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Laying hen production and welfare in a cage-free setting is impacted by the northern fowl mite

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Primary Audience: Flock Supervisors

SUMMARY

The northern fowl mite (*Ornithonyssus sylviarum*) is a common blood-feeding ectoparasite of poultry that can cause decreased egg production and reduced hen welfare. The blood-feeding behavior of the northern fowl mite (**NFM**) elicits an immune response in hens that leads to decreased egg production, anemia, irritation to flocks and personnel, profit loss, and death to hens in extreme cases. As the egg industry moves toward extensive housing systems, it is important to consider management implications of this switch, such as the impact of NFM infestations. In the present study, hens were infested with NFM in two trials. Production and welfare parameters were monitored throughout the duration of the trials, beginning at 18 wk of flock age, and ending at 47 and 49 wk of age for Trials 1 and 2, respectively. A cannibalism issue in Trial 1 resulted in a severe loss of hens by the end of the trial and a low overall NFM infestation, while Trial 2 had consistent NFM infestation levels. In Trial 1, NFM negatively affected hen-day percentage, body weight, and feather coverage of some body regions. In Trial 2, the NFM infestation negatively affected hen-day percentage, mortality, body weight, shell thickness, and feather coverage of some body regions.

Key words: mite, poultry, cage-free, welfare, cannibalism

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DESCRIPTION OF PROBLEM

As concern for animal welfare in confined production systems grows (Lay et al., 2011; Zhao et al., 2015; Ochs et al., 2018), the shift toward extensive housing systems in the laying hen industry, such as cage-free aviaries or barns, has been encouraged by consumers, retailers, and legislation in the United States and elsewhere (such as the European Union). As this shift occurs, consideration of the impacts on hen welfare, egg production, and ease of management of the system must be weighed. While cage-free systems have many advantages for birds, such as greater opportunities to exhibit natural behaviors and more space to roam, they can also have drawbacks, such as greater difficulty in managing and cleaning the system which can provide opportunities for ectoparasites (such as the northern fowl mite) to flourish (Lay et al., 2011; Heerkens et al., 2015).

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The most common ectoparasite of poultry in the United States, the northern fowl mite (*Ornithonyssus sylviarum*), has the potential to cause significant economic losses to producers. The mites' blood-feeding on the bird induces a host immune response that can lead to reduced egg production and feed conversion efficiency (Harris et al., 2000; Murillo et al., 2016; Vezzoli et al., 2016). The northern fowl mite was discovered in 1917 in Beltsville, Maryland and then spread throughout the United States before being recognized as a detrimental poultry pest by Wood in 1920 (DeVaney et al., 1977). These mites have a short life cycle (5−12 d) that completes on-host, allowing large populations to build rapidly on hens (DeVaney, 1979; Axtell and Arends, 1990; Harris et al., 2000; Mullens et al., 2004; Mullens et al., 2009). Down-like feathering in the vent region of birds provides a nest-like environment where the mites can grow and flourish; mites will reside on vent feathers and will move to the skin to blood-feed (DeVaney and Augustine, 1987; Owen et al., 2009; Vezzoli et al., 2016). This causes inflammation and scabbing in the vent region, causing bird irritability (Loomis et al., 1970; Owen et al., 2009).

In addition to bird discomfort, northern fowl mite infestations can cause significant economic losses. In one study, $0.07 to 0.10 was lost per hen over a 10-wk period, equivalent to a decrease in hen-day production of 2.1 to 4.0% (Mullens et al., 2009). Laying hen flocks or breeder facilities are common areas for a northern fowl mite infestation to occur due to the long period of time flocks are kept, allowing mite populations time to grow and flourish (Mullens et al., 2009). Mites can be introduced into a flock via infested pullets, personnel, wild birds and rodents, and equipment, causing irritation to flocks and workers in poultry houses (Axtell and Arends, 1990; Kells and Surgeoner, 1997; Murillo and Mullens, 2016). As the industry switches to more complex, extensive environments, the risk of parasite burden may increase, which can reduce hen welfare and can lead to profit loss (Harris et al., 2000; Kilpinen et al., 2005; Lay et al., 2011; Murillo et al., 2020).

While there is information regarding the impact of the northern fowl mite on hen performance and welfare in caged egg-production, a better understanding of northern fowl mite impact in a cage-free setting is necessary as the industry moves to new housing systems. The present study aimed to investigate the impact of the northern fowl mite on laying hen performance and welfare in a cage-free housing system. Unanticipated flock challenges with cannibalism, another problem that has the potential to increase in cage-free systems, are also described.

MATERIALS AND METHODS

General Management and Laying Hen Room Information

All procedures were approved by the Purdue University Institutional Animal Care and Use Committee (IACUC Protocol Number: 1706001582). Two flocks (Trials 1 and 2) each consisting of 800 Tetra Brown hens housed in 4 cage-free rooms at the Purdue University Poultry Unit ($n = 200$ hens per room). Two of the 4 cage-free rooms (Rooms 3 and 4) served as a control and 2 rooms (Rooms 1 and 2) were infested with northern fowl mites (**NFM**). In order to prevent the spread of mites from the 2 treatment (NFM infested) rooms to the 2 control rooms, the four rooms were located at the end of one wing of the laying hen building. This allowed personnel to move in descending order from Room 4 to Room 1 and then to the exit of the building. The 4 cage-free rooms were fitted with the communal kick-out nest boxes (Colony 2+ System, Big Dutchman USA, Holland MI), a single-tier aviary system with 10 automated nest boxes, litter space with 12 perches, and a slatted floor area. Figure 1 provides a schematic of the room, while Table 1 summarizes resource allotment. Over the slatted area there were 6 hanging tube feeders and 2 bell drinkers. Eggs were able to roll to the back of the nest boxes for daily collection. The four cage-free rooms met the Guidelines for Cage-Free Housing as provided by United Egg Producers (United Egg Producers, 2017). Water was supplied to the hens ad libitum and hens were managed and hand-fed daily according to the Tetra Management Guide (Tetra Americana, 2017).

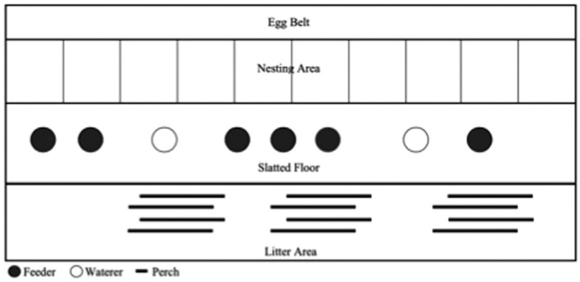

Figure 1. Schematic of the Colony 2+ cage-free room used in control and northern fowl mite infested rooms for Trials 1 and 2.

Trial 1. Pullets were reared at the Purdue University Poultry Unit in cage-free housing according to the Tetra Management Guide (Tetra Americana, 2017). Upon placement, chicks were observed to have not been beak treated at the hatchery, nor were they beak treated during rearing. A vaccination schedule used for cage-free flocks was followed during the rearing phase (Gast et al., 2021). At 17 wk of age, pullets were moved to the layer facility, where they were placed in Rooms 1 through 4 as described above. The flock in Trial 1 was monitored from 18 to 47 wk of age. Due to the absence of beak treatment, a cannibalism outbreak occurred during the pullet phase that continued throughout the duration of Trial 1. At 20 wk of age, in an attempt to mitigate the cannibalism, all hens' beak tips were removed using a Dremel (Dremel 4000 Series, Dremel, Racine, WI). At 21 wk of age, 25 replacement pullets (sisters of the original flock) were added to offset early mortality due to cannibalism. One pullet was added to Room 4, nine pullets were added to Room 3, fifteen pullets were added to Room 2, and no replacement pullets were added to Room 1. Two percent of the original hen population in the 2 NFM rooms (Rooms 1 and 2; 4 hens per NFM treatment room or 8 hens total across the treatment) were infested with NFM at 24 wk of age. This was done to mimic a commercial farm setting, where hens are unlikely to be uniformly infested with NFM. As a result of cannibalism and feather pecking behavior, NFM populations remained low after initial infestation, so a second mite infestation took place at 35 wk of age (on 2% of the initial hen population; 4 hens per NFM treatment room or 8 hens total across the treatment). NFM populations continued to remain low so a final attempt to infest with NFM occurred on all hens in Rooms 1 and 2 at 41 wk of age to increase mite prevalence.

Table 1. Resource allotment per hen (laying phase) in each of the four cage-free rooms fitted with the Big Dutchman Colony 2+ System.

Item	Area	Number	Total	Per hen
Total area	27.8 m^2	1	27.8 m^2	1,393 cm^2
Feeder space	127 cm	6	762 cm	3.8 cm
Bell drinker	109 cm	2	218 cm	1.0 cm
Nest area	0.55 m^2	10	5.57 m^2	278.7 cm
Perch space		12	3,048 cm	15.2 cm

Trial 2. Trial 2 involved a flock purchased as cage-free started pullets at 15 wk of age that were beak treated at the hatchery. This second flock was transported to the same 4 cage-free rooms in the laying hen building at the Purdue University Poultry Unit. This flock was monitored from 18 to 49 wk of age. Hens were infested with NFM at 24 wk of age (2% of the initial hen population, as in Trial 1), and a second mite infestation occurred at 30 wk of age (2% of the initial hen population) to boost mite infestation levels.

Mite Infestation

Mites were applied to individual birds using a method adapted from Martin and Mullens (2012). During infestation, hens were chosen at random from each of the 2 NFM treatment rooms (Rooms 1 and 2) and were inverted to expose the vent area. Approximately 30 mites were shaken onto the vent area from either glass pipettes or 50 mL conical tubes. The newly infested hen remained upside down for approximately 10 s to allow mites to crawl from the feathers to the vent skin. Northern fowl mites were sourced from commercial laying hen farms in the Midwest and from a research facility at the University of California (Riverside, CA).

Production Factors

Data Collection and Procedures. For both Trials 1 and 2, egg production and mortality data were recorded daily using electronic databases (FileMaker Pro 18, Claris International, Inc., Santa Clara, CA) developed for this study. Every 28-d period, 30 eggs collected from the nest box in each room (n = 120 eggs) were analyzed for egg component data following the procedure established by Karcher et al. (2019). Briefly, this involved recording individual intact egg weights, yolk weights, and shell weights. Rinsed and dried eggshells were used to take three measurements along the equator using a shell thickness gauge (Model 25-5; B. C. Ames, Inc., Melrose, MA).

To investigate initial spread and presence of mites, weekly mite checks were performed from the time of NFM administration to 28 d after. This involved always assessing 50 random hens from each of the 4 rooms. Hens were given a “yes” if mites were present and a “no” if mites were not. Mite populations were not quantified in the mite check procedure. NFM counts began 28 d after introduction and continued once every period (28 d). This involved using a scoring system adapted by Arthur and Axtell (1982) and Owen et al. (2009; Table 2) to estimate mite populations on hens. All hens in Rooms 1 and 2 were evaluated for mites and 40 hens each in Rooms 3 and 4 were examined to determine if any mites spread to the control rooms.

Table 2. Scoring system used for mite counts adapted from Arthur and Axtell (1982) and Owen et al. (2009).

Score[1]	Population[2]
0	0 mites
1	1−10 mites
2	11−50 mites
3	51−100 mites
4	101−500 mites
5	501−1,000 mites
6	1,001−10,000 mites
7	Greater than 10,000 mites

[1]Mite count score.

[2]Mite population level related to score.

Welfare Measures

Data Collection and Procedures. Body weights and Welfare Quality (**WQ**) assessments (Welfare Quality® Consortium, 2009) were conducted once every period (28 d) in a similar manner to Regmi et al. (2018) and Weimer et al. (2019). During WQ assessments, all hens in Rooms 1 and 4 and 40 hens each in Rooms 2 and 3 were examined for feather damage, comb abnormalities, footpad condition and toe damage, keel deformities, beak condition, presence of skin lesions, or abnormal conditions such as panting, enlarged crop, enteritis, or parasites. Feather damage, comb abnormalities, footpad condition, keel deformities, beak condition, and presence of skin lesions were scored as a 0, 1, or 2, with 0 indicating no damage, 1 indicating some or moderate damage, and 2 indicating severe damage. Toe damage and abnormal conditions such as panting, enlarged crop, enteritis, or parasites were scored as a yes (present) or no (absent).

Statistical Analysis

Weekly hen-day production percentage (an indicator of production rate of a flock over a given period) and eggs per hen housed were calculated for Trials 1 and 2. The GLIMMIX procedure of SAS (Version 9.4, SAS Institute Inc., Cary, NC) was used to detect effects from treatment (northern fowl mite or control), period (28 d), and interaction effects on each response variable. The experimental unit was either room or hen depending on the type of data being collected. Statistical significance was accepted at a $P \leq 0.05$. A random statement was included in the model to account for repeated measures. In Trial 2, mite population was added to the model (PROC GLIMMIX) for production parameters (hen-day production percentage, livability, and body weight) as a covariate to understand how mite population levels impacted production parameters. This covariate was significant ($P < 0.05$) for livability and body weight. The response variables for production factors were hen-day production percentage, percent livability, body weight, egg weight, and percent egg components. A post hoc test using the Bonferroni procedure was used to detect treatment and period differences in production factors.

For welfare measures, the PROC FREQ procedure of SAS was used to investigate overall treatment-specific frequencies of scores from Welfare Quality assessments across all periods. PROC GLIMMIX was then used to detect treatment- and period-specific frequencies of WQ scores for welfare measures using a binary or a multinomial distribution, depending on the specific scoring system used. The response variables for welfare measures were keel bone damage, presence of tip fractures, foot condition, presence of comb wounds, and feather score.

RESULTS AND DISCUSSION

Study Limitations

Stressors. Environmental stressors were present in both trials, such as high humidity in Room 1 that led to high litter moisture. This impacted foot condition and likely affected the NFM populations in Trial 1 in combination with cannibalism, further hindering the mites' ability to flourish. Trial 1 dealt with a severe cannibalism issue that developed in the pullet phase of the flock. These cannibalistic behaviors impacted hen behavior and caused excessive amounts of mortality in the flock. This contributed to differences in production metrics (body weight and hen-day percentage) and did not allow for a flourishing NFM population. While hens in Trial 2 did not experience cannibalism, the flock developed egg-eating behaviors that impacted hen-day percentage through decreased daily egg production values. Environmental stressors and negative behaviors such as cannibalism and egg-eating are identified challenges in cage-free or extensive housing systems. These stressors contributed to the flock-to-flock variability observed between Trials 1 and 2.

Trial 1. Throughout Trial 1, no hen was found to have a mite count score greater than 1 (1−10 mites; Table 2), and at peak infestation 33 hens (8.25% of the initial hen population) were found to harbor a mite count score of 1. Peak infestation occurred at 25 wk of age after initial infestation at 24 wk of age (period 2). By 33 wk of age (period 5), only 1 hen was found with mites (score 1). At 35 wk of age (period 5), the second attempt to infest with NFM occurred on 4 hens in each of the 2 NFM rooms (Rooms 1 and 2), and by 36 wk of age (period 5), mites were detected only on 4 hens (score 1). A final attempt to infest with NFM occurred on all hens (approximately 108 hens) in the 2 NFM rooms at 41 wk of age (period 7), and by 47 wk of age (period 8) only 3 hens were found with mites (score 1).

The flock developed cannibalistic behaviors as pullets that led to an average loss of 55% of the flock upon conclusion of the trial at 49 wk of age (period 8). As shown in Figure 2A, percent livability was 39.6 and 50.4% for the control and NFM groups at period 8, respectively, while the projected livability for Tetra Brown hens was 97.5% (Tetra Americana, 2017). Treatment ($P < 0.0001$) effect on livability was observed with the control treatment having more mortality than the NFM treatment. Period ($P < 0.0001$) had an impact throughout the duration of the trial with differences occurring at periods 6 to 8 ($P < 0.05$). Hen-day percentage (Figure 3A) was affected by period ($P < 0.0001$) and treatment ($P = 0.0003$). The NFM treatment peaked at 89% and control at 97% in period 4. The control group was higher throughout the duration of the experiment with period 6 being 13.8% higher than the NFM treatment ($P < 0.05$). Hen-day percentage from the Tetra Brown management guide (Tetra Americana, 2017) was added to Figure 3A as a baseline with an average hen-day percentage of 11.75% for period 1 (18−21 wk of age). Both the NFM and control treatments outperformed the management guide in period 1, with hen-day percentage at 51.04 and 56.40%, respectively; however, the Tetra Brown hen-day percentage increased and plateaued over time, ending with an average projection of 89.33% in period 8 (46 −49 wK of age). The interaction ($P = 0.002$) of period and treatment had an effect on body weight (Figure 4A) with NFM having smaller body weights in Periods 2 through 4. Egg weight (Figure 5A) was affected by period ($P <$

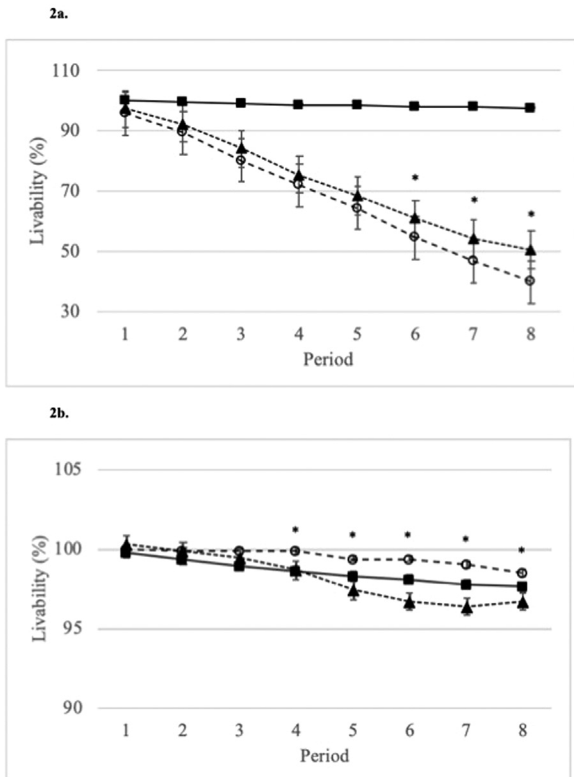

Figure 2. Percent livability for Tetra Brown hens from periods 1−8 for Trial 1 (A; 18−47 woa) and Trial 2 (B; 18−49 woa). Lines represent: Tetra Brown management guide (—■—), control hens (- -o- -), northern fowl mite infested hens (--▲--). Each data point is a mean of values for the period (28 days) with SEM. *: differences between control and NFM ($P < 0.05$).

0.0001) with NFM eggs increasing in weight while the control eggs decreased in weight at period 5 before increasing again. Percent yolk, albumen, and shell, expressed as a percentage of intact egg weights between 22 and 45 wk of age were affected by period ($P < 0.0001$). Period 4 revealed a lower percent yolk ($P < 0.0001$) in the control group (23.38%) compared to the NFM treatment (24.41%) and lower percentage of albumen ($P < 0.0001$) in the NFM treatment (65.74%) compared to the control (66.56%). Shell thickness observed an interaction effect ($P < 0.0001$; Figure 6A) with period 3 and 5 being different between treatments. Egg components, as well as individual egg weights, were not recorded in periods 1 and 6.

Environmental stressors such as high humidity can negatively impact performance (Talukder et al., 2010). Additionally, feather pecking and cannibalism behaviors can lead to fear and stress in a flock that can result in decreased egg production (Mertens et al., 2009; Sun et al., 2014). Cannibalistic behaviors may have led to slower body weight growth in the NFM treatment due to decreased time at the feeder or increased exercise from hens attempting to escape cannibalistic behavior.

The Welfare Quality assessment illustrated a change in all parameters over time (Tables 3−23). Keel bone damage worsened over time, regardless of treatment (Table 3). Presence of tip fractures increased over time, with a higher number of tip fractures found in the control treatment in period 4 (Table 5). Foot condition worsened over time, with better foot condition in the control treatment when compared to the NFM treatment at period 6 (Table 7). Comb wounds and abnormalities became more prevalent over time, regardless of treatment (Table 9). Litter in the four cage-free rooms became damp due to high humidity, especially in Room 1,

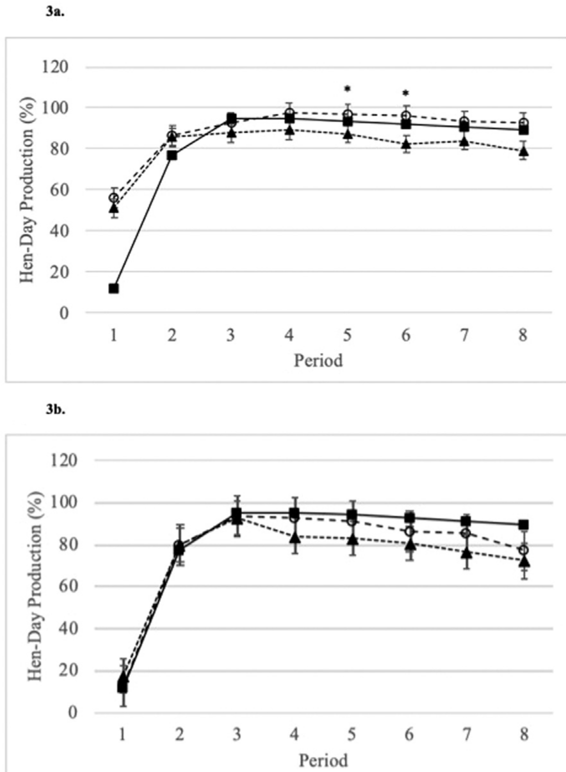

Figure 3. Hen-day egg production percentage for Tetra Brown hens from periods 1−8 for Trial 1 (A; 18−47 woa) and Trial 2 (B; 18−49 woa). Lines represent: Tetra Brown management guide (—■—), control hens (- -○- -), northern fowl mite infested hens (--▲--). Each data point is a mean of values for the period (28 days) with SEM. *: differences between control and NFM ($P < 0.05$).

which could have led to worse foot condition in the NFM treatment (Rooms 1 and 2) when compared to the control treatment. Feather damage worsened throughout the duration of Trial 1. Feather damage on the crop was more prevalent in the NFM treatment than the control in period 6 ($P < 0.01$; Table 11); however, at period 7, feather damage on the crop was greater in the control treatment ($P < 0.01$; Table 11). Feather damage on the keel region worsened over time but was greater in the control treatment at period 7 ($P < 0.001$; Table 13). Feather damage was greater on the belly region in the control treatment at periods 6 and 7 ($P < 0.05$; Table 15), while feather damage was greater on the head region in the NFM treatment in periods 5 and 6 ($P < 0.01$; Table 17). Feather damage on the neck was greater in the NFM treatment in periods 5, 6 $P < 0.01$) and 7 ($P < 0.05$; Table 19). Feather damage on the back was more prevalent in the NFM treatment when compared to the control in periods 2, 4 ($P < 0.05$), and 6 ($P < 0.01$) but was worse in the control treatment at period 7 ($P < 0.01$; Table 21). Feather damage on the rump was more prevalent in the NFM treatment in periods 4 ($P < 0.05$) and 6 ($P < 0.01$) and was more prevalent in the control treatment at period 7 ($P < 0.01$; Table 23).

Cannibalism and extreme feather loss in Trial 1 could be attributed to the intact beaks on hens (Lay et al., 2011; Tablante et al., 2000; Heerkens et al., 2015). Feather damage in the belly area could be linked to increased feather pecking behaviors in the flock (shown in Tables 11−23) as well as the hens' increased ability to groom themselves with intact beaks if irritation from mites was occurring. It has been reported that birds with intact beaks are able to groom themselves more efficiently than hens with modified beaks (Mullens et al., 2010) which could result in better control of NFM

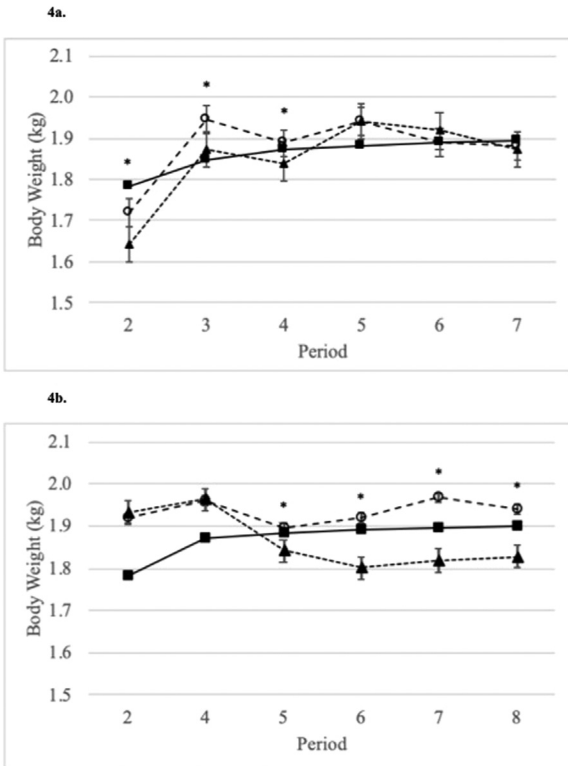

Figure 4. Body weight for Tetra Brown hens from periods 2–8 for Trial 1 (A; 22–47 woa) and Trial 2 (B; 22–49 woa). Lines represent: Tetra Brown management guide (—■—), control hens (- -o- -), northern fowl mite infested hens (--▲--). Each data point is a mean of values for the period (28 days) with SEM. *: differences between control and NFM ($P < 0.05$).

populations by the individual hens (Chen et al., 2011) Lay et al. (2011) wrote that beak trimmed hens can harbor 3 to 10 times more ectoparasites than hens with intact beaks. The large amounts of feather loss due to cannibalistic and feather-pecking behaviors in the flock led to a loss of the ideal environment for the northern fowl mite, as the mites prefer the down-like feathering in the vent region of hens (DeVaney and Augustine, 1987; Vezzoli et al., 2016).

Rather than the low NFM infestation, challenges with cannibalism and feather pecking as well as observed environmental stressors in all four cage-free rooms, such as high humidity and litter moisture, likely depressed production and welfare parameters.

Trial 2. No hen was found to have a mite count score of 7 (greater than 10,000 mites, Table 2) in Trial 2; however, NFM scores as high as 6 (1,001 to 10,000 mites, Table 2) were detected at peak infestation levels (Figure 7). The mite infestation in Trial 2 was much greater and more consistent than in Trial 1, with many hens harboring hundreds of mites at a time. However, a score of 1 was most frequently detected overall. An example of an infested hen is presented in Figure 8. The high NFM infestation levels negatively impacted hen-day percentage, percent livability, and body weight.

Percent livability was much higher in Trial 2 than in Trial 1 (approximately 98.5% and 96.7% for the control and NFM treatments, respectively, by 49 wk of age; Figure 2B), as hens were beak modified at the hatchery and cannibalism was not an issue in Trial 2. An interaction between period and treatment ($P < 0.0001$) was observed in percent livability where the NFM group was 1.8% lower than the control in periods 4 through 8 ($P < 0.002$). The presence of NFM as a covariate ($P < 0.0001$) would suggest that a mite infestation has the capability to impact percent livability in a negative fashion. Livability in Trial 2 was higher

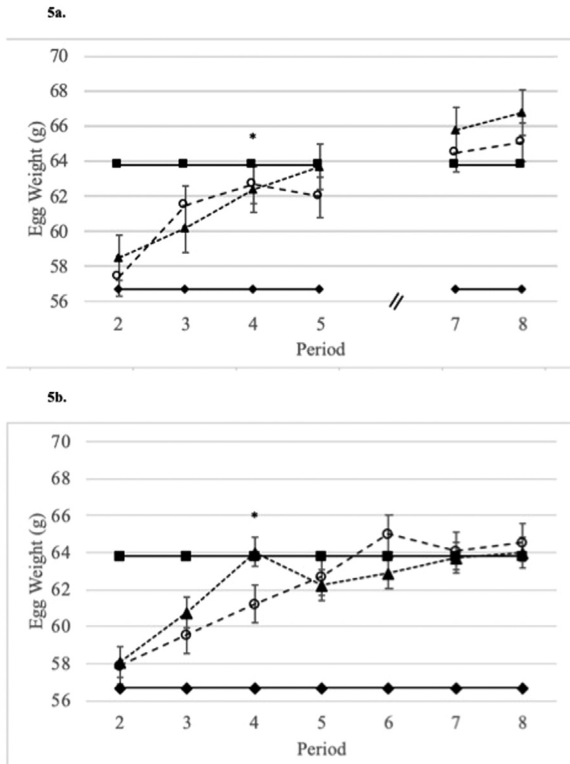

Figure 5. Egg weight for Tetra Brown hens from periods 2-8 for Trial 1 (A; 22−47 woa) and Trial 2 (B; 22−49 woa). Lines represent: control hens (-○-), northern fowl mite infested hens (--▲--) and the US egg industry minimum individual egg weight for large and extra large eggs (—◆— and —■—, respectively; USDA Agricultural Marketing Service, 2017). Each data point is a mean of values for the period (28 days) with SEM. *: differences between control and NFM ($P < 0.05$).

than in Trial 1 and remained above 96.5% for both treatments at the end of the trial; however, the lower livability in the NFM group was below the management guide (Tetra Americana, 2017).

Hen-day percentage (Figure 3B) was impacted by period ($P < 0.0001$). Mullens et al. (2009) reported a 2.1 to 4.0% decrease in hen-day production due to an NFM infestation, while Trial 2 saw a 5.5% decrease in overall hen-day production percentage in the NFM treatment when compared to the control. Hen-day percentage for both the control and NFM groups in Trial 2 remained below the Tetra Brown management guide (Tetra Americana, 2017) after period 2. Egg eating behavior was observed in all 4 cage-free rooms, which likely depressed egg production values and impacted hen-day percentage. Hens in all 4 rooms showed an observed preference for laying eggs in the first 2 colony nests closest to the front of the room, which led to an increased presence of eggs in those nest boxes. Due to the nature of the animal rooms, egg collection belts were static resulting in eggs piling back into the colony nest from the collection belt. This allowed hens to have easy access to the eggs.

Body weight (Figure 4B) was affected by the interaction ($P < 0.0001$) and covariates of mite presence ($P < 0.0001$). Lower body weights were observed in the NFM treatment when compared to the control in periods 5 to 8 ($P < 0.003$). NFM depressed body weight gain after period 4, as mite populations increased on hens. This is consistent with published research reports of depressed body weight gain as a result of NFM infestations (Loomis et al., 1970; Mullens et al., 2009; Owen et al., 2009; Vezzoli et al., 2016).

Egg weight (Figure 5B) was impacted by the interaction of the main effects ($P = 0.003$), with larger egg weights in the NFM group in period

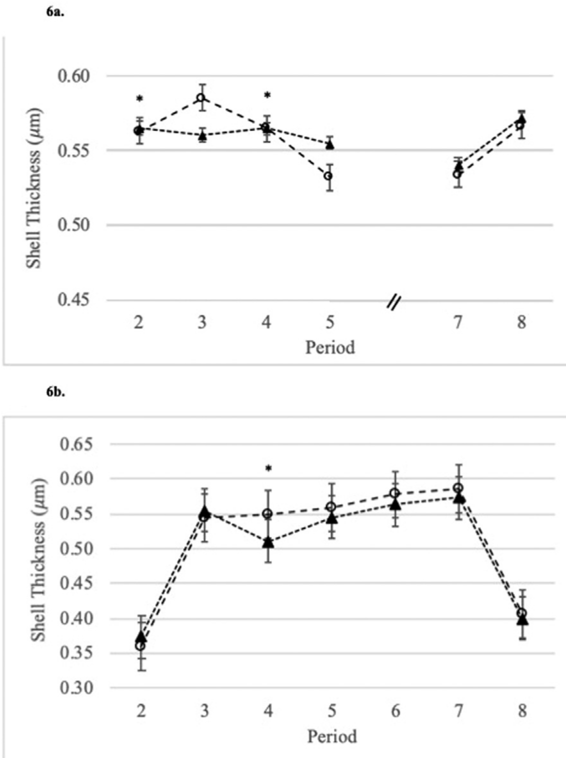

Figure 6. Shell thickness for Tetra Brown hens from periods 2−8 for Trial 1 (A; 22−47 woa) and Trial 2 (B; 22−49 woa). Lines represent: control hens (- -o- -), northern fowl mite infested hens (--▲--). Each data point is a mean of values for the period (28 days) with SEM. *: differences between control and NFM ($P < 0.05$).

4 (P = 0.02). However, NFM egg weight dropped 1.8 g by period 5 resulting in smaller egg weights compared to the control for the remainder of the trial. Percent yolk and albumen were impacted by period ($P < 0.0001$) while percent shell was influenced by period ($P < 0.0001$) and treatment ($P = 0.02$). The overall percent shell for NFM (9.4%) was lower than the control (9.6%; P = 0.02). Shell thickness was affected by the interaction ($P < 0.0001$), and post-hoc analysis showed a thinner shell in the NFM treatment starting in period 4 (P <

Table 3. Percentage of laying hens with corresponding keel palpation scores for Tetra Brown hens from periods 2−7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	78.8	18.8	2.4	79.8	19.0	1.2	$P > 0.05$
3	35.7	61.9	2.4	38.8	59.2	2.0	$P > 0.05$
4	18.2	60.8	21.0	33.9	62.0	4.1	$P > 0.05$
5	25.1	59.0	15.9	45.4	49.5	5.1	$P > 0.05$
6	17.2	70.7	12.1	22.8	69.4	7.8	$P > 0.05$
7	0.8	85.4	13.8	1.4	95.9	2.7	$P > 0.05$

[1]Period is a 28-d span of time; period 2−7 corresponds to 22−47 wk of age.

[2]0 − No keel deviation or deformation. 1 − Keel deviation or deformation < 2 cm. 2 − Keel deviation or deformation ≥ 2 cm. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).

[3]Northern fowl mite.

Table 4. Percentage of laying hens with corresponding keel palpation scores for Tetra Brown hens from periods 2−8[1] in Trial 2. Welfare quality assessments were not recorded during period 3.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	3.1	93.8	3.1	8.2	91.2	0.6	$P = 0.0179$
4	6.9	90.5	2.6	6.0	92.7	1.3	$P > 0.05$
5	1.7	96.6	1.7	7.8	90.9	1.3	$P = 0.007$
6	2.5	95.8	1.7	1.3	96.9	1.8	$P > 0.05$
7	7.4	86.5	6.1	8.4	88.6	3.0	$P > 0.05$
8	0.4	97.5	2.1	0	96.4	3.6	$P > 0.05$

[1]Period is a 28-d span of time; period 2-8 corresponds to 22−49 wk of age.
[2]0 − No keel deviation or deformation. 1 − Keel deviation or deformation < 2 cm. 2 − Keel deviation or deformation ≥ 2 cm. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

Table 5. Percentage of laying hens with tip fractures for Tetra Brown hens from periods 2−7[1] in Trial 1.

	Welfare score[2]				
	Control treatment		NFM[2] treatment		
Period	No	Yes	No	Yes	Treatment effect
2	96.4	3.6	97.5	2.5	$P > 0.05$
3	83.3	16.7	91.8	8.2	$P > 0.05$
4	60.3	39.7	71.3	28.7	$P = 0.0207$
5	72.2	27.8	74.9	25.1	$P > 0.05$
6	52.3	47.7	54.4	45.6	$P > 0.05$
7	3.3	96.7	5.5	94.5	$P > 0.05$

[1]Period is a 28-d span of time; period 2−7 corresponds to 22−47 wk of age.
[2]No − No presence of a tip fracture. Yes − Presence of a tip fracture.
[3]NFM: Northern fowl mite.

0.0001) and sustained through the end of the trial (Figure 6b). A drop in shell thickness of approximately 0.18 μm was observed in period 8 in both the NFM and control treatments. This change in shell thickness could be attributed to increasing egg size over time.

An increase in keel bone damage (Table 4) and tip fractures (Table 6) was found over time, as well as worsening foot condition and increased presence of comb wounds (Tables 8 and 10). Keel bone damage was worse in the control group when compared to the NFM

Table 6. Percentage of laying hens with tip fractures for Tetra Brown hens from periods 2−8[1] in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[2]				
	Control treatment		NFM[3] treatment		
Period	No	Yes	No	Yes	Treatment effect
2	8.6	91.4	15.1	84.9	$P > 0.05$
4	10.8	89.2	7.8	92.2	$P > 0.05$
5	2.2	97.8	7.4	92.6	$P > 0.05$
6	3.0	97.0	1.8	98.2	$P > 0.05$
7	24.0	76.0	26.9	73.1	$P > 0.05$
8	0.4	99.7	1.4	98.6	$P > 0.05$

[1]Period is a 28-d span of time; period 2−8 corresponds to 22−49 wk of age
[2]No − No presence of a tip fracture. Yes − Presence of a tip fracture.
[3]Northern fowl mite.

Table 7. Percentage of laying hens with corresponding foot pad condition scores for Tetra Brown hens from periods 2–7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
3	100	0	0	100	0	0	$P > 0.05$
4	82.5	17.1	0.4	78.2	21.8	0	$P > 0.05$
5	42.1	56.4	1.5	48.0	52.0	0	$P > 0.05$
6	79.4	20.6	0	54.7	45.3	0	$P > 0.05$
7	19.5	78.1	2.4	21.1	78.9	0	$P > 0.05$

[1]Period is a 28-d span of time; period 2–7 corresponds to 22–47 wk of age.

[2]0 – No or minimal epithelium proliferation and no wounds. 1 – Epithelium proliferation on the foot pad or presence of necrosis or bumble foot, with no or moderate swelling. 2 – Swollen foot pad that is visible from above the foot. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).

[3]Northern fowl mite.

Table 8. Percentage of laying hens with corresponding foot pad condition scores for Tetra Brown hens from periods 2–8 (22–49 wk of age) in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[1]						
	Control treatment			NFM[2] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	79.6	20.4	0	81.8	18.2	0	$P > 0.05$
4	69.8	30.2	0	71.1	28.5	0.4	$P > 0.05$
5	89.7	10.3	0	84.9	14.3	0.8	$P > 0.05$
6	61.7[a]	38.3[a]	0[a]	80.6[a]	18.1[a]	1.3[a]	$P < 0.0001$
7	75.6[a]	24.0[a]	0.4[a]	84.1[a]	14.1[a]	1.8[a]	$P = 0.0301$
8	5.6	93.6	0.8	13.2	84.1	2.7	$P > 0.05$

[1]Period is a 28-day span of time; period 2–8 corresponds to 22–49 wk of age.

[2]0 – No or minimal epithelium proliferation and no wounds. 1 – Epithelium proliferation on the foot pad or presence of necrosis or bumble foot, with no or moderate swelling. 2 – Swollen foot pad that is visible from above the foot. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).

[3]NFM: Northern fowl mite.

Table 9. Percentage of laying hens with corresponding comb wound scores for Tetra Brown hens from periods 2–7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
3	100	0	0	100	0	0	$P > 0.05$
4	95.3	4.3	0.5	96.9	3.1	0	$P > 0.05$
5	96.6	2.6	0.6	95.4	4.6	0	$P > 0.05$
6	97.7	2.3	0	97.8	2.2	0	$P > 0.05$
7	94.3	5.7	0	98.0	2.0	0	$P > 0.05$

[1]Period is a 28-d span of time; period 2–7 corresponds to 22–47 wk of age.

[2]0 – No pecking wounds. 1 – Less than 3 pecking wounds. 2 – 3 or more pecking wounds. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).

[3]Northern fowl mite.

Table 10. Percentage of laying hens with corresponding comb wound scores for Tetra Brown hens from periods 2−8[1] in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	96.3	3.7	0	97.5	2.5	0	$P > 0.05$
4	96.1	3.5	0.4	96.5	3.5	0	$P > 0.05$
5	92.7	7.3	0	91.4	8.2	0.4	$P > 0.05$
6	93.2	6.8	0	91.6	8.4	0	$P > 0.05$
7	98.7	1.3	0	95.6	4.4	0	$P > 0.05$
8	74.8	24.8	0.4	84.1	14.1	1.8	$P > 0.05$

[1]Period is a 28-d span of time; period 2−8 corresponds to 22−49 wk of age.
[2]0 − No pecking wounds. 1 − Less than 3 pecking wounds. 2 − 3 or more pecking wounds. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

Table 11. Percentage of laying hens with corresponding welfare scores for feather damage on the crop region for Tetra Brown hens from periods 2-7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
3	100	0	0	100	0	0	$P > 0.05$
4	95.3	3.8	0.9	93.3	6.2	0.5	$P > 0.05$
5	93.9	4.1	2.0	91.8	4.6	3.6	$P > 0.05$
6	95.4	4.0	0.6	65.4	18.1	16.5	$P < 0.0001$
7	6.5	22.8	70.7	16.3	28.6	55.1	$P = 0.0046$

[1]Period is a 28-d span of time; period 2−7 corresponds to 22−47 wk of age.
[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality® Assessment Protocol (WQ® Consortium, 2009).
[3]Northern fowl mite.

Table 12. Percentage of laying hens with corresponding welfare scores for feather damage on the crop region for Tetra Brown hens from periods 2−8[1] in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
4	99.6	0.4	0	100	0	0	$P > 0.05$
5	99.1	0.9	0	99.1	0.9	0	$P > 0.05$
6	99.1	0	0.9	94.3	5.7	0	$P = 0.0114$
7	85.6	11.3	3.1	68.7	22.5	8.8	$P < 0.0001$
8	54.3	40.6	5.1	27.2	60.6	12.2	$P < 0.0001$

[1]Period is a 28-d span of time; period 2−8 corresponds to 22−49 weeks of age.
[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality® Assessment Protocol (WQ® Consortium, 2009).
[3]Northern fowl mite.

Table 13. Percentage of laying hens with corresponding welfare scores for feather damage on the keel region for Tetra Brown hens from periods 2−7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
3	100	0	0	100	0	0	$P > 0.05$
4	94.7	5.3	0	98.5	1.5	0	$P > 0.05$
5	99.0	1.0	0	97.5	1.5	1.0	$P > 0.05$
6	97.1	1.7	1.2	98.9	0.6	0.5	$P > 0.05$
7	13.8	29.3	56.9	25.2	43.5	31.3	$P < 0.0001$

[1]Period is a 28-d span of time; period 2−7 corresponds to 22−47 wk of age.
[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

Table 14. Percentage of laying hens with corresponding welfare scores for feather damage on the keel region for Tetra Brown hens from periods 2−8[1] in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
4	97.0	3.0	0	88.4	11.6	0	$P = 0.001$
5	88.9	9.8	1.3	96.1	3.0	0.9	$P = 0.0052$
6	80.5	15.3	4.2	70.5	26.0	3.5	$P = 0.0195$
7	52.9	34.9	12.2	26.9	44.0	29.1	$P < 0.0001$
8	29.9	58.1	12.0	15.5	66.8	17.7	$P = 0.0004$

[1]Period is a 28-d span of time; period 2−8 corresponds to 22−49 wk of age.
[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

Table 15. Percentage of laying hens with corresponding welfare scores for feather damage on the belly region for Tetra Brown hens from periods 2−7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
3	97.6	2.4	0	98.0	2.0	0	$P > 0.05$
4	88.6	7.1	4.3	88.7	7.2	4.1	$P > 0.05$
5	99.5	0.5	0	98.5	0	1.5	$P > 0.05$
6	94.9	1.1	4.0	99.5	0	0.5	$P = 0.032$
7	6.5	8.1	85.4	12.2	12.2	75.6	$P = 0.0439$

[1]Period is a 28-d span of time; period 2−7 corresponds to 22−47 wk of age.
[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

Table 16. Percentage of laying hens with corresponding welfare scores for feather damage on the belly region for Tetra Brown hens from periods 2–8[1] in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
4	100	0	0	100	0	0	$P > 0.05$
5	94.0	6.0	0	94.4	5.6	0	$P > 0.05$
6	94.9	3.8	1.3	88.1	11.5	0.4	$P = 0.0121$
7	93.4	5.7	0.9	94.7	5.3	0	$P > 0.05$
8	81.2	17.9	0.9	77.7	21.4	0.9	$P > 0.05$

[1]Period is a 28-d span of time; period 2−8 corresponds to 22−49 wk of age.

[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).

[3]Northern fowl mite.

Table 17. Percentage of laying hens with corresponding welfare scores for feather damage on the head region for Tetra Brown hens from periods 2–7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[2] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
3	100	0	0	100	0	0	$P > 0.05$
4	99.1	0.9	0	95.9	3.6	0.5	$P > 0.05$
5	86.8	12.7	0.5	68.9	11.7	19.4	$P < 0.0001$
6	71.4	26.3	2.3	58.3	29.1	12.6	$P = 0.0024$
7	26.8	35.0	38.2	30.6	40.1	29.3	$P > 0.05$

[1]Period is a 28-d span of time; period 2−7 corresponds to 22−47 wk of age.

[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality® Assessment Protocol (WQ® Consortium, 2009).

[3]NFM: Northern fowl mite.

group in periods 2 ($P < 0.05$) and 5 ($P < 0.01$; Table 4), and foot condition was worse in the control group when compared to the NFM treatment in periods 6 ($P < 0.0001$) and 7 ($P < 0.05$; Table 8). The increased keel bone damage and worse foot condition in the control treatment was due to environmental differences between the groups, such as activity levels or litter

Table 18. Percentage of laying hens with corresponding welfare scores for feather damage on the head region for Tetra Brown hens from periods 2–8[1] in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
4	96.5	2.6	0.9	97.8	2.2	0	$P > 0.05$
5	97.0	2.1	0.9	97.4	2.2	0.4	$P > 0.05$
6	97.0	2.1	0.9	96.9	2.2	0.9	$P > 0.05$
7	95.6	1.8	2.6	97.4	0.9	1.7	$P > 0.05$
8	92.3	6.0	1.7	78.3	19.5	2.2	$P < 0.0001$

[1]Period is a 28-d span of time; period 2−8 corresponds to 22-49 wk of age.

[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality® Assessment Protocol (WQ® Consortium, 2009).

[3]Northern fowl mite.

Table 19. Percentage of laying hens with corresponding welfare scores for feather damage on the neck region for Tetra Brown hens from periods 2–7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
3	100	0	0	100	0	0	$P > 0.05$
4	98.1	1.4	0.5	96.9	2.6	0.5	$P > 0.05$
5	73.6	17.3	9.1	57.1	23.5	19.4	$P = 0.0004$
6	34.3	48.0	17.7	17.6	40.1	42.3	$P < 0.0001$
7	4.9	17.1	78.0	7.5	28.6	63.9	$P = 0.0144$

[1]Period is a 28-d span of time; period 2–7 corresponds to 22–47 wk of age.
[2]0 – No wear or damage on feather coverage. 1 – One or more featherless areas < 5 cm in diameter. 2 – One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality® Assessment Protocol (WQ® Consortium, 2009).
[3]Northern fowl mite.

Table 20. Percentage of laying hens with corresponding welfare scores for feather damage on the neck region for Tetra Brown hens from periods 2–8[1] in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
4	97.8	1.7	0.5	99.6	0.4	0	$P > 0.05$
5	98.3	1.3	0.4	98.3	1.3	0.4	$P > 0.05$
6	97.0	2.1	0.9	97.8	1.3	0.9	$P > 0.05$
7	94.8	4.4	0.8	96.0	3.1	0.9	$P > 0.05$
8	85.0	14.1	0.9	37.6	57.5	4.9	$P < 0.0001$

[1]Period is a 28-d span of time; period 2–8 corresponds to 22–49 wk of age.
[2]0 – No wear or damage on feather coverage. 1 – One or more featherless areas < 5 cm in diameter. 2 – One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

condition in the rooms. Feather damage worsened over time, with more frequent feather damage in the NFM treatment when compared to the control treatment in later periods (Tables 10–24). Post-hoc analysis showed increased feather damage in the NFM treatment when compared to the control on the crop in periods 6 ($P < 0.05$), 7, and 8 ($P < 0.0001$; Table 12);

Table 21. Percentage of laying hens with corresponding welfare scores for feather damage on the back region for Tetra Brown hens from periods 2–7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	97.6	2.4	0	86.1	6.3	7.6	$P = 0.0152$
3	64.3	26.2	9.5	83.7	10.2	6.1	$P = 0.049$
4	82.0	12.8	5.2	54.9	25.1	20.0	$P < 0.0001$
5	57.9	18.2	23.9	52.6	14.8	32.6	$P > 0.05$
6	58.9	30.8	10.3	31.9	25.8	42.3	$P < 0.0001$
7	7.3	40.7	52.0	23.8	42.9	33.3	$P = 0.0001$

[1]Period is a 28-d span of time; period 2–7 corresponds to 22–47 wk of age.
[2]0 – No wear or damage on feather coverage. 1 – One or more featherless areas < 5 cm in diameter. 2 – One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

Table 22. Percentage of laying hens with corresponding welfare scores for feather damage on the back region for Tetra Brown hens from periods 2−8[1] in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
4	99.1	0.9	0	100	0	0	$P > 0.05$
5	98.3	1.7	0	99.6	0	0.4	$P > 0.05$
6	96.6	3.4	0	99.1	0.9	0	$P > 0.05$
7	91.7	5.2	3.1	93.4	6.2	0.4	$P > 0.05$
8	79.1	19.6	1.3	62.0	36.2	1.8	$P = 0.0001$

[1]Period is a 28-d span of time; period 2−8 corresponds to 22−49 wk of age.
[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

Table 23. Percentage of laying hens with corresponding welfare scores for feather damage on the rump region for Tetra Brown hens from periods 2−7[1] in Trial 1.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	63.5	4.7	31.8	70.9	6.3	22.8	$P > 0.05$
3	14.3	47.6	38.1	28.6	40.8	30.6	$P > 0.05$
4	24.2	34.6	41.2	17.4	30.3	52.3	$P = 0.0205$
5	3.6	33.5	62.9	3.6	35.2	61.2	$P > 0.05$
6	12.0	41.1	46.9	4.4	26.9	68.7	$P < 0.0001$
7	0.8	9.8	89.4	5.4	15.7	78.9	$P = 0.0186$

[1]Period is a 28-d span of time; period 2−7 corresponds to 22−47 wk of age.
[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

on the keel in periods 4 to 8 ($P < 0.05$; Table 14), on the belly in period 6 ($P < 0.05$; Table 16), and on the head in period 8 ($P < 0.0001$; Table 18). Post-hoc analysis also showed increased feather damage on the neck, rump, and back in period 8 ($P < 0.001$) in the NFM treatment when compared to the control (Tables 20−24. The NFM infestation could

Table 24. Percentage of laying hens with corresponding welfare scores for feather damage on the rump region for Tetra Brown hens from periods 2−8[1] in Trial 2. Welfare quality assessments were not recorded at period 3.

	Welfare score[2]						
	Control treatment			NFM[3] treatment			
Period	0	1	2	0	1	2	Treatment effect
2	100	0	0	100	0	0	$P > 0.05$
4	99.6	0.4	0	100	0	0	$P > 0.05$
5	97.4	2.2	0.4	98.7	0.9	0.4	$P > 0.05$
6	95.3	4.2	0.4	95.1	4.9	0	$P > 0.05$
7	86.0	10.9	3.1	82.4	15.9	1.7	$P > 0.05$
8	50.4	48.7	0.9	34.4	62.9	2.7	$P = 0.0004$

[1]Period is a 28-d span of time; period 2−8 corresponds to 22−49 wk of age.
[2]0 − No wear or damage on feather coverage. 1 − One or more featherless areas < 5 cm in diameter. 2 − One or more featherless areas ≥ 5 cm in diameter. From the Welfare Quality Assessment Protocol (WQ Consortium, 2009).
[3]Northern fowl mite.

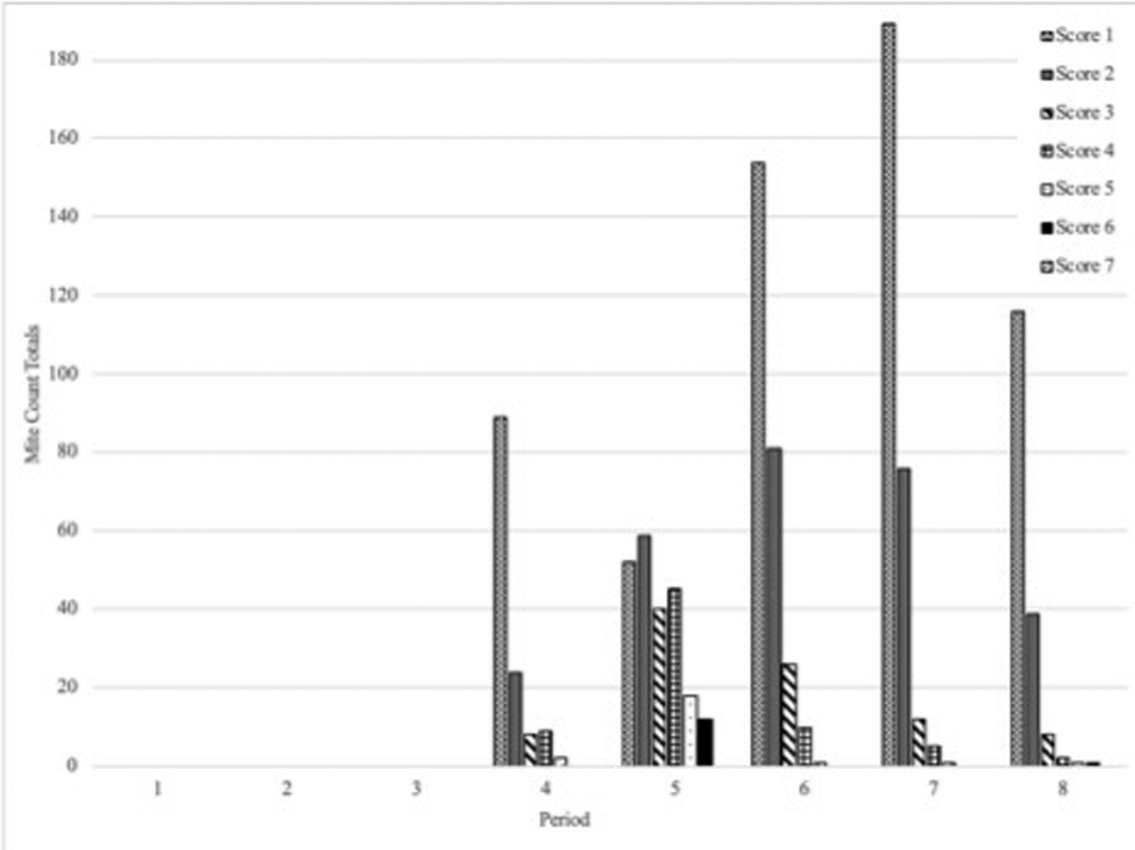

Figure 7. Frequency of scores collected during mite count data between periods 4−8, after introduction of mites had occurred at period 3 in Trial 2.

Figure 8. Vent feathers on a northern fowl mite infested Tetra Brown laying hen. Mite casings and waste can be seen on the feathers, while mites can be seen on the skin of the bird, just below the vent feathers.

have led to increased hen grooming behavior as a result of irritation from mites' blood-feeding on the hens.

Summary

Trial 2 experienced higher percent livability than Trial 1. Percent livability hovered above 90% in Trial 2, while it steadily decreased in Trial 1, ending around approximately 47.5% between the 2 treatments due to extreme cannibalism (Figures 2A and 2B). Hen-day percentage in Trial 2 was lower overall than Trial 1 (between 73 and 78% compared to 81 and 89%; Figures 3A and 3B). Body weight was more uniform in Trial 2, hovering between approximately 1.8 and 2.0 kg, while body weight hovered between approximately 1.6 and 2.0 kg in Trial 1 (Figures 4A and 4B). Egg weights and egg components (shell weight, albumen weight, and yolk weight expressed as percentage of intact egg weights) were similar between Trials 1 and 2 (Figures 5A and 5B). However, shell thickness values were lower overall (regardless of treatment) in Trial 2 compared to Trial 1; the average shell thickness between treatments was 0.507 mm in Trial 2 and was 0.501 mm in Trial 1 (Figures 6A and 6B). Trial 2 experienced greater keel bone damage and tip fractures than in Trial 1 (Tables 3 and 4; Tables 5 and 6), while Trial 1 experienced greater foot damage and comb wounds than Trial 2 (Tables 7 and 8; Tables 9 and 10). Feather damage overall was more prevalent in Trial 1 than in Trial 2 (Tables 11−23; Tables 12−24) and was due to feather pecking and cannibalistic behaviors.

CONCLUSIONS AND APPLICATIONS

1. NFM negatively impacts percent livability, body weight, and results in an increase in feather damage and decreased hen-day percentage when infestation levels are consistent and high.
2. Environmental and management differences in cage-free systems play a role in NFM populations including how they spread and how they impact flock welfare and production.
3. Cannibalism challenges in Trial 1 and egg eating behavior in Trial 2 were limitations to this study, as they impacted production numbers, but are a reality of both small-scale and commercial cage-free egg production, especially where hens are not beak treated.
4. Variations of cage-free systems could result in different hen welfare and production findings depending on management, hen genetic strain, and complexity of the system.

DISCLOSURES

The authors have no conflicts of interest to report.

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SUPPLEMENTARY MATERIALS

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.japr.2022.100290.

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