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Stress and fear responses of slow-growing broiler chickens to light stimulation initiated at different phases of embryonic development

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Abstract Photo-incubation can influence the fear and stress responses of poultry. However, it is unclear how photo-stimulation initiated at different phases of development influences the welfare status of slow-growing broiler birds. 500 Sasso eggs were assigned to 4 treatments; some were incubated in the dark throughout incubation (TA), while TB, TC and TD were photo-stimulated (12L:12D) from days 1, 7, and 14 of incubation, respectively, until hatch using a 6,500k LED at 788 clux intensity. Birds were raised in 5 replicates per treatment with 16 birds per replicate using a 6,500k LED (at 28 clux) and a photoperiod of 16L:8D. Fear (emergence, tonic immobility, isolation and inversion tests) and stress response (physical asymmetry) of 10 birds per treatment were examined. At the end of the three-week brooding, all parameters measured were not significantly influenced ($P > 0.05$) by the onset of photo-incubation. At slaughter age (12 weeks), physical asymmetry was significantly higher ($P < 0.05$) in TA compared to the other treatments. The frequency of isolation vocalisation was significantly lower ($P < 0.05$) in TB compared to TA, and latency to rightness during tonic immobility was significantly higher ($P < 0.05$) in TA compared to the other treatments. Latency to emerge was significantly longer ($P < 0.05$) in TA compared to TC and TD. The frequency of wing flaps during inversion was significantly higher ($P < 0.05$) in TA and TD. Conclusively, photo-incubating eggs reduce stress and fear, and initiating photo-incubation during the first phase of incubation is more beneficial.

Keywords: incubation, light, welfare

1. Introduction

Animal welfare, which is of utmost importance in the poultry industry, could be influenced by prenatal and perinatal experiences. Environmental conditions during incubation, such as darkness, have been identified as a potential stressor impacting poultry welfare (Archer and Mench 2013). Commercial incubation is often done in the dark, perhaps due to concerns about electricity consumption or hatching problems that could arise due to secondary heating from the light source (Archer and Mench 2014). The possibility of secondary heating from light-emitting diode (LED) bulbs has been ruled out, and no detrimental effects of LED bulbs on poultry birds have been highlighted in the literature (Oso et al 2022ab). It has also been demonstrated that hatching performance is positively influenced by photo-incubation (El-Sabrout 2017), although disruptive synchronisation of the hatch is not improbable depending on the strain (Hannah et al., 2019).

In natural incubation, eggs are exposed to natural light whenever hens leave the nest for food, although it has been proven that the exposure period is inadequate to modulate post-hatch fear and stress responses (Archer and Mench 2014).

In artificial incubation, it has been demonstrated that adequate light exposure can positively impact stress and fear behaviour, and these benefits transcend early post-hatch life and have been reported for up to six (6) weeks in fast-growing broilers (Huth and Archer 2015, Archer 2018). In the literature, two possible mechanisms have been identified as the underlining factors responsible. Firstly, pre-hatch lateralisation occurs in the brain when the eye is photo-stimulated during the developmental process (Rogers 2000). It is important to note that the presence of light induces cerebral lateralisation, and the direction of lateralisation is determined by the timing of the exposure (Archer and Mench 2014). Secondly, Ozkan et al (2012b) suggested that melatonin rhythms established during the last phase of embryonic development due to photo-stimulation can alter physiological responses of the hypothalamic-pituitary-adrenal (HPA) axis, thus improving stress susceptibility. Melatonin rhythm established during incubation can affect behavioural rhythm, decrease stress and anxiety, and

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corticosterone production (Ozkan et al 2012b). Thus, there is a greater possibility of influencing asymmetries (Archer et al 2009). These highlighted mechanisms suggest that the stage of embryonic development at the time of initiation of photo-incubation is an important factor when considering welfare. Various timings or onsets of photo-incubation have been used as a tool to influence stress and fear levels in fast-growing broiler birds (Archer and Mench 2013, 2014), but not in slow-growing. Peixoto et al (2020) highlighted that both fear and stress responses in poultry birds are highly dependent on genotype and that the strain effect may also have a role to play due to phenotypic diversity arising from intense selection for productivity. Given this, it is hypothesised that initiating light exposure at different stages of development might invoke different fear and stress responses in slow-growing broilers.

2. Materials and Methods

2.1. Experimental site and Ethical approval

The experiment was carried out at the Regional Centre of Excellence in Avian Sciences (CERSA), University of Lome, Togo. The animal care guidelines recommended by the Animal Ethics Committee of the University of Lome in Togo were followed (ref: 008/2021/BC-BPA/FDS-UL). The authors have read the policy relating to animal ethics and confirm that this study complies.

2.2. Experimental design

Five hundred hatching eggs of the Sasso broiler (naked neck) strain were obtained from 75-week-old breeders from the research farm of CERSA, University of Lome. Eggs of 52.5±2.5 g average weight were randomly allotted to 4 incubation treatments, 125 eggs per treatment: conventional incubation in the dark (TA); light exposure from day 1-21 (TB); light exposure from day 7-21 (TC) and light exposure from day 14-21 (TD) using a photoperiod of 12 hours of light and 12 hours of darkness (12L: 12D). A 6500k Correlated Colour Temperature (CCT) LED bulb with an intensity of 788 clux at the egg level was used. The light intensity was measured using a poultry lux meter.

2.3. Experimental management

At hatch, 320 chicks were weighed and divided into five replicates per treatment (16 birds per replicate) while maintaining their incubation treatment groups in a completely randomised design (CRD). The birds were supplied with a 6,500k LED and a light intensity of 28 clux at bird level. The birds were brooded on a photoperiod of 23L:1D and gradually decreased until 16L:8D was achieved. The brooding temperature ranged between 30-32 ºC for the first six days before it was gradually reduced until 27-28 ºC was achieved at three weeks. At four weeks of age, the birds were moved to an outgrow pen: a non-environmentally controlled open-sided facility where the birds were exposed to 12 hours of natural light and 4 hours of artificial light from 6 pm -10 pm daily at an intensity of 28 clux. Birds were raised on deep litter using wood shavings with a 4 cm depth and a stocking density of 11 m^2 per bird. The birds were fed *ad libitum* with a starter diet containing metabolisable energy of 2962.37 kcal/kg and 21.08% crude protein. The finisher diet contained 3044.81 kcal/kg and 18.26% crude protein. Vaccinations and medications were duly administered across the treatments.

2.4. Stress (Physical asymmetry)

Stress response was carried out using a minimally invasive approach. At weeks 3, 7, 10 and 12, the physical asymmetry of 10 birds per treatment (2 birds from each of the five replicates) was assessed using the method of Archer and Mench (2013). Birds were randomly selected for the test at week three and identified with tags for subsequent tests. The middle toe length, metatarsal length, and metatarsal width measurements were taken for both the right and left legs using a digital calliper. The physical asymmetry score was calculated using the formula:

$$\text{Physical asymmetry score} = (|L\text{-}R|_{MTL} + |L\text{-}R|_{ML} + |L\text{-}R|_{MW})/3$$

where: $|L\text{-}R|_{MTL}$ is the absolute difference between left and right middle toe length; $|L\text{-}R|_{ML}$ is the absolute difference between left and right metatarsal length; and $|L\text{-}R|_{MW}$ is the absolute difference between left and right metatarsal width.

2.5. Fear test

For each fear test, ten birds were selected from each treatment (2 birds from each of the 5 replicates) and labelled for easy identification. The test was carried out on the ten birds in an empty pen (out of sight of conspecifics). The birds were collected at the 3rd, 7th, 10th, and 12th weeks of age.

2.6. Emergence

A method described by Huth and Acher (2015) was slightly modified. A perforated basket was wrapped in brown paper and slanted on its side. The sliding cover of the basket was used as a door. One bird at a time was placed in the basket and closed. The observer sat behind the basket with a timer. After 1 minute of closure, the basket door was opened while the observer monitored the time of emergence. To calculate the latency to emerge, the 1 minute of basket closure was subtracted from the time of emergence. Birds were allowed to remain in the basket for as long as they desired.

2.7. Isolation

A day after the emergence tests, an isolation test was carried out using a modified method by Huth and Archer (2015). The birds marked for the isolation test were placed in a large container and moved to the entrance of an empty pen. A tall bucket was placed in the middle of the empty pen, and one bird was selected at a time and placed in the bucket for 2 minutes. The number of vocalisations made by the isolated bird within 2 minutes was counted and recorded.

2.8. Inversion

This was conducted a day after the isolation test. The inversion test was conducted with a slight modification to the methods discussed in Newberry and Blair (1993) and Archer and Mench (2014b). Each bird was held upright, and one hand supported the breast while the other held the two legs. The bird was inverted, and the hand on the breast was removed, so the bird hung freely upside down for 2 minutes. The wing flap was video recorded, and the number of occurrences of wing flaps was noted. More prolonged and intense flapping indicated more fearfulness (Newberry and Blair 1993).

2.9. Tonic immobility

A day after the inversion test, tonic immobility was carried out on the birds marked for tonic immobility. The method of Huth and Archer (2015) was slightly modified. A hollow container with a 25 cm diameter was placed on a table. One bird at a time was placed on the container with the head of the bird covered with one hand while the breast was held with the other hand for a minute to induce tonic immobility, after which the hand was removed, and a timer was started. Whenever the bird righted itself under 10s, the timer was reset, and the bird was placed on its back again to induce. After a maximum of 3 failed induction attempts per bird, both the latency to first head movement and the latency to rightness were recorded as zero. The time of first head movement and eventual rightness were recorded for successful induction attempts. However, to calculate latency, the one minute of tonic inducement was subtracted from the recorded time. Longer times to the first head movement and righting indicated more fearfulness (Jones 1986). The time interval between the first head movement and rightness was calculated by finding the differences between the two parameters.

2.10. Statistical analysis

The data were subjected to a one-way analysis of variance (ANOVA). A *P*-value of less than 0.05 ($P < 0.05$) was considered significant. Tukey test was used for comparison of means. Data were subjected to the Shapiro-Wilk test for normality and Levene's test for homogeneity of variance. The data met the assumptions of ANOVA; thus, no transformation was required. All analysis was carried out using Minitab 17 (2017).

3. Results

3.1. Physical asymmetry

Physical asymmetry was not significantly influenced ($P > 0.05$) by incubation treatment at 3 and 7 weeks of age (Table 1). However, at ten weeks of age, physical asymmetry was significantly lower ($P < 0.05$) in TB compared to TA. At 12 weeks of age, physical asymmetry was significantly higher ($P < 0.05$) in TA compared to the other treatments.

3.2. Isolation

In Table 2, isolation vocalisation was not significantly ($P > 0.05$) influenced by the onset of photo-incubation at week three and week 10 of age but was significantly ($P < 0.05$) influenced at weeks 7 and 12. Isolation vocalisation was significantly higher ($P < 0.05$) in TA compared to TB at weeks 7 and 12.

Table 1 Effect of varying onset of photo-incubation on post hatch physical asymmetry.

Treatment	Age of birds			
	Week 3	Week 7	Week 10	Week 12
TA	0.18	0.53	1.01^{a}	1.60^{a}
TB	0.04	0.19	0.38^{b}	0.56^{b}
TC	0.04	0.24	0.49^{ab}	0.79^{b}
TD	0.10	0.35	0.66^{ab}	0.83^{b}
SE	0.05	0.09	0.134	0.14
P-Value	0.060	0.567	0.013	0.000

[a,b]Different letters indicate significant differences between means within columns ($P < 0.05$); SE: Standard error.
TA- Dark incubation, TB- Exposed to light from day 1, TC- Exposed to light from day 7, TD- Exposed to light from day 14.

3.3. Tonic immobility

The tonic immobility responses are shown in Table 3. Latency to first head movement and latency to rightness were not significantly ($P > 0.05$) influenced by the onset of photo-incubation in the 3rd week of life. However, at weeks 7 and 12, the latency to rightness and latency to the first head movement were significantly ($P < 0.05$) influenced by the onset of photo-incubation. At week 7, the first head movement latency was significantly lower ($P < 0.05$) in TB and TC compared to TA. Latency to rightness was significantly ($P < 0.05$) longer in TA compared to the other treatment groups. Latency to the first head movement was not significantly ($P > 0.05$) influenced at week 10. However, latency to rightness was significantly higher ($P < 0.05$) in TA compared to TB and TC. At the 12th week of age, latency to the first head movement was significantly higher ($P < 0.05$) in TA and TD compared to TB and TC. At the same time, latency to rightness was significantly higher ($P < 0.05$) in TA in contrast to the other treatment groups.

Table 4 reveals that the time intervals between head movement and rightness were not significantly ($P > 0.05$) influenced by the onset of photo-incubation at the 3rd, 7th, 10th, and 12th weeks of age.

3.4. Emergence and Inversion

Table 5 shows the effect of varying the onset of photo-incubation on the latency to emerge and the frequency of wing flaps. The photo-incubation treatment groups had no significant influence ($P > 0.05$) on latency to emerge during the emergence test at the 10th week of life. However, latency to emerge was significantly ($P < 0.05$) influenced by the onset of photo-incubation at weeks 7 and 12. At 7 weeks of age, latency to emerge was significantly shorter ($P < 0.05$) in TB and TC compared to TA, but at week 12, latency to emergence was significantly longer ($P < 0.05$) in TA compared to TC and TD. At week 12, the frequency of wing flap during inversion was significantly higher ($P < 0.05$) in TA and TD.

Table 2 Effect of varying onset of photo-incubation on isolation vocalisations.

	Age of birds			
Treatment	Week 3	Week 7	Week 10	Week 12
TA	43.50	52.50^{a}	25.20	182.40^{a}
TB	21.67	6.67^{b}	2.17	52.67^{b}
TC	37.00	30.33^{ab}	9.50	61.00^{ab}
TD	40.20	37.70^{ab}	14.50	52.67^{b}
SE	17.90	12.90	13.00	62.60
P-Value	0.635	0.015	0.369	0.012

^{a,b,c}Different letters indicate significant differences between means within columns ($P < 0.05$); SE: Standard error.
TA- Dark incubation, TB- Exposed to light from day 1, TC- Exposed to light from day 7, TD- Exposed to light from day 14.

Table 3 Effect of varying onset of photo-incubation on post hatch tonic immobility response.

	Latency to first head movement				Latency to rightness			
Treatment	Week 3	Week 7	Week10	Week 12	Week 3	Week 7	Week 10	Week 12
TA	34.80	270.00^{a}	218.20	108.20^{a}	41.30	272.00^{a}	278.20^{a}	223.10^{a}
TB	0.00	56.00^{b}	35.30	20.67^{c}	0.00	80.00^{b}	38.80^{b}	39.33^{b}
TC	0.00	60.70^{b}	52.70	53.83^{b}	0.00	71.00^{b}	85.70^{b}	67.00^{b}
TD	12.80	89.20^{ab}	125.20	91.33^{a}	24.00	100.00^{b}	171.50^{ab}	101.83^{b}
SE	13.70	72.70	68.40	11.40	20.40	75.00	79.00	57.50
P-Value	0.062	0.023	0.057	0.000	0.155	0.046	0.033	0.020

^{a,b,c}Different letters indicate significant differences between means within columns ($P < 0.05$); SE: Standard error.
TA- Dark incubation, TB- Exposed to light from day 1, TC- Exposed to light from day 7, TD- Exposed to light from day 14.

Table 4 Effect of varying onset of photo-incubation on time interval between first head movement and rightness.

	Age of birds			
Treatment	Week 3	Week 7	Week10	Week 12
TA	6.50	1.00	60.00	114.80^{a}
TB	0.00	24.70	3.50	18.64^{b}
TC	0.00	11.00	33.00	13.17^{b}
TD	11.20	10.80	46.30	10.20^{b}
SE	8.56	19.60	40.30	46.20
P-Value	0.504	0.692	0.553	0.024

^{a,b,c}Different letters indicate significant differences between means within columns ($P < 0.05$); SE: Standard error.
TA- Dark incubation, TB- Exposed to light from day 1, TC- Exposed to light from day 7, TD- Exposed to light from day 14.

4. Discussion

Directional symmetry and anti-symmetry are linked to genetics, while stress primarily influences fluctuating asymmetry. Physical asymmetry is a non-invasive measure of developmental stress (Archer et al 2009). It is a combined asymmetry measurement that gives a more reliable effect of a stressor than a single measurement (Leung et al 2000) or any other form of asymmetry. In this study, physical asymmetry was comparable in all treatments in weeks 3 and 7. Our findings at these ages contradict that of researchers who found that at two weeks of age (Archer and Mench 2013) and six weeks old (Archer and Mench 2014), birds exposed to LED from day 1 of incubation had lower asymmetry compared to those incubated in the dark. In week 10, physical asymmetry was lower in TB compared to TA. At ten weeks old, physical asymmetry was distinct in TB, unlike TC and TD. At week 12, physical asymmetry was significantly lower in all photo-incubated treatments compared to TA. The observed trend suggests that asymmetry was present even early, but the differences became evident as they grew. The effect was more pronounced in TB earlier than in TC and TD. Archer et al (2009) noted that asymmetry differences emerge post-hatch as birds react differently to stressors throughout the growing period. This suggests that TB treatment is more efficient in combating early stressors. The result of our experiment at week 10 corresponds to that observed by Archer and Mench (2014) in 6-week-old fast-growing broilers. This also suggests that the rate of development of each strain may also determine the level of asymmetry, so differences in physical asymmetry may emerge at an earlier age in fast-growing birds than in slow-growing ones.

Table 5 Effect of varying onset of photo-incubation on latency to emerge and inversion test.

	Latency to emerge			Wing flap
Treatment	Week 7	Week10	Week 12	Week 12
TA	753.00[a]	1017.00	514.80[a]	11.75[a]
TB	63.50[c]	704.42	410.50[ab]	3.08[b]
TC	106.70[bc]	645.10	400.80[b]	4.15[b]
TD	638.00[ab]	385.00	390.00[b]	10.07[a]
SE	266.00	361.00	54.00	1.61
P-Value	0.032	0.400	0.010	<0.000

[a,b,c]Different letters indicate significant differences between means within columns ($P < 0.05$); SE: Standard error.
TA- Dark incubation, TB- Exposed to light from day 1, TC- Exposed to light from day 7, TD- Exposed to light from day 14.

Isolation from familiar conspecifics is capable of inducing fear in animals. The Isolation test works on the principle of social interaction and loneliness. Isolation often results in changes in behavioural repertoires, such as vocalisation. When an animal is separated from the flock, there is usually a vocalisation in an attempt to re-connect with the conspecifics. Increased vocalisation frequency during isolation has been associated with fearfulness, although alarm calls and normal vocalisation have not been differentiated during this test. No difference was observed in isolation vocalisation at week three and week 10. This is similar to the findings of Huth and Archer (2015), who at one week recorded a comparable frequency of vocalisation in birds isolated from photo-incubation treatment compared to the dark incubated.

In contrast, at three weeks, Archer and Mench (2014) recorded lower vocalisation in those photo-stimulated from the first day of incubation compared to those exposed from day 14 and those incubated in the dark. In our experiment, at week 7, birds isolated in TA made more frequent vocalisations than those isolated in TB. In contrast, those isolated in TC and TD were comparable to all the others. At week 12, birds isolated from TB and TD groups made vocalisations less frequently than TA. This suggests that photo-incubation reduces fear associated with separation from conspecifics, but exposing eggs to LED from the first day of incubation seems to reduce social fear.

Tonic immobility is another fear measure in poultry that can be described as a reflex characterised by temporary motor inhibition or inactivity in response to extreme fear. The longer the period of tonic immobility, the higher the level of fear. Huth and Archer (2015) highlighted tonic immobility as the most common fear measure in poultry, and it has been established that this could be influenced by light. On week 7, birds in TB and TC groups had less latency to their first head movements during tonic immobility, signifying that when faced with sudden fear, they tend to make head movements faster, perhaps to monitor the environment. Latency to eventual rightness was comparable in all photo-incubated treatments at week 7, and the same result was obtained at week 10. This disagrees with the findings of Archer and Mench (2014), who revealed that at five weeks, latency to rightness was shorter in those exposed to light within the first seven days compared to those exposed to light from day 14 and those incubated in the dark. In our study, at week 12, birds in TB made their first head movements faster than those in the other treatments, while those in TC made their first head movements earlier than TA and TD. At this age, latency to rightness was less across all photo-incubated treatments. All photo-incubated birds overcame tonic immobility in a shorter time than those in the dark. However, a more consistent trend was established in the TB and TC groups compared to the TD group. This suggests that exposing eggs to LED within the first seven days of incubation may be more beneficial to fear associated with invasion, intrusion, or sudden disturbance. The time interval between the first head movement and eventual rightness was significantly shorter in light-incubated treatments, suggesting that after taking cognisance of the environment and head mobility is initiated, the birds could overcome immobility faster than those incubated in the dark.

Fear of predation leads to predator avoidance behaviour (Archer 2018). The "emergence test" is one of the measures used to evaluate anti-predator behavioural repertoires in chickens. By instinct, hiding from predators is an animal's behavioural response in a defensive situation. The latency to emerge from hiding after an invasive stimulus suggests the level of fear. The higher the latency to emerge after introducing the stimuli, the greater the fear experienced by the bird. The emergence latency was significantly shorter in TB compared to TA and TD at week 7. This agrees with the report of Archer and Mench (2014) at three weeks of age.

On the contrary, Huth and Archer (2015) reported comparable results in photo-incubated and dark incubated treatment at one week of age. In our study at week 12, emergence was shorter in TC and TD compared to TA. It is obvious that photo-incubation influenced emergence, but there was a consistency in the TC group compared to TB and TD groups.

Practically, reducing fear responses may make handling and transportation of chickens easier (Huth and Archer 2015), and chickens can easily adapt to a new environment (Ozkan et al 2012ab). The frequency and intensity of wing flaps during inversion give an idea of how chickens respond to fear associated with being caught. Catching is an unavoidable activity, especially when there is a need for crating and transportation to the slaughterhouse. A higher occurrence of wing flaps shows greater fear. At slaughter age, birds in TA and TD treatment groups birds were more fearful than those in TB and TC. This aligns with the results obtained by Archer and Mench (2014) at the slaughter age of fast-growing broiler chickens.

Generally, post-hatch fear responses were better in chickens photo-stimulated early during incubation than in those exposed to light from day 14. It suggests that the lateralisation that occurred from the late exposure may not be enough to produce a consistent trend of fear response but could nonetheless reduce fear at some point. This corroborates the assertion of Archer and Mench (2014), who discovered that regardless of the time of photo-exposure, all light-incubated chicks were lateralised and the dark-incubated were not, although the pattern of lateralisation differs based on the timing of exposure. This suggests that visual pathways might not be the key mechanism influencing fear response since the photo-related lateralisation pattern differs based on the timing of exposure.

5. Conclusions

It is of benefit to photo-stimulate eggs during incubation as they reduce the physical asymmetry level response at slaughter. Still, early exposure from the first day of incubation may be more beneficial to birds. Exposing eggs to LED within the first seven days of incubation appears to reduce the level of fear associated with intrusion and catching or human handling. Emergence from hiding was more favoured by exposing eggs to LED on day seven compared to the other onsets of photo-incubation. Exposing eggs to light within the first incubation phase reduces post-hatch fear, and the mechanism(s) involved should be further researched.

Conflict of Interest

The authors have no competing interests.

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References

Archer GS (2018) Color temperature of light-emitting diode lighting matters for optimum growth and welfare of broiler chickens. Animal 12:1015-1021.

Archer GS, Mench JA (2013) The effects of light stimulation during incubation on indicators of stress susceptibility in broilers. Poultry Science 92:3103–3108.

Archer GS, Mench JA (2014) Natural incubation patterns and the effects of exposing eggs to light at various times during incubation on post-hatch fear and stress responses in broiler (meat) chickens. Applied Animal Behavior Science 152:44-51.

Archer GS, Shivaprasad HL, Mench JA (2009) Effect of providing light during incubation on the health, productivity, and behavior of broiler chickens. Poultry Science 88:29-37.

El-Sabrout K (2017) Effect of LED lighting during incubation of fayoumi eggs on hatchability and chick performance. Kafkas Universitesi Veteriner Fakultesi Dergisi 23:1007-1009.

Hannah WA, Astatkie T, Rathgeber BM (2019) Hatch rate of laying hen strains provided a photoperiod during incubation. Animal 14:353-359.

Huth JC, Archer GS (2015) Comparison of two LED light bulbs to a dimmable CFL and their effects on broiler chicken growth, stress, and fear. Poultry Science 94:2027-2036.

Jones RB (1986) The tonic immobility reaction of the domestic fowl: a review. World's Poultry Science Journal 42:82-96.

Leung B, Forbes MR, Houle D (2000) Fluctuating asymmetry as a bioindicator of stress: Comparing efficacy of analyses involving multiple traits. American Naturalist 155:101–115.

Newberry RC, Blair R (1993) Behavioral-responses of broiler-chickens to handling - effects of dietary tryptophan and 2 lighting regimens. Poultry Science 72:1237-1244.

Oso OM, Metowogo K, Oke OE, Tona K (2022a) Evaluation of light emitting diode characteristics on growth performance of different poultry species: a review. World's Poultry Science Journal 78:337-351.

Oso OM, Metowogo K, Oke OE, Tona K (2022b) Influence of LED bulb on reproductive and production performance of different poultry species: a review. World's Poultry Science Journal 78:515-529.

Ozkan SS, Yalcin E, Babacanoˇglu S, Uysal F, Karada S, Kozanoglu H (2012) Photoperiodic lighting (16 hours of light: 8 hours of dark) programs during incubation: 2. Effects on early post hatching growth, blood physiology, and production performance in broiler chickens in relation to post hatching lighting programs. Poultry Science 91:2922–2930.

Ozkan S, Yalcin S, Babacanooglu E, Kozanooglu H, Karada S, Uysal F (2012) Photperiodic lighting (16 hours of light: 8 hours of dark) programs during incubation: 1. Effects on growth and circadian physiological traits of embryos and early stress response of broiler chickens. Poultry Science 91:2912–2921.

Peixoto MR, Karrow NA, Newman A, Widowski TM (2020) Effects of maternal stress on measures of anxiety and fearfulness in different strains of laying hens. Frontiers in Veterinary Science 7:128.

Rogers LJ (2000) Evolution of hemispheric specialization: advantages and disadvantages. Brain and Language 73:236–253.