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Applied research note: comparison of the locomotor activity and circadian rhythm in 3 strains of laying hens as measured by tri-axial accelerometers

J. A. Abecia ,*,[1] C. Palacios,† J. Plaza ,† J. Nieto ,† and F. Canto *

**IUCA, Departamento de Producción Animal y Ciencia de los Alimentos, Universidad de Zaragoza, 50013 Zaragoza, Spain; and †Departamento de Construcción y Agronomía, Área de Producción Animal de la Facultad de Ciencias Agrarias y Ambientales, Universidad de Salamanca, Filiberto Villalobos, 119, 37007, Salamanca, Spain*

Primary Audience: Veterinarians, Researchers, Plant Managers

SUMMARY

The objective of this study was to use tri-axial accelerometers to quantify circadian changes in the locomotor activity of 3 strains of laying hens. Animals were from either the White, Brown or Black strain of a farm that breeds free-range laying hens. Hens were fitted with commercially available sensors that record high resolution raw acceleration data, which were attached to the back of the hen by nylon harnesses and remained in place for 7 d. Separately, animals from each of the strains were allocated to an indoor hen house (density = 0.5 m^2/hen), which had an artificial photoperiod (16L:8D), and an adjacent outdoor pen (4 m^2). Minute-by-minute activity data values (Vector Magnitude, VM) were calculated from the activity counts of each of the 3 axes. Mean (± S.E.) activity (counts/min) of the White strain (was significantly ($P < 0.001$) higher than that of the other 2 strains. Hens were quiet in the dark period of the day, and were significantly ($P < 0.001$) more active in the light period. Locomotor activity differed significantly (P<0.001) among strains in both the dark and the light periods. All hens exhibited a 24-h circadian rhythm in activity, and significantly different MESOR and acrophases ($P < 0.001$). In conclusion, the tri-axial accelerometers tested in this study were useful for measuring locomotor activity in laying hens, and the animals adapted quickly to wearing the devices attached to harnesses. Hens from the 3 strains exhibited the same pattern in locomotor activity throughout the day, although they differed in the intensity of their activity

Key words: laying hen, accelerometer, locomotor activity, circadian rhythm

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DESCRIPTION OF PROBLEM

Poultry farming has come a long way from traditional methods to advanced practices that incorporate the latest technology. Among the many technological innovations, accelerometers have become helpful in monitoring and managing poultry, particularly hens (Casey-Trott and Widowski, 2018; Pullin et al., 2020). The sensors, which measure acceleration, have found their way into poultry farming as versatile instruments for tracking and recording vital data, and have gained substantial attention because of their capacity to contribute to

[1]Corresponding author: alf@unizar.es

various aspects of poultry production; e.g., health monitoring. Pullin et al. (2020) used tri-axial accelerometers to record acceleration events in hens, which provided insights into the effects of pullet rearing on collisions and perch use in enriched colony cage layer housing. Casey-Trott and Widowski (2018) demonstrated that accelerometers have the potential to enhance animal welfare research by quantifying the effects of pain or sickness on activity, measuring daily activity patterns, and quantifying individual differences in general activity. Monitoring of poultry behavior is essential for assessing welfare and optimizing production conditions, and accelerometers provide valuable insights into the daily activities and social interactions of birds (Daigle et al., 2012), or to assess the effects of environmental enrichment on laying hen behavior, which provided insights into how to improve animal welfare in commercial egg production.

Social interactions, facility design, and genetic makeup can cause differences in the circadian pattern of locomotor activity of the animals; therefore, a study of these differences might be helpful in the design of a farm, for a particular breed or strain of hens, and for different animal densities. A temporal evaluation of a fluctuating variable that has a rhythmic variation, such as locomotor activity, can provide a forecast for making decisions because biological rhythms and the health status of an individual animal or a population are correlated. Data from accelerometers can be analyzed by cosinor analysis, which often is used in the analysis of biological time-series that demonstrate predictable rhythms that fit sine wave to a time series. Its advantages are the resistance to outliers, an accommodation of unevenly spaced or unbalanced data, and an increased power for estimation of parameters compared with nonparametric analyses. Changes in feeding time, housing conditions, and social cues can affect the biological rhythms of livestock; therefore, we hypothesized that strains of laying hens might differ in their accelerometer-based locomotor pattern. The objective of this study was to use tri-axial accelerometers to quantify the circadian patterns in locomotor activity of 3 strains of laying hens that were housed under the same conditions, and were subjected to the same feeding and management.

MATERIAL AND METHODS

Animal Ethics Statement

The study was performed at the experimental farm of the University of Zaragoza (Zaragoza, Spain; latitude 41°41′N) following a protocol approved by the Ethics Committee of the University of Zaragoza that was based on the requirements of the European Union for Scientific Procedure Establishments.

Animals

Twelve hens (10 mo of age) from 1 of 3 commercial hybrid egg layer strains (GRAPISA, Pinseque, Zaragoza, Spain) were used in the experiment. Animals were from either the White strain (n = 4; liveweight (LW) (± S.D.): 1.7 ± 0.2 kg), Brown strain (n = 4; LW: 2.2 ± 0.3 kg), or Black strain (n = 4; LW: 2.5 ± 0.2 kg). In the first week of February, hens were fitted with a commercially available sensor that records high resolution raw acceleration data (ActiGraph wGT3X-BT; ActiGraph, FL) (46 mm × 33 mm × 15 mm in size, mass = 19 g), which was attached to the back of the hen by nylon harnesses (Figure 1), and remained in place for 7 d. The sensors were equipped with an integrated light sensor that records lux along with the motion data, which confirmed whether the animal was indoors or outdoors.

Hens were allowed 1 wk to acclimate to wearing the sensor before the start of the experiment. Animals that had the devices attached were housed with 2 other hens of the same strain in an indoor hen house (3 m^2, density 0.5 m^2/hen) that had an artificial photoperiod (16L:8D, light from 08:00 h to 24:00 h), feeders and automatic drinkers, perches, and a nest. The hens had access to an adjacent outdoor pen (4 m^2) that was equipped with dust baths. Access to the outdoors was through 35 cm × 24 cm doors. The walls between the indoor and the outdoor spaces had windows, which provided natural light in the facility. Water was available ad libitum. At the time of the experiment, sunrise and sunset were at 08:16 h and 18:18 h, respectively.

Accelerometer Data Collection

The sensors record accelerations (activity) based on the individual's amplitude (g) and

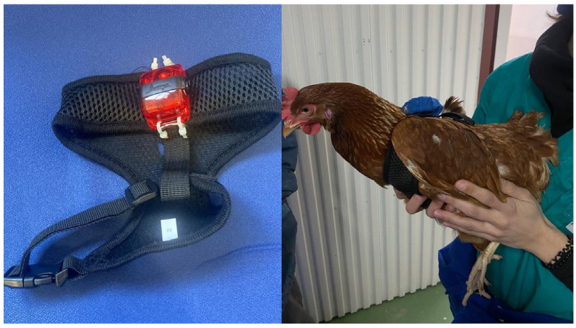

Figure 1. The tri-axial accelerometer, which was attached to the back of the hen by a nylon harness.

frequency (Hz) of movement along 3 axes (x = front-to-back, y = side-to-side, and z = up-down). Sensors were programmed to collect data at a rate of 30 Hz; i.e., 30 samplings per second.

The recorded activity data were downloaded as activity counts per 1 min intervals via BT to ActiLife software (ActiGraph, LLC, Pensacola, FL). Actigraph provides 3 columns of data; i.e., activities in the x-, y-, and z-axes. The activity counts for the 3 axes were combined to create minute-by-minute activity Vector Magnitudes (**VM**), which are the magnitudes of the vectors that are calculated from the combination of the accelerations from the 3 axes on any device. VM was calculated as follows:

$$\text{VM} = \sqrt{(x\_axe)^2 + (y\_axe)^2 + (z\_axe)^2}$$

Statistical Analysis

The statistical significance of the effects of hen strain and time of the day on activity variables (locomotor activity and circadian rhythm parameters) were evaluated by an ANOVA. Differences between groups were evaluated by post-hoc Least Significant Difference (**LSD**) tests. Before those statistical tests, a Kolmogorov−Smirnov test was performed to confirm whether the data were normally distributed.

Circadian rhythms in VM were graphed by fitting the time-series measurements of each hen to the cosine curve of a 24-h activity rhythm, which was obtained by the cosinor method at the Cosinor on-line platform. Midline Estimating Statistic of Rhythm (**MESOR**, the average around which the variable oscillates), amplitude (the difference between the peak and the mean value of a wave), and acrophase (the time of peak activity) were calculated for each variable for each individual. To test for rhythmicity, an F-test compared the (re-parameterized) cosine model and the nonrhythmic model. A $P < 0.05$ indicated that the time series fit a 24-h rhythm. Thereafter, the data were pooled and the mean 24-h cosinor curve, to describe patterns of cyclical activity such as circadian rhythms, for each of the 3 variables was calculated, and the cosinor values of the 3 hen strains were compared statistically by an ANOVA.

RESULTS AND DISCUSSION

Accelerometers indicated that the White strain had significantly ($P < 0.001$) higher mean (± S.E.) activity (counts/min) than did the other 2 strain (Brown = 56.8 ± 0.8, Black = 34.0 ± 0.3; White: 57.8 ± 0.8) (Figure 2A). Hens were mostly inactive in the dark period of the day (1.35 ± 0.07), and were significantly ($P < 0.001$) more active in the light period (63.66 ± 0.42). In both the dark and the light periods, the strains differed significantly ($P < 0.001$) in locomotor activity. Khalil et al. (2004) used behavioral observations and radiotelemetry data to confirm that light and dark periods induce a diurnal rhythm in locomotor activity such that, in the light period, the hens typically engaged in more active behavior such as preening early in the morning, followed by feeding and drinking because the hens

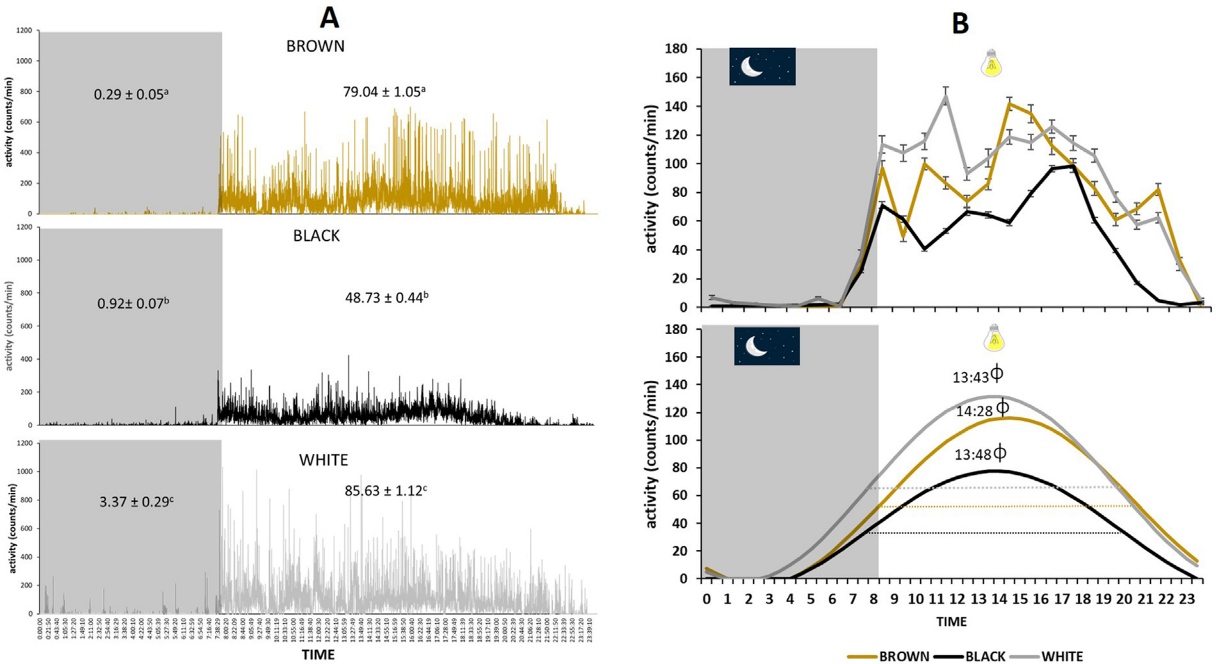

Figure 2. Mean 24-h locomotor activity (counts/min) (A), mean (± S.E.) hourly activity (counts/min/h), and the corresponding cosinor curves of the 24-h activity rhythms (B), as measured by actigraphy, of Brown, Black, and White hen strains (gray areas represent night).

tended to eat most of their feed soon after it had been distributed, which resulted in the highest locomotor activity at that time. Locomotor activity was lowest and remained relatively stable throughout the night because the hens were in a state of muscular relaxation during physiological sleep (Blokhuis, 1984). Kozak et al. (2019) demonstrated that individuals from different hen breeds differed in the degree of excitation and emotional reactivity and, importantly, in their preferences for environment-enriching elements, which indicated that laying hens are not an homogenous group that has the same environmental requirements.

Based on the data provided by the light sensor on the devices, the hens in our study were outdoors intermittently from 10:00 h to 16:00 h, but hens of the Black strain spent significantly ($P < 0.001$) more time outdoors (4.26 ± 1.23 h) than did the hens of the Brown (2.84 ± 0.90 h) and White (2.94 ± 0.74 h) strains. Abouelezz et al. (2014) reported that Rhode Island Red hens spent their time outdoors in foraging, exploring, roaming, standing, primping, and resting, and the range was used most early in the daylight period. Indeed, providing laying hens with access to an outdoor run provides the highest welfare potential in terms of behavioural freedom and some health aspects of all the housing systems evaluated.

In our study, all hens exhibited a 24-h circadian rhythm in activity, and had a mean MESOR of 55.8 ± 0.44, 35.5 ± 0.25 and 65.1 ± 1.54 counts/min ($P < 0.001$), amplitude of 59.89 ± 0.65, 40.68 ± 0.33, 66.63 ± 1.05 counts/min ($P < 0.001$), and acrophases at 14:28 h, 13:48 h, and 13:43 h, for Brown, Black and White strains, respectively (Figure 2B). Light regimes affect the frequency and duration of circadian behaviors in laying hens (Ohtani and Leeson, 2000), and laying behavior has a significant but transitory effect on the heart rate and body temperature of hens, which suggests that oviposition probably is associated with intense locomotor activity (Khalil et al., 2004). Singh et al. (2009) assessed the production performance and egg quality of 4 strains of beak-trimmed layers and found not only significant differences in egg production among strains, but also in the use of nest-boxes. A high proportion of floor eggs among brown egg strains suggest that genotype × environment interactions should be considered when designing a housing system. Furthermore, the genotypes of different hen strains influence the response to heat stress, and selection programs should incorporate behavioral and physiological traits of hen strains to improve their productive performances (Mack et al., 2013). The differences in the activity circadian rhythms of the strains used in our study indicate that that trait should be taken into consideration in designing and scaling laying hen facilities.

The presence of the harness and the accelerometer in some hens could have affected their normal behavior and the social interaction with the rest of the hens, and in particular those non-wearing harnesses. Daigle et al. (2012) concluded that no differences between harnessed and nonharnessed hens with regard to resource use and agonistic interactions at the individual and flock level were detected, and no differences in levels of agonistic interactions between hens wearing and not wearing sensors were reported.

In conclusion, the tri-axial accelerometers tested in our study were useful for measuring locomotor activity in laying hens, and the animals adapted to be wearing the devices attached to harnesses. Hens from the 3 strains exhibited the same pattern in locomotor activity throughout the day, although they differed in the intensity of their activity.

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DISCLOSURES

Authors have no conflicts of interest to disclose.

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