Ultraviolet Reflectance as a Signal of Individual Quality in Prothonotary Warblers

Emma R. Robinson

Undergraduate Thesis

School of Environment and Natural Resources

The Ohio State University, Columbus, Ohio 43210, USA

Research Advisor: Christopher M. Tonra, Ph.D.

22 April 2023

ABSTRACT

The Prothonotary Warbler (*Protonotaria citrea*) is a Neotropical migratory songbird that breeds in forested wetlands of the eastern United States and winters in the mangroves of Central and South America. Prothonotary Warbler populations have declined by approximately 31 percent in the last 50 years, leading to their listing as a Species of Concern in the U.S.A. Their decline is largely attributed to loss of habitat throughout their entire range; however, the effects of global climate change have also caused disruptions in migration processes, including the timing of arrival to breeding grounds, and first egg date. For many species such as the Prothonotary Warbler, earlier arriving birds are considered "more fit" than other potential mates, as they have the advantage in securing high quality resources. As migration cues are disrupted, understanding different features contributing to mate selection helps us understand what signals of individual fitness may be compensatory for those lost due to anthropogenic factors. This study's purpose is to provide a foundation for studying plumage as a driver of mate-choice and reproductive output in order to advance research towards understanding the behavioral impacts from climate change on this species. I tested the hypothesis that ultraviolet (UV) reflectance is a signal of individual quality and predict that the brightness of the white patches on tail feathers would positively correlate with arrival date in males, first egg date in females, and fledging success in both. Using tail feather samples and data collected from arrival surveys and nest monitoring of Prothonotary Warblers between 2021-2022 at Hoover Reservoir in Galena, Ohio, I conducted spectrophotometer analysis on 117 feathers for readings on UV reflectance, as well as mean brightness across spectra. Linear mixed-effect models were used to analyze the degree of reflectance against the arrival date, first egg date, and fledging data. The results show no evidence that higher levels of UV reflectance indicate an individual who is "more fit" for males or females. I discuss the implications of this on our understanding of this species and conservation concerns.

KEY WORDS: Prothonotary Warbler, UV Reflectance, Brightness, Arrival Date, First Egg Date, Success

TABLE OF CONTENTS

INTRODUCTION

North American bird populations have declined significantly for several decades (Rosenberg et al. 2016). These declines are induced by multiple factors, many rooted in anthropogenic causes such as global climate change, habitat loss and fragmentation, and environmental degradation. Due to these factors, bottomland forests are among the most impacted ecosystems (Londe et al., 2023). Wetland loss and degradation has an observed negative effect on biodiversity, especially of migratory birds relying on wetlands for breeding, wintering, and migratory stopover (Londe et al., 2023; Rushing et al., 2016). In Ohio, wetland habitat has decreased by 90% since the 18^{th} century (Ohio Environmental Protection Agency Division of Surface Water, 2023).

One species highlighted on many state lists is the Prothonotary Warbler (*Protonotaria citrea*). Prothonotary Warblers are a small neotropical migratory warbler, wintering in neotropical mangroves, breeding in forested wetlands of the midwestern and eastern United States. The decline of these habitats coupled with the effects of anthropogenic climate change leave this species vulnerable, emphasizing the importance of researching this species to inform conservation action (Tonra et al., 2019). Due to the high degree of specialization Prothonotary Warblers have on forested wetland habitat, this species has high potential as an indicator of ecosystem health and habitat availability (Hoover, 2009). By understanding the intricate biological mechanisms of this species, we further our knowledge of the world from a bird's perspective, enhancing our ability to steward wetland ecosystems.

Researchers have observed a relationship between the arrival time and breeding success of migratory birds, with earlier arrival time at breeding sites providing a fitness advantage (Reudink et al. 2009; Tonra et al. 2011). For males, arriving at the optimal time cascades into selecting the optimal territory, so that when females arrive the males have already established territories with suitable resources for nesting (Mobrey and Ydenberg, 2001). However, effects from anthropogenic climate change, such as changing weather patterns, interferes with the natural cues that trigger a bird to begin migration (Both and Visser, 2001).

To further understand these effects, this study seeks to underpin some other signals of individual quality birds may use in the process of mate selection. Previous studies examine the brightness of carotenoid-based plumage, identifying a positive relationship between brightness and the relative fitness of an individual (Hill and McGraw 2006). In Prothonotary Warblers, existing research examines the brightness of yellow plumage, a carotenoid-based pigmentation, which is subjectively bright to human eyes (Beck, 2013; Bulluck et al., 2016; Slevin et al., 2019). However, while carotenoid-based plumage tends to be colorful to the human eye, bird vision is tetrachromatic, spanning beyond our vision and into the UV-spectra (Burns and Shultz, 2012). Understanding the sensory perception and associated behaviors within avian social systems and environments with decreased bias towards human sensory perception is critical to making appropriate management decisions. Less known is the details of the vision spectrum of this species and how it is used to assess individual quality. To test this, I analyzed the UV reflectance of Prothonotary Warbler retrices (tail feathers) using photo spectrometry, and performed statistical analyses to understand if there is any correlation to plumage brightness with arrival date, first egg date, and nesting success.

METHODS

Study species

During the breeding season, Prothonotary Warblers breed in the wetland forests/bottomland hardwood forests of the eastern United States. They are believed to be mostly monogamous throughout the breeding season, and both males and females defend territories (Petit 2020). They are primarily insectivorous, including both terrestrial and aquatic insects, and other aquatic prey (Dodson et al., 2016). Prothonotary Warblers are cavity nesters but will utilize nest boxes that are over or near standing water (Petit 2020).

Study Area

The feathers used in this study were collected during arrival surveys at Hoover Nature Preserve in Delaware County, Ohio, United States. This area is protected and managed as an upland forest buffer along the northern border of Hoover Reservoir. The breeding Prothonotary Warbler population here has been part of the long-term monitoring efforts of the Tonra Avian Ecology Lab and Prothonotary Warbler Working Group, and therefore has several years of breeding data, as well as previous studies conducted on the population (Tonra et al., 2019; Ames 2021).

Field Methods

Warblers were marked with aluminum bands issued by the US Geological Survey and a unique combination of color bands for resighting during arrival surveys. This enabled subsequent identification of territories and nests of color-banded individuals. During these captures, additional data such as age, sex, morphometric measurements, and retrice were collected.

After processing, birds were then released. We would check territories and search for active nests adult observation and by checking potential cavities or boxes for signs of nest building or nests with eggs. When an active nest was located, GPS coordinates were recorded for further monitoring. Throughout this process, number of eggs, nestlings, fledglings, and date of first egg laying, hatch date, and fledge date were recorded. To reduce predation caused by our presence, nest checks occurred swiftly by following best practices outlined in Martin and Geupel (1993) and field protocol from Ames (2021).

Spectrometry Methods

UV reflectance and spectrometer work was performed post hoc on feather samples collected from banding efforts. Spectrometer readings were taken using the Ocean Insight Flame Spectrometer and Ocean Insight PX-2 Pulsed Xenon Light Source, which analyzes reflectance across visible light and UV spectra. Using the OceanView Spectroscopy Application Wizard for Reflectance, the spectrometer was calibrated using light and dark standards. Each feather was mounted on non-reflective black paper, and ten readings were taken along the white patch of the feather, with care to exclude any part of the rachis. Once all readings were obtained, they were processed using the R 4.2.3 Perceptual Analysis, Visualization, and Organization of Spectral Colour Data ‘pavo’ package for spectral analysis (Maia et al. 2019; R Core Development Team 2017). Values were organized into a comma-separated values (CSV) text file format where spectral readings corresponded with the appropriate unique band number associated with the bird whose feather was analyzed. From this, the B2 (mean brightness across entire spectral range) and S1U (relative contribution of ultraviolet to the total brightness) for each bird was extracted to another file with the compiled breeding data metrics arrival date, first egg date, total number of Prothonotaries fledged, and total number of Brown-Headed Cowbird (*Molothrus ater*) fledged. I also created a column for success as a binary with 1 representing that an individual successfully fledged any number/species of fledglings throughout the breeding season, and 0 representing individuals all of whose nesting attempts failed.

Statistical Analysis

Utilizing the condensed spectral data and breeding data, t-tests were utilized to first find there was no statistically significant difference in reflectance or brightness between male and female Prothonotaries. To visualize this relationship, boxplots were generated to show the comparison of data between sexes and mean brightness data (Figure 1), and sexes and UV reflectance data (Figure 2). After establishing this relationship, further t-tests were conducted to analyze whether there was any statistical difference in brightness and UV reflectance within a singular sex.

Mixed-effect linear modeling was used to examine the relationship between fixed and random effects. A total of four models were created, utilizing band number as the random effect to account for variation among individual birds.

RESULTS

117 feathers were analyzed for UV reflectance and mean brightness across UV and visible spectra. To further assess these results, linear mixed-effect models using the band number as random effect with first egg date as a measure of fitness for females yielded an R-squared value of < 0.01, meaning less than 1 percent of the variation in first egg date is accounted for by UV reflectance among females (Figure 7). However, mean brightness was slightly higher with an R-squared value of 0.10, or 10 percent of the variance in first egg date was accounted for by mean brightness (Figure 8). For male arrival date and UV reflectance, the R-squared value is 0.03, which signifies UV reflectance does not strongly predict the arrival date of male Prothonotary Warblers (Figure 9). Male brightness was similarly not a predictor of arrival date, with R-squared value < 0.01 (Figure 10).

DISCUSSION

The results of this study provide a framework for UV reflectance analysis for Prothonotary Warblers. While no evidence was found that UV reflectance in the white patch of the retrice predicts individual quality was found, there is still much to explore in the links between plumage color, structure, and reflectance in relation to life history. One factor unexplored in the dataset is the effect age has on feather brightness. Potential next studies may incorporate this information into the compiled dataset in order to draw further conclusions as to how plumage varies from year to year. As birds in their first year of breeding will have grown their feathers in the nest the previous year, they are likely to be 'lower quality' than that of a bird in its second or third breeding season. Additional areas of study include estimating the total area of white within the whole tail and utilizing the single feather as a baseline for color data, or analyzing other structural color within Prothonotary Warblers, such as the blue-gray wings, or yellow reflectance throughout other areas of the body. Building our understanding of plumage and potential biological linkages enables further analysis into how it may be used as a signal for conspecifics or other species, which lays the foundation for understanding how these signals are being disrupted by anthropogenic noise. As increasing anthropogenic pressures cause Prothonotary Warblers and other species towards decline, it is critical that we work to understand them within the context of their senses to inform decisions made for their conservation.

ACKNOWLEDGEMENTS

Special thanks to C. Tonra for his support as my research advisor and mentorship in my chosen career in wildlife ecology, as well as the Tonra Lab of Avian Ecology for allowing me to use their lab space. A massive thank you to B. Tsuru for his incredible guidance through field work and patient assistance in statistical analysis, and I. Ciaburri and A. Rose for their help in spectrometer analysis methods.

LITERATURE CITED

Ames, E. M. (2021). Linking events across the annual cycle, in a Neotropical migratory songbird of conservation concern, the Prothonotary Warbler (Protonotaria citrea).

Beck, M.L. 2013. Nest-box aquisition is related to plumage coloration in male and female Prothonotary Warblers. The Auk, Vol. 130, No. 2 pp. 364-371.

Both, C. and Visser, M.E., 2001. Adjustment to climate change is constrained by arrival date in a long-distance migrant bird. *Nature*, *411*(6835), pp.296-298.

Bulluck, L.P., Foster, M.J., Kay, S., Cox, D.E., Viverette, C., Huber, S. 2017. Feather carotenoid content is correlated with reproductive success and provisioning rate in Prothonotary Warblers. The Auk, Volume 134, Issue 1, Pages 229-239.

Burns, K.J. and Shultz, A.J., 2012. Widespread cryptic dichromatism and ultraviolet reflectance in the largest radiation of Neotropical songbirds: implications of accounting for avian vision in the study of plumage evolution. The Auk, 129(2), pp.211-221

Dodson, J.C., Nicholas, M.J., Bulluck, L.P. 2016. Prothonotary warbler nestling growth and condition in response to variation in aquatic and terrestrial prey availability. Ecology and Evolution, Volume 6, Issue 20, pp. 7462-7474.

Hill, G.E. and McGraw, K.J. eds., 2006. *Bird coloration* (Vol. 1). Harvard University Press.

Hoover, J. (2009). Prothonotary Warblers as indicators of hydrological conditions of bottomland forests. *Proceedings of the Fourth International Partners in Flight Conference*, 128-137. Partners in Flight

Londe, D.W., Davis, C. A., Scott, L.R., Robertson, E.P., Haukos, D.A., Hovick, T.J. 2023. Climate change causes declines and greater extremes in wetland inundation in a region important for wetland birds. Ecological Applications: Ecological Society of America.

Maia R, Gruson H, Endler JA, White TE (2019). “pavo 2: new tools for the spectral and spatial analysis of colour in R.” *Methods in Ecology and Evolution*, 10(7). doi:10.1111/2041-210X.13174.

Martin, T. E., and G. R. Geupel (1993). Nest-Monitoring Plots: Methods for Locating Nests and Monitoring Success (Métodos para localizar nidos y monitorear el éxito de estos). Journal of Field Ornithology 64:507–519.

Morbey, Y.E. and Ydenberg, R.C., 2001. Protandrous arrival timing to breeding areas: a review. *Ecology letters*, *4*(6), pp.663-673.

Ohio Environmental Protection Agency Division of Surface Water. 2023. Ohio Wetlands.

Petit, L. J. (2020). Prothonotary Warbler (Protonotaria citrea), version 1.0. Birds of the World. https://doi.org/10.2173/bow.prowar.01

R Core Team (2017). R: A language and environment for statistical computing. R Foundation for Statistical Computing, Vienna, Austria. http://www.r-project.org/

Reudink, M.W., Marra, P.P., Kyser, T.K., Boag, P.T., Langin, K.M. and Ratcliffe, L.M., 2009. Non-breeding season events influence sexual selection in a long-distance migratory bird. *Proceedings of the Royal Society B: Biological Sciences*, *276*(1662), pp.1619-1626.

Rosenberg, K.V., Kennedy, J.A., Dettmers, R., Ford, R.P., Reynolds, D., Alexander, J.D., Beardmore, C.J., Blancher, P.J., Bogart, R.E., Butcher, G.S., Camfield, A.F., Couturier, A., Demarest, D.W., Easton, W.E., Giocomo, J.J., Keller, R.H., Mini, A.E., Panjabi, D.N., Pashley, T.D., Ruth, H., Stabins, H., Stanton, J., Will, T. 2016. Partners in Flight Landbird Conservation Plan: 2016 Revision for Canada and Continental United States. Partners in Flight Science Committee 199 pp.

Rushing, C. S., Ryder, T. B., Marra, P. P. 2016. Quantifying drivers of population dynamics for a migratory bird throughout the annual cycle. Proceedings of the Royal Society B: Biological Sciences Volume 283, Issue 1823.

Slevin, M.C., Bulluck, L.P., Matthews, A.E., Boves, T.J. 2019. Spatial variation in carotenoid plumage coloration and relationships between female coloration and quality in Prothonotary Warblers (*Protonotaria citrea*). The Auk, Volume 136, Issue 2.

Tonra, C.M., Marra, P.P. and Holberton, R.L., 2011. Migration phenology and winter habitat quality are related to circulating androgen in a long-distance migratory bird. *Journal of avian biology*, *42*(5), pp.397-404.

Tonra, C.M., Hallworth, M.T., Boves, T.J., Reese, J., Bulluck, L.P., Johnson, M., Viverette, C., Percy, K., Ames, E.M., Matthews, A., Slevin, M.C., Wilson, R.R., Johnson, E.I. 2019. Concentration of a widespread breeding population in a few critically important nonbreeding areas: Migratory connectivity in the Prothonotary Warbler. The Condor, Volume 121, Issue 2.

FIGURES AND TABLES

Figure 1 Boxplot modeling t-test data of success of each sex with mean brightness

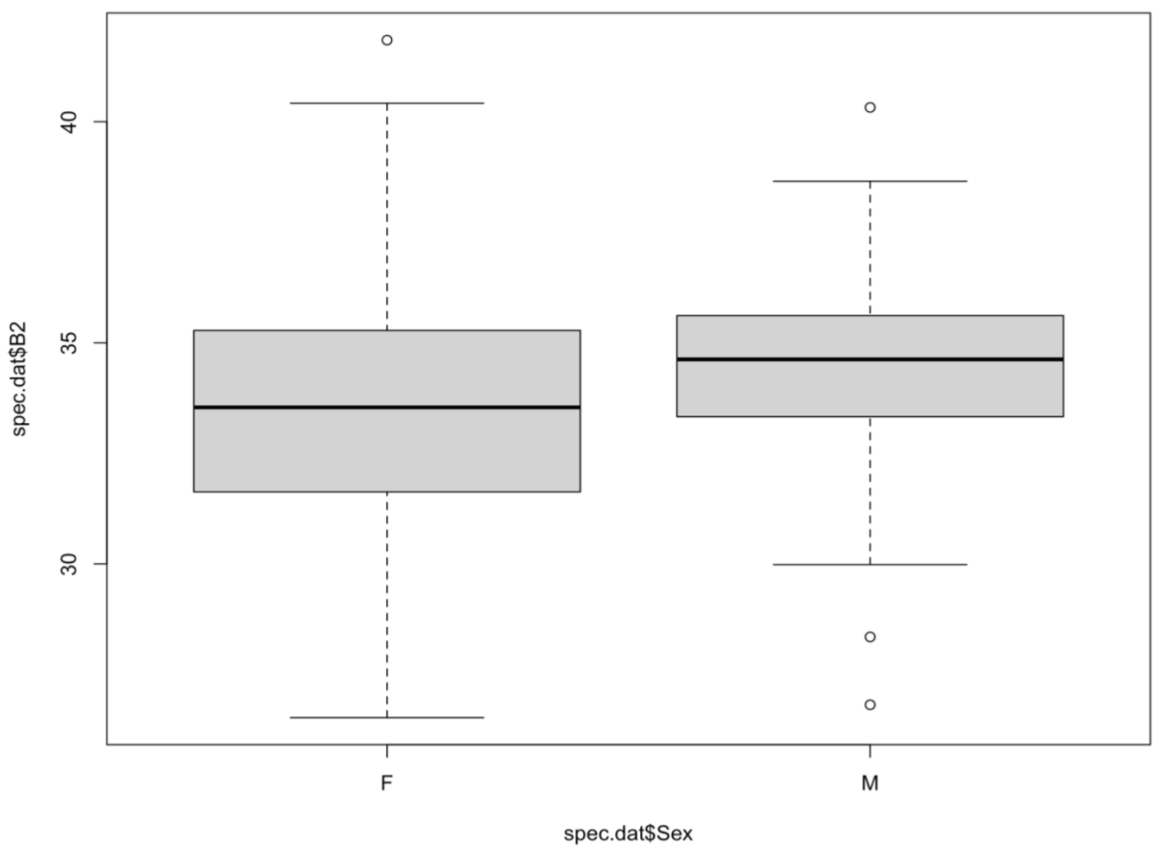

Figure 2: Boxplot modeling t-test data of success of each sex with UV reflectance (S1U).

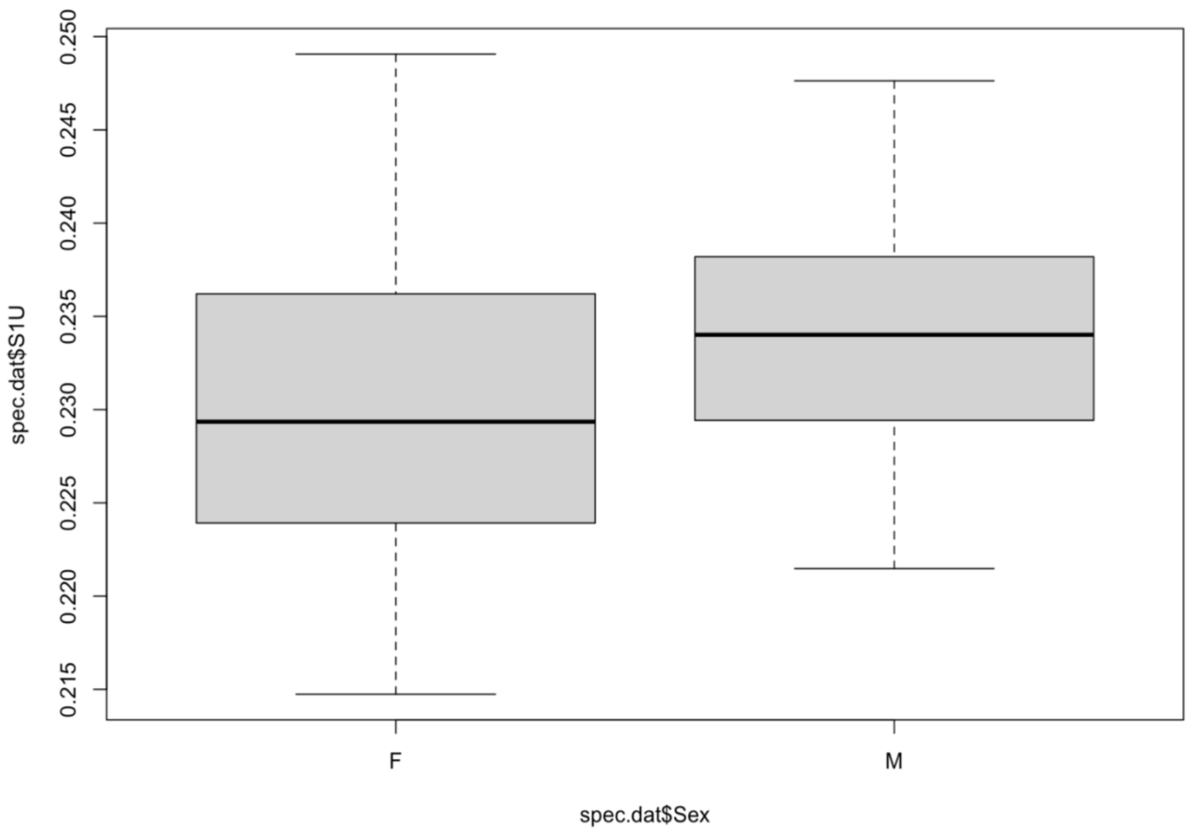

Figure 3: Boxplot modeling t-test of mean brightness data with male nest success

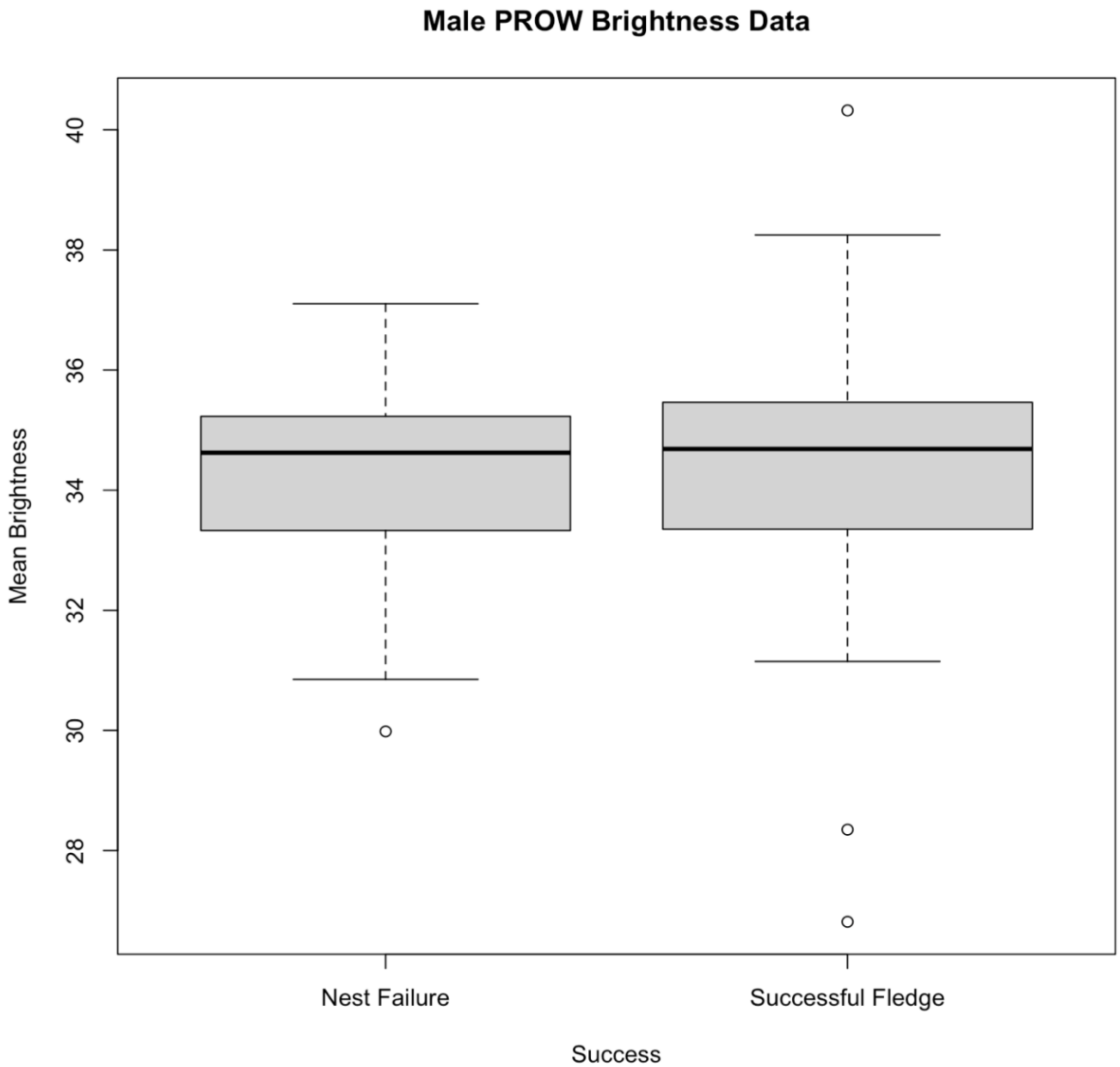

Figure 4: Boxplot modeling t-test of UV reflectance data with male nest success

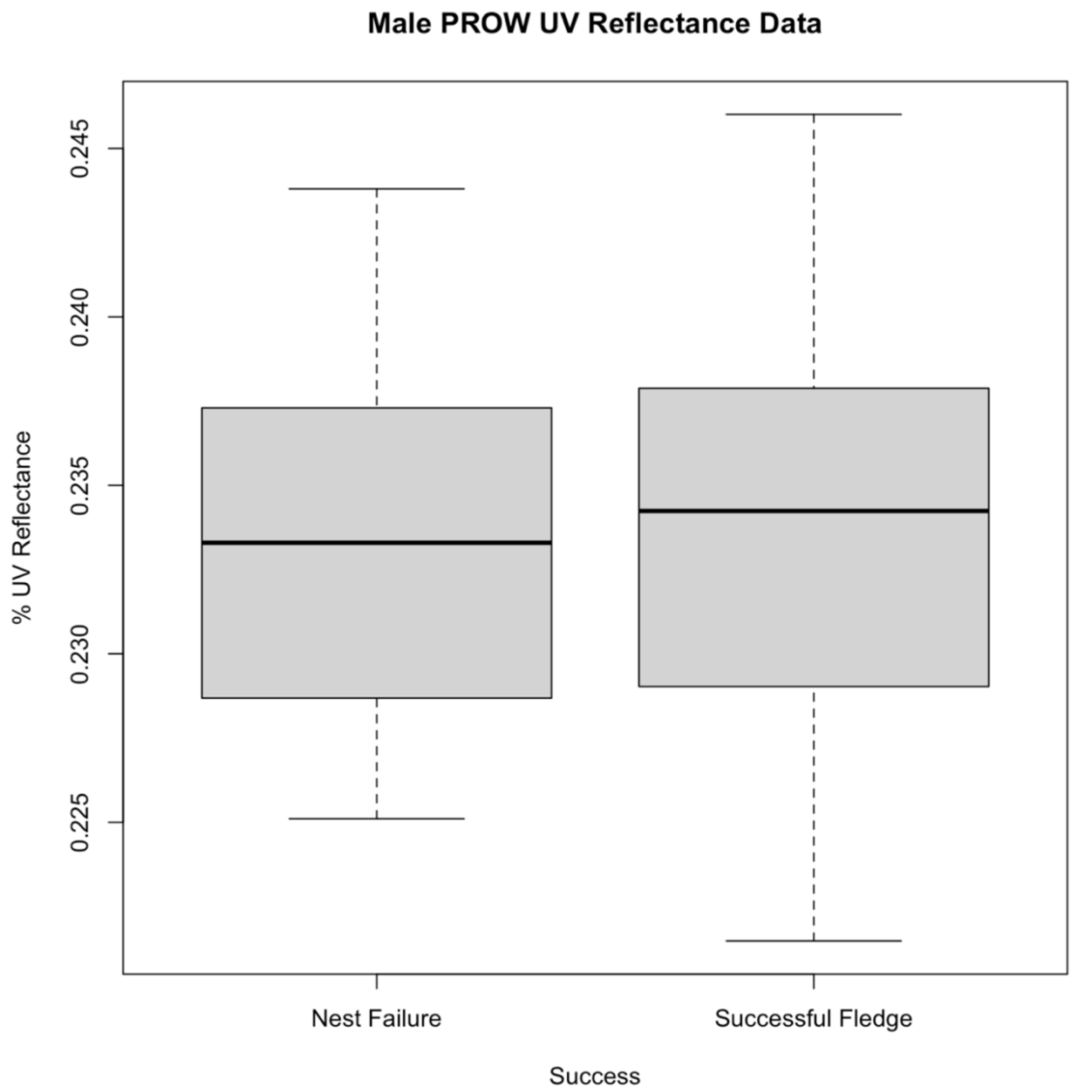

Figure 5: Boxplot modeling t-test of mean brightness data with female nest success.

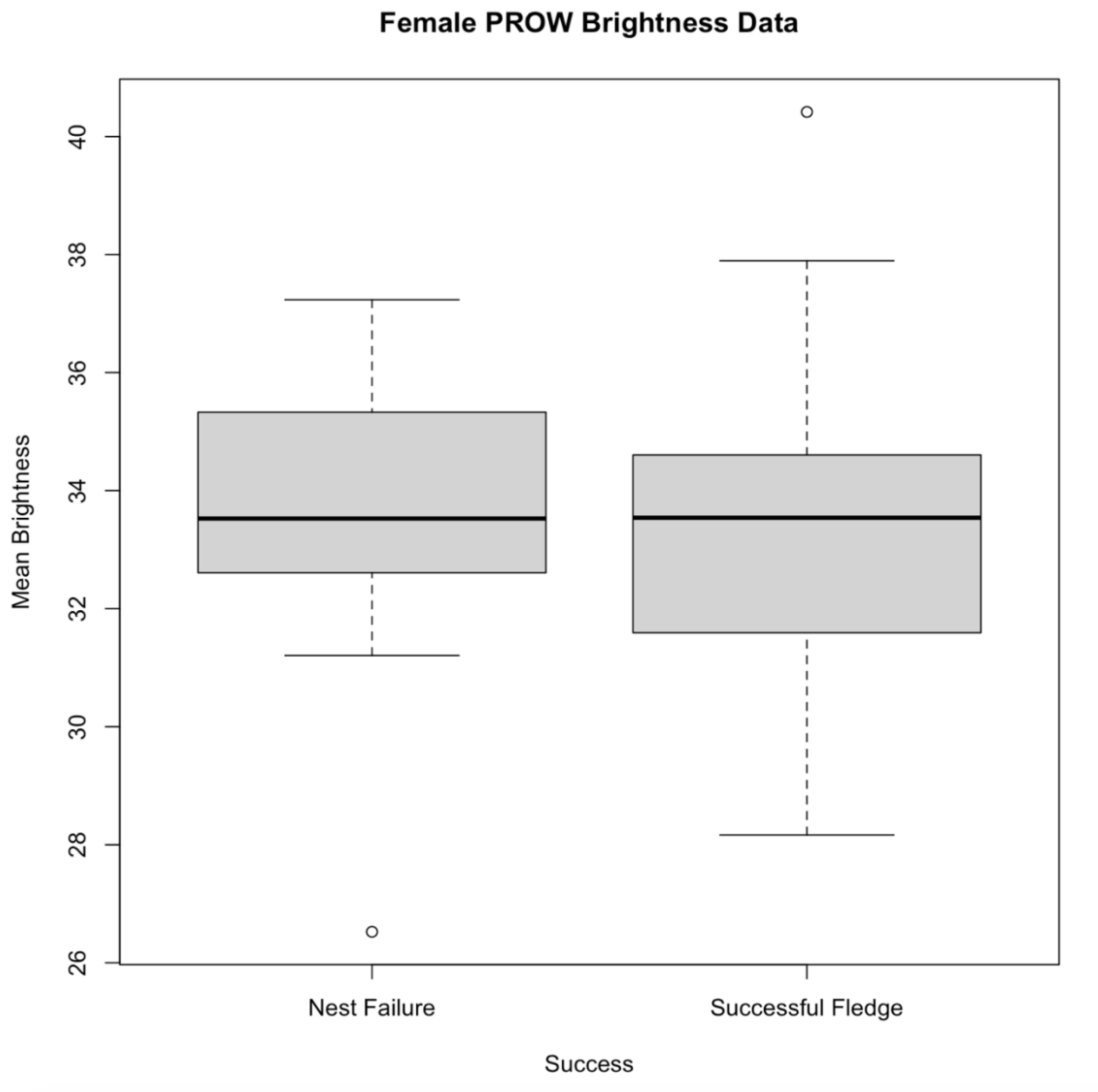

Figure 6: Boxplot modeling t-test of UV reflectance data with female nest success

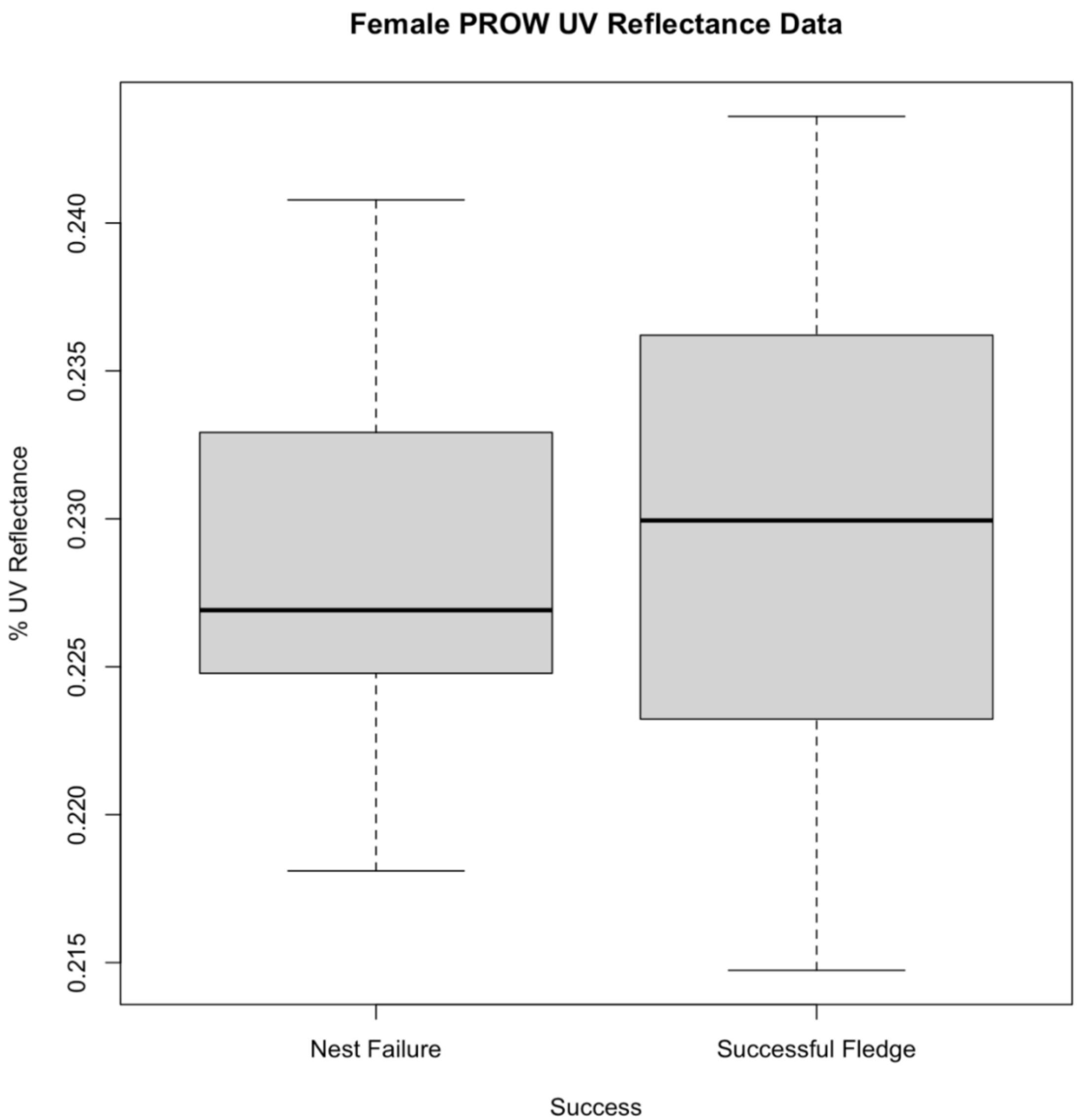

Figure 7: Mixed-effect linear model with line of best fit for the two variables: female arrival date based on the Julian calendar on the y-axis, with UV Reflectance values along the x-axis (B2). No evidence of UV reflectance affecting variation in arrival date.

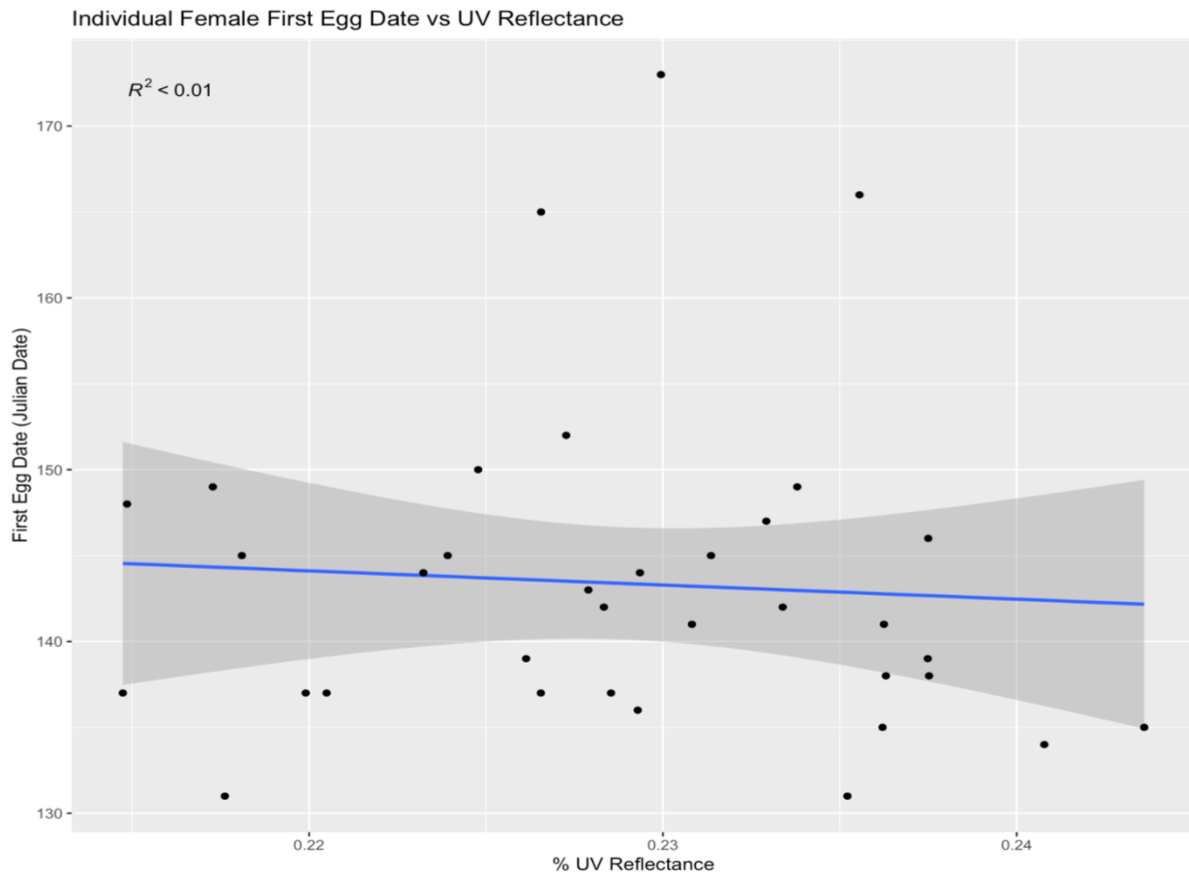

Figure 8: Mixed-effect linear model with line of best fit for the two variables: female arrival date based on the Julian calendar on the y-axis, with mean brightness values along the x-axis (B2). No evidence of brightness affecting variation in arrival date.

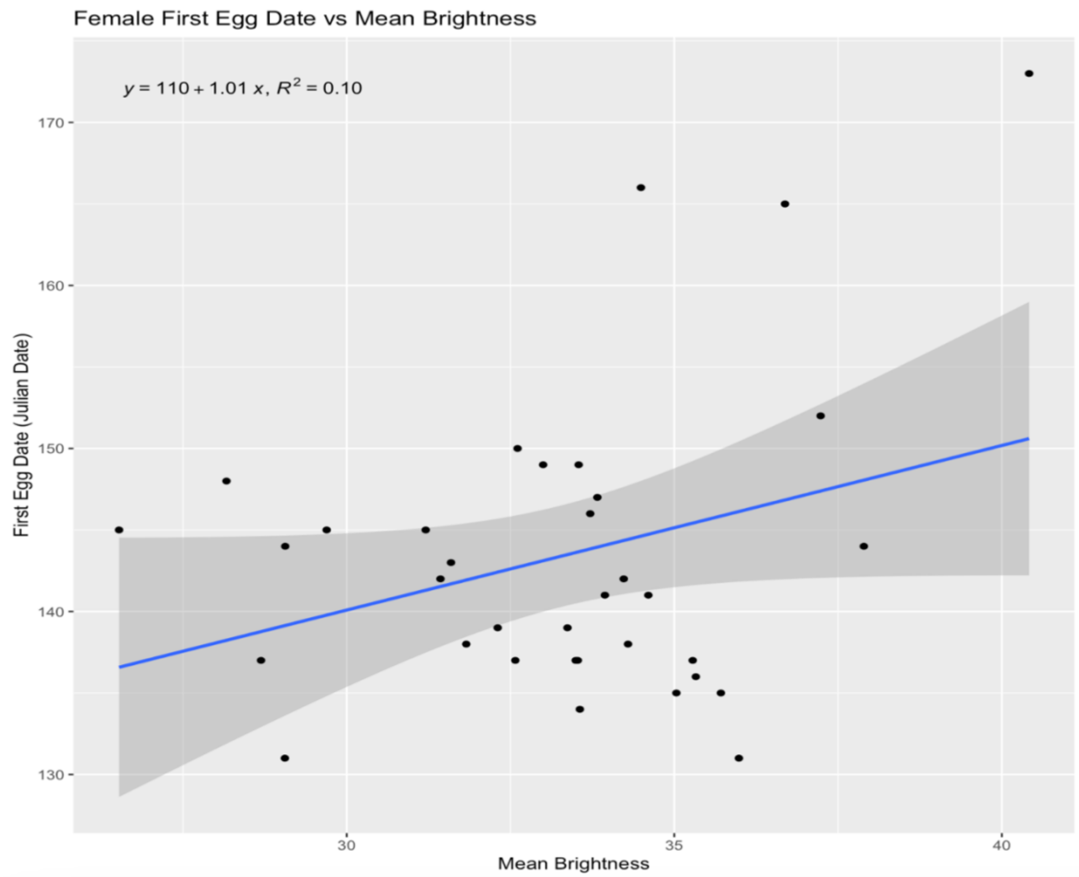

Figure 9: Mixed-effect linear model with line of best fit for the two variables: male arrival date based on the Julian calendar on the y-axis (ARRJD), with UV Reflectance (S1U) values along the x-axis. No evidence of UV Reflectance affecting variation in arrival date.

Male Arrival Date vs UV Reflectance

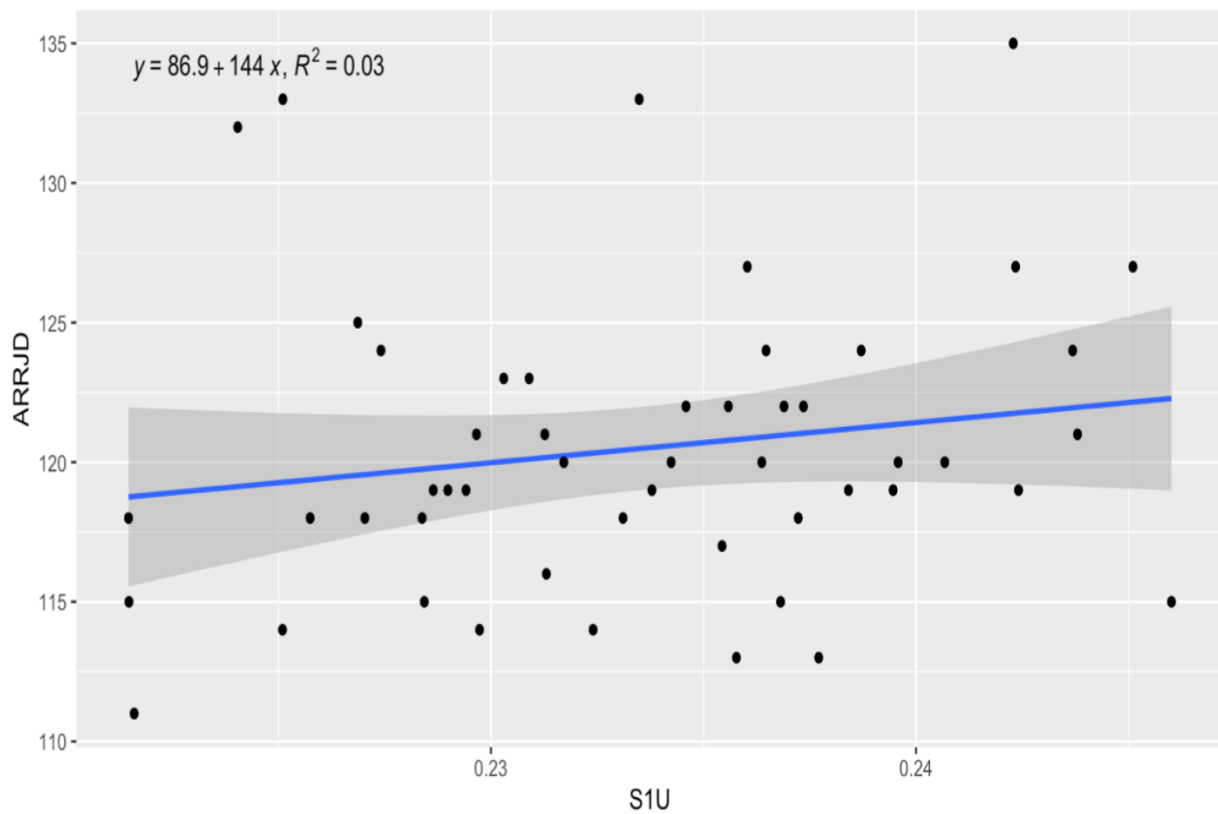

Figure 10: Mixed-effect linear model with line of best fit for the two variables: male arrival date based on the Julian calendar on the y-axis (ARRJD), with mean brightness values along the x-axis (B2). No evidence of brightness affecting variation in arrival date.

Male Arrival Date vs Mean Brightness

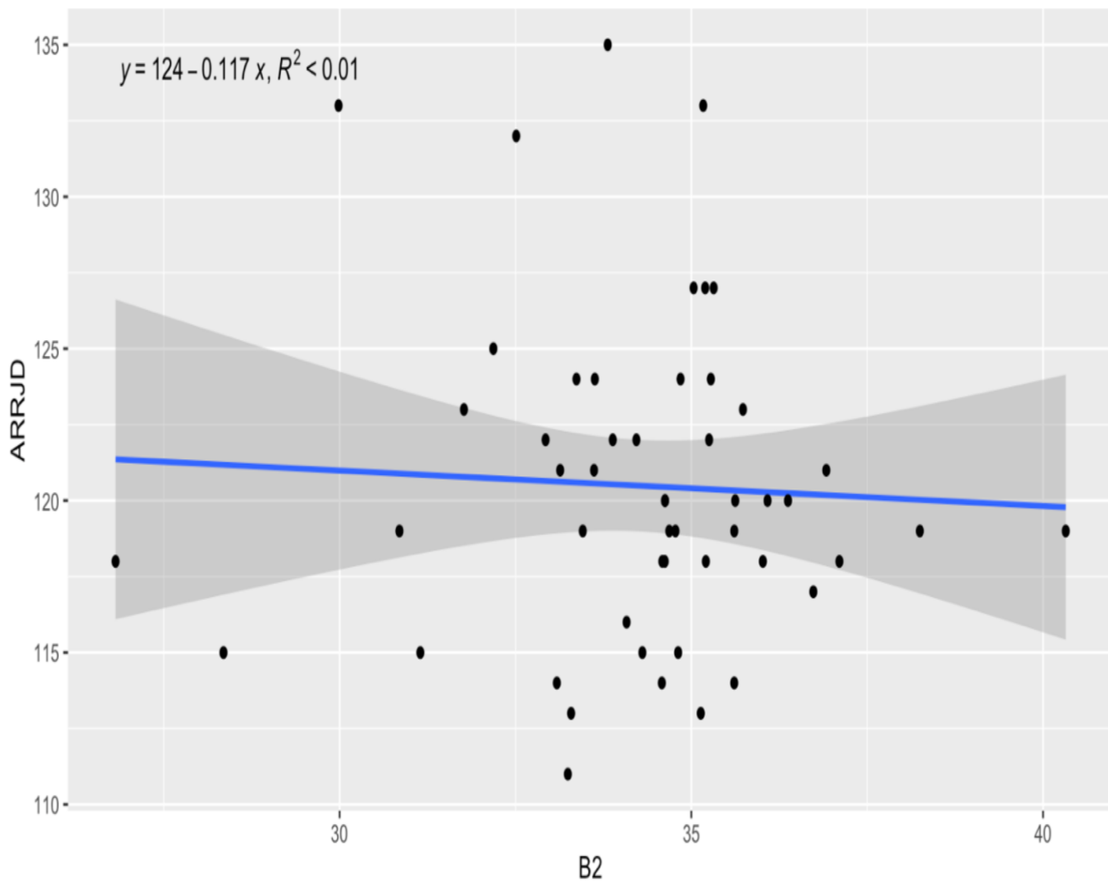

Table 1: t-test results

Dataset	***t-value***	***df***	***p-value***
B2 ~ Success (both sexes)	0.37136	91.739	0.7112
S1U ~ Success (both sexes)	-0.48656	88.724	0.6278
S1U ~ Female First Egg Date	-0.30758	21.474	0.7614
B2 ~ Female Success	0.11586	16.098	0.9092
S1U ~ Male Arrival Date	-0.14369	45.85	0.8864
B2 ~ Male Success	0.028648	48.052	0.9773