Evaluation of Behavioral Response to The Presence of Novel Objects in Young Pullets Housed in A Pulsed Alternating Wavelength System (PAWS) Environment

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Abstract

Pulsed Alternating Wavelength System (PAWS) Environment is an important tool which could help us know more about how the pullets will react to novel objects in different light environments. The treatments in the study are natural light and five PAWS environments from G2 to G6. The scan sampling data based on the experimental design with repeated measures over time on each tent. The research goal in the study is what will pullets react to PAWS light environments in a tent with novel objects? In the study, I set the natural light group as the control group and used Dunnett's test to identify the significant difference in reactions between each treatment and control group for all the time intervals and draw conclusions on pullets' reactions.

1.Introduction

To reduce the fear of adapting to environmental change is an important task in animal welfare science, which relates to animals' well-being. Novel objects may terrify animals, for example, young pullets. The main topic of the study is to evaluate the behavioral response of young pullets housed in a pulsed alternating wavelength system (PAWS) environment to the presence of novel objects. This topic could help us find a good environment for pullets that can reduce the impact of a novel object and make their life better. The PAWS is a kind of lighting system which could elicit a positive response from animals. The research goal in the study is what will pullets react to PAWS light environments in a tent with novel objects?

The experimental unit is a tent with 16 -18 bovan pullets of around 10 weeks of age. The novel objects are white wiffle ball and black wiffle ball which fasten on two sides of the tent respectively. Six treatments will be in the study: five different PAWS recipes as compare treatments and one natural light environment as a control treatment. Each light environment separately assigned to an environmentally controlled tent with duplicate (control) or triplicate (PAWS). The scan sampling data based on the

experimental design with repeated measures over time on each tent. The researchers kept track of data in each tent every five minutes in a 30-minute interval, therefore each tent would have six records for each variable.

2. Summary Statistics and Summary Figures

Here are some summaries about the data based on box plots, which give straight forward conclusions about the research, but the results still need further study to make a conclusion on statistical results.

2.1 Scan sampling data

Scan sampling data measured the proportion of time spent performing specific behaviors on each tent within six of five-minute intervals. The specific behaviors included movement toward the novel object (black or white), movement away from the novel object (black or white), eating or drinking, no movement/standing still, neutral movement, neither toward the novel object. The response variables will be all observed behaviors mentioned above and predict variables will be PAWS treatments, intervals, and tents.

2.2 Graphs and interpretation

Pullets spent more time on approaching white wiffle ball in G4 or G6 light environment when comparing control light environment during interval 1 (fig 1. (A)). Pullets spent more time on approaching black wiffle ball in G4 or G5 light environment when comparing control light environment all the time (fig 1. (B)). Pullets spent less time on staying away from the novel object in all other light environments when comparing control light environment during interval 1 but spent more time on it in the G3 light environment during interval 2 (fig 1. (C)). Pullets spent less time on eating or drinking in all light environment when comparing control light environment during interval 3 (fig 1. (D)). Pullets spent more time on standing still in G6 light environment when comparing control light environment during interval 3 and 6 (fig 1. (E)). Pullets spent less time on neutral movement in G2 light environment when comparing control light environment during interval 4 and spent less time in G6 light environment during interval 6 (fig 1. (F)).

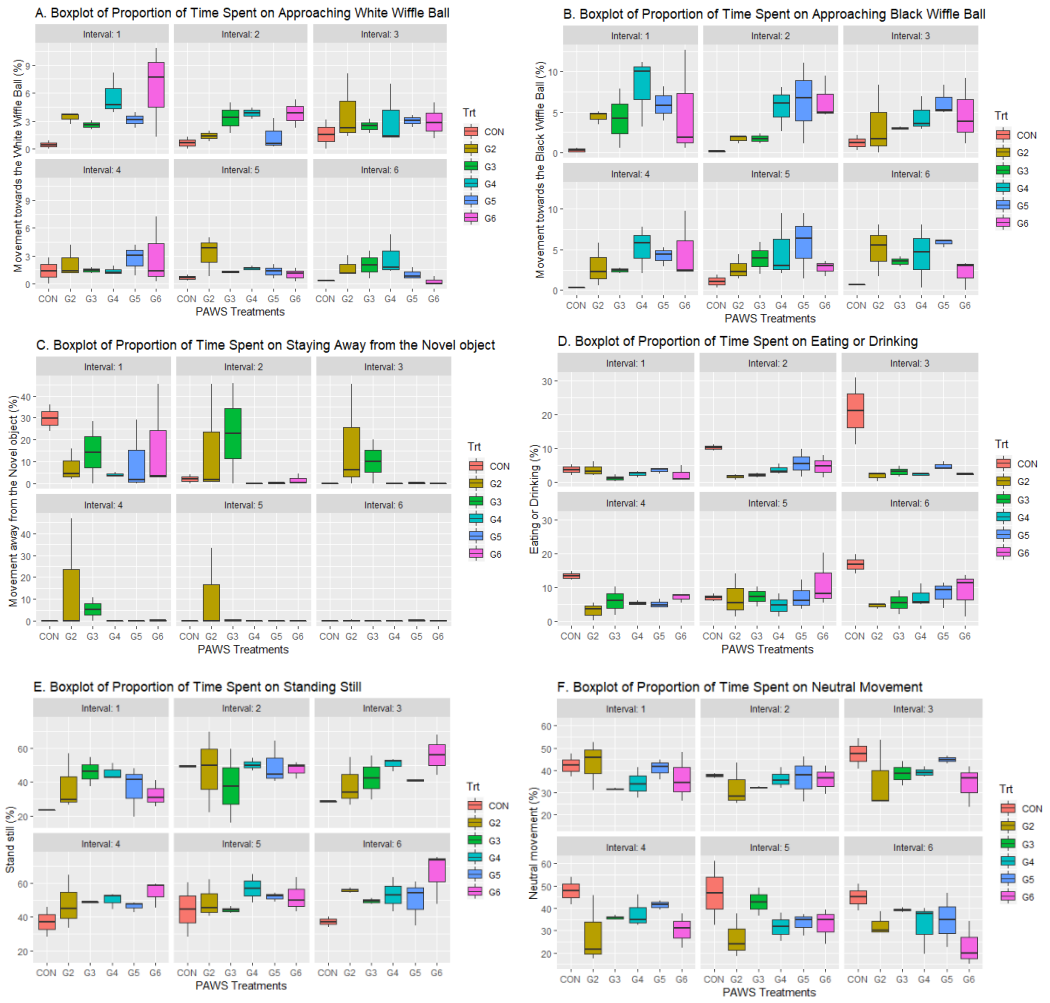

Fig 1. Boxplot of proportion of time spent on approaching white wiffle ball (A), boxplot of proportion of time spent on approaching black wiffle ball (B), boxplot of proportion of time spent on staying away from the novel object (C), boxplot of proportion of time spent on eating or drinking (D), boxplot of proportion of time spent on standing still (E), boxplot of proportion of time spent on neutral movement (F).

3. Methods/Analysis

3.1 Methods

To analysis the research goal, I would like to set the response of pullets to novel object in control light environment as normal reaction (control group), and this could help us know more about if we want to change the environment, which PAWS light environment is better for pullets to live. The Hypothesis is "there is no difference in the mean of the percentage of behaviors on the novel object (black or white) between occurring in PAWS light environments and occurring in control light environment by time."

3.1.1 Model

$$Behavior\ response = int + Trt + Interval + Trt * Interval + (1/Tent) \quad (Equation\ 1)$$

To predict all six response variables in scan sampling data, six linear mixed models were made respectively (Equation 1). Each model contains interaction of treatments and intervals as fixed effects, tents as random effects. The interaction of the fixed effects make sense, for the stress reaction of the pullets will change over time and this change will depend on what treatment environment the pullets live in. It is necessary to make tents as random effects because not every tent of pullets appears same reaction (even though pullets in each tent have a similar physical condition).

3.1.2 Model revise

$$Behavior\ response^{k} = int + Trt + Interval + Trt * Interval + (1/Tent) \quad (Equation\ 2)$$

Where k is root.

In order to meet the model assumptions, the linear mixed model is suitable for the sampling scan data after transformation (Equation 2), for example, we need to use square root for pullet towards white wiffle ball data and eating or drinking data, and four-time root for the data of pullets away from wiffle balls.

3.2 Assumption check

Two assumptions are needed for making linear mixed model: normality of residuals and equal variance of residuals. The results show the revised mixed models fit the data well.

3.2.1 Normality of residuals

From the Shapiro-Wilk test (Table 1), all the p-values are greater than 0.05, fail to reject H_0 that the residuals came from a normal distribution. From the normal Quantile-Quantile Plots (Fig 2), most of the points lie on the red lines, which means the residuals of six mixed model are normally distributed.

Table 1. Shapiro-Wilk test for residuals of the six mixed models with scan sampling data

Variable	*P-value*
Approaching white wiffle ball	0.55134960
Approaching black wiffle ball	0.06373894

Staying away	0.35305486
Eating or drinking	0.49728585
Stand still	0.54423859
Neutral movement	0.17285960

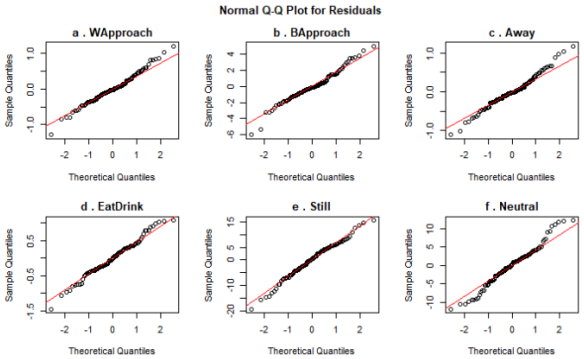

Fig 2. Normal Q-Q plots for residuals of the six mixed models with scan sampling data

3.2.2 Equal variance of residuals

From the residuals versus fitted values plots (Fig 3), the points scatter evenly on the graph, which means the residuals have constant variance.

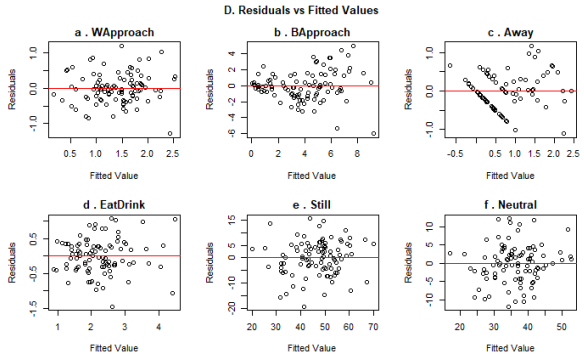

Fig 3. Residuals vs fitted values plots for residuals of the six mixed models with scan sampling data

3.3 Comparison

I used Dunnett's test to compare each response in each PAWS treatment with the control light environment and filtered out significant different comparison results based on p-value < 0.05.

4. Results and Conclusions/Discussion

4.1 Result for behavior analysis

The results give significant different comparisons of event that pullets spend most of their time on when comparing the control light environment (Table 2).

For approaching white wiffle ball, there is a positive significant difference between G4, G6, and control light environment during interval 1, which means pullets spend more time on approaching white wiffle ball in G4 and G6 light environments during interval 1 when comparing with control light environment.

For approaching black wiffle ball, there is a positive significant difference between G4 and control light environment during interval 1, which means pullets spend more time on approaching black wiffle ball in G4 light environment during interval 1 when comparing to in control light environment.

For eating or drinking, there is negative significant difference in eating or drinking between G2 and control light environment during interval 2, which means when comparing to control light environment, pullets spend less time on eating or drinking in G2 environments during interval 2; there is negative significant difference between all other and control light environment during interval 3, which means when comparing to control light environment, pullets spend less time on eating or drinking in all other light environment environments during interval 3; there is negative significant difference in eating or drinking between G2 and control light environment during interval 4, which means when comparing to control light environment, pullets spend less time on eating or drinking in G2 environments during interval 4; there is negative significant difference in eating or drinking between all G2, G3, and control light environment during interval 6, which means when comparing to control light environment, pullets spend less time on eating or drinking in G2, G3 environments during interval 6.

For standing still, there is a positive significant difference between G6 and control light environment during interval 6, which means when comparing to control light environment, pullets spend more time in standing still in G6 light environment during interval 6.

For neutral movement, there is a negative significant difference between G6 and control light environment during interval 6, which means when comparing to control light environment, pullets spend less time in neutral movement in G6 light environment during interval 6.

Table 2. Significant different comparison of event that pullets spend most of their time on between the PAWS light environment and the control light environment in each interval

Proportion of time	*Contrast*	*Interval*	*Estimate*
Approaching white wiffle ball	G4 - CON	1	1.870036
Approaching white wiffle ball	G6 - CON	1	1.932409
Approaching black wiffle ball	G4 - CON	1	7.840000

Eat or drink	G2 - CON	2	-1.943774
Eat or drink	G2 - CON	3	-3.227762
Eat or drink	G3 - CON	3	-2.726969
Eat or drink	G4 - CON	3	-2.933197
Eat or drink	G5 - CON	3	-2.268212
Eat or drink	G6 - CON	3	-2.907490
Eat or drink	G2 - CON	4	-2.066381
Eat or drink	G2 - CON	6	-1.969459
Eat or drink	G3 - CON	6	-1.861769
Standing still	G6 - CON	6	28.465000
Neutral movement	G6 - CON	6	-22.036667

4.2 Concern and improvement

4.2.1 Concern

The concern is that the study involves too many treatment levels, but with relatively small sample size, I highly suggest collect a large data set.

4.2.2 Improvement

A linear mixed model with an AR(1) random effects correlation structure could make the model more precise, though the study doesn’t focus on comparing difference over the time intervals.

References

• Duncan, I. A. 2012. Laying Hen Scenario. Animal Welfare Judging and Assessment Contest. AVMA. Available at: https://awjac.org/prepare/past-scenarios.html#.XNEna3dFzRM

• Forkman, B., A. Boissy, M.-C. Meunier-Salaün, E. Canali, and R. B. Jones. 2007. A critical review of fear tests used on cattle, pigs, sheep, poultry and horses. Phys. & Behav. 92:340-374.

• Gałecki, Andrzej T., and Tomasz Burzykowski. Linear mixed-effects models using R: A step-by-step approach. New York, NY: Springer, 2013. ISBN:1461438993 9781461438991

• Gallup Jr., G. G. and S. D. Suarez. 1980. An ethological analysis of open-field behavior in chickens. Anim. Behav. 28:368-378. Academic Press. San Diego, CA.

• Jones, R. B. 1996. Fear and adaptability in poultry: insights, implications and imperatives. World's Poultry Sci. J. 52:131-174. doi: 10.1079/WPS19960013

• Linear mixed effect models. Available at: https://www.st-andrews.ac.uk/media/capod/students/mathssupport/mixedeffectsknir.pdf

• Ploger, B. J., and K. Yasukawa. 2003. Exploring animal behavior in laboratory and field: An hypothesis testing approach to the development, causation, function, and evolution of animal behavior.

• Webster, A. B. and J. F. Hurnik. 1990. Behavior, production and well-being of the laying hen. 1. Effects of movable roosts, relationship of cage mates, and genetic stock. Poult. Sci. 69:2118-2127.