Light wavelength and its impact on broiler health

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ABSTRACT Light is a powerful management tool in poultry production systems, affecting productivity, physiology, and behavior. The objective of this study was to understand the impacts of three light colors (blue, green, or white) on broiler health. Broilers (N = 14,256) were raised in floor pens with fresh litter from 0 to 35 d in 9 rooms (2 blocked trials). Additionally, 2 genotypes (Ross YPMx708 and EPMx708) and sex were studied (6 room replications per lighting treatment and 18 pen replicates per sex × genotype × lighting program). Blood samples and tissue samples from the retina and the pineal gland were collected from birds (16−18 d of age) 9 times in one 24-hr period per trial, then analyzed to determine melatonin levels (pg/mL). Mobility was assessed via gait scoring, using a 0 to 5 scale at 31 to 32 d of age. Footpad dermatitis was assessed using a 0 to 4 scale, and litter quality by a subjective scoring system (scores ranging from 0−4). Mortality and morbidity causes were identified through necropsies performed by pathologists. Data were analyzed as a 3 × 2 × 2 factorial design, with trial as a random variable block and lighting treatment nested within rooms (MIXED procedure, SAS). Birds raised under blue light had lower serum melatonin levels during one time-point during the scotophase, but no other differences were noted. No effect of light color was observed for melatonin produced in the tissues, nor mobility and footpad dermatitis. An interaction was noted for litter quality where a higher percentage of pens housing YPM-708 broilers had litter categorized into dry, but not easily moved with the foot (category 1). Males had higher incidence of infectious and metabolic deaths than females. Interactions were observed between light and sex, where males raised under white light had a higher incidence of skeletal causes of mortality. Overall, the results showed that light color had minor impacts only on melatonin levels, mobility, footpad dermatitis, litter quality, and cause of mortality.

Key words: wavelength, light color, broiler, melatonin, welfare

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INTRODUCTION

Management of lighting programs in broiler production has significant influences on many aspects of bird lives, including production, physiology, welfare, and behavior. The use of light-emitting diode (**LED**) light bulbs as a light source has become an area of interest due to various benefits, such as increased bulb life span and low energy consumption (Parvin et al., 2014). LED bulbs can also provide monochromatic light colors, a topic that has been the object of numerous studies over the past decade. The impact on production has been one of the primary focus in previous research. For example, research has been conducted to understand the impacts of red light on the productivity of laying hens (Huber-Eicher et al., 2013; Li et al., 2014), and the impacts of short wavelengths treatments, such as blue and green light, on broiler weight gain and meat yield (Wathes et al., 1982; Prayitno et al., 1997; Rozenboim et al., 1999; Cao et al., 2008; Ke et al., 2011; Mohamed et al., 2014). However, when assessing animal welfare, health is an important parameter to consider (Fraser, 2008). Therefore, it is essential to understand the effects of light wavelength on poultry physiology and health.

Diurnal rhythms are included in a series of behaviors or physiological events that occur approximately every 24 hr. They are primarily entrained by regular exposure to light and darkness (Duffy and Czeisler, 2009) and are regulated by the neurohormone melatonin. Melatonin is produced in a cyclic fashion, with production increasing during the scotophase and decreasing during the photophase (Tähkämö et al., 2019). Thus, light exposure,

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which is frequently manipulated in poultry production, is an important regulator of melatonin secretion.

The mechanisms by which light wavelength influences melatonin synthesis are not completely understood. Various wavelengths possess distinctive capabilities of penetrating the skull of birds, which potentially could impact the amount of light reaching sections of the brain involved in control of diurnal rhythms, such as the pineal gland and the hypothalamus (Lewis and Morris, 2000). Light composed of short wavelengths, such as those that result in blue light, can activate melanopsin-producing cells, leading to suppression of melatonin synthesis (Alkozi, 2019). Changes in melatonin levels lead to impacts on biological cycles entrained by this neurohormone, such as control of hormonal levels, body temperature, quality and duration of sleep, and affective states (Wahl et al., 2019).

Changes in diurnal rhythms can be linked to additional effects, such as variations in behavioral expression (Schwean-Lardner et al., 2014). These behavioral changes may affect other common broiler welfare issues. Broilers that show a significant reduction in activity levels may exhibit a higher incidence of leg disorders (Bradshaw et al., 2002). Litter quality may be decreased, due to a reduced frequency of turnover of the litter due to a decrease in exercise (Kristensen et al., 2004). Poor litter quality may lead to an increase in footpad dermatitis (**FPD**) (Dunlop et al., 2016). The effect of light duration on timing of body weight gain throughout the production cycle, which could influence the incidence of leg disorders, has been the focus of previous research (Schwean-Lardner et al., 2013), however, research studying the relationship between light wavelength and production have been inconclusive (Wathes et al., 1982; Prayitno et al., 1997; Rozenboim et al., 1999; Cao et al., 2008).

Wavelength of light may also affect broiler immune function. In a study comparing blue, green, red, and white lights, broilers raised under short wavelengths (blue and green) had higher T-lymphocyte proliferation and greater antibody production, hence improved immune function, when compared to broilers raised under longer (red) wavelengths (Xie et al., 2008). This suggests that light color could affect mortality, however, contradictions exist in the literature. According to Sadrzadeh et al. (2011), T-lymphocyte proliferation was increased when broilers were raised under white light, as compared to blue and green light. In another study, Kim et al. (2013) reported that light color (white, blue, red, green, and yellow lights) had no direct impact on broilers immune function.

To further understand the different impacts of light wavelength on broiler health, our objective was to assess melatonin levels, bird mobility, footpad dermatitis, litter quality, and causes of mortality when broilers were raised under different light regimes. This study is a part of a more extensive research project, focusing on the impact of light color, genotype, and sex on broiler production, behavior, and physiology.

MATERIALS AND METHODS

This experiment was approved by the Animal Care Committee of the University of Saskatchewan. It was conducted following the guidelines of the Canadian Council on Animal Care (2009) as specified in the Guide to the Care and Use of Experimental Animals.

Housing and Management

The experiment, conducted in two trials, studied the effect of light wavelength, sex, and genotype (Ross EPMx708 and Ross YPMx708) on health parameters. For each trial, 7,128 broilers (N = 14,256) were raised from d 0 to 35. Broilers were housed in nine individually controlled rooms for the distribution of light color (3 room replications per wavelength treatment per trial). Each room was subdivided into 12 pens (2 × 2.3 m each) to distribute sex and genotype. Final density in each pen was calculated based on predicted weight at 35 d (31 kg/m^2; 62 males or 70 females per pen).

Wheat straw was used as litter material and was evenly distributed in the pens (depth of 7.5−10 cm). Water and commercial feed (starter [0.5 kg per bird], grower [2 kg per bird] and finisher [remainder]) were provided ad libitum. Birds were fed using aluminum tube feeders (110 cm of pan circumference from 0 to 30 d and 137.5 cm circumference from 30 d to market), and water was provided using pendulum nipple drinkers, with 6 nipples per pen. Diets were formulated based on Aviagen's Ross 708 recommendations (Aviagen, 2019). Supplemental feeders and drinkers were provided during wk 1.

Heat was delivered to rooms via hot-water pipes running along the walls. Room temperature was 32.1°C on d 0, gradually reduced (approximately 0.5°C daily) until reaching 21°C by 25 d and was maintained at this temperature for the remainder of the trial. To provide the recommended 40 to 60% relative humidity during the early brooding period, humidifiers were added to each room at the time of chick placement and removed by d 4. Temperature and humidity were monitored daily via computer system and by observations of behavioral thermal comfort indicators.

Lighting

LED bulbs (11W Alice Non-Directional LED Lamps, Greengage Agritech Limited, Roslin Innovation Centre, University of Edinburgh, Easter Bush Campus, Midlothian, EH25 9RG, United Kingdom) provided specific wavelength treatments in each room (3 rooms per light color). The wavelength treatments included blue (peak at 455 nm), green (peak at 510 nm) or white light (combination of wavelengths). Light spectrum in each room was assessed to confirm the spectral outcome of each lamp (Figure 1) using a light meter (Lighting Passport, Asensetek Incorporation, New Taipei City, Taiwan).

Each lighting program started at 23L:1D on d 0, and the duration of the photoperiod decreased 1 hr daily,

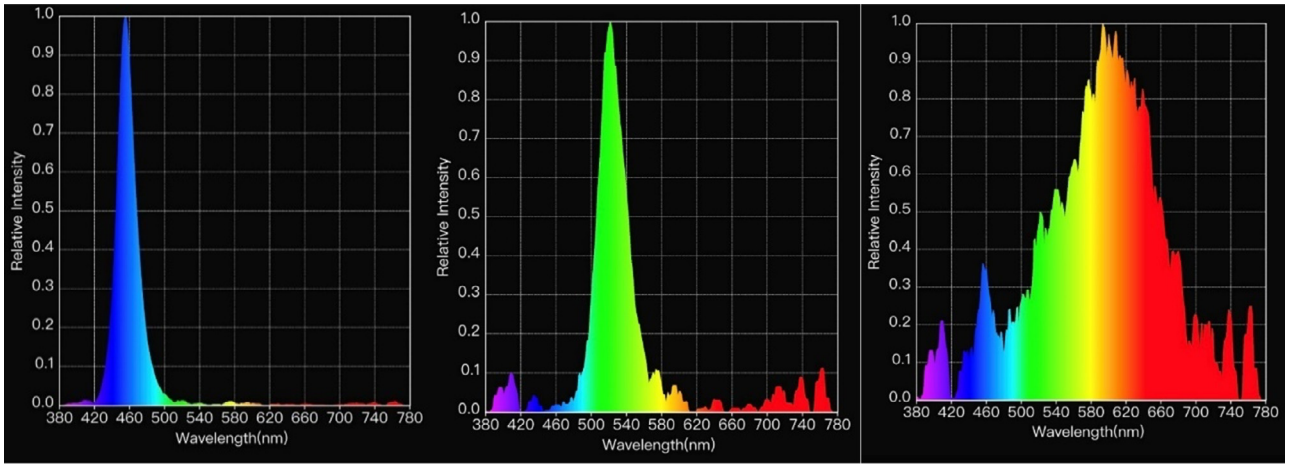

Figure 1. Measurements of light spectrum from blue, green, and white rooms, respectively.

reaching 18L:6D by d 5. Dawn and dusk periods of 15 min were provided daily before lights were fully on or off, which was included in the photophase time period. Light intensity was assessed in galilux or clux, which is based on the specific spectral sensitivity of birds within each wavelength. For trial 1, light intensity was 9.6 ± 0.4 clux for the entire period. For trial 2, additional lighting was added to allow the intensity for the first wk to reach 14.3 ± 0.1 clux and the remaining weeks had an intensity of 9.6 ± 0.4 clux (Galilux Light Meter, Hato Agricultural Lighting, Sittard, The Netherlands).

Data Collection

Serum Melatonin Levels Melatonin was measured for one sex and one genotype only. Blood samples were collected from 54 Ross YPMx708 males per wavelength. Samples were collected via decapitation of birds at 9 time points within a 24-hr period (4:00 pm, 7:00 pm, 10:00 pm, 1:00 am, 3:00 am, 5:00 am, 8:00 am, 11:00 am, and 2:00 pm) at d 17 and 18 for trial 1 and d 16 and d 17 for trial 2. The technique for sample collection was used previously in our research group (Schwean-Lardner et al., 2014), with sample-collection procedures completed in the dark during the scotophase, to ensure that light did not affect melatonin levels. Collection periods were scheduled to include three collections during the schotophase with the remainder equally spaced within the photophase. For each collection time, one bird per pen from 2 pens per room was randomly selected (6 birds per light treatment per time). Blood samples were centrifuged, and the supernatants were collected. Serum was stored at −20°C until the test was conducted. Samples were analyzed using the Chicken Melatonin (**MT**) ELISA kit (Elabscience Biotechnology Inc, Houston).

Tissue Melatonin Levels In trial 2, immediately after blood samples were collected for serum melatonin levels, the pineal gland and retina from both eyes were removed at 2 collection times, one during the scotophase (1:00 am) and the second during the photophase (7:00 pm). Samples were immediately placed in a container with liquid nitrogen and were then stored at −80°C until the analyses were conducted. Tissues were minced after being weighed then homogenized in a phosphate buffered saline buffer. The homogenates were centrifuged for 5 min to obtain the supernatant. Samples were analyzed using the Chicken Melatonin (MT) ELISA kit (Elabscience Biotechnology Inc, Houston).

Mobility Mobility was assessed utilizing the gait scoring (**GS**) methodology described by Garner et al. (2002), Table 1. A total of 48 birds per light wavelength treatment (12 birds per light color × genotype × sex) were tested at d 32 in trial 1 and d 31 in trial 2. To conduct the test, the home pen was divided into 2 equal sections. Birds were restricted to one section, while the empty section served as the runway for the birds to walk. Birds were individually selected at random, placed in the empty area and individually encouraged to walk. They were then scored on a scale of 0 (normal) to 5 (unable to stand) by 2 trained observers. Because a gait score of 3 or above is considered painful to animals (Danbury et al., 2000), the fractions of animals with a GS ≥3 were also calculated for statistical analyses.

Footpad Dermatitis (FPD) The birds chosen for GS were also evaluated for FPD. Subjective scoring was conducted using the photographic system described in

Table 1. Scoring system for the assessment of walking ability, as measured by gait scoring (adapted from Garner et al., 2002).

Score	Criteria
0	No impairment. Smooth locomotion.
1	Impairment detectable, but unidentifiable abnormality. The leg problem cannot be identified in the first 20 s of observation.
2	Identifiable abnormality, with little impact on overall function. The leg problem can be identified within the first 20 s of observation.
3	Identifiable abnormality which impairs function. Bird moves away from the observer but does not run and squat within 15 s.
4	Severe impairment of function, but still capable of walking. Bird remains squatting when the observer approaches. The observer gently touches the animal for 5 s. They may appear to rise but are still on their hocks.
5	Complete lameness. The bird cannot walk and shuffles along on its hocks. The animal is unable to stand.

the Welfare Quality Assessment (WQ) Protocol for Poultry (Welfare Quality Consortium, 2009). Broilers were assessed for the severity of hock burns according to scoring categories 0 (no lesion) to 4 (severe lesions) (Welfare Quality Consortium, 2009 [Figure 2]).

Litter Quality Litter quality was assessed on d 32 in the pens housing the broilers assessed for GS and FPD (one pen per genotype × sex per room) by one observer. Subjective scoring was conducted using the system described in the Welfare Quality Assessment (WQ) Protocol for Poultry (Welfare Quality Consortium, 2009 (Table 2)) in 4 areas in each pen: close to the entrance, along the right edge, beside the drinker, and in the center. Scores within a pen were averaged for statistical analyses.

Mortality and Morbidity Birds were monitored twice daily, and any runts or birds who were ill or displayed a physical abnormality that indicated suffering were culled via manual cervical dislocation. Pathologists at an independent lab (Prairie Diagnostic Services at the Western College of Veterinary Medicine, Saskatoon, Canada) performed necropsy on birds that were culled and that died during the rearing stage to determine the primary cause of morbidity or mortality.

Cause of death was classified into the following categories: *infectious (*runts, air sacculitis, arthritis, hepatitis, emaciation, liver necrosis, osteomyelitis, pericarditis, peritonitis, polyserositis, yolk sack infection), *metabolic (*ascites, sudden death syndrome,), *skeletal (*valgus varus, rotated tibia, tibial dyschondroplasia), *unknown (*no visible lesions), and *other (*intestinal accident, pendulous crop, starve out, dehydration).

Statistical Analyses

The two trials were treated as experimental blocks, which resulted in a total of 6 replicate rooms per lighting treatment and 18 replicate pens per wavelength × sex × genotype. Data were statistically analyzed using SAS (SAS 9.4, Cary, NC). The main effects were wavelength, sex, and genotype. Before analyses, all data were tested for normality using the UNIVARIATE procedure, and if not normally distributed, were transformed to meet these assumptions. All categorical data were log transformed to achieve normality. Data were analyzed as a 3 × 2 × 2 factorial design with light nested within room using the MIXED procedure. For melatonin data, an ANOVA was used for testing the differences in concentration at each time interval. Tukey's range test was used to separate means when significant differences were found. Differences were considered significant when $P \leq 0.05$.

Table 2. Scoring system for the assessment of litter quality (Welfare Quality Consortium, 2009).

Score	Criteria
0	Completely dry and flaky, easily moved with the foot
1	Dry but not easy to move with the foot
2	Left imprint of foot and formed a ball if compacted, but the ball did not stay together well
3	Stuck to boots and stuck readily in a ball if compacted
4	Stuck to boots once the cap or compacted crust is broken

RESULTS

The data included in this manuscript was part of a larger study. Productivity results have been previously published (Remonato Franco et al., 2022b), however Table 3 summarizes those results.

Melatonin

Light color had a minor effect on serum melatonin. The only time period with a difference occurred at 5 am, when birds raised under blue light had lower melatonin concentration compared to those reared under green or white light (Table 4). At that time point only, blue light suppressed broiler serum melatonin concentration by 40.7% compared to green light and 41.5% compared to white light. No differences existed for the remaining time points. Light color had no effect on melatonin concentration in either the retina or the pineal gland during the photophase or the scotophase (Table 5).

Mobility

Light color and genotype did not influence the percentage of birds falling within specific GS categories, nor on the total percentage of birds falling within the combined categories of 3, 4, and 5 (Table 6). A higher

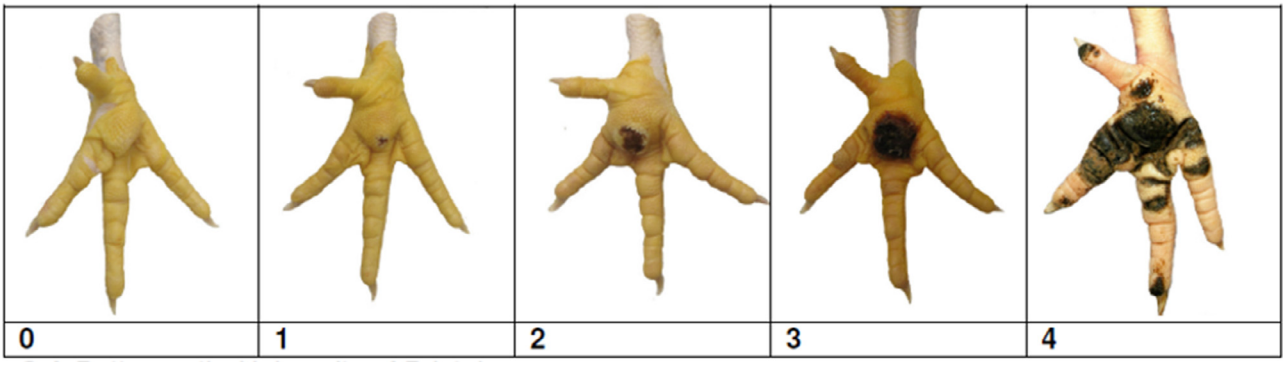

Figure 2. Scores of footpad dermatitis (Welfare Quality Consortium, 2009).

Table 3. Effects of wavelength treatments[1], genotype and sex, and their interactions, on broiler body weight (kg), feed intake (kg/bird) and gain to feed corrected for mortality (Remonato Franco et al., 2022b).

	Light				Genotype			Sex			Interactions	
	Blue	Green	White	*P* Value	Y-708	E-708	*P* value	Male	Female	*P* value	Genotype x sex	SEM[2]
Body Weight (kg)												
35d	2.457	2.447	2.464	0.77	2.488[a]	2.430[b]	<0.0001	2.594[a]	2.318[b]	<0.0001	NS	0.0121
Feed intake (kg/bird)												
0–35d	3.533	3.533	3.554	0.46	3.538	3.542	0.72	3.699[a]	3.381[b]	<0.0001	<0.0001	0.0202
Gain:Feed (mortality corrected)												
Total	0.715	0.715	0.714	0.97	0.720[a]	0.710[b]	<0.0001	0.726[a]	0.704[b]	<0.0001	NS	0.0011

Interactions between genotype and sex on feed intake

	Y-708 Male	Y-708 Female	E-708 Male	E-708 Female
0–35d	3.786[a]	3.286[c]	3.635[a]	3.261[b]

[a,b]Means with common letters in the same row do not differ significantly ($P \leq 0.05$).
[1]Dominant wavelengths for the blue treatment ranged from 435-500 nm, while the green treatment was dominated by 500-565 nm, and a combination of wavelengths produced white light.
[2]SEM = Standard error of the mean.

percentage of females compared to males were scored in the category 0 (normal mobility), and a higher percentage of males scored in categories 2 and 3 and categories 3, 4, and 5 combined. No significant interactions between light color, sex nor genotype were noted.

Footpad Dermatitis

Light color and genotype had no effect on FPD (Table 7). There was an increased percentage of males with FPD scoring 4 (most severe) compared to females. No significant interactions were noted.

Litter Quality

There was an interaction between light color and genotype (Table 8), where a larger percentage of pens containing YPM-708 broilers reared under green light were dry but not easy to move with the foot (category 1) compared to pens housing EPM-708 broilers under blue light. A higher number of pens housing EPM-708 broilers were dry and flaky (category 0) compared to pens housing YPM-708 broilers. With respect to sex, an increased percentage of pens with males fell within category 2 (left imprint of foot and formed a ball if compacted, but the ball did not stay together well), while more pens with females were noted in category 3 (stuck to boots and stuck readily in a ball if compacted).

Mortality and Causes of Mortality/Morbidity

Lighting treatment had no effect on the percentages of deaths/culls due to infectious, metabolic, unknown or "other" causes (Table 9). Genotype had no effect on total mortality nor cause of mortality. A higher percentage of infectious and metabolic related diseases were noted in males as compared to females. Sex reacted differently to wavelength treatments, where males raised under white light had a higher incidence of skeletal causes of mortality and had higher total mortality/morbidity.

DISCUSSION

Melatonin

Melatonin is an important neurohormone, responsible for regulating diurnal rhythms present in several behaviors and physiological or biochemical processes in the body (Alkozi, 2019; Cassone et al., 2017). It is known that light is one of the most important *zeitgeber*s of

Table 4. The effect of light color[1] on serum melatonin concentration in Ross YPMx708 males at 17d and 18d (trial 1) and 16d and 17d ([trial 2] pg/μL).

	Light				
	Blue	Green	White	*P*-value	SEM[2]
1:00 am	3835.3	3928.4	3714.4	0.96	312.15
3:00 am	4662.9	7941.9	10405.0	0.54	1405.16
5:00 am	8592.9[b]	14489.0[a]	14691.0[a]	0.02	1263.71
8:00 am	6159.1	4239.6	7182.7	0.31	797.89
11:00 am	5883.4	6453.2	7021.2	0.69	512.73
2:00 pm	5136.5	4274.9	8892.5	0.51	1726.78
4:00 pm	2475.4	3482.9	3962.5	0.58	431.40
7:00 pm	3377.2	2946.7	2909.2	0.85	415.05
10:00 pm	1957.9	3198.3	4218.1	0.16	518.09

[a,b]Means with common letters in the same row do not differ significantly ($P \leq 0.05$).Photophase: from 6:00 am to 12:00 pm. Scotophase: 12:00 am to 6:00 am.
[1]Dominant wavelengths for the treatment with blue light ranged from 435 to 500 nm, while the dominated wavelengths in the treatment with green light were 500−565 nm, and a combination of wavelengths produced white light.
[2]SEM = Standard error of the mean.

Table 5. The effect of light color[1] on melatonin concentration in the pineal gland and retina (pg/μL).

		Light				
		Blue	Green	White	*P*-value	SEM[2]
Retina	1:00 am	4207.5	5189.5	5415.4	0.65	305.81
Pineal Gland		1043.7	1266.8	1320.3	0.79	104.87
Retina	7:00 pm	3729.6	3975.7	5427.3	0.15	458.27
Pineal Gland		1178.7	1698.4	1167.1	0.38	121.73

[1]Dominant wavelengths for the treatment with blue light ranged from 435 to 500 nm, while the dominated wavelengths in the treatment with green light were 500−565 nm, and a combination of wavelengths produced white light.
[2]SEM = Standard error of the mean.Photophase: from 6:00 am to 12:00 pm. Scotophase: 12:00 am to 6:00 am.

Table 6. The effect of light color[1] on the percentage of broilers in each gait score category and the sum of categories 3, 4, and 5 on 32d (trial 1) and 31d (trial 2) as described by Garner et al (2002).

	Light				Genotype			Sex			
Categories	Blue	Green	White	*P*-value	Y-708	E-708	*P*-value	Male	Female	*P*-value	SEM[2]
0	27.8	20.8	25.0	0.78	23.9	25.2	0.79	15.5[b]	33.6[a]	0.001	3.24
1	51.0	53.1	45.8	0.67	50.3	49.6	0.89	45.5	54.5	0.09	3.26
2	18.9	21.9	21.9	0.85	22.3	19.5	0.56	31.3[a]	10.5[b]	<0.0001	2.66
3	2.2	3.1	4.2	0.78	1.42	4.9	0.09	5.6[a]	0.7[b]	0.02	1.09
4	0.0	0.0	1.0	0.41	0.7	0.0	0.32	0.7	0.0	0.32	0.35
5	0.0	1.0	2.1	0.37	1.4	0.7	0.57	1.4	0.7	0.57	0.59
3+4+5	2.1	4.2	7.3	0.31	3.5	5.6	0.42	7.7[a]	1.4[b]	0.01	1.34

[a,b]Means with common letters in the same row do not differ significantly ($P \leq 0.05$).
[1]Dominant wavelengths for the treatment with blue light ranged from 435 to 500 nm, while the dominated wavelengths in the treatment with green light were 500−565 nm, and a combination of wavelengths produced white light.
[2]Standard error of the mean.

diurnal rhythms, working as an external cue to help the body entrain its biological clock with a rhythmic cycle. Studies have been conducted on the impact of some aspects of light and its influence on melatonin production in poultry, including daylength, (Lewis et al., 2001; Zawilska et al., 2007; Schwean-Lardner et al., 2014) and light intensity (Deep et al., 2012). In humans, light color has an influence on melatonin production, as shorter-wavelength monochromatic light, such as blue light, suppresses melatonin (Lockley et al., 2003; Kazaki et al., 2016). This is also the case in other species. In hamsters, blue light was the most efficient at suppressing melatonin, followed by green light, when compared to yellow, near ultraviolet, and red light (Brainard et al., 1984). Blue light also suppressed melatonin in sea bass (Bayarri et al., 2002). In broilers however, little data are available in the literature to describe the influence of light spectrum on melatonin.

In our study, raising broilers under monochromatic blue light decreased melatonin concentration at one time period during the scotophase (5:00 am) compared to when birds were raised under green or white light. Although this result agrees with previous research conducted in humans, where exposure to blue monochromatic light attenuated the secretion of nocturnal melatonin (Lockley et al., 2003; Cajochen et al., 2005; Kazaki et al., 2016; Tähkämö et al., 2019; Wahl et al., 2019), no impacts were noted at other scotoperiod time points. Therefore, this suggests only a minor effect of light color on melatonin concentration. A suppression of melatonin can lead to a weakening of the circadian pacemaker (Gwinner at al., 1997), influencing sleepiness and alertness (Cajochen et al., 2005). However, because our results show only minor differences, the decrease in melatonin concentration observed are not likely to have a detrimental effect.

Gait Scoring, Footpad Dermatitis, and Litter Quality

Leg health is an important component of broiler welfare. The incidence of lameness in broiler flocks has been reduced in the past years, primarily due to targeted genetic selection for this issue at the primary breeder level, however, it remains an important factor to be monitored (Kapell et al., 2012). Depending on the severity, leg disorders reduce the welfare of affected birds, as they may be painful and lead to reduction of mobility, which could also impact access to food and water (Danbury et al., 2000; Bessei, 2006).

Leg issues may have genetic and infectious causes; however, it is known that other factors may be related to its etiology. Melatonin appears to be important, since it promotes bone development and impairs the development of osteopenia (Cardinali et al., 2003). Additionally, exercise and increases in activity can lead to the prevention of leg disorders through the promotion of bone development (Reiter and Bessei, 1996; Bradshaw et al., 2002). Data from previous work in our research group

Table 7. The effect of light color[1] on the percentage of broilers in each footpad dermatitis category at 32d (trial 1) and 31d (trial 2).

	Light				Genotype			Sex			
Categories[2]	Blue	Green	White	*P*-value	Y-708	E-708	*P*-value	Male	Female	*P*-value	SEM[3]
0	40.6	42.7	29.2	0.13	38.2	36.8	0.81	41.7	33.3	0.15	3.24
1	17.9	23.9	26.0	0.52	24.4	20.9	0.39	19.5	25.8	0.12	2.16
2	30.2	19.8	21.9	0.14	22.2	25.7	0.44	21.5	26.4	0.28	2.34
3	10.5	11.5	12.5	0.92	10.4	12.5	0.53	10.4	12.5	0.53	1.78
4	1.2	2.0	10.4	0.39	4.9	4.2	0.77	7.0[a]	2.1[b]	0.04	1.74

[a,b]Means with common letters in the same row do not differ significantly ($P \leq 0.05$).
[1]Dominant wavelengths for the treatment with blue light ranged from 435 to 500 nm, while the dominated wavelengths in the treatment with green light were 500−565 nm, and a combination of wavelengths produced white light.
[2]Category 0: no evidence of footpad dermatitis. Categories 1 and 2: minimal evidence of footpad dermatitis, Categories 3 and 4: evidence of footpad dermatitis (according to the photographic scale provided by Welfare Quality Consortium (2009).[3]Standard error of the mean.
[3]Standard error of the mean.

Table 8. The effect of light color[1] on the percentage of pens classified into different litter quality categories on 32d (trial 1) and 31d (trial 2) as well as the interactions between light and genotype.

	Light				Genotype			Sex			Significant interaction *P*-values	
Categories[2]	Blue	Green	White	*P*-value	Y-708	E-708	*P*-value	Male	Female	*P*-value	Light x genotype	SEM[3]
0	59.0	50.0	50.0	0.28	47.8[b]	58.2[a]	0.02	56.1	49.9	0.17	-	3.045
1	16.7	23.9	21.9	0.29	24.3	17.4	0.08	17.4	24.3	0.08	0.04	2.662
2	19.4	19.8	17.7	0.84	20.7	17.2	0.19	23.5[a]	14.5[b]	0.001	-	1.441
3	4.9	6.3	10.4	0.56	7.1	7.1	1.00	3.0[b]	11.3[a]	0.0003	-	1.417

Litter quality categories interactions between light x genotype

	Blue − Y708	Blue − E708	Green − Y708	Green − E708	White − Y708	White − E708
Category 1	20.8[ab]	12.5[b]	33.3[a]	14.6[ab]	18.8[ab]	25.0[ab]

[a,b]Means with common letters in the same row do not differ significantly ($P \leq 0.05$).No pens received a score of 4.

[1]Dominant wavelengths for the treatment with blue light ranged from 435 to 500 nm, while the dominated wavelengths in the treatment with green light were 500−565 nm, and a combination of wavelengths produced white light.

[2]Category 0: completely dry and flaky, category 1: dry but not easy to move with the foot, category 2: leaves an imprint of foot and will form a ball if compacted, but the ball does not stay together well, category 3: stick to boots and sticks readily in a ball if compacted, category 4: stick to boots once the cap or compacted crust is broken.

[3]Standard error of the mean.

Table 9. Effect of light color[1], genotype, and sex on causes of mortality from 0-35d and the interactions between light and sex.

	Light				Genotype			Sex			Significant interaction *P*-values	
	Blue	Green	White	*P*-value	Y-708	E-708	*P*-value	Male	Female	*P*-value	Light x sex	SEM[2]
Infectious	3.62	3.64	3.63	0.92	3.99	3.29	0.07	4.17[a]	3.09[b]	0.007	-	0.047
Metabolic	1.00	1.43	1.50	0.15	1.33	1.29	0.91	1.90[a]	0.71[b]	<0.0001	-	0.021
Skeletal	0.63	0.49	0.66	0.48	0.55	0.63	0.39	0.77[a]	0.42[b]	0.01	0.04	0.014
Unknown	0.53	0.50	0.51	0.84	0.60	0.41	0.09	0.70	0.34	0.06	-	0.012
Other	0.22	0.38	0.29	0.48	0.28	0.31	0.47	0.41	0.20	0.07	-	0.009
Total	6.00	6.44	6.59	0.78	6.75	5.93	0.13	7.95[a]	4.78[b]	<0.0001	0.01	0.055

Causes of mortality indices interactions between light x sex

	Blue - Male	Blue - Female	Green - Male	Green - Female	White - Male	White - Female
Skeletal	0.56[ab]	0.67[ab]	0.73[ab]	0.24[b]	0.99[a]	0.36[ab]
Total	7.23[ab]	7.74[bc]	7.10[ab]	5.56[bc]	8.97[a]	4.05[c]

[a,b]Means with common letters in the same row do not differ significantly ($P \leq 0.05$).

[1]Dominant wavelengths for the treatment with blue light ranged from 435 to 500 nm, while the dominated wavelengths in the treatment with green light were 500−565 nm, and a combination of wavelengths produced white light.

[2]Standard error of the mean.

revealed that light color affected behavioral expression, where broilers raised under blue light spent more time resting and less time performing active behaviors, such as walking (Remonato Franco et al., 2022a). With the observed decrease in broiler activity (Remonato Franco et al., 2022a) and melatonin concentration during one measured period in the scotophase, impacts on lameness might be expected in broilers raised under blue light. This was not the case however, which agrees with previous research in broilers raised under different light colors (Senaratna et al., 2011). This suggests that the differences in exercise and melatonin concentration were not large enough to impact gait scores.

Bird behavior can impact litter quality. A reduction in activity can decrease litter quality, as litter is not turned over to the same extent, limiting drying ability. Poor litter quality can increase footpad dermatitis (Bessei, 2006). However, wavelength treatments did not influence the incidence of footpad dermatitis, suggesting again that the reduction in activity was likely not enough to affect these parameters.

Causes of Mortality/Morbidity

Management processes that affect immunity of broilers could have impacts on mortality levels. Results in the literature are inconsistent about the impact of light color on immune function. Although previous work demonstrated that short wavelengths (green or blue light) could lead to an enhanced immune response in broilers (Xie et al., 2008), others found no significant effect of light color (Kim et al., 2013). In this research, light wavelength had no effect on infectious causes of mortality. Likewise, light color did not affect metabolic or skeletal causes of mortality/morbidity. There are many potential factors that can affect metabolic disorders, and one might be growth, or more specifically, timing of growth (Schwean-Lardner et al., 2013). Data from our previous work demonstrated that light color had no impact on timing of weight gain (Remonato Franco et al., 2022b), therefore no significant changes in metabolic causes of death related to increased growth rate were expected. Melatonin levels have also been implicated in Spiking Mortality Syndrome in broiler chickens, where broilers raised under brief dark periods developed hypoglycemia (Julian, 2005). This was not observed in the current study, which could support the suggestion that impacts on melatonin concentration were not large enough to affect glycemic levels.

Overall, the exposure of broilers to a lighting program consisting of monochromatic blue light led to only minor effects on the secretion of melatonin during the scotoperiod. Exposure to blue, green, and white light in this study did not impact other health parameters assessed, such as gait score or footpad dermatitis and had minor impacts on litter quality and cause of mortality/morbidity. Likewise, genotype and sex showed minor impacts on the health parameters assessed in this study. In conclusion, the results of the study demonstrate that utilizing light of a specific wavelength, such as blue or green, in commercial broiler facilities, is not likely to affect the health status of broilers reared to 35 d.

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DISCLOSURES

The authors declare no conflicts of interest.

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