

Effects of Light Color on Growth Performance, Histomorphometric Features of Small Intestine and Some Blood Parameters in Chukar Partridges (*Alectoris chukar*)^[1]

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Abstract

The study was conducted in order to compare the effects of light-emitting diode (LED) light with a different color on performance parameters, histomorphometric features of the small intestine and some blood parameters in growing partridges. For this purpose, a total of 450 Chukar partridges (*Alectoris chukar*) were perdivided into three experimental groups including green light (GL), daylight (DL) and blue light (BL), and 250-lumen lighting. Each group contained 150 partridges with 5 repetitions per group. Experimental groups balanced according to initial live weight and raised under the same feeding and environmental conditions within the scope of the needs of partridges during 35 days. GL group showed higher live weight ($P<0.05$), live weight gain ($P<0.01$) and feed intake ($P<0.05$). Feed conversion and viability rates were similar among the groups ($P>0.05$). Higher villus height (VH), villus width, villus surface area were obtained in GL group, followed by DL and BL ($P<0.001$). The differences in crypt depth (CD) and VH/CD were not significant ($P>0.05$). Serum glucose ($P<0.01$) and lactate dehydrogenase levels (LDH) ($P<0.05$) were increased in GL groups. Serum high-density lipoprotein level (HDL) was found to be higher in DL group ($P<0.01$). Consequently, GL group had better results in growing partridges. GL could be utilized under intensive farming conditions for partridges.

Keywords: Fowl, Illumination color, Biochemical parameters, Growth performance, Histological examination

Kımalı Kekliklerde (*Alectoris chukar*) Işık Renginin Büyüme Performansı, İnce Bağırsak Histomorfometrik Özellikleri ve Bazı Kan Parametreleri Üzerine Etkileri

Öz

Bu çalışma, farklı renkte ışık yayan diyot lambaların (LED) kekliklerde büyüme performansı, ince bağırsağın histomorfometrik özellikleri ve bazı kan parametreleri üzerine etkilerini karşılaştırmak amacıyla yapılmıştır. Bu amaç için, toplam 450 Kımalı keklik (*Alectoris chukar*) yeşil ışık (YI), gün ışığı (GI) ve mavi ışık (MI) grupları olmak üzere 3 gruba ayrılmış ve 250 lümen aydınlatma uygulanmıştır. Her grup, grup başına 5 tekrar içeren 150 keklikten oluşmuştur. Deney grupları, kekliklerin başlangıç canlı ağırlıklarına göre belirlenmiş olup, büyüme dönemleri göz önünde tutularak, 35 günlük deneme süresince her gruba eşit beslenme ve çevresel koşullar sağlanmıştır. YI grubu canlı ağırlık ($P<0.05$), canlı ağırlık artışı ($P<0.01$) ve yem tüketimi ($P<0.05$) açısından yüksek değerler göstermiştir. Yem dönüşümü ve yaşama gücü oranları gruplar arasında benzer tespit edilmiştir ($P>0.05$). YI grubunda villus yüksekliği (VY), villus genişliği, villus yüzey alanı daha yüksek olup, bu grubu sırasıyla GI ve MI ($P<0.001$) grupları takip etmiştir. Kript derinliği (KD) ve VY/KD'deki farklılıklar önemli bulunmamıştır ($P>0.05$). YI gruplarında serum glukoz ($P<0.01$) ve laktat dehidrojenaz seviyeleri (LDH) ($P<0.05$) artmıştır. Serum yüksek yoğunluklu lipoprotein düzeyi (HDL) GI grubunda yüksek tespit edilmiştir ($P<0.01$). Sonuç olarak, YI grubundan kekliklerin büyüme döneminde daha iyi sonuçlar elde edilmiştir. Entansif koşullarda keklik yetiştiriciliği yaparken YI'dan faydalanılabilir.

Anahtar sözcükler: Av kuşu, Aydınlatma rengi, Biyokimyasal parametreler, Büyüme performansı, Histolojik inceleme



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INTRODUCTION

In poultry farming, the light is determined to have crucial functions to carry out rhythmic and synchronized functions including important metabolic events of the body, to control body temperature, to control activities such as feed consumption, growth, maturity, and reproduction, as well as releasing, stimulating, and controlling hormones [1,2]. While conventional incandescent and fluorescent lamps have been used as the source of light until the recent past, there have been technological advancements in illumination in the last few years and these lamps have started to be substituted with light emitting diodes (LED). The most important advantages of LEDs are that they have high energy saving (consumption of energy of 80% lesser compared to incandescent lamp, 50% lesser compared to fluorescent lamp), are long-lasting, have high reliability, low costs of maintenance and the wavelength to ensure sufficient light stimulation for poultry species [3-5].

Color of light is the associations caused by beams at different wavelengths in the brain. Like mammals, poultry can see the part of light whose the wavelength varies between 380 nm and 780 nm and which is described as color [6]. The eye has different perception sensitiveness for lights with diverse wavelengths. Eye of poultry has a structure different from mammals. The light is detected more densely by retinal and extra-retinal photoreceptors [7]. Extra-retinal pineal gland and hypothalamus play a role in certain homeostatic, physiological, and reproductive events; whereas, retina directs growth and behaviors [1].

Perception levels of the light with different wavelengths vary across the poultry species [8]. In this sense, it is important to identify the wavelength that is appropriate to the goal of farming for species. Previous studies revealed that generally light with long wavelength or higher density contains higher energy and can reach to hypothalamus because of higher capability to penetrate in skull, thereby better results have been obtained in development of sexual organs, reaching to sexual maturity, activation of sexual hormones, egg production, and metabolic activities such as longer laying periods [1,7,9,10]. It was determined that the light with shorter wavelength or lower density could be easily detected by retinal receptor and retinal stimulation was sufficient in terms of growth and fattening performance [6,11,12]. On the other hand, poultry performance is also significantly affected by behaviors of poultry. Density, wavelength, and source of the light are known to be the important environmental factors that have an effect on behaviors and physiology of poultry animals [13,14]. It was found for laying hens that red light decreased aggressiveness [4], green light relieved and rested broilers [15], white light elevated plasma corticosterone levels in songbirds [16], also color of the light are effective on blood biochemistry such as glucose, triglyceride, cholesterol, uric acid [12,17] and immunological parameters [11,18].

Because it is thought that a different species such as partridge may react differently to the light spectrum. The study was aimed to determine the effect of green light, daylight and blue light on the growth performance, some blood parameters, and the intensity of histomorphometric characteristics of the small intestine during the growth period (1-35 days) of the Chukar partridges (*Alectoris chukar*).

MATERIAL and METHODS

Ethical Approval

The study was conducted in Chukar partridge production station affiliated with Turkish Republic Ministry of Forestry and Water Affairs. Approval of the Ministry and Firat University Animal Experimentation Local Ethics Committee was obtained for the study (FUHADYEK, Approval number: 2016/127).

Experimental Design

Daily chicks from incubator which was available in the station were weighed randomly and distributed into experimental groups to equalize initial live weights. Experimental groups were formed as green light (GL), daylight (DL), and blue light (BL) led groups (Single-chipped, outdoor, 60 leds/M, DC 12 V, 4.4 W/M). Every experimental group was planned with 5 repetitions. The study was conducted by using 30 partridges in each repetition, 150 partridge in each group, and 450 Chukar partridges (*Alectoris chukar*) in total. The ingredients and composition of the diet are shown in Table 1. Feed and water were supplied *ad libitum* during the study (1-35 days). Conditions of poultry house were regulated without causing the differences between experimental groups within the scope of the needs of partridges. Partridge chicks were raised in the special wire cages with 5 tiers in the size of 95 x 50 x 22.5 cm (length x width x height). Each section of the cage was illuminated using 250-lumen lighting. The 24 h light was applied during first three days, later it was gradually lowered to 16 h light/8 h dark program. During the trial, live weights were measured weekly. On the day of weighing, the feed was removed from in front of partridges at 12 a.m. and all partridges in the study were recorded weighing in groups with 5 birds at 8 a.m. The 5 birds in each group were weighed at once. Feeds were supplied by weighing every day and remaining feeds were recorded weekly and feed conversion was calculated. Dead animals were recorded daily. Viability rate of experimental groups was calculated (Fig. 1). At the end of 35 days, the animals were slaughtered by randomly choosing 2 partridges from each repetition, 10 from each group, and 30 partridges in total. Bloods of slaughtered partridges were collected in serum tubes with gel. Serum were removed by centrifuging blood samples at 1792 g for 10 min. Small intestine samples (duodenum) were taken following the slaughter and kept under appropriate conditions until the analyses were performed.

Table 1. Ingredients and composition of diet	
Feed Ingredients	(%)
Maize	40.50
Wheat	9.51
Soybean meal, 48%	30.20
Maizebran	9.95
Vegetableoil	4.70
DL-Methionine	0.34
Dicalciumphosphate	2.90
Calciumcarbonate	1.00
L-Lysinehydrochloride	0.25
Salt	0.40
Vitamin*-mineral** mixture	0.25
Composition	(%)
Drymatter	90.2
Crude protein	20.4
Crudecellulose	3.17
Crudefat	6.72
Crudeash	7.28
Calcium	1.18
Availablephosphorus	0.64
Sodium	0.19
Methionine + cystine	1.00
Lysine	1.32
Threonine	0.72
Metabolicenergy, kJ	12552.00
* Vitamin mix (per 1 kg): Vit. A 5.000 IU, Vit. D ₃ 500 IU, Vit. E 10 mg, Vit. K ₃ 2 mg, Vit. B ₂ 4 mg, Vit. B ₁₂ 10 mg; ** Mineral mix (per 1kg): Manganese 120 mg, ferrous 40 mg, zinc 16 mg, copper 16 mg, cobalt 200 mg, iodine 1.25 mg, selenium 0.30 mg	

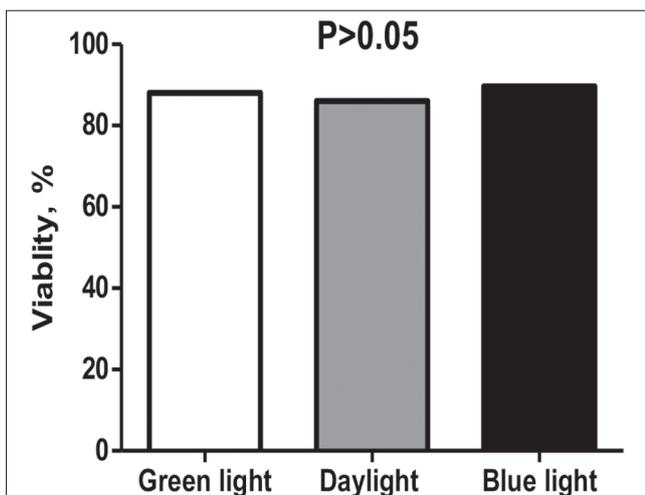


Fig 1. Effect of light color on viability of Chukar partridges

Morphometric Analyses of Small Intestine Segment

Tissue samples were taken from the duodenum and fixed in 10% neutral-buffered formalin for 24 h. They were then

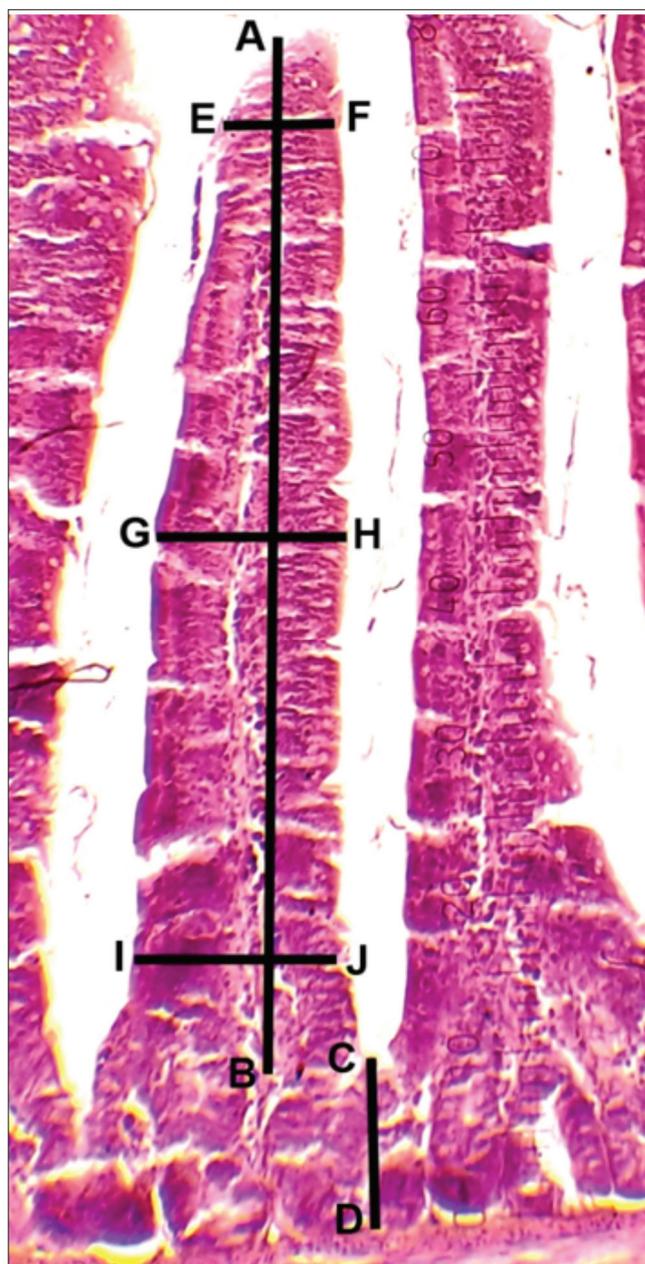


Fig 2. Section of the duodenum hematoxylin-eosinstained, showing the measured parameters. Villus height [AB], crypt depth [CD], villus edge widths [EF], villus middle width [GH] and villus bottom widths [IJ] (x200)

dehydrated through graded ethanol and embedded in paraffin. Five micrometers thick sections were obtained by a microtome and placed on glass slides. Cross sections were processed according to conventional hematoxylin and eosin staining method [19]. Sections were examined by a light microscope (Olympus CX31, Olympus USA) equipped with a digital imaging system (Olympus DP20, Olympus USA). Villus surface area was calculated according to Solis de los Santos et al. [20]. In all histomorphometric parameters, villus heights (VH), villus bottom widths, villus edge widths and crypt depths (CD) related to villi were measured from 7 consecutive villus-crypt complexes. Villus heights (AB) was measured from joining point of villi

Table 2. Treatment groups means and standard errors as well as P-values of performance traits of Chukar partridges in different ages

Days	Green Light (GL)	Day Light (DL)	Blue Light (BL)	Significance (P)
Live weights (LW), g/day/bird				
1	14.92±0.08	14.92±0.03	14.92±0.05	NS
7	28.62±0.22	28.87±0.34	28.76±0.25	NS
14	60.02±0.54	60.08±0.68	59.74±0.41	NS
21	95.58±0.87 ^a	92.44±0.59 ^b	90.33±1.22 ^b	**
28	138.36±1.32 ^a	135.48±0.75 ^{ab}	134.44±1.06 ^b	*
35	178.54±1.71 ^a	175.68±2.19 ^{ab}	171.67±1.26 ^b	*
Live weight gains (LWG), g/day/bird				
1-7	1.95±0.06	1.99±0.04	1.97±0.06	NS
8-14	4.48±0.08	4.45±0.03	4.42±0.04	NS
15-21	5.08±0.19	4.62±0.16	4.37±0.24	NS
22-28	6.11±0.24	6.14±0.04	6.30±0.33	NS
29-35	5.74±0.12	5.74±0.12	5.31±0.12	NS
1-35	4.67±0.03 ^a	4.59±0.03 ^{ab}	4.47±0.03 ^b	**
Feed intake (FI), g/day/bird				
1-7	4.41±0.09	4.69±0.14	4.29±0.11	NS
8-14	16.11±1.34	15.39±1.71	15.06±0.71	NS
15-21	24.22±0.10 ^a	19.44±0.20 ^b	21.57±1.51 ^{ab}	**
22-28	22.26±1.93	19.44±1.46	17.84±0.27	NS
29-35	22.56±2.29	20.48±2.19	17.27±0.18	NS
1-35	17.91±0.99 ^a	15.96±0.46 ^{ab}	15.20±0.30 ^b	*
Feed conversion ratio, g FI/g LWG				
1-7	2.27±0.09	2.35±0.12	2.17±0.11	NS
8-14	3.59±0.26	3.45±0.41	3.40±0.15	NS
15-21	4.76±0.21	4.20±0.18	4.93±0.23	NS
22-28	3.63±0.41	3.16±0.21	2.85±0.05	NS
29-35	3.93±0.45	3.56±0.30	3.25±0.08	NS
1-35	3.83±0.21	3.47±0.07	3.40±0.06	NS

NS: Non-significant, * P<0.05, ** P<0.01, ^{a,b} The differences among the mean values with different superscripts (a, b: P<0.05) in a row within the different illumination groups are significant according to the ANOVA and post-hoc Tukey HSD tests

and crypts to top points of the villus. CD was measured from the middle point of two neighboring villi to base of crypts. While villus edge widths (EF) was measured from just below the top point of the villus, villus bottom widths (IJ) was measured from just above the joining point of villi and crypts. Also, the middle widths (GH) was measured from the middle of the villus heights (Fig. 2). By using the measured values, ratios of villus height/crypt depth (VH/CD) were also calculated.

Blood Parameters

Serum glucose, triglyceride, high density lipoprotein (HDL), creatine, uric acid, urea, lactate dehydrogenase (LDH), and blood urea nitrogen (BUN) values were measured via autoanalyzer (Mind-Way B5-2000 M), calcium level via atomic absorption spectrophotometer (Perkin Elmer AAnalyst 800 Atomic Absorption Spectrometer-Flame), Vitamin D level via high performance liquid chromatography (HPLC)

(Shimadzu) (25-OH Vitamin D₂/D₃, ImmuChrom GmbH-IC3401-160114).

Statistical Analyses

After tests of Shapiro-Wilk for normality, Levene's test for homogeneity of variance was used. All data including performance parameters, blood parameters and histomorphometric features of small intestine were subjected to analysis of variance. Significant differences were compared with Tukey HSD test. Viability rate of groups were compared with Chi-square test. All analyses were performed using SPSS ver. 21 for Windows [21]. Differences were important at P≤0.05. Data were presented as means ± SEM.

RESULTS

The effect of light colors on performance parameters in Chukar partridge is shown in Table 2. The live weight of GL

Table 3. Treatment groups means and standard errors as well as P-values of histo-morphometric features of small intestine of Chukar partridges

Parameters (μm)	Green Light (GL)	Day Light (DL)	Blue Light (BL)	Significance (P)
Villus height (VH)	112.91 \pm 2.93 ^a	109.10 \pm 3.67 ^a	94.10 \pm 2.80 ^b	***
Crypt depth (CD)	11.50 \pm 0.47	10.64 \pm 0.60	10.29 \pm 0.53	NS
Villus edge width	13.02 \pm 0.46 ^a	12.02 \pm 0.46 ^{ab}	9.68 \pm 0.33 ^b	***
Villus middle width	16.37 \pm 0.54 ^a	14.93 \pm 0.51 ^{ab}	11.22 \pm 0.42 ^b	***
Villus bottom width	19.77 \pm 0.70 ^a	17.50 \pm 0.67 ^b	13.77 \pm 0.63 ^c	***
VH/CD	9.81 \pm 0.65	10.25 \pm 0.65	9.14 \pm 0.55	NS
Villus surface area (VSA)	5813.60 \pm 230.79 ^a	4989.54 \pm 178.61 ^b	3431.81 \pm 145.25 ^c	***

VSA: (2x3.14) x (Mean of Villus width/2) x (Villus height), NS: Non-significant, *** P<0.001, ^{a,b,c} The differences among the mean values with different superscripts (a, b, c: P<0.05) in a row within the different illumination groups are significant according to the ANOVA and post-hoc Tukey HSD tests

Table 4. Treatment groups means and standard errors as well as P-values of blood parameters of Chukar partridges

Parameters	Green Light (GL)	Day Light (DL)	Blue Light (BL)	Significance (P)
Vit D, mg/dL	11.92 \pm 1.58	14.24 \pm 1.56	10.70 \pm 0.59	NS
Calcium, mg/dL	6.81 \pm 0.29	6.55 \pm 0.30	6.46 \pm 0.39	NS
Glucose, mg/dL	350.26 \pm 6.42 ^a	321.00 \pm 9.96 ^{ab}	304.96 \pm 5.34 ^b	**
Triglyceride, mg/dL	68.94 \pm 7.64	58.84 \pm 4.03	55.74 \pm 7.37	NS
HDL, mmol/dL	108.28 \pm 7.48 ^b	141.60 \pm 4.11 ^a	117.38 \pm 5.45 ^b	**
Uricacid, $\mu\text{mol/dL}$	1.91 \pm 0.42	2.12 \pm 0.50	3.54 \pm 0.57	NS
Urea, mg/dL	6.75 \pm 0.94	4.79 \pm 0.53	5.33 \pm 0.51	NS
LDH, $\mu\text{kat/L}$	23.35 \pm 1.67 ^a	18.30 \pm 1.42 ^{ab}	18.20 \pm 0.80 ^b	*
BUN, mg/dL	3.20 \pm 0.37	2.20 \pm 0.35	2.60 \pm 0.24	NS

HDL: High density lipoprotein, LDH: Lactatedehydrogenase, BUN: Blood urea nitrogen, NS: Non-significant, * P<0.05, ** P<0.01, ^{a,b} The differences among the mean values with different superscripts (a, b: P<0.05) in a row within the different illumination groups are significant according to the ANOVA and post-hoc Tukey HSD tests

group was found to be higher in days of 21 (P<0.01), 28 and 35 (P<0.05), followed by DL and BL. Live weight gain was also different among the groups in the period of 1-35 days. GL group showed better performance in live weight gain than DL and BL (P<0.01). Feed intake in days 15-21 (P<0.01) and 1-35 (P<0.05) were significantly higher in GL group. However, feed conversion (Table 2) and viability rates (Fig. 1) were similar among the groups (P>0.05).

When the effect of light colors on histo-morphometric features of the small intestine is examined in Table 3, villus height, villus width in the edge, middle and bottom of the duodenum, and villus surface area were found to be superior in GL group compare with DL and BL, respectively (P<0.01). The crypt depth of duodenum and the ratio of villus height to crypt depth were similar among the groups (P>0.05).

Table 4 shows the blood parameters of partridges subjected to different light color. Serum glucose (P<0.01) and LDH (P<0.05) levels showed statistically higher value in GL group, followed by DL and BL. However, serum HDL was found higher in DL group than another experimental group. The differences in levels of Vit D, calcium, triglyceride, uric acid, urea, and BUN weren't significant among the experimental groups (P>0.05).

DISCUSSION

The studies on the sensitivity of poultry to light spectrum have been continuing intensively [10,22-24]. The superiority of LEDs to other sources of light increases the use of this light source in poultry houses day by day [5]. Because perception levels of LED lamps at various wavelengths are different, a lot of researchs in broiler [1,9,25] and less in other poultry species [10,17,23] were determined to have different effects on performance in recent years, however, no information could be reached in partridge species. Investigating in the present study showed that green LEDs increase feed intake of partridges significantly (Table 2). In parallel with feed intake, live weight gains of partridges and live weights on the day of 35 were significantly high in this group. There were also no significant differences among the groups in terms of feed conversion and viability rates. On the other hand, better results were obtained from daylight illumination compared to blue light and the lowest values were obtained in the blue light group. When the other studies on growth and fattening performance were reviewed, green light was emphasized to lead to an increase in skeletal muscle cells of broilers, expression of growth hormone receptor genes, the number and uniformity of myofibrils in early period [12,25]. Sarica [26] stated

that green illumination had a delaying effect on the ovary and testicle development of quails, therefore, increased the growth performance compared to red and white light because it retarded sexual maturation. In different studies revealed that melatonin, which has important effects on regulation of body rhythm, cell regeneration, and immunity, was reported to be synthesized mostly under green light followed by red, white, and blue light [27]. Associated with increased levels of melatonin, the fact that the present study provided higher feed consumption and better live weight gain for partridge chicks grown under green light may be associated with feeling better and coping with environmental factors better. Similarly, Pan et al. [8] stated that while the light with a short wavelength (blue and green) induced the growth of broilers in early periods, the light with a long wavelength (yellow) induced the growth in the late period. It was found that every 100 nm (455 to 620 nm) increase of light's wavelength in early period decreased live weight by 15.4 g, every 100 nm of increase in the late period elevated live weight by about 16.4 g. Hassan et al. [17] also reported that the highest body weight and weight gain of duck were obtained in green light treatment compared with the white, yellow and blue lights during first 21 days. During d 22-42, weight gain increased in both the green and blue treatments in a like manner. Poultry behaviors are another factor influencing poultry performance. Studies on broilers revealed that red and white light increased activity in chicks, white light increased aggressiveness compared to red therefore negatively influenced growth, and broilers felt more peaceful at a wavelength shorter than 580 nm (blue, green, yellow) [15].

In addition to above-mentioned results, challenging effect of green light on growth was considered to be arising from the changes in bowel structure. When the results in Table 3 were examined, there was a substantial increase in villus height and villus width of bowel in the green light group and villus width was higher than other groups all along the whole villus. These developments in intestinal villi increased the surface area of villi. These results were thought to influence digestion and absorption positively [20,28]. Similarly, compared to white and blue light, green light increased intestinal villus length and the ratio of villus length to crypt depth in early periods in broilers (days of 1-21) and decreased crypt depth and blue light had similar results in the late period (until 49) [11]. In addition, green light in early period and blue light in late period reinforced intestinal immunologic barrier by influencing the number of intestinal intraepithelial lymphocytes, goblet cells, and IgA+ cells [11], and also caused birds to become more resistant to environmental stress by improving general immunity [18].

When the findings of blood parameters were examined (Table 4), it was observed that all parameters studied were within the normal ranges reported for partridges [29].

However, blood glucose and LDH levels were significantly higher in the green light group compare with blue light. Blood HDL level was higher in daylight group, conversely. Light applications were stated to have an important effect on the release of hormones and on glucose level [30,31]. Elevated LDH might have related that it induces the energy transformation process which is required to reversibly convert pyruvate, in parallel to increasing glucose concentration, into lactic acid and for cellular usage of glucose [32]. In other studies, their results were similar to blue light group of the present study, reported that there were significant relationships between blood pressure, blood lipids and blood glucose; exposure to blue light causes a decrease in blood pressure and positively effects blood glucose level and blood lipids [33]. Hassan et al. [17] also stated lower cholesterol level in blue light than green in duck. On the contrary, Yang et al. [12] determined that in broilers green light alone significantly increased blood high-density lipoprotein cholesterol (HDL-CH) level; whereas, blue light alone low-density lipoprotein cholesterol (LDL-CH) level, a combination of both colors increased both performance parameters and blood glucose levels.

In conclusion, the green light was identified to stimulate the growth of partridges during the growth period. It also improved the intestinal structure in the direction of increasing absorption and changed hormonal mechanism positively. Accordingly, the use of green light during growth period in the enterprise performing intensive partridge farming is considered will be improve performance.

CONFLICT OF INTEREST

We declare that there is no conflict of interest with any financial organization regarding the material discussed in the manuscript.

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