

To be a chick mother

–Variation in maternal care and its effect on chick behavioural development

Att vara en hönsmamma - variationen i moderhönans omvårdnad och dess påverkan på kycklingens beteendeutveckling

Linnéa H Christenson

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Linnéa H Christenson

Supervisor: Carlos E. Hernandez, SLU, Department of HUV **Supervisor:** Jenny Yngvesson, SLU, Department of HMH **Examiner:** Harry Blokhuis, SLU, Department of HMH

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Faculty of Veterinary Medicine and Animal Sciences Department of Animal Nutrition and Management

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Abstract

In Swedish poultry production today over 7 million chicks are incubated, housed and reared without maternal care. Studies indicate that chicks behavioural development and welfare may benefit from hen maternal care and potentially maternal care could be artificially provided in the commercial situation. However, hen maternal abilities vary, which affect chick behavioural development. The aim of this study was to investigate maternal variation (social status and type of broodiness) in the maternal care between different hens (H) and how this variation affect the behavioural development of the chicks (C). The hens' social status (dominant or submissive) in the adult group affect the learning ability in other adults, the hens type of broodiness (induced broody or natural broody) affect when the maternal hormones in the hens body are released, both of which might affect the chicks behavioural development.

Old Swedish bantam hens and Bovans Robust chicks were used in this trial. Hen social status (dominant D or submissive S) in the adult group was determined with a social interaction test. Family groups were formed by putting six chicks with a hen. Control groups were six chicks without a hen. Chicks were brooded either under naturally broody hens (natural broody NB) or in an incubator and later put with the hen (induced broody IB). Behavioural testing of the chicks was made on day four, five, 10, 18 and 26 and feather scoring was done at day 27 post hatch. The chicks wound score was analyzed in the end of each group's testing period.

In total seven family groups and three control groups were recorded and analyzed in this trial. Four hens were defined as dominant, three as submissive, four as natural broody and three as induced broody. Hens differed significantly in purring and chick latency to approach. The chick group spread varies with the chicks ability to thermoregulate, close at first and further away for older chicks, unexpectedly the dominant hen's chicks keeps the same distance the entire time. Foraging behaviour did not differ, however on recording day 10 and 18 feather pecking behaviour differed between the family groups and control groups and between the natural broody and the induced broody groups.

Hen individual maternal behaviour showed large variation e.g. showing as different feather and wound scores. Based on the chicks feather and wound plumage the best family groups where ID3(S and NB) and ID7(D and IB) and the worst was ID6 (D and IB). Based on this study it is important to choose the best individual mother hens for the chicks. However, further research is necessary to define which mother hen characteristics that benefit chick behavioural development.

As a general conclusion, the control chicks in this trial showed less variation compared to chicks reared with a mother hen. In conclusion, during rearing the individual hen can favor while another can disfavor chick behavioural development, compared to chicks reared without a mother hen.

Keywords: characteristic, fearful, feather peck, inter-individual distance, purring

Sammanfattning

De 7 miljoner kycklingar som föds upp i den Svenska värphönsproduktion har ingen kontakt med någon modersgestalt. Flertalet studier påvisar att kycklingarnas beteendeutveckling gynnas av att ha en hönsmamma närvarande under uppväxten, beteenden såsom distansering, födosök, och fjäderplockning. Hönans individualitet påverkar både modersegenskaper, kommunikationen och kycklingens utveckling. Syftet med den här studien var att undersöka den individuella variationen hos hönsmammor och hur variationen påverkade kycklingarnas beteendeutveckling.

Tidigare studier visar att egenskaper såsom hönsmammans sociala status (dominant D eller undergiven S) och typen av ruvning (naturlig NB eller inducerad IB) kan påverkar kycklingens utveckling. Inlärningen bland andra vuxna värphöns påverkas av den sociala statusen och typen av ruvning påverkar hönsmammans hormonutsöndring, vilket påverkar moders egenskaperna.

Gammal Svensk dvärghöna och ägg/kycklingar från rasen Bovans Robust användes i detta försök. Hönans sociala status bestämdes genom ett socialt samspelstest. En familjegrupp (H) i den här studien innehöll sex kycklingar och en höna. Kontrollgruppen (C) innehöll bara sex kycklingar. Äggen kläcktes under hönan för NB och i inkubator för IB och C. Beteende test på kycklingarna utfördes dag fyra, fem, 10, 18 och 26, fjäderdräktspoäng sattes dag 27 och sårpoäng sattes i slutet för respektive grupp.

I studien fanns tre C och sju H, hönsmammorna delades in som fyra D, tre S, fyra NB och tre IB. Signifikanta resultat påvisades för hönans purrande och kycklingens snabbhet att närma sig. Kycklingarnas distansering påvisade ett oväntat mönster för gruppen med D hönor och gruppen med IB hönor. Inga signifikanta resultat i födosök hittades. Signifikanta resultat påvisades mellan H och C samt inom gruppen NB och IB för fjäderplockning och fjäderdräktspoäng.

Hönans egenskaper och individualitet har stor påverkan på kycklingens utveckling. De bästa familjegrupperna var med ID3 (S och NB) och ID7 (D och IB) baserat på fjäderplockning och fjäderdräktspoäng. Den sämsta familjegruppen som var med hela studien var ID6 (D och IB) baserat på fjäderplock och fjäderdräktspoäng. Det är alltså extra viktigt att välja rätt individ för kycklingarna. Det behövs mer forskning och i större skala för att få fram vilken typ av egenskaper som är fördelaktiga för kycklingarnas utveckling.

Den generella slutsatsen är att kontrollgruppen i denna studie var jämnare som grupp jämfört med kycklingarna i de olika hönsgrupperna. En hönsmamma kan alltså gynna och en annan hönsmamma missgynna kycklingarnas beteende utveckling jämfört med kycklingarna som uppfostras ensamma.

Nyckelord: hönsmamma, höna, moders gestalt, karaktärsdrag, fjäderplock, närhet till gruppen, purrande

Preface

Information for this master project was collected in summer 2018 at the Swedish university of Agriculture in Uppsala.

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Concept explanation

Behavioural display	A combination of vocalization and body language
Control group	One control group containing six chicks
Demonstrator	One bird demonstrates a certain behaviour, and other bird is mimicking.
Dominance (D) in so-	The hens, which received a score higher than 0 in the social status test based on social
cial status	interactions.
Family group	One family group containing one hen and six chicks
Group spread	A measurement of dispersal of chicks from the hen or how closely together the chicks
	in a group are.
Induced broody (IB)	Hens which received day old chicks during the night, the chicks were hatched in an
	incubator.
Natural broody (NB)	Hen that brooded eggs for at least 24h pre hatch.
Observer	A bird which observe a certain behaviour of the demonstrator, and mimic that behav-
	iour.
Rejected chick	Hen rejects chick during induced broodiness, she walks away fast, trying to get away
	from the chick or remove the chick from underneath her.
Social status	Hierarchy within the adult group
Submissive (S) in so-	The hens which received a score lower than 0 in the social status test based on social
cial status	interactions
Thermoregulation	The ability to maintain a stable body temperature
Tidbitting	The hen's action of picking up and dropping feed of interest with combination of vo-
	calization and/or scratching to encourage chicks to eat.

1 Literature review

1.1 General information

In Swedish poultry production today over 7 million chicks are incubated, housed and reared without maternal care (Sveriges officiella statistik; 2018 JO20SM1801). The main reason for the exclusion of the mother's presence, is the risk of diseases spreading across generations (Liljeholm 2017). Biologically the chick is precocial and has the ability to see, hear, walk and eat from the moment they hatch (Appleby *et al.*, 2004). The domesticated chick and hen (Gallus gallus domesticus) have similar needs and behaviours as their wild relatives, the red jungle fowl (Gallus gallus) (Appleby *et al.* 2004; Jensen 2006). In commercial production sufficiently warm temperature, isolation from predators and accessible feed and water is sufficient for chick survival, but the absence of a mother hen may have large detrimental effects on the behavioural development.

The first week in life is the most crucial for the chicks survival and behavioural development (Brown 1964; Nicol 2006; Shimmura *et al.* 2015). In the wild the chicks survival depends on the mother hen's guidance in feed search (Hogan, 1994 in Riber *et al.* 2007), her predator warnings (Appleby *et al.* 2004; Jensen 2006) and her thermoregulation (Appleby *et al.*, 2004). The hen stays with the chicks in isolation (Appleby *et al.*, 2004; Jensen 2006; Haas *et al.* 2014b) during the first 8 weeks post hatch (Jensen 2006; Haas *et al.* 2014). The isolation with the mother has a large impact on the chicks future behavioural development (Field *et al.* 2007; Rodenburg *et al.* 2008) and environmental adaption (Field *et al.* 2007). Studies indicate that the chicks behavioural development (Roden & Wechsler 1998) in communication (Wauters *et al.* 2002a), group spread, foraging (Blokhuis 1986; Riber *et al.* 2007) and pecking (Vestergaard *et al.* 1993; Rodenburg *et al.* 2009a; Shimmura *et al.* 2014).

Furthermore, there are individual variations in the frequency and magnitude of maternal care among hens (Pittet *et al.* 2013, 2014; Edgar *et al.* 2015; Zidar *et al.* 2017), which affect the chicks behavioural development (Pittet *et al.* 2014).

Perré *et al.*, (2002) discovered that the chicks' own social status was more pronounced, if the chick was raised with a mother hen compared if it was raised without a mother. The hens' individuality is correlated to the hens social status (Pittet *et al.*, 2014). The social status affects transmission of behaviours because it influences if a hen is seen as a demonstrator for other adults. A hen observing a dominant (D) demonstrator hen, performed a more distinct feeding, compared to the observer with a submissive (S) demonstrator (Nicol & Pope 1999; Croney *et al.* 2007).

A common trait showing individual variation in hens is broodiness. The main part of the scientific studies use induced brooders (IB) as mothers for the chicks' (Wauters et al. 2002b; Pittet et al. 2013; Shimmura et al. 2015), while others investigate the maternal care of natural brooders (NB) (Tuculescu & Griswold 1983) and some investigated both conditions for example Noel et al. (2012). The induction process starts with feed depriving the hen, to make stop her egg laying (Richard-Yris et al. 1987). After feed deprivation the chicks are introduced to the non-laying hens during night, which is the least harmful way for the day old chick (Richard-Yris et al. 1983, 1987). It is important that audio recognition occur before the visual introduction. If the hen hears the chicks before she sees them, her aggressiveness towards the chick will be decreased (Richard-Yris & Leboucher, 1987). However, there are individual hormonal differences between the induced broody hens and the natural broody hens (Richard-Yris et al. 1987). The natural broody hens have significantly higher levels of prolactin and lower levels of testosterone compared to the induced broody hens (Richard-Yris et al. 1987). Naturally broody hens preform more maternal care behaviours during the first days post hatch, compared to induced broody hens (Richard-Yris et al. 1987).

1.2 Behavioural interactions

Communication and the latency for chicks to approach the hen

Chick communication starts pre-hatch, the chick communicates with the eggs closest and develops a bond to those chicks. After the first week the chicks start to mimic each other's behaviour (Jensen 2006).

A newly hatched chick imprint on the first thing they see, this can be any hensimilar object or any hen, which is why family isolation is so important (Appleby *et al.* 2004; Trillmich & Rehling 2006). As early as five minutes post hatch a separated chick emit distress calls and search for the imprinted mother (Kent 1987). The special bond created by imprinting enables communication recognition (Wauters *et al.*, 2002a) and hasten chick behavioural development (Appelby et al, 2004). Both vocalization and body language is used by the hen to influence her chick (Wauters *et al.*, 2002a).

Vocalization is mainly used to communicate predator warnings, feed calls and follow calls (Appelby et al, 2004; Jensen, 2006;). The first seven days the chicks' response is slow but as the chicks grow older their response speed will be faster (Fält 1981; Wauters & Richard-Yris 2002). During the chicks' life the mother hen's vocalization changes (Wauters *et al.* 2002b) as well as the chicks response to the vocalization. Each hen's vocalization is different (Fält 1981), in frequency and duration (Wauters *et al.* 2002b). Chicks prefer their own mothers vocalization compared to other hens' vocalization (Fält 1981).

Body language is used by the hen to amplify a behaviour or to demonstrate a particular interesting feed (Wauters & Richard-Yris, 2002; Gajdon *et al.*, 2015). For example tidbitting display, where the hen picks up and drop feed of interest after attracting the chicks attention with a feed call (Evans & Evans, 1999; Marino, 2017). The hen will continue with the tidbitting display until the chick approaches the hen and mimic her (Wauters & Richard-Yris 2002). The intensity of the hens' behavioural display can be adjusted due to the needs, reactions and physical placement of the individual chick (Nicol & Pope, 1996; Trillmich & Rehling, 2006).

The hens' impression and experiences of communication in early life affects her interaction as an adult (Zidar *et al.* 2017).

Chicks reared by induced broody hens have a lower recognition of their mother due to the lack of communication pre-hatch (Fält 1981). According to Richard-Yris *et al.*, (1987) the natural broody hens express more cluck sound compared to the induced broody hens. According to Trillmich & Rehling, (2006) brooding is important for communication, for example chicks beg to be heated (to thermoregulate) under the hen. The communication difference, between chicks reared with the natural broody hen and chicks reared with the induced broody hen, will even out when the chicks have learned their mothers individual vocalization (Fält 1981).

Group spread

The physical distance within the group the first days is affected by thermoregulation. Thermoregulation is the individual's ability to maintain a stable body temperature. In the wild, the brooding hen provides the required heat for the chicks (Buntin 2010). When the hen is brooding, she stays put or moves around in a crouching position with the wings slightly lifted (Hess et al., 1976 in Chaiseha & El Halawani 2015) while the chick stays under or as close to the hen as possible (Fig. 1). The

first four days post hatch the chicks have almost no ability to thermoregulate themselves (Fält, 1981; Jensen, 2006; Haas et al., 2014b) and require an external heating source. The first seven days the chicks stay close to the mother hen (Fält, 1981). However, external heat is required for the first four weeks post hatch and the chicks have to stay somewhat close to the heating source this time (Shimmura *et al.*, 2010). In poultry production, a heating lamp can be used for chicks raised without a mother hen (Liljeholm, 2017). Chicks raised without a mother hen are less active in the end of the thermoregulating period compared to a chick raised with a mother hen (Shimmura *et al.* 2010).

When the chicks thermoregulating under a moving heat source (their mother) in contrast to a heating lamp (not moving), they become more physically active. Even later when thermoregulating is not as important, chicks stay close to the mother (Roden & Wechsler 1998). There is individual difference between the different hens of how much the mother hen encourage the chicks to explore the surroundings (Riber *et al.*, 2007) and investigate new objects (McBride *et al.* 1969). Chicks raised



with a hen spent more time moving and less time warming (Wauters *et al.* 2002b). A mother hen which investigate more, has more physical active chicks which explore more of their surroundings (Shimmura *et al.* 2015).

Figure 1. Photo screenshoot camera by Linnéa Christenson. This hen is brooding her chicks (thermoregulating).

A closer physical distance between the mother hen and chicks indicate a lower level of aggression in the hen (Pittet *et al.*, 2014), and a more submissive social status in the hen (Rushen 1982). The older hen spend more time thermoregulating (keeps a closer physical distance to) their chickens compared to a younger hen during the first week post hatch (Pittet *et al.* 2012).

The natural broody hens have a higher level of contact with their chicks than induced broody hens (Richard-Yris *et al.* 1987)

Foraging

Chicks' raised without a hen as a heat source have to walk between the feed, water and the heater (Shimmura *et al.*, 2015). The chick can live on their yolk for the first three days post hatch (Entenman *et al.* 1940). However, according to Kornasio *et al.*, (2011) foraging should occur within the first 6h. Chicks under a heater will not feed as often as a chick with a mother hen (Shimmura *et al.* 2015). Ground foraging is expressed less by chicks raised without a mother hen (Shimmura *et al.* 2010). Chicks raised with a mother hen foraged at similar feed, as the mother hen demonstrates, compared to the chicks raised without a mother hen which can foraged more non-eatable materials (Wauters *et al.* 2002b). The mother hen teaches the chicks about visual food particles and where to find them (Gajdon *et al.* 2015) and chicks raised with a mother hen pecked more at the feed container (Wauters *et al.* 2002b). The mother hen can redirect the chick attention to specific feed particles (Nicol 2006) and during the first days she can even relocate her chicks (due to thermoregulation) close to attractive feed and water (Shimmura *et al.*, 2015). Each hen has their own preference for what is considered as attractive feed (Newberry *et al.* 2007), which is correlated to each hen's past experience (Zidar *et al.* 2017).

Natural broody hens are more motivated to redirect and teach their chicks the first days post hatch, than the induced hens (Richard-Yris *et al.* 1987).

Feather pecking

Feather pecking is a severe abnormal behaviour in commercial hens causing animal welfare problems and economic loss. It has been found to be caused by re-directed foraging behaviour (Blokhuis 1986). Chicks reared without a mother hen will start to peck at everything to investigate if it is edible (Gajdon *et al.* 2015). According to Shimmura *et al.*, (2010) chicks without a mother hen spend more time with gentle feather pecking as well as aggressive feather pecking, fighting and threatening compared to chicks raised with a mother hen.

Chicks reared with a mother hen early in life are less prone to develop feather pecking compared to chicks reared as a control group (Rodenburg *et al.* 2009a; Shimmura *et al.* 2015). The mother hen canalize her own pecking preference to her chicks (Gajdon *et al.* 2015) and redirect the chicks' attention away from non-advantage items such as feathers (Nicol 2006). However, neither Roden & Wechsler (1998) nor Riber *et al.*, (2007) found that the mother hen affected the feather pecking behaviour in the chickens, indicating that mother hens may differ in the effect they have on the development of feather pecking in their chicks.

Chicks reared with natural broody hens peck less than induced broody hens (Richard-Yris *et al.* 1987) and therefore it is important to investigate the effect of hen broodiness on chick feather pecking behaviour.

Aim and hypothesis

The aim of this study is to investigate the individual variation in maternal care between different hens and how this variation affects the behavioural development of the chicks. Areas of interest include mother hen communication, chicks latency to approach their mother when called upon, groups spread in each family group, the chicks' foraging behaviour, the chicks' feather pecking behaviour and the chicks' plumage and wound score at the end of the trial. The maternal variation used was social status (dominant or submissive) in the adult group and type of broodiness (natural or induced).

Hypothesis 1 (communication)

Mother hens with the social status dominant vocalize less compared to mother hens with the social status submissive. Mother hens that were natural broody hens vocalize less compared to the induced hen.

Hypothesis 2 (group spread)

The chicks group spread, or physical distance will be less if reared with a mother hen than as a control group. Chicks reared with a dominant mother hen will spread more/further away compared to chicks raised by a submissive mother hen. Chicks reared with a natural broody mother hen keep a closer distance, compared to chicks raised by an induced broody mother hen.

Hypothesis 3 (foraging)

Chicks reared with a mother hen spend more time foraging compared to the control group. The same was predicted (more time foraging) for chicks reared with a dominant mother hen compared to chicks raised with a submissive.

The natural broody hens' chicks spend more time foraging, compared to chicks raised by an induced broody mother hen.

Hypothesis 4 (feather pecking):

Less feather pecking will be performed by chicks reared with a mother hen compared to the control group. Chicks reared with a dominant mother perform less feather pecking compared to chicks raised by a submissive mother hen. Less feather pecking is performed by natural broody hens' chicks and they also have better feather condition score compared to chicks raised by an induced broody mother hen.

2 Material and Method

2.1 General information

All procedures were approved by the Regional Animal Ethics Committee in Uppsala (diary number: 5.8.18-00610/2018). 160 eggs were purchased from Swedfarm facility Linghem in JUNE. The eggs were layed by 40 weeks old Bovan Robust hens. The eggs were incubated in a J.Hemel Brutgeräte GmbH & Co incubator. The first 40 eggs were incubated day one after arrival to Lövsta (Fig. 2). A week later 60 other eggs were incubated from the same batch and two weeks after arrival the last 60 eggs were incubated. Eggs not incubated day one, were stored in a refrigerator. The eggs were manually turned twice a day by staff employees at SLU Facility Lövsta where the incubator was standing. One week after the incubation started, the



eggs were candled for live embryos. The hatching day was named as day 1 post hatch.

Figure 2. Eggs in the incubator. Photo by Carlos Hernandez July 2018.

The animals

Ten observed broody hens and one rooster of the breed "old Swedish Bantam" (Fig. 3) were purchased in April 2018 from two different herds in Stockholm's University research facility Tovetorp. The old Swedish bantam are defined as hardy, lively, good feed finders, good brooders and good mothers (Chibreew 2018). Four of the hens were four years old and six of them were one year old. The rooster was one year old. Both hens and rooster had been incubated and reared without a mother. None of the hens had previous to this study reared chicks. The hens were individually marked with a metal ring with a number and one- or two-colored rings on their legs.



Figure 3. Old Swedish Bantam hens were used as mothers. Photo by Carlos Hernandez 2018.

The chicks' beaks were intact during the entire trial and both sexes were used in this trial. Except the control groups each family group also contained a hen. If a hen had to be put down before the final test day her family was included in the analyzing until the day she was excluded.

Six chicks were assigned to each hen comprising seven family groups. The remaining three hens could for various reasons not be included in the family groups. However, the remaining 18 chicks were divided into three control groups.

Stables

This experiment was located at SLU's research facility at Lövsta during summer 2018, group pen and single pen (Fig. 4). The birds where first housed in a group pen $(0.36m^2/adult bird)$. The hens were later on moved to individual pens $(1m^2/family group)$ with a nestbox (Fig. 5) and artificial eggs. The temperature in the stable was 23°C and the humidity was 35%, additional information about the stable conditions is found in Appendix 1.



Figure 4. Individual pen for each family group or each control group. Photos by Carlos Hernandez and Linnéa Christenson.

The social status in the adult group

Observational study of the social status was done during the first five weeks after arrival (Fig. 3). Pairwise encounters were staged between the hens and scored, using continuous, direct observations. Mealworms were used as the competition resource. Food and water was available ab libitum during this trial.

The social status test was based on attack behaviour according to Zuk *et al.*, (1998). The behaviours pecking, chasing, displacement (avoidance) and fighting were recorded to determine social rank. A hen pecking at another hen's head, with the reaction of avoidance from the other hen, the pecker gained a social status score (+1) and the avoider lost a social status score (-1). If a hen avoided another hen, the avoided hen gained a social status score (+1), and the hen avoiding lost a score (-1). At least three interactions between the pairs were observed. The total number of interaction social status scores was added together and the mean value of them gave the hen a mean social status score. If the score was > 0 the hen was considered dominant (D) if the score was < 0 she was considered submissive (S).

Brodiness- natural or incubated broody

If a If a hen had been observed broody (Fig. 5) (natural broody, five hens) for two weeks she was moved to a single pen. Four of the five (one died) naturally broody hens received 10 eggs each during one night (10.00 p.m.) between day 18 and 19 of incubation. The incubated eggs were switched with the fake eggs she already laid upon. A total of six chicks per hen was the goal for the hen and if one hen had more or fewer, cross-fostering was applied.



Figure 5. A broody hen in the nestbox. Photo by Linnéa Christenson.

If the hen did not get broody within the time the eggs were incubated, she was induced to broodiness according to (Richard-Yris *et al.* 1987). However, no food or water deprivation was applied before inducing, even if the hen was still laying eggs. The induced hen was kept alone with 10 fake eggs in the nestbox one night before she was induced to broodiness. In the morning the hen could walk around in her individual pen. The second night the hen received day old chicks from the incubator.

The hen was placed inside the nestbox when she was asleep, later that night ten newly hatched chicks were placed under her. After the chicks settled and the hen did not try to pull out the chicks the nestbox door was closed. According to the instructions from Richard-Yris & Leboucher (1987) the handler remained inside the single pen for 30 minutes listening to sounds from the chicks. If any sound might have suggested that the chick was harmed, the handler removed the chicks. The following morning the handler let the hen and the chicks out when the light turned on at 4.40 a.m. The hen and her chicks were observed for one hour after the nestbox was opened, to make sure no harm was done to the chicks. If the chicks were rejected or harmed they were removed to the heating lamp area and the hen was excluded from the trial, in total two hens were excluded.

2.2 Behavioural interactions

The interactions and behaviours in the family and control groups were observed in a custom made, sound proof recording shed on days three, four, five, ten, eighteen and twenty-six post hatch. A Go-Pro Hero 4 camera (located 1.5m above the floor) was used (Fig. 6). The birds were moved from their home pens to the recording shed in a closed box (plywood and dimensions of width 25cm* height 35cm* depth 50cm), with peat as bedding. Additional information in Appendix 1. The recordings lasted for 45 minutes with only feed present. The experimental setup inside the recording shed consisted of two wire net cages. The cages, a "food cage" and a "bird cage", were intended to prevent the chicks reaching the food before the commencement of recordings. Both cages could be lifted to the top of the ceiling and out of view by means of ropes. The "food cage" was lifted first and when the hen was close to the feed the "bird cage" lifted and the chicks were let out. Detailed information in Appendix 2.



Figure 6. Screenshot from GoPro camera 2018 details in Appendix 1 and 2. Photo by Linnéa Christenson illustrating a family group in the recording shed, during communication recording.

Statistics

The different treatment groups' (dominant, submissive, natural broody or induced broody) behavioural interactions (communication, groups spread, foraging and feather pecking) mean values and standard error (std err) where compared in a two-tailed T-test. The feather score for individual chicks was tested in a mixed model with the hen as a random effect. Significance level was set to p < 0.05.

In this trial there were two fixed treatments the social status and the broodiness. The social status of the hen was dominant or submissive and the broodiness was naturally broody and induced broody, with a randomized individual difference. Three control groups were used as well. The trial had an unbalance in the different treatments. Fixed factors were used such as hen and chick breed, hen previous preference of motherhood, feed, movement, housing and observer. The eggs and chicks were somewhat randomized to the individual hen. Al the videos were analyzed by the same observer and according to a pre-made description of behaviours analyzed (Appendix 3). Behavioural interactions were analyzed with The Observer XT 14 software package (Noldus Information Technology B.V., Wageningen, The Netherlands).

Communication

The communication between hen and chick were analyzed with 40 minutes continuous recording on day four and five post hatch. Here the vocalization bout was analyzed. One vocalization bout was considered from the start of the vocal until three seconds has past, or from the start of the vocal until a vocalization change occurred.

Latency to Approach

Each day the first part of the recordings was analyzed continuously for latency to approach mother, for each chick in seconds after the cages was lifted. The chick was considered at the mother hen's side when the chick was maximum one square (11.5cm*11.5cm) away from the hen (Appendix 3). For the control groups the chicks' latency to approach the feed or the heater was analyzed.

Group spread and foraging

Day four, five, ten, eighteen and twenty-six post hatch the behavioural interactions of foraging and distance between hen and chick (Fig. 7) were analyzed with oneminute interval for the first 35 minutes of the recordings. The different levels of distance between center and each chick where "same square as center", "one square away", "two squares away" and "more than two squares away". The center was considered as the hen in the groups with a hen and as the most centered chick in the control groups. The first image illustrates five chicks at level "more than two squares away" from the hen and one chick in the "same square as center". The second image illustrates all six chicks in the "same square as center". The third image illustrates the control group distance "same as center". More information in Appendix 3.



Figure 7. Screenshot from GoPro camera 2018 by Linnéa Christenson illustrating group spread.

Feather pecking

Pecking behaviour included both feather pecking and gentle pecking at hen or chick, the behaviour was analyzed continuously on day four and five. On day 10, 18 and 26 post hatch the pecking behaviour was analyzed with one-minute interval for the first 35 minutes of the recordings.

If a hen was discovered aggressive or a chick was discovered wounded or bloody the hen and the chick was removed from the family group.

Feather covering and wounds on each chick was photographed and scored on the last day of the trial, mainly on day 27 or 28 post hatch according to Tauson *et al.*, (2005). Feather covering concerning back, tummy, two sides of the neck, comb, wings and tail feathers were analyzed. The wounds on the chick's comb, rear part of the body and foot score were analyzed. Each body part was given a number between one and four, the best score was four. In the test, the higher the number the better the feather covering and the fewer wounds. The test score systems were calculated as mean for the hen and her family group. The chick could have a total feather plumage score between six and 24 and a total body wound score between three and 12, where 24 was the highest feather plumage score and 12 was the highest wound score given.

If the hen was aggressive towards chicks or expressed cannibalistic behaviour, she was euthanized. The hen was euthanized according to Swedish legislations, the hen was first made unconscious and then euthanized by the stable staff.

3 Results

3.1 General information

The number of family groups varied during the trial, due to exclusion. All excluded family groups developed feather pecking and was removed from the trial when the feather pecking behaviour was first discovered. For example hen family ID1 was excluded day 11 and was in all recordings until day 10 but no recordings was done day 11.

Social status test

The social status test showed that six hens were considered dominant and four were considered submissive in the adult group (Table 1 & Table 2).

Table 1. The social encounter test was done in the adult group. At least three interactions were observed of the observer during feeding. The hen gained points and lost points based on interactions of pecking, chasing, displacement (avoidance) and fighting. The mean social status score is the sum of all scores for each hen, as a mean value (mean social status score). If the mean social status score ≥ 0 the hen is considered dominant (D) in the adult group. If the mean social status score ≤ 0 the hen is considered submissive (S) in the group.

Hen ID	1	2	3	4	5	6	7	8	9	10
Mean social status score	4.2	-2.8	-4.3	0.1	-3.5	3.8	2.9	-8	0.1	7.5
Result	D	S	S	D	S	D	D	S	D	D

Broodiness

Out of the 10 hens four became naturally broody within the time for the eggs to hatch, one naturally broody died before she received any real eggs. Only three of the five non-broody hens accepted the live chicks and was considered as induced broody (Table 2), the other two were excluded due to aggressive behaviour. The induced broody hens expressed maternal behaviour directed to their chicks after the first night together.

Table 2. Background information for each Hen ID, hen weight in gram and age in years. Including a summary of each hens result from the social ranking test (D= dominant, S=submissive) and type of broodiness (NB= naturally broody, IB= induced broody). Number of chicks in each family group and the family groups last day in the chick trial. A full trial period were 28 days. The family group could be excluded due to feather pecking or wounds. Control groups number of chicks in each group and their last day in chick trial included as well. No results are indicated with an (-). Hen ID8, ID9 and ID10 were not included in the chick trial due to rejection and other difficulties.

Hen ID	Hen age(years)	Hen weight in gram	Type of broodiness	Social ranking test	Number of chicks	Number of days in chick trial
1 ¹	4	872	NB	D	6	10
2	1	942	NB	S	6	28
3	1	797	NB	S	6	28
4 ²	1	784	NB	D	6	5
5	1	866	IB	S	6	28
6	4	882	IB	D	6	28
7	4	810	IB	D	6	28
8	1	629	-	S	-	-
9	1	878	-	D	-	-
10	4	917	-	D	-	-
Control ID						
C1	-	-	-	-	6	28
C2	-	-	-	-	6	28
C3 ³	-	-	-	-	6	28

3.2 Behavioural interactions

Communication

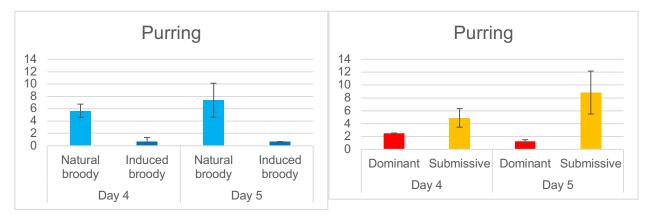
There were no significant differences between the different treatment groups, during continuous observation on hen vocalizations calling, clucking, other vocalizations and total number (included purring) on day four and five post hatch. However, there were a significant difference of the hen vocalization "purring" between the different treatment groups. The purring sound was not discovered in the literature supporting this paper. The submissive mother hens gave 0+more purring sounds than the dominant mother hens on day five post hatch (T-test; 2.11; $P \le 0.10$). The natural broody hens vocalized more purring sound than the induced broody hens (T-test: 4.62; $P \le 0.10$).

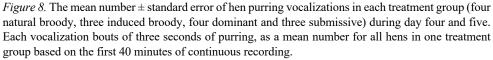
^{1.} Only trial day 4, 5, 10 + wound score due to feather pecking

^{2.} Only trial day 4, 5 + wound score due to feather pecking

^{3.} Only trial day 5 + feather score and wound score due to technical reasons

0.01) on day four and day five (T-test: 2.43; $P \le 0.10$). The mean numbers and standard error illustrated below (Fig. 8).





During continuous observation of vocalizations no significant differences between the different hens' vocalizations on day four and five post hatch were found.

Chick latency to approach

No significant differences were found between the control and the family groups on day five and 18. However, there were significant differences between the control groups and the family groups on day four (T-test: 2.21; $P \le 0.10$) and day 10 (T-test: 4.24; $P \le 0.05$). Where the control chicks were slower to leave at the start test compared to the family groups (Fig. 9).

Individual differences occurred during day five and 10, where the family group ID6 had the slowest chicks in the latency test compared to the other family groups. On day 10 family group ID7 had the fastest chicks in the latency test compared to all the other family groups.

No significant differences were found between the treatment groups in social status or broodiness, in chick latency to approach hen.

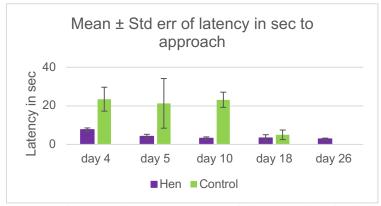


Figure 9. The mean values (Mean sec) \pm standard error (Std err) for the latency in the start test for family group hen (H) and family group control (C). The mean values in seconds(sec) for all the chicks in one treatment group to approach hen/feed or heater when "bird cage" is lifted in the beginning of the recording. Number of families in each treatment group day four (7H, 2C), day five (7H, 3C), day 10 (6H, 2C), day 18 (5H, 2C) and day 26 (5H).

Group spread

The group spread did not differ significantly between the different treatment groups. However, as the results for each day are shown in percentage of the number of chicks in relation to the center (hen or center chick), a descriptive trend can be seen between the different treatment groups (Fig. 10, 11 and 12).

There were individual differences between the family groups where family group ID6 showed the biggest difference in group spread. On day five family groups ID6 had the fewest chicks in its vicinity compared to all the other family groups. ID6 also had most chicks at a distance of more than two squares away compared to the other family groups. The family groups ID3 and ID7 had the most chicks in its vicinity and fewest at distance on day five compared to all the other family groups. On day 18 and day 26, the family groups ID6 had the most chicks in its vicinity compared to the other hen family groups. On day 26 family group ID6 even had the fewest chicks at distance more than two squares away compared to all the other family groups.



Figure 10. Illustrating the group spread for the hen group and the control group at "same square as centre" and "more than two squares away". Each day illustrate the percentage of chicks in that distance for each treatment group. Number of families in each treatment group day four (7H, 2C), day five (7H, 3C), day 10 (6H, 2C), day 18 (5H, 2C) and day 26 (5H). Measurement in percentage for all chicks in one treatment group once every minute during a 35 minutes interval.

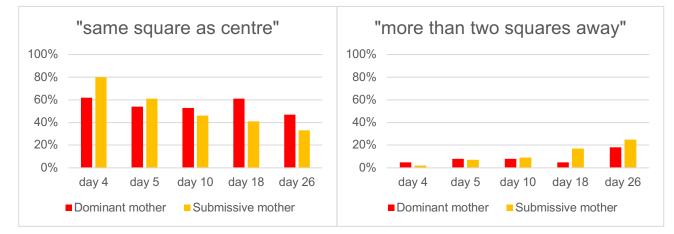


Figure 11. Illustrating the group spread for the dominant group (D) and the submissive group (S) at "same square as centre" and "more than two squares away". Each day illustrate the percentage of chicks in that distance for each treatment group. Number of families in each treatment group day four (4D, 3S), day five (4D, 3S), day 10 (3D, 3S), day 18 (2D, 3S) and day 26 (2D, 3S). Measurement in percentage for all chicks in one treatment group once every minute during a 35 minutes interval.

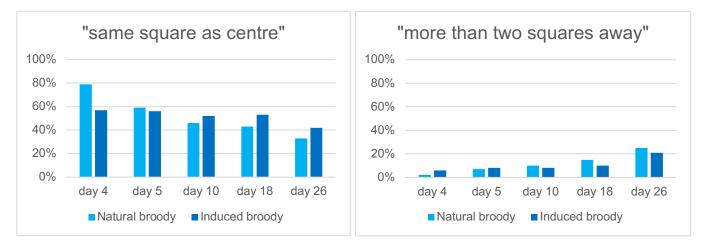


Figure 12. Illustrating the group spread for the natural broody group (NB) and the induced broody group (IB) at "same square as centre" and "more than two squares away". Each day illustrate the percentage of chicks in that distance for each treatment group. Number of families in each treatment group day four (4NB, 3 IB), day five (4NB, 3 IB), day 10 (3NB, 3 IB), day 18 (2NB, 3 IB) and day 26 (2NB, 3 IB). Measurement in percentage for all chicks in one treatment group once every minute during a 35 minutes interval.

Foraging behaviour

There was no significant difference between the different treatment groups in the foraging behaviour.

Individual differences between the hen family groups occurred. On day four the family group ID6 performed the fewest total number of foraging interactions. During day four family group ID7 had the highest total number of foraging interactions. During day five family group ID3 performed the fewest total number of foraging interactions. On day 10 the hen family groups ID3 and ID7 performed the highest total numbers of foraging interactions. However, on day 10 both control groups individually performed more foraging interactions compared to the hen family groups.

Feather pecking

The total number of pecking interactions included both feather pecking and gentle pecking at each other. No significant difference was found on day four, five and 26 (Table 3). However, there was individual variation in pecking behaviour between the family groups for day four and five. One of the family groups ID3 with the highest feather score (Table 4) had the fewest individually number of pecks on day four and five compared to the other family groups. The hen from family group ID1 and ID4, feather pecked the chicks and was removed and euthanized from the trial (Fig. 13). The chicks wounds from both family group ID1 and ID4 were scored according to Tauson *et al* (2005) and was included in the wound score for each treatment group in the end of the trial (Table 4 and 5).

During day 10 and 18 there was a significant difference between the treatment groups (Table 3). On day 10 chicks raised with a hen pecked significant more (Ttest; 7.77 and P \leq 0.0002) than the control chicks. This was also the case on day 18 (T_{test}; 24.90 and P ≤0.0001).

The chicks reared with a dominant hen pecked less than the chicks reared with a submissive hen on day 10 (T_{test}; 5.45 and P \leq 0.01).

On day 18 the chicks raised with a natural broody hen pecked less than chicks raised with an induced mother hen (T_{test}; 4.49 and P ≤ 0.05).



Figure 13. Photo by Giacomo Berteletti and Linnéa Christenson illustrating family groups removed from the trial. Hen aggression on the left image hen ID4, feather pecking on the right image hen ID1.

Table 3. The mean values and standard error (Std err) of total number of pecking for different treatment groups are present here. Total pecking include both gentle pecking and feather pecking. The mean value and standard error for all the chicks in one treatment group. The measurements was done continuously for 35 minutes on day four and five and once each minute during a 35 minutes interval on day 10, 18 and 26. Number of families in each treatment group day four and five (7H vs 2C, 4D vs 3S, 4 NB vs 3 IB) day 10 (6H vs 2C, 3D vs 3S, 3NB vs 3 IB), day 18 (5H vs 2C, 2D vs 3S, 2NB vs 3 IB) and day 26 (5H, 2D vs 3S, 2NB vs 3 IB).

10.51	iD) und u	uy 20 (5	11, 20 1	5 50, 21	0 10 5	1D).							
Pecking													
Treatment group		Contro	ol	All her	ns	Domin	ant	Submis	ssive	Natural	l broody	Induced	broody
		(C)		(H)		(D)		(S)		(NB)		(IB)	
Family ID		(1,2,3)		(1,2,3,	5,6,7)	(1, 6, 7)		(2,3,5)		(1,2,3)		(5, 6, 7)	
	Test	Mean	Std	Mean	Std	Mean	Std	Mean	Std	Mean	Std err	Mean	Std
	day		err		err		err		err				err
Continuous obser-													
vation	Day 4	7.33	± 2.69	5.62	± 1.51	5.88	±3.24	4.28	± 1.20	3.75	± 0.97	8.57	± 2.56
	Day 5	5.58	±2.52	6.97	±1.96	6.13	± 0.97	8.08	± 3.28	7.38	± 1.83	6.41	±2.14
One minute inter-													
val observation	Day 10	1.60	±0.10	2.34	±0.17	1.40	± 0.07	3.28	± 0.28	2.17	±0.17	2.51	± 0.18
	Day 18	1.00	± 0.00	1.72	±0.05	1.80	±0.12	1.67	± 0.00	1.50	± 0.00	1.87	± 0.08
	Day 26			1.60	± 0.00	1.50	±0.00	1.67	± 0.00	1.50	± 0.00	1.33	± 0.00

Feather scoring and wounds

The chicks' individual feather and wound score affected the family group score. The feather and wound score was calculated as a mean for each family group (Table 4) and each treatment group (Table 5). The feather score mean \pm standard error did not include family group ID1 and ID4. However, the wound score mean \pm standard error includes family group ID1 and ID4.

Table 4. The individual family groups feather (T) plumage and wound (W) score according to Tauson et al (2005) score system. The values presented are the mean value \pm standard error for each family group's six chick. The control groups (C) where named with the letters c, f or k. No feathers wounded or pecked had a score of 24 and naked birds due to feather pecking had a score of 6 in T. Score 12 in W indicated no wounds and 3 meant big wounds.

Individual fai	mily	ID1 ⁴	ID2	ID3	ID4 ⁵	ID5	ID6	ID7	C1	C2	C3
Feather plumage	mean		22.42	23.33		20.50	19.00	23.23	23.08	22.92	22.75
score	$\pm std \ er$		± 0.58	± 0.19		± 0.20	± 0.80	± 0.28	± 0.25	± 0.30	± 0.23
Wound score	mean	10.17	11.83	12.00	11.67	11.33	11.67	12.00	12.00	12.00	12.00
	\pm std er	± 0.44	± 0.15	± 0.00	± 0.30	± 0.30	± 0.19	± 0.00	± 0.00	± 0.00	± 0.00

Table 5. The score mean value \pm standard error according to Tauson et al (2005) score system for each treatment groups feather plumage. The mean value is for all the chicks in one treatment group. Number of families in each treatment group in the end of the trial in feather scoring (5H vs 3C, 2D vs 3S, 2NB vs 3 IB), in wound scoring (7H vs 3C, 4D vs 3S, 4NB vs 3 IB).

Feather plumage and wound score in each treatment group

Treatment groups	Control (C)	All hens (H)	Dominant (D)	Submissive (S)	Natural broody (NB)	Induced broody (IB)
Feather score	22.92±0.15	$21.70{\pm}0.37$	21.12±0.74	22.08±0.35	22.88 ± 0.33	20.91±0.51
Wound score	12.00±0	11.52±0.13	11.38 ± 0.50	11.72±0.13	11.42 ± 0.20	11.67 ± 0.14

The control chicks (three families) had a significantly better feather cover (T; 2.31 and $P \le 0.06$) and wound score (T; 3.62 and $P \le 0.01$) compared to the group raised with a mother hen (five families) day 27 post hatch.

The individual feather score of the two best family groups ID3 and ID7 (mean \geq 23.23) were higher compared to the control chicks (mean \leq 23.08).

No significant difference could be found between the social status treatment groups in feather covering or wounds score.

The chicks from the natural broody hens had a tendency for a significantly better feather plumage (T; 2.34 and $P \le 0.10$) but no difference occurred in the wound score compared to the induced broody chicks around 27 days post hatch.

^{4. &}amp; 5.No feather scoring was possible due to exclusion before end of trial, the chicks were younger and their feather covering could therefore not be compared to the other family groups.

4 Discussion

4.1 General information

The main part of the literature compared non-brooded chicks to natural reared chicks. The most common conclusion was an advantage if a mother hen reared the chicks, but also that there are differences in maternal behaviour (Perré *et al.* 2002; Field *et al.* 2007; Riber *et al.* 2007; Shimmura *et al.* 2010; Angevaare *et al.* 2012; Pittet *et al.* 2012, 2013, 2014; Gajdon *et al.* 2015; Edgar *et al.* 2016). In this study I found firstly variation in natural broodiness of the hens, and secondly variations in the possibility to induce broodiness.

Only four hens were naturally broody, three hens could be induced and of these two hens were excluded during the trial due to feather pecking and hence only five family groups remained until the end. However, the excluded hen groups wounds were scored and are included in the wound scoring.

There were large individual differences between the different family groups and different hens. This is important to consider when choosing key factors of the mother hen behaviour to potentially replicate under the commercial situation. It is important to choose the correct mother hen abilities. Two hens (ID3 and ID7) in this study reared chicks with better feather score compared to the control groups. However, the mothers (ID1 and ID4) had to be excluded due to feather pecking, and the family group with the lowest feather score (ID6) had a lower feather score compared to the control groups.

There were other, smaller, differences for each behaviour measured. However, no general conclusions can be drawn from this study due to small samples sizes.

Social status

According to Cloutier & Newberry, (2000) the social rank within the adult group can be determined both with hen social encounter and without using weight and age. However, the social status within the group varies, the social status in a moment for each individual hen, are based on the social interactions between the group members (Cloutier & Newberry, 2000). The hens in this trial were scored according to their pairwise encounters when competing for a desirable food resource. However, the effect of dominance in this context on the maternal ability is not completely straightforward. Due to feather pecking two of the dominant hens in this study had to be excluded before the end of the trial. Hence, dominance need to be investigated more carefully and potentially in other situations.

Broodiness

The imprinting starts within the egg and is fulfilled 96 hours post hatch (Fält 1981). The natural broody hens in this study got their eggs 48 hours pre-hatch and the induced hens received their chicks 12 hours post hatch, hence imprinting was likely not complete for the induced chicks. When the hen where induced to broodiness only three of five (induced broody) hens accepted the chicks and remained in the study. Even so there were differences when comparing the naturally broody and the induced broody family groups, indicating that the naturally broody hen has a more preferable maternal behaviour. Early observations of the induced family groups showed that the chicks dispersed further from the hen, compared to the chicks with the naturally broody hen, possibly caused by inadequate imprinting⁶.

4.2 Behavioural interaction

Communication

As mentioned before the mother hen affect the chicks behavioural development through communication (Field *et al.* 2007). The hen uses a lower frequency sound (Hill *et al.* 2014) and deeper rhythmic repetition which can create a deeper learning for the chick (Field *et al.* 2007) if expressed more often. The younger the brain and the more repetition, the easier it is to affect the memory (Field *et al.* 2007) in the young chicks, which could favour the vocalization and speed response for the natural broody family groups compares to the induced family groups. The difference in rhythmic and frequency could, with more research, clarify the hens' individuality and make it more specific to the chick. This study found significant differences between the treatment groups in the purring sound. During the literature research I have not found any literature focusing on purring. However, it seems to be a very important sound, due to our significant results. Previous studies have found that hen vocalization is of great importance (Appleby *et al.* 2004; Woodcock *et al.* 2004;

⁶ Giacomo Berteletti, 22/6-2018, personal communication

Edgar *et al.* 2015), hence vocalization is a key factor worth exploring more and potentially use in commercial rearing.

No literature found indicated any differences between the vocalization between a dominant hen and a submissive hen. However, there are studies indicating a higher frequency of maternal calls when the chicks where far away from the hen (Wauters & Richard-Yris, 2002), and the dominant hen reared more independent chicks that kept a longer distance to the mother hen (Pittet *et al.* 2014). Which suggests that the dominant mother should have a higher frequency of vocalization towards her chicks. However, the submissive hens in our study vocalized more purring sounds during thermoregulating, than did the dominant hens, which indicate that social status may not be a useful indicator of maternal abilities. Broodiness are far more important than social status.

The naturally broody hens expressed significantly more purring sounds during brooding on post hatch day four and a tendency could be seen on day five compared to the induced broody hen, indicating that broodiness could be a valid measurement of maternal ability.

Latency to approach

In this study the latency to approach the feed or heat, was shorter for chicks reared with a mother hen (Fig. 9), which is confirming earlier research (Wauters *et al.* 2002b; Edgar *et al.* 2015).

The studies of Perré *et al.*, (2002) and Pittet *et al.*, (2014) indicates a higher social motivation in chicks reared with a dominant mother. Wauters & Richard-Yris, (2002) saw a faster chick response with chick age. This study did not indicate any differences between the treatment groups (submissive and dominant) for chick latency to approach.

According to Field *et al.*, (2007) the chicks can feel a rush of noradrenaline secreted in their brain when their own mother hen is calling, which will increase their speed of response. Fält, (1981) and Mellor & Diesch, (2007) described the imprinting importance of the first day post hatch. However, our study did not indicate any differences between the treatment groups natural broody and induced broody in the chicks' latency to approach, potentially because the induced broody hens got their chicks quite early post hatch and the natural broody hens got their eggs quite late pre hatch.

Group spread

In the literature the natural situation is that chicks keep a close distance to each other (Angevaare *et al.* 2012) or the hen during the first period in life, due to the chicks dependence for external heat (Fält, 1981; Wauters *et al.* 2002b). Factors which

increase with chick age and can contribute to a bigger distance are, the chicks' ability to thermoregulate (Fält, 1981; Jensen, 2006; Shimmura *et al.* 2010; Haas et al., 2014b) and the chicks' curiosity for the surroundings (Wauters *et al.* 2002b; Riber *et al.* 2007) and other individuals (Appleby *et al.* 2004). In this study the chicks raised with a hen dispersed further from the hen with increasing age, as predicted due to their improved thermoregulation ability and increasing exploration.

However, chicks reared in family groups with a dominant hen deviated from this pattern and remained on the same distance to the hen during all test days. Furthermore, the induced hens' chicks followed a different pattern where the chicks dispersed less and followed the hen more closely. Hence, dominance and induced broodiness seem to affect the chicks behavioural development in a direction further away from the expected natural situation.

Foraging behaviour

The chicks raised with the mother hen are more efficient in their foraging behaviour compare to control groups (Wauters *et al.*, 2002b; Gajdon *et al.*, 2015). In this study both groups foraged more in the end compared to the beginning of the study. However, in this trial no differences could be found between the different treatment groups.

Feather peck

As Rodenburg *et al.*, (2009) and Shimmura *et al.*, (2015) suggested in their research, a mother hen can prevent the development of feather pecking if introduced early in the chicks life. According to Shimmura *et al.*, (2010) chicks reared with a mother hen spend less time with behaviours such as gentle feather pecking compared to the control chicks reared without a mother hen. In this trial the feather pecking results include both gentle and severe feather pecking. This study indicated that, the chicks reared with a mother hen feather pecked significantly more on day 10 and 18 compared to the control chicks. The control chicks had a significantly better feather and wound score compared to the chicks raised with a mother hen. According to Roden & Wechsler (1998) and Riber *et al.*, (2007) the mother hen did not affect the feather pecking behaviour in their chicks.

According to Appleby *et al.*, (2004) a hen provides the chicks with a more clear social status, which decreases the amount of feather pecking in the family. One older study found that dominant birds feather peck more compared to the submissive birds (Vestergaard *et al.* 1993). The maternal behaviour may perhaps not be directly transferred to chicks as in this study the chicks with a dominant mother hen pecked less compared to the chicks reared with a submissive hen. However, our data were on

the chicks' feather pecking behaviour, not the hens'. The results here indicated that there was no difference between the social status treatment groups in feather or wound score. However, two dominant hens were excluded during the study and were not recorded in feather scores which might have affected the results.

In this study the induced broody hens got their chicks later than the naturally broody. Chicks with induced mother hens pecked significantly more at each other and the hen during day 18 and had a tendency for a significantly worse feather scoring compared to the chicks reared with the natural broody hen. Which is similar to the results by Richard-Yris *et al.*, (1987). However, two natural broody were excluded before day 18 due to injurious pecking.

4.3 The best mother hen

Individual differences such as the mother hens' activity, affects the chicks rearing and group spread (Shimmura *et al.* 2010). The aim of this study was to identify key maternal behaviours during early rearing that could affect the chicks' behavioural development. In this study the bird breed old Swedish Bantam was used to ensure reliable maternal behaviour. Maternal characteristics investigated here where the social status (dominant versus submissive) and broodiness (natural broody versus induced broody). Behaviours measured were hen communication, chick latency to approach, chick group spread, chick foraging, chick feather pecking and the chicks' feather and wound scoring. However, the individuality of the mother hen differed in other behaviours as well, but were not included in this study.

Previous studies indicate that mother hen vocalization favors the chicks behavioural development (Appleby *et al.* 2004; Woodcock *et al.* 2004; Edgar *et al.* 2015). The best maternal variation was not completely confirmed by our results, more studies are needed. However individual variation between hens occurred.

4.4 Further studies

For further studies it would be interesting to see if there would be a maternal difference between chicks reared with a prerecorded hen and chicks reared with a real hen. It would be interesting to see if there is a beneficial effect to be reared with the best pre-recorded hen from this trial in a commercial production system.

Further research is needed to identify the best mother hen and to define her characteristics, since the best mother hens in this trial were very different. As previously mentioned differences between the hens occurred for each behaviour measured, however a bigger study with more family groups should be used for further research on individual characteristics of the mother hen. It would be interesting to know if different hen breeds were specifically designed in vocalizations and behavioural display for their own chick. Could it be harder for chicks to learn from a different mother, than that of the same breed?

5 Conclusion

5.1 General information

As a general conclusion, the control chicks raised without a mother hen in this trial are more even and healthy compared to chicks reared with a mother hen. Chicks reared with a hen can be both better or worse off than control chicks.

The hen individuality affects the maternal characteristics. However, the individuality of the hen is more complicated and further research in a larger scale is needed. The results of this trial were that family group hen ID3 (submissive and naturally broody) and ID7 (dominant and induced broody) were the best mother hens based on feather and wound score. The worst mother hen still remaining in the end of the trial based on feather and wound score, was hen ID6 (dominant and induced broody), which complies with the literature.

Previous research has found a connection between the foraging behaviour and the feather pecking behaviour, a higher foraging decreases feather pecking as adults (Blokhuis & Arkes, 1984; Huber-Eicher & Wechsler, 1997). Even if previous research found that foraging is important for feather pecking this study did not indicate any difference between the treatment groups in foraging behaviour. Instead it seems that the individuality of the mother hen affects the feather pecking, and that individuality in this study is not affected of the social status or the broodiness.

5.2 Behavioural interaction

Hypothesis 1 (communication)

In this study there were no differences in total vocalization between the hen groups (dominant, submissive, natural broody and induced broody). However, there were significant differences between the treatment groups in the sound "purring". The prediction that a dominant mother hen vocalize less was confirmed concerning the "purring". The chicks raised with a dominant mother was faster in their latency to approach compared to the chicks raised with a submissive hen, which could indicate that the dominant mother does not have to vocalize as much as the submissive and proves the hypothesis.

In contradiction to the hypothesis the natural broody hen expressed significantly more purring sounds during brooding compared to the induced broody hen. There was no difference between the treatment groups on the chicks' latency to approach the hen, which could indicate that both treatment groups vocalized equally and achieved the same results.

Hypothesis 2 (group spread)

Hypothesis 2 is confirmed as chicks raised with a hen stayed closer to the center in group spread compared to control chicks raised without a mother. Chicks raised with a dominant mother stayed indeed further away from the hen. However, this pattern continued during the entire trial not just day four and five, which was unexpected. The natural broody hens chicks stayed closer to the hen during the first days (four and five) compared to the chicks reared with the induced broody hens. The natural broody hens' chicks increased their distance to their mother hen as they grew older.

Hypothesis 3 (foraging)

Hypothesis 3 was not proved because no significant differences were found in foraging behaviour in any of the treatment groups.

Hypothesis 4 (feather pecking)

This hypothesis was proved in some points and not in other. This study even indicates that a mother hen might not counteract feather pecking, but instead the mother hen might even favor feather pecking behaviour in her chicks. However, the feather pecking between the chicks in this trial was proven to be less in the dominant family groups on day 10. Indeed the hypothesis of the chicks reared with a natural broody hen was correct, they feather pecked less on day 18 and had a better feather scoring in the end of the trial. However, there were no differences in the wound scoring which contained all hens in each treatment group.

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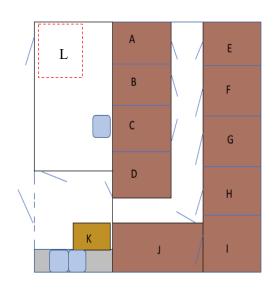
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Appendix 1; Measure points and values



Stable design and measures

Decibell was measured with a Velleman, DVM401, Environment Meter, No 12060373, Lo=35~100dB

Measure points	Distance between	dB
Κ	2 cm	77
L (open door)	4.5m	55.7
А		55.1
F		50.2
Н		52.5

Sound used was "Basic Tone" on the highest volume of alarm in a Samsung Gallaxy A5 (2016), model No SM-A510F. The phone was placed on the measure point K.

The doors were closed and peat mixed with wood shaving was covering the floor where the chicks stayed. The ventilation was kept at a low rate to minimize the sound, but enough to ventilate the room.

Lux was measured on a cloudy day with a LT Lutron lux meter, (L627837), 0-1995, LX-101. The lux was the same between 4:40 a.m. and 21:00 p.m. and were maintained of non-flickering led light lamp (iLOX, Xena Pro 7W, warm white 3000K, 770 Lumen, 50/60 Hz, 37mA, Vechta Germany).

Measure points	А	В	С	D	Е	F	G	Н	Ι	J
Lux value 1	13	16	15	39	6	12	12	21	7	8
Lux value 2	11	14	17	22	6	11	19	23	6	12
Lux value 3	8	13	13	17	5	13	11	24	13	14
Mean lux value	10.7	14.3	15	26	5.7	12	14	22.7	8.7	11.3

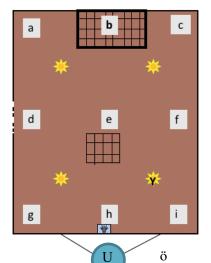
The lux in a nestbox inside measure point H got the values of 3, 25 and 14 depending on how far in the nestbox the measurement where. Mean lux for the nestbox inside H where 21 Lux. A closed nestbox for transportation had a lux value of 0.

Shed design and measures

The same gear for lux measurement and dB was used inside the shed. The shed was stored within a larger building. The floor was covered with litter about 5-10 cm of

peat. There were four lamps (Non-flickering led light lamp (iLOX, Xena Pro 7W, warm white 3000K, 770 Lumen, 50/60 Hz, 37mA, IVechta Germany)), in the roof and the "wire cages" was in the roof during the measurements. Both doors were closed for the lux test. Measurement in dB. The phone was used for noise.

	Phone on e	Phone on U
Door open	U=47.6	e= 49.3
Door closed	U=46.0	e= 42.7



Lux measurements in the shed:

Measure points	а	b	с	d	e	f	g	h	i
All lamps turned on	103	134	108	163	198	162	120	147	130
Only lamp Y turned on	12	21	19	33	45	51	28	48	56

The dB was measured with the same gear and the same phone and volume as in the stable. The measure points used for the dB was U and e. The measurement point e without any sound was registered a sound of 42.5dB. The distance between U and e was 4.5m. The phone was placed on measure point U or e and the measurement device was placed on the opposite measure point. The door ö was opened in some cases and closed in other. The measurement of 76.5dB was recorded, when both the phone and the measurement device were placed on measurement point e with a distance of 2 cm and the phone the same settings as before. The sound can be comparable to the background sound in a library of approx. 40 dB (Temple University Department of Civil/Environmental Engineering 1992).

All measurements were done at chick level around 5cm from the floor/sur-face/litter.

The recording shed was located 23 m from the stable, with doors and walls in between. The sheds inside temperature was 19°C, with a humidity of 35%. During the control recordings a heating lamp was used and the inside temperature of 21 °C was reached at the most distant corner.

Appendix 2; Protocol vocal and video recordings

Behaviour interactions recording from roof view was done with the GoPro Hero4 Silver camera, a resolution of 1440 pixel and a frame rate of 24 frames per second with the wide angle, were used during the recordings. The GoPro camera was installed in the roof (1.5m above the floor) on a built self, the camera was placed in the center of the shed.

Procedures

The birds were gently placed inside the nestbox, with peat as litter and carried to the shed, trough the inside of the birdhouse. Day 4, 5, 10, 18 and 26, the hen is placed outside the "wire bird cage" and the chicks are gently placed inside the "wire bird cage". The birds walks over themselves or are helped to walk across, through open doors (close to each other), feed presented in "feed cage" no water present during the recorded 45 minutes. The recordings for each family group starts between 9:00-11:00. Information of the family group presented on whiteboard which hen is there and their respectively post hatch day, shown to GoPro after started recording. Handler leave the shed and close the doors. "Food cage" is lifted and secured to the roof, when the hen approaches the feed (beak within the brown plate beneath the feed the "bird cage" is lifted (the chicks are free) and the "bird cage" is secured to the roof and wall, out of view for the GoPro camera. One handler watch that no chicks are still presented in the cage before secured to the roof. After the recording the birds are moved (gently gathered in one of the corners, and the nestbox is opened for them to walk inside), then the recorder is ended. Thereafter the hen and chicks are transported to the stable within the nestbox.

Appendix 3; Definitions

Square calculations

The area of the behavioural shed is 42300cm^2 (235 cm*180 cm). The area was divided into squares, each squares had a floor area of 132.19cm^2 with the measurements of 11.50 cm*11.50 cm, due to the cameras position 1.5m above the floor.

Behavioural interactions

Video analysis (GoPro recordings) 40 minutes recordings starts when wire bird cage leaving the floor, (p.m. recordings day 4 and 5 a.m. recordings day 10,18 and 26).

Latency to approach

The latency in seconds for each chick, from when the "bird cage" was lifted to when chicks is one square away (11.5*11.5cm) from external heat or feed.

Continuous recordings

Recordings analyzed continuously for 35 minutes during day 4 and 5. Behaviours peck at hen or peck at chick in feather pecking (gentle and hard) and hen vocalizations cluck, feed calling, <u>purring</u> and other (analyzed as a 5 secunds bout.

Instantaneous sampling

Every minute for 35 minutes day 4, 5, 10, 18 and 26. If two behaviour same time during 5 sec, record the longest or the first one.

- Feather pecking, peck at hen (both gentle and hard), peck at chick (both gentle and hard).
- Each chicks distance from center (hen or center chick), if chick is under heat lamp distance to closest part of lamp distance in squares (11.5*11.5cm) location in cage, same square as center, 1 square away from center, 2 square away from center or more than 2 square away from center.
- Foraging litter (pecking more than one square away from brown feeding place), foraging feeder (pecking maximum one square away from brown feeding place), same behaviour as mother/go to mother, run or fly to her if 2 squares or more away, copying her if closer than 2 squares (or to center chick if chick group).
- Vocalizations (clucking, calling (high sound close to when she starts to forage, one or more chicks run to her), purring (when brooding), other vocalization).