

## EFFECTS OF MONOCHROMATIC LIGHT ON PERFORMANCE, EGG QUALITY, YOLK CHOLESTEROL AND BLOOD BIOCHEMICAL PROFILE OF LAYING HENS

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### ABSTRACT

The present study was conducted to investigate whether monochromatic light affects the production performance, egg quality and some plasma parameters of layers. In this study, white and green fluorescent lamps were used to illuminate *Brown-Nick* hens for 8 weeks. Light sources made uniform to a light intensity of 15 lx. Forty-seven-week-old layers were divided into 2 groups of similar mean weight comprising 16 birds each and housed in individual cages. A 16:8 hours light:dark photoperiod was employed. The layers were exposed to conventional ambient temperature and 50-60% relative humidity for 24h per day. Performance was evaluated by recording feed intake, egg weight and egg production on daily basis; egg quality and egg cholesterol level on weekly basis; and blood parameters on 56<sup>th</sup> day of the experiment. The results indicated that green light application did not have any significant effect on feed intake, feed conversion ratio, egg production, total number of eggs, total egg weight, average egg weight, cholesterol level of egg yolk and egg quality of laying hens. However, green light application affected egg yolk color a\* and b\* values, but decreased serum albumin concentration. The results suggest that green light application may improve the yolk color of laying hens.

**Keywords:** Light color, egg cholesterol, egg quality, Brown-Nick hen, serum parameters.

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### INTRODUCTION

Light is effective for controlling physiological and behavioral processes that influence egg production and egg quality of laying hens. The chicken eye is superior to the eyes of other livestock, has the ability to see light wavelength (380-760 nm) in a narrow range and can discriminate light color (Prescott and Wathes, 1999). Apart from the eyes, the extra-retinal photoreceptors, located in the hypothalamus and in other sites of the brain, are involved in transduction of photo stimulation (Rozenboim *et al.* 1998). Previous studies show that the effects of monochromatic light on egg production and egg quality of laying hens are different. Li *et al.* (2019) reported that layer pullets preferred blue light the most compared with white, green and red lights. Similarly, in another study light color influenced egg production (Hassan *et al.* 2013). Mudhar and Tabeekh (2016) also demonstrated blue light increased FSH but red light increased LH hormone in laying hens. Zhang *et al.* (2017) suggested that breeders exposed to light of 740 nm improved performance of laying hens. Lewis *et al.* (2007) reported that pullets mature much earlier under light of short wavelength than under long wavelength light. Er *et al.* (2007) reported that egg weight was decreased significantly in hens that underwent red monochromatic light treatment, while eggshell quality

improved under green light treatment. Kim *et al.* (2010) claimed that red LED light causes improvement in eggshell thickness. On the other hand, it has been shown that monochromatic lights are not efficient in laying hens. Rozenboim *et al.* (1998) and Lewis *et al.* (2007) observed that light treatment did not affect the performance, egg production and egg quality of laying hens. Groot (2018) presented that different light spectra did not affect behavior of laying hens. Li *et al.* (2015) showed that monochromatic light did not affect the melatonin receptors in laying hens. Use of white or red LEDs did not cause any negative effect on egg production of laying hens (Borille *et al.*, 2013). Borille *et al.* (2015) demonstrated that the use of different colors of LED light in the rearing environment of laying hens in the second cycle did not affect egg production and quality.

Research on the effect of various light colors on broiler chicks have been also conducted previously. It has been demonstrated that blue and green lights improved growth performance and immune system (Cao *et al.*, 2008; Sultana *et al.*, 2013). Astuti *et al.* (2015) observed that body weights were increased under blue light treatment without adversely affecting stress indicators. Similarly, Fayoumi chickens maintained under blue light feel better welfare than exposed to yellow, red, green lights (Hesham *et al.*, 2018). Green monochromatic LED resulted in higher body weight marketed up to 7 or 5

week of age male or female respectively (Guevara *et al.*, 2015). Whereas Leigh *et al.*, (2017) reported that broiler performance was similar under white, green, blue, red LED lights.

Studies regarding the effects of monochromatic light on egg quality, egg cholesterol and blood biochemistry of laying hens are still contradictory. For this reason, the purpose of this study is to investigate the effects of monochromatic light on egg production, egg quality, egg cholesterol levels and some plasma metabolites of laying hens.

## MATERIALS AND METHODS

The present study was carried out in the February-March period of 2014, in the Layer Chicken Trial Unit of the Department of Animal Science, Faculty of Agriculture, University of Cukurova, Adana, Turkey. Prior to the study, thirty-two 47-week-old Brown-Nick layers were fed *ad libitum* with a standard layer diet for a week, egg production and egg weight were recorded daily. When the birds were 48 weeks old, they were divided into 2 experimental groups of similar mean body weight (1880 vs 1869 gm) and laying performance (egg weight 60.81 vs 60.62 gm; egg number 5.81 vs 5.86), comprising 16 birds each. During the experiment, two light regimes were employed in two experimental rooms, which were identical but different in environmental light color. The birds were housed in individual layer cages (40x40x40 cm) of three-tier battery blocks in a completely randomized design at a conventional ambient temperature (20-24°C) with a relative humidity of 50-60%. White (400-760 nm) and green (560 nm) fluorescent lamps (36 W) were used to illuminate Brown-Nick hens for 8 weeks (48-56 wk). Lamps were installed on the ceiling and side walls to achieve an average of 15 lx of light. The light intensity was measured with a luxmeter and light sources were equalized at the light intensity of 15 lx. A 16:8 hours light:dark photoperiod was employed. Light was supplied 16 hours (from 05:00 to 21:00) each day. The birds were fed a standard layer diet (17.8% CP; 2650 kcal/kg ME) obtained from a commercial feed industry. The experimental procedure was approved by the Animal Care and Ethic Committee of Cukurova University, Adana, Turkey. The performance of layers was determined by measuring the feed intake, egg mass, feed conversion ratio (egg mass:feed intake) and egg production (in house, number and weight) on a daily basis. The egg-shape index (width/length), egg weight, shell weight, shell thickness, yolk weight, albumen weight, albumen height, yolk index (weigh/height) and yolk color score L (lightness),  $a^*$  (redness),  $b^*$  (yellowness) (Hunter Lab) of each egg obtained on the third day of every week were calculated for egg quality parameters (32 eggs). Shell samples were taken from the top, middle and bottom of the eggs, shell

membranes extracted and the thickness of the shell was measured with a micrometer. The means of shell were calculated from recorded data before the statistical analysis.

Five birds from each group were randomly chosen, and blood samples were collected by puncture of the vena brachialis biweekly throughout the experiment. After centrifugation (1500 rpm, 5 minutes, 4°C), serums were carefully harvested and stored at -20°C until analysis. The serum concentrations of AST (Aspartat aminotransferaz), ALT (Alanin aminotransferaz), total protein, total cholesterol, albumin and globulin were measured with commercial kits on an automated KEYLAB LiquiVet Analyzer. The egg yolk cholesterol (6 eggs from each group) content (Boehringer Mannheim GmbH Biochemica 1995) was determined weekly using eggs from hens used for the blood biochemistry analysis with a specific kit. The egg yolks were hard-boiled for 10 minutes, homogenized and divided into 0.1g yolk fractions. After lipid extraction with isopropanol (4 mL, incubation at 37°C for 10 minutes and centrifugation at 3000 rpm for 5 minutes at 37°C), and the yolk cholesterol concentration was determined in the filtered samples by UV spectrophotometer by using commercial kits and calculated by the method of Boehringer Mannheim GmbH Biochemica.

Statistical data processing was performed using the software program SAS/STAT (SAS Institute, 2000). Statistical parameters were expressed as the arithmetic mean ( $\bar{x}$ ), and standard error (SE). Testing of significance of differences for the performance, quality of eggs and some blood parameters originating from two laying hens keeping lighting systems was carried out by using the t-test for independent samples. Significance of means was accepted at  $P \leq 0.05$ .

## RESULTS AND DISCUSSION

The effects of monochromatic light on performance, egg quality and some serum parameters of laying hens are summarized in Table 1, 2 and 3 respectively.

The results show that green light application did not have any significant ( $P > 0.05$ ) effect on feed intake, feed conversion ratio, egg production, total number of eggs, total egg weight, average egg weight, egg yolk cholesterol level, serum ALT, AST, total cholesterol, total protein and globulin concentration of laying hens. However, green light application affected egg yolk color  $a^*$  ( $P \leq 0.01$ ) and  $b^*$  values ( $P \leq 0.05$ ), so that the yolk has become darker but decreased serum albumin concentration ( $P \leq 0.05$ ).

Literature on the effect of light color on laying hens performance, egg quality and blood characteristics are very limited. In the study of Hassan *et al.* (2013, 2014) red and green light combination treatment

improved feed intake and feed conversion ratio in laying hens. Riber (2015) observed that final live body weight and the yield from the breast muscle tenders were increased in cold white (6,065 K) color temperature compared to neutral white (4,100 K) color temperature. Ke *et al.* (2011) determined a higher body weight in broilers illuminated with blue LED light compared to with red, green and blue LED lights. Rozenboim *et al.* (1999) reported that green light stimulate growth at early age, whereas blue light stimulate growth in older birds. Rozenboim *et al.* (2014) also suggest that shifting birds to a different light environment at 10 or 20 day of age might further stimulate growth. Light wavelength is very important factor in regulating development of sexual organ and laying performance in breeders exposed to light of 740 nm had better performance in egg laying (Zhang *et al.*, 2017). However, according to some authors (Lewis *et al.*, 2007; Gongruttananun, 2011), there is no differences in laying performance parameters. Huber-Eicher *et al.* (2013) indicated that in laying hens the light color did not affect the body weight gain, feed consumption and age at first lay but red light showed better early laying performance. According to Borille *et al.* (2013) the effect of light sources or period on feed intake is associated to birds had the same visual sensitivity to all tested light sources, and did not change their feeding behavior as a function of light source. Similarly, we did not find any significant differences of performance under green light when compared with control group (white light). Inconsistent responses of poultry to expose to light may be related mainly to inter-strains differences, age, sex, different light combinations, duration of time exposed to the light.

The monochromatic light effects on the egg quality results in poultry are still contradictory. Egg weight (Rozenboim *et al.*, 1998; Svobodová *et al.*, 2015), eggshell breaking strength, albumin height, haugh unit and yolk color (Hassan *et al.*, 2013) were not influenced by either the monochromatic or the combined light color. On the other hand red light produces smaller eggs, while the white light makes possible to produce large eggs (Er *et al.*, 2007). Egg weight has already tended to increase numerically under green light when compared with white light in the current study.

Yang *et al.* (2016) showed that red light has expedited sexual maturation and promotes egg production in birds. Birds maintained under green light produced fewer eggs and better egg quality than exposed to white, red and blue lights (Li *et al.*, 2014). The eggshell quality in green light was better than those in white, red and blue lights (Er *et al.*, 2007; Hassan *et al.*, 2014; Li *et al.*, 2014). Er *et al.*, (2007) determined a higher color interactions in laying hens reared under white light compared to those reared under blue, green and red light. Our results indicated that egg yolk color  $a^*$  and  $b^*$  values increased significantly in hens that underwent green light

treatment. Yolk color primarily depends on the intake of plant pigment in diet (Long *et al.*, 2014). Carotenoids contain large numbers of double bonds in their molecules, and as a result, they may oxidize, depending on storage time, room temperature and the effect of lighting (Barbosa *et al.*, 2011). Harvesting, milling, extruding, and separating to the flour fraction of corn can result in total carotenoid loss (Ortiz *et al.*, 2018). And these loses can decrease the absorption and deposition of carotenoids by birds in egg yolk (Moreno *et al.*, 2016).

Our results show that yolk color  $a^*$  and  $b^*$  values increased under green light treatment, possibly due to the improvement in absorption and deposition of carotenoids or the higher pigment contents in the green light spectra during the rearing period. The other possible reason might be that green light treatment enhanced some pigment synthesis in yolk. In addition, light influences formation and isomerization of carotenoids which is cis-trans isomerization leads to a decrease of color intensity (Schieber and Carle, 2005). Probably, the cis-trans isomerization increase in yolk color when exposed to green light in laying hens. However, the modes of action of the green light color on the egg quality are still unclear. The effect of light color on the lipid response is poorly understood. Ibrahim *et al.* (2012) founded that green light group had significantly higher cholesterol level than birds exposed to yellow light color. Firouzi *et al.* (2014) also demonstrated that blue light decreased serum glucose, cholesterol and triglyceride levels and created a calming effect on birds. Elevated serum glucose, cholesterol and triglyceride levels are as indicators of stress (Thaxton and Puvadolpirod, 2000). In the present study, exposure to green light color did not affect both egg yolk and serum cholesterol levels. The level of serum cholesterol tended to increase numerically while yolk cholesterol level tended to decrease numerically in laying hens housed in green light.

AST, ALT metabolites provide information about the functional capacity of liver that are involved in a particular metabolic pathway. AST is the most sensitive indicator of liver disease in birds (Speer, 2015). Damage of liver causes the release of AST or ALT enzymes into the bloodstream. Hassan *et al.* (2016) mentioned that monochromatic lights on broiler chicks did not alter liver enzymes (AST and ALT) in a healthy flock. Olanrewaju *et al.* (2015) mentioned that using two color temperatures of LED light bulbs did not influence plasma AST and ALT concentrations in broiler. In our results, green light color did not affect plasma AST (165 vs. 127), ALT (41.60 vs. 24.20) content but there is a numerical decrease when exposed to green light in laying hens. This numerical improvement is probably due to the relaxing caused by green light on laying hens.

As known that albumin is the main constituent of total protein which is made specifically by the liver (McDonald *et al.*, 2011). In chronic liver disease or

nephrotic syndrome are decreased albumin levels. Low albumin levels can also be seen in inflammation, shock, and malnutrition. Albumin acts as the main reservoir of the protein and plays a role in colloidal osmotic pressure and acid-base balance; it acts as a carrier for small molecules such as vitamins, minerals, hormones and fatty acids (Wu, 2017). Serum albumin concentrations are indicators of the protein status of the blood. The results of this study show that serum albumin is significantly reduced in chickens grown under green light compared to chickens grown under white light. Smith (1978) suggested that serum albumin is the major protein reserve of the laying hen, which is degraded in response to the needs of protein synthesis in the oviduct. Stress causes hypoalbuminemia in animals. A reduction of plasma

albumin concentration can be consequence of reducing albumin synthesis in liver (Ruot *et al.*, 2000). Decreased serum albumin concentration was observed in this study. Total protein and globulin levels decreased numerically in laying hens exposed to green light. Serum total protein and albumin of broilers under Blue-Green mix light, globulin under Green Light and uric acid under Blue Light and triglyceride under Green Light have improved (Abu Tabeekh and Mudhar, 2016). Firouzi *et al.* (2014) found differences in total serum protein concentration between broilers exposed to yellow monochromatic light and those exposed to blue and red lights. However, they did not make any comments on the result. The data concerning the effect of green light are still contradictory.

**Table 1. Effects of monochromatic light on productive performance.**

Parameters	White Light (mean±SE)	Green Light (mean±SE)	T <sup>y</sup>
Initial Body Weight (g)	1880.63±40.55	1869.69±44.48	0.8570
Final Body Weight (g)	1853.75±43.55	1806.88±46.36	0.4669
Total Feed Intake (FI; g/bird/56 days)	6451.25±91.64	6390.50±96.94	0.6521
Egg Mass (EM; g/bird/56 days)	3184.86±56.31	3214.07±61.93	0.7296
Feed Conversion Ratio (FI/EM)	2.03±0.04	2.00±0.04	0.4986
Mean Egg Weight (g/day)	61.64±0.73	62.41±0.81	0.4854
Number of Eggs (bird/56 days)	52.38±0.48	52.06±0.67	0.7069

<sup>y</sup>: Mean values were significantly different at  $p \leq 0.01$  and  $p \leq 0.05$ .  
SE: Standard error

**Table 2. Effects of monochromatic light on egg quality traits.**

Parameters	White Light (mean±SE)	Green Light (mean±SE)	T <sup>y</sup>
Egg Weight (g/egg)	56.87±1.01	57.67±1.04	0.7291
Shell Weight (g/egg)	7.04±0.09	7.22±0.07	0.1397
Yolk Weight (g/egg)	15.78±0.28	16.30±0.36	0.2637
Albumen Weight (g/egg)	34.05±1.09	34.15±1.05	0.9481
Shape Index (%)	76.64±0.58	78.05±0.56	0.0907
Egg Yolk Index	46.44±0.36	45.81±0.26	0.1712
Albumen Index	8.04±0.44	7.76±0.28	0.5985
L (lightness)	59.20±0.27	59.36±0.34	0.7133
a (redness)	10.44±0.15	11.33±0.14	<b>0.0001</b>
b (yellowness)	56.82±0.59	58.55±0.53	<b>0.0370</b>
Haugh Unit	80.99±1.56	80.34±1.35	0.7532
Breaking Strength (kg/cm <sup>2</sup> )	4.59±0.17	4.86±0.15	0.2351
Mean Shell Thickness (µm)	374.53±3.97	378.99±3.97	0.3771
Egg Yolk Cholesterol Content (mg/g egg)	19.057±0.24	18.709±0.07	0.2348

<sup>y</sup>: Mean values were significantly different at  $p \leq 0.01$  and  $p \leq 0.05$ .  
SE: Standard error

Table 3. Effects of monochromatic light on some serum parameters in laying hens.

Serum Parameters	White Light (mean±SE)	Green Light (mean±SE)	T <sup>y</sup>
Alanine Aminotransferase, ALT (U/L)	41.60±9.37	24.20±8.44	0.849
Aspartate Aminotransferase, AST (U/L)	165.33±17.07	127.71±20.58	0.844
Total Cholesterol (mg/dL)	106.80±10.13	118.86±13.98	0.668
Total Protein (mg/dL)	5.13±0.27	3.98±0.35	0.769
Albumin (mg/dL)	1.50±0.08	1.22±0.04	<b>0.032</b>
Globulin (mg/dL)	3.63±0.20	2.76±0.33	0.388

<sup>y</sup>: Mean values were significantly different at  $p \leq 0.01$  and  $p \leq 0.05$ .

SE: Standard error

**Conclusion:** In conclusion, green light color did not influence laying performance and egg quality except egg yolk pigmentation. Green light application increased egg yolk color  $a^*$  and  $b^*$  values, but decreased serum albumin concentration. The results suggest that green light application has the potential to increase egg quality in terms of yolk pigmentation.

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