Follow the cues

Using visual and auditory stimuli to promote utilization of resources and behavioural synchrony in pullets

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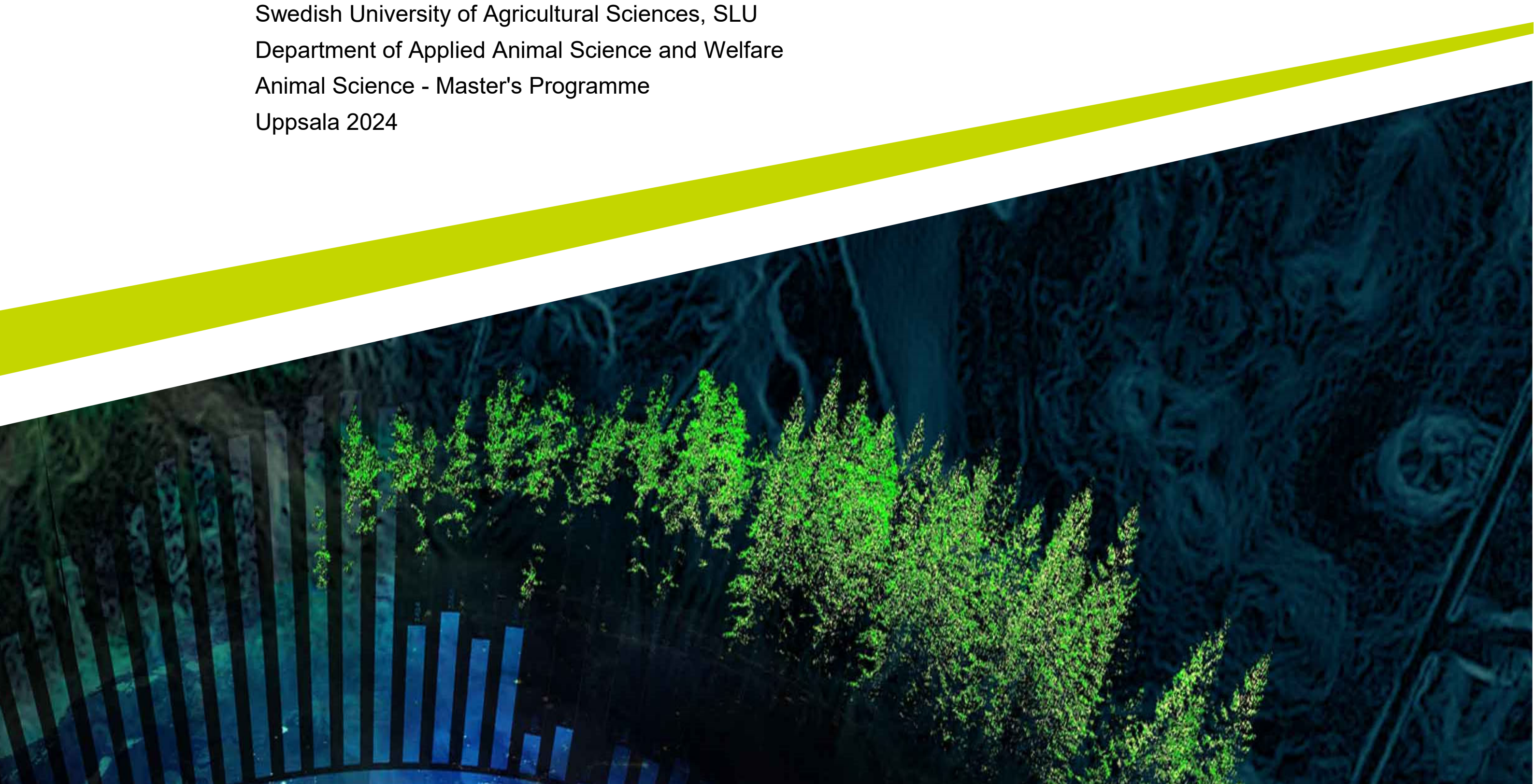

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Abstract

Rearing pullets without a broody hen poses some welfare challenges because the absence of a mother hen reduces the guidance provided to chicks, making it harder for them to find and use proper resources effectively, along with synchronizing their behaviour. This deficiency in guidance may lead to feather pecking in later stages, a significant concern within the layer industry.

This experiment aims to assess the efficacy of various methods providing visual and auditory cues of a broody hen, with the goal of identifying the most effective means of attracting chicks to resources and encouraging them to use the resources adequately.

A total of 240 1-day-old layer-type female chicks (Bovan white) were utilized in two replicates and randomly assigned to one of four rearing treatments: 1) the Audio group, involving auditory playback of broody hen feed and resting calls in the feed and rest zones; 2) the Video + Audio group, incorporating both audio and video playback of a broody hen in the feed and rest area; 3) the Video group, where only video playback is presented in the feed and resting areas; and 4) the Control group, featuring no audio or video playback. Feeding and resting cues were presented at different periods during the day. Chicks' behaviour was recorded for 5 h in the morning and 5 h in the evening. Video analysis was done on days 5, 10, and 15. Data were analysed using least square means and Tukey's HSD test for post hoc comparisons.

The results indicate that the number of birds observed in the high-response area in relation to feeding cues in the Video + Audio and Video groups was approximately twice that of chicks observed in the same area in the Audio and Control groups during the Feed period ($P \leq 0.05$). Around 71 percent of chicks in the Video + Audio group were attracted to the High Response area in relation to resting cues, with no significant difference between this group and the Video group ($P \leq 0.05$). While, this percentage for the Audio group was around 56%, and for the Control group, it was about 52%.Significantly more chicks exhibited feed pecking behaviour when feeding cues were presented in both Video + Audio and Video groups compare to the time with no cues or during resting cues at 5 and 10 days of age ($P \leq 0.05$).

Additionally, chicks showed noticeably increased laydown behavior in the nothing period (excluding the Video group), as well as during the resting period and in treatments without any screens ($P \leq 0.05$). The Video + Audio group and Video group (during the feed period and nothing period) displayed the lowest laydown behavior ($P \leq 0.05$). In conclusion, the Video+ Audio and Video groups proved to be the most effective means of attracting and encouraging chicks to engage in feed pecking in the feed area during the feed period. Chicks exhibited more laydown behavior during the nothing and resting periods, except for the Video group, and in treatments without screens ($P \leq 0.05$). The results further reveal that the Video + Audio and Video groups successfully synchronized the behaviour of chicks, with the majority exhibiting similar behaviours when specific cues were played. There were no differences in body weight between treatments.

Keywords: feather pecking, feed call, rest call, behavioural synchronization, visual cues, auditory cues.

Table of contents

List of tables

List of figures

Abbreviations

H	Hour
min	Minute
Sec	Second

1. Introduction

1.1. General overview

In a natural setting, during the first few weeks of a chick's life, the broody hen shares important lessons on finding food and interacting with others, ensuring her chicks stay warm and safe (Hewlett & Nordquist, 2019). Within the first 24 hours of life, chicks start pecking and gain an understanding of suitable food and pecking substrates. Additionally, chicks learn to respond to the maternal feeding call and purring sound that the hen makes when she settles down. Regular exposure to their mother's presence, along with the provision of food, guidance, and protection, reinforces the connection between the hen and her offspring.

In commercial setups, chicks undergo hatching and rearing processes in the absence of a mother. Chicks are exposed to a continuous phase of light and darkness in a commercial environment. They also receive static or whole-house heating, preventing the separation of active and inactive chicks. In such conditions, behaviour synchronization would not happen, and chicks might disrupt and target feather pecks toward other chicks that are resting (Gilani et al. 2012). Moreover, in a commercial setup where chicks are raised in large groups without maternal guidance, the absence of a mother hen can impact the formation of pecking behaviours. Chicks might direct their pecks towards undesired areas, such as the feathers of their conspecifics, as observed in studies by Rodenburg et al. (2008) and Riber et al. (2007).

1. 2 Statement of Problem

The welfare of laying hens is significantly influenced by the rearing method employed for pullets from hatch until they reach 15 to 18 weeks of age, marking their transition from the rearing system to the laying system. While some issues may heighten over time, especially impacting welfare during the laying phase, certain challenges are unique to the rearing period. Abundant evidence underscores the concept that early experiences play a pivotal role in shaping behaviour over the long term, leading to the manifestation of undesirable abnormal patterns (Janczak and Riber, 2015). Rodenburg et al. (2013) and Bilcik and Keeling (1999) suggest in their studies that feather pecking may result from redirected ground pecking, originating from either foraging or dust-bathing behaviours. Pecks aimed at exploring food and materials for dust bathing or foraging might inadvertently be directed towards inactive conspecifics. There's a risk of down feathers being mistaken for foraging materials, potentially leading to the development of feather pecking behaviour (Vestergaard & Baranyiová, 1996; Blokhuis, 1986). Feather pecking stands out as one of the most significant welfare and economic challenges in the layer industry (Sirovnik & Riber, 2022). Chicks are hatched and raised without the presence or care of a mother hen, leading to various welfare challenges, such as heightened occurrences of feather pecking and cannibalism (Rodenburg et al. 2004; Riber et al. 2007b), reduced feeding (Riber et al. 2007a), and the development of unsynchronized behavioural patterns (Jensen et al. 2006). Consequently, there is a growing focus on researching how to simulate various aspects of maternal care in commercial poultry rearing, becoming a significant area of interest in poultry welfare studies. One approach that has already been employed in commercial farms, as suggested by Gilani, Knowles, and Nicol (2012a), is to simulate the warmth and darkness experienced under a mother hen's feathers is the use of 'dark brooders . The utilization of dark brooders, simulating the experience of being under a mother's wings, has been proven to decrease feather pecking behaviour in adult layer hens (Riber & Guzman, 2017). Riber et al. (2007) proposes that the separation of active and inactive chicks within a defined area, while utilizing dark brooders during rearing, diminishes the likelihood of misdirected pecks. Dark brooders can also synchronize chicks' behaviour (Gilani et al. 2012a).

Economically and practically, rearing chicks with broody hens in commercial settings poses challenges. Broody hens may not provide the efficiency needed in space, feed, and overall management compared to controlled methods. This affects uniform growth rates crucial for meeting market standards. Moreover, broody hens increase disease risks and labour intensity compared to conventional management practices. Interestingly, chickens exhibit the ability to recognize both the video image and sound of a bird feeding, as indicated by findings from a study conducted by Keeling and Hurnik (1993). Therefore, it may be possible to use other aspects of maternal care such as feeding cues and resting cues playback of a broody hen to attract chicks to feed area and rest area and make chicks respond to these cues by performing the same behaviour as a broody hen and utilize the resource

1. 3 Aims and research hypothesis

The aim of this study is to determine the most effective method for attracting chicks to the feeding and resting areas, encouraging them to eat food in the feeding area, lay down in the resting area, and perform behaviours together, thereby increasing behavioural synchrony.

The first hypothesis of this study was that chicks will approach the resources when presented with a visual and/or auditory cue and will do so more than chicks that were not presented with any cues. The second hypothesis was, chicks will use resources appropriately (i.e., eat in the feeding area and rest in the resting area) when presented with a visual and/or auditory cue compared to birds not having any cues. The third hypothesis was that chicks will show more synchronized behaviour (eat, rest, etc. together) when presented with a visual and/or auditory cue compared to control birds with no cues. The fourth hypothesis was that there would be no differences in body weight among treatments.

2. Literature review:

2. 1 Maternal care in natural settings

In natural habitats, the survival of chicks depends on quickly forming a strong connection with their mother. A close bond forms when a broody hen is with her chicks.

A maternal connection between a broody hen and her chicks lasts between 5 to 12 weeks (McBride et al. 1969). During this crucial period, the maternal contact of a broody hen has a significant positive impact on the development of the chicks' behaviour, particularly on how they use resources (Edgar et al. 2016).

2. 2 Filial imprinting

During the first few days of chicks' lives, both domestic and wild Red Jungle fowl (Gallus gallus) chicks spend the first few days of their life living near or even under their mothers after hatching, particularly throughout the first four days (Edgar et al. 2016). This period is crucial for filial imprinting, a process where newly hatched chicks learn to recognize the appearance and sound of their mother and instinctively follow her. Filial imprinting is essential for fostering a strong maternal bond in chicks (Nakamori et al. 2013).

This imprinting process offers significant advantages, ensuring the establishment of appropriate feeding behaviour and behavioural synchrony (Edgar et al., 2016). It plays a vital role in the survival and social development of young chicks, as emphasized by Rosa-Salva et al. (2021).

Once filial imprinting is successfully completed, the chickens will follow the broody hen (Nakamori et al. 2013).

2.3 Broody hen vocalization:

Broody hens produce a variety of vocalizations aimed at their chicks right from the moment the chick hatch. These vocalizations play a significant role in shaping the behaviour of the chicks (Toukhsati et al. 2005; Kent, 1987). These vocalizations can be classified as attraction calls and alarm calls (Edgar et al. 2016; Field et al. 2007).

Maternal attraction vocalizations, including roosting calls, maternal cluck sounds, and feeding calls, serve to draw the chicks near and maintain the family unit (Edgar et al. 2016; Evans, 1975). These distinct vocalizations are used by the broody hen to attract her chicks and foster the connection within the family.

2. 3. 1 Feeding calls:

Domestic hens choose a wide diet of seeds, fruits, plants, and invertebrates when living in a variety of environments (Nicol, 2004b). Chicks must learn which objects are worth ingesting since they do not have the intrinsic capacity to distinguish between such a wide varieties of food kinds.

When a broody hen discovers food, she produces a distinctive high-pitched fast vocalization, which, together with pecking behaviour, draws the chicks and motivates them to eat (Stokes, 1971; Sherry, 1977). The mother's feeding behaviour not only prompts the chicks to peck but also draws them to the area where the broody hen is pecking, which helps the chicks develop adaptable foraging techniques and taste perception (Nicol, 2004; HOPPITT et al. 2008). Interestingly, hens produce louder and longer sounds when high-quality food is present (Moffatt & Hogan, 1992).

In their initial stages, young chicks randomly peck at food and non-food things with their beaks, displaying limited sensitivity to ingestion outcomes and learning minimally through conventional trial and error (Edgar et al. 2016; Nicol, 2004).

Observational studies demonstrating that chicks peck in the same areas as the broody hen further indicate the importance of the maternal feeding display in chick eating behaviour (Joos & Collias, 1953; Moffatt & Hogan, 1992).

Early exposure to a mother hen may avoid the initiation of feather pecking by introducing the chicks to direct their pecks in the direction of more suitable stimuli, like the ground or litter (Rodenburg et al., 2008; Riber et al., 2007), leading to a negative relationship between ground pecking and feather pecking (Rodenburg et al., 2008; Riber et al., 2007; Blokhuis and Arkes, 1984; Jones et al. 1995).

Studies comparing the feeding behaviour of chicks that are raised with broody hen and non-brooded chicks provide experimental support for a maternal influence. After a hen made a feeding display, the chicks showed increased eating behaviour, primarily directed at the same food source as the broody hen, and continued to consume food even after the hen's presentation was over (Riber et al., 2007) (Wauters & Richard-Yris, 2002).

Feeding calls and pecking actions provide the chicks with a mixture of both auditory and visual stimulation that raises the chicks' arousal level (Wauters, et al. 2002).

The auditory characteristics of feed calls are particularly beneficial to the chicks in helping them determine the kind and quality of food offered (Moffatt and Hogan, 1992; Wauters & Richard-Yris, 2003; Wauters et al. 2002).

Workman & Andrews propose that hens give the majority of their information regarding food palatability before the chicks are eight days old. As they get older during this time, chicks respond to food calls more quickly, further indicating that learning may be a factor in how they respond (Edgar et al. 2016; Wauters and Richard-Yris, 2002).

Furthermore, brooded chicks engage in longer-lasting eating behaviours (Wauters et al. 2002).

A small-scale study found that playing audio of maternal feeding calls close to the feeder boosted feed conversion and chick weight (Woodcock et al. 2004).

Surprisingly, no research has been done on the consequences of playing these calls on the welfare and behaviour of chicks.

2. 3. 1. 1 Visual characteristics of a broody hen during feed calls:

When engaging in feed calls, the broody hen develops a recognizable posture and makes a quick staccato call when it comes across an acceptable food source (Sherry, 1977). During this display, the broody hen turns forward while calling, pecking at the food, picking it up and dropping it again, pecking the ground, and pecking strongly on the food. Her back is sharply inclined, her breast is near the ground, and her tail is lifted and open (Sherry, 1977).

2. 3.2 Roosting calls:

A broody hen often attracts her remaining chicks to rest beneath her by producing long, low purring sounds when she has settled in a specific location (Collias and Joos, 1953).

These purring noises lack any discernible rhythm and serve as distinctive roosting calls, guiding the chicks to safety and warmth (Collias, 1987).

2. 4 Behavioural synchronizations:

Natural brooding fosters behavioural and diurnal synchronization among the brood, where all brood members exhibit active and inactive behaviours at the same time (Riber et al. 2007). This synchronized behaviour serves as a thermoregulation strategy in precocial birds, where mother hens act as mobile warmers (Edgar et al. 2016; Lumineau et al. 2000).

Mother hens, acting as mobile warmers, position themselves near food and water sources, motivating chicks to eat and further enhancing the synchronization within the brood (Campo et al. 2014).

Ultradian rhythms in precocial birds involve alternating active and inactive stages (Edgar et al. 2016; Serge Daan and Aschoff, 1981).

Synchronization of these ultradian rhythms facilitates group cohesion, enabling members to stay together and simplifying the grouping of individuals based on their various motivations (Conradt and Roper, 2000; RUCKSTUHL, 1998).

Riber et al. (2007) found no overall difference in the total time spent active between brooded and non-brooded groups of chicks. However, brooded chicks exhibited significantly longer activity bouts and enhanced synchronization.

Wauters et al. (2002) conducted a study comparing the activity levels of brooded and non-brooded chicks and discovered that both groups engaged in similar behaviours for about the same amount of time. Nevertheless, activity bouts in the brooded group were substantially longer. The behaviour of brooded chicks was more synchronized, and they also made better use of the space that was available (Edgar et al. 2016; Wauters et al. 2002).

These synchronized behaviours are critical as they serve as a mechanism to keep conspecifics that are resting separate from those actively pecking, both spatially and temporally. This maternal intervention likely plays a protective role against feather pecking. Perhaps not till the chicks are grown up will this maternal protection towards feather pecking appear (Edgar et al. 2016).

In studies comparing brooded and non-brooded chicks, Roden & Wechsler observed similar levels of feather pecking in one-week-old chicks. However, Riber et al. demonstrated higher mortality rates and increased feather pecking in chicks raised without a broody hen at 20 and 24 weeks old, underscoring the long-term consequences of maternal absence (Roden & Wechsler, 1998; Riber et al. 2007).

It's interesting to note that without a mother hen present, the early ultradian rhythms seen by non-brooded chicks disappear, highlighting the significant influence of maternal care on the biological rhythms of chicks (Edgar et al., 2016).

2. 5 Brooded chicks vs. non-brooded chicks:

Chicks that have been brooded exhibit distinctive behavioural patterns that emphasize the profound impact of maternal care in comparison to their non-brooded counterparts.

Activity level:

Compared to non-brooded chicks, brooded chicks are more active and engage in more ground pecking and dust bathing (Edgar et al. 2016; Riber et al. 2007; Shimmura et al. 2010).

Fear and Aggression:

Brooded chicks display less fear (Shimmura et al. 2010; Perré, Wauters and Richard-Yris, 2002), behave less aggressively (Fält, 1978), and are more motivated to engage in social interactions (Perré et al. 2002).

2. 6 Artificial rearing of chicks:

In spite of the advantages of mother care, it is not economically feasible to permit brooding on farms.

Given the crucial role played by the mother hen, artificially raising chicks may potentially have negative and long-lasting effects on the well-being of the chicks (Edgar et al. 2016).

In the commercial environment, chicks receive continuous light and artificial radiant heat via static brooders or whole-house heating. So active and inactive chicks aren't divided. Behaviours become unsynchronized in this circumstance, and chicks may disrupt and peck conspecifics who are resting (Gilani et al. 2012).

In Addition, in a commercial setting where chicks are raised in big groups without a mother, the absence of an experienced mother's guidance may have an impact on the formation of pecking choices, with chicks directing their pecks to undesired places like the feathers of conspecifics (Rodenburg et al. 2008; Riber et al. 2007).

2.6.1 Feather pecking:

Feather pecking is a significant concern in the poultry industry, impacting both economics and animal welfare (Edgar et al. 2016; Lambton et al. 2010; Gilani, Knowles, and Nicol, 2013).

During their first week of life, chicks naturally engage in foraging and gentle pecking behaviours as they explore their surroundings (HUBER-EICHER and WECHSLER, 1998).

Rearing settings during the first four weeks have a significant impact on how feather pecking develops in laying hens (Johnsen et al. 1998).

In an environment where foraging is hindered, abnormal feather pecking is likely to develop as redirected foraging behaviour (Blokhuis, 1986; Huber-Eicher and Wechsler, 1997, 1998).

Feather pecking presents in two distinct forms:

1. **Gentle Feather Pecking (GFP):** This behaviour involves pecking the tips of feathers, causing minimal harm and is mostly disregarded by the recipient. However, stereotyped gentle feather pecking might indicate underlying welfare issues (Rodenburg et al. 2013).

2. **Severe Feather Pecking (SFP):** SFP is aggressive feather pecking, leading to significant plumage damage and potential harm to the recipient (Rodenburg et al. 2013).

Attractive targets for pecking are feathers that have already been damaged. Studies have shown that compared to undamaged feathers, these injured feathers draw a lot more pecking attacks. Additionally, they contribute to the flock's development of feather-picking behaviour (McAdie & Keeling, 2000). Moreover, excessive feather pulling and plucking results in bald areas (McAdie &Keeling, 2002). Pecking at bare skin regions can result in bleeding, reinforce the undesirable habit, and quickly propagate this abnormal conduct within the flock through imitation (Hartcher et al. 2015; Bilcik and Keeling, 1999; WECHSLER, HUBER-EICHER and NASH, 1998). Tissue damage and significant blood loss through tissue pecking might eventually result in death (Rodenburg et al. 2013).

2.7 Behavioural Synchronizations in a Commercial Setting: Dark Brooders as Maternal Simulations

In commercial poultry farming, dark brooders have emerged as innovative solutions to behavioural synchronizations, replicating the warmth and darkness experienced under broody hens.
Dark brooders consist of heat sources beneath roofs made of dark plastic surrounds, effectively blocking out light.
This device is the initial and sole commercial use of a maternal simulation and serves as an illustration of the way natural maternal behaviour can result in the production of feasible on-farm options for welfare issues.
Crucially, studies indicate that dark brooders have no negative impact on production (Gilani et al. 2013).
Moreover, it has been shown that the usage of dark brooders, which simulate being under a mother's wings, helps to decrease adult layers feather pecking tendency (Riber & Guzman, 2017).

2.8 Domestic chicks' visual system:

Similar to humans, birds have highly developed colour vision and strong visual acuity, providing them with high-quality visual images that large portion of their behaviours are based on it (Zeigler & Bischof, 1993).

Chickens, as predatory animals, heavily rely on their keen vision for effective navigation inside commercial housing systems (Campbell et al. 2018).

Research by Nicol and Prescott et al. (2015) suggests that chickens exhibit visual abilities surpassing those of humans.

They see moving items more readily than stationary ones (Broom, 1969).

In both real-life (Broom, 1969), and in controlled displays (Vallortigara et al. in 2005 and Vallortigara and Regolin in 2006) young chickens have a natural inclination towards objects or stimuli displaying biological motion as observed in studies conducted by Regolin et al. (2000).

Chickens have the capability to recognize images from videos, including food items, other chickens, predators, and distinctions between moving and stationary objects (Campbell et al. 2018).

On the first day after hatching, chicks display pecking behaviour towards insect-like features shown on a video screen. They exhibit a tendency to peck more at sideways-moving insects compared to forward-moving ones (Clara et al. 2009).

Chicks change their feeding habits when watching videos of other members of their group eating from a specific food dish or when exposed to a stimulus resembling a predator (Keeling & Hurnik, 1993) (Dharmaretnam & Rogers, 2005).

In an experiment that was done by Keeling and Hurnik, (1993), the response of chickens to live bird during feeding and video display of same bird during feeding were compared. Their findings suggest that chickens have the ability to recognize the video image and sound of a bird feeding and respond to these stimuli similarly to how they respond to the sight and sound of a live bird eating.

Another experiment by Evans, and Marler in 1993 revealed that male chicks exhibited specific alarm responses depending on the type of predator presented in video images. When exposed to visual cues of an aerial predator, such as a hawk shown in a video image, the chicks displayed defensive behaviours: including lowering their bodies, smoothing their feathers, turning their heads upward with one eye, and emitting aerial alarm calls. Conversely, in the presence of a terrestrial predator like a raccoon shown on a screen, the chicks responded differently. They stood upright, focused on the screen, swiftly moved to the opposite cage end, and produced ground alarm calls while pivoting back and forth.

2.9 Domestic chicks' auditory system

The auditory system of chicks develops mostly during embryonic period and reaches maturity before other sensory systems do. Chicks start responding to sound around embryonic days 11 and 12 (Campbell et al. 2018). They show auditory imprinting from day 16 of incubation (Sirovnik et al. 2021).

In a study by Davila et al. (2011), the heterophil to lymphocyte (H/L) ratio of chicks was altered by playing classical music to them for up to 8 weeks, 5 hours per day, three days per week. This change, which was shown in comparison to control chicks exposed to no music, shows lower stress levels.

In an experiment by Zhao et al. (2021), the impact of short-term classical music stimulation on the behaviour of 10-week-old pullets was investigated. In comparison to the control group, birds exposed to classical music showed more comforting and preening activities but less aggressive behaviour and feather-pecking. The results indicate that auditory enrichment, like classical music, can improve the wellbeing of pullets by positively impacting their behaviour.

Chiandetti and Vallortigara (2011) found that, like humans and other species, chicks exhibit a preference for harmonic consonant sound intervals over dissonant ones. They suggested that this desire for harmonious consonant in chicks might be because those sounds are commonly heard in nature.

Studies involving humans have provided evidence that exposure to auditory stimuli like maternal sounds and music can enhance learning and memory in later stages of life (Chaudhury et al. 2013).

When domestic chicks hear rhythmic mother hen calls, noradrenaline is released in the brain, which improves memory (Field et al. 2007).

According to research done by Edgar et al. (2015), playing particular maternal cluck sounds to 15- to 16-day-old chicks lowered their stress response.

It has been observed that for the first nine days after hatch of broiler chick's life, playing broody hen vocalizations increased feed conversion and weight gain (Woodcock et al. 2004).

Chicks respond to food call playbacks with enhanced anticipatory eating activity, displaying higher exploration of the ground surface, in contrast to contact or alarm call recordings (Bessa Ferreira et al. 2022).

2.10 Using broody hen characteristics:

Because of the negative impacts of maternal care on certain production parameters, research should concentrate on identifying the key aspects of maternal care that can be artificially reproduced to enhance well-being and can be practically used in commercial settings (Edgar et al. 2016).

Few research is done to investigate other aspects of maternal care that can be reproduced to increase welfare of chicks later in life. In this study some aspects of maternal care, such as vocalizations and visual cues of a broody hen during feeding and resting are used to attract chicks to resources and increase the utilization of resources by chicks.

3.Materials and methods

3.1 Animal and housing

A total of 240 1-day-old layer-type female chicks (Bovan white) were used in this experiment. The chicks were housed in individual floor system pens (W 120 x L 240 x H 190 cm) at a research facility of the Swedish University of Agricultural Science. Pellets of crushed straw were used as litter from day 1 to day 21. The chicks were given free access to commercial pelleted feed on metal food trays (appropriate for their age), as well as water throughout the experiment. The location of the food plates was the same across all pens by placing them 20 cm in front of the screens or at a location that was equal in groups without screens. Throughout the experiment, the room's temperature was kept at 25 degrees. A 150-watt Elstein IOT/90 ceramic heat lamp (Elstein IOT/90, Germany) were used for each pen. The heat lamp was installed 10cm away from the wall precisely above the screen and set at 45cm height (bedding height was 4cm). The first day chicks had one hour of darkness (00:00-01:00h). On day 2, 3 and 4 they had 2 h (00:00-02:00h), 4h (23:00-03:00h) and 6 h (22:00-04:00h) of darkness respectively. From day 5 to the end of the experiment they had 8 hours of darkness (21:00-05:00h).

3.2 Visual and auditory cues

Videos and sounds of a broody hen with her chickens performing A) food calls and approaching food and B) resting calls with brooding behaviour (i.e., chickens hiding under hen) were presented near the food and resting areas, respectively. Video and audio cues were presented daily from 9:30 to 21:30.

HP Compaq LA2405x and LA2205wg computer monitors were used to display the visual cues, and compact stereo speakers were used for playback of the auditory cues (Logitech Z120).

The visual and auditory cues were presented in two separate locations inside the pen (Figure 2):

a) Feed area where the food plate was located.
b) Rest area underneath the brooder

To allow the birds to view the video images displayed on the screen at eye level, the monitors were placed outside the pens, standing directly on the ground, and leaning on a wooden panel with a cut-out for the screen. The 24 to 25-inch screens were large enough to display a hen at actual scale.

The speakers were placed outside the pens, leaning against a wooden panel with a speaker cut-out, and they were situated equally for all groups with regard to the location of heater lamp or feeders. Given that the bedding speakers were nearly at chicken height, the speakers were placed 16 cm from the floor, with the left and right speakers 64 cm apart (at either side of the screens).

3. 3 Treatments

This study was conducted in two replicates due to facility limitations, each with three pens per treatment per replicate, making a total of six pens per treatment. The birds were studied from 1 to 21 days of age. Twelve pens per prelicate were placed in the same room in the facility (Figure 1). Chicks in each pen visually were separated from others but not auditorially.
Chicks were randomly allocated to 1 of 4 rearing treatments that were used in this experiment.

1) Audio group, where audios playbacks are presented in the rest and feeding areas.
2) video + Audio group, where audios and videos playbacks are presented in the rest and feeding area
3) Video group, where videos playbacks are presented in the rest and feeding areas.
4) Control group where no audio or video playback is presented.

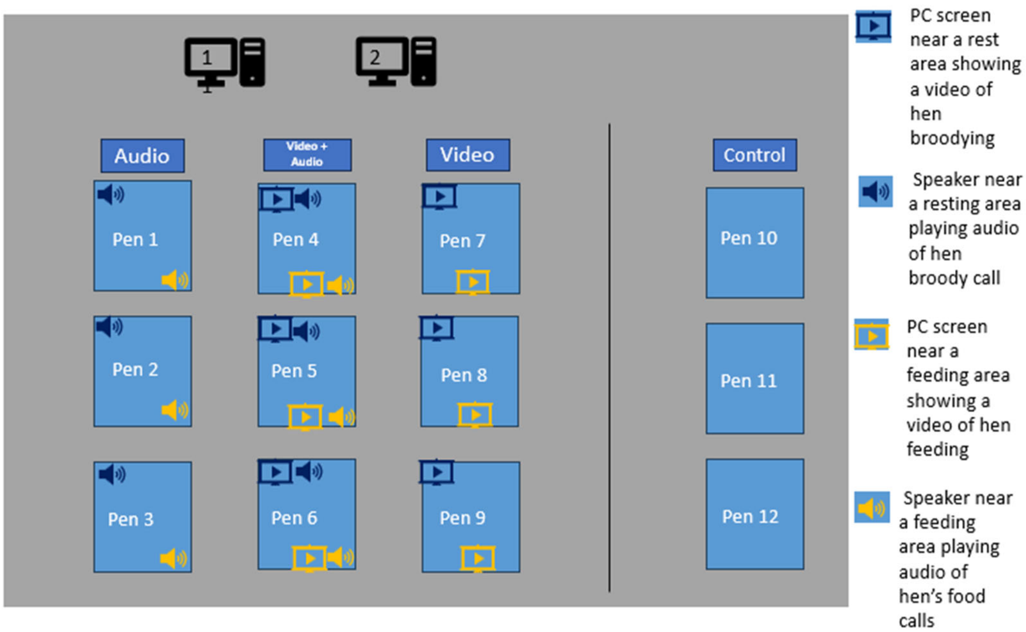


Figure 1. Setup design by Dr.Carlos E. Hernandez. This modified setup repeated 2 times to achieve 6 replicates. It shows 4 treatment group including Audio group, Audio + video group, video group and control group.

3.4 Recording and playing of cues setup

A camera was set at 190 cm above the centre of each pen. Behaviour of chicks were recorded every day from 9:30 (start of cues' playback) to 20:30 (end of cues 'playback).

A program was designed in a way that could play the cues based on the schedule given to it. Table 1 shows the schedule for feeding calls and resting calls.

Table 1. Cues' playback schedule

Cue	Duration
Feeding calls	10 minutes
No cue	8
Roosting calls	10 minutes
No cue	2 minutes
Feeding calls	10 minutes
No cue	8
Roosting calls	10 minutes
No cue	2 minutes

3.5 Behavioural analysis:

The behavioural analysis of chicks aimed to explore the attractiveness of the cues, identify which cues were more appealing to them, and to assess the synchronization of their behaviour in response to these cues.
Video analysis was done by observing the chick's behaviour on days 5, 10, and 15. Each day video analysis was started from 9:30 to 14:30 which was consider as morning analysis (AM), and 15:30 to 20:30 as evening analysis (PM).

The analysis was conducted using Observer XT software (Noldus). For behavioural analysis of chicks, this software was set up in a way that it was possible to write three values for each chick. These values were including "Period", "Behaviour", and "location".

3.5.1 Period:

The first value that was recorded for each chick was the period that observation was conducted during that.
Video analysis was done during three periods:

1) **Feed period:** Audios and/or videos playbacks of a broody hen feeding were presented in feeding areas.
2) **Nothing period**: None of the cues were presented during the nothing period.
3) **Rest Period:** Audios and/or videos playbacks of a broody hen brooding were presented in resting areas.

Each period was observed for 6 minutes, divided into three intervals of 2 minutes each. Each interval was observed for two seconds, during which the behaviour of each chick and its location were recorded.

3.5.2 Behaviour:

Chicks' behaviours were recorded based on ethogram of 15 mutually exclusive behaviours (Table 2).

Table 2. Ethogram of mutually exclusive behaviours

Behavioural category	**Definition**

Aggressive pecking	Pecking the head of another chick (Shimmura et al., 2010).
Drinking	Intake of water. Pecking with beak at the water in drinker with the pause to swallow water. Taking water in with their beak.
Dust bathing	Using feet, beak, and wings to make dirt of mound and lying down on litter and tossing dust and dirt with the wing and body movement on its wing and back.
Feed pecking	Peck toward feed in feed plate, eating from feed plate or peck at feed plate. Pecking at the ground within 1 chicken length (at 3d of age) from the feed plate was considered feed pecking as pelleted feed was always spilled out from feed plates.
Feather pecking	Pecking at other chicks' plumage or particles on plumage, feathers/skin. Grasping a feather, pulling it, and removing feather from body of another chick.
Ground pecking	Only pecks directed to the litter were classified as ground pecking. This included ground scratching as part of ground pecking as both are part of foraging behaviour.
Laydown	Sitting on the ground in a way their legs are bent, and their abdomen is on the ground. Also, if they neck is on the ground it is considered laydown
Locomotion activity	Birds changing location across the pen. Small changes in posture were not considered as locomotion activity and scored as standing still. All other behaviours like walking, running, jumping, flying was considered as locomotion activity.
Pecking screen or speaker	Any pecking behaviour directed toward the speakers or screens
Pecking at pen features	Pecking at walls or any object inside the pen including perches, doors, etc.
Preening	Arranging or oiling own feather with own beak (Shimmura et al., 2010)
Stand still	When chicks are in an upright position standing still. Small shifts in position are still counted as standing still.
Using perch	If a chick is seen on the perch.
Other	Any other behaviour that they perform not described above
Out of sight	If a chick was not visible

After observing each chick's behaviour for two seconds, the recorded behaviour became the second value for each chick.

3.5.3 Location:

To determine the location of each chick, the entire pen area was divided into 32 equal square sections. Each of these squares was assigned a different number between 1 and 32. Square number and location of resources was similar in all pens. (Figure 2). For this purpose, a transparent paper sheet was partitioned into 32 equal squares that was placed on screen during video analysis which covered the area of pen.

Figure 1. Locations number. This picture displays how the squares are numbered.

Initially, for each day video analysis, video observations were started with pens equipped with screens to identify potential technical issues related to video playback. This step was essential to determine precise timings for video analysis.

Video analysis was started 15 seconds after the start of videos on screen and at least 15 sec after the feed video turning off for Noting period. The reason for this was to avoid the transition period between stimuli.

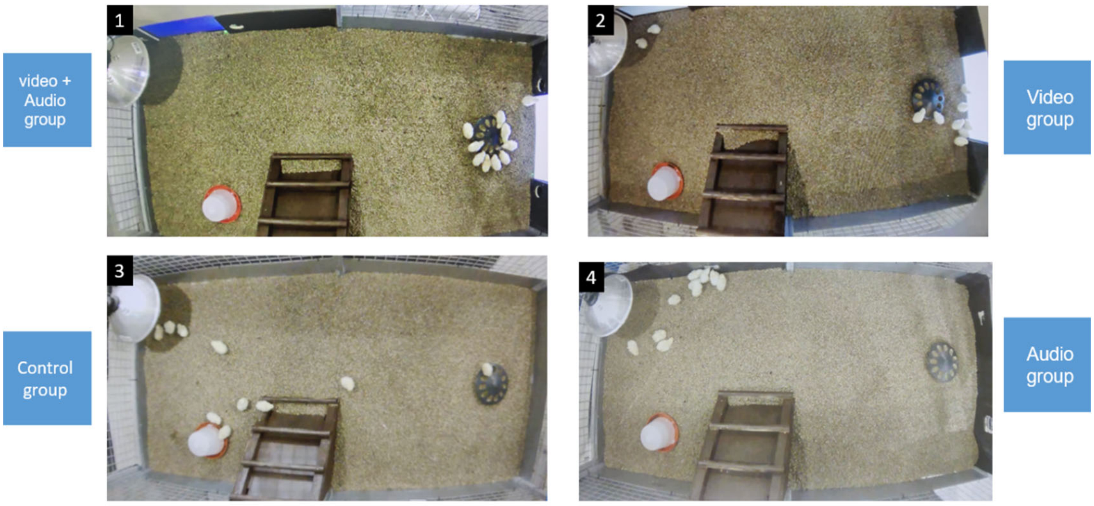


Figure 2. This image shows the four rearing treatments. It represents video+ audio group (Picture number 1), video group (Picture number 2), control group (Picture number 3), and audio group (Picture number 4).

3.6 Data Analysis:

All the videos were analysed by the same observer.
During video analysis, three values - "Period," "Behaviour," and "Location" - were recorded for each chick. The data collected was then extracted as an Excel file from this software.

3.6.1 Attractiveness of the cues:

To determine whether the chicks were attracted to the cues, their locations in relation to the cues were investigated. For this purpose, two methods were employed to define the feeding and resting areas.

Method 1

In this approach, the pen area was divided into three specific sections representing the intensity of the chicks' response. The squares in front of the cues that exhibited a high intensity of response from the chicks were categorized as "High response," indicated in red in Figures 4 and 5. This area included squares numbered 26, 27, 30, and 31 for the feeding area and 1, 2, 5, and 6 for the resting area (refer to Figure 5).

The squares located slightly further away but still near the cues were labelled as "Middle response," shown in blue. The middle response area comprised squares numbered 25, 29, 28, and 32 for the feeding area (Figure 4, and
3, 7, 9, and 10 for the resting area (Figure 5).
All other squares were categorized as "No response" areas, indicated in yellow.
Note: In this method, 'feeding area' refers to the high response area in relation to feeding cues, and 'rest area' indicates the high response area in relation to resting cues.

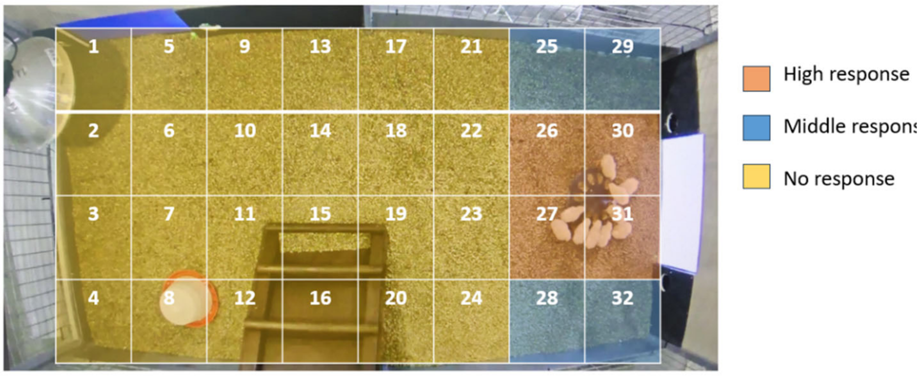


Figure 4, Three areas in relation to feeding cues. This picture illustrates the high response area (indicated in red), middle response area (shown in blue), and no response area (highlighted in yellow) in relation to the feeding cues.

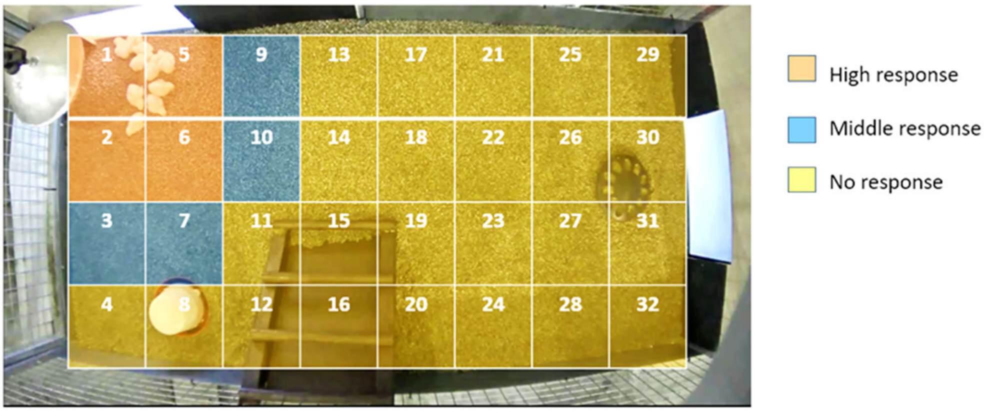


Figure 5. Three areas in relation to resting cues. This picture illustrates the high response area (indicated in red), middle response area (shown in blue), and no response area (highlighted in yellow) in relation to the resting cues.

Method 2:

In this method, an attempt was made to maintain an equal area size (i.e. number of squares) for each observed area. For this purpose, four specific areas were allocated for feeding cues, reflecting the varying distances of chicks from the feeding cues, as depicted in Figure 6.
Similarly, four specific areas were utilized to represent the distance of chicks from the resting cues, as shown in Figure 7.

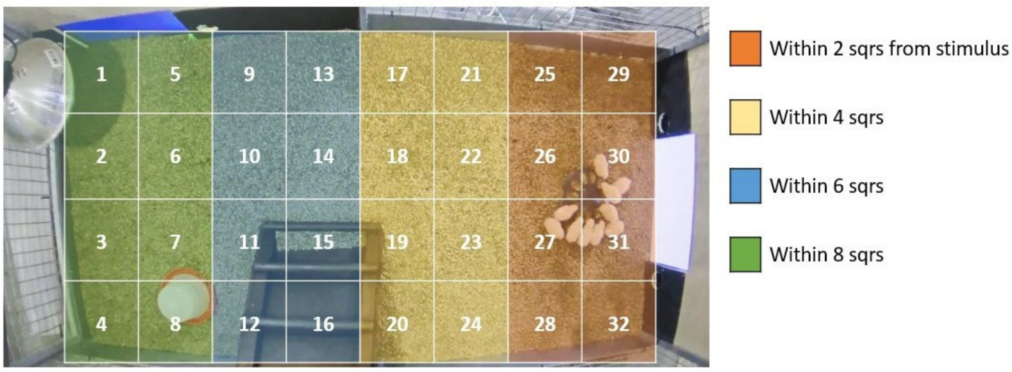


Figure 6, Distance from feeding cues. This photo demonstrates that the pen area is divided into four sections, each with an equal number of squares. Each section indicates the distance of the chicks from the feeding cues.

Figure 7. Distance from resting cues. This photo demonstrates that the pen area is divided into four sections, each with an equal number of squares. Each section indicates the distance of the chicks from the resting cues.

3.6.2 Behaviour of the chicks in response to the treatments

Another objective of this study was to investigate whether chicks exhibit social facilitation by performing the same behaviours as the ones performed in the videos. For this purpose, each behaviour was analysed separately.
To determine whether chicks exhibit feed pecking behaviour in response to feeding cues, in high-response areas, the analysis focused on the number of chicks engaged in feed pecking behaviour within this zone during various periods (feed, nothing, rest). A similar analytical approach was employed to determine the number of chicks lying down in high-response areas in relation to resting cues.

3.7 Ethical statement

The ethical committee of the Uppsala region within the Swedish Board of Agriculture (Jordbruksverket) granted authorization for all activities involving animals.

3.8 Statistical Analysis

All statistics were calculated using JMP Pro 17 software. The statistical analysis involved calculating Standard Least Square Means (LSM). Additionally, the Tukey's Honest Significant Difference (HSD) for post hoc test was used. Significance difference was set at $P \leq 0.05$.

For the statistical analysis of cue attractiveness, Treatment, Stimulus, Age (days), Replicate, AM/PM, Location, and up to 3 order interactions were used as fixed effects. In the Fixed Effect test table, P values greater than 0.05 were removed from the model in a stepwise manner. If Age, replicate, and AM/PM did not show statistical significance, they were retained in the model to account for variations across different time points.

Another aim of this study was to investigate whether chicks engaged in behaviors similar to those observed in the videos. For this purpose, the three specific locations in relation to feeding cues utilized with feed pecking behavior during different periods. A similar approach was applied to assess laydown behavior, with three specific areas in relation to resting cues. Each behaviour was analysed individually. Treatment, stimulus, age, replicate, AM/PM, and up to 3 order interactions were used as fixed effects. Other parts of the analysis were done similarly to the analysis of the attractiveness of cues.

To investigate the effects of treatments on other behaviours, location and stimulus were excluded from the data.
Behaviours included Dust bathing, Feather pecking, Ground pecking, Locomotion activity, Preening, pecking at pen fixtures (data of pecking at screen and speakers and pecking at pen fixture were combined), Stand still, and Using perch. Each behaviour was analysed independently. Treatment, age, replicate, AM/PM, up to 3 order interactions of treatment, age and replicate were used as fixed effects. Pen ID was used as a random effect. In the Fixed Effect test table, P values greater than 0.05 were removed from the model, starting with 3-way interactions and then 2-way interactions.

Residual of each analysis were used to assess whether data followed a normal distribution.

3.9 Potential challenges and solutions:

3.9.1 Technical problem regarding video playback

In situations where the screen was off at the beginning of feeding period or resting period (first interval), observation was done after 15 sec from the time the screen was turned on.
Additionally, if the screen was turn off in the second or third interval the observation took place as soon as the screen turned on.

Furthermore, if the screen was turned on in the beginning of nothing period (first interval) the observation was done after 15 sec from the part that screen turned off.

Since the computers were connected and played the videos almost at the same time, when the pens that had screen and speakers were analysed it is observed the parts that screen had problem were same in all the three pens that had screen. Consequently, observations for all other pens were conducted simultaneously. When the monitors displayed the cues, the audio playbacks in the pens equipped with speakers simultaneously played the cues.

3.9.2 Challenges regarding each chick's location

As explained in behavioural analysis section location of each chick was recorded based on the numbers assigned to squares covering the pen area.
If a chick was on the line between two squares, a square number was written for that chicken, where the majority of the chicken's body was positioned. If the chicken's body was equally placed in these two squares, a square number was written where the chicken's head was located.

Also, if the chicken's body was located in a part where every part of its body was placed in four squares, first, attention was paid to the point in which square the largest part of its body is located, and if it was placed relatively equally in these four squares, the number of square where the chicken's head was placed was written as the location of that chick.
Figure 3 represents how the location of each chick was recorded in these situations.

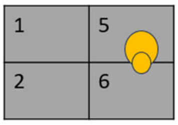
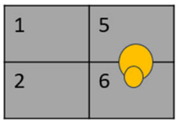
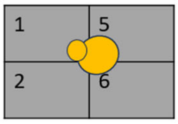
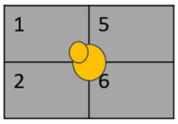
	Square number 5 is considered for this chicken location. Since the majority of its body is located on square number 5.
	Square number 6 is considered for this chicken location. Since its body is almost equally placed in the line between square number 5 and 6, a square number 6 show the location of this chick where the chicken's head is located.
	Square number 5 is considered for this chicken location. Since the majority of its body is located on square number 5.
	Square number 1 is considered for this chicken location. Since its body placed relatively equally in these four squares, the number of square where the chicken's head is placed is consider as the location of this chick.

Figure 3. This picture depicts how the location of each chick was recorded in case a chick is positioned between 2 or more squares.

4. Results

4.1 Attractiveness of cues

Method 1:

Three Areas in Relation to Feeding Cues

The number of birds observed in the high-response area during the presentation of feeding cues in the Video + Audio and Video groups was approximately twice the number of chicks observed in the same area in the Audio and Control groups (Table 3). Moreover, the number of chicks observed in the high-response area in the absence of any cues in the Video group was significantly higher than the number of chicks in this area in the three other treatment groups. Additionally, the results indicate that more than half of the chicks were located in the no response area, irrespective of the stimulus and treatment groups (Table 3).

Table 3. Three areas in relation to feeding cues. Effects of Treatments and stimulus on LSM of birds observed in three locations in relation to feeding cues, that characterize the intensity of chicks' responses.

	Stimulus								
	Feed			Nothing			Rest		
Treatment	High response	Middle response	No response	High response	Middle response	No response	High response	Middle response	No response
Audio	80.9 ±4.1 ^{ij}	4.5 ±4.4 ^{m}	214.2 ±4.1 ^{de}	65.6 ±4.1 ^{jk}	5.1 ±4.6 ^{m}	229.4 ±4.1 ^{bcd}	52.0 ±4.1 ^{kl}	4.8 ±4.4 ^{m}	243.8 ±4.1 ^{abc}
Video/Audio	123.1 ±4.2 ^{h}	6.2 ±4.3 ^{m}	166.6 ±4.2 ^{f}	69.3 ±4.1 ^{jk}	5.3 ±4.4 ^{m}	223.4 ±4.1 ^{cd}	31.9 ± 4.2 ^{l}	1.5 ±5.6 ^{m}	263.6 ± 4.1 ^{a}
Video	139.8 ±4.1 ^{gh}	7.6 ± 4.3 ^{m}	151.3 ±4.1 ^{fg}	95.4 ±4.1 ^{i}	4.0 ±4.3 ^{m}	200.2 ±4.1 ^{e}	46.9 ±4.2 ^{kl}	4.3 ± 5 ^{m}	247.4 ±4.2 ^{ab}
Control (no stimulus)	64.9 ±4.1 ^{jk}	7.2 ±4.5 ^{m}	227.7 ±4.1 ^{bcd}	62.3 ±4.1 ^{jk}	7.2 ±4.3 ^{m}	230.2 ±4.1 ^{bcd}	62.3 ±4.1 ^{jk}	5.7 ±4.3 ^{m}	229.6 ±4.1 ^{bcd}

Least square mean ± SEM.

Groups that do not share a letter are significantly different (P≤0.05).

Three areas in relation to resting cues:

When resting cues were played, approximately 71 percent of chicks in the Video + Audio group were attracted to the high response area, and there was no significant difference between this group and the Video group. This percentage for the Audio group was around 56 percent, and for the control group, it was approximately 52 percent (Table 4). Although Audio group attracted numerically more chicks to high response area concerning resting cues, there was no significant difference between this group and Control group ($P \leq 0.05$).

Table 4. Three areas in relation to resting cues. Effects of treatments and stimulus on LSM of birds observed in three locations in relation to resting cues, that characterize the intensity of chicks' responses.

	Stimulus								
	Feed			**Nothing**			**Rest**		
Treatment	High response	Middle respons	No response	High response	Middle respons	No response	High response	Middle respons	No response
Audio	154.8 ±5.2 ^{cde}	31.3 ±5.2 ^{ij}	113.0 ±5.2 ^{fgh}	173.0 ±5.2 ^{bc}	32.0 ±5.2 ^{ij}	94.6 ±5.2 ^{gh}	167.2 ±5.2 ^{bcd}	39.9 ±5.2 ^{ij}	93.0 ±5.2 ^{gh}
Video/Audio	117.7 ±5.3 ^{fg}	22.0 ±5.4 ^{j}	157.7 ±5.3 ^{cde}	160.0 ±5.2 ^{bcde}	31.7 ±5.2 ^{ij}	103.7 ±5.2 ^{gh}	211.1 ±5.2 ^{a}	29.9 ±5.2 ^{ij}	55.2 ±5.2 ^{i}
Video	98.6 ±5.2 ^{gh}	19.1 ±5.2 ^{j}	180.6 ±5.2 ^{bc}	136.4 ±5.2 ^{ef}	22.4 ±5.2 ^{j}	140.7 ±5.2 ^{def}	187.9 ±5.3 ^{ab}	25.5 ±5.3 ^{j}	84.6 ±5.3 ^{h}
Control (no stimulus)	162.2 ±5.2 ^{bcde}	32.4 ±5.2 ^{ij}	104.1 ±5.2 ^{gh}	162.0 ±5.2 ^{bcde}	36.1 ±5.2 ^{ij}	100.5 ±5.2 ^{gh}	156.1 ±5.2 ^{cde}	37.7 ±5.2 ^{ij}	104.0 ±5.2 ^{gh}

Least square mean and ± SEM.

Groups that do not share a letter are significantly different ($P \leq 0.05$).

Method 2:

Distance from feeding cues

The number of chicks that were within 2 squares from feeding cues during feeding period in both Audio and Control group were half of the number of chicks in the same area in Video+ audio and Video group ($P \leq 0.05$). There was a significantly higher number of chicks within 2 squares from feeding cues in Video group, when there were no cues compared to the three other groups. Also, regardless of the period, there were consistently more chicks within 8 squares from the feeding cues ($P \leq 0.05$). Furthermore, an equal number of chicks in the Audio and Control groups were distributed evenly across the pen in each area (Table 5).

Table 5. Distance from feeding cues. Effects of treatments and stimulus on Least Square Means of Bird Counts in four location concerning feeding cues.

	Stimulus											
	Feed				**No stimulus**				**Rest**			
	Distance from stimulus (squares)				Distance from stimulus (squares)				Distance from stimulus (squares)			
Treatment	2	4	6	8	2	4	6	8	2	4	6	8
Audio	84.9 ± 4.2 ^{g,h}	8.4 ± 4.3 ^{m}	18.1 ± 4.2 ^{l,m}	187.9 ± 4.2 ^{c,d}	70.2 ± 4.2 ^{h,i}	6.9 ± 4.3 ^{m}	16.3 ± 4.2 ^{l,m}	206.6 ± 4.2 ^{b,c}	56.3 ± 4.2 ^{i,j}	12.0 ± 4.3 ^{l,m}	28.9 ± 4.2 ^{k,l,m}	203.4 ± 4.2 ^{b,c}
Video/Audio	129.5 ± 4.3 ^{e}	9.4 ± 4.3 ^{l,m}	16.7 ± 4.3 ^{lm}	140.0 ± 4.3 ^{e}	71.9 ± 4.2 ^{h,i}	9.4 ± 4.2 ^{l,m}	20.3 ± 4.3 ^{l,m}	194.1 ± 4.2 ^{c,d}	33.4 ± 4.3 ^{j,k,l}	6.3 ± 4.4 ^{m}	17.6 ± 4.3 ^{l}	240.0 ± 4.2 ^{a}
Video	147.1 ± 4.2 ^{e}	9.5 ± 4.2 ^{m}	19.6 ± 4.3 ^{l,m}	122.7 ± 4.2 ^{e,f}	99.3 ± 4.2 ^{f,g}	9.0 ± 4.3 ^{m}	16.5 ± 4.2 ^{l,m}	175.0 ± 4.2 ^{d}	50.1 ± 4.3 ^{i,j,k}	8.6 ± 4.3 ^{m}	17.5 ± 4.3 ^{l,m}	221.9 ± 4.3 ^{a,b}
Control (no stimulus)	70.9 ± 4.2 ^{h,i}	6.9 ± 4.3 ^{m}	20.3 ± 4.2 ^{lm}	200.5 ± 4.2 ^{bc}	68.4 ± 4.2 ^{hi}	8.3 ± 4.5 ^{m}	18.4 ± 4.2 ^{l,m}	204.4 ± 4.2 ^{b,c}	68.2 ± 4.2 ^{h,i}	10.0 ± 4.4 ^{l,m}	24.8 ± 4.2 ^{lm}	196.6 ± 4.2 ^{cd}

Least square mean ± SEM.

Groups that do not share a letter are significantly different (P≤0.05).

Distance from resting cues

According to Table 6, approximately 80 percent of the chicks were observed within 3 squares from resting cues during the resting period ($P \leq 0.05$). This percentage was significantly higher compared to the other three treatment groups ($P<0.05$). Furthermore, while the Video group attracted more chicks within 3 squares from resting cues numerically, when resting cues were presented, there was no significant difference between this group, the Audio group, and the Control group.

Table 6. Distance from resting cues. Effects of treatments and stimulus on Least Square Means of Bird Counts in four location concerning resting cues.

	Stimulus											
	Feed				**No stimulus**				**Rest**			
	Distance from stimulus (squares)				Distance from stimulus (squares)				Distance from stimulus (squares)			
Treatment	2	4	6	8	2	4	6	8	2	4	6	8
Audio	186.1 ± 4.1^{c}	$19.9 \pm 4.1^{m,n,o,p}$	8.3 ± 4.2^{p}	$84.9 \pm 4.1^{h,i}$	$205.0 \pm 4.1^{b,c}$	$19.0 \pm 4.2^{m,n,o,p}$	6.9 ± 4.2^{p}	70.2 ± 4.1 i,j	$207.1 \pm 4.1^{b,c}$	$25.1 \pm 4.1^{m,n,o,p}$	$12.0 \pm 4.2^{m,n,o,p}$	$56.3 \pm 4.1^{j,k}$
Video/Audio	$138.2 \pm 4.2^{d,e,f}$	$18.6 \pm 4.2^{m,n,o,p}$	$9.4 \pm 4.2^{m,n,o,p}$	$129.5 \pm 4.2^{e,f}$	$191.6 \pm 4.1^{b,c}$	$22.3 \pm 4.1^{m,n,o,p}$	$9.4 \pm 4.1^{n,p}$	$71.9 \pm 4.1^{i,j}$	241.0 ± 4.1^{a}	$16.2 \pm 4{,}1^{m,n,o,p}$	6.3 ± 4.3^{p}	$33.4 \pm 4.2^{k,l,m,n}$
Video	$117.8 \pm 4.1^{f,g}$	$24.0 \pm 4.1^{m,n,o,p}$	$9.5 \pm 4.1^{o,p}$	$147.1 \pm 4.1^{d,e}$	158.8 ± 4.1^{d}	$32.7 \pm 4.1^{l,m,o}$	$9.0 \pm 4.2^{o,p}$	$99.3 \pm 4.1^{g,h}$	213.0 ± 4.2^{b}	$26.4 \pm 4.2^{l,m,n,o,p}$	$8.6 \pm 4.2^{o,p}$	$50.0 \pm 4.2^{j,k,l}$
Control (no stimulus)	$194.6 \pm 4.1^{b,c}$	$26.3 \pm 4.1^{l,m,n,o,p}$	6.9 ± 4.2^{p}	$70.9 \pm 4.1^{i,j}$	$198.1 \pm 4.1^{b,c}$	$24.7 \pm 4.1^{m,n,o,p}$	8.3 ± 4.4^{p}	$68.4 \pm 4.1^{i,j}$	$193.8 \pm 4.1^{b,c}$	$26.8 \pm 4.1^{l,m,n,o,p}$	$10.1 \pm 4.3^{m,n,o,p}$	$68.2 \pm 4.1^{i,j}$

Least square mean ± SEM.
Groups that do not share a letter are significantly different ($P \leq 0.05$).

4.2 Behaviour of the chicks in response to the treatments

Aggressive pecking

In this study, a low number of occurrences of aggressive pecking were recorded, totalling 56 instances. Table 7 shows the number of recorded aggressive pecking occurrences in relation to stimulus and treatments.

Table 7. Number of observed Aggressive pecking in each treatment and different period

	Stimulus		
Treatment	Feed	Nothing	Rest
Audio	2	2	3
Video+ Audio	6	3	5
Video	6	5	5
Control	2	12	5

Feed pecking

The findings from Table 8 reveal that, during the presentation of feeding cues, the number of chicks engaging in feed pecking behaviour in the Video + Audio and Video groups on days 5 and 10 was significantly higher compared to the number of chicks in these treatments during the nothing and resting periods of the same age Furthermore, in the Video + Audio group, the number of chicks exhibiting feed pecking behaviour when resting cues were played was approximately half of the number observed pecking on feed during the period when feeding cues were presented($P \leq 0.05$). In addition, significantly more chicks in video group of day 5 observed performing feed pecking compared to Audio and Control group during feed, nothing and resting period ($P \leq 0.05$).

Effects of stimulus on feed pecking behaviour were also significant ($P \leq 0.05$). It shows that during the feeding period, a significantly higher number of chicks performed feed pecking behaviour in high-response areas in relation to feeding cues compared to the nothing period and resting period ($P \leq 0.05$).

Chicks exhibited a significantly lower frequency of feed pecking behaviour at 15 days old ($P \leq 0.05$).

Our findings suggest that chicks exhibit a significantly higher frequency of feed pecking behaviour in the evening compared to the morning ($P \leq 0.05$).

Table 8. Effects of Treatments, Age and Stimulus on the LSM of bird counts performing feed pecking behaviour in Feeding area

	Stimulus											
	Feed				Nothing				Rest			
Age	**Treatment**											
	Audio	Video+ Audio	Video	Control	Audio	Video+ Audio	Video	Control	Audio	Video+ Audio	Video	Control
D5	62.8 ±5.3^{bcd}	87.1 ±5.3^{abc}	92.8 ±5.3^{a}	47.4 ±5.3^{defgh}	44.7 ±5.3^{defghi}	61.0 ±5.3^{cde}	58.8 ±5.3^{cdef}	46.4 ±5.3^{defghi}	44.0 ±5.3^{defghi}	23.7 ±5.3^{ghi}	31.5 ±5.3^{fghi}	41.8 ±5.3^{defghi}
D10	65.3 ±5.3^{abcd}	91.9 ±5.3^{ab}	86.8 ±5.3^{abc}	52.2 ±5.3^{defg}	51.9 ±5.3^{defg}	39.4 ±5.3^{defghi}	46.56 ±5.3^{defghi}	51.0 ±5.3^{defgh}	32.2 ±5.3^{efghi}	22.2 ±5.3^{hi}	31.0 ±5.3^{fghi}	51.3 ±5.3^{defgh}
D15	47.8 ±5.3^{defgh}	64.5 ±5.3^{abcd}	45.2 ±5.3^{defghi}	45.7 ±5.3^{defghi}	42.6 ±5.3^{defghi}	36.9 ±5.3^{defghi}	39.5 ±5.3^{defghi}	39.1 ±5.3^{defghi}	27.5 ±5.3^{ghi}	17.8 ±5.3^{i}	26.8 ±5.3^{ghi}	42.5 ±5.3^{defghi}

Least square mean ±SEM.

Groups that do not share a letter are significantly different ($P \leq 0.05$).

Laydown

Table 9 indicates that chicks exhibited significantly more laydown behaviour during the Nothing period (except for the Video group), during the resting period, and in treatments without any screens The groups with Video + Audio and Video (during the feed period and nothing period) demonstrated the least laydown behaviour (Table 9).

Table 9. Effects of Treatment and Stimulus on LSM of Bird Counts performing laydown behaviour in rest area.

	Stimulus		
Treatment	Feed	Nothing	Rest
Audio	87.6 ±4.8^{bcd}	106.7 ±4.8^{a}	98.6 ±4.8^{ab}
Video+ Audio	75.6 ±4.9^{cde}	87.0 ±4.8^{abcd}	102.0 ±4.8^{ab}
Video	58.8 ±4.8^{e}	70.4 ±4.8^{de}	92.8 ±4.9^{abc}
Control	99.1 ±4.8^{ab}	100.7 ±4.8^{ab}	93.9 ±4.8^{abc}

Least square mean ± SEM.

Groups that do not share a letter are significantly different (P≤ 0.05).

According to Table 10, chicks exhibited significantly more laydown behaviour on day 5 compared to days 10 and 15. However, in the Video group, significantly less laydown behaviour was observed on day 5. Despite this, chicks in the Video group on day 5 performed significantly more laydown behaviour compared to the Video groups on days 10 and 15. Furthermore, chicks in the Control and Audio groups on day 5 performed significantly more laydown behaviour than those in the Video + Audio and Video groups (Table 10).

Table 10. Effects of treatment and age on LSM of bird counts performing laydown behaviour in rest area.

High response area in relation to resting cues			
	Age		
Treatment	D5	D10	D15
Audio	125.2 ±4.8^{ab}	87.7 ±4.8^{de}	79.9 ±4.8^{de}
Video+ Audio	111.2 ±4.8^{bc}	77.6 ±4.9^{de}	75.8 ±4.8^{def}
Video	97.2 ±4.9^{cd}	55.0 ±4.8^{f}	69.8 ±4.8^{ef}

Control	134.8 ±4.8[a]	80.6 ±4.8[de]	78.4 ±4.8[de]

Least square mean ±SEM.
Groups that do not share a letter are significantly different (P≤ 0.05).

Significantly more chicks exhibited laydown behaviour in replicates 1 and 2 on day 5 compared to these replicates on days 10 and 15(P ≤0.05). There was no significant difference in the observation of laydown behaviour between replications on day 15(P ≤0.05).

Significantly more chicks exhibited laydown behaviour in the morning of day 10 compared to the evening.

Results from the interactions of replicate and stimulus show that replication 1 had the highest number of chicks laying down in the rest area during the resting period (P ≤0.05). There was a significant difference between Replication 1 and Replication 2 during this period. Additionally, Replication 1 had the least number of chicks performing laydown behaviour during the feeding period (Table 11).

Table 11, Effects of stimulus and Replication on LSM of Bird Counts performing laydown behaviour in rest area.

	Replication	
Stimulus	R1	R2
Feed	73.4 ±3.4[c]	87.1 ±3.4[bc]
Nothing	92.2 ±3.4[b]	90.2 ±3.4[b]
Rest	107.0 ±3.4[a]	86.6 ±3.4[bc]

Least square mean ±SEM.

Groups that do not share a letter are significantly different (P≤ 0.05).

Dust bathing

Table 12 shows that Video + Audio and Video of Replication 1 exhibited significantly more dust bathing behaviour than the Video group of Replication 2(P ≤0.05).

There was no significant difference between treatments for dust bathing behaviour (P ≤0.05). Chicks performed significantly less dust bathing behaviour at 5 days of age compared to 10 and 15 days of age (P ≤0.05). Moreover, chicks engaged in significantly more dust bathing behaviour in the afternoon compared to the morning (P≤0.05)."

Table 12, Effects of Replicate and Treatment on perfoming Dust bathing behaviour

	Replication	
Treatment	R1	R2
Audio	2.7 ±0.2^{ab}	2.3 ±0.3^{ab}
Video+ Audio	2.9 ±0.2^{a}	1.9 ±0.3^{ab}
Video	3.1 ±0.3^{a}	1.8 ±0.3^{b}
Control	1.9 ±0.3^{ab}	2.2 ±0.2^{ab}

Least square mean ±SEM.
Groups that do not share a letter are significantly different (P≤ 0.05).

Feather Pecking

Table 13 shows that chicks in the Control group on day 10 performed significantly more feather pecking than the Video + Audio group at the same age There was no differences between treatments in day 5 and 15 (P ≤0.05).

Chicks at 10 days old exhibited the highest feather pecking (P≤0.05), while those at day 5 showed the least feather pecking. Additionally, replicate 1 had significantly more feather pecking than Replicate 2 (P ≤0.05).

Table 13. Effects of Treatment and Age on feather pecking

		Age	
Treatment	D5	D10	D15
Audio	2.4 ±0.5^{b}	4.3 ±0.5^{b}	3.0 ±0.5^{b}
Video+ Audio	2.7 ±0.5^{b}	3.5 ±0.5^{ab}	4.0 ±0.5^{ab}
Video	2.7 ±0.5^{b}	4.6 ±0.5^{ab}	3.2 ±0.5^{b}
Control	2.6 ±0.5^{b}	6.2 ±0.5^{a}	3.1 ±0.5^{b}

Least square mean ±SEM.
Groups that do not share a letter are significantly different (P≤ 0.05).

Ground pecking

The occurrence of ground pecking increased with age (P≤0.05). Additionally, significantly more ground pecking was observed in the evening than in the morning

(P≤0.05). The 2-way interaction of age and replicate was significant (P≤0.05). Significantly more ground pecking behaviour was observed in Replication 2 of day 10 and 15 compared to Replication 1.

Locomotion activity

According to Table 14, significantly more chicks in the Video group exhibited locomotion activity during the feed period compared to other treatments (P ≤0.05). In the nothing period, significantly more locomotion activity was observed in the Video + Audio group compared to the Audio group. However, no significant differences were found between treatments when resting cues were playing (P ≤0.05).

The evening of Replication 1 showed significantly less locomotion activity compared to both the morning of Replications 1 and 2 (P≤0.05).

Significantly more locomotion activity was found in day 10 compared to day 5 and 15 (P≤0.05). Chicks perform significantly less Locomotion activity during day 15 (P≤0.05).

Table 14, Effects of Treatment and Stimulus on Locomotion activity

Locomotion activity			
	Stimulus		
Treatment	Feed	Nothing	Rest
Audio	28.9 ±1.9^{d}	29.6 ±1.9^{cd}	34.6 ±1.9^{abcd}
Video+ Audio	33.7 ±1.9^{bcd}	38.69 ±1.9^{ab}	37.9 ±1.9^{abc}
Video	42.7 ±1.9^{a}	35.3 ±1.9^{abcd}	39.0 ±1.9^{ab}
Control	33.1 ±1.9^{bcd}	35.2 ±1.9^{abcd}	36.6 ±1.9^{abcd}

Least square mean ±SEM.
Groups that do not share a letter are significantly different (P≤ 0.05).

Significantly more locomotion activity was observed during the rest period of replication 2 compared to the feed period of same replication (Table 15).

Table 15, Effects of Replicate and Stimulus on Locomotion activity

	Replication	
Stimulus	R1	R2

Feed	35.6 ±1.4^{ab}	33.6 ±1.4^{b}
Nothing	34.0 ±1.4^{ab}	35.4 ±1.4^{ab}
Rest	34.8 ±1.4^{ab}	39.3 ±1.4^{a}

Least square mean ± SEM.
Groups that do not share a letter are significantly different (P≤ 0.05).

Pecking at pen fixture

Chicks in the Video + Audio and Video groups pecked at pen fixtures significantly more than the Control group (P ≤0.05). Significantly fewer chicks pecked at pen fixtures on day 15 compared to days 5 and 10 (P≤0.05). In the morning, more chicks pecked at pen fixtures than in the evening (P≤0.05). More chicks pecked at pen fixtures in the morning of day 5 compared to the evening of the same day (P≤0.05).

Preening

More preening behaviour were observed with increasing age(P≤0.05). More preening behaviour were observed in Replication 1 than 2(P≤0.05). Result from 2-way interactions of Age and Replicate show that significantly more preening behaviour was observed in replication 1 of day 15(P≤0.05). Furthermore, more chicks performed preening behaviour in Replication 1 of day 5 than Replication 2.

Stand still

Significantly more chicks in the Video group exhibited standstill behaviour compared to the Video + Audio and Control groups, with no significant difference between the Audio group and the three other treatments (P≤0.05).
As the chicks aged, there was a decrease in the incidence of standstill behaviour. Additionally, an increased frequency of standstill behaviour in the evening was observed compared with the morning.
Table 16 illustrates that the highest number of chicks displaying standstill behaviour was observed in the Video group on day 5 (P≤0.05). Furthermore, significantly more chicks exhibited standstill behaviour in the Audio group on day 5 compared to day 10 and 15. Additionally, a higher incidence of standstill behaviour was observed in the Video + Audio group on day 5 compared to day 15.

A significantly higher number of chicks were observed standing still in day 5 Replication 1 compared to day 10 and 15 in both replications (P ≤0.05). Additionally, there was a significant increase in standstill behaviour in the evening of day 5 and 10 compared to the morning of these days.

Table 16, Effects of Treatment and Age on number of chicks performing stand still behaviour

Stand still			
		Age	
Treatment	D5	D10	D15
Audio	37.6 ±1.8^{b}	29.9 ±1.8^{cd}	27.3 ±1.8^{cd}
Video+ Audio	33.8 ±1.8^{bc}	28.2 ±1.8^{cd}	21.9 ±1.8^{d}
Video	46.5 ±1.8^{a}	33.8 ±1.8^{bc}	28.6 ±1.8^{cd}
Control	33.0 ±1.8^{bc}	30.1 ±1.8^{bcd}	27.5 ±1.8^{cd}

Least square mean ±SEM.
Groups that do not share a letter are significantly different (P≤ 0.05).

Using perch

Significantly more chicks used perches in the evening in the Control group than in the evening in the Audio group. There was no significant difference in perch usage between other treatments during both morning and evening.

There was a significant difference between Replication 1 and 2 on day 15 (P≤0.05). Significantly more chicks used perches in Replication 1 on day 15 compared to Replication 2 (P≤0.05). There was no significant difference in perch usage between replicates on days 5 and 10.

The use of perches increased with age (P≤0.05). There was no significant difference between Replication 1 and 2 in the morning and evening in perch usage.

In this study we did not find any significant difference between body weight of chicks in differen

5. Discussion

5. 1 Attractiveness of cues

The primary objective of this research was to assess various methods of providing visual and auditory cues, with the aim of determining the most effective approach for attracting chicks to specific resources.

Chicks' locations during stimulation with feeding cues

In this study we found out during the feeding cues presentation, notably, both the Video group and the Video + Audio group attracted a significantly higher percentage of chicks to the High Response area concerning feeding cues. Similarly, the second method demonstrated that when feeding cues were played, both the Video and Video + Audio groups effectively attracted significantly more chicks, to the locations within 2 squares from the feeding cues. This finding shows the attractiveness of visual and visual and auditory stimuli to attract chicks to feeding cues.

In the study conducted by Clarke and Jones in 2001, chicks were observed moving closer to the screen positioned over a picture of a goal box, indicating their ability to recognize conspecifics on video and respond to that by moving closer to the screen.

Our findings also indicate that adding auditory cues of food call to the visual cues did not demonstrate additive effects, implying that the approach was a reflection of attraction specifically to the visual cues.

A significantly greater number of chicks were observed in the no response area in relation to feeding cues compared to the high response area. This difference may be attributed to the no response area being approximately six times larger than the high response area. Consequently, a substantial proportion of animals were noted in this expansive area.

The Audio group showed results statistically similar to the Control group for attracting chicks to the high response area concerning feeding cues, which was significantly lower than the Video and Video + Audio groups. This finding was consistent with the second method, where the number of chicks within 2 squares from the feeding cues in the Audio and Control groups was approximately half of those in the Video and Video + Audio groups, suggesting that auditory stimuli alone did not make a difference compared to the group that did not have any stimuli. Although the Audio group numerically attracted more chicks than the Control group to the high response area during the feeding period, this difference was not significant. This could be due to the fact that auditory playback of feeding cues alone might not be strong enough to attract chicks to the feeding area. These results align with a study conducted by Van Kampen in 1994, where hens did not exhibit any inclination to move closer or approach the source of sound when food calls were played. Similarly, research by Evans and Evans in 1999 found no significant effects on hens' approach behaviour towards speakers when male food calls were played back. In this study, the researchers proposed that the way hens' approach is probably affected by visual signals from the male, such as particular displays like tidbitting. These visual cues might also contribute to stimulating the chicks' approach behaviour. In another study, chicks were presented with videos showing conspecifics engaged in feeding, either with or without accompanying sounds, or a video of the goal box, with or without the soundtrack of chicks. They approached the videos of other chicks faster than those of the goal box and there was no distinction in attractiveness between the versions with sound and the silent ones (Clarke & Jones, 2001). However, Kruijt (1964) and Andrew (1964) observed Gentle vocalizations or pleasure calls, have the capacity to elicit approach responses in chicks.

When looking at the resting area, it's clear that nearly half of the chicks in the Video group and approximately 43.8% of the chicks in the Video + Audio group were within 8 squares from the resting area (that means they were near feeding area and far away from resting area) when feeding cues were playing. This indicates a heightened attraction of chicks in these two groups towards the feeding cues.

Chicks' location during no periods without stimulation (i.e. no audio or video cues):

During the nothing period, there were significantly more chicks in the High Response area concerning feeding cues in the Video group compared to the other three groups. A similar result was found in the second method, where more chicks remained within 2 squares from the feeding area in the Video group, it was significantly higher than the other three treatment groups. This result demonstrates that since the nothing period immediately follows the feeding period, more chicks

stayed in the high Response area in the group that had only a screen. This could be because, without any auditory cues, the chicks did not notice when the feeding cues stopped, leading them to remain in the area.
Moreover, although behaviours were recorded during the time that there were not any cues, the software designed to play the cues encountered some problems, especially for the groups that had only a screen. For example, the screen suddenly turned on and off. These issues might have attracted the chicks to this area, and then the screen was turned off and chicks stayed there when the behavioural observation took place. Interestingly, more than half of the chicks were within 8 squares from feeding cues during this period, suggesting that when there were not any cues more chicks were in or close to the resting area.
Interestingly, more than half of the chicks were within 8 squares from feeding cues during this period, suggesting that when there were not any cues more chicks were in or close to the resting area.

Chicks' location in relation to the resting cues:

During the resting period, significantly higher number of chicks were attracted to both high response area concerning resting cues and within 3 squares from resting cues. This finding indicates the effectiveness of the combined visual and auditory stimuli of resting cues in attracting and maintaining the chicks in the desired area.
During the resting cues presentation, significantly fewer chicks were observed in the high response area concerning feeding cues in the Video+ audio group. Following that, fewer chicks were also observed in the High Response area in the Video group. This indicates that due to the resting cues being played, more chicks were attracted to the resting stimulations (no Response area), resulting in fewer chicks in the high response area concerning feeding cues. However, significantly more chicks were observed in the High Response area in the Control and Audio groups compared to the other two groups, and there were no significant differences between these two groups. The second method (which were used to investigate the attractiveness of cues) involved ensuring that each observed area had an equal number of squares, yielded similar results. Approximately 80% of the chicks were observed within 8 squares from the feeding cues in the Video + Audio group, and around 74.4% were in the Video group. These results emphasize the fact that the presence of a screen displaying resting cues, or the combination of visual and auditory stimuli of these cues, elicited a stronger response from the chicks, leading to a significant attraction towards the resting area. In the Audio group, although numerically more chicks were attracted to the resting cues compared to the control group, the difference was not statistically significant. This demonstrates the remarkable effects of visual and auditory stimuli together in comparison to the effect of auditory stimuli alone.

Interestingly, when considering the resting area across all methods, regardless of the periods (Feed, Nothing, and Rest), it becomes apparent that a high number of chicks were consistently found in this area. This observation suggests that it might be due to the fact that chicks spend more time in the resting area, away from the feeding area, throughout the day.

5.2 Behaviour of the chicks in response to the type of stimulation

Aggressive pecking

Chickens establish a social order through pecking, aiming mainly at the heads of others. This behavior, while forceful, can cause discomfort and prompt vocal responses or withdrawal from the receiving bird. (Savory, 1995). In the study conducted by (Fält, 1978), it was observed that the frequency of aggressive behavior during the period was higher in the non-brooded groups than in the brooded ones.

Due to the low number of instances of Aggressive pecking, a comparative analysis between treatments was not feasible. However, it appears that the control group during the Nothing period had significantly more Aggressive pecking occurrences. This outcome may imply that providing Visual and Auditory cues had a positive effect in reducing aggressive pecking.

Although the number of aggressive pecking instances was recorded during the 'nothing' period for the control group, this period lacks meaning for them. Unlike other groups, the control group was not subjected to feeding, nothing, or resting periods, as recorded in the program. Therefore, it may be appropriate to state that the overall number of aggressive pecking instances was higher in the control group. This outcome suggests that providing visual and auditory cues possibly had a positive effect in reducing aggressive pecking.

Feed pecking

Workman and Andrews (2016) propose that hens primarily communicate information regarding food palatability to their chicks before they reach eight days of age (Workman & Andrews, 2016). During this critical period, chicks exhibit increasingly rapid responses to food calls as they mature, implying a potential role for learning in shaping their reactions (Wauters & Richard-Yris, 2002).

In our study, a higher number of chicks exhibited feed pecking behaviour during the periods when feeding cues were presented in treatments with both screens and speakers, as well as in treatments with screens alone. Aligned with the research conducted by Riber et al. (2007) and Wauters & Richard-Yris (2002), it was observed that following a feeding display by a hen, the chicks demonstrated increased eating behavior, primarily directed towards the same food source as the brooding hen. Remarkably, this behavior persisted even after the hen's presentation had concluded.

A significantly higher number of chicks in the video group on day 5 were observed performing feed pecking compared to the audio and control groups during the feed period. This result suggests that chicks respond more effectively to visual cues than to auditory cues. Notably, there was no significant difference between the video + Audio groups and the audio or control groups. This indicates that the impact of the screen is predominant, and it can be concluded that even in the video + Audio group, chicks primarily respond to visual cues, and the addition of auditory stimuli does not increase the impact of visual cues.

Interestingly, the response to feeding cues, demonstrated by the higher performance of feed pecking behaviour in the Video groups, was higher in younger age groups. This observation suggests that chicks might respond more effectively to feeding cues when they are younger. Our results parallel those of Richard-Yris et al. (1998), showing that the frequency of maternal food calls decreased by age of chicks. During their first week, chicks typically show a strong preference for their mother's company. Studies suggest that during this time, they quickly learn food preferences through social cues and tend to mimic their mother's diet, as observed by Wauters et al. in 2002.

Laydown

Very young chicks lack the ability to effectively regulate their body temperature. In their natural habitat, they tend to spend a significant portion of their time resting beneath their mother, benefiting from her warmth, particularly in environments with reduced light. Research indicates that chicks in the early stages of development, when being brooded, allocate approximately 60% of their time beneath the hen (Shimmura et al. 2010a). This duration decreases notably during the initial two weeks, stabilizing at around 10% by day 13, and becomes minimal by day 25, correlating with the development of sufficient feather coverage for thermoregulation (Shimmura et al., 2010a). We hypothesized that playing Video and/or Audio of a broody hen resting, would lead to more chicks exhibiting laydown behaviour in the rest area. However, in this study, although chicks displayed more laydown behaviour when resting cues were played compared to the feeding period, there was no significant difference between treatments during the resting period, contrary to our hypothesis. This result, along with the findings from the attractiveness of cues, suggests that when the screen turned on in the resting area, it attracted chicks to this location, and by displaying a broody hen resting behaviour, it prompted chicks to exhibit laydown behaviour in this area. Other studies have demonstrated that chicks are capable of replicating behaviors observed on screens. For instance, televised representations of ground and aerial predators have triggered suitable anti-predator reactions in chicks, as evidenced by research conducted by Evans et al. (1993).

In this study, the lack of a significant difference between treatments during this period could be because, in the Control and Audio groups without a screen, the resting area was warm and dark, making it an suitable location for laydown behaviour. Therefore, no significant difference was observed between treatments. It is important to note that chicks may use perches later in life for resting or sleeping. Brantsæter et al. (2016) discovered that birds given vertical space tended to use perches and raised platforms more often by the age of 19 weeks, in contrast to birds without access to such structures. It could be argued that, since there was no significant difference between these treatments, even if feeding cues were played in the resting area, chicks would still be attracted to this area because the light of the screen and video of broody hen attracts them, and the warmth of brooder in this area makes chicks laydown in this area. Hence, this laydown behaviour in Video + Audio and Video group during resting period may not be due to chicks watching broody hen resting cues. This argument should be explored further in another experiment to determine which behaviours chicks will exhibit if the video of a broody hen eating and feeding cues are played in the resting area.

In addition, when there was not any screen to show feeding cues in feed area, and during the time that there were not any cues to play (nothing period), more laydown behaviour was observed compared to feeding period that Video of broody hen was presented. This could be attributed to the absence of disturbances like the video of broody hen eating in feed area allowing them to engage in undisturbed resting behaviour.

Additionally, the dark and warm environment under the brooder likely contributed to them choosing to sleep in this area during the Nothing period. Moreover, these findings indicate that when feeding cues were played, chicks were absorbed by the feeding cues and exhibited less laydown behaviour.

Although more laydown behaviour was observed on day 5 compared to days 10 and 15, chicks in the Video group on day 5 performed significantly less laydown behaviour. These findings bear resemblance to the results reported by Mascetti et al. (2004), where a gradual decline in sleeping time was noted during the initial two weeks post-hatching. However, Hess's (1959) findings indicated that peaks of sleeping were observed on Days 2, 5, 7, and 11.

In Video + Audio and Video groups with screens, chicks exhibited less laydown behaviour than the control groups. This difference could be attributed to the screen's light potentially disturbing their sleep and consequently their resting behaviour. The presence of screen light may lead chicks to engage in other behaviours around the screen in High response area in relation to resting cues, such as ground pecking, preening, pecking at the screen, and feather pecking, resulting in reduced laydown behaviour in these two groups.

Additionally, the significant difference in laydown behaviour observed in the Video group compared to the Control and Audio groups could be explained by previous findings regarding the attractiveness of cues. Results indicate that during the Nothing period, immediately following the feeding period, more chicks in the Video groups remained in the feed area, while chicks in other treatments moved to the High response area in relation to resting cues. Therefore, it can be inferred that chicks that moved to the rest area during the Nothing period were seeking a warm and dark place, prompting them to lay down. In contrast, chicks in the Video group, which remained in the high response area in relation to feeding cues, could be the reason for the fewer number of chicks in the Video group exhibiting laydown behaviour.

Dust bathing

The dust bathing behaviour in young birds undergoes a developmental process, and around 10 to 12 days of age, it becomes more consistent and follows a fixed sequence. As the birds grow, they establish a regular pattern in their dust bathing behaviour (Kruijt, 1964). In this study the highest number of dust bathing was observed in day 10 and 15. Therefore, the reason for less dust bathing behaviour in 5 days old could be because their dust bathing behaviour did not develop enough to establish a regular pattern.

Research comparing non-brooded and brooded chicks, indicate that brooded chicks exhibit more dust bathing activity in comparison to non-brooded chicks (Riber et al. 2007; Shimmura et al. 2010). However, we did not find any difference between treatments in performing dust bathing.

Feather pecking

In other studies, it is suggested that the presence of a broody hen during the early stages of life might act as a preventive measure against the development of feather pecking. According to these studies, the mother hen's presence is believed to guide the chicks, directing their pecks toward more appropriate stimuli, such as the ground or litter, thus potentially preventing the onset of feather pecking behaviour (Rodenburg et al. 2008; Riber et al. 2007). We predicted that visual and auditory cues of a broody hen during feeding and resting may decrease the incidence of feather pecking by synchronizing their behaviour, encouraging chicks to perform feeding behaviour together in response to feeding cues and rest at the same time in response to resting cues. Although the Video + Audio group had significantly less feather pecking compared to control group on day 10. There was no significant difference between other treatments in day 10. In addition, there was not any significant difference between other treatments on days 5 and 15.

The higher incidence of feather pecking on days 10 and 15 compared to day 5 could be explained by the increased occurrence of dust bathing during these days. Studies show that the frequency of feather pecking rises when birds engage in dust bathing (Vestergaard & Lisborg, 1993). When chicks start dust bathing, they peck and scratch the dust bathing spot, then sit there, gathering loose substrate particles around their bodies. While seated, the birds flap their wings, causing the particles to lift into the air and settle on their feathers (Costa et al., 2012). These particles on the feathers motivate other chicks to peck at them, which is sometimes associated with pecking on feathers and skin as well. In this study, pecking at particles on feathers is also scored as feather pecking. Therefore, the number of observed feathers pecking behaviours increased with the increasing exhibition of dust bathing behaviour. However, it's important to note that most of the pecks toward feathers and particles on feathers during dust bathing or afterward are not painful or aggressive. In this study, it is observed that when a chick pecks aggressively toward another chick, causing pain, the recipient chick typically leaves the area. In contrast, when more than two chicks peck at particles on the feathers of a chick that is dust bathing, that chick often continues dust bathing or sitting, rarely leaving the spot.

Ground pecking

Comparative studies between brooded and non-brooded chicks demonstrated that brooded chicks engaging in more floor pecking in comparison to non-brooded chicks (Riber et al., 2007; Shimmura et al., 2010). However, in this study we did not find any significant difference between treatments.

Feather pecking led to harm on feather and skin. However, when chicks peck the ground or substrate as part of exploration behaviour, it doesn't result in damage to feathers and skin (Xu et al., 2022). There is an inverse relationship between feather pecking and ground pecking, indicating that an increase in feather pecking is associated with a decrease in ground pecking, and vice versa (Blokhuis & Arkes, 1984; Blokhuis & Van Der Haar, 1989; Huber-Eicher & Audige, 1999). However, our results did not show any negative correlation between feather pecking and ground pecking. In this study both feather pecking and ground pecking increased by age of chicks.

Locomotion activity

Other experiments that compared brooded and non-brooded chicks have revealed that brooded chicks exhibit a higher level of activity compared to chicks raised without a broody hen (Riber et al. 2007; Shimmura et al. 2010).

In this study we found out, chicks in the Video group exhibited significantly more locomotion activity compared to the Video + Audio, Audio and Control groups during the feed period. However, there was no significant difference between the Control, Video + Audio, and Audio groups during this period. The higher number of locomotion activities in the Video group could be explained by our results from the attractiveness of cues, indicating that when feeding cues were played, a significantly higher number of chicks were found in the High response area in relation to feeding cues in the Video + Audio and Video groups. However, the number of locomotion activities was significantly higher in the Video group. The difference in locomotion activity between the Video and Video + Audio groups could be because chicks in the Video and Audio group responded to feeding cues faster. By playing feeding cues, they ran immediately to the High response area in relation to feeding cues. Therefore, at the time of scoring their behaviour, they were engaged in other activities in the feed area, including feed pecking, standstill ground pecking, or other behaviours. In contrast, the Video group may have had a slower response to the feeding cues, and at the time of scoring their behaviour, they were walking or running toward the High response area in relation to feeding cues. Consequently, the behaviour recorded for them was locomotion activity. Another reason for this difference could be that chicks in the Video group had more locomotion activity in the High response area in relation to feeding cues. This high locomotion activity could be due to the absence of auditory stimuli, and they were attracted to this area by the visual cues of a broody hen. They may have been confused about what they should do after going to this area and were walking around that area. Another study could compare the speed of their response to feeding cues to further explore this aspect.

We also did not find any significant difference in performing locomotion activity between treatments during the rest period. However, it is notable that all the treatments had significantly more locomotion activity during the resting period compared to the feeding period, except for the Video group, where there was not any significant difference between the Video group during the rest period and feed period.

Pecking at pen fixtures

In this study, we recorded the number of pecks that chicks directed toward the screen and speakers, as well as pecks toward pen fixtures. Our hypothesis was that chicks would respond to feeding or resting cues by pecking at the screen and speakers. However, since the Control group did not have screens or speakers, we were unable to compare the pecking behaviour in this group. To address this, we combined the data for pecking at screens or speakers with pecking at pen fixtures. We expected that if chicks responded to feeding or resting cues, treatments with these cues would exhibit more pecking at pen fixtures than the Control group. Interestingly, our results indicate that the Video + Audio and Video groups had significantly more pecking at pen fixtures than the Control group. This finding suggests that chicks recognized the cues, and the higher number of pecks at pen fixtures could be a result of increased pecking at screens and speakers. However, there was no significant difference between the Audio and Control groups.

Preening

In this study, there was an increase in the exhibition of preening behaviour with the age of the chicks. This could be attributed to the development of their feathers, leading to a higher frequency of preening, which is considered a comfort behaviour. Additionally, previous investigations have indicated that birds subjected to feather pecking tend to engage more frequently in preening activities (Keeling, 1995; Savory & Griffiths, 1997). In our study, we observed more feather pecking on days 10 and 15 compared to day 5, which could contribute to the increased exhibition of preening behaviour.

Stand Still

Shimmura et al. (2010) observed that in open field tests, non-brooded chicks at 4 weeks old showed a reduced duration of walking and an increased period of freezing compared to their brooded counterparts. Similarly, Rodenburg et al. (2009) reported increased activity during open-field examinations in five six-week-old chicks raised with a broody hen, suggesting a diminished level of fearfulness. However, in this study, the highest number of standstill behaviours was observed in the Video group on day 5, significantly higher than in other treatments. There was no significant difference between treatments on days 10 and 15.

On day 5, the increased standstill behaviour in the Video group could be due to chicks being drawn to resources when cues were played, and some chicks stood in front of the screen and watched the video of the broody hen. Another reason could be that, after chicks were attracted to the specific location based on the cues that were playing, they stood next to the screen and slept in a standing position as the screen emitted warmth. Despite the Video + Audio group also having screens, a significantly lower number of standstill behaviours were observed in this group. This difference could be attributed to the auditory stimuli, but further research is needed to investigate the specific impact of auditory stimuli.

Using perch

Riber et al. (2007b) suggested that an increased proportion of chicks utilizing perches could contribute to a reduction in feather pecking among brooded chicks. However, in this study, we did not find any significant difference between treatments in the utilization of perches.

5.3 Potential of use

The findings of this study not only carry practical implications for optimizing rearing conditions and improving chick welfare in commercial poultry settings but also hold broader significance for the understanding of animal behaviour. There is a possibility of using these visual and auditory stimuli in a commercial setting to make resource finding easier for chicks, synchronize their behaviour, and decrease feather pecking by separating the active and inactive phases during the rearing period. Consequently, we can improve the welfare of chicks during the rearing period, which has a lasting impact throughout the rest of their lives, potentially reducing feather pecking during the laying period as well.

The insights gained from analyzing chicks' responses to visual and auditory cues contribute valuable knowledge to the broader field of animal behaviour studies. This result can increase our knowledge regarding how chicks receive the video and audio cues and how they respond to them. Also, the strength of their response is compared in different treatments in this study, which could help researchers understand how chicks can receive these cues better and manipulate the intensity of their response in different environments. In this study, it is shown that chicks respond more strongly to visual and auditory stimuli.

This approach could be applied to a range of species, enhancing our comprehension of how animals, particularly poultry, perceive and respond to visual and auditory cues.

This study can also be a step toward using other maternal care behaviours of a broody hen and simulating them to improve the welfare of chicks. It can also be useful to use visual and auditory stimuli of other animals in commercial settings or for research purposes, not only to attract them toward resources but also to investigate more about their visual and auditory systems.

Future studies can investigate the effect of the feeding and resting cues of a broody hen in a commercial setting. It could also be possible to use visual and auditory cues for pets and attract them toward resources, especially for animals that have disabilities and have problems finding resources.

5.4 Limitations and room for improvement

One notable limitation of this study was related to the program designed for cue presentation, following the specified schedule. The program faced challenges in adhering to the schedule, especially in sections where screens were utilized for visual cues. This created difficulties in identifying parts suitable for analysis, demanding considerable time and effort. Additionally, the Observer XT program, configured for behaviour scoring at 2-minute intervals, encountered heightened complexity in segments with screen-related issues. To address this, behaviours in these segments were manually recorded and subsequently transferred to the Observer XT. In future studies, it is important to develop an improved program that smoothly aligns with the project schedule for cue presentation.
Moreover, the Observer XT and Nodules, designed primarily for scoring behaviours in laboratory animals like mice and rats, proved more suited for individual or smaller groups. However, applying these programs to score the behaviour of a group of chicks posed challenges. Future research would greatly benefit from creating a program specifically tailored for avian species, potentially minimizing errors compared to human observation.

Although chicks couldn't hear audio cues from other pens, they were sensitive to sounds from neighbouring pens, occasionally leading to chicks congregating in corners where these sounds were audible. Allocating a separate room for each pen, with an increased budget and space, could mitigate this issue.

Another path for potential improvement involves considering the arrangement of feeding and resting areas facing each other along the width of the pens. In future studies, aligning these areas in front of each other could simplify data analysis and facilitate comparisons of chick approach behaviours by ensuring a similar and equal number of squares for each area. It also makes it possible to define these areas for the software and score the number of birds that are entering this area.

For upcoming studies, employing coloured markings on chicks could significantly enhance the efficiency of behaviour scoring. This approach not only simplifies individual chick behaviour assessment but also opens up possibilities for using software capable of automatically recognizing and scoring behaviours based on colour markers.

Another limitation of this study was the method used to number each square in the area of the pen representing the location of each chick. A transparent paper sheet was employed on the screen, requiring adjustments to fit the entire pen area. Although the area was consistent across all pens, the adjustment process was time-consuming. Furthermore, due to a slight curve captured by the camera video at the top and bottom of the pen, adjustments became challenging. For future studies, it is advisable to implement a pre-experiment method for numbering each part or use software that displays these squares on the pen area and assigns numbers automatically.

5.5 Social and Ethical Aspects

The practice of rearing pullets without a broody hen raises pertinent social and ethical considerations. Firstly, it diverges from the natural behavior of chickens, which typically involves maternal care and guidance from a broody hen. This departure may impact the well-being of the chicks, potentially affecting their ability to navigate their environment and develop adaptive behaviors. Moreover, the absence of maternal interaction could lead to social deprivation among the chicks, as they may lack opportunities for social learning and bonding typical in a broody hen-reared setting.

In the realm of social and ethical considerations, the phenomenon of redirected foraging behavior among chicks reared without a broody hen holds significant implications. The absence of guidance from an experienced mother may result in chicks exhibiting behaviors that deviate from their natural foraging instincts, leading to instances of redirected pecking towards the feathers and skin of conspecifics. This behavior not only disrupts the natural social dynamics within the flock but also poses ethical concerns regarding animal welfare and well-being.

Ethically, redirected foraging behavior raises concerns about the welfare of both the chicks engaging in the behavior and the recipients of the pecking. The act of pecking towards feathers and skin can cause physical harm, injury, and distress to the targeted individuals, compromising their well-being. Additionally, this behavior may contribute to feather pecking, which is a significant problem within poultry farming, further highlighting the ethical imperative to address this issue.

The experiment undertaken seeks to address these concerns by exploring alternative methods to provide visual and auditory cues mimicking a broody hen's presence. Ethical considerations involve ensuring that these methods effectively fulfill the chicks' behavioral and developmental needs, mitigating any potential negative impacts associated with maternal deprivation.

5.6 Sustainability Aspects:

In commercial poultry farming, the traditional method of rearing layer-type chicks without a mother hen is driven by economic constraints and logistical challenges. However, this approach often leads to welfare issues such as chicks disturbing each other and developing redirected pecking behaviors. To address these challenges, this experiment investigated the feasibility of using Video and Audio of a broody hen to attract chicks toward feeding and resting areas, potentially offering a sustainable solution for commercial farms.

The integration of video and audio cues, specifically replicating a broody hen's presence, into commercial settings could revolutionize chick rearing practices. By using the nurturing behaviors of a mother hen, these cues could effectively guide chicks to feeding and resting areas, creating a more natural environment conducive to healthy development. This enhancement of chick welfare not only promotes sustainable farming practices but also optimizes resource utilization, contributing to overall sustainability in poultry production.

Moreover, by promoting natural behaviors and reducing stress, this technology aligns with the principles of organic farming, offering a sustainable solution that enhances both animal welfare and farm productivity.

Additionally, this method opens avenues for utilizing other behaviors of a broody hen to further improve chick rearing practices. By incorporating additional cues, such as those related to protection, socialization, and exploration, farms can create an enriched environment that fosters the development of natural behaviors and reduces stress among chicks. This holistic approach to chick rearing not only enhances animal welfare but also contributes to the long-term sustainability of poultry farming operations.

One notable aspect of reduced redirected foraging behavior is the potential decrease in feather pecking among the flock. When chicks exhibit less feather pecking behavior, there are fewer instances of feather damage and loss among hens. This reduction in feather pecking can have a positive impact on the energy consumption of the farm. Hens with intact feathers are better equipped to regulate their body temperature, requiring less energy to maintain warmth compared to those with damaged or missing feathers. Consequently, farms may experience decreased energy demands for heating facilities, leading to lower energy consumption and associated costs.

By minimizing feather pecking through the implementation of strategies like playing Video and Audio of a broody hen, farms can create a more energy-efficient environment while simultaneously promoting the welfare of their flock. This dual benefit underscores the sustainability of such practices, as they contribute to both environmental conservation and economic efficiency within the poultry production system.

5.7 Ethical Aspects of Research and Development

Ethical considerations were paramount in guiding the experiment investigating the use of Video and Audio of a broody hen to enhance chick rearing practices. The welfare of the chicks was prioritized, with measures taken to ensure their well-being throughout the experiment. The chicks were carefully monitored for any signs of distress, and procedures were adjusted accordingly. Transparency and accountability were maintained, with clear communication of the experiment's aims and potential implications. Overall, ethical principles guided the research, ensuring that scientific advances were made responsibly and with respect for animal welfare.

6. Conclusion

The principal aim of this study was to assess various methods for presenting feeding and resting cues resembling those of a broody hen to layer-type chicks, with the objective of identifying the most effective approach for attracting chicks to specific resources and encouraging simultaneous resource use.

The results support the first hypothesis, indicating that both the Video and Video + Audio groups successfully attracted more chicks to resources, while the Audio group showed a similar response to the Control group. In addition, the results strongly align with our second hypothesis, as chicks in both the Video and Video + Audio groups demonstrated an increase in feed pecking behavior during feeding period compared to periods with no cues or resting cues, at both 5 and 10 days old. This study also revealed that during the nothing period (except for the Video group), the resting period, and in treatments without any screens, chicks displayed significantly more laydown behavior ($P \leq 0.05$). In addition, Chicks in the Video + Audio and Video groups demonstrated a notably higher level of synchronized behavior, being more collectively attracted to the desired areas and displaying increased utilization of resource together. This synchronized behavior supports the third hypothesis. Furthermore, the absence of any significant difference in body weight between treatments provides support for our fourth hypothesis.

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Popular science summary

In natural environment, newly hatched chicks receive vital lessons from their broody hens during their initial weeks of life. The hen shares knowledge on successful foraging and provides warmth and safety. She communicates with her chicks through various auditory and visual characteristics. For example, when a hen finds food, she makes a special sound and pecks at food. This not only calls the chicks and attracts them to that area but also encourages them to eat, teaching them how to find food and what's good to eat. Also, when it is time to rest, she calls her chicks by making a kind of sounds that attract chicks to rest beneath her, and this sound means a warm and safe place for them to sleep. Furthermore, a broody hen synchronizes the behaviour of chicks in a way they do the behaviours together, for instance, feed together and rest together.

However, in commercial setups, chicks are raised without a mother hen to teach them about finding food and what is edible or not. Also, there is not a broody hen that chicks go beneath her to sleep in a warm and dark environment. Therefore, in the same place where they feed, some chicks are resting, and some are eating. This leads to chicks pecking at everything that attracts them during exploring their environment like feathers, toes, or skin of other chicks. Moreover, the chicks that want to rest are disturbed by chicks that are exploring and eating around them when they peck on their feathers or skin. This redirected pecking at the feathers of other chicks and causes chicks to get used to doing that and continue that in later life, which is called feather pecking that is one of the serious problems of a commercial setting. It not only has an economical problem but also is a serious welfare problem.

To address these challenges, in this study, video of a broody hen and her sound during feeding when she calls her chicks and during resting with resting calls is recorded. The aim of this study was to investigate which characteristics of the broody hen are more effective in attracting the chicks to resources and encouraging them to use resources together. Results indicated that the Video+Audio and Video of the broody hen were most effective in attracting chicks to feed areas and rest areas and encouraging feed pecking. However, there was no significant difference in inducing laydown behaviour in the rest area among treatments.

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