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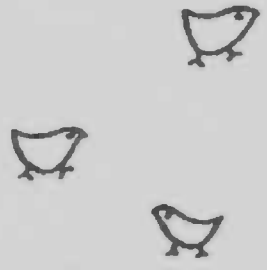
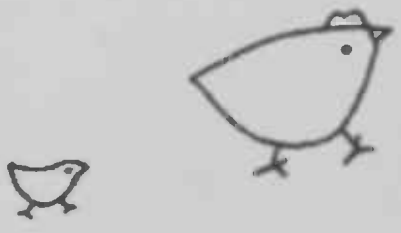


Rianne Lindhout
June 2000

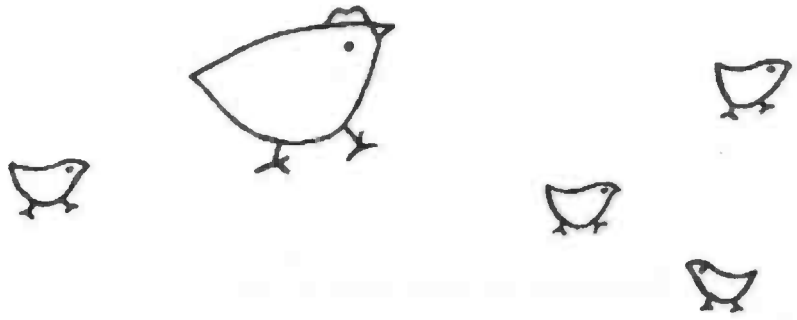
Supervised by Bernd Riedstra
and Ton Groothuis



Ontogenetic determinants of feather pecking in laying hens

The report that lies before you was written as a result of a study in which it was tried to find causes for feather pecking behaviour in laying hens and its relation with stress responses.
The study took place from October 1999 till April 2000 and was performed by drs. Bernd Riedstra, Ebele Zuidema and Rianne Lindhout, supervised by dr. Ton Groothuis who created the experimental design and helped to interpret the results.

Rianne Lindhout, June 2000



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White-Leghorn *Gallus domesticus*

1. Abstract

The development of feather pecking was studied in relation to laying background, social rearing condition and coping style in two strains of white leghorns (*Gallus domesticus)* that differ in their propensity to feather peck and in stress response. The strain with a high propensity to feather peck (high feather peckers or HP) reacts with a strong cathecholamine response to manual restraint whereas the strain with a low propensity to feather peck (low feather peckers or LP) reacts with a high corticosterone response.

The development of feather pecking was recorded in two experimental conditions: a semi-commercial and a semi-natural condition. In the semi-commercial condition chicks hatched from commercially produced eggs were raised in a large group without a mother. In the semi-natural condition chicks hatched from eggs produced by small groups of free-ranging hens were raised in small broods by a (foster) mother. Both strains were represented in both treatments by four replicates.

Over the course of twenty weeks pecking frequencies were scored on average once a week for thirty minutes per cage on individually marked hens and roosters. During the first four weeks there was no effect of housing condition, but high feather peckers showed more feather pecking then the low feather peckers. From week five to twenty the effect of strain persisted and an effect of condition became visible: semi-natural chicks showed less feather pecking then semi-commercial ones. This was especially apparent in the HP-strain.

To separate the effect of housing condition from laying and social rearing backgrounds one male and one female of each group were reallocated to one new cage after four weeks. The strain difference in feather pecking observed in the first four weeks disappeared; there was also no difference in feather pecking between the former semi-natural and semi-commercial chicks. The housing conditions therefore did not have a persistent effect on feather pecking.

The high peckers feather pecked less after reallocation. It seemed that the presence of low peckers 'diluted' the feather pecking: they did not join it and that probably inhibited the high peckers. We also found correlations between social pecking and feather pecking. From these and other results we conclude that feather pecking has an important social component.

In a standard stress test (open-field) we found a persistent effect of rearing condition on how individuals behave. Semi-naturally raised chicks behaved less inhibited than semi-commercially reared chicks. Most remarkable was the difference in righting time in a tonic immobility test performed six weeks after reallocation: the semi-naturally reared chicks had a shorter righting time than the semi-commercially reared chicks.

At twenty weeks after hatching the reallocation period was repeated with the female focal chicks from the home cages. No differences in feather pecking were found between the four groups then. The strong differences between the groups that had been found in coping in the open field and tonic immobility tests during the first reallocation period had disappeared in the second reallocation period.

Semi-natural housing conditions seem to have a diminishing effect on feather pecking, but this effect is not persistent after changing these conditions. The feather pecking by high peckers decreases after reallocation, probably because of the presence of low peckers. Housing conditions do have a persistent effect on stress responses, which are a part of coping style, at least until nine weeks of age. Semi-natural circumstances apparently help the chicks to be less inhibited in coping with stress factors. To solve the problems caused in poultry farms by feather pecking it is probably necessary to keep the groups small and to let a (foster) mother raise the chicks. We have not found a relation between Feather pecking and coping with stress.

2. Introduction

2.1 Feather pecking and its causes

Feather pecking is a major problem in commercial poultry farms (e.g. Blokhuis & Arkes, 1984). Chickens peck and pull at each others feathers, which can lead to severe feather damage and even death (cannibalism). In fact, up to 12 percent of all chickens in a farm may die of it (WB, 2-3-200). Besides mortality and increased food demand by bald chickens (Tauson & Svensson, 1980), cannibalism and feather pecking also constitute serious welfare problems.

As laying batteries will be forbidden in 2011 the problem of feather pecking will have to be solved then, because the frequency of feather pecking seems higher in big barns in which thousands of chickens are held than in the small cages that form laying batteries. Solutions already exist: beak trimming and dimming the light are general measures against feather pecking. Both measures only control the symptoms, and beak trimming may cause chronic pain.

Summary of studied questions
- To which extent can the occurrence of feather pecking be influenced by the manipulation of laying condition and social rearing condition?
- What developmental mechanisms underlie feather pecking and what is its relation with coping style and sensitivity to stress?

Several causes of feather pecking and cannibalism have been proposed. Sirën (1963) suggested that feather pecking was caused by too low arginine content in the food, but the results of studies investigating this factor have been inconsistent and inconclusive (Hughes, 1982). Aggression has been proposed as a cause, but Hoffmeyer (1969) demonstrated in pheasant chicks, *Phasianus colchicus*, that feather damage was caused by non-aggressive pecks and could be reduced if other substrates to peck at were provided. Feather pecking was seen as a substitute for normal feeding behaviour, but not as a consequence of nutritional deficiencies.

Blokhuis (1986, 1989[a]) concluded that feather pecking is a form of redirected ground pecking. Besides the intake of food, ground pecking is also an exploratory behaviour, serving to gather information (Wood-Gush *et al.*, 1983). Thus, different qualities of the ground(particles) such as visual, tactile or gustatory feedback signals, long-term effects of ingestion, as well as novelty, may all play a role in directing the pecking to the ground or to feathers of conspecifics (Blokhuis, 1989[b]). To prevent birds redirecting their ground pecks to the feathers of other birds, the peckability and scratchability of the ground is important but also other aspects like nutritive value or taste (Blokhuis & Van der Haar, 1990).

These examples of possible causes all are environmental factors. In this study feather pecking was studied in relation to environmental and genetic factors: individual characteristics of the animal. Previous research indicates that large individual differences exist in the propensity to feather peck (e.g. Hughes & Duncan, 1972) and this propensity is related to coping style*. Feather damage has also been found to be associated with increased (chronic) fear (Hughes & Duncan, 1972; Quarts & Adams, 1982). Therefore we analysed the development of feather pecking in individual birds and in relation to the development of their individual stress response and coping style. If certain factors influence the development of feather pecking do they influence the stress responses at the same time and in the same direction (less pecking = less fear and inhibition)? In this study we searched for causes of

* Coping style can be defined as the complex of individual behavioural and physiological characteristics that determine how an animal responds to environmental challenges.

feather pecking by doing research on leghorn chicks from the week they hatched until they were 23 weeks old. Young chicks were used because we wanted to find developmental mechanisms in ontogeny that underlie feather pecking. Except observing the pecking behaviour in a leghorn strain with a high and a low propensity to feather peck, we did behavioural tests to find differences between their coping styles.

2.2 Feather pecking and coping style

Breeds and lines of laying hens have been reported to show consistent differences in their propensity to feather peck (e.g. Hughes and Duncan, 1972). In our study we used two lines with different propensities to feather peck: a high feather peck line (HP) and a low feather peck line (LP). Besides the difference in feather pecking there are more differences between these lines. The high feather peckers react with a strong cathecholamine response to manual restraint whereas the low feather peckers reacts with a high corticosterone response (Korte *et al.*, 1997). In one case two lines that differed with respect to feather pecking damage have been reported to show different levels of fear measured by the tonic immobility test (Blokhuis & Beutler, 1992). The high peckers were more fearful at fourteen weeks of age than the low peckers. The birds of the different lines show behavioural and physiological differences that reveal profiles of peckers and non-peckers. We wanted to know whether manipulations influence feather pecking and these differences at the same time; whether feather pecking and coping style are related.

We wanted to know whether different 'coping profiles' exist for high peckers and low peckers. Therefore we needed experiments to measure aspects of coping style, such as coping with stressful situations. Bronson (1968) hypothised that heightened emotionality may inhibit rather than facilitate the expression of a variety of behavioural patterns associated with the avoidance of fear-arousing stimulation.

Tonic immobility, the response on a brief period of physical restraint, is widely used as a method of estimating fearfulness and it is considered positively related to fear (Jones, 1986). procedures intended to increase fear, such as shock, suspension over a visual cliff, simulated predatory encounters and loud noise, prolong the reaction whereas fear-reducers like taming, habituation, tranquillisers, the presence of social companions and conditioned safety signals, attenuate the response (Gallup, 1974, Jones, 1985). In our experiment we analysed the results of males and females together, as no sex differences in the duration of tonic immobility have been found (Benoff and Siegel, 1981).

Two white leghorns stocks that were selected for increased part-year egg mass showed more prompt head movement than unselected controls did (Craig *et al.*, 1984). Domestic chicks that were selected over several generations for high levels of activity in an open field or novel environment were subsequently found to show lower fear levels in a tonic immobility test than those of the inactive control lines (Faure, 1975; 1981).

The housing system can exert profound influence on tonic immobility. Adult white leghorn hens housed in groups of four in floor pens showed significantly shorter durations of tonic immobility than those cages in groups of four (Jones and Faure, 1981). Kujiyt *et al.* (1983) also observed shorter immobility reactions among birds housed socially in pens rather than in cages. Their differences were smaller than those of Jones and Faure (1981), but Kujiyat *et al.* (1983) housed their birds in groups with different sizes (one, five, fifteen and seventeen birds). So varying degrees of separation distress upon testing may have influenced their results (Jones, 1986). Indeed, hens caged in groups of seventeen showed longer tonic immobility responses than those caged in groups of five and, because area per hen was essentially equal in both environments, it was suggested that group size was a major factor

affecting tonic immobility (Kujiyat *et al.*, 1983). These results suggest that individually caged birds react more fearfully to disturbing stimuli than those pen-housed though it is not clear whether this effect is due to structural differences between cage and pen housing or to differences in social density.

From the discussed studies it can be concluded that genetic characteristics and housing conditions influence the way in which chickens cope with the stress of a tonic immobility test. In this study we are interested in the relation between feather pecking behaviour and coping with stress. Therefore a tonic immobility test took place with the experimental chicks.

Another way to measure fear is the open field test. Animals are placed in a novel environment individually and behavioural elements such as distress calls and locomotion are scored. According to Bronson (1968) the most fearful animals should display much freezing behaviour and little activity. Ginsburg *et al.* (1974) subjected handled and non-handled chicks to an open field test and a tonic immobility test and found (as expected) that the first were less fearful as they walked more and showed less freezing behaviour than the non-handled chicks. In the tonic immobility test they were faster to get up. In the open field test however, they also uttered more distress calls. This finding does not support the general assumption that a monotonic relationship exists between heightened levels of fear and an increased frequency of distress calls. It does support Bronson's (1968) hypothesis.

To measure other aspects of coping style than stress responses, such as competition behaviour, other behavioural tests took place with the experimental chicks of our study. Mealworm competitions were held and a strange chick was put in the cage. To measure the interest in feathers and hemparade (grinded hemp stalks on which the chicks lived) cards with the materials were hung in the cage. With these tests and scores of other pecks than feather pecks we hoped to find differences between chicks that are related with differences in feather pecking.

2.3 Rearing conditions

There are many indications that in mammalian species early experiences can induce persistent changes in stress responses at the level of physiology, brain and behaviour (Groothuis, 1997). Long term effects of stress during pregnancy of the mother on the offspring have repeatedly been found in rodents (Ward, 1991; Suchecki *et al.*, 1995). They might be induced by the social environment of the mother (Sachser & Kaiser, 1996). Prenatal gonadal hormones may be crucial for the development of different coping styles (Compaan, 1993). Early postnatal stress, such as handling and deprivation of contact with the mother, have often been found to influence later stress responses at the level of physiology and behaviour (Meany *et al.*, 1991; Kloet *et al.*, 1988).

The factors that influence development in mammals probably also play a role in fowl development (Groothuis, 1997). In several bird species newly laid eggs contain considerable doses of maternal gonadal steroids. Androgen levels vary considerably between eggs of different females, and correlate with testosterone plasma levels of the females at the time of laying (Schwabl, 1996[b]). Treatment of newly laid eggs with testosterone influences later behaviour and growth in canary chicks (Schwabl, 1996[a]).

Since early androgens influence the development of coping style in rodents (Compaan, 1993) and testosterone can suppress the development of feather pecking (Hughes, 1975) maternal gonadal steroids may be important determinants of individual differences in the propensity to feather peck. Stress hormones may also be important in this perspective. Laying hens used in commercial farms are produced by hens kept in stressful situations, most likely experiencing elevated corticosterone levels during laying. This may influence later stress

responses of the offspring by transfer of these maternal stress hormones (Groothuis, 1997). As the circumstances under which the mother of the chicks lives during laying are important we used two types of eggs in this project. Our chicks hatched from commercially produced eggs or from eggs that had been laid by hens that lived in small groups in large outdoor cages. We hypothised that chicks hatched from commercially produced eggs would be more stressful (e.g. show more freezing in the open field tests) and show more feather pecking, as we think that more stress leads to more feather pecking.

Secondly, like in mammals, early postnatal conditions may have a profound influence on later stress responses in birds. During an early sensitive period imprinting on the mother and siblings takes place in virtually all birds studied in this respect (Groothuis, 1997). The behaviour of the mother may guides the proper orientation of pecking behaviour of the chick. The mother very often performs ground scratching, ground pecking (food exploration) and dust bathing in front of her chicks. Evidence indicates that feather pecking is related to these behaviours (Huber-Eicher & Wechsler, 1997), suggesting that feather pecking might be wrongly or re-directed food exploration pecks or pecks that belong to the dust bathing system. Vestergaard *et al.* (1993) found that in small groups of junglefowl, *Gallus gallus spadiceus*, feather pecking during dust bathing was the main cause of feather damage. They suggested that the primary cause of feather pecking is an abnormal development of the perceptual mechanism responsible for the detection of dust for dust bathing.

With a hen in the cage chicks show less flight responses and they synchronize their activity (Roden and Wechsler, 1997). However, the mother is absent during commercial rearing, which takes place in abnormally large groups. Therefore the development of feather pecking and stress responses might be strongly influenced by the social rearing conditions. Roden and Wechsler (1997) found no difference in feather pecking between chicks with and without a hen. In our study we kept chicks with or without a (foster) mother as the presence of a hen seems to be an important factor in the development of pecking behaviour and coping style. Following from the proceeding information we hypothised that the presence of a mother would diminish feather pecking and stress.

The group size is another factor that may influence the development of feather pecking. In commercial housing conditions the normal process of imprinting is very likely to be disturbed, due to the lack of the mother and the enormous amount of birds in the group, hampering the formation of individual relationships (Groothuis, 1997). It has not been tested so far whether the lack of imprinting influences later stress responses and coping style. It may increase fear (Groothuis, 1997), and fear has been implied in the causation of feather pecking (Vestergaard *et al.*, 1993). It also has been shown to disturb the recognition of conspecifics in many studies (Groothuis, 1997), which may lead to non-social behaviour – feather pecking – at social companions. The group size is therefore the third factor that was manipulated in this project: we hypothised that a large group would feather peck more and contain more stressful chicks. We have tried to find a relation between rearing conditions (egg type, presence of mother and group size), feather pecking and stress responses.

3. Methods

3.1 Experimental design

Two strains of Hisex White Leghorns *Gallus domesticus* were used to study the development of feather pecking in relation to laying background and social rearing conditions. The strain with a high propensity to feather peck (high feather peckers or HP) reacts with a strong cathecholamine response to manual restraint whereas the strain with a low propensity to feather peck (low feather peckers or LP) reacts with a high corticosterone response. Three parameters (presence of a mother, group size and egg type, as described in the introduction) were used to maximize the difference between laying backgrounds and social rearing conditions. A two by two design with strain and treatment was created in order to measure gene-environment interactions. Table 1 summarizes the main differences between the four groups that were created this way.

Table 1.The four experimental groups, created by combining two treatments and two strains of chickens.

Strain / Treatment	High feather peckers (*HP*)	Low feather peckers (*LP*)
Semi-commercial (*SC*)	Commercially produced eggs 45 chicks per cage no mother *HPSC*	Commercially produced eggs 45 chicks per cage no mother *LPSC*
Semi-natural (*SN*)	Naturally produced eggs 6 chicks per cage mother *HPSN*	Naturally produced eggs 6 chicks per cage mother *LPSN*

In the semi-natural treatment (SN), eggs produced by chickens living in small groups in large outdoor aviaries were individually marked and put in incubators. After hatching six randomly selected chicks were weighed, individually marked with the colours black, blue or green on heads and necks, bled (for sexing) and put under a silky hen. Chicks and hen were transported to a standard aviary measuring 3×1.5 m and placed in an artificial container (to facilitate the adoption of the chicks). After 18 hours the hen with the chicks were allowed to freely use the entire aviary. The mother was present until the chicks were 20 weeks old.

In the semi-commercial (SC) condition 45 chicks, hatched from commercially produced eggs (NL, Hendrix Poultry Breeders), were placed in similar aviaries as the semi-natural treated chicks. At hatching eight chicks from individually marked eggs were weighed, marked and bled. For the first four weeks semi-commercial groups were restricted to 1 m^2. After four weeks the floor space was enlarged to 2 m^2 and after five weeks the groups were allowed to use the whole aviary freely for the remaining fifteen weeks.

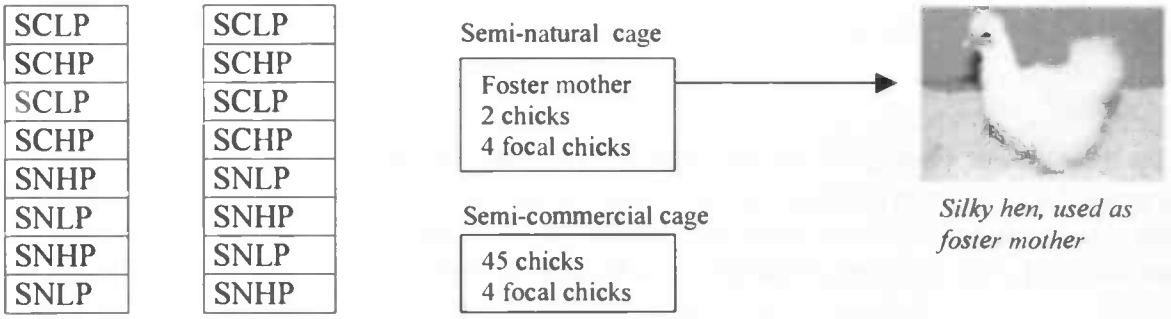


Figure 1. Distribution of groups over the aviaries. The cages were visually separated.

3.2 Housing, food and light

All groups were housed in similar aviaries measuring 3×1.5 m (the distribution of the groups over the sixteen different aviaries is depicted in figure 1). The floor in each cage was covered with hemparade (Hemp flax B.V.): grinded hemp stalks. Water and food were provided ad libitum. On the far end of each aviary was a plateau where a constant flowing water system was installed. Next to the entry of the aviaries either one (semi-natural condition) or two (semi-commercial condition) large feeders were installed. However, for the first five weeks chicks received food and water from smaller portable feeders. The chicks eat breeding crumbs: *dfk raising crumbs*, produced by Hendrix UTD. During the first four weeks the chicks eat crumbs 1; after that they received crumbs 2.

In each cage light was provided twelve hours a day for the entire period by a single 100 W light bulb. During the first four weeks two red brooding lamps (150 W) were used as heat sources in the semi-commercial condition and provided light for 24 hours. Because of them the temperature was about 37°C. In the semi-natural situation there were no brooding lamps: the foster mother kept the chicks warm. This mother was a silky hen (see figure 1). In each semi-natural cage there was an extra 40 Watt light bulb that remained on at night during the first four weeks. The cages were kept at approximately 20°C by fixed electric heaters on the far end of the cages. These systems were also used in the semi-commercial cages after the brooding lamps had been removed.

All differences between semi-commercial and semi-natural cages are summed in table 2.

Table 2. Differences between semi-commercial and semi-natural cages.

Semi-commercial cages	**Semi-natural cages**
Chicks hatched from commercial eggs.	Chicks hatched from semi-natural eggs.
Group size: 45 chicks.	Group size: six chicks.
No hen present.	A silky hen present.
High density of chicks.	Low density of chicks.
Cage size 1, later enlarged to 4,5 m^2.	Cage size 4,5 m^2 during the complete experiment.
Two cage enlargements experienced.	No cage enlargements experienced.
Two breeding lamps present during first four weeks.	No breeding lamps present.
Heat provided by breeding lamps the first four weeks; after that by one red heat source per two cages.	Heat provided by hen and one red heat source per two cages from the beginning.
Light dark cycle: 12:12 hours by turning a 100 W lamp on and off.	Idem, but in the dark stage there is a 40 W lamp to prevent total darkness.
Food: only breeding crumbs and – after a few weeks – gravel.	Food: breeding crumbs and gravel and laying feed meant for the silky hen.
Cages are cleaned more often (once a week after ten weeks).	Cages are cleaned only a few times.
Cages are dirtier (in spite of the weekly cleanings).	Cages are cleaner.
Focal chicks are coloured more often (once a week).	Focal chicks are coloured about once in three weeks.
More disturbances because of cleaning, wounded chicks that had to be treated or removed and floods caused by dirt in the water pipes.	Fewer disturbances.

Ethical note

If severe bleeding caused by feather pecking was observed the birds were treated with spray or removed, to prevent massive outbreaks of severe feather pecking possibly resulting in high mortality rates. Furthermore, culling of individuals took place for stock health reasons and individual well being, if chicks showed signs of a severe health deterioration. This was mostly apparent in the semi-commercial groups.

3.3 Obtained data

3.3.1 Home cage observations

Observations on pecking frequencies began when the chicks were one week old. Pecking behaviour was recorded between 9:00 and 15:00. Pecks directed to the ground, feathers (gently or severely), objects, particles on feathers, bills, weak parts (comb, feet and eyes) and tags were counted. An ethogram of the different pecks is depicted in table 3. Pecks received from cage mates were counted at the same time. In the first four weeks four focal chicks per cage were observed (two males and two females), each during 7.5 minutes twice a week. After four weeks due to reallocation of 2 focal chicks (see 3.3.2) two focal chicks were observed (one male and one female) each during fifteen minutes once a week until they were 20 weeks old. Appendix C1 shows our protocol form for registering pecking orientations.

Table 3. Ethogram of recorded pecks in different orientations.

Pecking behaviour	Definition
Ground	All pecks directed to the ground more than 3 cm from the feeding trough.
Feather (gentle)	Gentle pecks directed to plumage (Bilcic & Keeling, 1999).
Feather (severe)	Pecking and pulling severely at plumage (Bilcic & Keeling, 1999).
Head pecking	An aggressive, quick peck (mostly directed to the head).
Bill	Pecks directed to the bill.
Weak parts	Pecks directed to the eyes, feet or comb.
Tag	Pecks directed to the wing tag.
Objects	Pecks directed to objects such as the wall or the water trough (not drinking).
Particles on feathers	Pecks directed to particles on feathers, like hemparade or dust.

From the fifth to the fifteenth week after hatching we observed the behaviour of the two focal chicks per cage by one-zero sampling. During fifteen minutes per chick (i.e. thirty minutes per cage) we scored seventeen behavioural components. In table 4 these components are visible. One-zero sampling took place once a week, between 9:00 and 15:00. Each minute all performed components were noted on a protocol sheet which is visible in Appendix C2.

Table 4. Ethogram of recorded behavioural components during one-zero sampling.

Behavioural component	Definition
Walk	Gentle walking.
Run	Running through the cage.
Stand	Standing still.
Sit	Sitting or lying.
Fly	Flying.
Sleep	Sleeping
Eat	Eating from the feeding troughs, or pecking at the ground very near to it.
Drink	Drinking from the drinking pipe.
Comfort behaviour	Cleaning feathers and stretching wings and feet.
Dust bathing	Sitting and shaking with wings, so that the body gets covered with dust and hemparade. Mostly combined with ground pecking.
Alert	Concentrating on the environment. other chicks, insects or the observer.
Freeze	Instantly stopping any movement, mostly as a reaction on a sudden sound.
Distress call	Screaming, mostly as a reaction on an attack or severe peck from another chick.
Jump or threaten	Threaten another chick by jumping in the air, not always followed by an actual attack.
Aggressive peck	Quickly and severely pecking at feathers, head or other body parts or pulling hard at feathers.
Ground peck	Pecking at the ground. Also ground pecking during dust bathing is noted here; pecks very near the feeding through are scored as eating.
Feather peck	Gently pecking and pulling at feathers.

3.3.2 Reallocation experiments

In order to separate the effect of housing condition from rearing background one focal male and female from the home cages were put in one indoor cage, measuring 2.6×1.55 m. In this cage 32 chicks represented the four experimental groups: there were eight chicks per group. These chicks all lived under the same circumstances now but had different backgrounds (semi-commercial and semi-natural) and different propensities to feather peck (high peckers and low peckers).

At the same time the pecking behaviour of the chicks was observed the same way as in the home cages. Once a week the pecking behaviour of each chick was observed for fifteen minutes, i.e. thirty minutes per cage of origin. The reallocation experiment lasted five weeks. Afterwards the chicks were kept together: in week seventeen and eighteen their pecking behaviour was recorded again and in week twenty and 23 they did an open field test. Appendix B shows a time schedule in which an overview of the complete experiment is visible.

During the reallocation experiment we did four behavioural tests. The first test was the open field test with a novel object to measure the behavioural response to a standard stressor. In the open field tests that took place at the beginning of the reallocation experiment the chicks were put in a circular arena with a diameter of 1 m and a height of 0.5 m. After five minutes a novel object was lowered into the arena: a soft, grey toy seal, a bit smaller than the chicks. It hang on a long thread with which it could be moved up and down from a distance. The novel object was kept in the cage for five minutes. During both periods calls and the time spent walking were scored.

This test took place just before the chicks were put into their new cage (they were four weeks old then) and just before the reallocation experiment ended (at nine weeks of age). To compare the results from the open field tests of the reallocated group, the behavioural response to an open field test was also recorded for the focal chicks that remained in the original situation. The tests was performed four days and one day later respectively and did not include a novel object challenge after five minutes.

To preserve the aspect of novelty, the open field condition was changed as the test was repeated at the end of the reallocation experiment. The open field had a octangular shape (but the same area and height), was located in a different room; a match box was the novel object.

In the second behavioural test competition behaviour was measured. At six weeks of age mealworms were introduced to the chicks and after that a mealworm competition was organized. Two chicks of opposite strain but the same rearing background were put together in a wire mesh arena with an area of 1 m^2. It was placed in the cage with the other chicks, so the competing chicks could hear and see their cage mates. Five times three mealworms were offered by hand; each time it was scored which chick won the competition (eat most mealworms), which one was the first to eat a worm and how many worms each of them ate.

The third test existed in presenting the chicks in their cage two paper cards. The cards measured 10×15 cm and hung 10 cm apart on the wall 10 cm above the floor. One card was covered with feathers, the other with hemparade, the material used for ground cover. The cards were presented when the chicks were 6 weeks and when they were 9 weeks old. Directly after presenting the cards the number of pecks towards the feathers and the hemparade was scored for a period of 10 minutes. Also the time at which each chick pecked at the feathers and the hemparade for the first time was noted. Not all chicks took part at the same time: at six weeks we started with the males and at nine weeks with the females. In both cases chicks from the opposite sex were removed.

The last experiment was a tonic immobility test, which took place nearly at the end of the reallocation experiment, one day after the open field test. In random order each chick was laid on its back and held down for ten seconds. It was scored how long it took for it to get up. Also

the number of inductions were counted: the number of times a chick had to be laid down before it took longer then ten seconds to get up. There was no limit to number of inductions necessary nor a cut off time in righting time. The experiment was done near the cage, so the chicks could hear their cage mates.

A second group of chickens was reallocated to one new group at twenty weeks after hatching, when the home cage observations ended. This time the experiment lasted four weeks. The chickens were put in one of the home cages that had become available. The males were not included as they had become too aggressive: only the focal female from each cage was used. To create a group of the same size as in the first reallocation experiment sixteen non-focal females from the home cages were put in it: one female from each cage.

The pecking behaviour of the focal chickens was scored in the same way as in the home cages and the first reallocation experiment. To obtain thirty observation minutes per cage of origin again, each chick was observed for thirty minutes per week. This period was spread over two periods of fifteen minutes in each week, mostly between 9:00 and 15:00.

We did an open field test at the beginning and the end of the experiment. The first time all focal chickens that were observed in the home cages till then were put in the open field: one male and female per cage. Only the females that would take part in the reallocation experiment received a novel object after five minutes. To preserve novelty another open field was used than in previous open field tests. It was a box which measured 93×57×60 cm (l×w×h). The novel object consisted of three plastic green labels with a blue spot, made heavier by a piece of lead. The chickens that had taken part in the first reallocation experiment also did an open field test now, but without a novel object.

The second open field test, at the end of the second reallocation experiment, took place in the same box as at the beginning. The differences were that one of the white walls was painted in vivid colours and the arena stood in another part of the room. No novel object was presented. Except the females from the reallocation test there were no other chickens that did the open field test.

In the first and the last week of this reallocation experiment, the test with cards with feathers and hemparade took place. This happened in the same way as in the first reallocation experiment. The non-focal chicks were removed during the tests.

Another test existed in the introduction of an unfamiliar chick in the cage for fifteen minutes. The non-focal chicks were removed. The number of pecks from each individual towards the new chick were counted. Feather pecks, aggressive pecks, pecks towards the eye-region and pecks at the bill were scored. Also the latency-time to start pecking at the chick was noted for each pecking chick. To recognize the chick its wings were coloured green. The test was done in the first and the last week of the reallocation experiment: the second time another chick was used but its wings were green again.

Like in the first reallocation experiment a tonic immobility test took place. This happened 21 days after the reallocation experiment had ended. The experimental chicks and their non-focal cage mates were still together then, but they were in a cage that was twice as large as the cage they had lived in till then.

All data were analysed in Statistix for Windows and SPSS 9.0. In Statistix the data were transformed. Almost all measured variables were poisson distributed and were therefore transformed by the formula sqrt(X+0.5). The time the chicks spent walking in the open field test was measured as a proportion and therefore transformed by another formula: arcsin(sqrt(X/100)). In SPSS 9.0 the transformed data were analysed in univariate or repeated measures ANOVA, as is noted per dataset in the next chapter. The data obtained from the mealworm competitions were not transformed and analysed by a Wilcoxon signed rank test.

4. Results

4.1 Home cages

4.1.1 Pecking

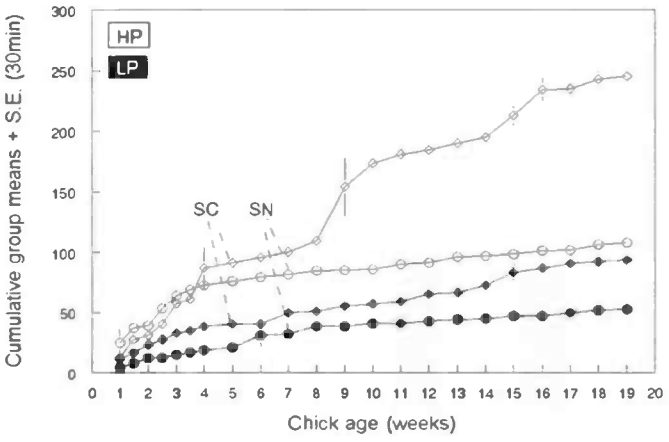


Fig. 2: Cumulative results of feather peck scores in the home cages.

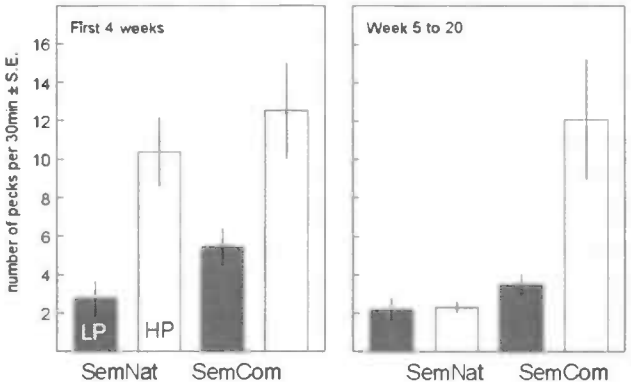


Fig. 3: Feather pecks in home cages: group means, divided over two periods.

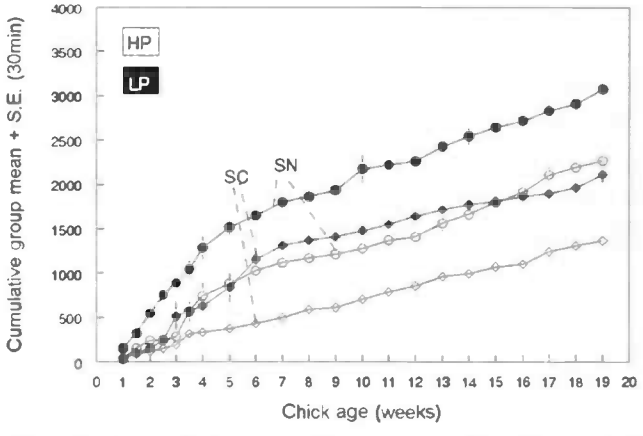


Fig. 4: Cumulative results of ground peck scores in the home cages.

Figure 2 shows the cumulative results of the scored feather pecks in the home cages. (Feather pecks are gentle and sever pecks added.) After four weeks the slope decreases in all groups except in semi-commercial high peckers. Especially because of many feather pecks in week 4, 9, 15 and 16 they end with the highest total amount of feather pecks. High peckers in both housing conditions end with more feather pecks than the low peckers; semi-natural low peckers showed the least feather pecking.

In figure 3 the mean results of feather peck scores in the home cages are shown divided over two periods: the first four weeks (the period before the first reallocation procedure) and the last fifteen weeks. In the first four weeks the high peckers pecked more than the low peckers (univariate ANOVA N=4 for all four groups; df=15; R^2=0.591; F=21.041; P=.001). There was no difference in the number of pecks between semi-commercial and semi-natural chicks.

In the last fifteen weeks the effect of strain persisted (LPSN: N=3; HPSN, LPSC, HPSC: N=4; df=14; R^2=0.533; F=4.794; P=0.051) and the semi-commercial chicks pecked more than the semi-natural ones (LPSN: N=3; HPSN, LPSC, HPSC: N=4; df=14; R^2=0.533; F=9.026; P=0.012. Per line the amount of pecking was independent of housing condition, but there was a trend towards an interaction effect: especially the high peckers caused the larger amount of pecks in the semi-commercial situation. (LPSN: N=3; HPSN, LPSC, HPSC: N=4; df=14; R^2=0.533; F=4.334; P=0.061).

Figure 4 shows the cumulative results of the scored ground pecks in the home cages. All groups except the semi-commercial high peckers started with much ground pecking and diminished it after six weeks. The semi-commercial high peckers kept their ground pecking at a rather constant rate which resembles the amount of ground pecking the other three groups showed as from six weeks. Therefore at last the smallest number of ground pecks was counted in semi-commercial high peckers. The semi-natural low peckers pecked most; the semi-natural high peckers and the semi-commercial low peckers both ended at the same amount of ground pecks.

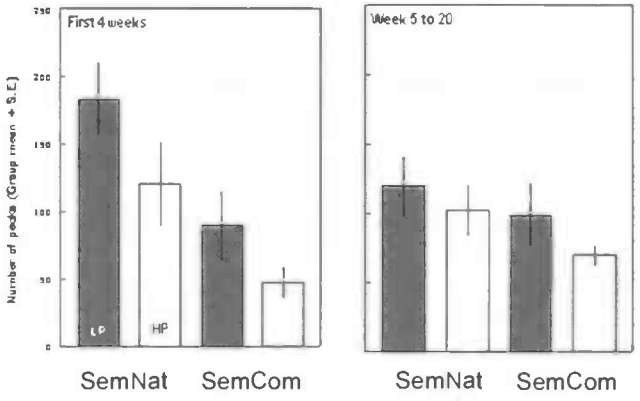


Fig. 5: Ground pecks in home cages: group means, divided over two periods.

Figure 5 shows the mean results of ground pecking rates in the first four weeks and the last fifteen weeks. Univariate ANOVA revealed that in the first four weeks the amount of ground pecking was independent of line, although there was a trend towards more ground pecking by low feather peckers (N=8; df=15; R^2=0.424; F=4.035; P=0.068). Semi-natural chicks pecked more at the ground then semi-commercial chicks (N=8; df=15; R^2=0.424; F=10.003; P=0.008). Per line the amount of ground pecking was not shown to be dependent of housing condition. In the period between five and twenty weeks after hatching ground pecking was equal in all groups.

4.1.2 One-zero sampling

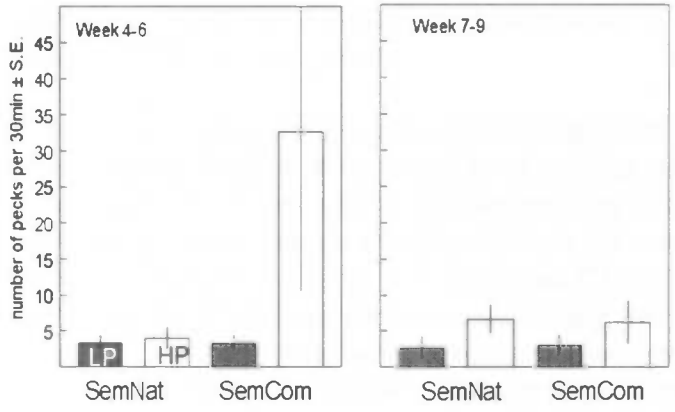
Correlations between feather pecking and other (pecking) behaviours were calculated by spearman rank correlations from the data obtained by one-zero sampling. We did this within the four experimental groups; the individual chicks were the sample units. Among semi-natural high peckers there was a significant negative correlation between feather pecking and aggressive pecking (N=6; P=0.0286). Among semi-natural high peckers feather pecking and ground pecking were correlated (N=10*; P=0.0061). Among semi-commercial low peckers feather pecking and dust bathing correlated (N=8; P=0.0235). Among semi-commercial high peckers no relevant correlations were found between feather pecking and other behaviours.

4.2 Reallocation experiments

4.2.1 Pecking

In figure 6 (next page) the scored feather pecks in the first reallocation period are depicted. The period is split up in the first three and the second three weeks. In table 5 the analysis by repeated measures ANOVA is depicted. Repeated measures ANOVA within subjects revealed no significant change in feather pecking between the periods. Repeated measures ANOVA between subjects only showed a trend towards high peckers pecking more at feathers than low peckers.

* One chick died during the period, so another was scored from then. In another cage a few times three chicks were scored. That is why the sample size is 10 instead of 8. The scored time was always half an hour per cage.



The two periods of three weeks were also analysed separately from each other with univariate ANOVA. In both periods of three weeks there were no significant differences between the lines and the conditions.

Fig. 6: Feather pecks during the first reallocation period, divided over two periods.

Table 5: Statistical analysis of feather pecking and ground pecking during the first reallocation period. LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8.

Variable	ANOVA rep. meas.	Source	Age	Type III Sum of squares	df	Mean Square	F	Sig.
Feather pecking	Within subjects contrasts	Age	Linear	5.605	1	5.605	1.377	.252
		Age•HP	Linear	2.015	1	2.015	.495	.488
		Age•SC	Linear	8.868	1	8.868	2.179	.152
		Age•HP•SC	Linear	13.433	1	13.433	3.300	.081
		Error(Age)	Linear	101.753	25	4.070		
	Between subjects effects	Intercept		440.844	1	440.844	60.788	.000
		HP		25.299	1	25.299	3.488	.074
		SC		2.501	1	2.501	.345	.562
		HP•SC		7.594	1	7.594	1.047	.316
		Error		181.304	25	7.252		
Ground pecking	Within subjects contrasts	Age	Linear	14.461	1	14.461	1.071	.311
		Age•HP	Linear	40.930	1	40.930	3.033	.094
		Age•SC	Linear	2.462	1	2.462	.182	.673
		Age•HP•SC	Linear	31.982	1	31.982	2.370	.136
		Error(Age)	Linear	337.409	25	13.496		
	Between subjects effects	Intercept		8911.947	1	8911.947	294.230	.000
		HP		82.496	1	82.496	2.724	.111
		SC		340.785	1	340.785	11.251	.003
		HP•SC		4.606	1	4.606	.152	.700
		Error		757.226	25			

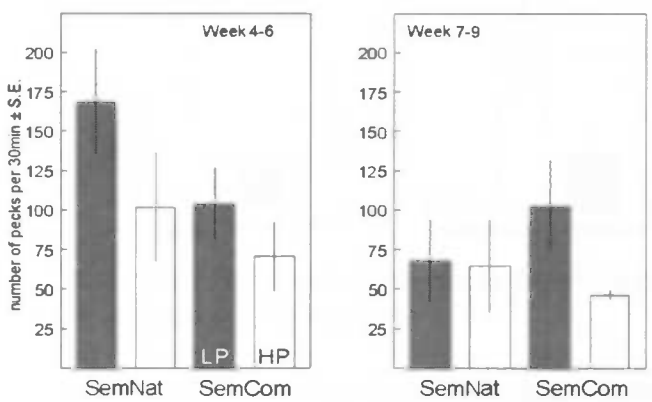


Fig. 7: Ground pecks during the first reallocation period, divided over two periods.

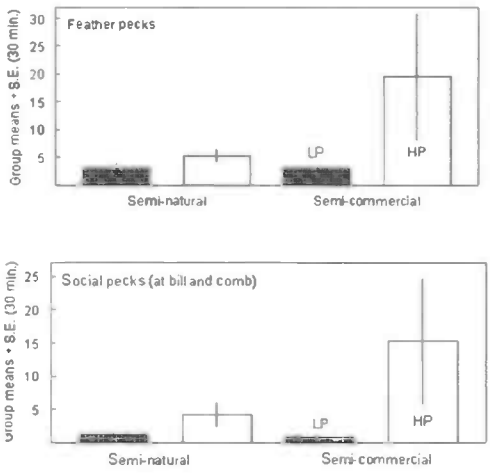
In figure 7 the ground pecks of the reallocated group are depicted. In table 5 the analysis by repeated measures ANOVA is shown. Repeated measures ANOVA within subjects revealed no significant change in ground pecking between the periods, only a slight trend towards low peckers pecking less in the second period. Repeated measures ANOVA between subjects revealed that semi-natural chicks pecked more at the ground than semi-commercial chicks.

The two periods of three weeks were also analysed separately from each other with univariate ANOVA. In the first period the

semi-natural chicks peck more at the ground than the semi-commercial ones (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8; df=28; R^2=0.349; F=12.627; P=0.002).In the second period this effect did not persist.

With a Pearson correlation test correlations were calculated between feather pecking and ground pecking, feather pecking and head pecking and feather pecking and social pecking in the first reallocation experiment. Feather pecking is the sum of gentle and severe feather pecking which were scored. Social pecking is the sum of bill pecking and pecking at weak parts (for definitions see table 3 or Appendix C1). In practice feather pecking consisted mostly of gentle feather pecking; social pecking was almost always bill pecking.

We found a strong correlation between feather pecking and social pecking (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8; F= 0.578; P=0.001). There was no correlation between feather pecking and ground pecking and between feather pecking and head pecking.



In figure 8 feather pecks and social pecks over the complete first reallocation period are shown (social pecks consist of pecks at bill and comb). Univariate ANOVA revealed no differences in amounts of social pecks or feather pecks between lines and conditions, but the patterns of both charts resemble each other strongly and as shown before there was a strong correlation between these pecking directions.

Fig. 8: Feather pecks and social pecks during the first reallocation period.

Figure 9 shows the scored feather pecks during the second reallocation procedure, divided over two periods of two weeks. Table 6 shows the statistical analysis by repeated measures ANOVA. Repeated measures ANOVA within subjects revealed no change in feather pecking between the first and the second two weeks. According to repeated measures ANOVA between subjects there were no differences in feather pecking between the lines and the housing conditions, there was only a trend towards high peckers pecking more often.

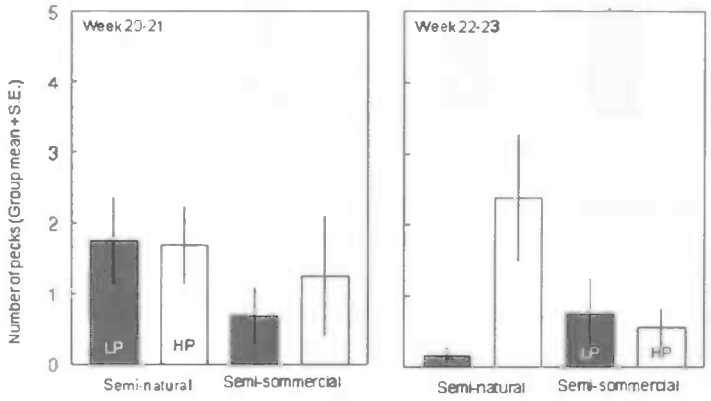


Fig. 9: Feather pecks during the second reallocation period, divided over two periods.

The feather pecking during the two periods of two weeks of the second reallocation procedure was also analysed separately by univariate ANOVA. No significant differences between the lines or housing conditions were revealed, there was only a slight trend that high peckers pecked more at feathers than low peckers during the second two weeks (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: *N=8*; df=14; R^2=0.359; F=3.294; P=0.097).

Table 6: Statistical analysis of feather pecking and ground pecking during the second reallocation period. LPSN: N=3: HPSN: N=4; LPSC: N=4; HPSC: N=4.

Variable	ANOVA rep. meas.	Source	Age	Type III Sum of squares	df	Mean Square	F	Sig.
Feather pecking	Within subjects contrasts	Age	Linear	2.161	1	2.161	1.581	.235
		Age•HP	Linear	.869	1	.869	.636	.442
		Age•SC	Linear	.127	1	.127	.093	.766
		Age•HP•SC	Linear	2.732	1	2.732	1.999	.185
		Error(Age)	Linear	15.033	11	1.367		
	Between subjects effects	Intercept		121.917	1	121.917	119.099	.000
		HP		3.752	1	3.752	3.665	.082
		SC		2.775	1	2.775	2.711	.128
		HP•SC		.410	1	.410	.401	.540
		Error		1.260	11	1.024		
ground pecking	Within subjects contrasts	Age	Linear	.199	1	.199	.004	.951
		Age•HP	Linear	8.057	1	8.057	.160	.697
		Age•SC	Linear	151.655	1	151.655	3.003	.111
		Age•HP•SC	Linear	111.650	1	111.650	2.210	.165
		Error(Age)	Linear	555.601	11	50.509		
	Between subjects effects	Intercept		6594.443	1	6594.443	180.478	.000
		HP		5.084	1	5.084	.139	.716
		SC		11.571	1	11.571	.317	.585
		HP•SC		1.484	1	1.484	.041	.844
		Error		401.927	11	36.539		

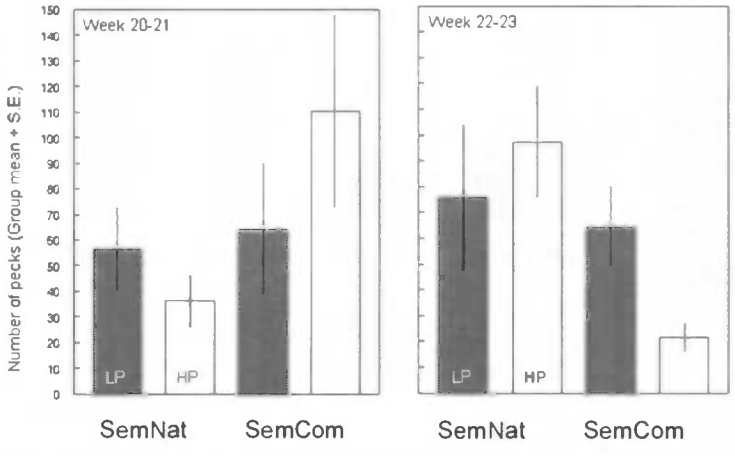


Fig. 10: Ground pecks during the second reallocation period, divided over two periods.

Figure 10 shows the ground pecking during the second reallocation period, divided over two periods of two weeks. In table 6 the statistical analysis by repeated measures are visible. Repeated measures ANOVA within subjects revealed no change in the amount of ground pecking between the first and the second two weeks. Repeated measures ANOVA between subjects showed no differences in ground pecking between the lines or the housing conditions.

The ground pecking during the two periods of two weeks of the second reallocation procedure was also analysed separately by univariate ANOVA. No significant differences between the lines or housing conditions were revealed, there was only a trend that semi-natural chicks pecked more often than the semi-commercial chicks during the second two weeks (N=7 for LP and SN; N=8 for HP and SC; df=14; R^2=0.425; F=4.568; P=0.056).

4.2.2 Competition for mealworms

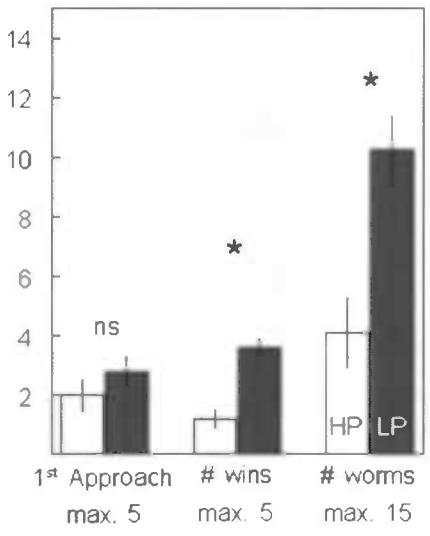


Figure 11 shows the results of the mealworm competition that took place during the first reallocation procedure. According to a Wilcoxon signed rank test there was no difference in the number of times both lines were the first to approach a mealworm. After five times three offered mealworms the low peckers mostly obtained the most mealworms (N=15; df=14; Z?=2.527; P=0.0115). Of the fifteen offered mealworms the low peckers eat more than the high peckers (N=15; df=14; Z=2.243; P=0.0249).

Fig. 11: Mealworm competitions between high peckers and low peckers in the first reallocation period. Which strain most often was the first to approach the worms, won most often and eat the most mealworms.

4.2.3 Interest in feathers and hemparade

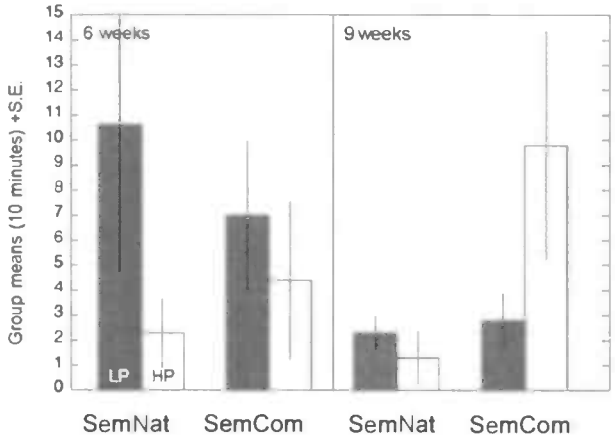


Fig. 12: Pecks at the feathers on a card during the first reallocation period, at six and nine weeks of age.

In figure 12 the number of pecks at feathers on a card is depicted. In table 7 the statistical analysis by repeated measures ANOVA is shown. Repeated measures ANOVA within subjects revealed no significant change in the amount of pecks at the card with hemparade, there was only a trend that low peckers pecked less the second time. Repeated measures ANOVA within subjects showed no significant differences neither between lines nor between housing conditions.

Table 7. Statistical analysis of pecks at feathers stuck on a card. LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8.

ANOVA rep. meas.	Source	Pecks at feathers	Type III sum of squares	df	Mean Square	F	Sig.
Within subjects contrasts	Age	Linear	40.214	1	40.214	.718	.405
	Age*HP	Linear	213.539	1	213.539	3.813	.062
	Age*SC	Linear	71.714	1	71.714	1.281	.269
	Age*HP*SC	Linear	12.847	1	12.847	.229	.636
	Error (age)	Linear	1400.104	25	56.04		
Between subjects effects	Intercept		1329.073	1	1329.073	18.009	.000
	HP		6.046	1	6.046	.082	.777
	SC		75.381	1	75.381	1.021	.322
	HP*SC		115.142	1	115.142	1.560	.223
	Error		1845.033	25	73.801		

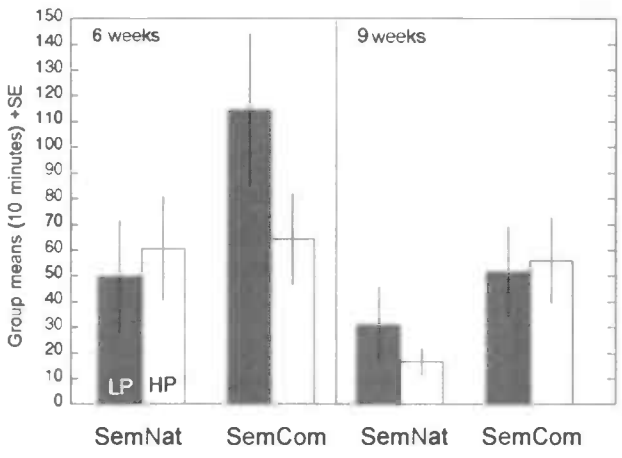


Fig. 13: Pecks at hemparade on a card during the first reallocation period, at six and nine weeks of age.

Figure 13 shows the number of pecks at hemparade on a card. In table 8 the statistical analysis is shown. Repeated measures ANOVA within subjects revealed that at nine weeks the pecking at the hemparade had decreased. This decrease was independent of line and housing condition, but per line the difference depended on housing condition. Repeated measures ANOVA between subjects showed no significant differences between the lines and conditions, only a trend towards the semi-commercial chicks pecking more often.

Table 8. Statistical analysis of pecks at hemparade stuck on a card. LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8.

ANOVA rep. meas.	Source	Pecks at hemparade	Type III sum of squares	df	Mean Square	F	Sig.
Within subjects contrasts	Age	Linear	17320.331	1	17320.331	15.811	.001
	Age*HP	Linear	1024.719	1	1024.719	.935	.343
	Age*SC	Linear	8.203	1	8.203	.007	.932
	Age*HP*SC	Linear	5078.591	1	5078.591	4.636	.041
	Error (age)	Linear	27385.711	25	1095.428		
Between subjects effects	Intercept		173542.972	1	173542.972	37.015	.000
	HP		2050.222	1	2050.222	.437	.514
	SC		15350.695	1	15350.695	3.274	.082
	HP*SC		1640.881	1	1640.881	.350	.559
	Error		117209.783	25	4688.391		

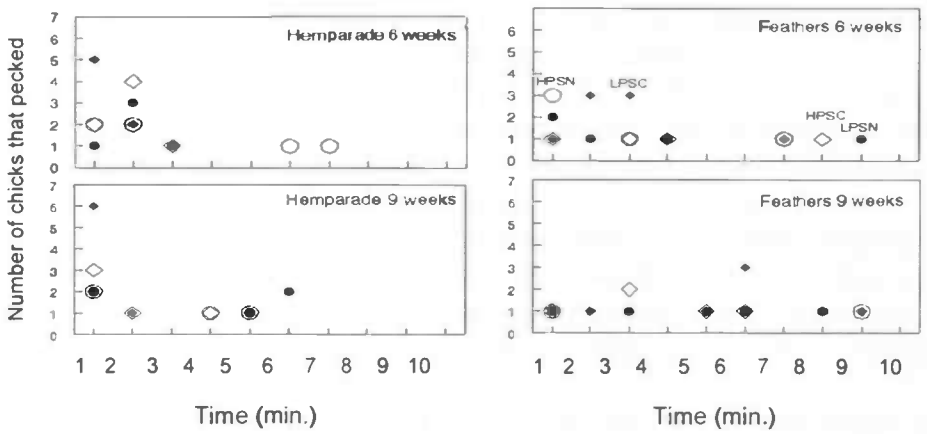


Fig. 14: When the chicks pecked at the feathers and hemparade on the cards during the first reallocation period.

In figure 14 is shown in which minute each chick pecked at the feathers and hemparade for the first time. At six weeks, when the test took place for the first time, seven chicks pecked at the feathers in the first minute; ten chicks pecked at the hemparade in the first minute. In total 22 chicks pecked at the feathers this time; 25 chicks pecked at the hemparade.

At nine weeks of age the experiment was repeated: then three chicks pecked at the feathers during the first minute whereas thirteen chicks pecked at the hemparade this minute. In total seventeen chicks pecked at the feathers during this experiment; 21 chicks pecked at the hemparade.

The experiment with the cards also took place during the second reallocation period, but as the chicks showed very little interest then, the results were not analysed. Also the experiment in which a strange chick was put in the cage during the second reallocation period was not analysed for this reason.

4.3 Coping with stress factors

4.3.1 Open field tests

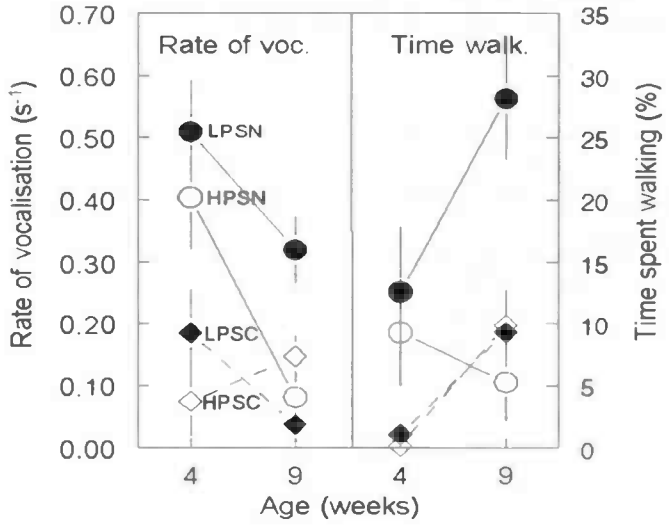


Fig. 15: Vocalisations and walking in open field: reallocated chicks before and after the first reallocation period.

In figure 15 the results of the open field tests at the beginning and the end of the first reallocation procedure are depicted. Table 9 shows the statistical analysis. Repeated measures ANOVA within subjects revealed that the rate of vocalisation was significantly reduced five weeks after reallocation. This reduction was not dependent of line, but on housing condition: semi-natural chicks lowered their calling rate more than semi-commercial chicks. There was also a 3-way interaction between line, condition and age, which means that the especially the semi-natural high peckers caused the lowering of pecking rate in semi-natural chicks.

Repeated measures ANOVA between subjects revealed that calling rate is independent of line (there was only a trend that low peckers called more), but semi-natural chicks uttered more vocalisations than semi-commercial chicks. The rate of vocalisation per line depended on housing condition.

According to repeated measures ANOVA within subjects the time that chicks spent walking increased five weeks after reallocation. This increase was independent of line and housing condition (although there is a trend towards more walking by semi-natural chicks). There was a 3-way interaction between line, condition and age.

Repeated measures ANOVA between subjects revealed a dependence of time spent walking on line and condition: low peckers walked more than high peckers and semi-natural chicks more than semi-commercial ones. The time spent walking per line depended on housing condition: among low peckers the different housing conditions caused larger differences than among high peckers.

Post-hoc the two open field test with the first reallocated groups were also analysed separately with univariate ANOVA (not shown in table 9). During the first open field test the semi-natural chicks had a higher calling rate (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8; df=28; R^2=0.490; F=22.129; P=0.000). The semi-natural chicks also spent most time

walking during the first open field test (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8; df=28; R^2=0.327; F=15.941; P=0.001).

During the second open field test with these chicks, at nine weeks after hatching, semi-natural chicks still had a higher calling rate than the semi-commercial ones (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8; df=28; R^2=0.539; F=9.547; P=0.005). There was a slight trend towards low peckers calling more often (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8; df=28; R^2=0.327; F=3.026; P=0.094). Per line it mattered whether it had a semi-natural or semi-commercial background: high peckers called more if they were semi-commercial and low peckers if they were semi-natural (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8; df=28; R^2=0.327; F=26.213; P=0.000).

The low peckers spent more time walking than the high peckers during the second open field test (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8; df=28; R^2=0.434; F=10.477; P=0.003). Per line it mattered whether it had a semi-natural or semi-commercial background: high peckers walked more if they were semi-commercial and low peckers if they were semi-natural (LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8; df=28; R^2=0.434; F=9.491; P=0.005).

Table 9: Statistical analysis of open field tests with reallocated chicks at 4 and 9 weeks. LPSN: N=6; HPSN: N=7; LPSC: N=8; HPSC: N=8.

Variable	ANOVA rep. meas.	Source	Age	Type III Sum of squares	df	Mean Square	F	Sig.
Calling rate	Within subjects contrasts	Age	Linear	62.396	1	62.396	6.932	.014
		Age•HP	Linear	1.641	1	1.641	.182	.673
		Age•SC	Linear	85.986	1	85.986	9.553	.005
		Age•HP•SC	Linear	91.101	1	91.101	10.121	.004
		Error(Age)	Linear	225.024	25	9.001		
	Between subjects effects	Intercept		2561.606	1	2561.606	241.762	.000
		HP		36.924	1	36.924	3.485	.074
		SC		277.831	1	277.831	26.221	.000
		HP•SC		55.960	1	55.960	5.281	.030
		Error		264.889	25	10.596		
Time spent walking	Within subjects contrasts	Age	Linear	.369	1	.369	14.426	.001
		Age•HP	Linear	.073	1	.0735	2.873	.103
		Age•SC	Linear	.085	1	.085	3.337	.080
		Age•HP•SC	Linear	.116	1	.116	4.530	.043
		Error(age)	Linear	.639	25	.026		
	Between subjects effects	Intercept		3.224	1	3.224	121.932	.000
		HP		.235	1	.235	8.893	.006
		SC		.355	1	.355	13.437	.001
		HP•SC		.144	1	.144	5.437	.028
		Error		.661	25	.026		

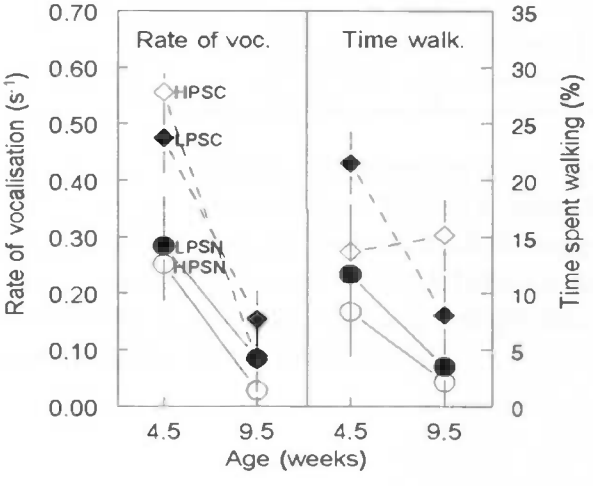


Figure 16 shows the result of the open field tests with home cage focal chicks at 4.5 and 9.5 weeks. Table 10 shows the statistical analysis of them. Repeated measures ANOVA within subjects revealed that the rate of vocalisation declined, independent of line or condition. There was no 3-way interaction between line, condition and age.

Fig. 16: Vocalisations and walking in open field: home cage focal chicks.

According to repeated measures ANOVA between subjects the rate of calling is equal in low peckers and high peckers but semi-commercial chicks call more than semi-natural chicks. Per line the results were not dependent of housing condition.

Repeated measures ANOVA within subjects revealed that at 9.5 weeks the chicks spent less time walking than at 4.5 weeks. In low peckers the time spent walking decreased more than in high peckers. The decrease was independent of housing condition ant there was no significant 3-way interaction.

The time the chicks spent walking was independent of line according to repeated measures ANOVA between subjects. Semi-commercial chicks walked more than semi-natural ones; per line the time spent walking was independent of housing condition.

The open field tests with home cage focal chicks were also tested separately with univariate ANOVA (not shown in table 10). During the open field test at 4.5 weeks of age the semi-commercial chicks had a higher calling rate than the semi-natural ones (LPSN: N=6; HPSN: N=8; LPSC: N=8; HPSC: N=8; df=29; R^2=0.344; F=12.962; P=0.001). The semi-commercial chicks also walked more than the semi-natural chicks in this open field test (LPSN: N=6; HPSN: N=8; LPSC: N=8; HPSC: N=8; df=29; R^2=0.217; F=4.426; P=0.045). During the second open field test, at 9.5 weeks, the semi-commercial chicks still called more often than the semi-natural chicks (LPSN: N=6; HPSN: N=8; LPSC: N=8; HPSC: N=8; df=29; R^2=0.173; F=4.242; P=0.050). They also spent more time walking (LPSN: N=6; HPSN: N=8; LPSC: N=8; HPSC: N=8; df=29; R^2=0.352; F=11.109; P=0.003).

Table 10: Statistical analysis of open field tests with home cage focal chicks at 4.5 and 9.5 weeks. LPSN: N=6; HPSN: N=8; LPSC: N=8; HPSC: N=8.

Variable	ANOVA rep. meas.	Source	Age	Type III Sum of squares	df	Mean Square	F	Sig.
Calling rate	Within subjects contrasts	Age	Linear	574.924	1	574.924	98.632	.000
		Age•HP	Linear	7.935	1	7.935	1.361	.254
		Age•SC	Linear	15.803	1	15.803	2.711	.112
		Age•HP•SC	Linear	.416	1	.416	.071	.791
		Error(Age)	Linear	151.553	26	5.829		
	Between subjects effects	Intercept		2811.209	1	2811.209	212.233	.000
		HP		.687	1	.687	.052	.822
		SC		156.540	1	156.540	11.818	.002
		HP•SC		2.799	1	2.799	.211	.650
		Error		344.392	26	13.246		
Time spent walking	Within subjects contrasts	Age	Linear	.253	1	.253	14.460	.001
		Age•HP	Linear	.0914	1	.0914	5.228	.031
		Age•SC	Linear	.0118	1	.0118	.674	.419
		Age•HP•SC	Linear	.0578	1	.0578	3.307	.081
		Error(age)	Linear	.455	26	1.749		
	Between subjects effects	Intercept		4.068	1	4.068	81.895	.000
		HP		.0077	1	.0077	.154	.698
		SC		.478	1	.478	9.621	.005
		HP•SC		.0055	1	.0055	.110	.743
		Error		1.291	26	.0497		

In figure 17 (next page) the results of the open field tests before and after the second reallocation period are shown. Table 11 shows the statistical analysis. Repeated measures ANOVA within subjects revealed no differences in calling rate between the tests at 20 and 24 weeks. Repeated measures ANOVA between subjects also showed no significant differences between the groups: there was only a slight trend towards semi-commercial chicks calling more often than semi-natural chicks.

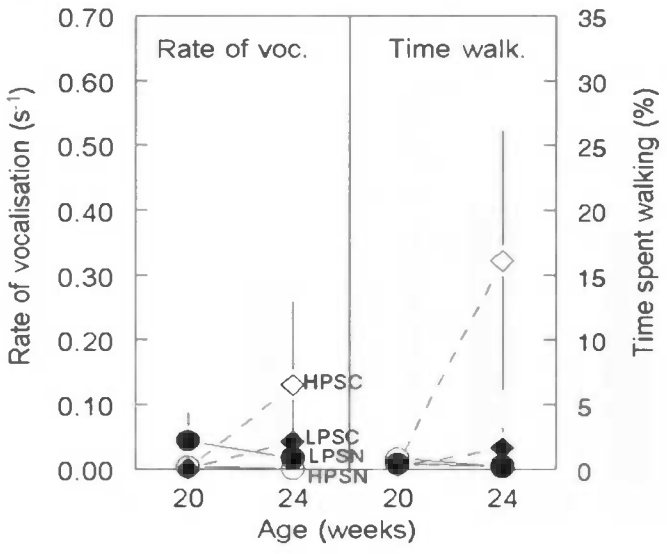


Fig. 17: Vocalisations and walking in open field: reallocated chicks before and after the second reallocation period.

According to repeated measures ANOVA within subjects there was no difference in time spent walking between the open field test before and after the reallocation procedure. Repeated measures ANOVA between subjects also showed no significant differences between the groups.

The both open field tests were also tested separately with univariate ANOVA (not shown in table 11). For both times no significant differences were found between the lines or the housing conditions, neither in calling rate nor in time spent walking.

Table 11: Statistical analysis of open field tests with reallocated chicks at 20 and 24 weeks. LPSN: N=3; HPSN: N=4; LPSC: N=4; HPSC: N=4.

Variable	ANOVA rep. meas.	Source	Age	Type III Sum of squares	df	Mean Square	F	Sig.
Calling rate	Within subjects contrasts	Age	Linear	10.314	1	10.314	2.106	.175
		Age•HP	Linear	3.438	1	3.438	.702	.420
		Age•SC	Linear	14.838	1	14.838	3.030	.110
		Age•HP•SC	Linear	1.473	1	1.473	.301	.594
		Error(Age)	Linear	53.870	11	4.897		
	Between subjects effects	Intercept		66.714	1	66.714	16.195	.002
		HP		1.781	1	1.781	.432	.524
		SC		13.574	1	13.574	3.295	.097
		HP•SC		3.902	1	3.902	.947	.351
		Error		45.315	11	4.120		
Time spent walking	Within subjects contrasts	Age	Linear	1.964	1	1.964	1.244	.289
		Age•HP	Linear	2.105	1	2.105	1.333	.273
		Age•SC	Linear	2.948	1	2.948	1.867	.199
		Age•HP•SC	Linear	3.164	1	3.164	2.003	.185
		Error(age)	Linear	17.371	11	1.579		
	Between subjects effects	Intercept		44.757	1	44.757	31.564	.000
		HP		1.240	1	1.240	.874	.370
		SC		4.010	1	4.010	2.828	.121
		HP•SC		1.002	1	1.002	.707	.418
		Error		15.598	11	1.418		

4.3.2 Tonic immobility tests

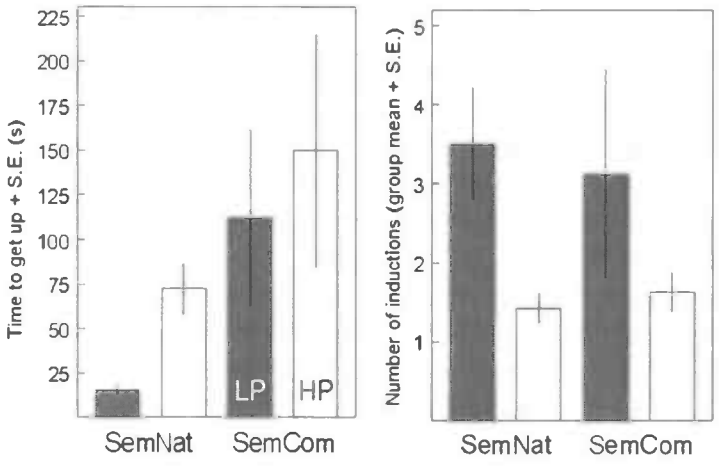


Fig. 18: Tonic immobility test after the first reallocation period.

Figure 18 shows the results of the tonic immobility test at the end of the first reallocation procedure. On the left the righting times after induction are visible; on the right the number of inductions before it took longer than ten seconds for the chick to get up. Univariate ANOVA revealed that the time to get up was independent of line, but semi-natural chicks needed less time to get to their feet (N=8; df=28; R^2=0.794; F=4.740; P=0.039). The number of inductions however, was independent of rearing condition but low peckers needed more inductions than high peckers (N=8; df=28; R^2=0.926; F=5.720; P=0.025).

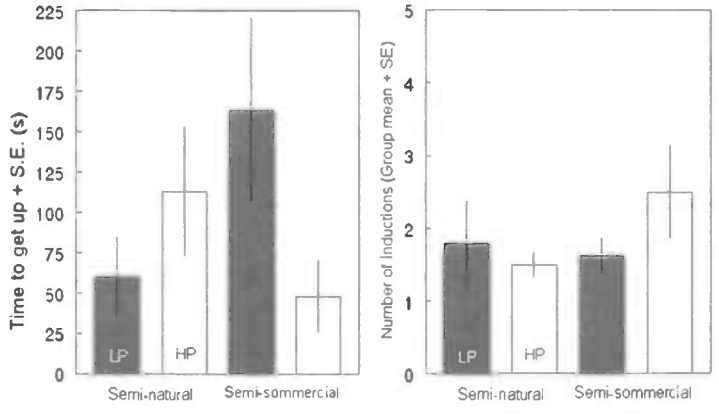


Fig. 19: Tonic immobility test after the second reallocation period.

In figure 19 the results of the tonic immobility test after the second reallocation procedure are depicted. The left chart shows the righting times; the right one shows the number of inductions needed to induce a tonic immobility of more than ten seconds. According to a univariate ANOVA test there were no differences in righting times between the lines or housing conditions but per line the righting times depended on rearing condition. Low peckers needed more time if they had come from semi-commercial conditions, whereas high peckers took longer if they had come from semi-natural conditions (LPSN: N=5; HPSN: N=8; LPSC: N=8; HPSC: N=8; df= 28; R2=0.085; F=4.401; P=0.046). The number of inductions was statistically equal for the four groups.

5. Conclusions

In the home cages semi-commercial high peckers feather pecked most. During the first four weeks the semi-natural high peckers feather pecked more than the last fifteen weeks. Therefore the semi-natural condition seems to influence the pecking behaviour of high feather peckers. In both periods the high feather peckers pecked more than the low peckers. In ground pecking no line effect was revealed, but the semi-natural chicks ground pecked more than the semi-commercial ones. After four weeks this difference had disappeared.

After the first reallocation no difference in feather pecking between semi-natural and semi-commercial chicks was found, whereas in the home cages it started to develop at that time. The effect of housing conditions was therefore not persistent. During the second reallocation period no significant differences in feather pecking was found between neither the lines nor the housing conditions, but this was also the case in the home cages at that time.

Differences in ground pecking only existed in the first four weeks: semi-natural chicks pecked more at the ground than semi-commercial ones. There was a trend towards more ground pecking by low peckers. Blokhuis (1989) concluded that feather pecking and ground pecking were negatively correlated. From comparing the figures of feather pecking results with those of ground pecking during the same periods the same could be concluded, but correlations in individual chicks only showed this for semi-natural high peckers in the home cages. Correlations between feather pecking and other behaviours were not found.

In the experiment in which a card with feathers and one with hemparade stuck on it were hung in the cage of the first reallocated group neither line-effects nor condition-effects were found in the amounts of pecks.

During the mealworm competition in the first reallocation period the low feather peckers won most of the five trials in which three mealworms were offered and at last they had eaten more mealworms than the high peckers.

In the open fields test with the first reallocated group before and after the reallocation period the semi-natural chicks called more often than the semi-commercial ones. They also spent more time walking during the first open field test. During the second open field test the low peckers spent more time walking than the high peckers. In the second open field test the overall calling rate decreased; the time spent walking increased.

The results of the open field tests with home cage focal chicks at about the same time as the open field tests with the first reallocated group also revealed a decrease in calling rate between the repeats, but also a decrease in time spent walking, especially caused by the low peckers. In both open field tests the semi-commercial chicks called more often and spent more time walking than the semi-natural chicks.

Two open field tests also took place with the second reallocated group, before and after the reallocation period. No significant differences between the four groups were then found neither in calling rate nor in time spent walking.

At the end of both reallocation periods a tonic immobility test took place with the involved chicks. The first one revealed that semi-natural chicks were faster in getting up; this effect was not there in the tonic immobility test after the second reallocation procedure. In the latter low peckers needed more time if they had come from semi-commercial conditions, whereas high peckers took longer if they had come from semi-natural conditions. In the first test the low peckers needed more inductions than the high peckers. This effect had disappeared in the tonic immobility with the second reallocated group.

6. Discussion

6.1 Methodology

Besides the experimental differences between the semi-commercial and semi-natural condition (rearing conditions: egg-type, group size, presence of mother) we were forced to accept several other differences in housing conditions to exist. Especially the factor group size implied differences like dirtier cages and more disturbances in the large groups (see table 12). It is important to reflect the consequences of these different housing conditions on feather pecking and stress responses.

Table 12. Differences between semi-commercial and semi-natural cages.

Semi-commercial cages	Semi-natural cages
Chicks hatched from commercial eggs.	Chicks hatched from semi-natural eggs.
Group size: 45 chicks.	Group size: six chicks.
No hen present.	A silky hen present.
High density of chicks.	Low density of chicks.
Cage size 1, later enlarged to 4,5 m^2.	Cage size 4,5 m^2 during the complete experiment.
Two cage enlargements experienced.	No cage enlargements experienced.
Two breeding lamps present during first four weeks.	No breeding lamps present.
Heat provided by breeding lamps the first four weeks; after that by one red heat source per two cages.	Heat provided by hen and one red heat source per two cages from the beginning.
Light dark cycle: 12:12 hours by turning a 100 W lamp on and off.	Idem, but in the dark stage there is a 40 W lamp to prevent total darkness.
Food: only breeding crumbs and – after a few weeks – gravel.	Food: breeding crumbs and gravel and laying feed meant for the silky hen.
Cages are cleaned more often (once a week after ten weeks).	Cages are cleaned only a few times.
Cages are dirtier (in spite of the weekly cleanings).	Cages are cleaner.
Focal chicks are coloured more often (once a week).	Focal chicks are coloured about once in three weeks.
More disturbances because of cleaning, wounded chicks that had to be treated or removed and floods caused by dirt in the water pipes.	Fewer disturbances.

The density of chicks was higher in the semi-commercial cages than in semi-natural cages, whereas after four weeks the cage sizes were equal. We wanted the cages to be equal, and the difference in density contributed to the resemblance of natural and commercial situations. The higher density in the semi-commercial cages enlarged the chance that chicks met each other and therefore the chance that they feather pecked at each other. As feather pecking was equal for example in semi-commercial and semi-natural high peckers during the first four weeks, we conclude that chance did not play a role in feather pecking. The large density may have been a stressor to the chicks, but it is difficult to judge that as in the first open field test with chicks that were reallocated afterwards the semi-commercial chicks were most fearful, but in the open field test with home cage focal chicks a few days later the semi-natural chicks were most fearful. As these last chicks had just met several challenges (SN: two cage mates taken away; SC: cage enlarged) it is most probable that the first open field test, with reallocated chicks, was most representative. The semi-commercial condition therefore was more stressful.

To imitate commercial conditions we put the semi-commercial chicks in an area of 1 m^2 the first three weeks after hatching. After that the available area was enlarged twice until the complete cage was available just like in the semi-natural cages. The enlargements may have caused stress, as a new environment was presented. During the exploring of this new environment feather pecking may have diminished. It appeared that the chicks soon

habituated to the new environments, so acute effects probably disappeared quickly. Stress responses might be influenced more persistently, as the home cage focal chicks showed responses in the open field that were very different from those shown by the reallocated chicks a few days before. In between the last enlargement of the semi-commercial home cages had taken place.

The semi-commercial cages were dirtier than the semi-natural cages, especially as the chicks grew older. Ground pecking was therefore less attractive, which may have had influence on the feather pecking. We think that when ground pecking is made impossible or unpleasant, the chicks might compensate for this by feather pecking. With a pleasant ground to peck at, ground pecking and feather pecking are not related (see 6.2).

The semi-commercial chicks were disturbed by us more often than the semi-natural chicks because the semi-commercial focal chicks needed to be marked more often, we sometimes had to treat a chick and because of the necessity of cleaning the cages more often. One could accept this as a 'natural' characteristic of the commercial condition, but it is not one of the three factors we wanted to influence. It may have caused more stress. Furthermore, newly coloured chicks received feather pecks at the coloured parts. Therefore we made sure that colouring and also other disturbances took place at least a day before pecking scores or afterwards.

As laying hens like leghorns don't get broody another type was needed as a foster mother for the semi-natural chicks. We used silky hens because they get broody very easily. Figure 1 shows a silky hen. Silky hens look and behave different from leghorns. They are smaller, have different feathers and they behave tranquil and friendly. The silky feathers possibly were more or less attractive to peck at, which could influence the feather pecking at siblings and the total amount of feather pecks. The tranquil behaviour of the silky hens may have influenced the chicks: nervous individuals may have been tranquillised by it, which changed the stress responses.

In most studies on feather pecking only hens are used. Except the practical fact that we could not sex the chicks directly after hatching there was another, more important reason why the males were also studied. From the fact that there are strains that differ in feather pecking and stress responses it can be concluded that these characteristics have a genetic component. Therefore both sexes are important to study.

It should be noted that after about six weeks behavioural differences became visible between hens and roosters. The latter became more aggressive, and during the threatening and jumping feather pecking was impossible. Aggression may have caused stress in the involved males, but it is also possible that it diminished stress, as the males learned to cope with aggression and the stress it causes. In addition males looked different from females after several weeks: their feathers looked different. An interesting fact is that males from the high peckers strain sometimes looked very dishevelled for several weeks: their feathers grew in all directions and they had no feathers on their tails and backs. Instead the juvenile down still covered these body parts. Though not noted the differences between the feathers of the chicks may have influenced feather pecking. As we found no differences in feather pecking or stress responses between males and females, we are inclined to think that the described differences in behaviour and appearance had no important influence on our results.

Despite the small sample size several significant differences were found between the four experimental groups. There were four cages per experimental group; during the two reallocation periods there were respectively eight and four studied chicks from each group. In the home cages the cages were the sample units (so N=4 per experimental group); during the

reallocation procedures the individual chicks were the sample units (N≈8 and N≈4 respectively per experimental group). Because of the small sample size we cannot be completely sure about for example the correlation between feather pecking and ground pecking. The charts give the impression that much ground pecking correlates with relatively low amounts of feather pecks, but computing correlations from one-zero sampling revealed that only in semi-natural chicks in the home cages feather pecking and ground pecking were negatively correlated. Perhaps larger sample sizes would have yielded a more definite answer.

Except the small sample size there was another problem in the semi-natural cages. Not all sample units were equal, as the number of chicks was not equal in each cage. Table 13 shows how many chicks were present in the semi-natural cages before and after the first reallocation. It is visible that males and females were not represented in equal amounts, but as we did not find differences between males and females in feather pecking and stress responses we are inclined to think that this is not a problem. There were 29 instead of 32 chicks in the first reallocation procedure.

Table 13. Numbers of chicks present in semi-natural cages.

Cage	Week 1-4 (before first reallocation)			Week 5-20 (after first reallocation)		
	♂	♀	Total	♂	♀	Total
B5 (HP)	0	5	5	0	4	4
B6 (LP)	3	2	5	2	1	3
B7 (HP)	3	3	6	2	2	4
B8 (LP)	3	3	6	2	2	4
C1 (LP)	0	3	3	0	2	2
C2 (HP)	3	3	6	2	2	4
C3 (LP)	<3	>1	4	-	-	-
C4 (HP)	2	3	5	0	2	2

In order to separate the effect of housing condition from rearing background the reallocation experiments took place. Forced by the small sample size a large disadvantage of this reallocation was that the change was different for semi-natural and semi-commercial chicks. Semi-natural chicks exchanged a large cage with only five cage mates and a foster mother for a smaller cage with many cage mates and no foster mother. The semi-commercial chicks had already experienced a change in cage size before, had already had many cage mates and no foster mother. It was impossible to solve this problem, because otherwise it would have been impossible to achieve the aim of separating housing conditions from rearing background. A larger sample size would have allowed us to create a group like we did and in addition some groups with more semi-natural characteristics. For commercial poultry industry the group we formed was most relevant; this was also a reason for us to choose for one larger group.

In the behavioural test during the reallocation periods in which cards with feathers or hemparade were offered to the reallocated group we let about sixteen chicks peck at the cards at the same time. A problem of this procedure is that except the motivation to peck the factor competition played a role. In the first reallocation almost all chicks were very eager to peck and some chased others away. For that reason it might have been better to make small groups, for example of four chicks. A disadvantage of this would be that the situation in each group is different because of the differing individuals.

The experiment with the cards with feathers or hemparade was not analysed for the second reallocation period because there were very few chicks that were interested in the cards. Comparing the charts with the open field results at week 4-9.5 and 20-24 (figures 15, 16, 17) it is visible that also in the open field little action is recorded. It seems very difficult to find differences between chicks at twenty weeks of age as they did not react on stimuli given by us

(on which they previously had a very strong reaction to) very strongly anymore. Only the tonic immobility test revealed any differences.

During the home cage pecking scores each cage was studied for half an hour per week (in the first four weeks this happened twice a week). During both reallocation periods we scored half an hour per former cage. This implies that the half hours were divided over four focal chicks during the first four weeks in the home cages; after that over the two remaining home cage focal chicks. In the first reallocation procedure the half hours were divided over two reallocated chicks per cage (or one chick, as there were three semi-natural cages from which only one chick had been reallocated). During the second reallocation period only one chicken per former cage was studied, so these individuals were studied half an hour each.

In this report we limited ourselves to analysing the ground and feather peck scores from the home cages and the two reallocated groups, the one-zero sampling in the home cages, correlations between pecking orientations in the first reallocated group, the open field tests (without the novel objects), the cards with feathers or hemparade and the mealworm competitions in the first reallocated group and the tonic immobility tests after both reallocation procedures. Many more important analyses could have been done on the enormous amount of data that have been obtained, but in the available time it was impossible to do so. Unfortunately the blood samples from which corticosterone levels would be determined had not been analysed at the time this report was written. They could have given useful information about the stress responses during the open field tests, as the behavioural responses were very contradicting between home cage focal chicks and reallocated chicks.

6.2 Results

6.2.1. Pecking

As expected the high feather peckers feather pecked most during the twenty weeks the pecking scores in the home cages took place. After four weeks the semi-natural high peckers diminished their feather pecking: it reached the same level as the feather pecking by low peckers. The semi-natural condition therefore seems to influence the feather pecking behaviour, but it is unknown which of the many differences between semi-commercial and semi-natural condition is or are responsible for this influence. As the influence became clear after four weeks and not immediately it is probable that not the laying condition of the mother was responsible for it, but instead of that the social rearing conditions after hatching (small group size and presence of a silky hen).

One could think that the removal of two chicks per cage had an important influence on the feather pecking behaviour of the remaining semi-natural chicks because the two chicks formed a large part of the total population in those cages. In figure 2 however it is visible that also the semi-commercial chicks diminish their feather pecking after four weeks, and it is not probable that the removal of two chicks from a population of 45 chicks had much influence on the feather pecking behaviour.

The importance of a small group size has been shown in our experiment. In semi-commercial cages we often found chicks with bleeding tails caused by feather pecking: this started in cages with high peckers but later the low peckers followed. It was clear that when a wound was caused practically all other chicks pecking at the bleeding, 'abnormal' looking wound. It is logic that such wounds were not easily caused and worsened by the few other chicks that were present even though they feather pecked in the same amount as the semi-commercial chicks.

At about eighteen weeks of age however, there was one semi-natural cage (with two males and two females) in which the females and the silky hen were permanently hiding themselves out of fear from one of the males. He constantly tried to attack and copulate with his 'sisters' and they were severely bleeding.* The other male did not seem to notice. This emphasizes the large differences that exist between individuals, which are also visible in the large standard errors in some feather pecking scores. It is not clear what causes these differences, but a consequence of them is that even small groups of poultry must regularly be checked as even there large social problems are possible.

During the first reallocation no differences in feather pecking were found between the four groups, in contrast to the home cage condition: also the previously found line-effect was not significant. High peckers from both housing conditions halved their feather pecking to the level of the low peckers (about five pecks per thirty minutes). This might be because low peckers 'diluted' the pecking population. Recruitment may play a role in feather pecking: one or a few chicks recruit the others to peck, but in low peckers do not start this process and/or they are more difficult to recruit. Feather pecking therefore seems to have a social component.

During the first reallocation procedure correlations were computed between feather pecking and ground pecking, aggressive pecking and social pecking (pecks at bill and comb). A correlation was only found between feather pecking and social pecking. The negative correlation between ground pecking and feather pecking that Blokhuis (1989) found has not been proven in our study. Comparing the charts with feather pecking results with those with ground pecking results (figures, 2 and 4, 3 and 5, 6 and 7 for example) this correlation could be concluded, but in individual chicks this relation was only found in semi-natural chicks in the home cages. This could be due to a small sample size, but as feather pecking changes after reallocation and ground pecking does not, and we found an important social component in feather pecking, we are inclined to think that the relation between ground pecking and feather pecking is not very important.

Similarity that were found between the charts of feather pecks ground pecks (a visible negative correlation) was also found between the charts of feather pecks and social pecks during the first reallocation period (a visible positive correlation). However, the latter was also supported by a significant computed correlation. Together with the 'dilution-effect' caused by putting low peckers and high peckers together this points in the direction of an important social component of feather pecking. Social behaviour is probably guided by the mother, as is shown in former research (Roden and Wechsler, 1998) and by the recent experiment by Bernd Riedstra.

There is another argument for the fact that feather pecking is social behaviour. Whereas there was a trend that high peckers feather pecked more than low peckers during the first reallocation procedure this trend was not there in the pecks at feathers on a card. From figure 12 it even seems that low peckers are more interested in the feathers than the low peckers. From this can be concluded that feathers on a card a totally different from feathers on a chick, which again implies a social component in feather pecking.

During the second reallocation period, like the first one, no significant differences in feather pecking were found between the four groups. There was only a trend towards high peckers pecking more. During the second reallocation period the semi-commercial high peckers feather pecked less than they did in the home cages. Again it is possible that this was caused by the dilution-effect, but figure 2 shows that in the home cages the larger amount of feather pecks by semi-commercial high peckers was especially caused in some particular weeks. A

* This highly aggressive rooster was removed from the cage and the wounds of the hens were treated.

decreased rate during four weeks seems therefore possible. Anyway we found no persistence of rearing background effects on the feather pecking behaviour.

6.2.2. Coping

In the mealworm competitions during the first reallocation period in which a low pecker and a high pecker from the same housing condition tried to obtain mealworms both strains were the first to approach the mealworms equally often. The low peckers however won most trials and obtained most worms in the end. From this can be concluded that both strains are equally fearful to approach the experimenter but the low peckers are more competitive. As low peckers are more successful in competitive situations, they seem to be faster.

That low peckers are faster can also be concluded from the results of the tonic immobility test after the first reallocation period. The low peckers need more inductions to lie down for more than ten seconds. After a successful induction they are not significantly faster to get on their feet. The time to get up is assumed to be related with fear (Jones, 1986): the longer it takes the more inhibited and the more fearful the animal is. According to the mealworm competitions and the tonic immobility test there is no difference in fearfulness between low peckers and high peckers about two months after hatching.

This result contrasts with the result Korte *et al.* (1997) found: their low peckers needed more time to get up and their corticosterone levels rose more then that of the high peckers. High corticosterone levels prolong tonic immobility. In the introduction (2.2) was shown that many factors influence tonic immobility, and it is unclear under which circumstances exactly the tonic immobility test took place in the experiment of Korte *et al.* (1997). It is possible for example that in their experiment the chicks could not hear their cage mates during the test, whereas thy could during ours. If low peckers are more sensitive to social isolation, this would probably have influenced the outcome.

The tonic immobility test after the first reallocation procedure did reveal that semi-commercial chicks need more time to get on their feet and therefore are more fearful. Whereas the differences in feather pecking between the four groups seem to disappear after reallocation (it could also be a consequence of the 'dilution-effect' by low peckers), differences in stress response are revealed in a tonic immobility test and are therefore caused by the rearing conditions rather than housing conditions, as housing conditions had been equal the last five weeks.

After the second reallocation period however, no differences in numbers of inductions were found between the groups. In time to get up an interaction effect was found: low peckers needed more time if they had come from semi-commercial conditions, whereas high peckers took longer if they had come from semi-natural conditions. Apparently the semi-natural conditions make high peckers more and low peckers less fearful after twenty weeks, or the semi-commercial situation is responsible in the other way round. It is also possible that domination plays a role at this age; the semi-commercial high peckers or the semi-natural low peckers might be dominant and therefore be less fearful and inhibited to get up.

In the open field test just before the first reallocation the semi-natural chicks uttered more vocalisations than the semi-commercial chicks. Are they less inhibited (fearful) or are they fearfully calling for their mother, who normally responds to them? After five weeks in the reallocated situation however, their calling rate has lowered. Time spent walking is a less dubious variable. During the first and the second time in the open field the semi-natural chicks spent more time walking, so the effect of their background was persistent on their fearfulness.

The open field results from the home cage focal chicks at 4.5 and 9.5 weeks contradict the results from the reallocated chicks. The time between both first times was only a few days,

but the semi-commercial chicks called more often and spent more time walking. These results were also found five weeks later. It has to be noted that during the few days between the first open field tests with the reallocated chicks and the home cage focal chicks important changes had taken place in the home cages. The semi-natural chicks missed two cage mates (a large proportion of the total population) and the semi-commercial cages had been enlarged. Because of these challenges it is most probable that the first open field test with reallocated chicks was most representative. The semi-commercial condition therefore probably was more stressful. It seems however that many factors influence the responses in the open field, so they are difficult to interpret. In spite of that it has been proven that persistent differences exist between stress responses in semi-commercial and semi-natural chicks.

From the open field tests before and after the second reallocation no significant differences between the experimental groups were revealed. Figure 17 shows that all chicks uttered very few vocalisations and spent little time walking. As noted before it is very difficult to reveal differences in twenty weeks old chickens as they hardly show responses in any test.

Our open field results prove that one has to be very careful to expose young animals to stress. The effects persist at least several weeks, and information obtained on a later age does not reveal much information. Therefore it is unknown whether these effects are still there then.

As we influenced three factors instead of one factor in our study we have done an explorative experiment, in which it has become clear that housing and/or rearing conditions influence feather pecking behaviour and stress responses. An example of interesting future research to find out the importance of a single factor would be to put chicks hatched from free range eggs in semi-commercial cages. If their background would diminish the feather pecking, the situation in poultry farms could improve by a relative simple solution. At the end of our experiment Bernd Riedstra experimented with chicks with or without a foster mother, and it seemed that the presence of a mother is important.

Summary of studied questions
- To which extent can the occurrence of feather pecking be influenced by the manipulation of laying condition and social rearing condition?
- What developmental mechanisms underlie feather pecking and what is its relation with coping style and sensitivity to stress?

In the first four weeks of our experiment it seems that none of the manipulated differences has any effect on feather pecking; only the line matters (high peckers peck more). Of the three other differences (egg type, mother present, group size) egg type had applied longer than the other two. If assumed that any difference between the groups should exert influence on feather pecking within a few weeks, one could think that social rearing condition has more influence on feather pecking than the laying condition of the mother, because the lowering effect of semi-natural conditions became visible after four weeks and not immediately, as group size and the present mother had (also) had time to exert influence.

As effects of semi-natural rearing conditions on feather pecking seemed not persistent after leaving these conditions, feather pecking in poultry farms probably cannot be diminished by putting the chicks in semi-natural cages for just a short period.

Probably a combination of group size and presence of a mother is needed to prevent damage by feather pecking. As feather pecking has an important social component the mother is probably needed to raise the chicks and teach them when to peck and where to peck at. Future research will have to show whether it is sufficient to for example put fifteen chicks with one foster mother in a small cage or six chicks without a mother.

The permanence of a small group is probably needed, as a small amount of feather pecks per chicks is enough to worsen coincidently caused wounds if the group is large. After all we have seen that when there is a wound all chicks peck at it. In small groups there is little

chance that wounds are caused and they are not worsened by thirty (thousand) other chickens. As we have also seen that very aggressive individuals exist (although in our case this was a male which in most barns are not present, but perhaps also hens exist with extreme aggressive or pecking behaviour) those small groups should be checked regularly.

The present study also revealed that feather pecking diminishes when semi-commercial and semi-natural chicks of the two lines are mixed in one group. This could also be important in solving the problem of feather pecking and cannibalism in poultry farms.

The other topic that was studied in this experiment was the relation between feather pecking and coping style and sensitivity to stress. Do certain factors influence feather pecking and coping style at the same time and in the same direction, which implies that less sensitivity to stress and less feather pecking go together? Whereas effects of housing conditions were not persistent for feather pecking, they were so for responses in open field tests and tonic immobility-tests after the first reallocation period. We found no statistical evidence for feather pecking and coping style being related. Whereas feather pecking differences disappear after reallocation, the influence of rearing conditions on stress responses are persistent in ten week old chicks.

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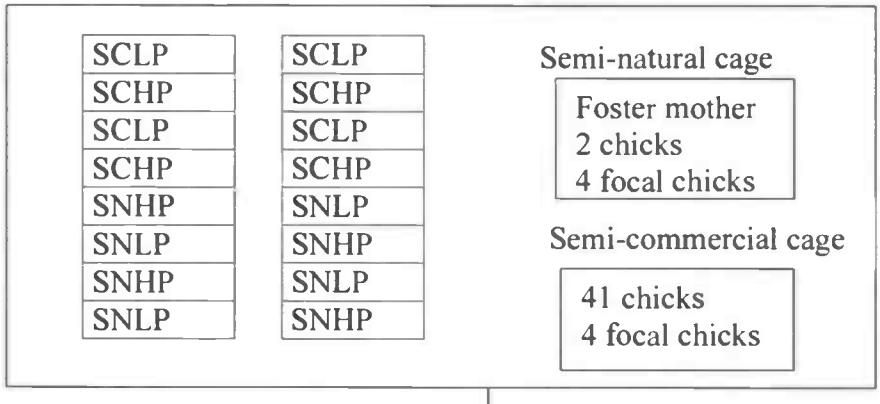
8. Appendix

A. Overview of the experiment

1. The four groups of chicks

	High feather peckers	Low feather peckers
Semi-commercial	**HPSC**	**LPSC**
Semi-natural	**HPSN**	**LPSN**

2. Experiment



Home cages

week 1-20 → Weekly pecking scores (during 30 minutes per cage)

Home cages

Reallocation procedures:
- Pecking scores
- (Repeated) behavioural tests
  Open field and novel object
  Competition for mealworms
  Cards with feathers and hemparade
  Introduction of strange chick
  Tonic immobility test

week 5-9 → From each cage:
1 focal ♀ + 1 focal ♂
= 32 chicks

Reallocatio procedure 1

week 20-24 → From each cage: 1 focal ♀
=16 chicks

Also 1 non focal ♀ per cage
= 32 chicks

Reallocation procedure 2

B. Time schedule

Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
17-21/10	0	Focal chicks are weighed and measured. Blood is taken to sex them.	Chicks are put in home cages; focal chicks are coloured.	Chicks hatch.
22/10	0		Vaccination of all chicks against Marek and New Castle disease.	
25/10	1	Pecking scores in two cages (see 8.3).	Colouring all focal chicks.	
26/10		Pecking scores in the rest of the cages.		
27/10				Dead chicks: B4 (1) and C7 (1).
28-29/10		Pecking scores home cages.	Colouring if needed.	Dead chicks: C6 (1); C5 (1); B3 (2).
1-2/11	2	Pecking scores home cages.	Cleaning semi-commercial cages (= putting in new hemparade).	Dead chick: B1.
3/11				Dead chick: C3.
4-5/11		Pecking scores home cages.	Colouring all focal chicks.	
8-9/11	3	Pecking scores home cages.	Colouring all focal chicks.	Dead silky hen: C1 (a new one is put in the cage). Dead chick: B5.
10/11		Weighing and measuring focal chicks. They receive a tag in their wing.		
11/11			first enlargement of semi-commercial cages.	
12/11		Pecking scores home cages.		
14/11				Flood in B4 (cage is cleaned).

Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
15/11	4	Pecking scores home cages.		Dead chicks: B5 (2).
16/11		Blood samples of group 1, to measure[corticosterone].	Colouring group 1.	
18/11		Group 1: open field test, novel object, weighing, blood samples for measurement of [corticosterone].	FIRST REALLOCATION Breeding lamps in semi-commercial cages are removed; heating is turned on there.	
19/11		Pecking scores group 1.	Second enlargement of semi-commercial cages; colouring the chicks.	
22/11	5	Pecking scores home cages.		
23/11		Pecking scores group 1.		
24/11		Open field test, weighing and colouring the two focal chicks in each home cage that are observed weekly. Group 1: attempt to test mealworm competition. Failed.		Flood in B3.
25/11		1/0 sampling of overall behaviour in home cages.	Injection of PHA in wing of one observed focal chick per cage.	
26/11			Group 1: cleaning cage. Measuring the swellings of PHA-injected wings (25/11).	
28/11				Flood in C8.
29/11	6	Pecking scores home cages.	Cleaning semi-commercial cages. Group 1: colouring.	

Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
30/11		Pecking scores group 1.	Repairing water systems in two semi-natural cages (B7 and B8). Taking blood and injecting sheep red blood cells (SRBC) in one observed focal chick per cage (the same one as on 25/11).	Dead chick: B2.
1/12			Colouring focal chicks in home cages.	
2/12		1/0 sampling home cages. Group 1: mealworm competition and cards with feathers and hemparade.	Injection of PHA in wing of one non-focal chick per semi-commercial cage.	
3/12			Semi-commercial chicks and group 1 receive extra hemparade. Measuring the swellings of PHA-injected wings (2/12).	
6/12	7	Pecking scores home cages.	Group 1: colouring. Taking blood from SRBC-chicks (30/11).	Dead chick: C3 (killed because it was lame).
7/12		Pecking scores group 1.	Injection of SRBC in the PHA-injected chicks on 2/12.	
8/12		Weighing and measuring group 1, focal chicks in home cages and the chicks firstly used on 2/12.	Cleaning two semi-commercial cages (C7 and C8).	
9/12		1/0 sampling home cages. Group 1: victim of feather pecking is introduced; pecks are counted for 15 minutes.	C7 receives extra hemparade.	Flood in B4. Dead chick: C8 (killed because it was pecked severely).
10/12			Taking blood from SRBC-chicks from 30/11.	Flood in B4.

Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
13/12	8	Pecking scores home cages. Group 1: introduction of silky hen. Pecks are counted for 30 minutes.	Colouring group 1 and focal chicks in home cages. Taking blood from SRBC-chicks from 7/12.	
14/12		Pecking scores group 1.		
15/12			Extra hemparade in all cages.	
16/12		1/0 sampling home cages.	Heating in home cages is turned lower.	
17/12			Taking blood from SRBC-chicks from 7/12.	
20/12	9	Pecking scores home cages. Group 1: introduction of silky hen (second time).	All home cages except C1, 2 and 4 receive new hemparade.	Flood in B4.
21/12		Pecking scores group 1. Group 1: cards with feathers and hemparade (second time).	Group 1: colouring.	Flood in B2 and C4.
22/12		Group 1: open field test, novel object, weighing, blood samples for measurement of [corticosterone] (second time).	END OF FIRST REALLOCATION PERIOD	Dead chick: C6 (killed because it was pecked severely).
23/12		1/0 sampling first half of home cages. Open field test, weighing and colouring the two focal chicks in each home cage that are observed weekly (second time).	Group 1: move to empty cage C3. Two males in B4 have pecking wounds; they are treated with anti-feather peck spray.	Flood in B1. Dead chicks: B4 (3; killed because they were pecked severely).
24/12		1/0 sampling second half of home cages.	Extra hemparade in all home cages. Four males in B4 receive anti-feather peck spray.	Flood in C6.
27/12	10			Flood in C1.

Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
29/12		Pecking scores home cages.		
30/12		1/0 sampling home cages.	Extra hemparade in semi-commercial cages.	Flood in C3.
2/1				Flood in B4.
3/1	11		Cleaning semi-commercial cages. Colouring focal chicks. Anti feather peck spray for: 1 female (C8), one male (C6) and one male (group 1, groenrug from cage).	Dead chicks: C4 (2; killed because they were lame. One focal male); C8 (1).
4/1		Pecking scores home cages.	Anti feather peck spray for one male in C6. Taking blood from the other weekly observed chick in each home cage then used on 25/11.	Flood in B8 and B6.
5/1		1/0 sampling home cages.		Flood in C8.
6/1			Injection of PHA in all observed focal chicks in home cages.	
7/1			Extra hemparade in all home cages. Anti feather peck spray for groenrug and tag 236 from group 1. Measuring swellings of PHA-injected wings (6/1).	
10/1	12		Cleaning semi-commercial cages and one semi-natural cage (B8); colouring semi-commercial focal chicks. Two males in B2 and three females in B3 are treated with anti-feather peck spray. New bulb in C1.	Flood in B1 and B2 (10 cm water in both cages!) Dead chicks: B1 (4, drowned); B4 (1: killed because it was lame).

Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
11/1		Pecking scores home cages.	Injecting SRBC in the same chicks that received PHA on 6/1	
12/1				Flood in B3.
13/1		1/0 sampling home cages.	Preventing Floods in semi-commercial cages by cleaning the water systems and boring holes in the walls to let water out.	Dead chick: B2.
14/1			Extra hemparade in all home cages.	
15/1				Dead chick: B2.
17/1	13		Cleaning semi-commercial cages. Taking blood from SRBC-chicks (11/1).	Flood in B2 and B3.
18/1		Pecking scores home cages.	Colouring focal chicks. Lately coloured necks are pecked at (especially blue ones), so in semi-commercial cages we only colour heads now.	Flood in B2 and C6.
19/1		Counting all chicks in semi-commercial home cages.	Anti feather peck spray for one male and female in B3.	
20/1		1/0 sampling home cages.	Anti feather peck spray for two males in C7, of which one focal. New bulb in C8.	Flood in C7.
21/1			Extra hemparade in semi-commercial cages. Anti feather peck spray for four males and one female (B1); one female (B3); one male (B4).	B4: one male dead.

Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
24/1	14		Cleaning semi-commercial cages; colouring semi-commercial focal chicks. Replacing the lamp in B6. Anti feather peck spray for seven males (B3); five males of which one focal (B1); one male (C5).	
25/1		Pecking scores home cages.		
27/1		1/0 sampling home cages.	Feather peck spray for one female in B1.	Dead chick: C8 (killed because it was lame); small flood in C6.
28/1			Cleaning water pipes in semi-commercial cages; clean hemparade added in all cages.	
31/1	15		Cleaning semi-commercial cages; colouring semi-commercial focal chicks.	
1/2		Pecking scores home cages.		Flood in C6.
4/2		1/0 sampling home cages (last time).		
7/2	16		Cleaning semi-commercial cages; colouring all focal chicks and group 1.	Dead chick (C6; killed because it was lame).
8/2		Pecking scores group 1 and home cages (first half).		
9/2		Pecking scores group 1 and home cages (second half).	Feather peck spray for one chick in C8.	
11/2		Cleaning water tubes and adding clean hemparade in semi-commercial cages.		

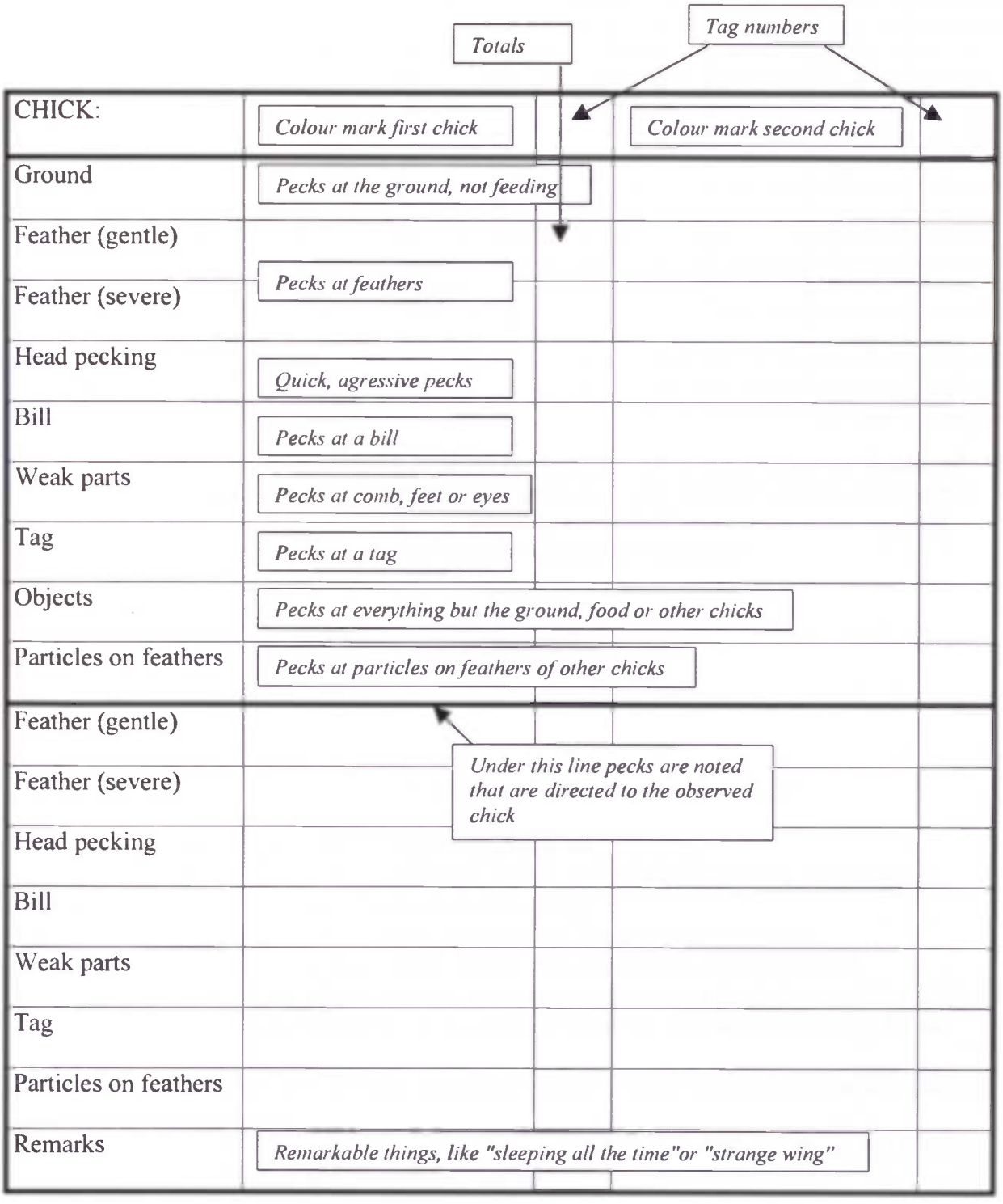
Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
14/2	17		Cleaning semi-commercial cages; colouring all focal chicks in semi-commercial cages and group 1. Feather peck spray for two chicks (C5, C6)	Severe flood in B4.
15/2		Pecking scores group 1 and home cages.	New bulb in B7 and C6.	
16/2			New bulb in B8. Feather peck spray for two chicks (C7, C8) Males removed in semi-commercial cages except C7 (52 chicks: five till ten per cage; in C6 one female).	Flood in C6. First eggs are layed.
17/2			Taking blood from focal chicks in home cages; injecting PHA.	
18/2			Colouring focal chicks and measuring swells in their wings. Clean hemparade added in semi-commercial cages. Feather peck spray for one chick in C8.	Flood in B4.
21/2	18		Cleaning semi-commercial cages; colouring all focal chicks in semi-commercial cages.	
25/2				Small flood in B2.
26-2			Seven non-focal chicks in home cages receive SRBC.	

Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
28/2	19		Cleaning semi-commercial cages and C2. New bulb in B5. Taking blood from Focal chicks and the seven non focal chicks in home cages. Feather peck spray for the focal chicks in B7.	Severe flood in C3 and B4; smaller flood in C2.
29/2		Pecking scores in home cages (last time).	Non focal male in B7 removed (too aggressive)	
2/3		Open field test group 1; measuring body mass, head-bill, tarsus and wing length. Blood is taken to measure corticosterone levels.		
3/3		Open field test home cage focal chicks. Females that will be followed in the new reallocated condition also receive a novel object and a new colour mark. Same measurements as yesterday.	SECOND REALLOCATION. (situated in cage B3)	
6/3	20		Cleaning the cage.	
7/3		Pecking scores.		
8/3		Cards with feathers and hemparade (first time).	Adding clean hemparade.	
9/3		Pecking scores. Strange chick in the cage (first time).		
13/3	21		Cleaning the cage.	
14/3		Pecking scores.		
16/3		Pecking scores.		
20/3	22		Cleaning the cage.	

Date	Age (weeks)	Observations and measurements	Interventions	Occurrences
21/3		Pecking scores.		
31/3		Pecking scores.		
3/4	23		Cleaning the cage.	
4/4		Cards with feathers and hemparade (second time); strange chick in the cage (second time). Open field test without novel object.		
25/4	26	Back test	END OF THE EXPERIMENT	

C. Observation protocols

1. Pecking scores



2. One-zero sampling

Date: Cage: Individual (tag): Observer: Time:

Behav. Minute	walk	run	stand	sit	fly	sleep	eat	drink	comfort
1									
2									
3									
4									
5									
6									
7									
8									
9									
10									
11									
12									
13									
14									
15									
total									

Above the left half of the observation protocol for one-zero sampling is depicted. In each of the fifteen minutes all showed behaviour components that were showed were marked. In the last row the numbers of minutes in which each component had been shown were summed. Down below, all behaviour components that were scored are described.

Walk: gentle walking.
Run: running through the cage.
Stand: standing still.
Sit: sitting or lying.
Fly: flying.
Sleep: sleeping
Eat: eating from the feeding troughs, or pecking at the ground very near to it.
Drink: drinking from the drinking pipe.
Comfort behaviour: cleaning feathers and stretching wings and feet.
Dust bathing: sitting and shaking with wings, so that the body gets covered with dust and hemparade. Mostly combined with ground pecking.
Alert: concentrating on the environment: other chicks, insects or the observer.
Freeze: instantly stopping any movement, mostly as a reaction on a sudden sound.
Distress call: screaming, mostly as a reaction on an attack or severe peck from another chick.
Jump or threaten: threaten another chick by jumping in the air, not always followed by an actual attack.
Aggressive peck: quickly and severely pecking at feathers, head or other body parts or pulling hard at feathers.
Ground peck: pecking at the ground. Also ground pecking during dust bathing is noted here; pecks very near the feeding through are scored as eating.
Feather peck: gently pecking and pulling at feathers.

D. Exact numbers of chicks in home cages on 19/01/2000

Cage	Group	Males	Females	Mother	Total chicks
B1	LPSC	20	17	-	37
B2	HPSC	22	18	-	40
B3	LPSC	25	12	-	37
B4	HPSC	15	18	-	33
B5	HPSN	0	4	1	4
B6	LPSN	2	1	1	3
B7	HPSN	2	2	1	4
B8	LPSN	2	2	1	4
C1	LPSN	0	2	1	2
C2	HPSN	2	2	1	4
C3	First reh. cond.	13	16	-	29
C4	HPSN	0	2	1	2
C5	LPSC	21	23	-	44
C6	HPSC	24	16	-	36
C7	LPSC	15	24	-	39
C8	HPSC	20	20	-	40

9. Epiloog

Deze epiloog schrijf ik in het Nederlands, omdat ik denk dat hem dat ten goede zal komen en omdat hij geheel los staat van dit verslag. Behalve de taal is ook de vorm anders: geen lijdende vormen meer! Ik heb mezelf er echt toe moeten zetten de rest van het verslag niet te populair te schrijven, omdat ik tijdens het grootste deel van mijn studie nu juist heb geleerd dat wel te doen. Hier wil ik op mijn eigen manier vastleggen hoe ik het afgelopen half jaar heb ervaren: wat ik ervan vond en wat ik heb geleerd.

Toen ik begon met deze stage, had ik eigenlijk geen idee wat wetenschap is. Als in een artikel een onderzoek werd beschreven, snapte ik nooit waarom de verschillende steekproeven niet altijd even groot waren. Nu weet ik het: er gaan beesten dood! Het is echt ongelofelijk hoe weinig voorstelling ik had van de onderzoekspraktijk. Na het vastleggen van duizenden kuikenminuten, het stelpen van enkele rampzalige overstromingen en het schoonmaken van stinkende kippenhokken, ben ik letterlijk met mijn neus op de feiten gedrukt. De semi-commerciële kuikens waren vaak vreselijk om te zien. Ik werd heen en weer geslingerd tussen gevoelens als 'smerige rotbeesten' en 'arme kuikentjes'.

Behalve dat er dieren doodgaan en onder vreselijke omstandigheden leven, kan ook de theoretische opzet van een onderzoek lang niet altijd zijn zoals je wilt. Ik heb gezien dat je vaak moet kiezen tussen twee kwaden. Zo hadden wij bijvoorbeeld onze 'standaard verenpik test', waarbij we uit elk hok twee kuikens haalden om samen één groep te vormen die we konden testen. Die groep was dus (ongeveer) 32 kuikens groot: voor de semi-natuurlijke kuikens een veel grotere overgang dan voorde semi-commerciële. Kleinere groepjes maken zou echter tot gevolg hebben gehad dat we weer meerdere situaties hadden, en dat was nu juist niet de bedoeling.

Zo'n methodologisch dilemma is leuk om over te filosoferen, vind ik. Het was erg goed om te discussiëren over wat nu de beste oplossing was. Waar ik me wel vreselijk aan heb geërgerd, is het eeuwige geldgebrek. Ik kan er niemand concreet de schuld van geven, maar ik baalde ontzettend toen 'onze' waterbuizen in die tochtige kippenhokken steeds overstroomden. Een fatsoenlijke oplossing was te duur, dus maakten we ze maar steeds schoon om verdere rampen te voorkomen. Wat dat betreft ga ik later liever ergens werken waar tijd óók geld is.

Het lijkt nu alsof ik alleen maar negatieve dingen heb geleerd bij het onderzoek, maar dat is zeker niet waar. Zoals gezegd vond ik het leuk te filosoferen over de beste manier om het onderzoek te doen. Verder leerde ik met ups en downs de vele data te verwerken tot grafieken en ze (tot op zekere hoogte) statistisch te analyseren. Ik ben blij dat ik dat nu een beetje kan, al vind ik dat dat laatste een beetje ondergesneeuwd is geraakt onder de vele andere dingen die moesten gebeuren. Naast het 'zien van de praktijk' was het leren en begrijpen van statistisch analyseren namelijk een doel dat ik voor mezelf had gesteld.

Al met al denk ik dat ik een stuk wijzer ben geworden van het afgelopen 'half jaar' (ondanks mijn sterke wil op tijd klaar te zijn toch nog uitgelopen), en ik vond het ook een erg leuke periode. De tijd is omgevlogen. Natuurlijk wil ik op deze plaats Bernd Riedstra bedanken. Veel van de dingen die ik de afgelopen periode heb geleerd, leerde ik van hem. Ik denk dat hij het ver gaat schoppen in de wetenschap. Hij is namelijk even intelligent als eigenwijs, en dat zijn volgens mij belangrijke eigenschappen voor een wetenschapper.Ook wil ik Ton Groothuis bedanken: de gesprekken met hem over de resultaten en methodologie van het onderzoek waren erg leerzaam.

Rianne Lindhout, juni 2000