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Identification of key nest site stimuli for Japanese quail (*Coturnix japonica*)

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Abstract

For both egg production and laboratory research, Japanese quail (*Coturnix japonica*) are typically housed in battery cages without nest boxes. In such cages the quail hens show symptoms of pre-laying restlessness. The inability to perform normal pre-laying behaviour is regarded as one of the most important problems for the welfare of caged laying hens. The present study aimed at identifying key nest site stimuli for quails, to enable nest boxes to be designed for alternative housing systems. Groups of hens were kept in pens containing litter, nest boxes and dustbathing boxes. The type of nest box was varied between pens, and the incidence of eggs laid outside the nest boxes (floor-eggs) was recorded over periods of 15 days. In experiment 1 (8 groups of 30 hens), both the nature of the top of the nest boxes (closed or with slits) and the type of substrate in the nest boxes (artificial turf or hay) had a significant effect on the percentage of floor-eggs. Nest boxes with a top with slits and filled with hay were best accepted for laying. Nest boxes situated in the corners of the pens were significantly selected more often for egg laying than more central nests. In experiment 2 (8 groups of 14 hens), neither the nature of the sides of the nest boxes (closed or with slits) nor the colour of the nest boxes (green or brown) had a significant effect on the incidence of floor-eggs. In experiment 3 (16 groups of 14 hens with 2 or 3 cocks), the percentage of floor-eggs was significantly lower in pens with high (170 lux) than with low (15 lux) light intensity. There was also a tendency for nest boxes filled with chaff to be better accepted than nest boxes filled with hay. In experiments 2 and 3, in pens with floors half litter and half of perforated plastic more floor-eggs were found on the litter. Few eggs were laid in the dustbathing boxes in any experiment. This study shows that Japanese quail lay up to 90% of eggs in nest

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boxes provided these are properly designed. It should therefore be feasible to develop housing systems with nests for Japanese quail which may replace conventional battery cages. © 1998 Elsevier Science B.V.

Keywords: Japanese quail; Reproductive behaviour; Nest site selection

1. Introduction

When kept for egg production or as laboratory animals, Japanese quail (*Coturnix japonica*) are typically group-housed in cages arranged in batteries with several tiers (Yamashina, 1961; Cooper, 1987; Gerken and Mills, 1993). The cages do not contain nest boxes, and their floor is sloped to allow the eggs to roll out. Inability to perform normal pre-laying behaviour is considered as one of the most important problems for the welfare of laying hens (*Gallus domesticus*) in cages (Appleby and Hughes, 1991). Hens in battery cages show pre-laying restlessness (Wood-Gush and Gilbert, 1969; Ramos and Craig, 1988) and this behaviour is interpreted as indicating frustration caused by the lack of an adequate nest site (Wood-Gush, 1972; Brantas, 1980). As a consequence, alternatives to battery cages (Kuit et al., 1989) as well as modified cages containing nest boxes (Appleby and Hughes, 1995) have been developed, and the hens' preferences with respect to nest box design have been investigated in detail (Huber et al., 1985; Appleby and McRae, 1986; Duncan and Kite, 1989; Appleby and Smith, 1991; Reed and Nicol, 1992).

Quails housed in battery cages show symptoms of pre-laying restlessness as well (Gerken and Mills, 1993). To our knowledge, however, only one experimental study on nest site selection in Japanese quail has been published. Michel (1989) found that quails housed singly prefer cubicles with vertical grey stripes over cubicles with horizontal grey stripes and green over ochre cubicles for laying. In the wild, Japanese quail build their nests among the tufts of grass in the dry grassy lands (Taka-Tsukasa, 1967).

The aim of the present study was to identify key stimuli for nest site selection in Japanese quail. Groups of 30 hens (experiment 1), 14 hens (experiment 2) or 14 hens and 2 or 3 cocks (experiment 3) were kept in pens containing nest boxes. The nature of the substrate in the box, the presence of openings on the top or in the sides of the box, the colour of the box and the light intensity in the pen was varied between groups. As the present study is part of a project that aims at developing an alternative to battery cages for Japanese quail, the nest boxes were presented in enriched pens and not in cages. In this project, the design of nest boxes was only one objective among others (Schmid, 1997). As a consequence, several factors were altered simultaneously between the experiments described in the present study.

2. Experiment 1

Based on observations of quail nests in outdoor aviaries (unpublished data) and the description of nest sites in the literature (Stevens, 1961; Taka-Tsukasa, 1967; Orcutt and

Orcutt, 1976), we assumed that cover and substrate quality may be important factors for nest site selection in Japanese quail. In experiment 1, we therefore examined the effects of substrate type (hay or artificial turf) and the degree of cover provided by the nest box (top closed or with slits) on the incidence of floor-eggs. Both factors were varied independently in a 2 × 2 factorial design. After a first period of data collection, the nest boxes were exchanged between pens to check whether the use of a specific type of nest box is influenced by previous experience with a different type of nest box.

2.1. Methods

Eight groups of 30 female quails were kept in pens with a floor area of 2 × 1 m and a height of 0.5 m. The birds were of a strain used for both egg and meat production. They had been reared in cages on a commercial farm and were introduced into the pens when

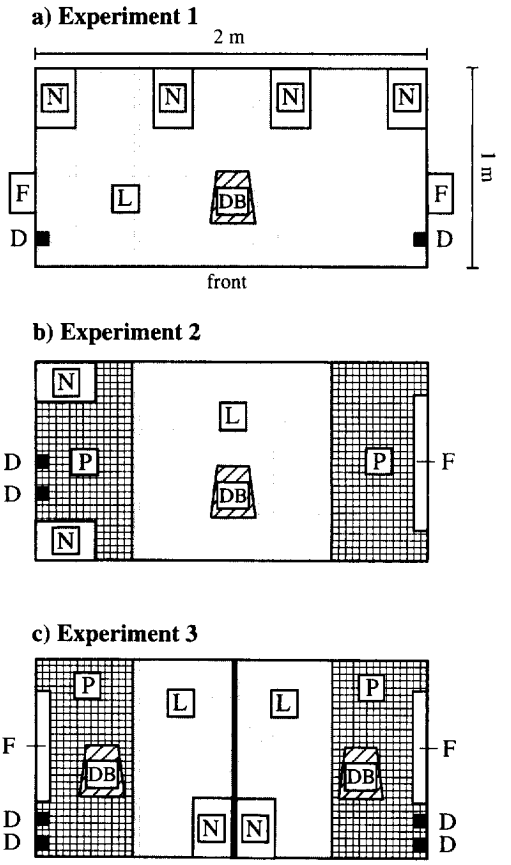

Fig. 1. Pen design and location of the nest boxes (N) in (a) experiment 1, (b) experiment 2 and (c) experiment 3. The pens contained feeders (F), drinkers (D) and a dustbathing box (DB). In experiment 1, the whole floor area was covered with litter (L). In experiments 2 and 3, part of the floor was made from perforated plastic (P). The quails could enter the nests on the narrow side of the nest boxes.

they were 8 weeks old. About one quarter of the quails had already started laying eggs at this age.

The pens were of wood, with solid walls on three sides and a solid floor covered with a mixture of wood-shavings and chaff. Both front and top were of wire mesh. Food and water were available ad libitum at each side of the pen (Fig. 1a). In the centre of the pen, the quails had access to a transparent plastic box (dimensions 25 × 21 × 15 cm) filled with sand (3 cm deep) for dustbathing.

The pens were arranged in two tiers of four pens each. A fluorescent tube (36 W) was suspended 53 cm above the pen floor. The lights came on at 05:30 h, and the photo period was 15 h with a 15 min twilight phase at the start and the end. Light intensity ranged from 39 lux in the corners to 370 lux in the centre of a pen (average values of six measurements in all six directions at 15 cm above the pen floor). Temperature varied between 12 and 31°C. Ventilation rate was increased as the temperature rose.

At the rear of each pen, there were four nest boxes on the floor (Fig. 1a). They were made of a white plastic basket (30 × 21 × 6.5 cm) filled with substrate and a light brown cardboard box (23 × 19 × 12 cm) set in the basket (Fig. 2). Cardboard boxes were used for experimental purposes and are not recommended for quail farming. At the entrance of the nest box, there was a curtain made of strips (2 cm wide, 1 cm apart) of green plastic tablecloth. The plastic basket contained either hay or a rectangle of artificial turf as a substrate. The second independent variable was the top of the cardboard box, which was closed (Fig. 2, type A) or had slits (six rectangular openings of 17 × 1.5 cm, type B). The four types of nest box were assigned to two pens each (i.e., one type per pen) and placed into the pens before the introduction of the quails.

The numbers and locations (in the nest box, in the dustbathing box, in the litter) of eggs laid in each pen were recorded every morning for two periods of 15 days each. The first period of data collection started 9 days after the introduction of the quails. Nine days after the end of this period, the nest box types were exchanged between the pens. Both the substrate in the basket and the nature of the top of the cardboard box were

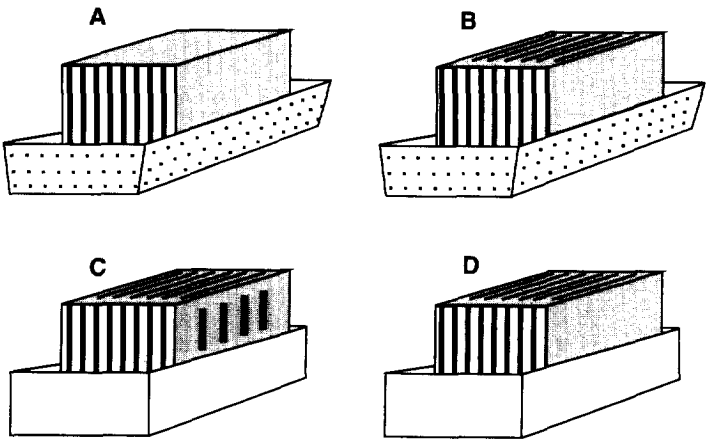

Fig. 2. Types of nest boxes used in experiment 1 (A, B), experiment 2 (C, D) and experiment 3 (D).

altered in each pen. After 2 days of acclimatisation to the new nest box types, the second period of data collection was started. All eggs were removed every day.

During statistical analysis, the percentage of floor-eggs (i.e., eggs laid outside the nest boxes) was calculated separately for each pen and each period of data collection. After arcsine square-root transformation, data from each period were analysed using a two-way ANOVA (Systat; Wilkinson, 1992). Differences in the transformed data on floor-eggs between the two periods of data collection were similarly analysed. The number of eggs found in the two corner nests and in the two central nests, respectively, was compared using the Wilcoxon-matched pairs-signed ranks test (Siegel and Castellan, 1988). All P values are two-tailed.

2.2. Results

In the first period, the quail hens laid between 11 and 73% of the eggs outside the nest boxes (Fig. 3a). Both type of substrate ($F_{1,4} = 51.50$, $P = 0.002$) and nature of nest box top ($F_{1,4} = 39.12$, $P = 0.003$) had a significant effect on the percentage of floor-eggs. The interaction between the two factors was not significant ($F_{1,4} = 0.45$, $P = 0.54$). Nest boxes with hay and a top with slits were most attractive. In addition, in all 8 pens, a significantly higher proportion of the 1470 eggs found in nests was laid in the nest boxes situated in the corners (average 78.9%) than in the more central nests (Wilcoxon-matched

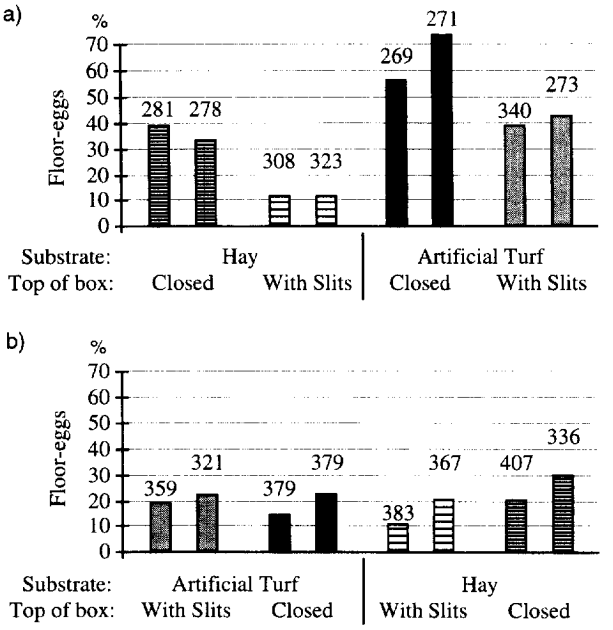

Fig. 3. Percentage of floor-eggs in pens ($N = 8$) with different types of nest boxes in (a) the first and (b) the second period of experiment 1. The nest boxes differed in the nature of the substrate and the nature of the top of the boxes. The pens are presented in the same order in both graphs. Figures above the bars indicate total number of eggs laid in a pen during the period of data collection (15 days).

pairs-signed ranks test, $N = 8$, $T^+ = 36$, $P < 0.01$). Only eight (0.3%) out of 2343 recorded eggs were laid in a dustbathing box.

The independent variables also had significant effects on the difference in the percentage of floor-eggs between the two periods of data collection (substrate: $F_{1,4} = 59.30$, $P = 0.001$; top of box: $F_{1,4} = 63.47$, $P = 0.001$; interaction: $F_{1,4} = 1.13$, $P = 0.35$). A decrease in the percentage of floor-eggs was observed in all four pens in which the quality of the substrate was changed from artificial turf to hay and in all four pens in which the quality of the top of the cardboard box was changed from closed to slits (Fig. 3).

In the second period (after exchange of nest box types), the quails laid between 11 and 30% of the eggs outside the nest boxes (Fig. 3b). In contrast to the first period, the percentage of floor-eggs was similar for all four types of nest boxes. Neither substrate ($F_{1,4} = 0.01$, $P = 0.93$) nor nest box top ($F_{1,4} = 0.83$, $P = 0.41$) had a statistically significant effect on the proportion of floor-eggs. However, in all 8 pens, the quails still laid significantly more eggs in the two corner nests (average 79.5%, $N = 2339$ eggs found in nests) than in the two central nests (Wilcoxon-matched pairs-signed ranks test, $N = 8$, $T^+ = 36$, $P < 0.01$). Out of 2931 eggs recorded during the second period, 24 (0.8%) were found in a dustbathing box.

3. Experiment 2

In experiment 2, the effects of nest box colour (green or brown) and side wall-type (closed or with slits) on the incidence of floor-eggs (2×2 factorial design) were examined.

3.1. Methods

This experiment was carried out with eight groups of 14 female quails. The birds were from the same breeding stock as in experiment 1, but had been reared in the experimental pens from day 1. In comparison to experiment 1, the pen design was slightly changed (Fig. 1b). In the centre of the pens, the floor was still covered with a mixture of wood-shavings and chaff, but on each side of the pens, there was an area of 1×0.5 m made from perforated plastic flooring, to reduce soiling of the litter floor. There were two drinkers, one feeder and one dustbathing box in each pen.

On the left of each pen, there were two nest boxes on the perforated plastic floor, made from a wooden box ($33 \times 21 \times 8$ cm) filled with chaff (3 cm deep) and a cardboard box (same dimensions as in experiment 1). In four pens, the nest boxes were entirely painted green (RAL 6018 yellow green, glossy) or brown (RAL 8003 loam brown, glossy). The second independent factor was presence of side-wall slits: the cardboard box had slits both on the top and in the sides (four vertical rectangular openings of 6×1.5 cm; Fig. 2, type C) or only on the top (type D). Each of the four nest box types was assigned to two pens. The nests were placed into the pens when the quails were 3 weeks of age.

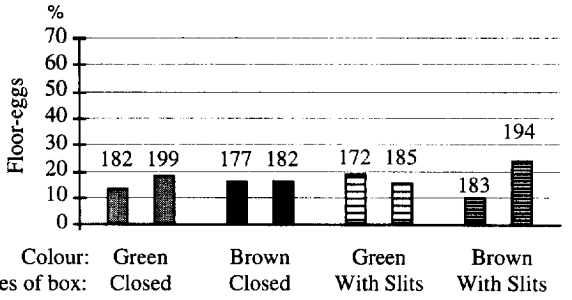

Fig. 4. Percentage of floor-eggs in pens ($N = 8$) with different types of nest boxes in experiment 2. The nest boxes differed in the colour and in the side wall type. Figures above the bars indicate total number of eggs laid in a pen during the period of data collection (15 days).

At the start of data collection, the quails were of the same age as those in experiment 1 (9 weeks). Data on egg laying places were recorded on 15 successive days. It was recorded whether an egg was laid in a nest box, in a dustbathing box, on the litter floor or on the perforated plastic floor. The percentage of floor-eggs was calculated for each pen. After arcsine square-root transformation, data were analysed using a two-way ANOVA. The number of eggs found in the nest box situated in the front and in the rear, respectively, was compared using the Wilcoxon-matched pairs-signed ranks test (Siegel and Castellan, 1988). All P values are two-tailed.

3.2. Results

The quail hens laid between 10 and 24% of the eggs outside the nest boxes (Fig. 4). Neither colour of nest box ($F_{1,4} = 0.01$, $P = 0.91$) nor presence of slits in the sides ($F_{1,4} = 0.06$, $P = 0.81$) had a significant effect on the proportion of floor-eggs. There was also no significant difference in the percentage of eggs found in nest boxes situated at the front or the rear of the pens (Wilcoxon-matched pairs-signed ranks test, $N = 8$, average values are 56.5 and 43.5, $T^+ = 25$, $P = 0.38$). Only 12 (0.8%) out of 1474 recorded eggs were laid in a dustbathing box. More floor-eggs ($N = 234$) were found on the litter floor (88.0%) than on the perforated plastic floor (12.0%).

4. Experiment 3

In experiment 3, the effects of substrate type (hay or chaff) and light intensity in the pen on the incidence of floor-eggs were examined in a 2×2 factorial design.

4.1. Methods

The pens previously used in experiments 1 and 2 were divided into two (Fig. 1c). There were thus 16 pens with a floor area of 1×1 m each. The groups consisted of 14

females and two males (8 pens) or 14 females and three males (8 pens). The number of males was varied between groups to check for differences in egg fertility (Wechsler and Schmid, in prep.). The quails had been reared in cages on the same commercial farm as the birds used in experiment 1. They were introduced into the experimental pens at the age of 7 weeks. The pen design was similar to experiment 2. Half the area was perforated plastic floor. The other half was covered with wood-shavings. There were two drinkers, one feeder and one dustbathing box in each pen.

In a corner in the front of the pens, there was one nest box on the litter floor. The wooden box was light brown and of the same dimensions as in experiment 2. All cardboard boxes were light brown and had slits on the top (Fig. 2, type D). Half of the nest boxes were filled with chaff as a substrate, the other half with hay. The second independent variable was light intensity, which was high (170 lux, average value of six measurements in all six directions at 15 cm above the pen floor at the brightest spot in the pen) or low (15 lux). Each of the four combinations of factors was assigned to four pens.

Data collection started 9 days after the introduction of the quails into the pens when the birds were 8 weeks of age and lasted for 15 successive days. It was recorded whether an egg was laid in a nest box, in a dustbathing box, on the litter floor or on the perforated plastic floor. The percentage of floor-eggs was calculated for each pen. After arcsine square-root transformation, data were analysed using a two-way ANOVA.

4.2. Results

The quail hens laid between 16 and 58% of the eggs outside the nest boxes (Fig. 5). There were fewer floor-eggs in pens with high light intensity than in pens with low light intensity ($F_{1,4} = 11.11$, $P = 0.006$). The effect of the substrate failed to reach statistical significance ($F_{1,4} = 3.71$, $P = 0.08$), but there was a trend for the percentage of floor-eggs to be lower for chaff than for hay. The interaction between the two factors was not significant ($F_{1,4} = 0.20$, $P = 0.66$). Only 16 (0.7%) out of 2369 recorded eggs

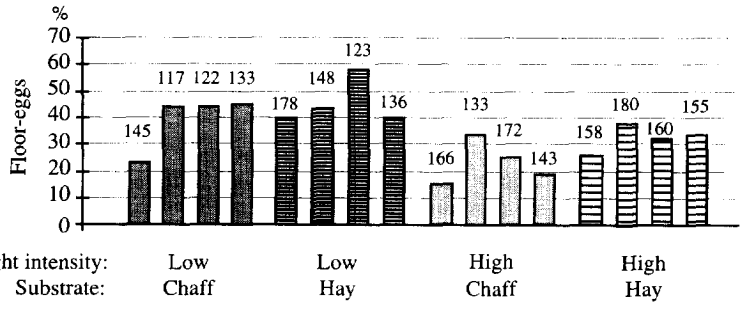

Fig. 5. Percentage of floor-eggs in pens ($N = 16$) with different light intensities and with nest boxes filled with different substrates in experiment 3. Figures above the bars indicate total number of eggs laid in a pen during the period of data collection (15 days).

were found in a dustbathing box. More floor-eggs ($N = 797$) were found on the litter floor (73.5%) than on the perforated plastic floor (26.5%).

5. Discussion

The results show that Japanese quail prefer laying eggs in sheltered sites. Although the nest boxes covered only 12.6% and 6.9% of the pen floor area in experiments 1 and 2/3, respectively, the quails laid up to 90% of the eggs in nest boxes. A preference for sheltered nest sites has also been observed in Japanese quail kept in semi-natural outdoor aviaries (Stevens, 1961; Orcutt and Orcutt, 1976; Schmid and Wechsler, 1997) and may be related to the use of tufts of grass as cover in the wild (Taka-Tsukasa, 1967).

The quails laid significantly fewer eggs outside the nests when the top of the nest boxes was not closed but had slits (experiment 1). This could indicate either that the quails seek sheltered nest sites where they can still monitor the sky, or that they tend to avoid nest sites which are too dark. The second explanation is supported by the observation that nest boxes with a top with slits were significantly better accepted in pens with high than with low light intensity (experiment 3). This result could also indicate that quails are reluctant to lay out in the open in very bright light. The addition of slits to the sides of the nest boxes did not further increase the use of nests with slits on the top (experiment 2).

Quails housed in outdoor aviaries show a marked preference to lay eggs in corners of the enclosures (Schmid and Wechsler, 1997). In accordance with this observation, significantly more eggs laid in nest boxes were found in nests situated in the corners of the pens than in more central nests (experiment 1), possibly because corners were perceived as more enclosed. On the other hand, the quails hardly ever used the dustbathing boxes for egg laying in all three experiments, although these boxes were also enclosed. As the dustbathing boxes were made of transparent plastic, they were perhaps not enclosed enough to be attractive egg laying sites. Alternatively, quails may avoid laying eggs on sand.

In experiment 1, nest boxes containing hay were significantly better accepted than nest boxes containing artificial turf, and there was a tendency for nests filled with chaff to be more attractive than nests containing hay (experiment 3). In the wild and in outdoor aviaries, Japanese quail nests typically consist of a shallow, scratched out depression in the ground filled with a small amount of dead straw or weed stems (Stevens, 1961; Taka-Tsukasa, 1967; Orcutt and Orcutt, 1976). As nests filled only with chaff were frequently selected in our study, the possibility of forming a hollow may be of more importance for quails than the availability of grass stems that can be built into the nest. Outside the nest boxes the selection of egg laying places was also influenced by substrate type. More floor-eggs were laid on the litter floor than on the perforated plastic floor (experiments 2 and 3).

In contrast to the results of Michel (1989), nest box colour (green or brown) had no significant effect on the percentage of floor-eggs (experiment 2). There was, however, a major difference in the design of the two studies. Whereas in the present study the colour of the nest boxes was varied between the pens, the quails in the former study

could choose between a green and an ochre cubicle provided in the experimental enclosure simultaneously. It is therefore possible that the quails in our experiment were mainly looking for an enclosed nest site irrespective of its colour.

In experiments 1 and 3, the quails were reared in battery cages and introduced into the experimental pens when they were 8 and 7 weeks of age, respectively; whereas in experiment 2, the quails were reared in the experimental pens and could visit the nest boxes from week 3 after hatching. Although it should be taken into account that there were differences in the types of nest box and in light intensity between the three experiments, the percentages of floor-eggs were not substantially lower in experiment 2. This suggests that the use of nest boxes for egg laying is not enhanced by giving the quails access to such nest sites during the rearing period. We can, however, not exclude that there may be a sensitive period for the development of nest site preferences during the first three weeks after hatching.

In experiment 1, our results suggest that the use of a specific type of nest box is influenced by previous experience with a different type. In the second period, after the exchange of the nest boxes, there were no longer significant effects of substrate or nest box top on the percentage of floor-eggs. This was mainly due to quails which had been given the most attractive nest box type (hay, with slits) in the first period continuing to lay most of their eggs in nest boxes during the second period, although the nests were now of the least attractive type (artificial turf, closed). The birds had probably built up a site preference in the first period that still lasted in the second period. This suggests that experiments on nest box preferences in Japanese quail should only be carried out with naive birds.

There are similarities with respect to nest site preferences in Japanese quail and laying hens. For example, domestic hens typically prefer enclosed nests over exposed ones (Appleby and McRae, 1986; Reed and Nicol, 1992) and nests containing litter over nests without litter (Huber et al., 1985; Duncan and Kite, 1989). Also in accordance with our observations on quails, laying hens can develop site preferences that may persist when the quality of the nest box is experimentally changed (Sherwin and Nicol, 1994; Hughes, 1993), and hens kept in modified cages containing both a nest box and a dust bath lay only a few eggs in the dust bath (Appleby et al., 1993; Smith et al., 1993).

In summary, properly designed nest boxes were well accepted resulting in percentages of floor-eggs as low as 10% even though part or all of the pen floor was covered with litter and despite the presence of a dustbathing box. Our results therefore indicate that it should be feasible to develop housing systems with incorporated nests for Japanese quail which may replace conventional battery cages. For practical reasons, however, it should be attempted to decrease further the incidence of floor-eggs in such housing systems. With free range hens the percentage of floor-eggs can be as low as 2–3.5% (Plank, 1989) and hens kept in modified cages containing both a nest box and a dust bath lay up to 99% of their eggs in the nest boxes (Appleby et al., 1993).

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