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# A novel environmental enrichment device improved broiler performance without sacrificing bird physiological or environmental quality measures

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# A novel environmental enrichment device improved broiler performance without sacrificing bird physiological or environmental quality measures

## Abstract

Modern commercial broilers have been genetically selected for fast growth and heavy breast muscling, contributing to a top-heavy phenotype and increased leg lameness. A quick-growing phenotype coupled with poor leg health fosters inactivity. The objective of this study was to stimulate broiler movement using novel environmental enrichment and determine the impact of movement on production, leg health, and environmental parameters. A total of 1,200 Ross 308 broilers were housed in 40 pens with 30 birds/pen for 6 wk in 2 separate rooms (laser enrichment or control). Each enrichment device was mounted above 2 adjoining pens, projected 2 independent, randomly moving laser beams at the floor to stimulate innate predatory behavior, and was active 4 times daily in 4-min periods. Performance outcomes were calculated by pen and averaged per bird for each performance period and overall days 0 to 42. A total of 70 randomly selected focal birds were examined for breast blisters and footpad dermatitis each week and euthanized on day 42 for tibia quality measures. Air quality and litter moisture were sampled by week. Laser-enriched pens had greater average bird feed intake in starter ( $P < 0.001$ ), grower ( $P = 0.004$ ), finisher periods ( $P = 0.004$ ), and overall days 0 to 42 (0.19 kg/bird;  $P = 0.0003$ ). Average bird weight gain was also increased in enriched pens in each performance period: starter ( $P = 0.043$ ), grower ( $P = 0.001$ ), finisher ( $P < 0.001$ ), and overall days 0 to 42 (0.24 kg/bird;  $P < 0.001$ ). Enriched pens had improved feed conversion ratio (**FCR**) vs. control with a decrease of 3 FCR points in the grower ( $P = 0.031$ ), 18 points in the finisher ( $P < 0.001$ ), and 7 points overall ( $P < 0.001$ ). Enriched pens had higher ADG during starter ( $P = 0.048$ ), finisher ( $P < 0.001$ ), and overall (5.7 g/bird/d;  $P < 0.001$ ). No differences were found in breast blister, footpad dermatitis, tibia, air, or litter quality measures ( $P > 0.05$ ). In summary, a novel enrichment device based on bird visual feeding and predatory instincts positively affected performance through decreased FCR and increased ADG without sacrificing external animal-based measures, tibia quality, or air or litter quality.

## Keywords

broiler, welfare, environmental enrichment, performance, sustainability

## Disciplines

Agriculture | Animal Sciences | Poultry or Avian Science

## Comments

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## BROILER ENRICHMENT

### **A novel environmental enrichment device improved broiler performance without sacrificing bird physiological or environmental quality measures**

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## BROILER ENRICHMENT

**ABSTRACT** Modern commercial broilers have been genetically selected for fast growth and heavy breast muscling, contributing to a top-heavy phenotype and increased leg lameness. A quick-growing phenotype coupled with poor leg health fosters inactivity. The objective of this study was to stimulate broiler movement using novel environmental enrichment and determine the impact of movement on production, leg health, and environmental parameters. Twelve-hundred Ross 308 broilers were housed in 40 pens with 30 birds/pen for 6wk in 2 separate rooms (laser enrichment or control). Each enrichment device was mounted above 2 adjoining pens, projected 2 independent, randomly-moving laser beams at the floor to stimulate innate predatory behavior, and was active 4 times daily in 4-min periods. Performance outcomes were calculated by pen and averaged per bird for each performance period and overall d0-42. Seventy randomly-selected focal birds were examined for breast blisters and footpad dermatitis each wk and euthanized on d42 for tibia quality measures. Air quality and litter moisture were sampled by wk. Laser-enriched pens had greater average bird feed intake in starter ( $P<0.001$ ), grower ( $P=0.004$ ), finisher periods ( $P=0.004$ ); and overall d0-42 (0.19 kg/bird;  $P=0.0003$ ). Average bird weight gain was also increased in enriched pens in each performance period: starter ( $P=0.043$ ), grower ( $P=0.001$ ), finisher ( $P<0.001$ ), and overall d0-42 (0.24 kg/bird;  $P<0.001$ ). Enriched pens had improved feed conversion ratio (**FCR**) vs. control with a decrease of 0.03 FCR points in the grower ( $P=0.031$ ), 0.18 points in the finisher ( $P<0.001$ ), and 0.07 points overall ( $P<0.001$ ). Enriched pens had higher ADG during starter ( $P=0.048$ ), finisher ( $P<0.001$ ), and overall (5.7 g/bird/ d;  $P<0.001$ ). No differences were found in breast blister, footpad dermatitis, tibia, air, or litter quality measures ( $P>0.05$ ). In summary, a novel enrichment device based on bird visual feeding and predatory instincts positively affected performance through decreased FCR and

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increased ADG without sacrificing external animal-based measures, tibia quality, or air or litter quality.

**Key words:** broiler, welfare, environmental enrichment, performance, sustainability

# BROILER ENRICHMENT

## INTRODUCTION

Due to intense genetic selection, modern broilers weigh 4 to 5 times more than broiler lines from the 1950's at the same timepoints, and are 2 to 3 times more feed efficient (Havenstein, et al., 2003; Zuidhof, et al., 2014). However, this selection for increased growth rate has contributed to up to 30% of modern commercial broilers being affected by leg lameness or poor locomotion (Knowles, et al., 2008; Bassler, et al., 2013). Lameness leads to increased time spent lying down, which in turn increases litter contact and could result in a higher breast blister occurrence and contact dermatitis (Weeks, et al., 2000; Nääs, et al., 2009; Bassler, et al., 2013). Further, lack of activity compounds lameness by negatively affecting bone strength, mass, and ability to bear weight properly (Lanyon, 1993; Rath, et al., 2000). Contact dermatitis-driven tissue damage may be caused by urea in the litter generating ammonia, creating a chemical burn effect and is likely painful. The prevalence of this issue, affecting 21.87% of Ross 308 broilers in a recent year-long study, may be reflective of air and litter quality in the house (Haslam, et al., 2006; Dinev, et al., 2019). The National Chicken Council (NCC, 2017) Animal Welfare Guidelines and Audit Checklist for Broilers require that ammonia in the air never exceeds 25 ppm at bird height, and litter must be evaluated for friability and moisture.

Severe lameness hinders birds from accessing feed and water, and negatively impacts the industry economically, necessitating up to 2% culls in a \$30 billion industry (Dunkley, 2007; USDA, 2017). In addition, studies have reported that broilers with severe leg lameness eat more analgesic-containing feed than healthy birds, and birds fed an analgesic diet showed improved speed of walking, indicating relief from pain and discomfort caused by leg abnormalities (McGeown, et al., 1999; Danbury, et al., 2000). The 2017 NCC broiler audit guidelines have recognized lameness as a detrimental welfare issue and recommend gait scoring 100 birds per flock to evaluate leg health within 1 wk of slaughter, and footpad scoring 200 paws at slaughter.

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Past research in laying hens has shown that restricting exercise had a clear, negative impact on bird skeletal health (Shipov, et al, 2010), and work in broilers has shown that motivating physical activity increased tibia strength and decreased lameness (Reiter and Bessei, 2009). Prayitno and others (1997) concluded that activity driven by red light, early and late in the rearing period, improved locomotion compared to a blue light treatment where broilers were less active. Birds are visual feeders and prefer red and orange colors over green and blue (Ham and Osorio, 2007). Bizeray et al. (2002) studied the effects of red, blue, green, and yellow spotlights moving across the floor but did not see a change in broiler physical activity, and the authors concluded that that the spotlights moved too quickly. Baxter and others (2019) implemented perches and dust baths but did not see an effect of enrichment on foraging, play, or activity, nor, in a separate paper published on the same study, leg health (Bailie et al, 2018). Platform use by broilers in Norring et al. (2016) likewise did not increase overall activity. A study by Jordan and others (2011) showed that broiler activity and foraging was increased by scattering feed in the litter, but broilers in the enriched treatment had 13% lower weights at harvest.

Certainly, some forms of environmental enrichment have been shown to improve broiler welfare outcomes, as in Ventura et al. (2012), where barrier perches stimulated natural perching behavior and reduced aggressive interaction and rest disturbances compared to the control. Recent work by BenSassi and others (2019) showed that increasing environmental complexity was associated with fewer skin concerns, lower mortality, fewer underweight birds, a lower overall rejection rate at harvest, and less welfare problems overall. However, an enrichment option designed to motivate broiler activity and improve physical and performance outcomes is still lacking in the published literature. Thus, we developed a novel form of environmental enrichment designed to motivate physical movement through visual stimulation. The objectives

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of this work were to determine the impact on bird physiology (leg lameness and footpad and breast condition), performance, and environment (air and litter quality).

### **MATERIALS AND METHODS**

All live bird procedures were approved by the Iowa State University Institutional Animal Care and Use Committee.

#### ***Animals***

Twelve hundred and sixty straight-run Ross 308 broiler chicks (day of hatch; BW  $47.38 \pm 0.14$  g) were obtained from a commercial hatchery and transported to the Poultry Research and Teaching Unit at Iowa State University (International Poultry Breeders Hatchery, Bancroft, Iowa). Twelve hundred were randomly assigned to treatment groups and the remainder were culled. A subset of 70 birds were randomly assigned upon arrival as focal birds, identified with wing-bands, and marked with unique animal-safe food coloring (red, blue, green, purple, and black; Wilton, Woodridge, Illinois). Half of the focal birds were assigned to laser-enriched pens, and half were assigned to control pens (n=5 focal birds/pen in 14 pens). Food coloring was applied to a cotton ball, rubbed on the back of the chick's head and neck, and reapplied on an as-needed basis throughout the trial.

#### ***Housing and Feeding***

Birds were housed in 40 floor pens (30 birds/pen) measuring 1.22 by 2.44 m across 2 rooms in the barn (20 pens/room). One room contained 20 enriched pens (exposed to laser device), and the other contained 20 control pens, with an anteroom separating the 2 so no crossover of enrichment device was possible. Approximately 10 cm deep fresh wood shavings provided bedding over the solid concrete floor, and PVC pipe dividers with mesh walls (1.22 m



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height) separated pens. High and low temperatures and humidity were monitored daily in the enriched and control rooms of the barn. Average temperatures are listed from the starter, grower, and finisher periods respectively from the enriched room: 85.47°C, 77.39°C, and 71.71°C, and the control room: 85.53°C, 77.46°C, and 71.50°C. Average relative humidity is listed from the starter, grower, and finisher periods respectively from the enriched room: 23.86%, 27.21%, and 33.93%, and the control room of the barn: 19.89%, 23.93%, 27.75%.

Birds were gradually adjusted from 24 h light on day 0-7 (30-40 lux) to 20 h light (20-30 lux) from days 8-42. Chicks were brooded with 2 heat lamps/pen (22.9 cm reflectors with porcelain socket) using 125-watt heat bulbs (Sylvania, Wilmington, MA) for the first wk. Birds were fed an *ad libitum* diet formulated for Ross 308 commercial recommendations (Table 1) out of a hanging chicken feeder (BRHF151, Brower Equipment, Houghton, IA) gradually raised to accommodate bird height. Water was provided *ad libitum* from a hanging nipple water line (8 nipples/pen).

### ***Laser Enrichment Device***

A total of 10 laser enrichment devices designed and built specifically for this research were affixed over 20 pens in 1 room of the broiler barn. Each device was designed and calibrated to cover 2 adjoining pens. The enrichment device consisted of 2 independent red 650 nm lasers contained within a 20.5 by 20.5 cm metal box with a glass bottom mounted on a custom-designed structure made of 3 wooden beams (2.4 m height) raised above the pens. The lasers projected in the direction of the pen floor and moved in a random pattern at a variable speed between 7.6-30.5 cm/s for 4-min “laser periods”: 05:30 to 05:34, 11:30 to 11:34, 17:30 to 17:34, and 23:30 to 23:34 daily for the entirety of the trial. Overhead snapshots of the activated laser in the pens are shown in supplementary Figures 1A-D for day 2, 16, 30, and 37. As this device was

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novel, and there is no explanation of broiler attention span in the current literature, the 4-min length of laser periods was tested with the knowledge that it would need to be validated and may need fine-tuning in future studies. The decision to expose broilers to laser periods 4 times/d was based off work by Jones and others (2000), which showed that laying hens exposed to environmental enrichment in the form of strings for limited daily time periods (10 min), rather than constant exposure, maintained interest in pecking the strings for 14 weeks.

### *Performance*

The 6-wk trial was separated into a starter, grower, and finisher period that were 2-wk in length. All birds in each pen were weighed as a group, and then focal birds were weighed individually at the start of each period to determine weight gain. Feed disappearance/intake (**FI**) was recorded throughout. Feed conversion ratio (**FCR**) and ADG were calculated by pen and averaged by number of birds in the pen.

### *Breast Blisters and Footpad Dermatitis*

Focal birds were examined the same day each week of the trial by the same researcher, on a different day than birds were weighed, in their home pens for breast blisters and footpad dermatitis, with all birds examined on d42. Both examinations took place at the same day and time each wk and were done by the same researcher. Footpad dermatitis was scored pass/fail using the American Association of Avian Pathologists Paw Scoring system (2015), where a normal yellow color or slight discoloration with hyperkeratosis on an area less than 1/2 of the footpad was scored a pass, and erosions, ulcerations, scabs, hemorrhages, or swelling on an area greater than 1/2 of the footpad was scored a fail. Breast blisters were scored on a present/absent basis based on methods use by Greene and others (1985) where blisters were considered present

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when a blister was equal to or larger than 1.27 cm<sup>2</sup>, when there were 1 or more breast burns, or when there were scabs on breast skin. A brownish-colored scab would be considered “mild” and an ulcer with black exudates was considered “severe”.

### *Tibia Quality*

On day 42, focal birds were euthanized using carbon dioxide and the right tibia was collected from each bird and frozen at -20°C until further analysis. Tibia (n=70) were thawed overnight, weighed, and scanned using dual energy x-ray absorptiometry (**DXA**, Hologic, Marlborough, Massachusetts). The bones were scanned in groups of seven using the validated “rat whole body scan” protocol for bone mineral density (**BMD**) and bone mineral content (**BMC**).

Bone breaking strength of focal bird tibia was measured using the tensile test and compression method on an Instron 3367 Universal Test Machine (Norwood, Massachusetts). The machine had a 30 kN load capacity and 2 platons controlled to fracture the bone between them. Each tibia was individually fractured in a plastic bag wrapped in cheesecloth to prevent contamination of the machine or slippage due to the bag. Each tibia was placed on the bottom platon with the lateral/medial condyle end of the bone intentionally placed over the edge, out of reach of the platons, and the bend of the tibia facing down. The test was set up so that the top platon moved vertically downwards towards the bone at a rate of 10 mm/min and a 15% rate of load. The machine was stopped at the distinct rapid decline in force (visualized on the monitor) and simultaneous sound of the bone fracturing. Load (kgf) was recorded at the point of break, and divided by area of tibia (cm<sup>2</sup>, obtained from DXA scanning) to calculate bone breaking strength per manufacturer recommendations (Instron; Norwood, MA).

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### *Air and Litter Quality*

Ammonia in the air (ppm) was measured at bird height in the front, middle, and back of each room on one d/wk wks 2-6 with a hand-held ammonia monitor (GasAlert Extreme, BW Technologies, Schaumburg, Illinois) and ammonia test strips. The ammonia monitor was titrated every 14 days with an ammonia tank and provided an exact value, while the strips provided a range of 5 ppm. Litter quality was analyzed weekly according to the NCC Audit Guidelines. Litter moisture was evaluated in 3 randomly-selected pens in the front, middle, and back of each room of the barn. One handful of litter sample was gathered from 3 sections; litter within 15 cm of the water line of each pen was intentionally excluded. Litter quality was scored pass/fail by the same researcher weekly; to pass litter must be “loosely compacted when squeezed in the hand. If the litter remains in a clump when it is squeezed in the hand, it is too wet” (NCC, 2017).

### *Statistical Analysis*

In this experimental design, individual control pens (n=20) were considered the experimental units, and laser-enriched pens were analyzed as a group of 2 pens with a shared laser device (n=10). Room within the barn was confounded by laser treatment, thus was not included in the model, but environmental conditions, management, and feeding were kept as identical as possible between both rooms. All data were analyzed using SAS software version 9.4 (SAS Institute Inc.; 2016). PROC UNIVARIATE was used to assess the distribution of data prior to analysis. Performance and tibia quality data were normally distributed, hence were analyzed using PROC MIXED, a mixed linear model, with treatment as a main effect. Principal Component Analysis (PROC PRIN COMP) was used to test for redundancy and correlation within the bone quality measures, and then Multidimensional Preference Analysis (using PROC PRIN QUAL) was performed to visualize the correlation between variables and reduction to 2

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components. Air quality measures were analyzed using PROC FREQUENCY and CHI SQUARE to determine the distribution and association of readings by treatment. For all measures, a value of  $P \leq 0.05$  was considered significant and differences between means were detected using PDIFF.

### RESULTS

#### *Performance*

All performance measures, including FI, weight gain, FCR, and ADG were averaged per bird by each 2-wk performance period and overall (d0-d42). Feed intake was increased in laser-enriched birds in all periods compared to the control: 4% increase in the starter,  $P < 0.001$ ; 3.1% in the grower,  $P = 0.004$ ; 5.1% in the finisher,  $P = 0.004$ ; and 3.9% overall,  $P = 0.003$  (Table 2). Enriched birds had an increased intake of 5.52 kg/pen overall compared to the control ( $P = 0.006$ ). Weight gain was also increased in laser-enriched birds in each performance period when compared to the control: 2.6% in the starter,  $P = 0.043$ ; 5.5% in the grower,  $P = 0.001$ ; 13.8% in the finisher,  $P < 0.001$ , and 7.9% overall,  $P < 0.001$ , (Table 2). Enriched pens showed increased gains of 7.19 kg/pen overall compared to the control ( $P < 0.001$ ).

Enriched birds had improved FCRs compared to control birds with a decrease of 0.03 FCR points in the grower ( $P = 0.031$ ), 0.18 points in the finisher ( $P < 0.001$ ), and 0.07 points overall ( $P < 0.001$ , Table 2). When averaged per bird, laser-enriched bird ADG was increased by 2.9% ( $P = 0.048$ ) in the starter period, 13.2% ( $P < 0.001$ ) in the finisher, and 7.9% overall ( $P < 0.001$ , Table 2), and was increased overall on a pen basis when compared to the control (0.17 kg/day;  $P < 0.001$ ).

#### *Breast Blisters and Footpad Dermatitis*

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Under our research conditions, no control or laser-enriched birds displayed breast blisters or footpad dermatitis.

### *Tibia Quality*

DXA scan results of focal bird tibia showed no changes in BMC or BMD, although the enriched tibia were numerically higher in both categories compared to the control (Table 3). BMD and BMC were strongly correlated (Figure 1), as were BMC and tibia weight ( $r=0.720$ ). Interestingly, BMD and tibia weight were only moderately correlated ( $r=0.479$ ). Bone mineral content of the tibia and bird body weight were strongly correlated ( $r=0.677$ ), but BMD and bird weight were again only moderately correlated ( $r=0.456$ ).

Bone breaking strength, determined using the Instron 3367 Universal Test Machine compression method and reported as load (kgf)/area (cm<sup>2</sup>), was numerically higher in enriched focal bird tibia than control, but the difference was not significant (Table 3). Bone breaking strength was moderately negatively correlated with tibia weight ( $r=-0.486$ ) and bird weight ( $r=-0.325$ ). The correlation between all bone measures can be visualized in Figure 2 using a Multidimensional Preference Analysis.

### *Air and Litter Quality*

The birds started on fresh, dry litter that remained friable throughout, and litter scored “pass” in all pens for all 6 wk the birds were on trial. Ammonia strip readings were identical on both the enriched and control rooms of the barn weekly. Before birds arrived, the baseline ammonia levels were 0ppm in both rooms of the barn. The averaged readings were wk 2, 5ppm; wk 3, 5 ppm; wk 4, 10 ppm; wk 5, 10 ppm; and wk 6, 16.67 ppm. Variable readings only occurred on wk 6, with 2 readings of 20 ppm and 1 reading of 10 in each room of the barn.

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Means from the ammonia monitor were compared using a simple frequency and chi square distribution with enrichment treatment as a fixed effect. Treatment effect was not significant ( $P=0.112$ ). In the control room of the barn, 60% of readings were 0 ppm, 20% were 3 ppm, and 20% were 8 ppm. In the enriched room, 60% of readings were 0 ppm, 20% were 3 ppm, and 20% were 4 ppm. Thus, according to the more accurate GasAlert ammonia monitor, the control room peaked at 8 ppm and the enriched room never surpassed 4 ppm at bird height.

### DISCUSSION

At the commercial level, improved FCR is arguably the most valued production trait as it translates to greater weight gain from the same or lesser amount of feed, a cost savings in production, and thus improved sustainability (Stenholm and Waggoner, 1991). Reducing FCR by 0.17 points could equal to more than a 5% decrease in feed costs (Emmerson, 1997), which account for >70% of the costs of broiler production (Banerjee, 1992). The laser enrichment device successfully decreased FCR by 0.18 points in the finisher period of our study vs. control. The increased weight gain of 0.24 kg/bird overall could be translated to between \$0.71 to \$1.39 more saleable product/bird, using current breast meat prices as an example (USDA, 2019). Improved feed conversion may be attributed to decreased maintenance requirements, or more energy partitioned towards growth (Urdaneta-Rincon and Leeson, 2002).

Laser-enriched birds showed significantly increased physical movement during laser periods (Meyer, et al., unpublished data). Increased physical activity has been reported to reduce leg disorder parameters in broilers (Prayitno, et al., 1997; Reiter and Bessei, 2009) and laying hens (Shipov, et al., 2010), but has also been associated with worse feed conversion (Akbar, et al., 1985), or no change in weight gain or FCR (Prayitno, et al., 1997; Reiter and Bessei, 2009; Ruiz-Feria, et al., 2014). Indeed, feed conversion in chickens may have a behavioral component,

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as was suggested in laying hen genetics research by Fairfull and Gowe (1979), but whether this effect may be positive or negative based on exercise has remained unclear. Similar results to ours were seen by Sorensen and others, (2000) where broilers raised at a lower stocking density showed both increased weight gains and improved walking ability. It was hypothesized by Lewis and Hurnik (1990) that there is likely a “locomotory-neutral zone or comfortable upper limit” in broiler movement; meaning that there is an ideal activity level somewhere between the bare minimum distance traveled to access necessary resources and overexertion.

Researchers have successfully forced broilers out of the bare minimum range of movement by increasing distance or introducing barriers between feeders and waterers without compromising performance (Ventura, et al., 2012; Ruiz-Feria, et al., 2014), but other, non-resource-based methods have been less successful. For example, Bizeray and others (2002) tested wheat scattered on the pen floor and colored, moving spotlights but concluded that “forcing animals to exercise more...was more effective for increasing physical activity than was attempting to stimulate foraging activities”. Shields and others (2005) hypothesized that broiler exercise would increase, and leg lameness would decrease, when provided sand bedding, but they were unable to support this as birds rested and displayed more inactive behavior on the sand.

Thus, it appears that the success of the novel laser device in motivating broiler physical activity, while simultaneously improving FCR and ADG, is among the first to accomplish this goal. Further, we may speculate that the 4-min laser periods induced a suitable amount of physical activity without increasing maintenance requirements, hence sacrificing FCR or incurring negative changes to footpad quality, but different lengths of time would need to be tested to validate if this is the most ideal duration. The increased FI observed in laser-enriched



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pens may be attributed to the laser motivating the broilers to move, thus driving them towards the feeders. It has been established by Yngvesson and others (2017) that when resting broilers are disturbed by their conspecifics (a common occurrence), they get up and walk away. We hypothesize that a similar effect was driven by laser-enriched birds, who were physically active at a level considerably greater than control birds at the same time during laser periods, triggering other birds in the pen to move and ultimately move towards the feeders, much like the presence of humans walking a commercial barn motivates broilers to rise and head towards feed.

Yngvesson et al. did not record feeding behavior post-disturbance for comparison, but this hypothesis is supported by our finding that 71% of laser enriched focal birds were at the feeder at least once either during or within 5 min following laser periods (Meyer, et al, unpublished data).

It is thought that most skeletal support is established in broiler birds by day 18, following “intensive bone formation to provide rapid mineralization”. However, bone porosity or density changes more slowly over time to support increasing bird weight (Williams, et al., 2000). Further, bone strength and mass are increased with activity (Rath, et al., 2000), and load-bearing bones need to develop bearing weight, or they will immediately fail to do so when given the opportunity (Lanyon, 1993). Thus, we hypothesized that the increased movement, activity, and growth seen in laser-enriched birds may have been reflected in improved tibia quality measures (Meyer, et al, unpublished data). DXA scanning has been used successfully in broilers to measure BMD and BMC (Swennen, et al., 2004; Shim, et al., 2012; Castro, et al., 2019), and bone breaking strength has been used to detect treatment differences as well (Rowland, et al., 1970; McDevitt, et al., 2006; Shim, et al., 2012). Bone mineral density is believed to be reflective of mineral content (Rath, et al., 2000); our highly correlated ( $r=0.857$ ) BMD and BMC

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values agree with this. Further, BMD and BMC of the tibia obtained from DXA scanning were within normal range, similar to values seen in broiler tibia by Shim and others (2012).

Body weight, tibia weight, BMD, and BMC collectively explain 71.25% of the total experimental differences seen in these variables (Figure 2). Body weight and tibia weight positively influence overall bone measures and tightly cluster, while BMC and BMD cluster with an overall weak negative influence on the collective bone-related outcomes. Although BMD is commonly used as an indicator of bone strength, as bone mineralization is believed to provide compression strength (Rath, et al., 2000), our data indicate a weak negative correlation between BMD and BMC of tibia with bone breaking strength (Figure 2), an outcome that was not expected based on previous work in poultry (Leterrier and Nys, 1992). However, work done in humans (Divittorio, et al., 2006) has indicated that increases in bone density are not consistently correlated with decreased occurrence of fractures, and work in non-human primates (Vahle, et al., 2015) stated that BMD is not always indicative of whether bone will fail in “repetitive loading, as in a stress fracture, or when subjected to high impacts”.

Our bone breaking methods using compression and a constant rate of load would have reflected this repetitive loading, and may contribute to the lack of correlation seen between these bone quality measures. Further work is necessary to validate this unexpected, negative correlation. It is important to note that BMD, BMC, and breaking strength were each numerically higher in laser-enriched birds. These outcomes have previously been seen by Shim and others (2012) in fast growing broilers compared to slow growing, but when analyzed in terms of body weight of the birds, the slow growing birds ultimately had better bone quality. However, in our study the body weight of birds was weakly negatively correlated with bone breaking strength,

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( $r=-0.325$ ), indicating that this measure was not simply reflective of tibia weight or bird weight, but rather a possible true numerical increase of bone quality in the enriched birds.

Commercially, contact dermatitis including breast blisters and footpad dermatitis necessitates downgrading of 15-30% of broiler carcasses/wk (Greene, et al., 1985). Breast conditions range from “mild” brownish colored scabs to “severe” exudate and litter filled ulcers that are aggravated because broilers rest 60% of their body weight on the keel while lying (Nielsen, 2004). Footpad dermatitis is a similar condition, but on the bottom of broiler feet and toes, with symptoms of inflammation and necrotic lesions. This is an obvious animal welfare concern, but also represents a considerable economic loss to the industry, where paws are “the third most important economic part of the chicken behind the breast and wings...accounting for approximately \$280 million a year” (Shepherd and Fairchild, 2010). However, in our clean research environment, neither of these conditions occurred. Other researchers have assessed these on commercial broilers at the slaughterhouse to gather a true representation of the issue (Allain, et al., 2009), and considering that our birds were housed on fresh pine shavings with a relatively lower stocking density and number of birds than a commercial broiler house (0.24 m<sup>2</sup>/bird recommended by the NCC; 0.33 m<sup>2</sup>/bird provided), we were not expecting a high occurrence in our flock. However, we were able to successfully show that the enrichment device did not negatively influence the birds’ health by worsening breast or feet condition compared to the control.

Tied in with breast blisters and footpad dermatitis are air and litter quality. These conditions may be caused by ammonia, originating from urea in the litter compounded by mixing with leaked water from drinkers, causing a chemical burn effect. Haslam and others (2006) showed that percent of birds with footpad concerns was correlated with ammonia concentrations

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in the house and litter moisture, although they did not find this association with breast burns. In the current study, ammonia levels were low, peaking at 8 ppm and 4 ppm on the control and enriched rooms of the barn, respectively, using the GasAlert monitor. This is well within the acceptable range of up to 25 ppm accepted by the NCC audit. Likewise, litter remained dry and friable throughout the 6 wks in both rooms of the house. These outcomes indicate that the larger, more active laser-enriched birds were not generating more moisture in the litter from increased waste or stirring up greater quantities of ammonia over the course of the experiment.

Regarding impact of the laser on bird welfare, data from this study have thus far provided no evidence that animal welfare was negatively impacted by laser enrichment. There was not an increase in lameness, dermatitis, nor mortality; nor did we see a decrease in body weight, tibia quality, or environmental conditions due to laser treatment. In the behavior companion paper (Meyer et al., unpublished data) the Human-Approach Paradigm was utilized as a measure of fearfulness. Results showed that a greater number of laser-enriched birds were closer to the unfamiliar human in the pen during wk 6 of the trial than the control birds, hence an increased fear response was not observed. Future work in this area should include taking physiological stress measures, such as serum or feather corticosterone, to determine if laser enrichment is causing a stress response in broilers. However, it has been shown that broilers are interested in exploring novel objects (Newberry, 1999), and have a propensity to peck at small objects, (Hogan, 1973), so although it is certainly possible that in some cases birds may have been moving away from or unduly stressed by the lasers, thus far our data indicate they were not negatively impacted and were rather interested in the novel nature and small size of the laser dots.

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In summary, these data have provided a strong indication of positive performance effects related to this novel environmental enrichment device. Furthermore, the environmental enrichment device did not result in any unintended negative consequences on the birds' tibia quality, breast and feet condition, or living environment. This unique device improved gains and feed conversion compared to the control, with peak performance results seen in the crucial grower and finisher periods. Following future validation in research as well as in a commercial setting, this enrichment option may be effective for producer implementation. The device does not come into contact with birds, therefore reducing the potential for disease vectors (as in perches/straw bales/tiles) and can be used across multiple flocks. Further work is needed to refine the device as well as performance outcomes, and this work also needs to be extended to commercial conditions.

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Table 1. Starter, grower, and finisher diets provided *ad libitum* to Ross 308 broilers. Each diet was fed for 14 days: starter diet wks 0-2, grower wks 2-4, and finisher wks 4-6. Analyzed values are presented on as as-fed basis.

<b>Ingredients<sup>2</sup></b>	<b>Starter</b>	<b>Grower</b>	<b>Finisher</b>
Corn (%)	55.32	58.69	62.78
Soybean Meal (%)	37.15	33.40	28.59
Soy Oil (%)	2.02	2.98	3.97
Salt (%)	0.40	0.40	0.40
DL-Methionine (%)	0.33	0.30	0.27
Lysine HCl (%)	0.25	0.23	0.21
Threonine (%)	0.15	0.15	0.15
Limestone (%)	1.30	1.01	1.00
Dicalcium Phosphate (%)	2.05	1.81	1.60
Choline Chloride 60 (%)	0.40	0.40	0.40
Vitamin Premix <sup>1</sup> (%)	0.63	0.63	0.63
<b>Calculated Values</b>			
Crude Protein (%)	23.05	21.50	19.50
ME (kcal./kg)	3000	3100	3200
Fat (%)	4.59	5.59	6.64
Digestible Lysine (%)	1.30	1.19	1.06
Digestible Threonine (%)	0.92	0.87	0.80
Digestible Arginine (%)	1.39	1.28	1.14
<b>Analyzed Values (As fed)</b>			
Dry Matter (%)	89.40	89.81	89.23
Crude Fat (%)	6.42	7.63	8.74
Crude Protein (%)	24.17	21.66	19.89

<sup>1</sup>Vitamin and mineral premix provided per kg of diet: Selenium 200 µg; Vitamin A 6,600 IU;

Vitamin D<sub>3</sub> 2,200 IU; Vitamin E 14.3 IU; Menadione 880 µg; Vitamin B<sub>12</sub> 9.4 µg; Biotin 33 µg;

Choline 358 mg; Folic acid 1.1mg; Niacin 33 mg; Pantothenic acid 8.8 mg; Pyridoxine 880 µg;

Riboflavin 4.4 mg; Thiamine 1.1 mg; Iron 226 mg; Magnesium 100 mg; Manganese 220 mg;

Zinc 220 mg; Copper 22 mg; Iodine 675 µg

<sup>2</sup>Calculated according to NRC (1994)

## BROILER ENRICHMENT

Table 2. Ross 308 straight run broiler<sup>1</sup> performance outcomes including feed intake, weight gain, feed conversion ratio (FCR), and ADG by each 2-wk performance period and overall. Starter period indicates wks 0-2, grower wks 2-4, and finisher wks 4-6. Values presented as LSMMeans (pooled SEM) averaged per bird (apart from FCR) with treatment as the main effect

<b>Performance measure</b>	<b>Control<sup>2</sup></b>	<b>Laser<sup>3</sup></b>	<b>Pooled SEM</b>	<b>P-value</b>
<b><i>Feed intake (kg)</i></b>				
Starter	0.48	0.50	0.003	<0.001
Grower	1.56	1.61	0.012	0.004
Finisher	2.62	2.76	0.030	0.004
Overall	4.69	4.88	0.041	0.003
<b><i>Weight gain (kg)</i></b>				
Starter	0.37	0.38	0.004	0.043
Grower	1.04	1.10	0.013	0.001
Finisher	1.37	1.59	0.018	<0.001
Overall	2.80	3.04	0.026	<0.001
<b><i>FCR<sup>4</sup></i></b>				
Starter	1.29	1.31	0.009	0.119
Grower	1.49	1.46	0.008	0.031
Finisher	1.92	1.74	0.024	<0.001
Overall	1.68	1.61	0.010	<0.001
<b><i>ADG<sup>5</sup> (kg)</i></b>				
Starter	0.0265	0.0273	0.001	0.048
Grower	0.0740	0.0756	0.001	0.390
Finisher	0.0992	0.1143	0.001	<0.001
Overall	0.0666	0.0723	0.001	<0.001

<sup>1</sup>Broiler chicks transported from International Poultry Breeders Hatchery (Bancroft, IA) on day of hatch to Iowa State Poultry Research and Teaching Farm: BW 47.38± 0.14g

<sup>2</sup>Control describes pens not exposed to laser enrichment

<sup>3</sup>Birds exposed to laser enrichment device

<sup>4</sup>FCR calculated by dividing kilogram of feed by kilogram of bird weight gain per pen, averaged by treatment for each performance period and overall

## BROILER ENRICHMENT

<sup>5</sup>ADG calculated by dividing bird weight gain averaged per bird by number of days in each performance period and overall

## BROILER ENRICHMENT

Table 3. Focal bird (n=70) right tibia quality measures and weight (LSMeans, pooled SEM) using treatment as a fixed effect

<b>Measure</b>	<b>Control<sup>1</sup></b>	<b>Laser<sup>2</sup></b>	<b>Pooled SEM</b>	<b>P-value</b>
Bone mineral density <sup>3</sup> (g/cm <sup>2</sup> )	0.129	0.138	0.005	0.203
Bone mineral content <sup>3</sup> (g)	0.975	1.107	0.078	0.237
Bone breaking strength <sup>4</sup> (kgf/cm <sup>2</sup> )	9.941	11.143	0.693	0.225
Right tibia weight (g)	14.97	15.75	0.470	0.250

<sup>1</sup>Control describes birds from pens not exposed to laser enrichment

<sup>2</sup>Laser describes birds in pens exposed to laser enrichment

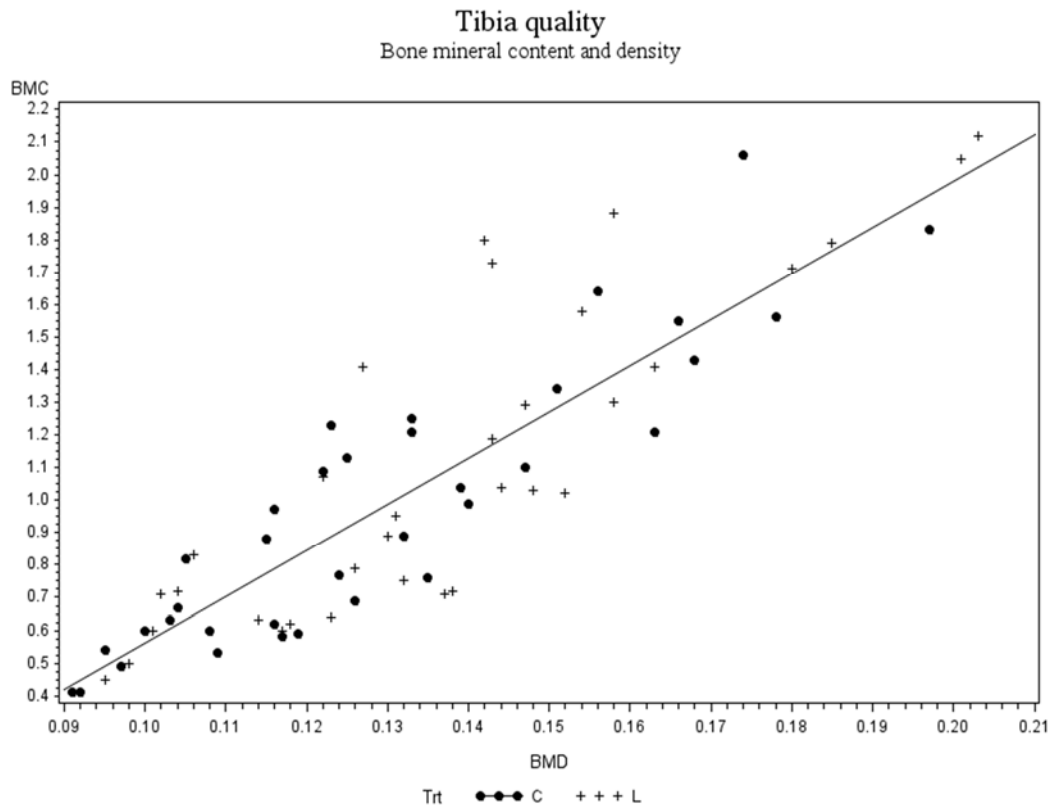
<sup>3</sup>Tibia were scanned for bone mineral density and content using the DXA “rat whole body scan” in groups of 7

<sup>4</sup>Bones were fractured individually using the compression method on an Instron 3367 Universal Test Machine at a rate of 10 mm/min and a 15% rate of load. The machine was stopped at the distinct rapid decline in force as visualized on the monitor, and value is presented as load (kgf) divided by area of tibia (g/cm<sup>2</sup>)



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Figure 1. Regression of focal bird tibia (n=70) bone mineral content (BMC, g) and bone mineral density (BMD, g/cm<sup>2</sup>). Content and density are highly correlated (r=0.857)



<sup>1</sup>Control describes focal birds not exposed to laser enrichment device

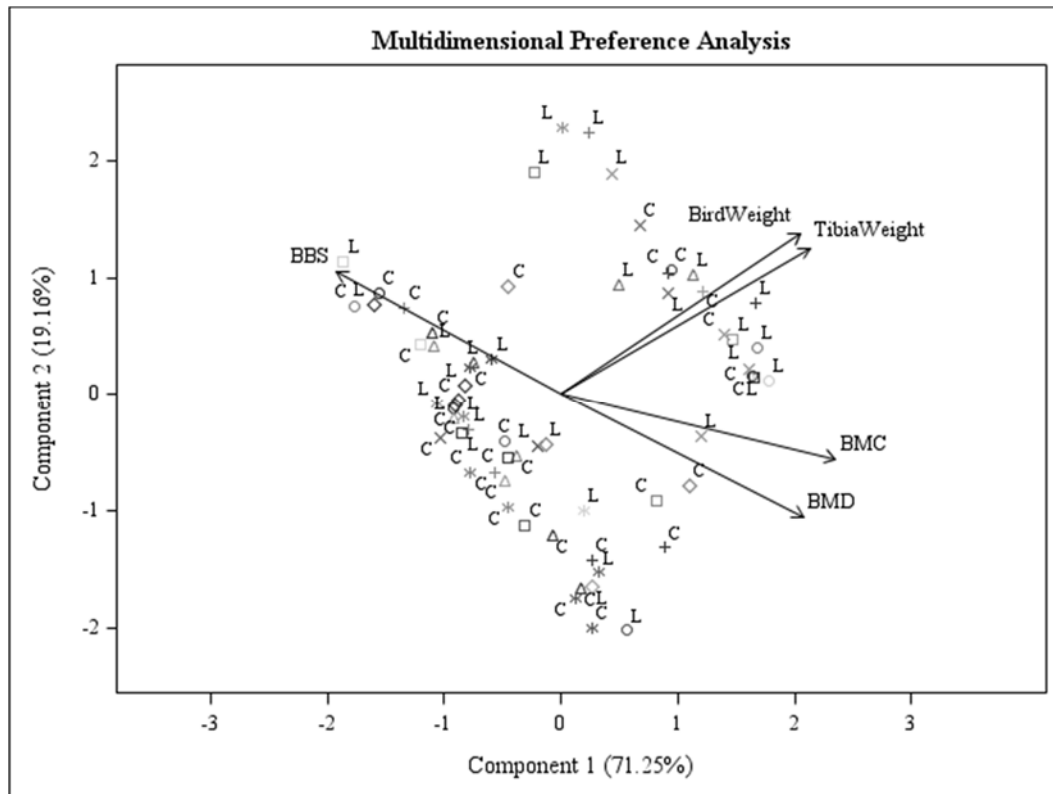
<sup>2</sup>Laser describes focal birds exposed to enrichment device 4 times daily for 4-min laser periods

<sup>2</sup>Bone mineral content and density of the tibia were obtained using the DXA “rat whole body scan” in groups of 7

<sup>4</sup>BMC denotes bone mineral content (g), BMD denotes bone mineral density (g/cm<sup>2</sup>)

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Figure 2. Multidimensional preference analysis of all focal bird tibia (n=70) measures<sup>1-3</sup>: bone breaking strength (BBS), bird weight, tibia weight, bone mineral content (BMC), and bone mineral density (BMD). Abbreviations: C: control bird; and L: laser-enriched bird. Symbols denote individual focal birds



<sup>1</sup>Right tibia were collected from 70 focal birds on d42, weighed, DXA scanned for BMC and BMD, and fractured using an Instron 3367 Test Machine compression method for bone breaking strength

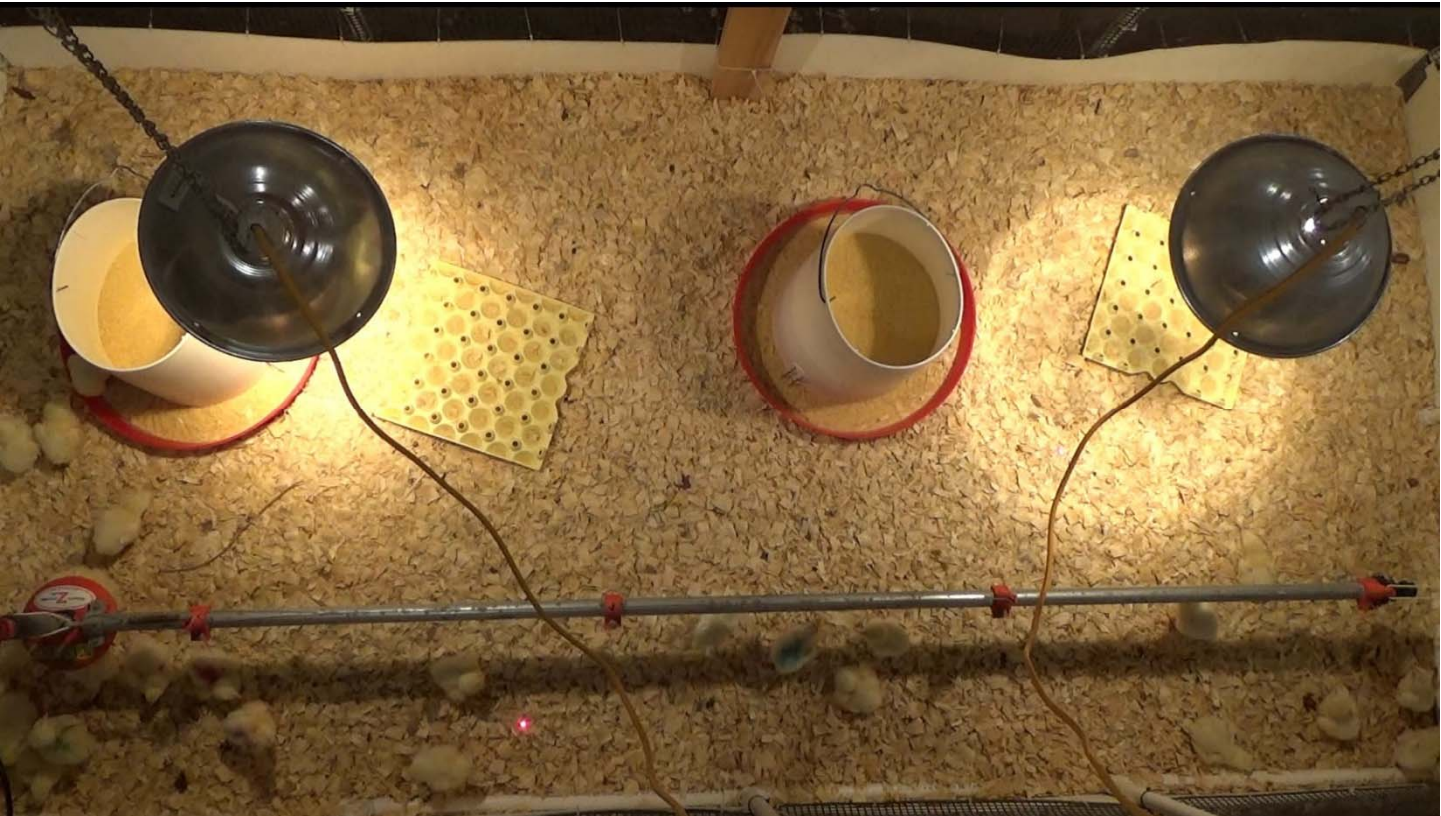
<sup>2</sup>The cosine between 2 variables indicates the correlation between the variables, the length of the arrows reflect the variance of the original variables. This is a 2-dimensional approximation of the dimensions

<sup>3</sup>Original variables have been transformed into new variables (Component 1 and 2) that account for most of the variance

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Supplementary Figure 1A-D. Overhead snapshots of the activated laser in the pens A: d2, B: d16, C: d30, and D: d37

A.



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B.





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C.



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D.

