



Full-Length Article

Dark brooders during rearing affect fearfulness but not production parameters of broiler chickens

Sara Forslind ^a, Emily M. Leishman ^{b,*}, Carlos E. Hernandez ^a, Anja B. Riber ^b, Helena Wall ^a, Eva Wattring ^c, Harry J. Blokhuis ^a

^a Department of Applied Animal Science and Welfare, Swedish University of Agricultural Sciences, SE-750 07 Uppsala, Sweden

^b Department of Animal and Veterinary Sciences, Aarhus University, Blichers Allé 20, DK-8830 Tjele, Denmark

^c Department of Microbiology, Swedish Veterinary Agency, SE-75189 Uppsala, Sweden

ARTICLE INFO

Keywords:

Antibody production
Environmental enrichment
Fear
Poultry
Production

ABSTRACT

Dark brooders are often suggested as enrichment for broiler chickens because they promote natural motivated behaviors compared to conventional barren environments in broiler production. Although the use of brooders is thought to convey benefits for animal welfare, it is not well described how they might affect parameters related to production and health. The objective of this study was to investigate if providing dark brooders affects production, health, behavior, and welfare compared to a control group without dark brooders. Six pens of each treatment were used, keeping 60 mixed-sex chickens (Ross 308) per pen for a full rearing period of 35 days. Data on growth, mortality, gait score, litter quality, infectious bronchitis virus (IBV)-specific antibody titers, and fearfulness were collected. There were no differences in BW at day 35 (brooders: 2857.3 ± 40.8 g, control: 2831.9 ± 39.6 g, $p = 0.55$) or feed conversion ratio (FCR) (brooders: 1.44, control: 1.45, $P = 0.87$). Nor were there any differences in mortality, gait score, or litter quality ($P > 0.05$). However, birds reared with brooders showed lower IBV titers compared to the control at 15 days after vaccination ($P = 0.0448$), although there were no other differences between treatments at 0, 22, or 29 days after vaccination. Chickens reared with dark brooders were less fearful as shown by shorter latency to first head movement in the tonic immobility test (brooders: 197.2 ± 15.9 s, control: 307.6 ± 17.9 s, $P = 0.0002$), shorter latency to turn itself around in tonic immobility test (brooders: 234 ± 19.7 s, control: 351 ± 29.5 s, $P = 0.0012$), more chickens near the object in the novel object test (brooders: 2.6 ± 0.25 birds, control: 1.5 ± 0.16 birds, $P < 0.0001$), and more chickens near the observer in the adapted touch test (brooders: 9.8 ± 0.28 birds, control: 4.8 ± 0.28 birds, $P < 0.0001$). The results of this study indicate that the use of dark brooders does not affect production or health of broiler chickens, however, birds with brooders were less fearful. Fear is a negative emotion and thus reducing fear should increase bird welfare. Therefore, dark brooders may be a useful tool to increase broiler welfare without negatively affecting production or health.

Introduction

Broiler chickens are generally raised in large flocks in a barren environment consisting of feeders, drinkers and with litter on the floor. This, together with the broilers' rapid growth, raises various welfare concerns, such as health and locomotory problems and behavioral restrictions (EFSA, 2023). To increase broiler welfare, environmental enrichment can be used to satisfy behavioral needs or improve specific welfare aspects. For example, environmental complexity could increase the level of activity of the broiler chickens which could result in better

leg health but also satisfy behavioral needs (reviewed in Riber et al., 2018). A type of environmental enrichment that has rarely been studied regarding behavior and welfare of broiler chickens is dark brooder use. Such dark brooders provide warmth and darkness and add structure to the birds' environment. These additions serve as valuable enrichment items because they simulate natural comfort or predator avoidance behaviours performed in the wild such as seeking shelter under the mother hen or hiding in vegetation (Mench, 2017). Dark brooders have previously been shown to reduce feather pecking (Gilani et al., 2012; Riber & Guzmán, 2017) and fearfulness in layers. However, one study by Stadig

* Corresponding author: Department of Animal and Veterinary Sciences, Blichers Allé 20, Aarhus University, Tjele, Denmark, 8830.

E-mail address: emily.leishman@anivet.au.dk (E.M. Leishman).

<https://doi.org/10.1016/j.psj.2025.105682>

Received 11 March 2025; Accepted 12 August 2025

Available online 13 August 2025

0032-5791/© 2025 The Authors. Published by Elsevier Inc. on behalf of Poultry Science Association Inc. This is an open access article under the CC BY-NC-ND license (<http://creativecommons.org/licenses/by-nc-nd/4.0/>).

et al. (2018) found no differences in fearfulness in slow-growing broilers raised with or without brooders. Another behavior affected by dark brooders is resting, where Forslind et al. (2022) found that the resting quality increased due to a reduction in disturbances in broilers reared with dark brooders, even after the removal of the brooders at 21 days old.

To further investigate the effects and feasibility of using dark brooders as environmental enrichment for broilers, the present experiment not only focused on some relevant welfare parameters but also on parameters that are essential for the profitability of broiler production like growth, feed conversion, and health. When dark brooders are used as a heating source, the ambient temperature of the barn is typically reduced to 20–24°C, while the temperature under the brooders follows the temperature program normally used for whole-house heating, i.e., starting at 33–34°C at chick placement, with a gradual reduction to 20°C around three to four weeks of age. If the broilers do not use the brooders as expected, this method of heat supply will risk the chickens experiencing cold stress, which in turn can affect broiler health, growth and immune responses (Lara & Rostagno, 2013; Olfati et al., 2018). Changes in the environment that induce behavioral alterations may also involve the immune system (reviewed in Hofmann et al., 2020). As dark brooders improve the quality of rest (Forslind et al., 2022), it is likely that they also affect sleep. In mammals, it has been shown that sleep is important for optimal immune function, reducing the susceptibility to infection as well as increasing antibody responses to vaccination (Schmitz et al., 2022). Young birds have a great need for rest (Malleau et al., 2007) and good quality of rest is important for welfare (e.g., immune system, Schmitz et al. 2022).

The aim of the present study was to investigate if access to dark brooders affected broiler production, welfare, and capacity to mount specific antibody production in response to vaccination. We hypothesized that broilers kept in an environment with brooders would have better welfare, in terms of improved clinical animal-based welfare measures, improved immune response, and reduced fearfulness, and show similar or better production performance.

Materials and methods

Animals and housing

The study was approved by the Animal Research Ethics Committee in Uppsala, Sweden (Dnr 5.8.18-17765 2018). The experiment was conducted at Lövsta Research Center, Swedish University of Agricultural Sciences, Uppsala, Sweden. In the building, one room was divided by a movable wall into two identical sections (6 m x 30 m), each section contained six pens (12 pens in total) of 2 m x 3.5 m (7 m²). The pens were separated by 60 cm high wired fences and the floor was covered with a 4 cm layer of wood shavings.

Mixed-sex Ross 308 broilers were picked up as day-old from a commercial hatchery (Swehatch AB, Väderstad, SWE) and driven by car to the research facilities. Mothers of the chicks were vaccinated according to the company standard protocol that among other immunizations included three vaccinations against infectious bronchitis virus (IBV) at 4, 8 and 16 weeks of age. Upon arrival, the chicks were randomly allocated into 12 pens. In one section, the ambient temperature was kept according to commercial practices with a starting temperature of 34 °C and gradually decreased to reach 20 °C at 21 days old and this temperature was maintained until the end of the growing period. The other section kept an ambient temperature of 20 °C throughout the whole growing period and in each of these pens three dark brooders (20 chicks/brooder, 40 cm x 60 cm, vidaXL, Netherlands) were provided with a starting temperature of 34 °C measured on the floor under the brooder. The temperature of the dark brooders was gradually decreased to reach 20 °C at 21 days old and the dark brooders were then removed from the pens. The stocking density was kept at an expected 20 kg/m² at slaughter, which resulted in 60 chicks per pen.

This density corresponds to the requirements for organic broiler production in Sweden (Statens jordbruksverksförfattningssamling, 2019). Water was provided ad libitum by nipple drinkers (10 broilers/nipple) and feed was provided in round feeders (2 cm feeder space per bird). Birds were fed a commercial grower diet, with an estimated energy content at 12.6 MJ per kg feed and a crude protein content of 198 g/kg, ad libitum (feed company Lantmännen, SWE). At one day of age, the light schedule was programmed for 24L:0D. The dark period was gradually increased to 6 h at 6 days old (18L:6D) and maintained until the end of the experiment (the light was on 04:30–22:30). The light intensity was 27 lux at bird level and 0–2 lux under the brooders. The study ended at 35 days old.

Treatments

Two treatments were included in this study, dark brooders (Fig. 1A) and control without brooders (Fig. 1B). A brooder is a horizontal panel, held up by four plastic legs with adjustable height, with an electrical heating source giving heat under the panel (Fig. 2). In the treatment with brooders, each pen had three dark brooders (40 cm x 60 cm) with the sides of the brooders covered with flaps of tarp to make the area under the brooders dark. The brooders were removed at 21 days old, when it was no longer possible for all the chicks to fit simultaneously under the brooders and the heat provided was no longer necessary.

Data collection

Ten chicks per pen were individually marked using color spray and then weighed at a weekly basis, to monitor growth of the birds. Weight gain was calculated as the final body weight (35 days) minus the initial weight. The amount of feed provided to each pen was automatically measured through the feeding system, and the remaining amount in each pen was weighed manually every week. Data on mortality and culls were collected during the experiment. Feed intake was calculated per pen and adjusted according to mortality on a weekly basis. Feed conversion ratio (FCR) was calculated at the end of the study, 35 days old,

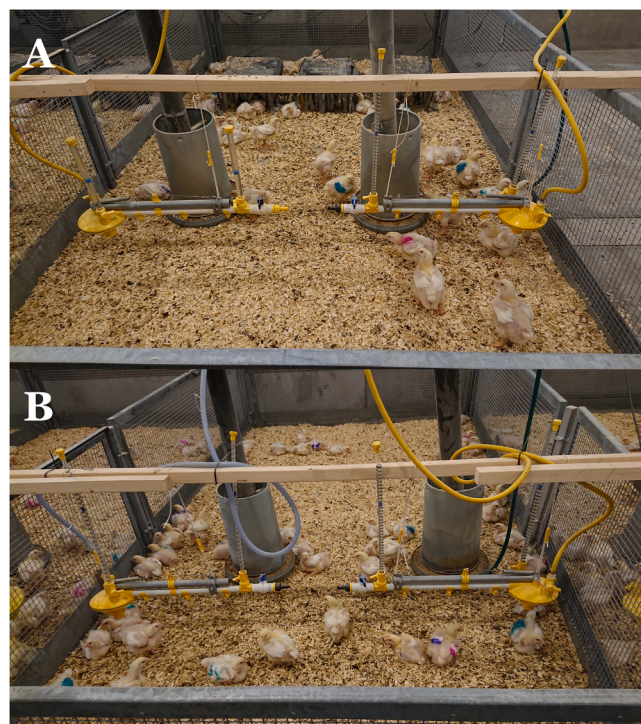


Fig. 1. Photo of experimental pet set-up for the brooder (A) and control (B) treatments.



Fig. 2. Photo of the dark brooders used in the brooder treatment.

by dividing feed consumption by weight gain of the chickens.

All chicks received a commercial live vaccine against IBV (Nobilis® IB Ma5 vet, MSD Animal Health) at 7 days old. The vaccine was delivered in the drinking water according to the manufacturer's instructions. Ten individually identified chickens per pen were monitored for antibodies to IBV in serum during the experiment. Blood samples were collected from the jugular vein at 7 days old, prior to vaccination, and subsequently at 22, 29 and 36 days old. Blood was collected into test tubes without additive, stored at room temperature overnight, centrifuged and sera were collected and stored at -20°C until analysis. The levels of antibodies to IBV in sera were quantified by a commercial ELISA-test (IDEXX IBV Ab Test) performed according to the manufacturer's instructions.

A tonic immobility test was done at 24 days old to test for fear responses (Bertin et al., 2008; Forkman et al., 2007) where a longer duration spent in tonic immobility indicates increased fearfulness (Jones, 1986). Five individuals per pen, that had not previously been handled or tested in any other individual-level test, were chosen randomly and were tested one at a time. A cradle was made of cardboard and placed on a table in a separate room in the building to avoid disturbances. The test was performed by the same observer for all birds. A bird was picked up and turned on its back when carefully placed in the cradle. The bird was gently touched on the belly with one hand and the head was covered with the other hand for 10 s. If the bird stayed on the back for at least five seconds, tonic immobility was considered induced whereas it otherwise was redone for a maximum of three times. The number of attempts needed to induce tonic immobility. The duration until the bird moved its head as well as the duration until the bird turned around were noted. If the bird had not yet turned around after 10 min a gentle nudge on the side made it turn around and the trial ended. A maximum duration of 10 min was then noted.

A novel object test was done at 10 and 32 days old to test for fear and explorative responses in a group setting (Forkman et al., 2007). A camera (GoPro Hero 7 White, GoPro, United States) was placed at one side of the pen giving a view of the whole pen. A novel object (colorful rope with knots or a small silicone cow) was placed in the pen, between the feeders. As the pens were not visually separated and therefore a risk that some chicks from the adjacent pen would see the object placed half of the pens within each treatment got the rope and half got the cow at the first test, changing to the other object at the second test to have the chicks encounter a novel object. The observer left the room and recorded the pen for 10 min. The number of individuals within three bird lengths from the object were noted every 60 s for 10 min.

Using the WelfareQuality protocol (WelfareQuality®, 2009), five different measures were taken near the end of the experiment. The touch test was adapted from the WelfareQuality protocol and was done at 35 days old to assess fear response. An observer walked slowly through the pen, approached a group of chickens and stopped at a bird length from where the closest bird was at the start of the approach. This was repeated in four trials at different positions in each pen. At every stop, the

observer squatted for 10 s and then counted the number of birds within an arm's length. In the WelfareQuality protocol, this test also includes counting the number of birds that could actually be touched. However, this additional recording was not done in this study.

Data on walking abilities were collected at 35 days old using the gait score assessment described in the WelfareQuality protocol where 0 = Normal, dextrous and agile, 1 = Slight abnormality, but difficult to define, 2 = Definite and identifiable abnormality, 3 = Obvious abnormality, affects ability to move, 4 = Severe abnormality, only takes a few steps, 5 = Incapable of walking (WelfareQuality®, 2009). Using an L-shaped wire mesh wall (LxH: 1.5 × 0.6 m + 0.5 × 0.6m) 20 birds from each pen were captured in a corner of the pen. Thereafter, one by one, the birds were allowed to walk back to the group in the pen and the gait score were given while doing so. The 20 birds gathered in the corner of each pen for assessing walking ability were also checked for hock burns and footpad dermatitis after the gait assessment. For both measures, in contrast to the WelfareQuality protocol, only presence/absence was noted.

Litter quality was assessed weekly in five areas in each pen: near feed and water, under the brooders, near walls and in open areas. In contrast to the Welfare Quality protocol, hands were used instead of the boots, but the same scoring system was used, from very wet (4) to dry (0). With one hand, the litter was squeezed between the fingers. If the litter ended up in a dense ball it was considered very wet and score 4, if the litter formed a ball that was not dense or falling apart it got score 3, if the litter formed a ball but fell apart it got score 2, if the litter did not form a ball but were dense to pick up it scored 1 and if the litter were easy to pick up and would just fall apart through the fingers it was considered dry and scored 0.

Statistical analysis

Statistical analyses were performed using SAS (SAS Institute Inc., Cary, NC, USA). The alpha level of significance used in this study was 0.05.

Bodyweight

A mixed model was used to compare the body weight of the birds in the two treatments which included the fixed effects treatment (control vs. brooder), age (1, 7, 21, 28, 35 days old), as well as the random effect of pen. The interaction between treatment and age was initially included in the model but was removed due to non-significance. A repeated statement was also included to account for the repeated measurements on the same birds at each age. A lognormal distribution was used due to the non-normal distribution of model residuals with a gaussian distribution. The replicate unit for this model was the individual bird. Results are presented as back transformed least-square means (LSmeans) ± SEM.

IBV Titers

A mixed model was used to compare the IBV titer results including the fixed effects of treatment (control vs. brooder), days post-vaccination (0, 15, 22, 29 d), and their interaction. A repeated statement to account for the measurement of the same individuals at each age was also included. Due to the non-normality of the linear model residuals, IBV titers were log transformed prior to analysis. The replicate unit for this model was the individual bird. Results are presented as LSmeans ± SEM.

Tonic immobility – turning around

To examine the effect of treatment on turning around during the tonic immobility test, a mixed model was used including the fixed effect of treatment (control vs. brooder) and the random effect of pen. Due to the non-normality of the linear model residuals, a lognormal distribution was used. The replicate unit for this model was the individual bird. Results are presented as back transformed LSmeans ± SEM.

Novel object test

To examine the effect of treatment on the outcome of the novel object test, a mixed model was constructed including the fixed effects of treatment (control vs. brooder), age (10 vs. 32 days old), and object (rope vs. cow) used during the test. The interaction between treatment and age was initially included in the model but was removed due to non-significance. The random effect of pen was also included. Due to the nature of the data (counts) and the mean being similar to the variance, a Poisson distribution was chosen for this model. The replicate unit for this model was the individual bird. Results are presented as back transformed LSmeans \pm SEM.

Adapted touch test

To examine the effect of treatment on the outcome of the touch test, a mixed model was used including the fixed effects of treatment (control vs. brooder) and trial (1-5) and the random effect of pen. A gaussian distribution was used due to the normal distribution of the residuals based on the Shapiro Wilk statistic. The replicate unit for this model was the individual bird. Results are presented as LSmeans \pm SEM.

Other

For the variables feed intake, FCR, mortality, culls, gait score, litter quality and tonic immobility head movements and number of inductions, a Kruskal-Wallis test was conducted as there was insufficient data or data variability for more complex models and a non-normal distribution of linear model residuals as determined by the Shapiro-Wilk statistic. In these tests, the fixed effect of treatment was tested, and results are presented as descriptive (raw arithmetic) means \pm standard deviation.

Results

Bodyweight and feed intake

There was no effect of treatment on body weight ($F_{1,569} = 0.36$, $P = 0.5489$). As expected, there was a significant effect of age on body weight (Fig. 3) with significant increases in bodyweight being observed at each measurement ($F_{4,569} = 55795.5$, $P < 0.0001$); 39 ± 0.4 g (day 1), 212 ± 2.1 g (day 7), 1261 ± 12.8 g (day 21), 2018 ± 20.6 g (day 28), 2828 ± 28.9 g (day 35). There was no difference in feed intake at 35 days old ($\chi^2(1) = 2.08$, $P = 0.1488$) or in FCR between the treatments

($\chi^2(1) = 0.0256$, $P = 0.8728$, Table 1).

Mortality and culls

There was no difference in combined mortality and culls between treatments ($\chi^2(1) = 0.7491$, $P = 0.3868$). Combined deaths were 27 birds in the brooder treatment (0.75%) and 29 (0.81%) birds in the control treatment where culls accounted for 12 birds in the brooder treatment and 10 birds in the control treatment. The majority of chickens died or were culled during the first week (10 of 56) and last week (23 of 56). During the last week, 14 chickens were found dead. Throughout the experiment, there were no birds found dead under the dark brooders.

Antibodies to IBV

There was a significant interaction between treatment and days post-vaccination ($F_{3,326.6} = 3.73$, $P = 0.0116$). Serum levels of IBV specific antibodies, measured prior to vaccination at 7 days old and at 15 (22 days old), 22 (29 days old), and 29 days (36 days old) after vaccination, are shown in Fig. 4. Results show that most chickens in both groups had maternally derived antibodies to IBV prior to vaccination (98% of birds in both groups had a titer ≥ 100 ; 81% and 84% of brooder and control birds, respectively, had a titer ≥ 300). At 15 days after vaccination (22 days old) serum titers to IBV were significantly decreased compared to those measured before vaccination for both groups ($t_{327.5} = 10.73$, $P_{\text{Control}} < 0.0001$; $t_{326.3} = 15.13$, $P_{\text{Brooder}} < 0.0001$). At 22 days after vaccination, titer levels were significantly increased from day 15 for both groups ($t_{326.3} = -4.50$, $P_{\text{Control}} = 0.0003$; $t_{326.3} = -6.87$, $P_{\text{Brooder}} < 0.0001$). There was no difference in IBV titers between 22 and 29 days post-vaccination for both groups ($t_{326.3} = -1.41$, $P_{\text{Control}} = 0.8541$; $t_{326.3} = -1.76$, $P_{\text{Brooder}} = 0.6461$). The only difference between treatments at

Table 1

Feed intake (g) and FCR of broiler chickens (Ross 308) provided with dark brooders or not at 35 days old (mean \pm SD).

Variable	Brooder	Control	P-value
Feed intake (g)	4053.5 \pm 12.1	4042.7 \pm 13.9	0.5489
FCR	1.44 \pm 0.04	1.45 \pm 0.03	0.8728

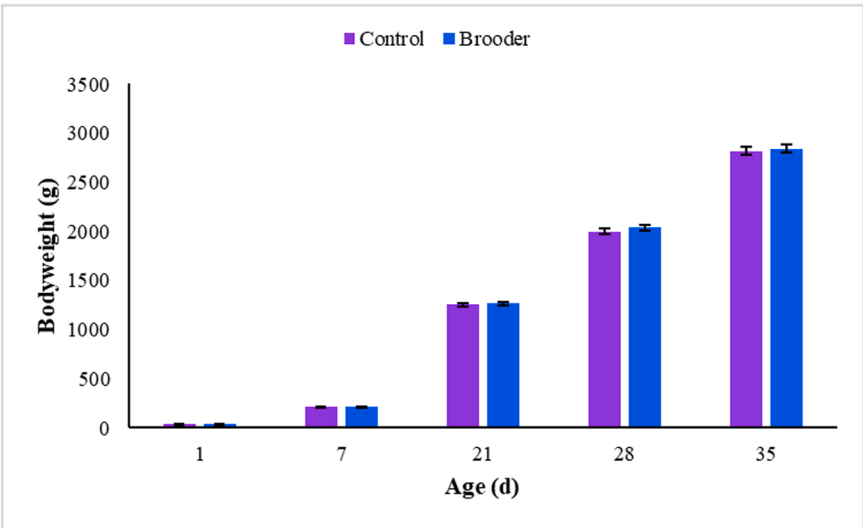


Fig. 3. Bodyweight of broiler chickens (Ross 308) with (blue) and without (purple) access to dark brooders over the experimental period (0-35 d). Data is presented as backtransformed LSmeans \pm SEM from the interaction between treatment and age for illustrative purposes, although the interaction was statistically nonsignificant ($P = 0.6899$). Across treatments, bodyweight increased significantly with age at each measurement interval ($P < 0.0001$). The effect of treatment was not significant ($P = 0.5489$).

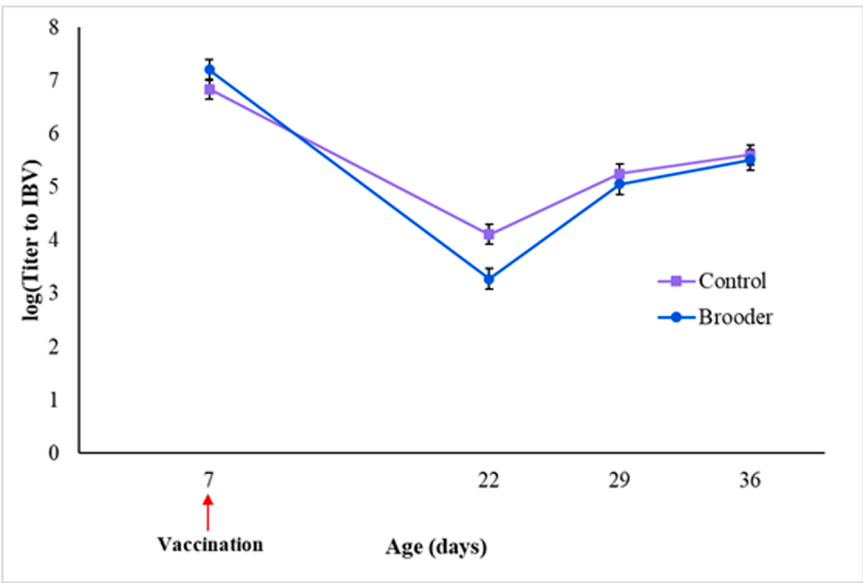


Fig. 4. Serum titers to infectious bronchitis virus (IBV) collected on the indicated days old from broiler chickens (Ross 308) with access to dark brooders (blue circles) or chickens without brooder access (purple squares). All chickens were vaccinated with a live IBV vaccine at 7 days old (arrow). Values are log-transformed LSmeans \pm SEM. There was a significant interaction between treatment and age ($P=0.0116$).

the same sampling point was at 15 days post-vaccination when titers to IBV were lower in the group with brooders compared to the control ($t_{419,5} = -3.09$, $P = 0.0448$).

Gait score

The proportion of birds in the different gait score categories is shown in Table 2. No birds displayed a gait score worse than 2 (definite and identifiable abnormality) in either treatment. The assessment of walking ability at 35 days old showed a tendency of a better gait score (lower = better) in the brooder treatment (brooders: 0.42 ± 0.05 , control: 0.59 ± 0.06 ; $\chi^2(1) = 3.8268$, $P = 0.0504$).

Litter quality, hock burns, and footpad dermatitis

No difference was found in litter quality between different treatments (brooders: 0.20 ± 0.07 , control: 0.23 ± 0.08 ; $\chi^2(1) = 0.0966$, $P = 0.7560$). Litter quality was good throughout the experiment. No hock burns were found in any of the treatment. Occurrences of footpad dermatitis were found in only three birds (two in the control treatment and one in the treatment with brooders).

Tonic immobility

There was a difference between the treatments for the latency to turn around in the tonic immobility test ($F_{1,40}=12.14$, $P = 0.0012$). Birds in the control group had a duration of 351 ± 29.5 s whereas birds in the

brooder group had a significantly shorter duration of 234 ± 19.7 s.

A longer duration of tonic immobility until first head movement was also shown in the control group compared to the group with brooders (brooders: 197.2 ± 15.9 s, control: 307.6 ± 17.9 s, $\chi^2(1) = 13.52$, $P = 0.0002$). The number of attempts needed to induce tonic immobility did not differ between the treatments (brooders: 1.1 ± 0.06 , control: 1.2 ± 0.08 , $\chi^2(1) = 1.4651$, $P = 0.2261$).

Novel object test

There was no effect of age ($F_{1,225} = 0.18$, $p = 0.6700$) or object ($F_{1,225} = 0.02$, $P = 0.8894$) on performance in the novel object test. However, there was a significant difference between treatments in the performance in the novel object test ($F_{1,225} = 40.49$, $P < 0.0001$). More chickens in the brooder treatment were within 3 bird lengths of the object (2.6 ± 0.25 birds) compared to the control group (1.5 ± 0.16 birds).

Adapted touch test

There was no effect of the trial number on the outcome of the adapted touch test ($F_{1,33} = 0.70$, $P = 0.5587$), but there was a difference between treatments ($F_{1,33} = 153.59$, $P < 0.0001$). More birds in the brooder treatment were within an arm's length of the observer (9.8 ± 0.28 birds) compared to the control group (4.8 ± 0.28 birds).

Discussion

This study shows positive effects of the use of dark brooders as an environmental enrichment to improve welfare. Chickens reared with brooders were less fearful than chickens reared without brooders in all three performed fear tests. This is in line with previous results showing that environmental enrichment provides animals the opportunity to regulate their environment and thus be less fearful (Jones & Waddington, 1992). Stadig et al. (2018) found no differences in fear between broilers reared with or without dark brooders but in that study slow growing broilers (SASSO) were used. Possibly, slow growing birds are generally less fearful (Abdourhamane & Petek, 2024), reducing the effect of brooders. Another possible explanation for the lack of effects on fearfulness may be that Stadig et al. (2018) did a tonic immobility test

Table 2
Distribution of gait scores using the WelfareQuality protocol (higher score = worse) for broiler chickens (Ross 308) with (brooder) and without (control) dark brooders where score 0 indicates normal, dextrous and agile, 1 indicates slight abnormality, but difficult to define, and 2 indicates definite and identifiable abnormality. No scores greater than 2 were observed in this experiment. There was a tendency for birds in the brooder treatment to have a better (lower) gait score at 35 d ($P = 0.0504$).

Score	Control	Brooder
0	60 (60%)	72 (50%)
1	49 (38%)	46 (41%)
2	11 (2%)	2 (9%)

but only ran it for a maximum of 300 s per bird compared to our 600 s. As our mean duration was more than 300 s for birds reared without brooders, this could explain the difference in outcome.

Since the chickens in both treatments gained weight at equal rates, and the FCR did not differ between treatments, the lower ambient temperature did not cause a higher feed consumption. This suggests that the low ambient temperature in the treatment with brooders was probably compensated by the heat from the brooders and did not affect the birds negatively. The flocks of both treatments grew faster than the performance objectives by [Aviagen \(2019\)](#) and had a slightly better FCR. Both the fast growth and the lower FCR could relate to the low stocking density used in our study as lower stocking densities have been seen to have a positive effect on the FCR in broilers ([Abudabos et al., 2013](#)). Also, at Lövsta research facility there are often long periods of empty holding between flocks, reducing the pressure from pathogens compared to commercial practice.

Antibody responses to vaccination did not differ between treatments, except at 15 days post-vaccination when chickens in the brooder treatment had lower IBV titers compared to the group without brooders. Most chickens had prominent titers of maternally derived antibodies to IBV persisting at one week of age. Therefore, the contribution of vaccination induced IBV antibodies at day 15 after vaccination, i.e. 22 days old, were difficult to evaluate. However, from 21 days after vaccination it was clear that chickens in both groups had mounted IBV-specific antibody responses. Considering the similar vaccine induced responses at the latter time points it seems likely that the lower IBV titers observed in chickens reared with brooders at 15 days after vaccination were due to a lower contribution of maternal antibodies at that time. It is possible that this difference was due to increased activity of the birds in the brooder treatment which increases metabolism and consequently the turnover of proteins such as maternally derived serum antibodies. The vaccine induced responses indicated that access to dark brooders did not affect the capacity to mount specific antibodies and thus does not entail specific health risks ([Hofmann et al., 2020](#)). Similar antibody responses to the vaccine in both the control and brooder groups may also imply no differences in stress levels in the chickens, or that this trait was not suitable to detect minor differences in stress level. Stress status needs to be kept low in the flocks to have an appropriate immune response, which is important for welfare, health and production ([Hofmann et al., 2020](#)).

A very low prevalence of walking difficulties was found. Walking difficulties mainly occur in the later stage of the rearing period. This study ended when the birds were 35 days old, which is the standard length of the production period in Sweden, and this might be a reason for the low prevalence of walking difficulties compared to other studies where birds were kept until a later age ([Wilhelmsson et al., 2019](#); [Yngvevsson et al., 2017](#)). Walking difficulties become more common with increasing age ([Henriksen et al., 2016](#); [Rasmussen et al., 2022](#)). However, walking difficulties are also related to body weight, and the body weight of the birds at the end of our study, at day 35, was comparable to that in other studies, where much higher incidences of walking difficulties were registered ([Kaukonen et al., 2017](#)). Heavier birds of the same age also show more leg problems ([Fernandes et al., 2012](#)). Possibly, the lower stocking density used in this study could have been a reason for the low incidence of walking difficulties ([Tahamtani et al., 2020](#)). The birds had more space to move undisturbed and a higher activity has been shown to reduce leg problems (reviewed in [Bessei, 2006](#); [Riber et al., 2018](#)).

No hock burns and only three instances of footpad dermatitis were seen in this study which is likely related to the good quality of the litter in this study. Contact dermatitis can be linked to the quality of the litter, where wet litter may induce contact dermatitis ([De Jong et al., 2014](#); [Meluzzi et al., 2008](#)). The prevalence of footpad dermatitis is considered an important welfare indicator ([Meluzzi & Sirri, 2009](#)), and the litter quality is considered a key to ensure welfare ([Meluzzi et al., 2008](#)). A common concern with the use of dark brooders is the idea that a high number of birds congregating under dark brooders for extended periods

of time could lead to poor litter quality (and consequently reduced footpad health). However, this was not the case in this experiment where the provision of dark brooders during rearing did not adversely affect litter quality or footpad health. The low prevalence of health problems does not match the high mortality during the experiment. The majority of the mortality consisted of birds found dead, especially during the last week. There was no apparent reason for the deaths which might suggest the occurrence of sudden death syndrome or ascites. However, no post-mortem examinations were done to determine the cause(s) of death, so it is not possible to say with certainty.

To increase the likelihood of being applied in practice, the costs of measures to improve bird welfare should be compensated for by an improvement of production parameters, reduction of other costs, or a higher market price. The scale of the current experiment does not allow conclusions on farm scale application of brooders but the data support earlier results ([Sirovnik & Riber, 2022](#)) indicating that giving access to dark brooders can improve animal welfare without negative effects on production. Also, in layers the dark brooders reduce injurious feather pecking, reduce mortality and increase the total egg production (reviewed in [Sirovnik & Riber 2022](#)). Based on the present results, the cost of the brooders (and the related improved welfare) is not compensated by better production. Possibly a higher market price can be realized based on improved welfare. Moreover, in some situations, it is possible that brooder use may save heating costs, but this is highly dependent on barn efficiency and alternative supplemental heating methods available (i.e., burning straw).

Conclusion

In agreement with our hypothesis, we found positive effects on the affective state of the broilers, as they were less fearful. The clinical welfare measures, mortality, antibody responses and FCR were not affected by using dark brooders. As the low ambient temperature in combination with heated brooders did not affect the FCR or health parameters, the use of brooders could decrease the cost for heating while not affecting production. The improved welfare may result in a marketing advantage. Using dark brooders in broiler rearing has positive effects while not showing any negative effects on the broilers. Practical application and economic evaluation must be carried out on a larger scale.

Declarations of interest

None

CRediT authorship contribution statement

Sara Forslind: Investigation, Writing – original draft, Writing – review & editing. **Emily M. Leishman:** Formal analysis, Visualization, Writing – original draft, Writing – review & editing. **Carlos E. Hernandez:** Conceptualization, Methodology, Supervision, Writing – review & editing. **Anja B. Riber:** Conceptualization, Methodology, Supervision, Writing – review & editing. **Helena Wall:** Conceptualization, Methodology, Supervision, Writing – review & editing. **Eva Watrang:** Conceptualization, Methodology, Supervision, Writing – review & editing. **Harry J. Blokhuis:** Conceptualization, Funding acquisition, Methodology, Supervision, Writing – review & editing.

Disclosures

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

Acknowledgements

The research described in this paper was funded by the Swedish Research council Formas and the Swedish Board of Agriculture. We thank Linnea Forslind for assistance during the data collection.

References

- Abdourhamane, I.M., Petek, M., 2024. Health-based welfare indicators and fear reaction of slower growing broiler compared to faster growing broiler housed in free range and conventional deep litter housing systems. *J. Appl. Anim. Welf. Sci.* 27 (3), 442–453.
- Abudabos, A.M., Samara, E.M., Hussein, E.O., Al-Ghadi, M.a.Q., Al-Atiyat, R.M., 2013. Impacts of stocking density on the performance and welfare of broiler chickens. *Ital. J. Anim. Sci.* 12 (1), e11.
- Aviagen, 2019. Ross 308 performance objectives. Aviagen.
- Bertin, A., Richard-Yris, M.-A., Houdelier, C., Lumineau, S., Möstl, E., Kuchar, A., Hirschenhauser, K., Kotschal, K., 2008. Habituation to humans affects yolk steroid levels and offspring phenotype in quail. *Horm. Behav.* 54 (3), 396–402.
- Bessei, W., 2006. Welfare of broilers: a review. *World's Poult. Sci. J.* 62 (3), 455–466.
- De Jong, I.C., Gunnink, H., Van Harn, J., 2014. Wet litter not only induces footpad dermatitis but also reduces overall welfare, technical performance, and carcass yield in broiler chickens. *J. Appl. Poult. Res.* 23 (1), 51–58.
- EFSA, 2023. Welf. broilers. farm 21.
- Fernandes, B.C.d.S., Martins, M.R.F.B., Mendes, A.A., Paz, I.C.d.L.A., Komiyama, C.M., Milbradt, E.L., Martins, B.B., 2012. Locomotion problems of broiler chickens and its relationship with the gait score. *Rev. Bras. Zootec.* 41, 1951–1955.
- Forkman, B., Boissy, A., Meunier-Salaün, M.-C., Canali, E., Jones, R.B., 2007. A critical review of fear tests used on cattle, pigs, sheep, poultry and horses. *Physiol. Behav.* 92 (3), 340–374.
- Forslind, S., Hernandez, C.E., Riber, A.B., Wall, H., Blokhuis, H.J., 2022. Resting behavior of broilers reared with or without artificial brooders. *Front. Vet. Sci.* 9, 908196.
- Gilani, A.-M., Knowles, T.G., Nicol, C.J., 2012. The effect of dark brooders on feather pecking on commercial farms. *Appl. Anim. Behav. Sci.* 142 (1-2), 42–50.
- Henriksen, S., Bilde, T., Riber, A., 2016. Effects of post-hatch brooding temperature on broiler behavior, welfare, and growth. *Poult. Sci.* 95 (10), 2235–2243.
- Hofmann, T., Schmucker, S.S., Bessei, W., Grashorn, M., Stefanski, V., 2020. Impact of housing environment on the immune system in chickens: A review. *Animals* 10 (7), 1138.
- Jones, R.B., 1986. The tonic immobility reaction of the domestic fowl: a review. *World's Poult. Sci. J.* 42 (1), 82–96.
- Jones, R.B., Waddington, D., 1992. Modification of fear in domestic chicks, *Gallus gallus domesticus*, via regular handling and early environmental enrichment. *Anim. Behav.* 43 (6), 1021–1033.
- Kaukonen, E., Norring, M., Valros, A., 2017. Perches and elevated platforms in commercial broiler farms: use and effect on walking ability, incidence of tibial dyschondroplasia and bone mineral content. *Animal* 11 (5), 864–871.
- Lara, L.J., Rostagno, M.H., 2013. Impact of heat stress on poultry production. *Animals* 3 (2), 356–369.
- Malleau, A.E., Duncan, I.J., Widowski, T.M., Atkinson, J.L., 2007. The importance of rest in young domestic fowl. *Appl. Anim. Behav. Sci.* 106 (1-3), 52–69.
- Meluzzi, A., Fabbri, C., Folegatti, E., Sirri, F., 2008. Survey of chicken rearing conditions in Italy: effects of litter quality and stocking density on productivity, foot dermatitis and carcase injuries. *Br. Poult. Sci.* 49 (3), 257–264.
- Meluzzi, A., Sirri, F., 2009. Welfare of broiler chickens. *Ital. J. Anim. Sci.* 8 (supl), 161–173.
- Mench, J., 2017. Behaviour of domesticated 11 birds: chickens, turkeys and ducks. *Ethol. Domest. Anim.: Intro. Text* 153.
- Olfati, A., Mojtahedin, A., Sadeghi, T., Akbari, M., Martínez-Pastor, F., 2018. Comparison of growth performance and immune responses of broiler chicks reared under heat stress, cold stress and thermoneutral conditions. *Span. J. Agric. Res.* 16 (2) e0505-e0505.
- Rasmussen, S.N., Erasmus, M., Riber, A.B., 2022. The relationships between age, fear responses, and walking ability of broiler chickens. *Appl. Anim. Behav. Sci.* 254, 105713.
- Riber, A.B., Guzmán, D.A., 2017. Effects of different types of dark brooders on injurious pecking damage and production-related traits at rear and lay in layers. *Poult. Sci.* 96 (10), 3529–3538.
- Riber, A.B., Van De Weerd, H., De Jong, I., Steinfeldt, S., 2018. Review of environmental enrichment for broiler chickens. *Poult. Sci.* 97 (2), 378–396.
- Schmitz, N.C., van der Werf, Y.D., Lammers-van der Holst, H.M., 2022. The importance of sleep and circadian rhythms for vaccination success and susceptibility to viral infections. *Clocks, sleep* 4 (1), 66–79.
- Sirovnik, J., Riber, A.B., 2022. Why-oh-why? Dark brooders reduce injurious pecking, though are still not widely used in commercial rearing of layer pullets. *Animals* 12 (10), 1276.
- Stadig, L., Rodenburg, T., Reubens, B., Ampe, B., Tuytens, F., 2018. Effects of dark brooders and overhangs on free-range use and behaviour of slow-growing broilers. *Animal* 12 (8), 1621–1630.
- Statens jordbruksverks föreskrifter och allmänna råd om fjäderfåhållning inom lantbruket m.m., (2019).
- Tahamtani, F.M., Pedersen, I.J., Riber, A.B., 2020. Effects of environmental complexity on welfare indicators of fast-growing broiler chickens. *Poult. Sci.* 99 (1), 21–29.
- WelfareQuality®, 2009. WelfareQuality® Assessment Protocol for Poultry (Broilers, Laying Hens). The WelfareQuality® Consortium.
- Wilhelmsson, S., Yngvesson, J., Jönsson, L., Gunnarsson, S., Wallenbeck, A., 2019. Welfare Quality® assessment of a fast-growing and a slower-growing broiler hybrid, reared until 10 weeks and fed a low-protein, high-protein or mussel-meal diet. *Livest. sci.* 219, 71–79.
- Yngvesson, J., Wedin, M., Gunnarsson, S., Jönsson, L., Blokhuis, H., Wallenbeck, A., 2017. Let me sleep! welfare of broilers (*Gallus gallus domesticus*) with disrupted resting behaviour. *Acta. Agric. Scand. A—Anim. Sci.* 67 (3-4), 123–133.