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# Influence of nest site on the behaviour of laying hens

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#### ABSTRACT

Low rates of nest acceptance by laying hens are a major problem in commercial poultry farming operations with aviary systems, leading to costly manual collection and cleaning of mislaid eggs. To gain knowledge about factors affecting nest use, laying hens' preferences for different nest locations were tested. Nests are normally installed at one of two sites: against a wall of the hen house or integrated into one tier of the aviary rack. The preferences of laying hens for different nest sites have never been examined under commercial conditions. The aim of this study is to investigate whether behavioural differences can be detected between the different nest sites. The study consists of two consecutive trials involving 5027 Lohmann Selected Leghorn hens (LSL) and 601 layer hybrids selected for extensive housing conditions (EXT). The hens were randomly assigned to eight compartments per trial in groups of 355-360 LSL or 300 EXT in a laying hen house. Four compartments were equipped with a Volito Voletage® aviary system (VV), and four were equipped with a Rihs Bolegg®II aviary system (RB), both of which contained either integrated or wall-placed nests when the experiments started. A strongly balanced crossover design with four periods was used. At 36, 44 and 52 weeks of age, the nest site in four out of the eight compartments was switched. Before each change, the fronts of half of the nests were videotaped during the light period, and the behaviour throughout the main laying period was analysed. Furthermore, the numbers of nest eggs and mislaid eggs in each compartment were recorded every day. No differences in the number of mislaid eggs between the two nest sites could be detected, except at the age of 20/21 weeks when hens in VV aviaries mislaid more eggs when nests were integrated (P = 0.0012). More hens stood simultaneously in front of the integrated nests than in front of wall-placed nests (P=0.015). Activity of the laying hens increased (P = 0.0073), and stationary behavioural patterns declined (P = 0.0093), when the nests were placed by the wall. Hens inspected integrated nests for a longer duration than wall-placed nests, but wall-placed nests were visited more frequently. In addition to the nest site, the width of the platform in front of the nest influenced laying hen behaviour. Compared with narrower platforms, balance movements decreased on wider ones. Additionally, the platform design had to be taken into account as well, given that hens could not stand or walk as securely on wooden slats as on a grid floor.

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# 1. Introduction

Pre-laying behaviour, including searching for a suitable nest site and the inspection of potential nests, is one of the most important behavioural patterns in a hen's life and has barely changed throughout domestication (Nicol,

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2004; Kruschwitz, 2008). It begins up to 3 h before oviposition, with increased locomotion and inspection of several potential nest sites (Huber et al., 1985; Sherwin and Nicol, 1993; Cooper and Appleby, 1996). These behavioural patterns enable a hen to find an optimal place for incubation and hatching of young. Their natural environment provides a large variety of possible nest sites, whereas in commercial non-cage laying houses, like aviary systems, their choice is limited to one type of standardised roll-off group nest. Farmers expect that these nests meet the hens' requirements while avoiding eggs not laid into the nest boxes, called mislaid eggs thereafter. These eggs cannot be collected automatically and are often dirty. Mislaid eggs also present the added risk of the eggs being pecked and eaten.

To maximise nest use, laying hens' preferences for different nest designs have previously been tested to identify suitable laying nests for the majority of hens under commercial conditions (Buchwalder and Fröhlich, 2011). The quality of the nest floor (Huber et al., 1985; Appleby and Smith, 1991; Petherick et al., 1993; Struelens et al., 2008), nest seclusion (Appleby and McRae, 1986; Struelens et al., 2008) and nest colour (Zupan et al., 2007) are important factors for nest choice. Ontogenetic influences, such as nest experience early in life and rearing conditions (Cooper and Appleby, 1995; Colson et al., 2008), should be taken into account, as well as social interaction (Lundberg and Keeling, 1999) and the phenomenon of gregarious nesting; i.e. when an occupied nest is preferred over an unoccupied one. (Appleby et al., 1984; Appleby and McRae, 1986; Riber, 2010). Due to hormonal influences, egg-laying happens during a short time period of day and therefore pre-laying behaviour of different individuals is shown simultaneously (Appleby and Smith, 1991; Odén et al., 2002).

Little is known about the influence of the nest site on nest selection by laying hens. Duncan et al. (1978) described the laying behaviour of a population of domestic fowl established on an uninhabited island and found that hens chose nests of quite different types (e.g. with varying degrees of vegetative cover) for successive clutches but placed these successive clutches in the same area. Although discrimination between clutches is impossible under commercial conditions, hens tend to show a preference for certain nest sites. For example, consecutive eggs are laid in nests nearby each other (Rietveld-Piepers et al., 1985; Appleby et al., 1986), and high-placed nests and corner nests are favoured (Lundberg and Keeling, 1999; Riber, 2010). In conclusion, the location of nests within the housing system could be very important, offering hens the possibility to express their pre-laying behaviour and to find a vacant nest at the relevant time of day. Furthermore, preferences for suitable nest sites differ among individuals. Thus, consistent floor layers and consistent nest layers can be recognised (Cooper and Appleby, 1996, 1997; Kruschwitz et al., 2008; Zupan et al., 2008).

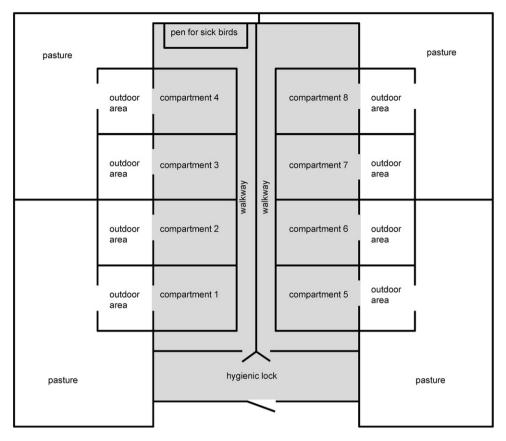
In aviary systems, two different nest sites are possible. Nests can be either installed outside the aviary rack against the wall (or, if large groups of hens are housed in the same barn and more than one aviary rack is present, between two racks) or integrated into the aviary rack (Fig. 1). Aviary systems provide different areas of activity by offering resources such as food, perches, litter and nests in



**Fig. 1.** Pictures of a compartment with the Rihs Bolegg<sup>®</sup>II aviary system to illustrate the two different nest sites. The aviary rack can be seen on the left side of the pictures. The upper picture shows treatment A (wall-placed nests available) and the lower picture shows treatment B (integrated nests available).

discrete areas. As a consequence, individuals performing specific behaviours in their respective areas are spatially separated from conspecifics performing other behaviours. Therefore, hens performing different behaviours do not disturb each other.

The variation in accessibility between different nest sites under commercial conditions has never been examined. The aim of this study is therefore to investigate whether behavioural differences in pre-laying behaviour can be detected when nests are placed by the wall vs. being integrated in the aviary rack. We assumed that the accessibility of nests would be higher for nests placed by the wall because only hens motivated to lay are assumed to frequent the nest area, whereas hens that are motivated to engage in other activities like feeding or resting would rather avoid the nesting area. It was expected that hens would move up and down the aviary rack in front of the integrated nests even when they are not motivated to lay. Therefore, we hypothesised that more hens would be found in front of integrated nests than in front of wall-placed nests. We also assumed that the more hens stood in front of the nests the



**Fig. 2.** Layout of the laying hen house. Compartments 1–4 were equipped with the Rihs Bolegg<sup>®</sup>II aviary system and compartments 5–8 with the Volito Voletage<sup>®</sup> aviary system.

poorer the accessibility of nests would be and the more eggs would be mislaid. Furthermore, the number of agonistic interactions may increase in front of integrated nests because hens searching for suitable nest sites are potentially disturbed by conspecifics that stop at nest sites while travelling between different tiers of the aviary rack.

#### 2. Material and methods

#### 2.1. Animals and housing

The study was conducted in two consecutive trials with a total of 5628 non-beak-trimmed hens, including 2167 Lohmann selected leghorn (LSL) and, due to another experiment, 601 layer hybrids selected for extensive housing conditions (EXT) in the first trial and 2860 LSL in the second trial. All chickens were raised from day one until 18 weeks of age in a breeding barn with eight separate compartments equipped with two different aviary systems (four compartments with 'Landmeco Harmony' from Landmeco A/S, Ølgod, DK; the others were 'Inauen Natura' from R. Inauen AG, Appenzell, CH). All compartments were equipped with perches, nipple drinkers, manure belts, automatic chain feeding, room heating and a humidifier. At 18 weeks of age, the hens of each specific compartment of the rearing house were collectively assigned into one of the eight compartments in a laying hen house, in groups of 355–360

LSL or 300 EXT. EXT were 2 weeks younger than LSL so they were moved at 16 weeks of age. The stocking density was 8.7 hens/m<sup>2</sup> accessible floor and 97 hens/m<sup>2</sup> nest space for LSL and 7.8 hens/m<sup>2</sup> accessible floor and 85 hens/m<sup>2</sup> nest space for EXT. A wall divided the building into two halves, each with four compartments. Walkways for animal keepers ran along both sides of the wall (Fig. 2). Four compartments were equipped with Volito Voletage® aviary systems (VV) (Glogobal AG, Tannlihag, CH), and four compartments were equipped with Rihs Bolegg<sup>®</sup>II aviary systems (RB) (Rihs Agro AG, Seon, CH). The compartments were separated by mesh wire from one another, and the floor of each compartment was covered with wood shavings and straw. Every compartment had a separate covered outdoor area, and four compartments had additional pastures.

Both aviary systems had standardised roll-off group laying nests placed along the wall. VV was equipped with eight 'Globogal Roll-Off Nests', and RB had six 'Vencomatic Classic Sidebelt Nests' per compartment. The nests were built in two tiers. To change nest sites, additional nests of the same colours and dimensions were built and integrated into the aviary racks where needed (Fig. 1). Nests along the wall were closed and made invisible with a wooden cover mimicking the wall when not needed, whereas integrated nests were physically removed when not needed. Every nest had a grid platform in front of the entrances, except for nests

#### Table 1

Nest sites at different ages (A: wall-placed nests; B: integrated nests).

Trial 1								
Aviary	Rihs Bolegg <sup>®</sup> II				Volito Voletage®			
Compartment	1	2	3	4	5	6	7	8
Hybrid	EXT	LSL	LSL	EXT	LSL	LSL	LSL	LSI
18-36 weeks of age	A	А	В	В	В	А	В	А
Switch nest site	Yes	No	No	Yes	No	Yes	Yes	No
37-44 weeks of age	В	А	В	А	В	В	А	Α
Switch nest site	No	Yes	Yes	No	Yes	No	No	Yes
45-52 weeks of age	В	В	А	А	А	В	А	В
Switch nest site	Yes	No	No	Yes	No	Yes	Yes	No
53-60 weeks of age	А	В	А	В	А	А	В	В
Trial 2								
Aviary	Rihs Bolegg <sup>®</sup> II			Volito Voletage®				
Compartment	1	2	3	4	5	6	7	8
Hybrid	LSL	LSL	LSL	LSL	LSL	LSL	LSL	LSI
18-36 weeks of age	A	В	А	В	В	В	А	А
Switch nest site	No	Yes	Yes	No	Yes	No	No	Yes
37-44 weeks of age	А	А	В	В	Α	В	А	В
Switch nest site	Yes	No	No	Yes	No	Yes	Yes	No
45-52 weeks of age	В	А	В	А	А	А	В	В
Switch nest site	No	Yes	Yes	No	Yes	No	No	Yes
53-60 weeks of age	В	В	Α	А	В	А	В	А

placed along the wall in the VV aviary system, which had two wooden slats instead. The width of the platforms was 30 cm in the VV aviary system and 60 cm in the RB aviary system. Housing conditions were standardised with a lighting schedule comprising a day length of 15 h (2:00–17:00 h) by using artificial lighting with a 15-min twilight phase at the beginning and end of the light period. Window curtains were lifted from 5:00 to 17:00 h to bring additional natural light inside the laying house. The room temperature was maintained at a constant 18 °C ( $\pm 2$  °C). A commercial standard layer diet and water were provided ad libitum.

#### 2.2. Experimental design

To achieve high statistical power by using each experimental unit as its own control, a two-treatment crossover design with four periods was used (Díaz-Uriarte, 2001). It consisted of four different sequences: AABB, BBAA, ABBA and BAAB, where treatment A represents a period when nests were placed by the wall and treatment B represents a period when nests were integrated into the aviary rack. Each treatment appeared the same number of times within each sequence and within each period, thus the design was balanced according to the criteria of Díaz-Uriarte (2001).

At 36, 44 and 52 weeks of age, the nest sites in four of the eight compartments were switched. Nests along the wall were replaced by integrated ones and vice versa. Time and number of changes between nest sites depended on the individual sequence of each compartment (Table 1). The assignment of sequences was chosen randomly, except that compartments with the same facilities (hybrid, aviary system, pasture) received complementary sequences. The switching of nest sites was completed within 4 h and began 8 h after the artificial light was turned on to ensure that hens had enough time to lay their eggs in the nests before they were removed. Because of the large number of mislaid eggs after switching nest sites in the VV aviary system compartments, the parts of the floor where most of the eggs were found were temporary closed (day 5–21).

#### 2.3. Data collection

Every day, the number of eggs per compartment laid in nests and the number of eggs per compartment mislaid on the floor or elsewhere in the aviary rack was recorded. Average laying performance was determined for periods of every 4 weeks after 20 weeks of age by counting the number of eggs per compartment per day and dividing it by the number of individuals present on that day. In the course of a single day during weeks of age 34, 42, 50 and 58, eggs laid in nests were collected every 30 min to determine the time of egg-laying within the first hours after illumination (main laying period). To avoid disturbing laying hens, only eggs from wall-placed nests were collected for main laying period determination because those eggs could be collected from the walkway.

At 35, 43, 51 and 59 weeks of age, half of the nests were filmed with digital cameras (Samsung 200X WDR Power Zoom) during the light period. Two cameras were installed in each compartment 1 day before filming started so the hens could get used to the equipment. Recordings were made with an artec Multieye-Hybrid Recorder<sup>®</sup> and stored on external harddiscs. Video recordings were analysed by continuous focal animal sampling and scan sampling (Martin and Bateson, 1993) using The Observer XT<sup>®</sup> (Noldus Information Technology, Wageningen, NL).

A total of 48 min per compartment per age throughout the 4 h of the main laying period were observed, consisting

#### Table 2

Definitions of recorded behavioural patterns. For point events only frequencies were counted, for state events also durations were recorded.

Behaviour	Properties	Definition
Avoid	Point event	Evasion movement after being threatened or pushed by another hen
Balance	Point event	Hen makes body movement including wing flapping; tail feathers are spread
Body movement	Point event	Body shaking or wing-leg-stretching or wing flapping
Exit	State event	The 3-min interval is over or the hen left the tier
Fight	State event	Two hens face to face trying to hack, peck or kick each other; neck feathers are spread
Hack	Point event	Peck/being pecked from another laying hen at head or comb
Leave tier	Point event	Hen leaves the tier within a 3-min interval; a new focal animal is chosen
Nest inspection	State event	Head inside the nest and both feet outside the nest
Nest visit	Point event	Head inside the nest and one food in contact with the nestfloor
Object peck	Point event	Pecking against equipment
Pacing	State event	Hen walking fast (>5 steps per 3 s) up and down; duration recorded after the hen changes the direction for the first time
Peck	Point event	Directed pecking against the body of another hen or being pecked
Push	Point event	Body contact; counted if one of the hens shows reaction i.e. walk, peck, balance, etc.
Preen	State event	Hen directs its beak to its own plumage of several body parts and carries out pecking, nibbling movements (stops <5 s included)
Sit	State event	Body touches surface and neck upright
Sleep	State event	Compliant with 'sit' but hen has its head tucked backwards into its feathers behind the wing
Stand	State event	Hen remains >3 s at the same place in an upright position; body does not touch the surface; duration recorded after 3 s
Threat	Point event	Neck stretched staring at another hen or hacking at another hen without touching; counted when other hen shows 'avoiding'
Walk	State event	Feet attaching alternately the ground; hen moving back-/forwards; duration recorded after the first step

of the first 3 min of every quarter of an hour. At the beginning of every 3-min interval, the individual standing in the centre of the observed area (as defined by the position of its feet) was chosen as the focal animal. When the focal animal left the observation area during the 3-min interval or a new 3-min interval began, a new focal animal was chosen in the same manner. Behavioural patterns are listed in Table 2, with some behaviours counted as frequencies (point events) and other behaviours recorded as durations (state events). Scan samples were made at the beginning of each 3 min to count the number of hens in front of the nests. Climate data were automatically collected with a HOBO<sup>®</sup> data logger (Onset Computer Corporation, Bourne, USA).

#### 2.4. Statistical analysis

Data were analysed using the repeated measures ANOVA in the statistical program NCSS (NCSS 2007, J. Hintze, Kaysville, Utah). For behavioural analysis, data from focal animals were averaged within each compartment and nest site. After being tested for normality (Kolmogorov–Smirnov test) and circularity (Mauchly test) to meet the assumptions of a Repeated Measures ANOVA, square root transformation of the data was performed if necessary to meet those requirements. Transformation was required for the number of balance movements, the number of nest inspections and the number of nest visits. The statistical unit used in all analyses was compartment. Aviary system (VV or RB) and hybrid (LSL or EXT) were included as fixed effects in the model; different nest sites (wall or integrated) and trial (1 or 2) were included as repeated factors. The first model with all main effects and the interaction terms was produced, omitting the interaction between hybrid and aviary system because EXT were housed in the RB aviary system only. EXT were present because of another experiment. They differed in many

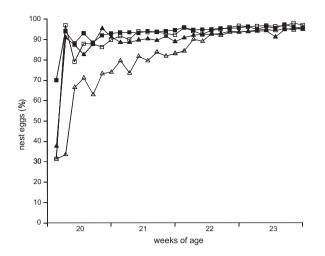
aspects and therefore were excluded from most analyses. The statistical model was adjusted accordingly by removing the fixed-effect hybrid and each affected interaction. Significance was never observed in multifactorial interactions; thus, results are not shown.

Results are reported as means and standard errors. In the case of square root-transformed data, raw data are presented. Durations of state events are presented in seconds out of a total of 2880 s (=48 min). Pace and walk were summarised as *moving behaviours*; sit, stand, sleep, preen and nest inspection were summarised as *stationary behaviours*; and push, pick, hack, avoid and fight were summarised as *agonistic interactions*. The proportion of mislaid eggs was analysed before the first change of the nest position (weeks of age 20 through 28) using the repeated measures ANOVA. Aviary system (VV or RB) and nest site (wall or integrated) were included as fixed effects, and age was included as a repeated factor in the model. A full model with all main effects and interaction terms was performed.

## 3. Results

### 3.1. Laying performance

At 19 weeks of age, the first eggs were laid. The laying performance continuously increased until a maximum of 95% was reached at 25 weeks of age. EXT were less productive; a maximum of only 85% was detected. At the age of 20/21 weeks, differences in the percentage of eggs laid in nests and mislaid eggs could be observed between nest locations while the hens habituated to the laying hen house and became familiar with laying (Fig. 3). The laying performance was in the range of 30–50% at that age. The percentage of mislaid eggs was affected by the interaction between nest sites and the aviary systems ( $F_{1,168}$  = 9.03, P=0.0132). Hens in VV aviaries mislaid more eggs when



**Fig. 3.** Percent of nest-eggs while hens became familiar with laying. Squares show average data of compartments with the Rihs Bolegg®II aviary; triangles show average data of compartments with the Volito Voletage® aviary (each dot represents 1 day). Black dots show percentages of nest-eggs laid in wall-placed nests; white dots show percentages of nest-eggs laid in integrated nests. A significant difference could be detected within 20 and 21 weeks of age.

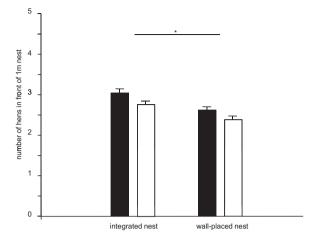
nests were integrated ( $F_{1,96}$  = 33.53, P = 0.0012). After 21 weeks of age, no differences in the percentage of mislaid eggs between the two nest sites could be detected.

Throughout the period of collecting behavioural data, hens showed a laying performance between 95% at 35 weeks of age and 90% at 60 weeks of age. The main laying period was identified as beginning 1 h after artificial lights were turned on and ending 4 h later, by which time a minimum of 80% of all eggs per day per compartment was laid.

#### 3.2. Behaviour

During the first trial, behavioural differences could be detected between the hybrids. In compartments with EXT, the animals showed fewer stationary behaviours  $(F_{1,16} = 8.15, P = 0.0462)$ . They tended to be more active  $(F_{1,16} = 4.93, P = 0.09)$  and less aggressive  $(F_{1,16} = 6.21, P = 0.0674; 27.84 \pm 6.2$  SE agonistic interactions (EXT);  $49.84 \pm 3.6$  SE agonistic interactions (LSL)). Furthermore, nest inspections of EXT were much shorter  $(F_{1,16} = 22.32, P = 0.0091)$ . The number of hens in front of the nests varied significantly between both hybrids  $(F_{1,16} = 165.53, P = 0.0002)$ . More hens were present in front of the nests in compartments housing LSL, on average  $2.87 \pm 0.07$  SE hens per running meter nest compared to only  $1.1 \pm 0.12$  SE EXT. Because of these significant differences, EXT were excluded from further analysis.

More LSL were present simultaneously in front of the integrated nests during the main laying period compared to nests placed against the wall ( $F_{1,36} = 11.52$ , P = 0.015; Fig. 4). The number of hens in front of the nest was also affected by compartment ( $F_{6,36} = 2.98$ , P = 0.018) and trial. More LSL were present in front of the nests during trial 1 than during trial 2 ( $F_{1,36} = 11.31$ , P = 0.0018).



**Fig. 4.** Number of hens in front of 1 running meter nest at the same time. Results are presented as mean (bars) with SE (lines on top of the bars). Black bars label trial 1; white bars label trial 2. Significance at \*P < 0.05; \*\*P < 0.01.

Results of the behaviour observations are presented in Table 3. LSL in RB systems performed longer nest inspections than LSL housed in VV aviaries ( $F_{1,36} = 42.54$ , P=0.0006). Nest inspections of LSL were influenced by nest site and trial. If integrated nests were present, LSL inspected them for longer durations than wall-placed nests  $(F_{1,36} = 44.63, P = 0.0005)$ . Inspections in trial 1 were of longer durations than in trial 2 ( $F_{1,36}$  = 8.32, P = 0.0066). Nest visits of LSL differed between nest sites. Wall-placed nests were visited more frequently in both aviary systems ( $F_{1,36}$  = 28.83, P = 0.0017). Balance movements of LSL were affected by the interaction between aviary system and nest site ( $F_{1,36}$  = 40.16, P = 0.0007). Increased balance movements were performed in the VV aviary system when the nests were placed against the wall. Exactly the opposite was found in the RB aviary system, where balance movements became more frequent if integrated nests were available. Activity of LSL increased and therefore stationary behavioural patterns declined when the nests were placed along the wall (moving behaviours:  $F_{1,36} = 15.81$ , P = 0.0073; stationary behaviours:  $F_{1,36} = 14.22$ , P = 0.0093). Regarding the activity level, differences between the trials could also be identified. During the first trial, LSL were more active (moving behaviours:  $F_{1,36}$  = 6.66, P = 0.0141; stationary behaviours:  $F_{1,36} = 6.80$ , P = 0.0131). Agonistic interactions were affected by an interaction between aviary system and trial ( $F_{1,36}$  = 8.82, P = 0.0053). No significant difference was found in the first trial, but in the second trial, significantly more agonistic interactions between LSL hens could be observed in the VV aviary system ( $F_{1,16}$  = 35.59, P = 0.0009).

# 4. Discussion

Both farmers and manufacturers of aviary racks with integrated nests often claim that integrated nests can be found more easily at the beginning of lay, resulting in fewer mislaid eggs because young hens do not need to leave the aviary rack to search for an adequate nest site. The results of

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Means, standard errors (SE) and significant differences among the observed behavioural patterns related to main effects and their interactions.

	Nest inspection Mean ± SE	Nest visit Mean ± SE	Balance Mean ± SE	Moving behaviours Mean ± SE	Stationary behaviours Mean±SE	Agonistic interactions Mean±SE
Aviary						
Bolegg	$771.41 \pm 32.51^{***}$	$8.25\pm0.94$	$1.75 \pm 0.69^{***}$	$341.34 \pm 10.33$	$2512.06 \pm 10.37$	$39.97 \pm 3.94^{*}$
Voletage	$500.66 \pm 28.16^{***}$	$9.28\pm0.82$	$9.47 \pm 0.60^{***}$	$357.13 \pm 8.95$	$2488.16 \pm 8.98$	$56.69 \pm 3.41^{*}$
Nest site						
Integrated	$771.81 \pm 28.23^{***}$	$6.50 \pm 0.66^{**}$	$4.88\pm0.40$	$284.31 \pm 22.08^{**}$	$2566.50 \pm 23.81^{**}$	$44.38 \pm 2.50$
Wall	$500.25 \pm 28.23^{***}$	$11.03 \pm 0.66^{**}$	$6.34 \pm 0.40$	$414.16 \pm 22.08^{**}$	$2433.72 \pm 23.81^{**}$	$52.28 \pm 2.50$
Trial						
1	$715.13 \pm 38.70^{**}$	$8.28 \pm 1.04$	$4.94\pm0.77$	$380.03 \pm 17.43^{*}$	$2468.94 \pm 17.46^{*}$	$49.84\pm3.02$
2	$556.94 \pm 33.52^{**}$	$9.25\pm0.90$	$6.28\pm0.66$	$318.44 \pm 15.09^{*}$	$2531.28 \pm 15.12^{*}$	$46.81\pm2.62$
Interaction						
Aviary x Nest site	ns	ns	***	ns	ns	ns
Compartment x Nest site	ns	ns	ns	ns	ns	ns
Aviary x Trial	ns	ns	ns	ns	ns	**
Nest site x Trial	ns	ns	ns	ns	ns	ns

Point events are given as counts, state events are given as durations in seconds and if based on transformed data they are presented as raw data. ns (Not significant).

\* Significance at *P* < 0.05.

\*\* Significance at *P* < 0.01.

\*\*\* Significance at *P* < 0.001.

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this experiment indicate the opposite. When hens started laying, more eggs were laid in wall-placed nests, whereas in compartments with integrated nests, the number of mislaid eggs was higher. As expected, the number of mislaid eggs increased after switching the position of the nests. Within 21 days, the hens seemed to get used to the new nest sites, no longer showing differences in the number of mislaid eggs compared to the period before the change (Lentfer et al., 2008). Thus, the 8-week period between the two data collection points was considered to be long enough. Animal keepers reported a considerable variation in the distribution of eggs from the different nests in each compartment. If nests were placed against the wall, more eggs were found in higher placed nests than in low-tiered nests, and when integrated nests were available, more eggs were laid in the ones facing the walkway than the ones facing the outdoor area. Corner nests at both nest sites, and the nests closer to the hygienic lock, seemed to be occupied most.

Moreover, nests placed along the wall were visited more often than integrated ones. Fewer hens stood in front of the wall nests simultaneously so that hens could perform more thorough nest-searching behaviours, including inspecting the nests and entering afterwards to decide whether a nest met individual prerequisites. By contrast, the accessibility of the integrated nests was limited because more hens were standing in front of the integrated nests at any given time. We assume that hens were not able to walk along the platform in front of the integrated nests. They primarily performed stationary behaviours, especially nest inspections of comparatively long durations and standing without any body movement. The phenomenon of gregarious nesting (Riber, 2010; Appleby and McRae, 1986) was directly observed during egg collection. One hen suffocated because too many hens were inside the same nest sitting on top of each other. Additionally, other conspecifics might have blocked the nest entry by standing in front of it, making it even more difficult for hens to move inside or outside the nest.

It is also possible that the hens behaved in a less active manner when integrated nests were present because of the combination of a high number of hens on the platform and the desire to avoid body contact between conspecifics while walking. Walking activity requires comparatively large inter-individual distances, and if available space decreases, walking frequency decreases with it (Keeling, 1995).

The present results also indicate that the aviary system, especially the design of nest platforms, influenced laying hen behaviour. In this experiment, hens experienced two different platform widths: a 30-cm-wide platform in the VV aviary system and a 60-cm-wide platform in the RB aviary system. As expected, hens walking on the 60-cm-wide platforms showed fewer balance movements in general, but balance movements occurred more frequently in front of integrated nests compared to nests along the wall. The reason may be that the increased density of hens in front of integrated nests resulted in more jostling between conspecifics. By contrast, when only 30-cm-wide platforms were available, hens showed more balance movements if the nests were placed along the wall. Because the 30-cmwide platforms in front of the wall-placed nests consisted of two wooden slats instead of a grid, the hens might not stand or walk as securely as on a grid floor because they could not grip the slats with their claws, resulting in more balance movements. Authors observed that this was especially pronounced where contact occurred with conspecifics. Considering the body size of a hen, avoidance behaviour to maintain with interindividual distances is nearly impossible on a 30-cm-wide platform, which may lead to increased body contacts and possibly increased agonistic interactions. In fact, more agonistic interactions were observed in the VV aviary system, which was equipped with 30-cm-wide platforms.

In accordance with previous studies (Odén et al., 2002; Eklund and Jensen, 2011), behavioural differences in the two hybrids were found. EXT showed fewer stationary behaviours, were more active and tended to show fewer agonistic interactions. Interpretation of these results is difficult because the highlighted behavioural differences could be explained by genetic variation or lower stocking density. It seems very likely that the increased activity of EXT was due to lower stocking densities in their compartments, with the result that fewer birds were in front of the nests, allowing more space in which to move and search for an appropriate nest.

#### 5. Conclusion

In laying hen farming with aviary systems, differences in laying hen behaviour can be detected according to the available nest site. Although the aviary system has an effect on the behaviour, we found that the nest site is of major importance for the hens. The design of nest platforms, especially the width, should be taken into account to provide adequate accessibility to nests where aviary systems are used at commercial farms. High animal densities on nest platforms, as is the case when nests are integrated into an aviary rack, require wider platforms in front of the nests to ensure appropriate pre-laying behaviour. Therefore, aviary systems with integrated nests should be equipped with platforms more than 30 cm in width.

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# References

- Appleby, M.C., McRae, H.E., Duncan, I.J.H., Bisazza, A., 1984. Choice of social conditions by laying hens. Br. Poult. Sci. 25, 111–117.
- Appleby, M.C., McRae, H., 1986. The individual nest box as a super stimulus for domestic hens. Appl. Anim. Behav. Sci. 15, 169–176.
- Appleby, M.C., Maguire, S.N., McRae, H., 1986. Nesting and floor laying by domestic hens in a commercial flock. Br. Poult. Sci. 27, 75–82.
- Appleby, M.C., Smith, S.F., 1991. Design of nest boxes for laying cages. Br. Poult. Sci. 31, 667–678.
- Buchwalder, T., Fröhlich, E.K., 2011. Assessment of colony nests for laying hens in conjunction with the authorization procedure. Appl. Anim. Behav. Sci., doi:10.1016/j.applanim.2011.06.005.
- Colson, S., Arnould, C., Michel, V., 2008. Influence of rearing conditions of pullets on space use and performance of hens placed in aviaries at the beginning of the laying period. Appl. Anim. Behav. Sci. 111, 286–300.
- Cooper, J.J., Appleby, M.C., 1995. Nesting behaviour of hens: effects of experience on motivation. Appl. Anim. Behav. Sci. 42, 283–295.

- Cooper, J.J., Appleby, M.C., 1996. Individual variation in prelaying behaviour and the incidence of floor eggs. Br. Poult. Sci. 37, 245–253.
- Cooper, J.J., Appleby, M.C., 1997. Motivational aspects of individual variation in response to nestboxes by laying hens. Anim. Behav. 54, 1245–1253.
- Díaz-Uriarte, R., 2001. The Analysis of Cross-Over Trials in Animal Behavior Experiments: Review And Guide to the Statistical Literature. Samizdat Press, http://samizdat.mines.edu.
- Duncan, I.J.H., Savory, C.J., Wood-Gush, D.G.M., 1978. Observations on the reproductive behaviour of domestic fowl in the wild. Appl. Anim. Ethol. 4, 29–42.
- Eklund, B., Jensen, P., 2011. Domestication effects on behavioural synchronization and individual distances in chickens (*Gallus gallus*). Behav. Process. 86, 250–256.
- Huber, H.U., Fölsch, D.W., Stähli, U., 1985. Influence of various nesting materials on nest site selection of domestic hen. Br. Poult. Sci. 26, 367–373.
- Keeling, L., 1995. Spacing behaviour and an ethological approach to assessing optimum space allocations for groups of laying hens. Appl. Anim. Behav. Sci. 44, 171–186.
- Kruschwitz, A., 2008. Evaluation des Legeverhaltens bei Legehennen und Untersuchungen zur Nestwahl unter Berücksichtigung der Motivation für den Nestzugang zu arbeiten. Inaugural-Dissertation, Institut für Tierhygiene und Öffentliches Veterinärwesen der veterinärmedizinischen Fakultät der Universität Leipzig und Bundesamt für Veterinärwesen, ZTHZ, Schweiz.
- Kruschwitz, A., Zupan, M., Buchwalder, T., Huber-Eicher, B., 2008. Nest preference of laying hens (Gallus gallus domesticus) and their motivation to exert themselves to gain nest access. Appl. Anim. Behav. Sci. 112, 321–330.
- Lentfer, T.L., Gebhardt-Henrich, S., Fröhlich, E., 2008. Changing nest positions: influence on laying behaviour of laying hens (*Gallus gallus domesticus*) in aviary systems. In: Erhard, M., et al. (Eds.), Association for Technology and Structures in Agriculture (KTBL), vol. 471. Current Research in Applied Ethology, Darmstadt, pp. 125–138.
- Lundberg, A., Keeling, L., 1999. The impact of social factors on nesting in laying hens (*Gallus gallus domesticus*). Appl. Anim. Behav. Sci. 64, 57–69.
- Martin, P., Bateson, P., 1993. Measuring Behaviour: An Introductory Guide. Cambridge University Press.
- Nicol, C., 2004. Development, direction, and damage limitation: social learning in domestic fowl. Learn. Behav. 32, 72–81.
- Odén, K., Keeling, L.J., Algers, B., 2002. Behaviour of laying hens in two types of aviary systems on 25 commercial farms in Sweden. Br. Poult. Sci. 43, 169–181.
- Petherick, J.C., Seawright, E., Waddington, D., 1993. Influence of quantity of litter on nest box selection and nesting behaviour of domestic hens. Br. Poult. Sci. 34, 857–872.
- Riber, A.B., 2010. Development with age of nest box use and gregarious nesting in laying hens. Appl. Anim. Behav. Sci. 123, 24–31.
- Rietveld-Piepers, B., Blokhuis, H.J., Wiepkema, P.R., 1985. Egg-laying behaviour and nest-site selection of domestic hens kept in small floor pens. Appl. Anim. Behav. Sci. 14, 75–88.
- Sherwin, C.M., Nicol, C.J., 1993. A descriptive account of pre-laying behaviour of hens housed individually in modified cages with nests. Appl. Anim. Behav. Sci. 38, 49–60.
- Struelens, E., Van Nuffel, A., Tuyttens, F.A.M., Audoorn, L., Vranken, E., Zoons, J., Berckmans, D., Ödberg, F., Van Dongen, S., Sonck, B., 2008. Influence of nest seclusion and nesting material on prelaying behaviour of laying hens. Appl. Anim. Behav. Sci. 112, 106–119.
- Zupan, M., Kruschwitz, A., Huber-Eicher, B., 2007. The influence of light intensity during early exposure to colours on the choice of nest colours by laying hens. Appl. Anim. Behav. Sci. 105, 154–164.
- Zupan, M., Kruschwitz, A., Buchwalder, T., Huber-Eicher, B., Ŝtuhec, I., 2008. Comparison of the prelaying behavior of nest layers and litter layers. Poult. Sci. 87, 399–404.