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Jill R. Nelson
Texas A & M University - College Station

Joey L. Bray
Stephen F Austin State University, jbray@sfasu.edu

Juliette Delabbio
ONCE Innovations Inc.

Gregory S. Archer
Texas A & M University - College Station

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Light emitting diode (LED) color and broiler growth: effect of supplementing blue/green LED to white LED light on broiler growth, stress, and welfare

Jill R. Nelson,* Joey L. Bray,† Juliette Delabbio,‡ and Gregory S. Archer*,[1]

*Department of Poultry Science, Texas A&M University, College Station, USA; †Department of Agriculture, Stephen F. Austin State University, Nacogdoches, TX, USA; and ‡ONCE Innovations Inc., Plymouth, MN, USA

ABSTRACT Light emitting diode (**LED**) lighting provides an affordable lighting option for use in commercial poultry production. However, more information is needed to understand the effects of LED color on broiler welfare and growth. Five consecutive flocks (1 in summer, 1 in fall, 2 in winter, and 1 in spring) of straight run Ross 708 × Ross 708 broilers were reared in commercial type barns for 45 D. For white only (**WO**) treatment, birds were reared under white LED only (Agrishift MLB). For white supplemented (**WS**) treatment, birds were reared under white LED (Agrishift MLB) in the center aisle, with supplemental blue/green LED lighting (Agrishift MLBg) above the feed and water lines on either side of the barn. Each barn housed 26,200 chicks, and there were 2 barns in each treatment (n = 52,400/treatment). Treatments were rotated among barns between each flock. On day 45, blood samples were collected from 20 birds/barn (n = 40/treatment) to assess the plasma corticosterone (CORT) level and heterophil/lymphocyte ratio. On day 45, 100 birds/barn (n = 200/treatment) were weighed individually and assigned scores for hock burn and foot pad dermatitis. All measures were affected by trial ($P < 0.001$). Plasma CORT and body weight were affected by the treatment × trial interaction ($P \leq 0.001$). Overall, birds in the WS treatment had higher day 45 live body weight ($P < 0.001$) and lower hock burn scores ($P = 0.032$) than birds in the WO treatment. Birds in the WS treatment had higher day 45 body weight overall ($P < 0.001$) and in trials 1, 3, and 5, although the overall body weight was lower in trials 1, 3, and 5 than in trials 2 and 4. Supplemental blue/green LED improved hock burn scores and increased overall day 45 body weight. However, trial differences suggest that more data are needed to determine whether supplementing blue/green LED to white LED improves body weight gain in mixed sex broiler chickens.

Key words: broiler, light emitting diode, growth, welfare, stress

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INTRODUCTION

Light source, spectrum, intensity, and photoperiod are important components of broiler housing which can affect economically significant performance traits (Andrews and Zimmermann, 1990). Growing interest in alternatives to inefficient light sources traditionally used in broiler houses has resulted in increased research on the effects of monochromatic light emitting diode (**LED**) lights on poultry growth, stress, and welfare. Environmental stressors can stimulate production of corticosterone (**CORT**), an indicator of short-term stress, by the adrenal cortex in order to increase readily available energy (Wodzika-Tomaszewska et al., 1982). However, increased CORT secretion for extended periods can depress growth rate and immune response (Virden and Kidd, 2009). Glucocorticoid-induced hypotrophy of the lymphoid organs results in a higher heterophil/lymphocyte (**H/L**) ratio, an indicator of long-term stress (Gross and Siegel, 1983). LED light has been shown to reduce measures of fear and stress in broilers compared to other light sources (Huth and Archer, 2015). More specifically, blue or blue/green LED have shown potential for reducing measures of stress and fearfulness in ducks (Sultana et al., 2013) and broilers (Archer, 2018; Archer and Byrd, 2018).

Different colors of monochromatic light from LED bulbs have also been shown to affect growth performance. Notably, Rozenboim et al. (1999) reported an

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[1] Corresponding author: garcher@poultry.tamu.edu

early and sustained increase in growth in broilers reared under green light, and a late increase in growth in broilers reared under blue light compared to birds reared under red or white light. These changes were accompanied by increases in plasma testosterone levels (Rozenboim et al., 1999). Similar results were reported by Cao et al. (2008a), who also showed that myofiber size of the pectoralis major was higher early on (day 0–26) in broilers reared under green light, and later (day 27–49) in birds reared under blue light compared to birds reared under red or white light. Birds reared under blue or green light have also demonstrated increased satellite cell density in the breast muscle as well as a 1.6-fold increase in expression of the growth hormone receptor over birds reared under red or white light (Halevy et al., 1998). Furthermore, broilers reared under green light have shown increased melatonin-mediated expression of growth hormone releasing hormone in the hypothalamus and, subsequently, increased plasma growth hormone levels (Zhang et al., 2016). Light-induced mammalian target of rapamycin (**mTOR**) activity in the brain plays a role in entrainment of the circadian rhythm (Cao et al., 2008b). mTOR is also involved in metabolic homeostasis (Khapre et al., 2014); more specifically, mTOR stimulates protein synthesis in response to testosterone, resulting in muscle hypertrophy (Basualto-Alarcón et al., 2013; White et al., 2013). As such, green and blue light may alter mTOR activity in the brain, thus affecting signaling to the hypothalamus, production of growth hormone and testosterone, and muscle growth compared to birds reared under red or white light (Lewis and Morris, 2000).

It is important to consider optimizing lighting conditions in order to reduce stress and its effects on animal welfare and production outcomes. Although previous research has primarily focused on comparing the effect of a single color of LED light to another, one study has shown that rearing birds under 2 colors rather than a single color of monochromatic light may improve feed conversion (Yang et al., 2016). In light of the effects of blue and green light on growth performance, this research aimed to compare the effect of providing additional blue and green light to white, or full-spectrum, light on commercial broiler performance. Therefore, the objective of this study was to determine whether supplementing blue and green LED to white LED improves measures of stress, welfare, and growth performance in mixed sex commercial broilers reared to 45 D of age compared to providing white LED only.

MATERIALS AND METHODS

Animal Rearing and Lighting Treatments

Day-of-hatch straight-run Ross 708 × Ross 708 broiler chicks were reared to 45 D of age in solid-side wall, tunnel-ventilated barns measuring 13.1 × 152.4 m at the Stephen F. Austin State University Broiler Research Center in Nacogdoches, TX. There were 5 consecutive trials consisting of 104,800 birds each. Trial 1 was reared in summer, trial 2 was reared in fall, trials 3 and 4 were reared in winter, and trial 5 was reared in spring. Birds were placed in one of 4 barns (n = 26,200/barn), resulting in a space allowance of 0.25 m^2/bird. Birds were cared for in accordance with the Guide for the Care and Use of Agricultural Animals for Use in Research and Teaching (FASS, 2012) and protocols were approved by the Stephen F. Austin State University Institutional Animal Care and Use Committee (AUP#2016-004). Birds in both treatments received the same diet in all 5 trials, and they had ad libitum access to feed and water for the duration of the rearing period. Diets consisted of a crumbled starter (day 1–18), pelleted grower (day 19–35), pelleted finisher phase 1 (day 36–42), and pelleted finisher phase 2 (day 43–45). Each barn was fitted with 2 feeder lines with 428 feeders each, allowing for approximately 61 birds/feeder, and 4 drinker lines with 572 nipples each, allowing for approximately 11 birds/nipple. Lights were placed in 3 aisles across the width of each barn and at 6.1 m intervals along the length of the barn. For white only (**WO**) treatment, birds were reared under white LED only (Agrishift MLB; Once Inc., Plymouth, MN). For white supplemented (**WS**) treatment, birds were reared under white LED (Agrishift MLB; Once Inc.) in the center aisle and received supplemental blue/green LED lighting (Agrishift MLBg; Once Inc.) above the feed and water lines on either side of the barn. Two barns were assigned to each treatment, and treatments were rotated among the barns between each trial. The spectrum output for each light source is presented in Figure 1.

Stress Measures

On day 45, blood samples were collected via the brachial vein from 20 birds/barn (n = 40/treatment): 1 to 2 mL of blood was collected from each bird and transferred to a lithium heparin separation gel vacutainer (367884; BD Medical, Franklin Lakes, NJ). One drop of blood from each sample was used to prepare a blood smear slide. Vacutainers were then inverted and stored on ice. Blood samples were centrifuged at 4000 rpm for 15 min (Centrifuge 5804; Eppendorf, Hamburg, Germany), and plasma was poured off into a microcentrifuge tube and stored at −20°C until analysis. Plasma was thawed overnight at 4°C and used to assess CORT using a commercially available ELISA kit (ADI-901-097; Enzo Life Sciences, Inc., Farmingdale, NY). Absorbance was read at 450 nm using a microplate absorbance reader (Tecan Sunrise; Tecan Trading AG, Männedorf, Switzerland) and analyzed using the Magellan Tracker software program (Tecan Trading AG). Dry blood smear slides were stained with a neat stain hematology stain kit (Cat. #25034; Polysciences, Inc., Warrington, PA). Heterophil/lymphocyte ratio was determined at 40× magnification under light microscopy (89,404-886; VWR International, Radnor, PA) by counting individual heterophils and lymphocytes up to a total of 100 cells/slide. Blood was collected from a different

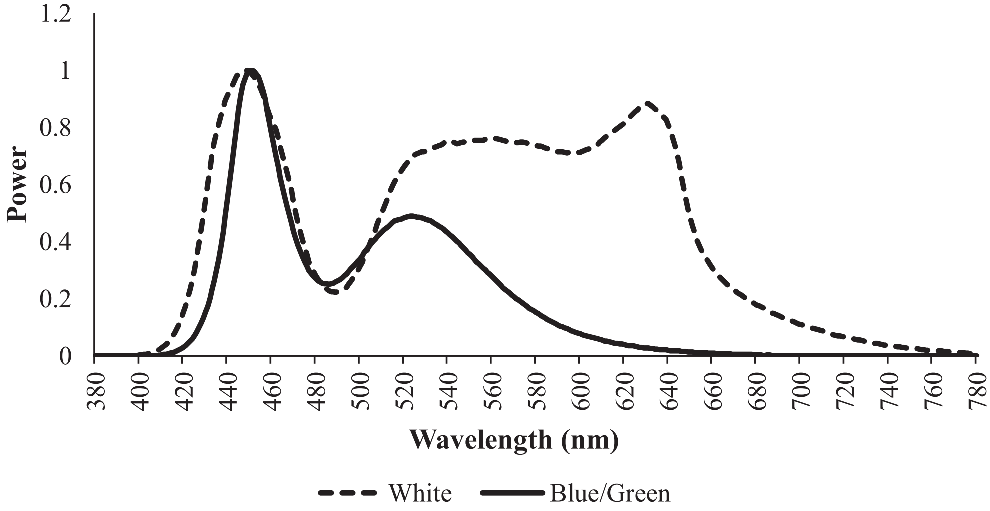

Figure 1. Spectrum readings for the white and blue/green light used in this study. In one treatment, only white light (WO) was provided. In the other treatment, white light was provided in the center aisle and supplemental blue/green light (WS) was provided above the feed and water lines on either side of the barn.

subset of birds than that which was used to obtain body weight and welfare measures.

Welfare Assessment

Welfare assessment was performed on 100 birds/barn (n = 200/treatment) on day 45 according to the procedures outlined by Arnould et al. (2009) and Welfare Quality (2009). Twenty birds were assessed in each of five 30.5-m sections of each barn and assigned scores for hock burn and foot pad dermatitis. Hock burn and foot pad dermatitis were both scored on a scale of 0 to 4, where a score of 0 indicated normal coloration and no visible lesions, a score of 1 indicated normal coloration and one lesion less than 0.5 cm in width, a score of 2 indicated one lesion 0.5 to 1.0 cm in width, a score of 3 indicated discoloration and one or more lesions larger than 1.0 cm in total width, and a score of 4 indicated severe discoloration and multiple severe lesions more than 1.0 cm in total width (Welfare Quality, 2009). Welfare assessment was performed on the same subset of birds which was used to obtain body weight measurements.

Body Weight

Live body weight was determined for 100 birds/barn (n = 200/treatment) on day 45. Twenty birds from each of five 30.5-m sections of each barn were weighed on a hanging scale with shackles (BW-2050; Weltech International, Ltd., Cambridgeshire, England).

Statistical Analysis

Ordinal data from the welfare assessments were analyzed for main effects of treatment and trial on the median in Minitab 17.1.0 (Minitab, LLC, State College, PA) by using Kruskal-Wallis test, adjusted for ties. Data for body weight, plasma CORT, and H/L ratio were analyzed for the main effects of treatment (WO or WS), trial (1–5), and treatment × trial interaction using the GLM procedure in Minitab 17.1.0 followed by mean separation using Fisher's least significant difference test, with individual birds as the experimental unit. Normality was confirmed by using the Shapiro-Wilk test and homogeneity of variances was confirmed by using Levene's test. All assumptions for ANOVA were met without transformation of the data. A significant difference was defined as $P \leq 0.05$.

RESULTS

Data for day 45 live body weight, plasma CORT, H/L ratio, and hock burn and foot pad dermatitis scores are shown in Table 1. There was a main effect of treatment on day 45 live body weight ($P < 0.001$), where birds in WS treatment weighed more on day 45 than birds in WO treatment. There was also a trial effect ($P < 0.001$) and an interaction effect ($P < 0.001$) on live body weight. Body weight was highest in trials 2 and 4 and lowest in trials 3 and 5. Live body weight was highest in WS treatment in trial 1, and lowest in WO treatment in trial 1 and in both treatments in trials 3 and 5.

There was a main effect of trial on plasma CORT ($P < 0.001$), which was higher in trial 5 compared to all other trials. The H/L ratio ($P = 0.001$), which was highest in trial 1 and lowest in trial 5, depended on the trial. There was also an interaction effect on H/L ratio, which was highest in birds in the WO treatment in trials 1 and 4. H/L ratio was lowest in the WO treatment in trial 2 and in both treatments in trial 5. Hock burn score was affected by both treatment ($P = 0.032$) and trial ($P < 0.001$). The WS treatment had lower overall hock burn scores than those in the WO treatment ($P = 0.032$). Hock burn scores were lowest in trial 3. There was an effect of trial ($P < 0.001$), but not treatment ($P = 0.482$), on foot pad dermatitis scores.

Table 1. Main effects of treatment and trial, and the treatment × trial interaction on day 45 live body weight, and stress and welfare measures for broilers.[1]

	Live body weight	Plasma CORT	H/L ratio	Hock burn	Foot pad dermatitis
Units	kg	pg/mL		0–4	0–4
Treatment × trial					
WO1	2.59^{g}	1272.72	0.95^{a}	0.45	1.24
WS1	3.11^{a}	1745.85	$0.91^{a,b}$	0.69	1.47
WO2	$3.05^{a,b}$	1284.11	0.25^{d}	0.94	0.76
WS2	$2.94^{d,e}$	2778.12	$0.68^{b,c}$	0.24	0.48
WO3	2.70^{f}	808.66	0.60^{c}	0.32	0.81
WS3	2.88^{e}	791.91	0.58^{c}	0.26	0.60
WO4	$3.03^{b,c}$	275.26	0.99^{a}	0.42	1.32
WS4	$2.96^{c,d}$	56.55	0.56^{c}	0.43	1.13
WO5	2.70^{f}	9211.80	0.27^{d}	0.41	1.02
WS5	2.88^{e}	10574.35	0.26^{d}	0.55	1.34
Pooled SEM	0.01	407.22	0.03	0.01	0.03
P-value treatment × trial	<0.001	0.925	0.001		
Main effect treatment					
WO	2.81^{b}	2570.51	0.62	0.51^{a}	1.03
WS	2.96^{a}	3189.36	0.60	0.42^{b}	1.00
P-value treatment	<0.001	0.403	0.788	0.032	0.482
Main effect trial					
1	2.85^{b}	1509.28^{b}	0.93^{a}	0.57	1.35
2	2.99^{a}	2031.11^{b}	0.47^{c}	0.59	0.62
3	2.79^{c}	800.28^{b}	$0.59^{b,c}$	0.29	0.70
4	2.99^{a}	165.90^{b}	$0.78^{a,b}$	0.42	1.23
5	2.79^{c}	9893.08^{a}	0.27^{d}	0.48	1.18
P-value trial	<0.001	<0.001	<0.001	<0.001	<0.001

$^{a–g}$Values within a column with different superscripts differ ($P < 0.05$).

Abbreviations: CORT, corticosterone; H/L, heterophil/lymphocyte; LED, light emitting diode.

[1] Parameters include treatment (WO or WS) and trial (1–5) (e.g., WO1 indicates WO treatment in trial 1, and so on). Treatment WO indicates birds reared under white LED only, whereas WS treatment indicates birds reared under white LED light in the center aisle only and blue/green LED on either side of the barn, above the feed and water lines.

DISCUSSION

Plasma CORT is a useful measure of short-term stress (Siegel, 1995), and responds to environmental stressors such as vaccination, crowding, acute heat stress, and short-term feed and water withdrawal (Nelson et al., 2018). Because blue and green light have been shown to reduce fearfulness in Pekin ducks (Sultana et al., 2013), it is presumed that plasma CORT would be lower in birds reared under supplemental blue/green light. However, similar to the results reported by Olanrewaju et al. (2015), plasma CORT did not differ between birds reared under only white LED (WO) and those reared under white light supplemented with blue/green LED (WS). Nevertheless, birds in trials 1, 2, and 5 had plasma CORT levels indicative of a stressed condition (Thaxton et al., 2005). Moreover, plasma CORT was significantly higher in trial 5 than in trials 1 to 4. It appears that in this study plasma CORT levels were more dependent on climatic conditions. Trial 5 was reared in spring, when the Texas climate experiences an increase in heat and humidity. Blue light has been shown to reduce measures of oxidative stress in heat-stressed broilers (Abdo et al., 2017). Because plasma CORT also increases under heat stress (Nelson et al., 2018) and increased plasma CORT can induce oxidative stress (Lin et al., 2004), it would be expected that birds in the WS treatment would have lower plasma CORT concentrations compared to the WO treatment, even in warmer temperatures. However, birds in trial 5 had higher plasma CORT levels compared to all other trials regardless of light treatment. Compared to other trials which were reared at the end of summer or during winter, the birds in trial 5 may have had difficulty in acclimating to changes in weather patterns.

Heterophil/lymphocyte ratio is commonly used to measure long-term stress: increased stress typically results in a higher ratio of heterophils to lymphocytes (Gross and Siegel, 1983). Rearing broilers under a combination of blue and green monochromatic light has been shown to stimulate lymphocyte proliferation compared to birds reared under a single color of light, potentially reducing the effects of stress on the immune system (Zhang et al., 2014). Therefore, it was anticipated that birds in the WS treatment would have lower H/L ratios across all trials. However, there was no such trend: the H/L ratio was only observed to be lower in the WS treatment in trial 4. Unlike previous studies in which both plasma CORT and H/L ratio increased concurrently in response to stress (Nelson et al., 2018), H/L ratio was lowest in trial 5, which had the highest plasma CORT concentration. Because H/L ratio is often used to measure long-term stress, and there was no effect of treatment on H/L ratio, differences in H/L ratio may be more indicative of long-term effects of climate or other environmental conditions rather than lighting treatment on broiler stress in this study. Thus, overall, neither light treatment significantly influenced measures of long- or short-term stress in this study.

Ducks reared under blue light have been observed to spend more time sitting, standing, and preening, and less time socializing, foraging, and walking than those reared under white light (Sultana et al., 2013). Thus, birds in the WO treatment were expected to have lower hock burn and foot pad dermatitis scores than birds given supplemental blue/green light. However, overall birds in the WS treatment had lower hock burn scores than those in the WO treatment. Although there were no treatment differences in foot pad dermatitis scores, both hock burn and foot pad dermatitis scores were affected by trial. In other words, hock burn scores tended to be higher (more severe) during summer (trial 1) and fall (trial 2), whereas foot pad dermatitis scores were higher (more severe) in summer (trial 1), late winter (trial 4), and early spring (trial 5). Rather than the result of a change in activity under different light treatments, trial effects on welfare measures may be the result of climate. To substantiate, measures to control barn temperature may have resulted in increased humidity and, therefore, increased contact with wet litter, leading to increased incidence or severity of hock burn and foot pad dermatitis (Haslam et al., 2007).

In some studies, rearing birds under cool LED has been shown to increase body weight and carcass yield, probably due to testosterone-induced muscle hypertrophy (Olanrewaju et al., 2016; 2018). In this study, birds reared under white LED supplemented with blue/green light had higher overall day 45 live body weight than birds reared under only white LED light. In addition, birds in the WS treatment were heavier than WO birds on day 45 in trials 1, 3, and 5. However, trial differences indicate that overall body weight was lower in trials 1, 3, and 5 compared to that in trials 2 and 4, suggesting that other factors besides lighting treatment alone may have contributed to differences in day 45 body weight. Other studies have reported no effect of light wavelength on body weight, feed conversion, or carcass yield (de Santana et al., 2014; Olanrewaju et al., 2015). In addition, climate, rather than light treatment, may have been more important in determining the trial differences observed in stress and welfare measures. Therefore, more data are needed to determine whether supplementing blue/green LED to white LED is effective in improving day 45 body weight in mixed sex broilers.

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