Dust bathing in laying hens: strain, proximity to, and number of conspecifics matter

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ABSTRACT As housing laying hens in aviaries becomes more common, understanding relationships between social context and performance of key behaviors, such as dust bathing (**DB**), is important. Expression of behaviors may be increased or repressed by the presence of conspecifics, and degree of behavioral synchrony can affect per hen resource allocation. We investigated relationships between number of hens on litter, number of hens simultaneously DB, and interbird distances (**IBD**) on space used to DB and duration of DB bouts across 4 laying hen strains (Hy-Line Brown [HB], Bovan Brown [BB], DeKalb White [DW], and Hy-Line [W36]) at 28 wk of age. Brown hens needed more space to DB than white hens (HB 1125.26; BB 1146.51 vs. DW 962.65; W36 943.39 cm^2; $P < 0.01$). More white hens occupied litter at once (43 DW, 41 W36 vs. 28 HB, 31 BB; $P < 0.01$), and more white hens DB simultaneously than brown hens (11 DW, 19 W36 vs. 4 HB, 4 BB; $P < 0.01$). Brown hens had larger average IBD (HB 13.99, BB 15.11 vs. DW 8.39, W36 7.85 cm; $P < 0.01$) and larger minimum IBD (HB 6.76, BB 7.35 vs. DW 1.63, W36 1.79 cm; $P < 0.01$) but shorter DB durations than white hens (HB 7.37, BB 9.00 vs. DW 13.91, W36 15.16 min; $P < 0.01$). White hens' DB area decreased if number of hens on litter increased (DW 0.85; W36 0.79 cm; $P < 0.05$) or minimum IBD decreased (DW 3.66, W36 2.98 cm; $P < 0.01$). Brown hens' DB bout duration decreased as number of hens on litter increased (HB 0.87, BB 0.95 min; $P < 0.01$), number of other hens DB increased (HB 0.75, BB 0.69 min; $P \leq 0.02$), or minimum IBD decreased (HB 2.39, BB 2.31 min; $P < 0.01$). In response to smaller IBD and more hens on litter simultaneously, DW and W36 hens minimize DB area while BB and HB hens shorten DB bouts, potentially terminating bouts before fulfilling their needs. Variations in DB behavior among strains should be considered when planning and stocking laying hen aviaries.

Key words: dust bathing, aviary, laying hen, welfare, behavior

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INTRODUCTION

The U.S. laying hen industry has begun to move from conventional cages to noncage systems, including open-concept aviaries. As this conversion occurs, it is important to understand how the birds utilize the space and resources provided in these cage-free environments and large social groups. Most aviary systems offer perches, nests, and litter areas to allow hens to perform highly motivated behaviors, making the additional space more valuable to the hens than would be gained by simply adding area (Keeling, 1995). However, birds tend to distribute themselves unevenly within these systems (Channing et al., 2001), clustering around certain resources (Collins et al., 2011), which may lead to crowding at some resources while others appear underused (Appleby, 2004; Ali et al., 2016). Thus, it is important to understand hens' behavior as affected by the social context in aviaries to identify birds' preferred distributions and behavior patterns when housed in groups to inform better aviary designs or more ideal stocking rates to facilitate optimal use of resources.

One of the important behaviors that aviaries promote is dust bathing (**DB**). This behavior generally occurs on litter-covered areas, where hens may spend up to 23% of their time each day (Carmichael et al., 1999). Dust bathing is a functionally important behavior, as it realigns feather structure and removes lipids from the skin of birds (Vestergaard, 1982; van Liere and Bokma, 1987). Hens are highly motivated to dust bathe and will work to gain access to litter (Widowski and Duncan, 2000).

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They will even dust bathe in the absence of an appropriate litter source, such as on wire flooring in conventional cages, which is unlikely to improve feather quality (Hughes and Duncan, 1988; Lingberg and Nicol, 1997). As an acknowledgment of the importance of DB to hens, various standards, laws, and welfare accreditation schemes require that hens have access to areas containing substrates suitable for DB. Thus, many aviary designs provide an open floor space that can be covered with litter to facilitate DB by laying hens.

Dust bathing is a dynamic behavior—composed of many active behaviors and hen movement around a litter area. As hens typically dust bathe in the afternoon, multiple birds in a group are likely to want to dust bathe at the same time (Vestergaard, 1982). There is also evidence for behavioral synchrony in DB among hens housed together (Hoppit et al., 2007). If many hens in a group perform DB concurrently, this may cause crowding on the litter areas (Campbell et al., 2016). It is not fully known how the synchrony of the behavior affects hens who are attempting to dust bathe, and hens of some strains may have a tendency to view a resource as crowded more than others (Keeling, 1995; Mench and Blatchford, 2014). Thus, stocking density and nearby conspecifics may influence hens' ability and desire to perform DB in an unconstrained manner at the time of day a hen would prefer to dust bathe. For example, as bird density increases on litter areas, DB decreases slightly (Carmichael et al., 1999).

Hens also express preferred interbird distances (**IBD**) between themselves and surrounding hens. Interbird distances vary depending on the behavior hens are engaging in, with more dynamic behaviors typically associated with larger distances and socially facilitated behaviors with smaller distances (Keeling, 1995). Space guidelines do not account for IBD that are preferred during performance of behaviors (Riddle et al., 2018).

Further, as there are multiple genetic strains of laying hen used by the egg industry, variation among these strains in weight and size leads to occupancy of different amounts of physical space when hens perform behaviors such as DB or perching (Riddle et al., 2018; Giersberg et al., 2019). Previous work in our lab found significant variations in space used by DeKalb White (**DW**), Hy-Line White, Hy-Line Brown (**HB**), and Bovans Brown (**BB**) hens during performance of key behaviors, including DB (Riddle et al., 2018). In addition to differences in the amount of space physically occupied by the body of a hen when performing a behavior, selective breeding for certain traits may have caused behavioral divergences among strains (Albentosa et al., 2003), which may include their preferred IBD, circadian rhythms, or desire to perform behaviors in synchrony. For example, previous studies have found distinctive distribution patterns, circadian rhythms, as well as different preferences for resources, in 4 of the more common genetic strains (Villanueva et al., 2017; Ali et al., 2019a, 2019b).

The goal of this study was to explore differences in DB behavior among 4 genetic strains of laying hens in an aviary. Building on previous work from our lab regarding areas used by hens of these same 4 strains while DB (Riddle et al., 2018), we investigated whether strain affected the degree to which hens would dust bathe synchronously and how the presence of conspecifics and their distance from a hen would affect her performance of a DB bout.

MATERIALS AND METHODS

Ethics

The methods used in this study were approved by the Michigan State University Institutional Animal Care and Use Committee before data were collected or animals were placed in the facilities (Animal use #01/15-025-00).

Hens and Housing

The subjects of this study were 2,304 hens from 4 commonly used genetic strains of laying hens in the U.S. egg industry: **HB**, **BB**, Hy-Line W36 (**W36**), and **DW** (n = 576 of each strain). The birds were reared in the pullet house at the Michigan State University Poultry Teaching and Research Center in East Lansing, MI. The pullet house was climate-controlled and contained 12 pens, each able to hold 225 to 250 chicks. Chicks were separated into pens based on strain (3 pens/strain; n = 657–750 chicks per strain). Pens were bedded with pine shavings (~7–10 cm deep), and a roosting area was added at 3 weeks of age (**WOA**).

At 17 WOA, the pullets were moved to a commercial-style aviary system (NATURA60, Big Dutchman, Holland, MI) in the Michigan State University Laying Hen Facility. Pullets were divided by strain across 4 rooms, with each room containing 4 separate aviary units with 1 unit/strain/room for 16 total units (4 units/strain). Each unit was initially populated with 144 pullets, as per the manufacturer's recommended stocking density. The 4 strains were placed into units in a balanced fashion within each room so that each strain was housed in a different unit location within each room to avoid location bias. Each aviary unit consisted of a wire-mesh enclosure with 3 tiers and an external litter area on the floor level. The litter area consisted of an open area in front of the tiered enclosure and an area that extended under the enclosure (see Figure 1). Hens were provided with 305 cm^2 of open litter area per bird. The stocking densities for each of the 4 strains based on the average live weights of adult hens (kg/m^2 as per Thaxton et al., 2006) were as follows: HB = 65.57 kg/m^2, BB = 61.64 kg/m^2, DW = 56.5 kg/m^2, and W36 = 50.16 kg/m^2. We also calculated the number of hens that could fit into the litter area based on their body size while standing or DB. In both cases, fewer brown hens would be expected to physically fit into the open litter area than white hens (Standing: DW = 77.4 hens, W36 = 76.1 hens, BB = 67.7 hens, and HB = 65.5 hens; DB: DW = 42.7 hens, W36 = 43.8 hens, BB = 37.2 hens, and HB = 36.9 hens).

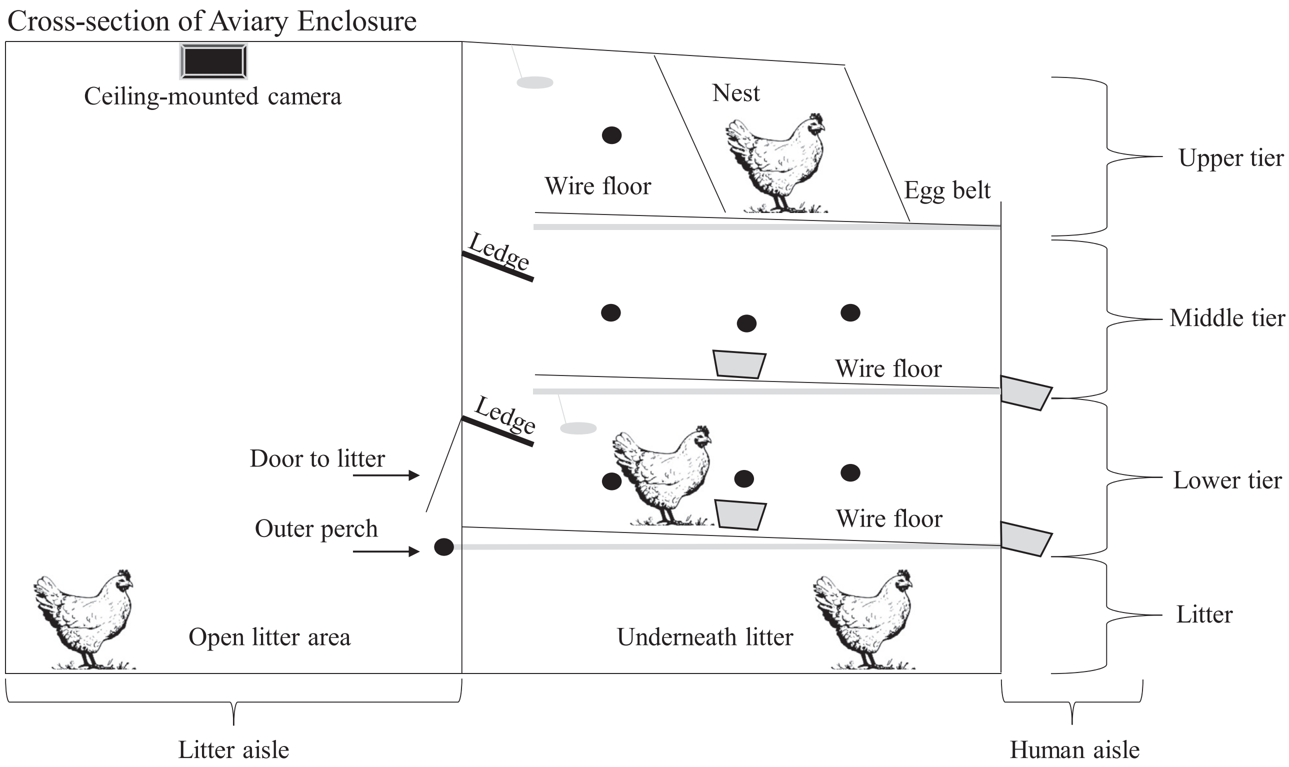

Figure 1. Cross-sectional diagram of the tiered aviary enclosure, showing the litter areas, aisles, manure belts (light Gy bars), and locations of the nest, drinkers (gray ovals), feeders (gray rectangles), perches (black circles), and ledges.

At the start of the laying cycle, the litter area was bedded with pine shavings (∼3–5 cm deep) similar to those provided during rearing. A round metal perch ran along the front of each enclosure to help the hens move between the floor litter area and the tiered enclosure. Light was provided by dimmable LEDs in the ceiling (Agrishift PL 12 W, ONCE, Inc., Plymouth, MN). For more details on the aviary design, food and water access, and space allotment per hen, see (Ali et al., 2016).

The hens remained enclosed in the tiered aviary until 26 WOA, when they were given access to the litter area. This delay allowed the hens to reach ∼90% egg production before allowing access to the litter in an attempt to train hens to use the nests and prevent egg-laying in the litter. Starting at 26 WOA, the doors on the lower tier of the enclosure began opening automatically at 11:30 each day to allow hens daily access to the litter area. The doors closed again at 01:00 once hens had returned to the aviary enclosure to roost. The lights turned on automatically each day at 05:00 and dimmed for 30-min before shutting off completely at 20:00. Hens had 2 wk to acclimate to the litter area before data were collected. For more details on feeding, cleaning, and lighting, see study by Ali et al. (2016).

Video Recording

Before the birds' placement in the aviary units, high-resolution digital video cameras (VF450: Clinton Electronics, Loves Park, IL) were affixed to the ceiling, centered above the open litter area of each unit, to record hen behaviors on litter during the day. All cameras were placed at the same distance from the litter. During the study, data were collected from approximately 11:30 to 15:00 as hens follow a circadian rhythm and most often dust bathe in the afternoon (Mishra et al., 2005).

Behavior Definitions

The behaviors examined in the current study are the key elements within the DB behavior sequence, as described by Kruijt (1964). These elements are bill raking, head rubbing, scratching with 1 leg, scratching with 2 legs, side-lying, ventral lying, and vertical wing shaking. A full ethogram describing these behaviors is provided in Table 1. The start of a bout was recorded when a hen's body touched the litter, and she performed any of the key elements of DB (Larsen et al., 1999; van Rooijen, 2005). When the hen stood up and did not resume any elements of DB behavior within 10 s, the bout was considered to have ended.

Data Collection

A statistical power analysis was conducted to determine the sample size needed for examining DB ("pwr" package in R, version 3.3.1 R Core Team, 2013). For all analyses, the experimental unit was the individual hen. The power of the test was set at 80%, with a beta of 20%, and an alpha of 0.05. The test yielded a desired sample size of n = 30 DB hens per strain. To balance collection of images across the 4 units per strain, we

Table 1. Ethogram of behaviors.

Behavior	Abbreviation	Description
Bill Raking	BR	The bill is first moved downward, and after touching the litter, it is quickly moved backward and then upward; in this way the litter is raked closer to the bird
Head Rubbing	HR	The side of the head is rubbed on the ground with 1 quick sweep
Scratching with 1 leg	S1L	One leg is moved backward at a time manipulating the litter while the bird has its body in contact with the litter
Scratching with 2 legs	S2L	Two legs are moved backward at a time manipulating the litter while the bird has its body in contact with the litter
Side Lying**	SL	One side of the bird remains flat against the litter while the bird remains still; occasionally, wing- and leg-stretching are present
Ventral Lying**	VL	The ventral side of the bird remains flat against the litter while the bird remains still
Vertical Wing Shaking	VWS	Nearly closed wings are held at a distance from the body and are moved vertically, both at the same time in the same direction, to sweep litter into the plumage

Adapted from Kruijt (1964). **created specifically for this project–no definition was present in the article.

collected DB information from 8 hens per unit for a total of 32 hens per strain and 128 hens in total (8 hens/unit × 4 units/room × 4 rooms = 128). All data were collected by the same trained individual.

Naturally occurring hen activity in the open litter area was recorded on video over the course of 3 d. Selection of DB bouts for analysis was done via convenience sampling using 2 criteria. First, a hen had to be demonstrating a key element of DB behavior as described in the ethogram (Table 1). Second, the hen needed to be roughly in the center of the open litter area (i.e., not touching any walls or gates or be fully or partially out of sight under the enclosure). Once a DB bout was identified, the duration of the bout was recorded, and images were captured from the beginning, middle, and end of that DB bout using the Snipping Tool (Microsoft Windows, 10.0.15063.13 tool kit). Images were then labeled by hen number (1–8), unit, time, and date and saved. We observed 32 focal hens DB per strain (8 hens per unit) and gathered measurements from each hen in the beginning, middle, and end of her DB bout. Therefore, we had a total of 96 observations for each strain of laying hen (32 hens/strain × 3 still images per hen).

From each selected image, the area each hen occupied while DB was calculated by drawing a line from the hen's most distal anterior point to the most distal posterior point (length); a second line was drawn across the widest part the hen's body (width), including potentially outstretched wings and legs. Interbird distances were measured between the focal hen and the nearest 5 to 7 surrounding hens in each captured image. The average IBD was calculated using the distances measured between each of these surrounding hens and the focal bird. The minimum IBD was the distance between the focal hen and the closest of these surrounding hens. The surrounding hens were labeled within the captured images to ensure accurate measurements and to avoid re-measuring the same birds (see Figures 2 and 3). The number of surrounding hens for which IBD was recorded varied between images, even sometimes from images recorded at different times within the same DB bout because the focal hen would occasionally relocate herself to an area with more or less conspecifics or because conspecifics would move away from the focal hen during the DB bout.

All measurements were recorded from captured images using ImageJ 1.50i software (Wayne Rasband, National Institute of Health, USA). We first set a scale in ImageJ by measuring a reference object within the open litter area of a known length, in this case the outer perch (244 cm). In each captured image, the scale was set to approximately the same value (595–600 pixels; each pixel then measured 2.4–2.5 cm) along the length of the outer perch to ensure each measurement produced an accurate length in cm within the pictures. All images used in this study captured the open litter area and the outer perch.

Finally, the total number of birds DB (including the focal hen) was counted within each image, and the total number of birds present on the open litter area was counted. Synchrony can be defined as multiple animals in a group performing a behavior simultaneously (Keeling et al., 2017). For this study, we looked at hens DB at the same time on the open litter. In an attempt to record the most accurate number of hens using the open litter area at once, a hen was considered to be on the litter when her body was at least 1/3 of the way past the outer perch in the open litter area.

Statistical Analysis

Statistical analyses were performed using R software (version 3.3.1), package "stats" (R Core Team, 2013). Descriptive statistics were calculated using the "psych" package, and data are presented as mean ± standard error of the mean. Counts of hens using litter area and DB, space used for DB by an individual hen, duration of DB, and average and minimum IBD between birds during DB were compared across the 4 strains of laying hens (HB, BB, DW, and W36). All comparisons among strains were performed using one-way ANOVAs using the "car" package, at the level of the individual hen, controlling for repeated measures from each hen, with strain as the main effect and unit (i.e., pen) as a random effect.

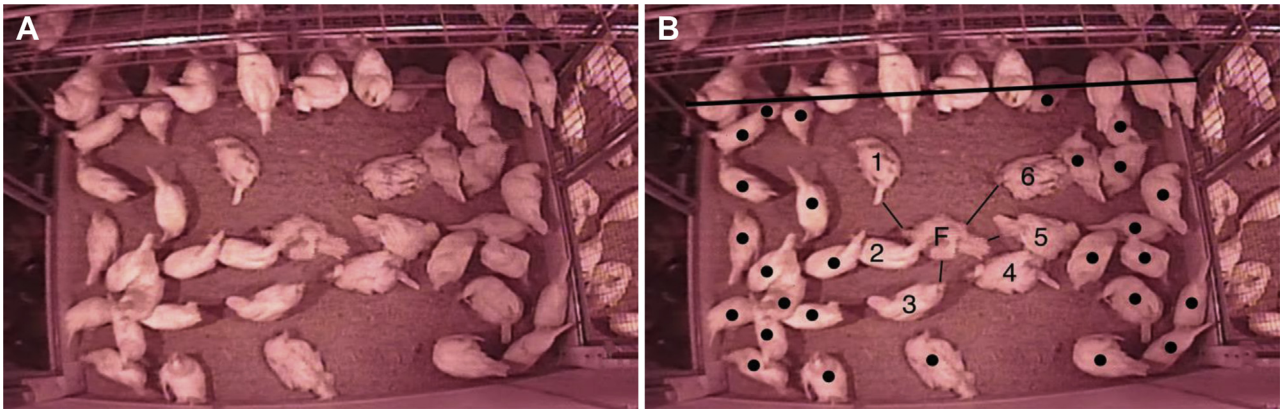

Figure 2. (A) Example of screen capture from video footage of the open litter area showing several hens performing DB. (B) F indicates the focal hen. Dots indicate hens counted on the open litter area (i.e., at least one-third of the hen's body was past the outer perch as indicated by the black line). Surrounding conspecifics analyzed for IBD are marked with a number (1-6), and lines indicate the closest part of their bodies to that of the focal hen. Abbreviations: DB, dust bathing; IBD, interbird distances.

$P \leq 0.05$ was considered significant. Analysis was done using hens as experimental units specifically to assess variability in individuals' behavior when DB on the open litter. Using individual animals as the experimental unit is appropriate when looking for variation in behavioral responses among individual animals (Robinson et al., 2006). Statistically significant effects were further analyzed using Tukey's honestly significant difference multiple comparison procedure using the "multcomp" package (Hothorn et al., 2008).

To explore influences of litter occupancy and IBD on the average duration and space occupied by individual hens while DB, mixed effect regression models were conducted using the "lme4" package (Bates et al., 2015). Aviary unit (i.e., pen) was included as a random effect in all regression models. The first regression model was generated to identify the relationships between number of hens on litter, number of hens DB, as well as the average and minimum IBD and the average duration of DB bouts. A second regression model was generated to identify the relationships between those same variables and the space occupied by individual hens while DB across the 4 strains of laying hens. Finally, coefficient estimates were transformed and presented as odd ratios (**OR**).

The Intraclass Correlation Coefficient for measuring agreement (**ICC**) was used following (Shrout and Fleiss, 1979) to measure intraobserver reliability using 10% of measurements for each behavior per strain using "ICC {cran}" package. Intraobserver reliability was calculated during the training period with the observer re-measuring the same birds twice in a random order, and a strong ICC of 0.98 (confidence interval = 0.898) was found.

RESULTS

Area

Hens of both brown strains occupied more space while DB than the hens of both white strains ($P < 0.01$ for all comparisons). Bovan Brown hens used the greatest area while performing a DB bout (1,146.51 ± 240.55 cm), followed by HB hens (1,125.26 ± 222.94 cm), and the areas used to DB by hens of these 2 brown strains were not different from each other ($P = 0.6$). Hens of both white strains occupied a similar amount of space to each other

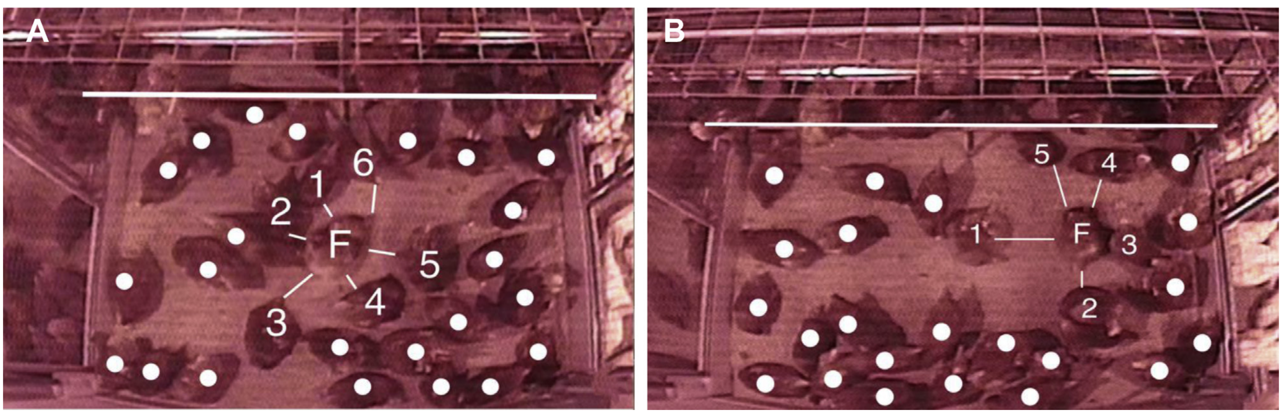

Figure 3. Examples of screen captures that show the beginning (A) and end (B) of a focal hen's DB bout. The focal hen is indicated by F. The hen moved throughout the DB bout as did other hens on litter, resulting in different numbers of hens being assessed for IBD at the beginning, middle and end points of the DB bout. These images also illustrate that it was difficult to distinguish the outline of brown hens from their shadows against the litter. Abbreviations: DB, dust bathing; IBD, interbird distances.

Table 2. Synchronous DB and litter occupancy, DB duration, and average and minimum IBD of focal hens among 4 laying hen strains.

Strain	Number of hens on litter[1]	Number of hens dust bathing simultaneously[2]	Average duration (minute)[3]	Minimum IBD (cm)[4]	Average IBD (cm)[4]
Hy-Line Brown	28.30 ± 8.47[a]	3.82 ± 3.27[a]	7.37 ± 6.98[a]	6.76 ± 3.67[a]	13.99 ± 4.65[a]
Bovan Brown	30.97 ± 8.46[a]	4.04 ± 3.92[a]	9.00 ± 5.11[a]	7.35 ± 3.89[a]	15.12 ± 7.34[a]
DeKalb White	42.8 ± 8.6[b]	11.26 ± 3.86[b]	13.92 ± 8.60[b]	1.63 ± 2.73[b]	8.39 ± 3.83[b]
Hy-Line W36	41.5 ± 6.59[b]	10.21 ± 3.53[b]	15.16 ± 8.58[b]	1.79 ± 1.74[b]	7.85 ± 3.65[b]

[a,b]Data are presented as means ± SEM. Different superscripts indicate statistical significance ($P < 0.05$).
Abbreviations: DB, dust bathing; IBD, interbird distances.
[1]Indicates the total number of hens present on the open litter area (i.e., at least one-third of the hen's body is past the outer perch on the open area).
[2]Indicates the total number of hens DB on the open litter area. (Note: the focal hen is included in both counts of both total number of hens on litter and the total number of hens DB.)
[3]Total duration of the focal hen's DB bout in minute.
[4]Minimum and average IBD in centimeters for each strain.

while DB (DW: 962.65 ± 165.13 cm; W36: 943.39 ± 152.90 cm; $P = 0.99$).

Relationships Between IBD and DB Area and Bout Duration

The W36 and DW hens spent an average of approximately 15 and 13 min per DB bout, respectively; whereas the BB and HB hens had average DB bout durations of approximately 9 and 7 min, respectively (Table 2). When looking at the average IBD between DB focal hens and nearby conspecifics on the litter, we found that, on average, DW and W36 hens DB with a smaller minimum IBD than BB and HB hens (Table 2). The DW and W36 hens were also more likely to show synchronous DB behavior, have more hens on the open litter area simultaneously, and have longer DB bout durations compared with the brown hens (Table 2).

Hens of the 2 white strains in the present study also responded differently to decreasing IBD than hens of the 2 brown strains. As the number of hens occupying the litter area increased, DW and W36 hens were more likely to reduce the amount of physical space they occupied during a DB bout compared with BB hens (Table 3). As the minimum IBD decreased (i.e., the "buffer zone") between the focal hen and nearby conspecifics decreased, DW and W36 hens were more likely to reduce the area they used to DB compared with BB hens.

In contrast, as the minimum IBD decreased between a focal DB hen and other hens on the litter, HB and BB hens were more likely to reduce the duration of DB bouts compared with DW hens (Table 3). As increasing numbers of hens occupied the litter area, HB and BB hens were more likely to decrease the duration of their DB bouts compared with DW hens. Finally, HB and BB hens were also more likely to decrease their DB duration compared with DW hens when the number of hens DB on the litter increased.

DISCUSSION

Area

Previous research, using the same flock of birds and aviary system, looked at the space used by laying hens of these 4 strains when performing certain behaviors, including DB (Riddle et al., 2018). In the current study, we re-calculated the area used by hens of the 4 strains when DB to verify that previous area estimates remained accurate. To do so, we used the same video footage used by Riddle et al. but took different still images and measurements from more (32 hens/strain vs. 16 hens/strain) and different individual birds (i.e., we used the same flock, but not the same hens). We found that the average areas occupied by hens of each of the 4 genetic strains during a DB bout were comparable to the results reported previously by Riddle and colleagues (2018). In addition, hens from both brown strains were again found to use more space to DB than did hens of both white strains (Riddle et al., 2018). BB hens, which occupied the largest average area while DB, used approximately 203.12 cm^2 more space than the smallest strain, the DW hens. This is likely because of the fact that brown hens are generally physically larger than white hens and may, therefore, require more space as they perform dynamic behaviors (Appleby, 2004).

Hens of both white strains in the present study decreased the amount of physical space they occupied while DB with closer proximity of conspecifics. The hens of the 2 brown strains did not significantly reduce the area they used to DB in response to closeness of conspecifics but instead shortened their DB bout duration.

Litter Occupancy

Hens from the 2 white strains examined in this study occupied the open litter area simultaneously to a greater extent than hens of the 2 brown strains. The number of hens that can fit on the open litter floor area will, of course, vary depending on what the birds are doing, but in general, more white hens would be expected to fit onto the litter than brown hens because of their smaller body sizes (Riddle et al., 2018). For example, 77.4 DW hens could fit onto the litter area while standing compared with 65.5 HB hens, and when DB 43.8 W36 hens could fit into the open litter area compared with 36.9 HB hens. Brown hens, therefore, would be expected to perceive the litter area as crowded at lower numbers of hens than white hens. In the current study, we found 40 white hens on average on the open litter

Table 3. Strain differences in area occupied by DB hens and duration of DB bouts.

Parameter	Odds ratio	Z	95% CI	*P*-value
Area of focal hen while DB				
Intercept [BB]	1.09	8.96	−0.37-0.75	0.00
Strain × Litter Occupancy				
DW	0.85	−4.25	0.24-1.43	0.004
W36	0.79	−6.96	0.15-2.34	0.003
Strain × Minimum IBD				
DW	3.66	3.99	1.58-4.48	0.00
W36	2.98	2.23	1.30-3.74	0.00
Duration of focal hen DB bout				
Intercept [DW]	1.14	15.52	−0.41-0.72	0.00
Strain × Litter Occupancy				
HB	0.87	−3.36	0.42-1.43	0.002
BB	0.95	−6.96	0.73-1.26	0.004
Strain × Number of Birds DB				
HB	0.75	−2.6	0.29-1.99	0.02
BB	0.69	−5.9	0.27-1.75	0.01
Strain × Minimum IBD				
HB	2.39	4.98	0.55-14.73	0.001
BB	2.31	4.52	0.68-10.59	0.003

Abbreviations: BB, Bovan Brown; DB, dust bathing; DW, DeKalb White; HB, Hy-Line Brown; IBD, interbird distances; W36, Hy-Line.

Results are presented as odds ratios and 95% confidence intervals. The first strain listed is tested against the baseline of the second strain (i.e., DW hens compared with BB hens).

at any 1 time, compared with an average of 30 brown hens, which suggests this to be the case.

Previously, hens from the same white strains had been found to often occupy the open litter area concurrently at higher numbers throughout the day compared with brown hens (Ali et al., 2016, 2019b). It should be pointed out, however, that Ali et al. (2019b) counted more brown hens in the litter area under the tiered enclosure as opposed to the open litter area, which was the location examined in the current study. Thus, observing only the open litter area may not give a full picture of litter occupancy, although it does provide evidence of strain differences in hen distribution.

Synchronous DB Behavior

Domestic fowl have previously been described to DB together, which might indicate that they feel safety in numbers when performing a vulnerable behavior like DB (Duncan, 1980; Keeling, 1995). Alternatively, their synchrony may arise as a result of social facilitation, which leads hens to DB more readily in the presence of other hens already executing DB bouts (Vestergaard et al., 1993). Rebound effect could be another possible explanation for DB synchrony in this system, as the hens did not have 24-h access to the litter and were kept enclosed in the aviary tiers from 01:00 to 11:30. Because the hens had delayed access to the litter, they may have an increased propensity to DB synchronously as soon as the doors opened (Dawkins, 1988; Hughes and Duncan, 1988).

Hens of the white strains were more likely to DB together with more conspecifics than hens of the brown strains—that is, roughly 10 white birds would DB at the same time compared with 4 brown birds. At present, it is unclear what underlying motivation causes the different degrees of DB synchronicity among the strains. Hens of the white strains in this study generally began to DB immediately upon the opening of the aviary doors, whereas the hens of the brown strains appeared less likely to DB right away. Hens of another white genetic strain, Lohmann Whites, have also been found to DB together at a higher rate during the morning hours (i.e., 11:00–13:00) than in the afternoon (Campbell et al., 2016). Specifically, the hens in that study would DB more often in the morning at approximately 27 WOA. The hens in our study were very close in age (28 WOA) to those hens during data collection. The hens of our 2 white strains would often DB as soon as the doors opened at 11:30, so it could be theorized that they DB in at a similar time to Lohmann Whites. Therefore, hens of these white strains may be more susceptible to effects of litter restriction than hens of brown strains.

However, Campbell et al. (2016) also used the same aviary system that closed from 05:30 to 11:00, so we cannot be sure if the white hens' inclination to DB early is because of rebound effect or to differences in circadian rhythm. These 4 strains (HB, BB, DW, and W36) also show variability in the time of oviposition. For example, hens of these white strains lay 55% of their daily nest eggs between 6:00 and 10:00 compared with 85% of nest eggs laid by the hens of the brown strains during this same period (Villanueva et al., 2017). Future studies should focus specifically on the circadian rhythm of DB among different strains of laying hen to parse out any distinctions in behavior because of genetics.

Duration

The W36 and DW hens in this study DB for an average duration of approximately 15 and 13 min, respectively; whereas the BB and HB hens had bout durations of around 9 and 7 min, respectively. Durations of DB bouts have previously been reported to last between 20 and 35 min in White Leghorn hens housed in deep-litter floor pens (Vestergaard, 1982). However, as the duration of a DB bout might vary depending on circumstances as well as strain of hen, it may be hard to determine whether reports from the literature represent an ideal bout length (van Rooijen, 2005). Hens housed in fairly unconstrained systems, such as those with larger available areas to use for DB, may show longer DB bout durations that are closer to those previously reported in the literature. For example, hens of another brown strain (Warrens) had a median DB duration of 16.8 min when provided with a larger DB area (1,200–1,800 cm^2/bird; van Liere et al., 1991). In contrast, other studies using smaller litter areas and varying litter quality have also found reduced DB bout durations, similar to those reported in the current study. For example, ISA Brown pullets housed in battery cages affixed to boxes containing sand, providing 259 cm^2/bird in each box, performed DB bouts lasting an average of 5 to 10 min (Smith et al., 1993). A second study, also using ISA Brown pullets in cages affixed to boxes but this time containing 375 cm^2/bird of usable litter, found median DB durations of 4 to 7 min (Appleby et al., 1993). Finally, Lohmann Brown and Lohmann Selected Leghorn hens, a brown and white strain, respectively, housed in compartments containing litter trays providing 1,000 cm^2/hen, DB for 2 to 15 min, with bout length influenced by diet or litter substrate (Scholz et al., 2011).

In the current study, hens of all 4 strains were provided 1,132 cm^2/bird, including the total litter area and tiered enclosure, with 305 cm^2/bird of this space in the open litter area. All hens were raised the same way, fed the same diet, and housed in the same environments (see Ali et al., 2016 for more information). Thus, the differences in DB durations among hens of BB, HB, DW, and W36 strains are likely because of genetic variation in behavior or in their sensitivity to disruption or proximity of conspecifics. For example, the shorter DB duration of brown hens may reflect greater sensitivity to social disruptions or the need for more room to DB successfully. Keeling (1995) suggested that a particular stocking density may be viewed as “crowded” for certain individuals but may be fine for others. In this case, the hens of the white strains in the current study appeared to DB together more often overall and to tolerate the presence of conspecifics while DB to a greater extent than hens of the brown strains. Given these results, we agree with Appleby’s (2004) conclusion that a standard space allowance broadly applied to all strains of laying hens is not be ideal for facilitating key behaviors by hens.

Alternatively, the strains of brown hens in the current study may also simply have shorter DB bouts than the hens of our white strains. It is also possible that the brown hens continued DB bouts under the tiered enclosure or performed several short DB bouts throughout the day, whereas the white hens may have been more inclined to start a DB bout and complete it to the best of their ability in 1 sitting—thus increasing their duration. In future, DB by hens of various strains should be examined under more optimal conditions (e.g., fewer total hens, more space per individual hen) to see if there are still differences in how hens of different strains perform DB behavior when less constrained.

Interbird Distances

DeKalb White and W36 white hens in the present study had the smallest average and minimum IBD—meaning that white focal birds were generally closer to conspecifics while DB. Conversely, BB and HB hens had the largest average and minimum IBD—indicating that focal birds had a larger buffer between themselves and others while DB. Hens have a tendency to cluster together instead of distributing themselves evenly over an area, but the degree of clustering can vary depending on the behavior being performed (Keeling, 1994; 1995). In the case of DB, conspecifics may be inclined to join a focal hen that is DB until large numbers of hens are synchronous in their behavior (Campbell et al., 2016). Conversely, hens may be motivated to perform DB behavior but cannot physically do so because of limited space or because of a conspecific entering their individual space. Because DB is a social behavior (Duncan, 1980; Hoppit et al., 2007), it is possible that hens wanted to DB together but could not fit within the litter area while maintaining their preferred IBD.

Regardless of the underlying cause, our results indicate that hens of the white strains were generally more likely to tolerate smaller IBD than hens of the brown strains. Even if the white hens in our study were unable to move their bodies in the most optimal manner during a DB bout, they still continued the behavior while crowded, whereas the brown hens terminated their DB bouts when they appeared to find IBD to be smaller than tolerable.

Limitations

We were not able to view the entire litter area and could not tell if hens were DB in the litter area under the tiered enclosure. Additionally, as litter access was restricted throughout the night and for part of the morning, our ability to observe differences in strain-based circadian rhythms in DB behavior was limited. More research should be done in aviaries with 24-h litter access tracking individual focal hens to further understand if differences we observed in DB bouts are because of distinctive circadian rhythms among the strains. There may also be sampling bias in our results, as we assessed only “ideal” DB bouts where the focal hen was centered on the open litter area. Thus, we may have neglected to analyze certain naturally occurring bouts throughout the day in our attempt to choose clear, higher-quality bouts for measurement. However, the data from all strains were likely equally biased. In future, all DB bouts that occur should be sampled for a more accurate picture of hen behavior and space use within an aviary system.

CONCLUSION

There are strain differences in how laying hens perform DB in an aviary system with respect to bout duration, varying IBD, synchrony of behaviors, and, potentially, circadian rhythms. Hens of the white strains used in this study (i.e., DW, W36) showed higher rates of litter occupancy and more synchrony in their DB behavior compared with hens of the brown strains (i.e., HB, BB). The white hens had smaller IBD while performing a DB bout, whereas hens of the brown strains had larger IBD and shortened the duration of DB bouts in the presence of more hens on the litter or with less space between nearby hens. This indicates spatial-social differences among strains of laying hen while performing key behaviors such as DB. These findings continue to support the growing number of studies indicating behavioral differences among strains of laying hens. Producers may want to use different strains depending on the housing system they are stocking.

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