

2015

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Wilkins, Nikki and Brown, William P. (2015) "The Accuracy of Eastern Bluebird (*Sialia sialis*) Nestling Age Estimates Produced from Three Different Aging Guides of Digital Images," *North American Bird Bander*. Vol. 40 : Iss. 1 , Article 1.

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The Accuracy of Eastern Bluebird (*Sialia sialis*) Nestling Age Estimates Produced from Three Different Aging Guides of Digital Images

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ABSTRACT

We examined the accuracy of age estimates produced from three different aging guides of Eastern Bluebird (*Sialia sialis*) nestlings. The “complete” guide presented digital images of dorsal and lateral views of the entire nestling from days 1 to 17 after hatching. The “wing” aging guide presented images of lateral views of wings and the “tail” aging guide presented dorsal views of tails for the same span of development. One of the three aging guides and one of 16 sets of 50 randomly selected images from a total of 596 images were assigned randomly to each participant ($n = 39$ participants resulting in 1,950 age estimates). Differences in average daily age estimates among the guides were explored with a linear mixed model with each observer specified as a random variable. Differences in overall accuracy (± 1 day of actual nestling age) among the three guides and differences in the proportion of accurate age estimates for each day of development among the guides were evaluated with corrected Chi-square tests. There were no differences in average daily age estimates among the three guides but overall accuracy differed among the guides ($p < 0.001$). Estimates generated by the complete guide (90.3% accurate) did not differ from those of the wing guide (88.2% accurate). Age estimates from the complete and wing guides differed from those generated with the tail guide (75.1% accurate). Generally, the proportion of accurately determined age estimates decreased for older nestlings.

INTRODUCTION

Aging guides based on photographs or digital images of nestlings have been used to inform studies of growth and development (Hanson and Kossack 1957, Jongsomjit et al. 2007, Brown et al.

2011, Brown et al. 2013). Morphological measurements have also been used to estimate nestling ages (Holcomb and Twiest 1971, Bechard et al. 1985, Brown et al. 2011) but may be less accurate than estimates produced only from images (Brown et al. 2013).

Factors that influence the accuracy of aging guides are related to those that influence differences in rates of nestling development within and among nests, including the sex of the nestling (Holcomb and Twiest 1971), diet (Paxton and Owen 2002), timing of breeding and breeding condition (Zach and Mayoh 1982), and environmental conditions (Bechard et al. 1985, Zach 1982). The accuracy of aging guides may also be influenced by the experience of users, the presence or absence of a scale reference in images, orientation of images in the guide (Brown et al. 2013), and other considerations regarding image size and quality. Whether images of the entire brood (<http://www.sialis.org/runt.htm>), the entire nestling (e.g., Brown et al. 2013), or only parts thereof, such as the wing, are presented (e.g., Bartholmai and Ready 2013) may also influence accuracy among guides.

Despite the increasing availability of aging guides based on nestling images, only one study has provided an accuracy assessment of age estimates produced from such images (Brown et al. 2013). Here, we assessed the accuracy of three different Eastern Bluebird (*Sialia sialis*) aging guides that included digital images from days 1 to 17 of development. These guides included a “complete” guide that presented dorsal and lateral views of the entire nestling for each day of development, a “wing” guide that presented lateral views of the nestling wing, and a “tail” guide that presented dorsal views of only nestling tails for each day of development.

METHODS

Data collection methodology followed that of Brown et al. (2013). Peterson-style nest boxes

(Davis and Rocca 1995) were established near Penn Yan, NY, before the breeding season began in 2011. Thirty-five boxes were available in 2011 and 2012, and 37 were available in 2014; no data were collected in 2013. Nest boxes were checked nearly every day until nestlings hatched in order to determine nestling age correctly. The toenails of each nestling were marked uniquely with colored paint markers. Digital images from a dorsal and lateral perspective were collected during the 2011, 2012, and 2014 breeding seasons from 35 different Eastern Bluebird nestlings from 10 different nests ($n = 596$ images). All images used in this study were from nestlings that survived until at least 10 days of age, and all images included a scale bar. The ages of nestlings in this study ranged from 1 to 17 days of age, with the day of hatching considered as day one.

A “complete” aging guide that included an image from both a dorsal and lateral perspective for each day of development through 17 days of age was created based on images judged to be representative for a particular age. Two other aging guides, one based on images of only nestling wings from a lateral perspective and the other based on images of nestling tails from a dorsal perspective, also were developed from the same source of 596 images. All images in the aging guides contained a scale bar. Each of 39 volunteer participants was randomly assigned one of the three aging guides and one of 16 sets of 50 images randomly selected from the 596 available images. Participants visually compared each image in their picture set to those in their aging guide and provided one age estimate for each of their 50 images ($n = 1,950$ total age estimates). Although age estimates differed from actual nestling age by as much as 7 days ($n = 2$), there was no basis to remove any estimates as outliers.

Statistical methodology - An age estimate was considered accurate if it was within one day of the actual age of the nestling in the image.

Overall differences in average nestling age estimates among the three aging guides were determined with a mixed model (van de Pol and Verhulst 2006). The year in which data were collected and the participants were specified as random effects. Because accuracy of age estimates

might be influenced by nestling age, the effects of guide and actual nestling age on age estimates were also assessed with a mixed model. The alpha level for these multiple post-hoc comparisons was determined using the Šidák-Bonferroni correction procedure (Abdi 2007): $\alpha = 1 - (1 - \alpha_{FW})^{1/c}$, where α_{FW} is the family-wise Type I alpha level (0.05) and c is the number of planned comparisons (17). Based on this procedure, comparisons with $\alpha < 0.003$ were determined to indicate significant differences in age estimates among guides for a specified nestling age.

Differences in the proportions of accurate age estimates among the three aging guides were evaluated with a chi-square test. Differences in the overall proportion of accurate age estimates between each pair-wise comparison of aging guides were also completed with chi-square tests. Comparisons of differences in the proportion of accurate daily age estimates among the three aging guides were completed with chi-square tests corrected with the Šidák-Bonferroni procedure (Abdi 2007); comparisons with $\alpha < 0.003$ were determined to indicate differences in the proportion of accurate daily age estimates among the three guides.

RESULTS

Aging guides produced different average-age estimates overall but estimates did not differ for any specific day of development based on the determined significance level of $\alpha = 0.003$ (Table 1, Fig. 1).

The proportions of accurate age estimates differed among the three aging guides (Fig. 2). Overall, age estimation accuracy was 90.3% for the complete guide, 88.2% for the wing guide, and 75.1% for the tail guide. For all three guides, accuracy tended to decrease as nestling age increased (Fig. 2). Age estimation accuracy between the complete and wing guide did not differ overall ($\chi^2 = 1.8$, $df = 1$, $p = 0.18$; Fig. 2). The complete guide and the wing guide were both more accurate than the tail guide overall ($\chi^2 = 42.9$, $df = 1$, $p < 0.0001$; $\chi^2 = 31.3$, $df = 1$, $p < 0.0001$, respectively). Accuracy among the guides differed for days 3, 4, 7, and 8 of nestling development (Table 1, Fig. 2).

Table 1. Comparisons of the proportions of accurate age estimates among the three aging guides for each day of development. Based on the conservative Šidák-Bonferroni correction for multiple comparisons ($\alpha = 0.003$), the guides differed overall and at ages 3, 4, 7 and 8 days.

Actual Age (days)	Complete Guide	<i>n</i>	Tail Guide	<i>n</i>	Wing Guide	<i>n</i>	χ^2	<i>df</i>	<i>p</i>
1	0.98	42	0.78	49	0.88	43	8.29	2	0.016
2	0.97	33	0.93	43	1.00	39	2.99	2	0.224
3	0.92	38	0.60	58	0.90	39	17.94	2	< 0.0001
4	0.81	54	0.61	82	0.92	77	22.86	2	< 0.0001
5	0.94	34	0.72	43	0.94	34	10.36	2	0.006
6	0.97	30	0.77	39	0.92	38	7.19	2	0.027
7	0.96	28	0.69	35	1.00	22	14.84	2	0.001
8	0.97	35	0.69	39	0.88	34	11.43	2	0.003
9	0.88	33	0.93	57	0.88	51	0.91	2	0.634
10	0.81	32	0.95	39	0.88	33	3.21	2	0.201
11	0.91	32	0.89	54	0.93	42	0.44	2	0.803
12	0.91	35	0.83	35	1.00	30	5.81	2	0.055
13	0.86	29	0.89	27	0.96	23	1.30	2	0.522
14	0.91	23	0.79	34	0.82	33	0.65	2	0.723
15	0.80	10	0.48	21	0.78	18	5.09	2	0.079
16	0.88	16	0.75	24	0.83	24	1.09	2	0.579
17	0.74	46	0.59	71	0.53	70	5.22	2	0.073
TOTAL	0.903	550	0.751	750	0.882	650	57	2	<0.0001

DISCUSSION

Aging guides based on images of nestlings have been used in a variety of studies (Hanson and Kossack 1957, Boal 1994, Jongsomjit et al. 2007, Brown et al. 2011, Brown et al. 2013), but an assessment of age estimation accuracy similar to the one conducted here was available only from a study of nestling House Wrens (*Troglodytes aedon*; Brown et al. 2013). Overall accuracy from the House Wren study, 88.4%, was similar to the overall accuracy of age estimates for nestling Eastern Bluebirds reported here (90.3%) and suggests that guides of digital images provide satisfactory age estimates of nestlings. If only bluebird nestlings aged 14 days of age and younger are considered, overall accuracy increased to 91.2%. Handling nestlings that appear fully feathered – such as those greater than 14 days of age – is not advised due to the potential to induce fledging.

Generally, the accuracy of Eastern Bluebird age estimates decreased as nestlings approached fledgling age. Variability in feather development, particularly wing feather development, became more difficult to discern at that time (see also Bartholmai and Ready 2013; Appendix). Variability in the development of the tail became more prominent as nestlings neared fledging age but, counter to expectations, the accuracy of age estimates generated with the tail aging guide did not improve for older nestlings relative to the other guides. In part, this may be due to the manner in which images were collected from nestlings: the focal point of nestling images usually included a wing or the dorsal area. Sometimes, the tail was not in focus, resulting in challenges to providing age estimates for nestlings in these images based on the tail guide. Developing a tail guide with images collected expressly for that purpose may result in greater accuracy of age estimates based on such a guide.

The accuracy of age estimates may be influenced by factors that influence variability in the development of young, including sex of the young (Holcomb and Twiest 1971), diet (Paxton and Owen 2002), environmental variability (Zach 1982, Bechard et al. 1985), the condition of the parents and timing of breeding (Zach and Mayoh 1982), and the number of young in the nest (Nilsson and Gårdmark 2001). Likewise, there are many factors that influence the application of aging guides. These include the quality of images (Bechard et al. 1985), image orientation, the presence or absence of a scale bar, the experience or motivation of users (Brown et al. 2013), and the images selected to be representative of specified ages. Given the potential for

confounding among these variables, we were nonetheless satisfied at the generally high rates of age estimation accuracy, to within one day of actual nestling age.

Aging guides based on digital images may be more convenient to use than guides based on morphology. Additionally, due to variability in the development of young (e.g., Brown et al. 2011), guides based on images may provide more accurate age estimates than estimates produced from measuring the nestlings (Brown et al. 2013). The development of a general, non-specific, image-based guide that can be calibrated to the developmental trajectories of individual species is a goal for future work.

Fig. 1. Aging guides produced different age estimates overall ($F_{2,1899} = 6.23, p = 0.002$), but did not differ for any specific nestling age based on a level of significance adjusted for multiple comparisons ($\alpha = 0.003$).

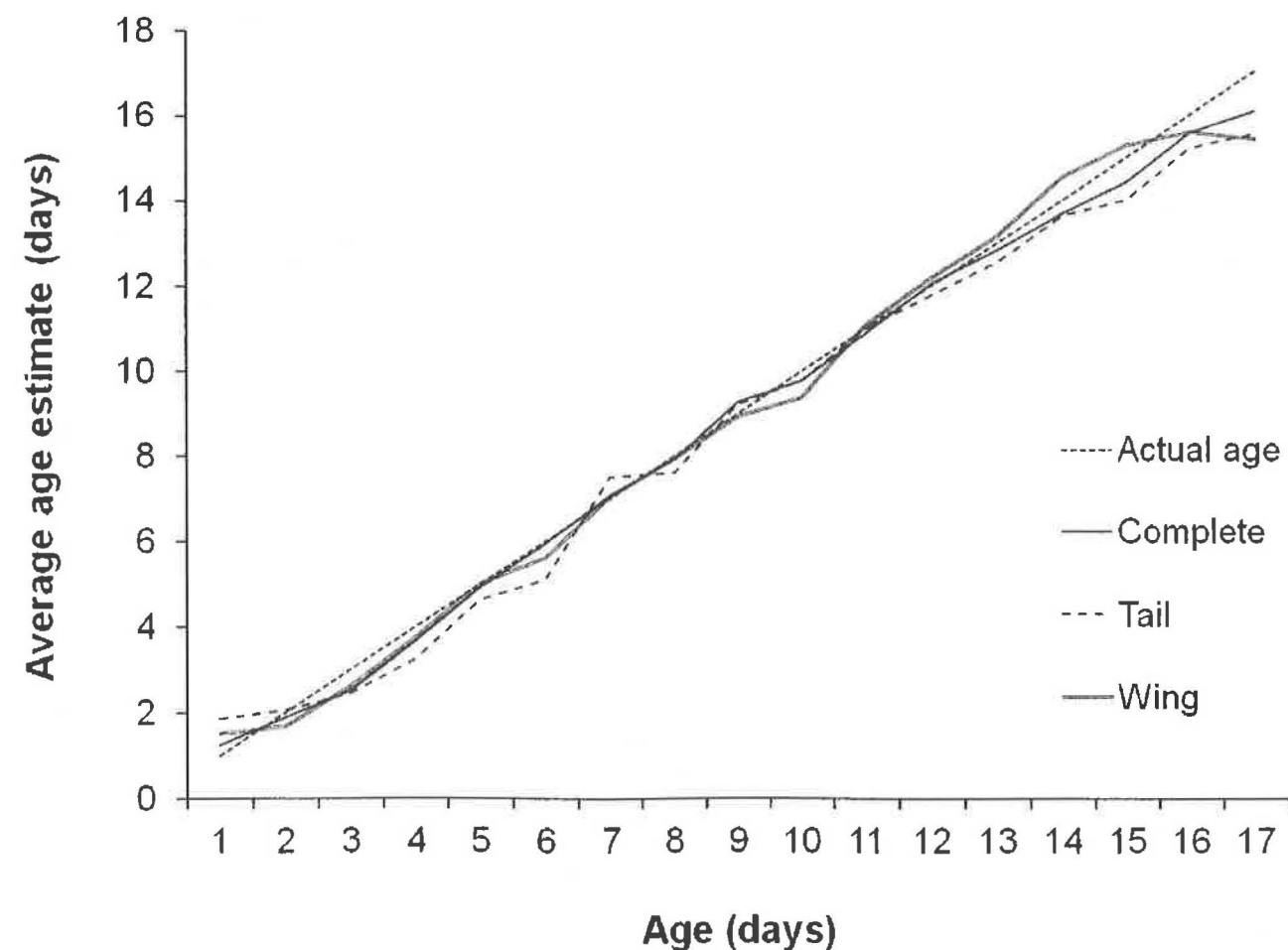
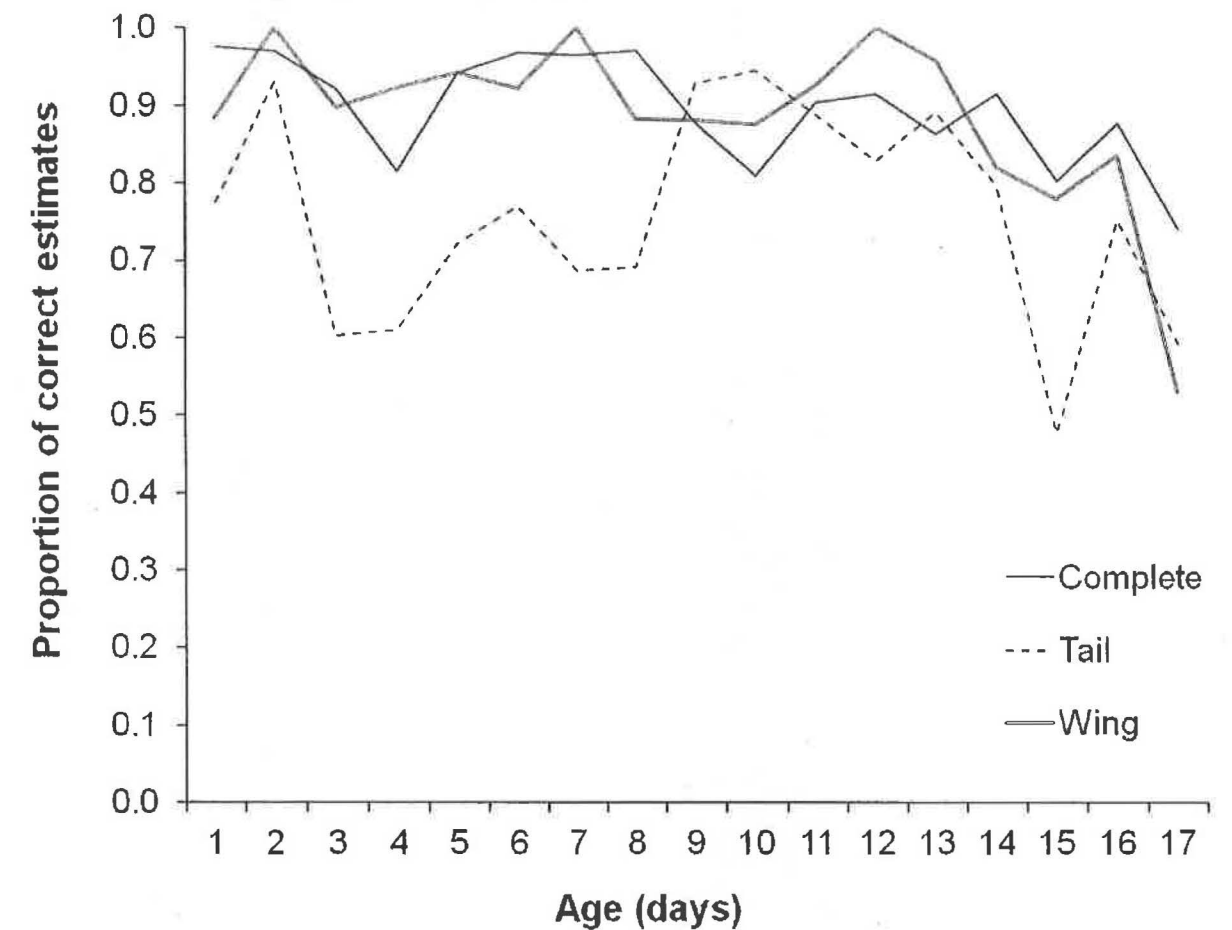


Fig. 2. The proportion of accurate age estimates differed among the three aging guides overall ($\chi^2 = 57, df = 2, p < 0.0001$). Based on an α level adjusted for multiple comparisons ($\alpha = 0.003$), the proportion of accurate age estimated differed among the guides on days 3, 4, 7, and 8 (also see Table 1).





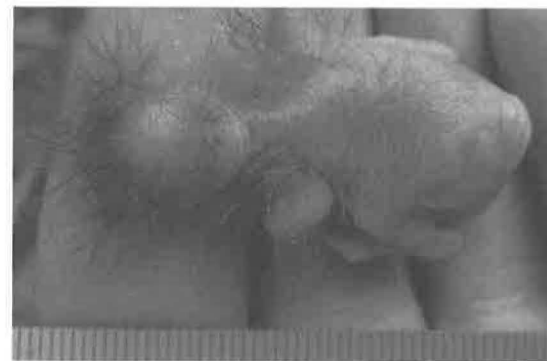
Day 1 Side



Day 1 Top



Day 2 Side



Day 2 Top



Day 3 Side



Day 3 Top



Day 4 Side



Day 4 Top



Day 5 Side



Day 5 Top



Day 6 Side



Day 6 Top



Day 7 Side



Day 7 Top



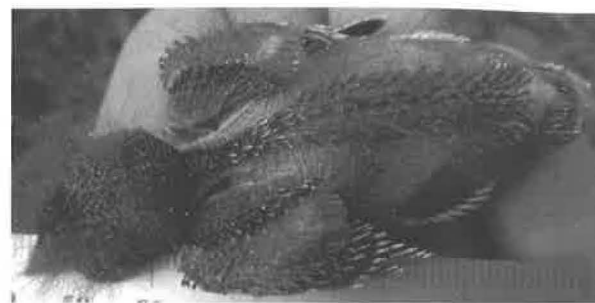
Day 8 Side



Day 8 Top



Day 9 Side



Day 9 Top



Day 10 Side



Day 10 Top



Day 11 Side



Day 11 Top



Day 12 Side



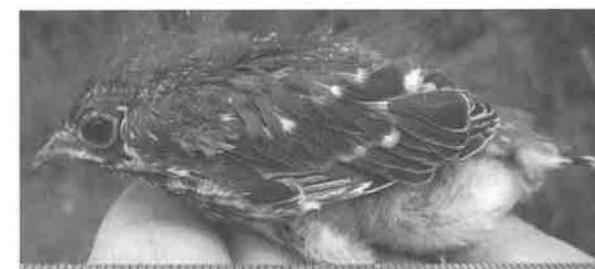
Day 12 Top



Day 13 Side



Day 13 Top



Day 14 Side



Day 14 Top



Day 15 Side



Day 15 Top



Day 16 Side



Day 16 Top



Day 17 Side



Day 17 Top

The photos for this manuscript can be viewed in color on EBBA's Website at <http://www.easternbirdbanding.org>. Go to About EBBA, click on Publications then click on Vol. 40 No.1.

ACKNOWLEDGMENTS

We thank Alex Morgan, Indigo De Jong, Amelia Gonnella, Ana Fajardo, Becky Chase, Carey Bradley, Courtney Lewis, DeNise Wilkins, Dominique Bucci, Kyle Michaud, Gina Ferruzza, Jeff Chapman, Jessica Chapman, Josh Makin, Kayla Curry, Kelsey Morgan, Kelsey Sage, Eric Hoffmaster, Lauren Wilkins, Liana Raguso, Marion Zuefle, Marquita Farrell, Michael Cook, Nick Cantando, Olivia Hudson, Rachel Feagley, Richie Weit, Sara Caruso, Sean Farrell, Stefanie Rienhardt, Tiffany Bane, Tom Lupo, Trace Wilkins, Travis Pelkey, Trevor Irby, and Tyler Skowronski for providing age estimates. Marion Zuefle, Patrick Ready, and Jack Bartholmai reviewed our work.

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Eye Coloration in Common Yellowthroat (*Geothlypis trichas*) to Determine Age

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ABSTRACT

We present a study on age determination of the Common Yellowthroat (*Geothlypis trichas*) through the use of eye coloration, which is compared with other known aging criteria: skull ossification, preformative molt and shape of rectrices. From a sample of 300 birds, it has been determined that the use of eye color is one of the most reliable criteria to determine the age of this species during fall. It is recommended that one gains experience observing captured birds to establish the characteristic.

INTRODUCTION

Although the change of eye color in the iris of several species of birds in relation to their age has been known for some time (Wood 1969, Rosenfield and Bielefeldt 1997, Geupel and Ballard. 2002, Ash 2004, Guallar et al 2009), the use of this criterion has been generally underutilized for passerine birds. Guallar et al. (2009) document the possibility of using eye color as a method of "aging through semi-quantitative variables". In many passeriformes species, such as Brown Thrasher (*Toxostoma rufum*) (Nichols 1953), Dark-eyed Junco (*Junco hyemalis*), White-throated Sparrow (*Zonotrichia albicollis*) (Yunick 1977), and Green-tailed Towhee *Pipilo chlorurus* (Leukering 2000), the eye coloration criterion can be used readily to differentiate young birds from adults, at least during fall when the iris color

difference between young and adult birds is evident. Techniques used to determine eye color go from empirical techniques (Guallar et al. 2009) to the use of references such as Munsell Book of Color (1973) or, more recently, the use of a digital technique (Garrod 2014).

From a total of the 73 passeriforme species treated in their publication, Guallar et al. (2009) reported 25 species in which the use of iris color to determine age of individuals is reliable. Guallar et al. (2009) reported 48 species in which iris color to determine age was not recommended, including the Common Yellowthroat.

Previous observations of specific characteristics of Common Yellowthroats (*Geothlypis trichas*) allowed us to note prominent age-related differences in eye coloration: dark gray in young birds and light brown in adults. Based on this observation, we determined and compared the age of individuals by using different characteristics independently, among these, eye color. The goal of this study is to corroborate that eye color in Common Yellowthroat is not only a valid but also a complementary characteristic to determine age in this particular species.

Study Area - Santa Alejandrina Swamp Monitoring and Banding Station (OAPSA) is located in Minatitlán, Veracruz, Mexico (17°59'27" N, 94°30'38" W) (7 m above sea level), in the coastal plains of the Gulf of Mexico. This site is characterized by wetlands with reed marshes and