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Source: Waterbirds, 40(4) : 334-343

Published By: The Waterbird Society

URL: https://doi.org/10.1675/063.040.0405

Development of a Survey Protocol for Monitoring Reddish Egrets (*Egretta rufescens*) in Florida, USA

W. Andrew Cox[1,*], Amy Schwarzer[1], Richard Kiltie[1], Ann Paul[2], Mark Rachal[2], Gina M. Kent[3], Kenneth D. Meyer[3], Jerome J. Lorenz[4], Peter E. Frezza[4], Heather Rafferty[4] and Suzy Roebling[4]

[1]Fish and Wildlife Research Institute, Florida Fish and Wildlife Conservation Commission, 1105 SW Williston Road, Gainesville, Florida, 32601, USA

[2]Florida Coastal Islands Sanctuaries, Audubon Florida, 410 Ware Boulevard, #702, Tampa, Florida, 33619, USA

[3]Avian Research and Conservation Institute, 411 NE 7th Street, Gainesville, Florida, 32601, USA

[4]Everglades Science Center, Audubon Florida, 115 Indian Mound Trail, Tavernier, Florida, 33070, USA

[*]Corresponding author; E-mail: Andrew.Cox@myfwc.com

Abstract.—The Reddish Egret (*Egretta rufescens*) is North America's rarest heron, and roughly 10% of its population resides in Florida. Its dark plumage, subcanopy nesting, and rarity make it difficult to count with aerial surveys, and assessments of ground- and boat-based methods for estimating the abundance of nesting pairs at breeding sites in Florida are lacking. The efficacy of flight-line surveys (boat-based counts of adults flying to and from colonies) and direct counts (ground- or boat-based counts of nests) were compared using data collected by multiple observers during repeated visits to 16 Reddish Egret colonies in three core breeding areas in the State of Florida, USA. Detection rates on direct counts were 77% for a single observer and 89% for two observers combined. Variance between repeated flight-line surveys was high (61%) for 1-hr surveys but substantially lower for 2-hr (18%) and 3-hr (14%) surveys. Estimated nest counts from flight-line surveys were substantially greater than those produced during direct counts, with mean differences of 85% for 1-hr counts, 134% for 2-hr counts, and 133% for 3-hr counts. Overestimates from flight-line surveys may be related to factors that can be mitigated by avoidance of sites for which use of the method is likely inappropriate (e.g., where breeding is substantially asynchronous or when a site includes an interior foraging lagoon). Survey recommendations are presented for monitoring the Reddish Egret on a large spatial scale. *Received 2 December 2016, accepted 24 February 2017.*

Key words.—breeding synchrony, *Egretta rufescens*, Florida, population estimation, Reddish Egret.

Waterbirds 40(4): 334-343, 2017

The Reddish Egret (*Egretta rufescens*) is North America's rarest heron with an estimated global population size of 5,000-7,000 individuals, of which roughly 10% reside in Florida, USA (Green 2006). The species' narrow coastal distribution makes it especially vulnerable to habitat loss, alteration, and disturbance (Wilson *et al.* 2012). The rarity of the species, its localized breeding distribution, and a potential negative population trend have led to a State listing status of Threatened in Florida.

Nesting Reddish Egrets have been monitored closely in parts of Florida (Hodgson and Paul 2011), but those efforts have not been matched elsewhere in the State, in part because of the cryptic nature and dispersed nesting of the species. Aerial surveys performed from fixed-wing aircraft are often used for colonial-nesting wading birds but are ineffectual for dark-plumaged and subcanopy-nesting species such as the Reddish Egret. One study on the efficacy of aerial surveys detected just 17% of nests and only 15% of known colonies of dark-plumaged wading birds (Frederick *et al.* 1996), and in another study aerial surveys failed to detect any of the Reddish Egrets that had been detected on ground surveys (Rodgers *et al.* 2005). These studies suggest that ground surveys of Reddish Egrets are more accurate and precise than aerial surveys, but ground-based surveys are time consuming and would be expensive to implement statewide. Furthermore, detection rates can vary widely among observers (Green *et al.* 2008), in part because the nests, eggs, and chicks of Reddish Egrets are similar in appearance to those of other heron species. Perhaps most important, colonial species can be extremely sensitive to survey-related disturbances and may abandon their nests or become more vulnerable to predation in response to visits by researchers (Tremblay and Ellison 1979; Frederick and Collopy 1989).

Researchers also use flight-line surveys to estimate sizes of wading bird colonies (Erwin and Odgen 1980; Erwin 1981). These surveys, which use counts of flights by adults to and from colonies to estimate colony sizes, resolve the challenges of plumage color, concealed nests, and research-related disturbances by shifting focus to counting birds that are off the nest. Flight-line surveys present their own challenges because nest visitation rates can vary substantially within and among days, which can increase variance and reduce precision of nest count estimates (Frederick *et al.* 2002). Nevertheless, flight-line surveys can offer reasonable counts for a group of nesting wading bird colonies even when individual colony counts vary widely (Erwin 1981). To date, however, no evaluations of the method have included counts of Reddish Egrets, and earlier efforts focused on larger colonies of wading birds (Erwin 1981; Frederick e*t al.* 2002), whereas Reddish Egrets in Florida, USA, nest in small colonies (usually < 10 pairs) or even singly (Powell *et al.* 1989; Hodgson and Paul 2011).

Variation in habitat, site access, and breeding synchrony makes the Reddish Egret a challenging species to survey and has led to the implementation of a variety of survey methods (Powell *et al.* 1989). The use of multiple survey methods can be problematic if the overall effort does not allow for a quantification of uncertainty, but careful use of multiple data types can produce estimates of population sizes (Martin *et al.* 2015). Our objectives were to: 1) estimate the variation between observers on direct counts and on flight-line surveys; 2) determine the optimal duration of flight-line surveys; 3) estimate the variation between multiple independent flight-line surveys performed at the same site; 4) estimate the correlation between flight-line surveys and direct counts; and 5) describe breeding synchrony of Reddish Egrets at an intensively monitored subpopulation of breeding birds.

METHODS

Study Area

The State of Florida has more than 2,000 km of coastline that includes thousands of natural and artificial islands in its estuarine and marine waters. We surveyed Reddish Egrets at 23 known colony sites in three focal regions (Tampa Bay, Florida Bay, and the lower Florida Keys) from October 2014 through April 2015 (Fig. 1). We conducted direct counts and/or flight-line surveys at 16 of the 23 colony sites and breeding synchrony surveys at all known colony sites in Florida Bay. Colony sites included natural and dredge spoil islands in estuarine/marine (n = 22) and freshwater (n = 1) systems and were typically dominated by red mangrove (*Rhizophora mangle*), black mangrove (*Avicennia germinans*), white mangrove (*Laguncularia racemosa*), or Brazilian pepper (*Schinus terebinthifolius*). Colony sites ranged in area from 0.3 to 69.3 ha ($\bar{x}$ = 14.9 ha ± 4.4 SE), and overall colony sizes varied in size (from < 10 to > 5,000 nests) and species composition (2-13 species).

Data Collection

To allow for comparisons among and within survey types, we typically first performed two independent (i.e., different observers and survey times) direct counts on the same day or on two consecutive days. The direct counts were followed by two flight-line surveys, each jointly performed by two or more observers stationed at opposing sides of a colony site, which generally occurred on two consecutive days. Finally, a third flight-line survey was performed by two observers at a single viewing point to compare results from different observers. We could not achieve this schedule at all 16 colony sites because of logistical challenges associated with weather and concurrent work, so sample sizes varied for each survey type: direct counts and flight-line counts both occurred at 11 colony sites, only direct counts occurred at an additional four colony sites, and at the sixteenth colony site data from flight-line surveys were compared to count data from a breeding synchrony monitoring protocol rather than from our direct count protocol.

Direct counts at 15 colony sites were conducted in close temporal proximity, with 12 counts conducted on the same day or the following day and the remaining three conducted 6, 13, and 14 days apart because of logistical constraints. Counts were often conducted by slowly circling a colony site by boat ~30 m away because colonies on small islands without interior flooded flats or ponds are often amenable to this approach (Paul *et al.* 2004). When likely nest sites (e.g., along an interior creek or pond) were not visible from a boat, counts were conducted by entering the colony on foot. On each direct count, the observer used a Garmin 78sc GPS receiver (95% of locations with ≤ 10 m error) to record nest locations and, when visible, nest contents. We used the GPS coordinates, nest contents and height, adult plumage color, and substrate to distinguish nests counted by one vs. both observers. Ground counts were

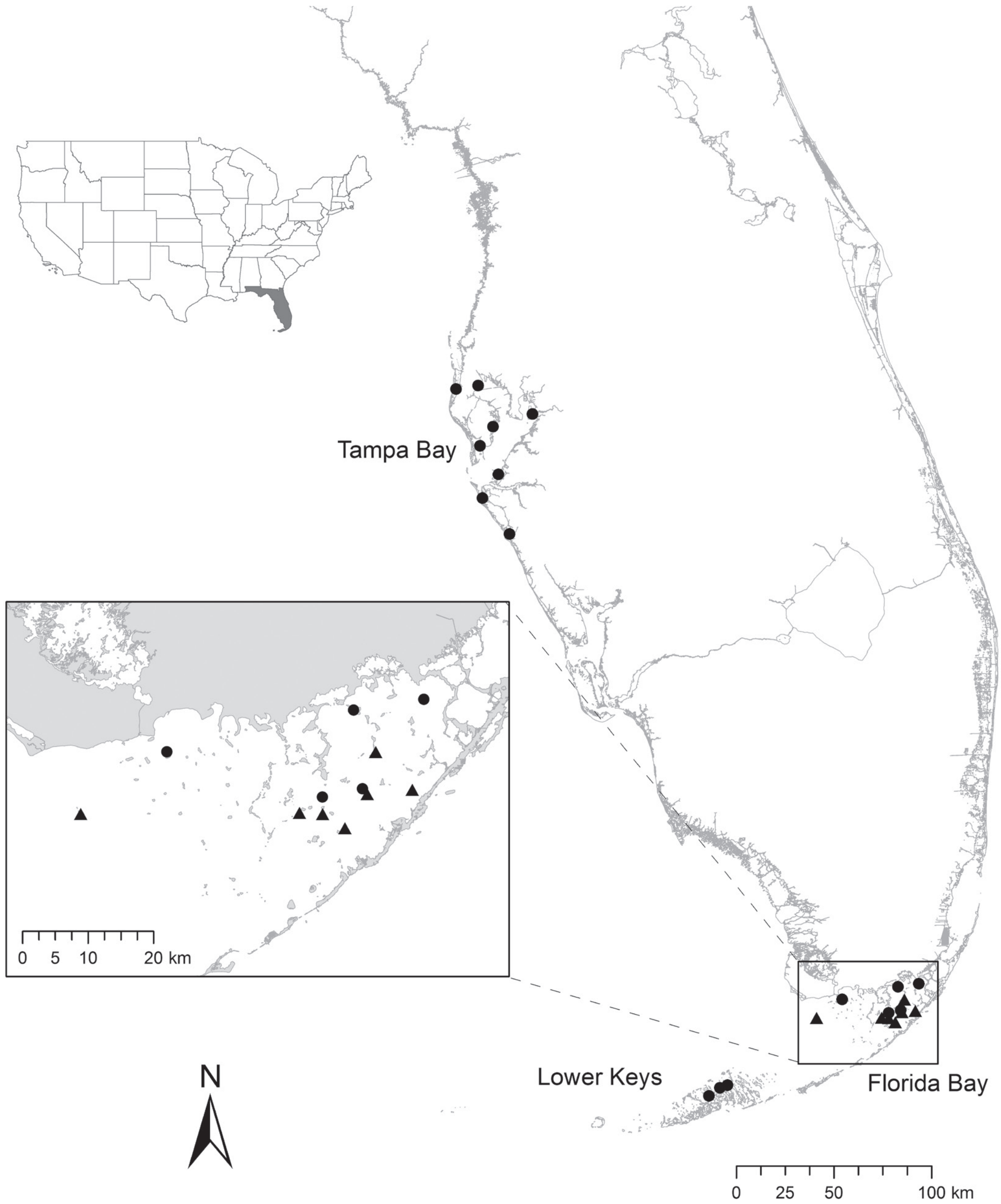

Figure 1. Reddish Egret (n = 23) colony sites in peninsular Florida surveyed during the 2014-2015 breeding season. Filled circles represent colony sites at which we performed direct counts or flight-line surveys. Breeding synchrony surveys occurred at all marked colony sites in Florida Bay (inset), but no direct counts or flight-line surveys were performed at colony sites marked as triangles.

done only when the level of disturbance to nesting Reddish Egrets and other species was deemed acceptable (e.g., when the canopy was sufficiently high) and never when Fish Crows (*Corvus ossifragus*) or other putative nest predators were present. The duration of direct counts ranged from 0.17 to 4.75 hr ($\bar{x}$ = 1.55 hr ± 0.99 SD; n = 30) depending on the size of the colony site and the number of nesting birds of all species.

The first of two flight-line surveys at each of 12 colony sites was performed shortly after the direct counts ($\bar{x}$ = 2.1 days ± 1.35 SD; n = 12), and in all but one case the second was performed within the same week ($\bar{x}$ = 3.75 days ± 6.32 SD; n = 12) following established protocols (Erwin and Ogden 1980; Erwin 1981). Two to four observers stationed themselves at locations that allowed for the entire colony site perimeter to be visible at a distance of approximately 100 m from the shoreline and recorded all incoming and outgoing Reddish Egret flights for 3 hr. Flight-line surveys began 1 hr after sunrise to preclude counting roosting birds, which typically leave near dawn, and to capture the nest exchange between incubating or nest guarding adults, which occurs once each morning during the incubation and early nestling periods (Lowther and Paul 2002). We did not double count flying birds that we confirmed had been recorded by multiple observers, nor did we double count successive flights if continuous surveillance or unique plumage (e.g., pied birds) verified that a single individual had already been observed. Occupancy and abundance survey designs often incorporate three repeat visits per colony site, but we limited our assessment to two because of the cost of visiting coastal colony sites and because simulations have shown that more than two visits result in only minor increases in the accuracy and precision of abundance estimates (Kéry and Royle 2016). Upon completion of the two flight-line surveys, we performed a third survey following a similar protocol except that the two observers were stationed at a single point to determine how variable observer counts were when exposed to the same birds arriving and departing from a colony site.

We conducted breeding synchrony surveys at known Reddish Egret nesting colonies in Florida Bay at least three times per season, with visits every 3 weeks for colony sites with known nesting activity during the past 5 years. We marked each active nest with a uniquely numbered 16 × 8-cm, waterproof, colored (red or blue) card attached with a spring loaded clip to a tree near the nest and revisited active colonies every 7-10 days until all nests failed or chicks became branchlings. Once a colony no longer had active nests, we revisited it once or twice by the end of April to document renesting effort (Lorenz 2014). We sometimes found nests during egg laying but otherwise backdated from hatching or fledging dates whenever possible to estimate nest initiation dates (i.e., the day the first egg was laid) based on a 21-day incubation and 21-day nestling phase (Lowther and Paul 2002; J. J. Lorenz, pers. commun.).

Analysis

We derived nest counts from flight-line surveys for a 3-hr time period because adults typically exchange incubation or nest guarding duties once between midmorning and noon during incubation and the early nestling stage (Paul *et al.* 2004). For each colony site, the nest count was calculated as ½ the total recorded number of incoming and outgoing flights over the 3-hr period (e.g., a male entering a colony to relieve a female that then leaves the colony results in a count of two adults for one nest). To determine whether the duration of surveys could be less than 3 hr and still produce acceptable results, we compared the 3-hr survey counts to those for extrapolated 1-hr surveys (i.e., survey nest counts from a single hour were multiplied by 3) and extrapolated 2-hr surveys (nest counts were multiplied by 1.5). We used a generalized linear mixed model (PROC GLIMMIX; SAS Institute, Inc. 2015), where the derived nest count was an exponential conditional-distributed response variable, survey length was a fixed effect, and colony site was a random effect (Stroup 2012). Finally, we compared nest counts from different days at the same colony site using the method of moments estimator (MME; Springate 2012). Our procedure was a modification of Dahlberg's (1940) formula for the standard deviation of measurement using two counts from the same colony site. We made this comparison for counts from extrapolated 1-hr surveys, extrapolated 2-hr surveys, and 3-hr surveys. Confidence intervals (95%) for measurement error were calculated from Springate (2012), and we applied van Belle's (2008) interpretation that less than 25% CI overlap suggests significance.

We also used the MME estimator of the standard deviation of observer error between the flight-line survey counts from two observers at the same point on a given survey morning. Within a 3-hr survey, we considered the counts of incoming and outgoing birds recorded by each of the two observers as trials that could be used to determine whether either observer consistently overcounted or undercounted Reddish Egrets during flight-line surveys. We assigned a sign (+ or –) to indicate that an observer over- or under-counted egrets relative to second observers they were paired with, and used a binomial sign test (Conover 1971) to assess the results.

We calculated three empirical detection probabilities and one model-based detection probability for the direct counts. The first empirical detection probability was simply the number of nests detected by the first of two observers divided by the total number of nests detected at a colony site. This included nests found only by the second observer and nests later found concurrent with our study but during unrelated surveys of other species. After calculating the same detection probability for the second observer, we combined their counts to estimate an empirical detection probability for a two-observer survey. We estimated a model-based detection probability and sample population size using the double-observer intercepts-only model (Magnusson *et al.* 1978) with the multinomPois function of the unmarked package (Fiske and Chandler 2011) in statistical package R (R Development Core Team 2015). We calculated 95% confidence intervals for the sample population size using a parametric bootstrap (Efron and Tibshirani 1993) with 250 resamples. We made a simple comparison of flight-line and direct counts by calculating the mean differences between them.

RESULTS

We detected 47 nests at 15 colony sites ($\bar{x}$ = 3.13; Range = 1-8) during double-observer direct counts. Sixty-four percent (n = 30) of nests were detected by both observers during the direct counts, and 25% (n = 12) were detected by just one observer. Eleven percent (n = 5) of nests not detected by either observer were found during surveys of other species during the same period. We were unable to exhaustively survey the largest colony site (Alafia Bank; > 14 ha) because once ashore we felt the presence of the observer posed a disturbance-related predation risk to other birds nesting in the colony. Empirical detection rates were 77% for a single observer and 89% for two observers combined. The empirical nest count and detection rate fell within the confidence intervals of the model-based abundance (43.2; 95% CI: 32.0-58.0) and detection probability (0.83; 95% CI: 0.72-0.91) estimates, but this modeled point estimate of average detection probability predicts only one nest missed by both observers ($42*(1-0.83)^2$) rather than the five we recorded.

Estimated nest counts from flight-line surveys at 12 colony sites (Table 1) were similar regardless of survey duration ($F_{2,58} = 0.70$, $P = 0.21$). When we compared two flight-line surveys at the same colony site, the MME estimates (95% CI) of count error for 1-hr, 2-hr, and 3-hr surveys were 4.86 (2.36-7.68), 1.80 (0.87-2.85), and 1.45 (0.70-2.29), respectively; the 1-hr estimate differed significantly from the others by van Belle's (2008) ≤ 25% overlap rule. Relative to the mean estimated counts (i.e., their coefficient of variation), the MME estimates resulted in considerable error (61%) for 1-hr surveys but substantially less error for 2-hr (18%) and 3-hr (14%) surveys. Observer error was considerable when two observers were stationed at the same point, with the MME estimate of 2.27 (1.10-3.59) resulting in an error estimate of 41% relative to the mean count of 5.52. Incoming and outgoing counts did not differ consistently between observers in 58% ($n = 7$) of the 12 two-observer counts, which suggests that individual observers were not consistently undercounting or overcounting Reddish Egrets during flight-line surveys (binomial $P = 0.77$).

Estimated nest counts from flight-line surveys were substantially greater than the actual mean nest count (4.33), with mean differences of 3.69 (85% greater), 5.82 (134% greater), and 5.75 (133% greater) for 1-hr, 2-hr, and 3-hr counts, respectively. The discrepancy between flight-line surveys and direct counts was especially large at two colony sites in Florida Bay that had interior lagoons in which Reddish Egrets foraged (Stake and Jimmy Keys; Table 1). Excluding these colony sites as well as Alafia Bank, the mean differences for 1-hr, 2-hr, and 3-hr counts were 0.72 (14%), 2.82 (56%), and 2.89 (58%) greater, respectively.

We monitored a total of 68 nests at five focal colonies and seven additional colonies in Florida Bay (Fig. 1) to describe breeding synchrony in the area. Breeding synchrony appeared to be somewhat low, with 24% ($n = 16$) of nests initiated in the 3 months prior to the peak of nesting initiation in January (Fig. 2).

DISCUSSION

The dark plumage and subcanopy-nesting habits of Reddish Egrets necessitate direct counts or flight-line surveys for estimating the number of birds breeding in a given area (Frederick *et al.* 1996; Rodgers *et al.* 2005). Our results suggest that direct counts are preferable to flight-line surveys because the latter may bias abundance estimates upward. Nevertheless, both approaches have important caveats, and flight-line surveys can still be useful when used judiciously.

It is clear from our results that a single observer is unlikely to detect all Reddish Egrets at a given colony site during a direct count. Reddish Egrets sometimes flushed from nests upon approach, and the subsequent lack of an attending adult probably led to some undercounts of nests, especially during the second count when birds were sometimes more wary of observers. Many nests were well concealed in dense vegetation, and the similarities between the eggs and nest structures between Reddish Egrets and other species may help explain different counts between observers. In one case, the large size of an island (Alafia Bank; > 14 ha), the number of nesting pairs of other wading birds, and the dense vegetation precluded thorough direct counts within a minimally disruptive time frame. Nevertheless, overall

Table 1. Estimated nest counts from two 1-hr, 2-hr, and 3-hr flight-line surveys, aggregate nest totals from two direct counts, and flight-line survey totals from two observers at a single point at 12 known colony sites with nesting Reddish Egrets in Florida, 2015. Direct count data from three colony sites that did not receive flight-line surveys are not presented.

	Flight-line Count #1				Flight-line Count #2				Two Observer Comparison	
Site	1 hr	2 hr	3 hr	Direct Count	1 hr	2 hr	3 hr	Direct Count	Observer 1	Observer 2
Tampa Bay										
Alafia Bank	10.00	11.50	12.50	3	12.00	14.25	15.00	3	5.50	4.00
Alligator Lake	6.00	4.00	4.00	4	3.00	3.25	3.50	4	3.00	2.50
Clearwater I 25	0.00	4.50	3.00	2	4.50	3.75	2.50	2	4.50	6.00
Coffeepot Bayou	0.00	2.25	2.00	4	0.00	0.75	2.50	4	3.50	1.50
Cortez Key	4.50	8.50	6.00	4	7.50	10.75	8.00	4	6.50	5.50
Lower Keys										
Budd NW	12.00	10.50	10.50	5	3.00	9.00	8.00	5	4.50	4.50
Happy Jack	7.50	1.50	3.50	5	0.00	0.00	1.00	5	2.00	2.50
Torch Key Mangrove	1.50	0.75	1.00	3	0.00	0.00	3.50	3	1.00	1.00
Florida Bay										
Central Jimmie	13.50	12.75	11.50	2	25.50	18.00	15.00	2	7.50	5.00
Eagle Key	1.50	6.75	6.50	3	0.00	2.25	5.00	3	4.50	5.50
Duck Key	28.00	36.25	36.00	16	24.00	36.00	35.50	14	18.00	27.50
Stake Key	7.50	22.50	22.00	2	21.00	24.00	24.00	2	4.50	2.00

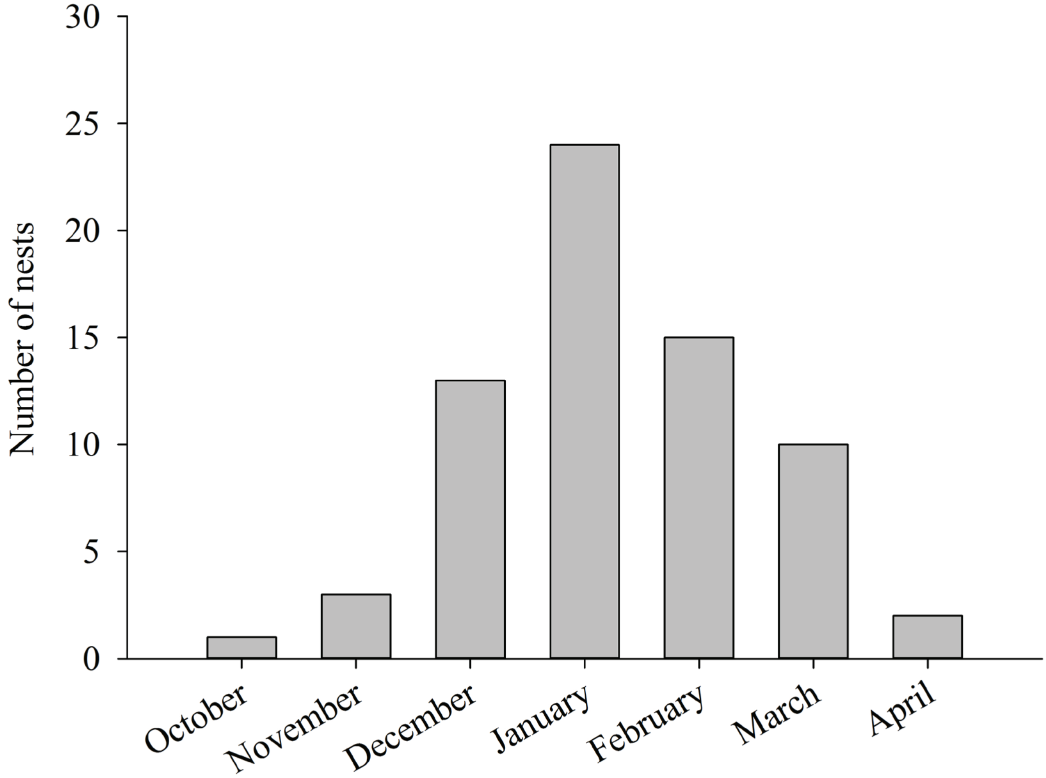

Figure 2. Estimated initiation dates by month for 68 Reddish Egret nests at 12 nesting sites in Florida Bay, Florida, USA, 28 October 2014-25 April 2015.

the two independent visits detected nearly 90% of known nests, and our intercept-only modeling approach to estimating detectability and abundances produced reasonable results. Future efforts that incorporate detection covariates such as island size should further refine nest estimates.

Observer error during flight-line surveys, as estimated by having two observers count incoming and outgoing Reddish Egrets at a single point, was greater than we had anticipated given the unique appearance of the species and the limited number of birds counted within each survey window. Some of the error may have been due to white-morph Reddish Egrets being mistaken for other species, and some colonies with relatively few Reddish Egrets had large breeding populations of other species that could have made counts difficult. Regardless of what caused the error, there are two reasons not to have multiple observers stationed at each point. First, it would at least double the labor costs of each survey and frequently require four or more observers because most colony sites require observers stationed at two separate points and the largest colony sites can require up to four. Second, individual observers did not consistently undercount or overcount Reddish Egrets during the paired incoming/outgoing trials, suggesting that imprecision in the counts will reduce precision but not substantially affect bias. Indeed, 2-hr and 3-hr flight-line surveys done twice at the same colony site produced an acceptably low error (< 20%). By contrast, error for 1-hr flight-line surveys was high (> 60%), and no birds were detected at 25% of the colony sites known to have nests. We recommend that flight-line surveys last at least 2 hr.

The direction of the bias we observed for flight-line surveys contrasts with a previous study in which flight-line counts tended to underestimate nest counts at relatively large wading bird colonies (Erwin and Ogden 1980). The difference may have occurred simply because it is harder to count birds at the larger colonies Erwin and Ogden (1980) surveyed. In addition, our assessment of flight-line survey bias is predicated on the assumption that the direct counts we performed were accurate. We know this is untrue to some extent because Reddish Egret nests were found at our colony sites concurrent with this study but during unrelated surveys, and at a rate that was somewhat higher than our model-based detection rates would predict. Nevertheless, our experience with the species at the colony sites suggests that our final nest counts were inaccurate at just one colony site, which was among the largest in acreage and hosted the greatest number of nesting wading birds. Another potential cause for the bias we observed relates the behavior of adult egrets. Our approach assumes that one nest exchange occurs per morning and that counts corrected for a 3-hr time period best reflect the behavior of the species. In reality, mates might exchange places more than once each morning during the incubation and early nestling guarding stages as the duration of foraging bouts can be quite variable in ardeids regardless of nest stage (Maccarone *et al.* 2012). More study is warranted because detailed data on nest attendance patterns of Reddish Egrets are lacking, and it is feasible that attendance rates may be especially variable given the species' dependence upon tidal flats.

Perhaps the most important contributors to the bias we observed related to site-specific factors such as a lack of breeding synchrony and/or interior lagoons that are suitable for foraging. Logistical constraints prevented us from assessing breeding asynchrony outside of Florida Bay, but the pronounced breeding asynchrony we observed there is consistent with many tropical species that have prolonged breeding seasons that are less closely tied to seasonal climatic constraints. This asynchrony likely leads to behavior by adults that violates a core assumption of the flight-line survey method: that only one exchange between adults occurs each morning. This will tend to bias count results upward during the later nestling stages as adults make more frequent trips to the nest to feed young. Paul *et al.* (2004) introduced an adjustment factor to account for breeding asynchrony in surveys of nesting wading birds. The factor requires that the nest contents be known for a substantial portion of a colony site, but we cannot gather this information directly from nests at colony sites with restricted access. It may be possible to develop a correction factor using the beak color of breeding Reddish Egrets, which is brightest during courtship and incubation and tends to fade as eggs hatch and nestlings grow, but doing so would require further study. It is likely that the presence of interior lagoons that are suitable for foraging, which may attract birds that are breeding elsewhere to a colony site, also contributed substantially to the bias we observed for flight-line counts. The lagoons may be less sensitive to tidally driven changes in water depth and as such may offer more reliable foraging habitat for Reddish Egrets, which prefer a narrow range of water depth for foraging (typically 5-15 cm; Lowther and Paul 2002). Indeed, the two most biased flight-line counts in this study both occurred at colony sites with interior lagoons (Central Jimmie and Stake Keys; Table 1).

In addition to the bias in our counts, the flight-line surveys were time consuming. A survey team could perform only one survey per day because the protocol requires that surveys start shortly after dawn. As such, relatively few surveys can be performed during the short time a synchronous population is in the incubation or nest-guarding stages. This, combined with the bias described above, suggests that it is infeasible to rely solely on flight-line surveys for a population survey. Nevertheless, direct counts simply are not an option at colony sites where land managers prohibit colony access by foot or when entry into a colony is deemed unduly risky to nesting birds.

The challenges outlined here are not unique to Reddish Egrets or to our chosen survey methods, but they do suggest that careful consideration of the appropriate survey method is warranted. We recommend that researchers avoid relying on flight-line surveys to the extent possible and use site-specific factors to determine when they are inappropriate. Specifically, we suggest that colony sites with low breeding synchrony and/or substantial internal foraging habitat should not be surveyed using flight-line surveys. Nevertheless, infrequent use of flight-line surveys may be a necessary component of a survey effort that largely relies on direct counts and should not substantially bias overall population estimates. We also recommend that colony sites be surveyed twice to characterize detectability and/or count error, which can be substantial. The use of multiple survey types requires the combining measures of error across survey types, which may result in wide confidence intervals around the overall population estimate and thus make it difficult to document minor changes in population trends. Nevertheless, we believe this approach acknowledges the challenges associated with monitoring Reddish Egrets and provides a flexible but repeatable protocol to allow for the estimation of Reddish Egret populations.

Acknowledgments

Funding for this study was provided by the State of Florida's Threatened and Nongame Species Management program and the Non-game Trust Fund. In-kind support was provided by Avian Research and Conservation Institute and Audubon Florida. We thank Brehan Furfey, Adam Emerick, Elaine Mason, Carol Cassels, Ed Kwater, and Sean Johnson-Boyce for their help in the field. The Florida Keys National Wildlife Refuge Complex provided housing and access to field sites. All applicable ethical guidelines for the use of birds in research have been followed, including those presented in the Ornithological Council's "Guidelines to the Use of Wild Birds in Research."

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