among others. Changes in acid-base balance are the early manifestation or clinical signs of many diseases in both domestic animals and human beings. Hence, it is necessary to examine these factors on blood physiological variables of broilers grown to heavy weights under these new light sources in combination with photoperiods to ensure the health and welfare of broilers. Our previous study[8] investigated the effects of light-sources and photoperiod on hemato-physiological indices of broilers grown to heavy weights. However, lack of comparison of these strains A and B on blood physiological indices begs the question as to whether they respond similarly to environmental adjustment changes in production practices of these two strains. To the knowledge of the authors, no previous research has been conducted regarding broiler strain comparison on the effects of LED light-sources in combination with photoperiod on blood physiological indices that relates to the welfare of strains A and B grown to heavy weights used in the current study. Therefore, the objective of this study was to evaluate the effects of light sources (CFL, LED, PS-LED) in comparison with incandescent (ICD) bulbs in combination with photoperiod on blood physiological indices that relates to the welfare of strains A and B grown to heavy weights (>3.0 kg), since establishing proper welfare practices are central to international trade negotiations of meat products.

MATERIALS AND METHODS

Bird husbandry: All procedures relating to the use of live birds in this study were approved by the USDA-ARS Animal Care and Use Committee at the Mississippi State location. In addition, unnecessary discomfort to the birds was also avoided by using proper housing and handling techniques[9]. The present study used birds from the same group examined in a recent study[10]. This experiment was repeated two times and in each, 480 (240 males/240 females) 1-d-old chicks of each strain were purchased from different commercial hatcheries. Upon arrival, the chicks were sexed and group-weighed. Birds were equally and randomly distributed into 16 environmental-control rooms (30 males and 30 females/room) at 50% RH. Each room was randomly assigned one of 16 treatments from d 1 to 56 d of age. Each environmental room had a floor area of 2.3×2.6 m (5.98 m^2) with a room volume of 14.95 m^3 (ceiling height = 2.5 m). The stocking density was 10.7 birds/m^2 (1 bird/ft^2), which resulted in a final stocking density of 52 kg/m^2 at the end of each trial. Each room contained approximately 7.62 cm depth of fresh pine shavings, tube feeders and a 7-nipple watering system. Chicks were vaccinated for Marek's, Newcastle and infectious bronchitis diseases at the hatchery. At 12 days of age, birds received a Gumboro vaccination via water administration. The chicks remained in their respective rooms from 1-d-old throughout the experimental period (1-56 days of age). All birds were fed the same diet throughout the study. Birds were provided a 4-phase feeding program (starter: 1-14 days; grower: 15-28 days; finisher: 29-42 days and withdrawal: 43-56 days of age). Diets were formulated to meet or exceed NRC[11] nutrient recommendations for each feeding phase. Starter feed was provided as crumbles and subsequent feeds were provided as whole pellets. Feed and water were offered *ad libitum*. Temperature and relative humidity (RH) on day 1 were maintained at 32±1.1°C and 50 ± 5%, respectively and RH was held constant across all treatments. Temperature was decreased as the birds progressed in age until 15.6°C was reached at 49 days of age.

Treatments: The experimental design was a 4×2×2 factorial consisting of 4 light sources (ICD, 2010k, Standard; CFL, 2700k; Neutral-LED, 3500k; Cool-PS-LED, 5000k) from 1-56 days of age and exposure to photoperiod consisted of continuous lighting (24L:0D) with 20 lx of intensity from placement to 7 days of age and then subjected to the following two photoperiods (regular/intermittent [2L:2D] and short/non-intermittent [8L:16D] from d 8 to 48 and [23L:1D] from day 49-56, respectively) and two strains (A, B). Each of the two photoperiod treatments was paired with one of the four light sources equally with the two strains so that each room represented a particular photoperiod:light source: strain combination for a total of 16 rooms of the 16 treatments. Neutral-LED light bulbs were purchased from NexGen Illumination Inc. (Fayetteville, AR), CFL light bulbs were purchased from Osram Sylvania (Danvers, MA) and Cool-PS-LED light bulbs, made specifically for poultry, were purchased from Once-Innovation (Plymouth, MN). Light spectra of the light sources and ICD along with light intensity settings utilized in this study have been reported previously[12].

Blood collections and chemical analyses: On d 14, 28, 42 and 56, venous blood samples were collected between 0800 and 0900 h on sampling d from a brachial vein of 6 (3 male and 3 female chicks/room) randomly selected birds from each room and the birds were then returned to the appropriate rooms by using our standard handling procedure[12]. Within 45 sec after birds were caught, blood samples were collected directly into heparinized syringes for immediate analysis of blood gas and electrolytes using a gas/electrolyte analyzer (ABL80-COOX-Flex, Radiometer America, Westlake, OH) while pH, pCO_2, pO_2 and HCO_{3-} values were corrected to reflect broiler body temperature of 41.5°C. Blood samples were then centrifuged at 4000 g for 20 min at 4°C to obtain plasma samples and 2 mL of each of the plasma samples were stored in 2.5 mL graduated tubes at -20°C for later chemical analyses. Plasma samples were removed from the freezer, thawed and analyzed for corticosterone using a universal microplate spectrophotometer (Bio-Tek Instruments Inc., Winooski, VT) with ELISA reagent assay test kits (EIA-CS Kit, Enzo Life Sciences, Farmingdale, NY), according to the manufacturer's instructions. Levels of plasma triiodothyronine (T_3) and thyroxine (T_4) concentrations were measured using a universal microplate spectrophotometer (Bio-Tek Instruments Inc.) with ELISA reagent assay test kits from ALPCO Diagnostics (Salem, NH) according to the manufacturer's instructions.

Statistical analysis: The experimental design was a randomized complete block design and two trials were conducted. Treatment structure was a 4×2×2 factorial arrangement with the main factors being four light sources (ICD, CFL, Neutral-LED, Cool-PS-LED), two photoperiods (regular/intermittent [2L:2D] and short/non-intermittent [8L:16D]) and two strains (A,B). Individual sample data within each of the replicate units were averaged before analysis and data from the two trials were pooled and analyzed together. A mixed model ANOVA employing PROC MIXED procedure of SAS software[13] was used to analyze the data. Trial was a

random effect, whereas the light sources, photoperiod and strains were the fixed effect. Room was considered the experimental unit with 60 birds/room and treatments were replicated over time. Rooms used were switched randomly among treatments between trials to remove room effects so that treatments were not confounded. Main effects of light sources, photoperiod, strains and the interaction of the three factors were tested. Means comparisons on day 14, 28, 42 and 56 were assessed by least significant differences and statements of significance were based on $p<0.05$ unless otherwise stated. Analyses of variance combined across days were performed to obtain treatment comparisons averaged across days and to test for treatment interactions with equal variances between days. In addition, ANOVAs for each of the 2 week interval sampling days was performed.

RESULTS

The combined main effects of light sources, photoperiod and strains on selected blood physiological variables are shown in Table 1. In comparison with ICD light, birds reared under CFL group had higher Na^+ ($p = 0.001$) and Osmo ($p = 0.002$), while birds reared under Neutral-LED light sources had lower pH ($p = 0.033$), pO_2 ($p = 0.038$), SaO_2 ($p = 0.032$) and higher pCO_2 ($p = 0.002$), K^+ ($p = 0.011$). Furthermore, in comparison with ICD light, birds reared under Cool-PS-LED light sources had higher BW ($p = 0.045$), HCO_3 ($p = 0.021$), McHc ($p = 0.023$), K^+ ($p = 0.011$), along with decreased SaO_2 ($p = 0.022$) and T_3 ($p = 0.002$) concentrations. In addition, no main effects of light sources on Hb, Hct, Ca^{2+}, angap, GLU and CORT were observed. In comparison with regular/intermittent photoperiods, short/non-intermittent photoperiod significantly reduced BW ($p = 0.032$), pH ($p = 0.023$), pO_2 ($p = 0.001$), SaO_2 ($p = 0.001$), Ca^{2+} ($p = 0.003$), Na^+ ($p = 0.001$), K^+ ($p = 0.004$), Cl^- ($p = 0.001$), Osmo ($p = 0.003$) and T_3 ($p = 0.033$) along with significantly elevated pCO_2 ($p = 0.025$), Hb ($p = 0.014$), Hct ($p = 0.043$) and McHc ($p = 0.002$) concentrations. In addition, there were no effects of photoperiod on HCO_3^-, anion gap, glucose and T_4. Moreover, in comparison with Strain A, Strain B had higher BW ($p = 0.030$), pH ($p = 0.032$), pCO_2 ($p = 0.044$), HCO_3^- ($p = 0.001$), McHc ($p = 0.032$), Ca^{2+} ($p = 0.005$), Na^+ ($p = 0.001$), K^+ ($p = 0.002$), Cl^- ($p = 0.023$), Angap ($p = 0.035$), Osmo ($p = 0.031$), T_3 ($p = 0.035$), along with significantly lower pO_2 ($p = 0.035$), SaO_2 ($p = 0.012$), Hb ($p = 0.012$) and Hct ($p = 0.011$). There were no effects of light source, photoperiod and strains on blood glucose and plasma corticosterone concentrations.

Unlike Table 1, which has ANOVA combined effects of treatments over day, Table 2-4 represented separate ANOVAs for each of the sampling days of 2 week interval. The influence of light sources, photoperiod, strains and their interaction on whole blood pH and pCO_2 is presented in Table 2. As shown in Table 2, there was no main effect of light sources found on pH and pCO_2 in any of the sampling days. However, in comparison with regular/intermittent photoperiods, short/non-intermittent photoperiod significantly reduced blood pH on d 28 ($p = 0.025$), 42 ($p = 0.023$) and an increase in pCO2 ($p = 0.027$) on day 42. In addition, in comparison with Strain A, Strain B had higher pH on d 28 ($p = 0.028$), day 42 ($p = 0.001$), day 56 ($p = 0.021$), along with higher pCO_2 on d 42 ($p = 0.009$).

Table 3 shows the effects of light sources, photoperiod, strains and their interaction on whole blood concentrations of pO_2 and SaO_2. Birds reared under CFL had higher blood concentration of pO_2 on d 56 ($p = 0.019$) along with increased blood concentration of SaO_2 on d 42 ($p = 0.043$) in comparison with the other two new light sources. In comparison with regular/intermittent, short/non-intermittent significantly reduced blood concentration of pO_2 ($p = 0.042$) on day 56 and SaO_2 on day 42 ($p = 0.016$) and d 56 ($p = 0.021$). In addition, in comparison with Strain A, Strain B had lower pO_2 on day 42 ($p = 0.001$), day 56 ($p = 0.039$), along with lower SaO_2 on day 42 ($p = 0.042$) and d 56 ($p = 0.039$). There were treatments interactions on pO_2 on d 42 ($p = 0.046$), day 56 ($p = 0.006$) and on SaO_2 on day 42 ($p = 0.032$) and day 56 ($p = 0.047$).

The influence of light sources, photoperiod, strain and their interaction on blood concentrations of Ca^{2+} and K^+ is presented in Table 4. In comparison with ICD, birds reared under Cool-PS-LED had significantly reduced blood concentrations of Ca^{2+} on day 56 ($p = 0.015$). Furthermore, in comparison with ICD, birds reared under Cool-PS-LED had significantly reduced blood concentrations of K^+ on day 14 ($p = 0.009$), 28 ($p<0.012$) and 42 ($p = 0.021$). Also, short/non-intermittent photoperiod in comparison with regular/intermittent, significantly reduced blood Ca^{2+} concentration on day 14 ($p = 0.020$), 28 ($p = 0.001$), 42 ($p = 0.024$), day 56 ($p = 0.023$), along with that of blood K^+ concentration on day 14 ($p = 0.024$), day 28 ($p = 0.027$) and day 42 ($p = 0.045$). In addition, in comparison with Strain A, Strain B had higher Ca^{2+} concentration on d 14 ($p = 0.001$), 28 ($p = 0.009$), 42 ($p = 0.007$), day 56 ($p = 0.016$), along with significantly higher blood K^+ concentration on day 14 ($p = 0.012$), day 28 ($p = 0.001$) and day 42 ($p = 0.044$).

As shown in Table 5, there were no effects of light sources on Na^+ and Cl^- on any of the sampling days. However,

Table 1: Combined main effects of light sources, photoperiod and strains on selected blood physiological variables of broilers grown to heavy weights[ab]*

	Light sources[2]						Photoperiod				Strains			
Variables[1]	ICD	CFL	Neutral-LED	PS-LED	SEM	p-value	Reg-Inter	Short-Non-Inter	SEM	pvalue	A	B	SEM	p-value
BW (kg)	3.962[b]	4.072[ab]	4.074[ab]	4.087[a]	0.028	0.045	4.047[a]	3.905[b]	0.021	0.032	3.923[b]	4.069[a]	0.021	0.030
pH	7.35[a]	7.34[ab]	7.33[b]	7.32[b]	0.004	0.033	7.35[a]	7.32[b]	0.003	0.023	7.36[a]	7.34[b]	0.003	0.032
pCO_2 (mmHg)	58.42[b]	58.47[b]	60.64[a]	60.65[a]	0.651	0.007	58.52[b]	60.67[a]	0.702	0.025	58.23[b]	60.40[a]	0.713	0.044
pO_2 (mmHg)	55.45[a]	56.75[a]	54.68[b]	54.58[b]	0.363	0.038	54.65[a]	53.23[b]	0.471	0.001	56.31[a]	54.42[b]	0.615	0.035
HCO_{3-} (mmHg)	27.56[b]	27.63[b]	27.66[b]	28.81[a]	0.140	0.021	27.83	28.46	0.121	0.076	27.14[b]	28.18[a]	0.117	0.001
SaO_2 %	74.84[a]	74.87[a]	72.77[b]	71.83[b]	1.011	0.034	74.45[a]	71.71[b]	0.331	0.000	74.56[a]	71.43[b]	1.031	0.012
Hb (g dL^{-1})	8.32	8.36	8.27	8.31	0.087	0.253	8.25[b]	8.56[a]	0.092	0.014	8.48[a]	8.28[b]	0.056	0.031
Hct (%)	25.87	25.98	25.71	25.79	0.261	0.264	25.45[b]	26.35[a]	0.262	0.004	26.35[a]	25.51[b]	0.135	0.011
McHc (g dL^{-1})	32.16[b]	32.15[b]	32.18[ab]	32.24[a]	0.026	0.023	32.14[b]	32.19[a]	0.001	0.002	32.14[b]	32.18[a]	0.012	0.032
Ca^{2+} (meq L^{-1})	3.01	3.01	2.99	2.99	0.001	0.523	3.01[a]	2.99[b]	0.007	0.001	2.98[b]	3.02[a]	0.007	0.005
Na^+ (meq L^{-1})	152.00[b]	154.45[a]	151.14[b]	151.12[b]	0.716	0.001	153.15[a]	151.12[b]	0.651	0.001	151.32[b]	153.35[a]	0.653	0.001
K^+ (meq L^{-1})	4.21[b]	4.26[b]	4.67[a]	4.55[a]	0.045	0.011	4.45[a]	4.24[b]	0.034	0.001	4.26[b]	4.55[a]	0.092	0.002
Cl^- (meq L^{-1})	107.46[ab]	108.26[a]	107.49[ab]	107.38[b]	0.644	0.043	107.61[a]	106.42[b]	0.241	0.002	107.44[b]	108.45[a]	0.334	0.023
Angap (meq L^{-1})	20.45	20.48	20.04	20.32	0.252	0.261	20.18	20.46	0.178	0.324	20.05[b]	20.59[a]	0.178	0.035
GLU (mg dL^{-1})	252.02	252.86	251.12	251.33	2.39	0.654	250.35	253.32	1.687	0.356	252.361	251.423	2.374	0.526
Osmo mOs kg^{-1})	317.78[b]	321.79[a]	317.44[b]	318.06[b]	1.234	0.002	319.16[a]	316.37[b]	0.751	0.003	317.42[b]	319.23[a]	0.572	0.031
CORT (pg mL^{-1})	2519.96	2437.63	2473.32	2381.80	183.4	0.624	2379.20	2327.20	129.710	0.264	2214.86	2391.47	129.710	0.513
T_3 (ng mL^{-1})	3.14[b]	3.18[a]	3.20[a]	3.20[a]	0.010	0.002	3.25[a]	3.16[b]	0.026	0.033	3.17[b]	3.21[a]	0.012	0.035
T_4 (µg dL^{-1})	2.71[a]	2.68[b]	2.67[b]	2.67[b]	0.064	0.023	2.74	2.82	0.141	0.231	2.75[a]	2.71[b]	0.012	0.034

[ab]Means within a row and treatment that lack common superscripts differ significantly ($p<0.05$); *Values are least squares of 8 replicate rooms with 60 birds per room. [1]BW: Body weight; pCO_2: Partial pressure of CO_2, pO_2: Partial pressure of O_2, HCO_{3-}: Bicarbonate, SaO_2: Saturated O_2, Hb: Hemoglobin, Hct: Hematocrit, McHc: Mean corpuscular hemoglobin concentration, GLU: Glucose, Osmo: Osmolality, angap: Anion gap, CORT: Corticosterone, T_3: Triiodothyronine, T_4: Thyroxine.[2]Light Sources; 1: Incandescent light (ICD, Standard), 2: Compact Fluorescent light (CFL), 3: Light Emitting Diode (Neutral-LED), 4: Poultry Specific-LED (Cool-PS-LED), [3]Photoperiod: Reg-Inter: Regular/intermittent (2L:2D), Short-Non-Inter: Short/non-intermittent (8L:16D)

Table 2: Influence of light sources, photoperiods, strains, and their interaction on blood pH and partial pressure of CO_2 (pCO_2) of broilers grown to heavy weights*

	pH				pCO_2 (mmHg)			
Item	14 day	28 day	42 day	56 day	14 day	28 day	42 day	56 day
Light sources[1]								
ICD	7.340	7.310	7.300	7.290	55.630	59.930	61.680	63.370
CFL	7.340	7.310	7.300	7.300	55.820	60.480	62.970	61.140
Neutral-LED	7.340	7.310	7.290	7.300	56.140	60.030	64.020	60.090
Cool-PS-LED	7.330	7.310	7.300	7.290	57.150	61.340	62.800	62.740
Photoperiod[2]								
Reg-inter	7.350	7.350[a]	7.310[a]	7.300	56.020	60.840	61.490[b]	62.250
Short-non-Inter	7.330	7.310[b]	7.290[b]	7.300	56.360	60.050	64.240[a]	61.420
Pooled SEM	0.008	0.006	0.004	0.006	0.820	1.280	0.906	1.467
Strains								
A	7.330	7.300[b]	7.290[b]	7.290[b]	56.040	60.000	62.460[b]	61.370
B	7.340	7.320[a]	7.310[a]	7.310[a]	56.330	60.890	63.270[a]	62.300
Pooled SEM	0.006	0.006	0.004	0.005	0.580	0.905	0.263	2..075
Interaction								
ICD-Reg-inter-A	7.350	7.290	7.270	7.270	55.790	60.190	65.560	67.340
ICD-Reg-inter-B	7.340	7.320	7.330	7.310	56.580	61.590	59.250	63.770
ICD-short-non-inter-A	7.350	7.230	7.260	7.260	57.310	64.920	69.120	66.570
ICD-short-non-inter-B	7.360	7.320	7.310	7.320	53.980	59.480	61.960	58.970
CFL-Reg-Inter-A	7.350	7.300	7.280	7.310	56.790	60.280	62.830	60.920
CFL-Reg-Inter-B	7.340	7.330	7.300	7.330	55.560	58.840	64.560	57.500
CFL-short-non-inter-A	7.340	7.310	7.320	7.290	57.790	60.360	59.320	62.730
CFL-short-non-inter-B	7.340	7.320	7.300	7.300	55.350	61.040	63.330	59.740
Neutral-LED-Reg-inter-A	7.330	7.320	7.290	7.290	53.380	57.230	62.140	62.930
Neutral-LED-Reg-inter-B	7.350	7.310	7.320	7.300	56.790	60.270	59.750	58.960
Neutral-LED-ICD-short-non-inter-A	7.310	7.320	7.290	7.320	56.970	59.070	60.680	55.570
Neutral-LED-ICD-short-non-inter-B	7.350	7.330	7.330	7.290	56.030	61.910	60.140	63.480
Cool-PS-LED-Reg-inter-A	7.340	7.300	7.290	7.300	53.520	59.070	62.550	58.600
Cool-PS-LED-Reg-inter-B	7.340	7.310	7.280	7.300	58.700	61.910	66.140	63.360
Cool-PS-LED-short-non-inter-A	7.330	7.300	7.290	7.280	56.830	61.950	63.980	63.250
Cool-PS-LED-short-non-inter-B	7.340	7.300	7.300	7.300	58.650	62.010	64.560	65.240
Pooled SEM	0.018	0.016	0.012	0.014	1.640	1.287	3.058	2.934
Source of variation	p-value							
Light sources	0.957	0.975	0.923	0.157	0.568	0.860	0.759	0.385
Photoperiod	0.221	0.025	0.037	0.893	0.681	0.540	0.027	0.574
Strains	0.604	0.028	0.001	0.021	0.690	0.505	0.299	0.182
Interaction	0.968	0.768	0.264	0.109	0.861	0.664	0.694	0.315

[a-c]Means within a column and effect that lack common superscripts differ significantly ($p<0.05$); *Values are least squares of 2 replicate rooms with 60 birds per room, [1]Light sources; 1: Incandescent light (ICD, Standard), 2: Compact Fluorescent light (CFL), 3: Light Emitting Diode (Neutral-LED), 4: Poultry Specific LED (Cool-PS-LED), [2]Photoperiod; Reg-Inter: Regular/intermittent (2L:2D), Short-Non-Inter: Short/non-intermittent (8L:16D)

short/nonintermittent photoperiod in comparison with regular/intermittent photoperiod showed significantly reduced blood concentration of Na^+ on day 14 ($p = 0.002$), day 28 ($p = 0.001$), day 42 ($p = 0.003$) along with Cl^- on day 14 ($p = 0.001$), day 28 ($p = 0.001$), day 42 ($p = 0.003$) and day 56 ($p = 0.007$). In comparison with Strain A, Strain B had higher Na^+ concentration on day 14 ($p = 0.001$), 42 ($p = 0.001$), day 56 ($p = 0.002$), along with significantly higher blood Cl^- concentration on day 14 ($p = 0.001$), day 28 ($p = 0.003$) and day 42 ($p = 0.010$) but no treatments interaction was observed on any of the sampling days.

DISCUSSION

Previous studies demonstrated the effects of light sources on blood physiological variables[14] and in combination with light intensity[12], ambient temperature[15] and photoperiod[8]. Similar to our previous studies, the present results under ANOVA combined over days (1-56 days of age) indicate that light sources in combination with regular/intermittent photoperiod with or without strain had a minor effect on most of the selected blood physiological variables of broilers grown to heavy weights. However, treatments had effect on most of the selected blood physiological variables as related to strains in this study. In comparison with Strain A, Strain B had higher BW, pCO_2, HCO_{3-}, McHc, Ca^{2+}, Na^+, K^+, Cl^-, Angap, Osmo, T_3, along with significantly lower pH, pO_2, SaO_2, Hb and Hct under the same experimental conditions. Also, exposure of broilers to the photoperiod significantly affected BW and acid-base balance. Broilers reared under short photoperiod with and without light sources had significantly increased in levels of pCO_2, Hb, Hct and McHc along with significantly decreased

Table 3: Influence of light sources, photoperiods, strains, and their interaction on blood partial pressure of O_2 (pO_2) and saturated O_2 (SaO_2) of broilers grown to heavy weights*

	pO_2 (mmHg)				SaO_2 (%)			
Item	14 day	28 day	42 day	56 day	14 day	28 day	42 day	56 day
Light sources[1]								
ICD	49.360	51.830	46.580	47.400^{ab}	72.490	75.180	72.250^{ab}	71.840
CFL	48.400	51.500	44.400	49.770^{a}	73.360	74.610	73.520^{a}	70.790
Neutral-LED	50.110	52.310	45.460	44.730^{b}	73.680	75.530	71.470^{b}	70.410
Cool-PS-LED	48.690	51.900	46.680	43.690^{b}	71.740	74.100	71.320^{b}	70.180
Photoperiod[2]								
Reg-Inter	49.620	52.180	45.030	47.750^{a}	72.670	75.260	74.870^{a}	72.810^{a}
Short-non-inter	48.660	51.600	46.690	42.290^{b}	72.460	74.450	70.280^{b}	69.300^{b}
Pooled SEM	1.985	2.080	4.159	1.592	2.472	1.242	0.743	0.121
Strains								
A	50.610	51.570	46.780^{a}	47.240^{a}	74.023	74.820	74.580^{a}	72.170^{a}
B	47.660	52.200	44.780^{b}	44.060^{b}	71.107	74.890	70.690^{b}	70.900^{b}
Pooled SEM	1.404	1.471	0.594	1.046	1.748	0.878	1.291	0.422
Interaction								
ICD-Reg-inter-A	45.170	46.420	41.250^{ab}	45.670^{abc}	70.870	72.170	67.130^{ab}	69.510^{ab}
ICD-Reg-inter-B	48.090	55.090	50.250^{a}	46.830^{abc}	69.560	76.740	76.600^{a}	72.780^{ab}
ICD-short-non-inter-A	44.920	48.340	30.410^{b}	33.170^{c}	72.130	66.190	58.600^{b}	60.490^{b}
ICD-short-non-inter-B	54.750	53.090	46.910^{a}	50.170^{ab}	77.580	77.290	73.240^{a}	75.200^{a}
CFL-reg-inter-A	47.500	49.590	44.160^{ab}	39.920^{bc}	73.780	73.900	69.190^{ab}	67.830^{ab}
CFL-reg-inter-B	52.830	55.420	48.830^{a}	49.420^{ab}	74.130	78.180	74.240^{a}	74.050^{a}
CFL-short-non-inter-A	47.000	53.250	48.170^{a}	46.330^{abc}	72.400	76.340	73.290^{a}	71.050^{ab}
CFL-short-non-inter-B	48.670	53.250	50.250^{a}	42.830^{abc}	70.950	73.290	73.240^{a}	67.610^{ab}
Neutral-LED-Reg-inter-A	58.920	58.170	49.340^{a}	49.090^{ab}	78.910	79.680	74.190^{a}	72.320^{ab}
Neutral-LED-Reg-inter-B	45.250	47.670	45.500^{a}	48.080^{ab}	70.600	72.130	71.060^{a}	72.730^{ab}
Neutral-LED-ICD-short-non-inter-A	52.250	56.670	53.670^{a}	54.090^{a}	72.070	79.800	77.130^{a}	75.700^{a}
Neutral-LED-ICD-short-non-inter-B	47.670	52.920	46.580^{a}	49.670^{ab}	67.660	75.150	73.100^{a}	71.790^{ab}
Cool-PS-LED-Reg-inter-A	58.000	57.000	48.000^{a}	49.170^{ab}	78.580	78.140	73.660^{a}	73.990^{a}
Cool-PS-LED-Reg-inter-B	42.080	47.250	40.830^{ab}	40.420^{abc}	68.230	71.930	68.790^{ab}	65.800^{ab}
Cool-PS-LED-short-non-inter-A	51.170	48.160	43.250^{ab}	43.080^{abc}	73.460	72.310	71.790^{a}	70.490^{ab}
Cool-PS-LED-short-non-inter-B	47.920	52.920	45.080^{a}	42.500^{abc}	70.160	74.470	71.570^{a}	67.580^{ab}
Pooled SEM	3.971	4.159	2.812	2.736	4.943	2.485	2.267	2.543
Source of variation	p-value							
Light sources	0.931	0.994	0.626	0.019	0.973	0.932	0.043	0.262
Photoperiod	0.602	0.587	0.291	0.042	0.876	0.152	0.016	0.021
Strains	0.143	0.276	0.001	0.039	0.572	0.691	0.042	0.039
Interaction	0.449	0.191	0.046	0.006	0.961	0.056	0.032	0.047

$^{a-c}$Means within a column and effect that lack common superscripts differ significantly ($p \leq 0.05$), *Values are least squares of 2 replicate rooms with 60 birds per room, [1]Light Sources; 1: Incandescent light (ICD, Standard), 2: Compact fluorescent light (CFL), 3: light emitting diode (neutral-LED), 4: Poultry specific filtered LED (Cool-PSF-LED), [2]Photoperiod; Reg-Inter: Regular/intermittent (2L:2D), Short-Non-Inter: Short/non-intermittent (8L:16D)

levels of BW, pH, pO_2, SaO_2, Ca^{2+}, Na^+, K^+, Cl^-, Osmo and T_3, in comparison with birds reared under regular/intermittent photoperiod. However, all these acid-base changes are still within the normal acid-base homeostasis and within physiological ranges.

The acid-base status of poultry is challenged daily by environmental factors such as light program, temperature, humidity and air quality among others and they influence respiratory and metabolic activities[2,8,16-18]. The primary organ systems used in acid-base homeostasis in birds are the lungs and kidneys, supported by the gastrointestinal tract and cardiovascular system that participate in thermoregulatory processes through modulation of heat dissipation and oxygen transport[19]. The pH of the blood is maintained within a very narrow range because sudden changes can result in cellular damage via protein ionization, while the carbonic acid-bicarbonate system is the most important buffer for maintaining blood acid-base balance[20]. The association of CO_2 retention, which we observed under photoperiod and strains may be due to the rate of CO_2 elimination as related to BW. For instance, when the respiration rate decreased in strain B in the current study, the levels of pCO_2 in the blood increased and more H^+ ions accumulated, causing the pH of blood to decrease, thereby pushing the equilibrium reaction to the right and creating more hydrogen ions. This increased hydrogen ion concentration that lowered blood pH and thus

Table 4: Influence of light sources, photoperiods, strains, and their interaction on blood calcium (Ca^{2+}) and potassium (K^+) of broilers grown to heavy weights*

	Ca^{2+} (meq L^{-1})				K^+ (meq L^{-1})			
Item	14 day	28 day	42 day	56 day	14 day	28 day	42 day	56 day
Light sources[1]								
ICD	2.938	3.005	3.001	3.097[a]	3.712[a]	4.394[a]	4.481[a]	4.480
CFL	2.943	3.018	2.991	3.084[ab]	3.611[ab]	4.378[ab]	4.461[ab]	4.518
Neutral-LED	2.936	3.013	2.981	3.068[ab]	3.534[ab]	4.325[ab]	4.373[ab]	4.357
Cool-PS-LED	2.926	2.988	2.896	2.847[b]	3.343[b]	4.311[b]	4.356[b]	4.475
Photoperiod[2]								
Reg-Inter	2.954[a]	3.033[a]	3.006[a]	3.059[a]	3.789[a]	4.433[a]	4.473[a]	4.363
Short-Non-Inter	2.917[b]	2.978[b]	2.968[b]	3.016[b]	3.513[b]	4.325[b]	4.408[b]	4.551
Pooled SEM	0.011	0.011	0.009	0.013	0.055	0.030	0.018	0.102
Strains								
A	2.897[b]	2.946[b]	2.975[b]	3.018[b]	3.556[b]	4.331[b]	4.359[b]	4.418
B	2.974[a]	3.006[a]	3.013[a]	3.075[a]	3.646[a]	4.428[a]	4.461[a]	4.496
Pooled SEM	0.011	0.011	0.009	0.018	0.025	0.031	0.065	0.072
Interaction								
ICD-Reg-inter-A	2.895	2.908	2.985	3.128	3.325	4.150	4.063	4.323
ICD-Reg-inter-B	2.958	3.043	2.913	3.108	3.760	4.525	4.650	4.640
ICD-Short-Non-Inter-A	2.840	2.935	3.010	3.028	3.313	3.948	3.988	3.938
ICD-Short-Non-Inter-B	2.930	3.018	2.940	3.108	3.803	4.415	4.545	4.590
CFL-Reg-Inter-A	2.870	2.963	3.035	3.023	3.338	4.178	4.202	4.055
CFL-Reg-Inter-B	2.988	3.045	2.950	3.050	3.833	4.555	4.788	4.583
CFL-short-non-inter-A	2.848	2.940	3.015	3.050	3.438	4.333	4.313	4.535
CFL-short-non-inter-B	3.005	2.978	3.000	3.015	3.908	4.500	4.678	4.245
Neutral-LED-Reg-inter-A	2.918	3.008	3.000	3.018	3.810	4.640	4.792	4.815
Neutral-LED-Reg-inter-B	2.982	2.990	3.000	3.055	3.535	4.060	4.138	4.135
Neutral-LED-ICD-short-non-inter-A	3.035	3.143	3.033	3.045	4.298	5.075	5.218	4.940
Neutral-LED-ICD-short-non-inter-B	2.965	2.978	3.001	3.155	3.430	4.075	4.015	4.605
Cool-PS-LED-Reg-inter-A	2.890	3.065	2.990	3.005	4.028	4.723	4.663	4.720
Cool-PS-LED-Reg-inter-B	2.995	2.978	2.960	3.073	3.355	4.115	4.238	4.070
Cool-PS-LED-short-non-inter-A	2.878	3.023	2.990	3.085	3.703	4.378	4.438	4.641
Cool-PS-LED-short-non-inter-B	2.973	3.013	3.028	3.038	3.525	4.403	4.32	4.480
Pooled SEM	0.032	0.030	0.027	0.033	0.191	0.196	0.184	0.204
Source of variation	p-value							
Light sources	0.891	0.517	0.570	0.015	0.009	0.012	0.021	0.149
Photoperiod	0.020	0.001	0.024	0.023	0.024	0.027	0.045	0.779
Strains	0.001	0.009	0.007	0.016	0.012	0.001	0.044	0.263
Interaction	0.268	0.093	0.489	0.897	0.478	0.235	0.358	0.354

[a-c]Means within a column and effect that lack common superscripts differ significantly ($p<0.05$); *Values are least squares of 2 replicate rooms with 60 birds per room, [1]Light Sources; 1: Incandescent light (ICD, Standard), 2: Compact Fluorescent light (CFL), 3: Light emitting diode (neutral-LED), 4: Poultry specific LED (Cool-PS-LED), [2]Photoperiod; Reg-Inter: Regular/intermittent (2L:2D), Short-Non-Inter: Short/non-intermittent (8L:16D)

the blood acid-base status in this study reflected acute respiratory acidosis (significantly increased level of pCO_2, decreased levels of pH with or without change in HCO_{3^-}) caused by hydrogen ion (H^+, acid) accumulation[21]. Sometimes, acute respiratory acidosis results from hypoventilation (low in blood pH and high pCO_2) that may be due to loss of respiratory drive, body mass, chest wall capacity, or rapid shallow breathing but this can be quickly corrected through a compensatory mechanism[22]. This is based on the fact that 3 systems (buffer, respiratory and renal systems) function interdependently to regulate and maintain acid-base balance. These compensatory changes result in increased oxygen transport to the tissues, increased chemoreceptor pO_2 with a concomitant decrease in CO_2 and H^+, increased vascular resistance, increased lactate production and decreased workload capacity.

Reduced pO_2 and SaO_2, as we observed in these same group of birds may be due to inadequate blood oxygenation and hypoxemia, which increases the risk of hypoxia[23]. Modern broiler chickens are able to consume large quantities of feed and grow rapidly due to genetic selection, resulting in high demand for oxygen. However, when oxygen intake is low (low pO_2, SaO_2) relative to BW size, the heart essentially pushes the blood through the lungs with more pressure to increase the amount of oxygen available to the bird's metabolism[24]. Due to the fact that the lung volume and cardiovascular

Table 5: Influence of light sources, photoperiods, strains, and their interaction on blood sodium (Na^+) and chloride (Cl^-) of broilers grown to heavy weights*

	Na^+ (meq L^{-1})				Cl^- (meq L^{-1})			
Item	14 day	28 day	42 day	56 day	14 day	28 day	42 day	56 day
Light sources[1]								
ICD	147.710	149.580	153.380	155.980	104.060	105.96	109.8/3	111.530
CFL	148.230	150.040	153.370	156.370	104.650	106.33	109.940	111.710
Neutral-LED	147.600	150.170	152.980	154.400	103.870	106.38	109.960	110.750
Cool-PSF-LED	147.730	149.290	153.170	154.730	103.960	105.85	109.350	110.360
Photoperiod[2]								
Reg-inter	148.320[a]	150.770[a]	153.780[a]	155.830	104.700[a]	107.24[a]	110.500[a]	111.650[a]
Short-non-inter	147.310[b]	148.760[b]	152.670[b]	154.910	103.570[b]	105.12[b]	109.040[b]	110.520[b]
Pooled SEM	0.216	0.252	0.256	0.611	0.203	0.277	0.333	0.322
Strains								
A	147.260[b]	149.470	152.380[b]	154.570[b]	103.760[b]	106.190	109.140[b]	110.670
B	148.370[a]	150.070	154.070[a]	156.170[a]	104.510[a]	106.180	110.410[a]	111.510
Pooled SEM	0.216	0.252	0.254	0.432	0.203	0.277	0.333	0.644
Interaction								
ICD-Reg-inter-A	147.250	146.330	151.750	156.750	103.340	103.160	107.170	110.670
ICD-Reg-inter-B	148.250	151.330	154.590	156.170	104.590	107.080	111.420	110.830
ICD-short-non-inter-A	145.830	148.420	150.330	153.830	101.830	104.160	107.250	109.590
ICD-short-non-inter-B	147.330	149.090	154.670	156.750	104.250	106.000	110.830	111.750
CFL-reg-inter-A	146.170	148.080	151.340	153.000	101.920	104.500	107.670	109.250
CFL-reg-inter-B	148.340	150.250	153.840	153.920	104.920	106.080	110.250	112.000
CFL-short-non-inter-A	146.750	147.500	150.920	154.330	102.590	104.000	107.330	109.340
CFL-short-non-inter-B	148.580	149.170	153.920	153.920	105.170	106.000	110.420	110.250
Neutral-LED-reg-inter-A	147.000	150.250	153.250	154.840	104.090	107.330	110.500	112.670
Neutral-LED-reg-inter-B	148.330	150.420	153.920	156.170	104.250	106.250	110.250	111.960
Neutral-LED-ICD-short-non-inter-A	150.660	153.170	154.500	155.000	107.750	109.920	111.580	112.330
Neutral-LED-ICD-short-non-inter-B	149.080	149.500	154.000	159.920	104.750	105.250	110.080	113.170
Cool-PS-LED-reg-inter-A	147.080	151.830	152.840	154.170	104.500	109.330	111.000	110.920
Cool-PS-LED-reg-inter-B	148.830	150.500	153.920	156.500	104.170	106.420	110.920	110.830
Cool-PS-LED-short-non-inter-A	147.330	150.170	154.080	151.500	104.080	107.080	110.590	110.080
Cool-PS-LED-short-non-inter-B	148.240	150.330	153.750	156.000	104.000	106.330	109.080	111.250
Pooled SEM	0.610	0.712	0.717	1.228	0.573	0.784	0.942	0.911
Source of variation	p-value							
Light sources	0.479	0.285	1.839	0.076	0.232	0.533	0.781	0.130
Photoperiod	0.002	0.001	0.003	0.085	0.001	0.001	0.003	0.007
Strains	0.001	0.096	0.001	0.012	0.001	0.003	0.010	0.072
Interaction	0.241	0.374	0.368	0.998	0.075	0.412	0.814	0.563

[a-c]Means within a column and effect that lack common superscripts differ significantly ($p \leq 0.05$); *Values are least squares of 2 replicate rooms with 60 birds per room, [1]Light sources; 1: Incandescent light (ICD, Standard), 2: Compact Fluorescent light (CFL), 3: Light emitting diode (Neutral-LED), 4: Poultry specific LED (Cool-PS-LED), [2]Photoperiod; Reg-Inter: Regular/intermittent (2L:2D), Short-Non-Inter: Short/non-intermittent (8L:16D)

volume within the lung tissue is fixed in birds, unlike in mammals, eventually a point may be reached whereby the lungs may no longer accommodate more blood being supplied by the heart and this may have negative effects on the body due to poor oxygenation[25]. Disturbances in venous blood acid-base status (pCO_2 and pH) are frequently observed in older birds due to age-dependent differences in ventilation rate or may reflect the consequences of an increased metabolic demand in the larger birds[26].

Body fluid electrolyte concentrations, such as Na^+, K^+ and Cl^- and acid-base balance are interconnected and are also associated with the condition producing acidosis or alkalosis in mammals, which may be true in birds[27]. In the present study, we observed significantly higher levels of Ca^{2+}, Na^+, K^+ and Cl^- in strain B in comparison to strain A. The decrease in Na^+ concentration in birds reared under short photoperiod might be related to concurrent loss of Na^+ due to decreased extracellular fluid volume. In addition, decrease Cl^- concentration may be interpreted to be secondary to a shift of Na^+. Respiratory acidosis is closely linked to Cl^- depletion, which leads to an increased reabsorption of HCO_3^- in the distal renal tubule in humans[28] and may also be possible in avians. In addition, changes in pH can alter the amount of Ca^{2+} bound by proteins during respiratory acidosis or metabolic alkalosis, which in turn, alters ionized Ca^{2+} levels leading to very low blood Ca^{2+}. Changes in blood Osmo observed in the present study may be associated with the higher blood Na^+ concentration, which is characterized by water shift from intra-

cellular fluid to extra-cellular fluid as reported by Freda *et al.*[29]. In addition, Na^+ is the main determinant of plasma osmolality and water moves toward body compartments with higher osmolality[29]. The pH of blood affects the distribution of ions throughout the body and changes in pH may be associated with changes in ion concentration, which may have effects on body systems.

The plasma T_3 levels of birds reared under the three light-sources were significantly higher in comparison with those reared under ICD light. Also, plasma T_3 level of broilers reared under the regular/intermittent (2L:2D) in the present study was significantly higher than those of broilers reared under the short/non-intermittent (8L:16D) photoperiods. Furthermore, the plasma T_3 level of strain B in the present study was significantly higher, whereas T_4 concentration was opposite that of strain A. Thyroid hormones especially tri-iodothyronine (T_3) and thyroxine (T_4) are the most important humoral factors that influence all major metabolic pathways of protein, carbohydrates and lipid metabolism and are involved in the regulation of the basal metabolism of the majority of tissues including liver, heart, kidney and brain among others[30]. It has been documented that T_3 is the main physiological thyroid hormone regulating oxygen consumption and daily activities, particularly in young chickens and is metabolically more active than T_4. It has been reported[31] that the T_3 hormone is closely associated with feeding and is also a key factor influencing conversion of T_4 to T_3 and that a higher T_3 level is associated with increased protein deposition. It is generally believed that T_4 is the predominant thyroid hormone in circulation but it has little biological activity in comparison with physiological T_3 that has more metabolically active activities[32]. Therefore the high level of T_3 in birds reared under new light sources, regular/intermittent photoperiod and strain B group may relate to feed intake during the light period and significant differences in their BW in Table 1 that support the classical role of the thyroid hormones in metabolic and physiological activities on growth and development. Concentrations of glucose and corticosterone among others have been suggested to be sensitive indicators of physiological responses in stressed broiler chickens[33,34]. In this study, there were no effects of treatments on blood glucose and plasma corticosterone concentrations, suggesting that these treatments and their interaction did not present stressors to broilers grown to heavy weights.

In summary, specific main effects of light sources in combination with photoperiods on selected blood physiological variables were observed on these two major broiler strains grown to heavy weights. It follows that the blood oxygen-carrying capacity of strain B assessed by pO_2 and SaO_2 values, was lower in comparison to strain A. However, all these acid-base changes are still within the normal acid-base homeostasis and physiological ranges. Furthermore, the results of this study supplement current knowledge of the blood gases, electrolytes, and metabolites of early- and late-developing broiler strains grown to heavy weights under environmentally controlled conditions. This data may be used to establish the normal blood values for these two commercial broilers strains.

CONCLUSION

In conclusion, the three light source bulbs evaluated in this study along with a regular/intermittent photoperiod in commercial poultry facilities would reduce energy costs and optimize production efficiency without compromising the welfare of broilers grown to heavy weights irrespective of strains.

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