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Cover: Silva et al. (in this issue) investigated the breeding biology of Hyacinth Macaws (Anodorhynchus hyacinthinus) nesting in tree cavities in southern Amazonia. Photo author: João Marcos Rosa.

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Beyond a feeding and thermoregulatory structure: toucan's bill as a sword and pincer

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ABSTRACT: Toucans (Ramphastidae) have always attracted researcher's attention for their exuberant bill shape and size, which function has been often related to feeding strategies and thermal regulation. In this study, we discuss the importance and use of the bill by Ramphastidae species in intraspecific disputes. We present novel data and video recordings on agonistic encounters between females of the Spot-billed Toucan *Selenidera maculirostris*, along with a compilation of data from the Brazilian citizen science platform WikiAves and previous studies on six other Ramphastidae species. Until now, only a couple of species was known for using their bill in fights against conspecifics. Our study highlights the between-species behavioral similarities and the widespread occurrence of such behavior in the family, suggesting that this may be more common than previously thought and proposing hypotheses on the likely roles of such intraspecific contests.

KEY-WORDS: agonistic behavior, citizen science, female disputes, Ramphastidae, Selenidera maculirostris.

INTRODUCTION

Since the observations of bill morphology variations in Darwin's finches and their relationships with feeding habits (Grant 1981), there has been a noteworthy interest for the relationships of this structure with birds' ecology and behavior (e.g., Podos 2001). Besides its main use for feeding, some bird species also use their bill as warfare tool against predators (e.g., mobbing) and in agonistic conspecific disputes for resource, territory and mate (e.g., Murphy et al. 2009, Rico-Guevara & Araya-Salas 2014). Despite intraspecific agonistic encounters also occur irrespective of individuals sex, literature is often biased towards male-male interactions (Clutton-Brock 2007). Nevertheless, studies show that in some species such as monkeys (Koenig et al. 2004) and anhingas (Sazima & D'Angelo 2012) females may fight for resources or even for dominance over each other (e.g., crickets; Delago & Aonuma 2006).

Females were historically perceived as passive, little aggressive, or coy (Darwin 1871), but modern less biased approaches have been revealing how improper this is (*e.g.*, Hrdy 2006). Besides the currently accepted assertion of females' role on sexual selection of male traits and ornaments (Andersson 1994, Andersson & Simmons 2006), females may also play key-roles in territory establishment and competition for mates (Clutton-Brock 2007 & 2009, Rosvall 2011). In this study we provide a jointly interpretation of the bills' function and the female role in ecological behavior, adding to the perspective of bills use as weapons by disputing females.

METHODS

The Neotropical toucans (Aves: Ramphastidae) have one of the most noteworthy bill morphology, which is not only tightly related to feeding behavior (Bühler 1995) - assisted by a strong leg musculature (Moermond & Denslow 1985) - and thermoregulation (Tattersall et al. 2009, Hughes 2014), but also seems to be a dueling weapon (Van Tyne 1929, Brydon 1995, Ehrlich et al. 2001, Ritterson & Stein 2011). During regular fieldwork days in July 2008, at an Atlantic Forest reserve, in southeast Brazil - Ilha do Cardoso State Park, Cananeia, São Paulo state -, we recorded two events of a novel agonistic dispute between females of the Spot-billed Toucanet Selenidera maculirostris. In the next lines, we describe these encounters in detail (video recordings available as supporting material (Suppl. 1), followed by the results of a comprehensive review for similar agonistic behavior in other Ramphastidae species. Then, and to the best of our knowledge, we conclude with the first ever discussion on the recurrence of this behavior and its likely functions in the family.

RESULTS & DISCUSSION

We registered – with binoculars (10×42) and camera (Sony DSC-H1) from about 15-25 m distance - the first agonistic encounter at a point where a trail crosses a lowland pristine forest area (approximate coordinate: 25°04'46"S; 47°55'36"W). At ~07:30 h (GMT -3) on 10 July 2008, we spotted and heard two females flapping and ruffling wings atop (~12 m) of a tree. Females (hereafter referred to as A and B) were repeatedly beating and pecking each other using their bill in an apparent attempt to dislodge each other from the perch. When females were spotted, female A was biting the right-side facial skin and feathers of female B. Both females were vigorously flapping their wings and shaking their body with their legs. Some seconds after this "bill wrestling" started, female A managed to push B so the later got hanging down the perch being solely suspended by A's bite. After ~40 s flapping its wings, B got rid of female A's bill, quickly reverting the situation: female B managed to bite A's face and push it hanging down the perch. Then, female A's feet slipped off the perch one at a time and she stood belly up while holding female B's head. After ~12 s in that position, female B's feet loosened from the perch and both individuals fell for ~5 m high. During the fall, female A remained still and holding B's head which opened the tail feathers and flapped wings four times in an apparent attempt to control the fall. When both hit some leaves and branches, they got loose and flew to opposite directions. During all the fight, a male remained quiet and still on a nearby tree (~15 m), apparently watching the fight, and flew to a perch near female B where both stayed for a few minutes until fly out of sight.

The second event occurred on 13 July, about 1.5 km far from the first (25°5'33"S; 47°55'35"W) in the under-canopy of a primary forest area and again involved two females (C and D). Despite all birds had no natural or artificial markings and observations occurred far from each other, it is uncertain these were independent behavior, *i.e.* performed by distinct pairs of females. At 08:05 h, we heard one female (C) perching on a tree at ~10 m high, apparently at the end of a pursuit flight after another female (D), which perched on a nearby branch. After a few seconds, female D went to the same branch C was and both started "bill wrestling" as did females A and B. In the beginning, both kept striking each other's bill, but at times females kept pulling each other while their bills were interlocked: while one was biting the other's lower mandible, the latter was biting the former's upper mandible. Females remained around 2 min in this dispute, when female C finally bit D's left-side facial feathers near to its throat. With that, female C pushed D until she got hanging down the perch. After -12 s, female D slipped off the perch, but differently from the dispute

between females A and B, female D could not hold herself on C, which kept flapping its wings at times and moved along the perch while twisting and shaking the hanging opponent. After ~13 s, female D beat its wings and managed to grab C's throat with its left foot, which in turn reduced the amplitude of the shaking movements. Females remained in this position for ~45 s, when D's feet got loose and she got being suspended again by C. Approximately 13 s latter, D slipped off C's bill (a single clap sound could be heard) and free fell about 2 m until she flew to a nearby lower perch -4 m away. After a couple of minutes, D flew back to the wrestling perch and restarted bill strikes and interlocking for another 3-4 min. Then, a male, which remained still and quiet on a nearby perch (~10 m) during the entire duration of the fight, flew and perched aside female C and on the opposite side of female D. At this point, females where facing each other, perched on different branches, and kept fighting with no noticeable behavioral change upon male's arrival. A few seconds after that, and despite leaves obstructed the observers' sight, female C seemed to have bitten D's, forcing its head down (noticed through its raising tail). After ~10 s, another bill clap could be heard, at the same time that female D flew away. Female C kept wiping its bill for a few times and then flew to a nearby upper perch. The male remained in the perch for another 1-2 min and then followed female C. This entire fight lasted ~11 min, vs. ~2 min in the first event, but we caution that these are incomplete data since we only spotted both fights when they were already ongoing. We heard no vocalization of the females or of any other individual during any of the events.

We run an extensive literature review for evidences of aggressive behavior in other Ramphastidae species. Google Scholar (www.scholar.google.com), we In ran searches combining the family name with all the following terms (one at a time), both in English and Portuguese: female, agonistic behavior, agonistic encounter, aggression, dueling, and fight. This resulted in only one reference explicitly describing aggressive behaviors in Ramphastidae (e.g., fence duelling in Ramphastos sulphuratus brevicarinatus; Van Tyne 1929) and one record of Pteroglossus aracari killing each other in captivity in two events with no further details (Pernalete 1989). Further unstructured literature review led us to three other references, two for P. torquatus erythropigius (Brydon 1995) and one for R. ambiguus swainsonii (Ehrlich et al. 2001, Ritterson & Stein 2011). In addition to the literature review, we also searched for evidences in two large citizen science databases, the Internet Bird Collection (www.hbw.com/ibc) and WikiAves (www. wikiaves.com.br). We narrowed the research to all species occurring in Brazil, for which there was more available data (27,707 records at the WikiAves as of 19 April 2016

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and 1023 at the IBC), totaling 22 species (Piacentini *et al.* 2015), about 60% of all 34 extant Ramphastidae (Remsen-Jr. *et al.* 2017). We analyzed each and all existing photos and videos for every species in those databases. With that, we successfully found a few additional evidences that four other species may have similar aggressive behavior: the Toco Toucan *Ramphastos toco* (record made in August at Cataguases, southeast Brazil; Filho 2012) the Channelbilled Toucan *R. vitellinus* (in September at Macapá, north Brazil; Albano 2012), the Red-breasted Toucan *R. dicolorus* (in October at Campos do Jordão, southeast Brazil; Rodrigues 2012), and the Chestnut-eared Aracari *Pteroglossus castanotis* (in June at Foz do Iguaçu, south Brazil; Bolivar 2012).

All three Ramphastos species showed agonistic behaviors very similar to those described above for the Spotbilled Toucanet. However, it was impossible to assign the sex of individuals involved in those records because sexes are identical or have inconspicuous dimorphism. Thus, insofar our report for the Spot-billed Toucanet aggression is the first in describing exclusively female-female fencing contests in Ramphastidae. In all cases involving Ramphastos species, one individual used its bill to hold the other suspended bellow the perch. Observer's notes for the R. vitellinus record describe that both individuals slipped off the perch, likely free falling while flapping its wings. In this case, there is no report of the presence of a third individual. For the other two Ramphastos species, the individual that remained perched held the other suspended by its throat, but what happened in R. dicolorus record after that remains uncertain. From this point on, we describe the disputes involving R. vitellinus and P. castanotis, extracting as many details as possible from the observers reports (Albano 2012, Bolivar 2012). Prior to the moment when one individual of *R. vitellinus* was held hanging by the other, both birds struck each other with their bills (Albano 2012). When individual A pushed and held B suspended by its throat, a third bird perched aside A and attacked B for ~90 s (Albano 2012). After that, B fell motionless to the forest floor, but it remains uncertain if the individual died (Albano 2012). In the P. castanotis record, two birds got their bills interlocked, with one of them being held suspended. The observer describes that all other four or five individuals in the flock were apparently trying to break the birds apart by hanging themselves on the suspended individual and striking both birds bills (Bolivar 2012). After ~4 min, the two birds were set apart, fell to the ground for a few seconds and all flew away (Bolivar 2012).

This is the first time that any intraspecific agonistic behavior is formally described for these five ramphastid species (*R. toco, R. vitellinus, R. dicolorus, S. maculirostris* and *P. castanotis*). Moreover, the resemblance of such behavior among species is noteworthy. In fact, the

confrontations reported for two species of the family -R. ambiguus swainsonii (Ehrlich et al. 2001) and P. torquatus erythropygius (Brydon 1995, Ritterson & Stein 2011) are very similar, the latter culminating in death of one individual. In all cases, authors suggested intraspecific within-group dominance hierarchy as the motif for the fights, perhaps associated with male-male disputes for females. Thus, our study is the first to confirm that females could also act in such within-sex disputes. Altogether, this behavior is currently recorded for ~23% of all Ramphastidae species (n = 9 R. toco, R. a. swainsonii, R. s. brevicarinatus, R. vitellinus, R. dicolorus, S. maculirostris, P. t. erythropygius, P. aracari, and P. castanotis) and is widespread through all family clades (Hughes 2014). Therefore, it is plausible that this behavior also occurs in other species of the family, representing an additional role of the bill morphology and a likely result of morphological and behavioral coevolution, which deserves further study. Ramphastidae have well developed leg muscles, which importance was so far related to their feeding strategy of reaching and picking fruits hanging upside down and at the tip of branches (Moermond & Denslow 1985). Our observations add to these findings, since such anatomical adaptation seems also to be an important individual trait in fights. Individuals with stronger legs should have higher success rates in disputes, as it would be more able to hold still while pushing and holding its opponent hanging below the perch. We also suggest additional and more comprehensive anatomical evaluations not only of leg, but also of jaw muscles (e.g., Bühler 1995), in an attempt to reveal further morphological specializations and their ecological and behavioral roles.

The context of all aggressive contests described in this study allows inferences on the adaptive value of this behavior within the family. In species which individuals often live alone (e.g., R. toco), flocks (e.g., R. vitellinus), or in (socially) monogamous pairs such as S. maculirostris (Sick 1997), such behavior may represent a dispute for matting and/or territory. Since only females of the latter species were actively observed fighting while males remained as spectators, we propose three non-excluding hypotheses that the aggression may be (i) a female's strategy to compete for food resources or even for nesting cavities (e.g., Christianini 2018), (ii) to reduce male extra-pair copulation, likely ensuring the highest male parental care to the offspring, and (iii) a consequence of sex ratio bias towards males in this species, leading to a more intense and frequent female dispute for a mate (Rosvall 2011). The aggression in S. maculirostris occurred a month prior to its known nesting period at Ilha do Cardoso (Guaraldo & Staggemeier 2009), which could indicate a role as pre-breeding dispute for mating and nesting territory establishment. This behavior may be more widespread among Ramphastidae species than previously documented, since all but one record (July, dry period in Costa Rica; Brydon 1995) occurred during each species' breeding period. An exception to a breeding-context disputes is *P. castanotis*, in which the role of fights is less clear and could range from contests for mating to withingroup hierarchical position establishment. We believe that future studies of marked individuals are mandatory for allowing researchers to test these hypotheses.

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Evidence of breeding activity of subadult *Turdus* thrushes in Argentina

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ABSTRACT: Little is known about the age at which many Neotropical bird species first attempt to breed. We evaluated the breeding condition of 515 adult and subadult males and females among four species of *Turdus* thrushes (*T. nigriceps, T. chiguanco, T. amaurochalinus* and *T. rufiventris*) in Tucumán, Argentina during three breeding seasons (2015 to 2018). We registered a total of 126 individuals with brood patches and cloacal protuberances, which accounted for 24.5% of thrushes in breeding condition that we sampled. Forty thrushes had a brood patch (31.7% of those in breeding condition), of which 11 were subadults (8.7%). Eighty-six thrushes (68.3%) had a swollen cloacal protuberance, 18 of which were subadults (14.3%). Only a few of the subadults in breeding condition built nests, incubated eggs or raised nestlings. Of 130 nests we found, only 3 belonged to a subadult thrush (in all cases, a subadult female with an adult social mate), one of which was successful. Further research on these patterns among various Neotropical bird species could lead to important insights into the life history strategies that characterize different populations, and how these ultimately affect their population dynamics.

KEY-WORDS: brood patch, cloacal protuberance, nest, reproduction, Tucumán.

INTRODUCTION

A substantial body of research has shown that a bird's reproductive performance can vary considerably over its lifetime (reviewed by Newton 1989). Reproductive success in passerines generally improves with age (e.g., Förschler & Kalko 2006), with older birds initiating nesting activities earlier, having larger broods, and lower rates of nest failure (Robertson & Rendell 2001). Females that breed in their first year tend to raise fewer offspring per season than older females (Nol & Smith 1987), although nest success eventually diminishes after peaking in middle-aged females (Robertson & Rendell 2001). Reproductive success is generally lower when one member of a breeding pair is younger, whereas experienced pairs, consisting of two adults, show higher reproductive performance (Geslin et al. 2004). Thus, the first year in a bird's life appears to be generally characterized by low reproductive success (Förschler & Kalko 2006).

Evaluating how age influences reproductive success is important for understanding the evolution of life history

strategies and the causes of population demographic patterns (Clutton-Brock 1988). According to life history theory, birds should begin breeding when the net benefits are greater than those that promote delaying breeding (Promislow & Harvey 1990). Breeding at an early age can be beneficial if it reduces the chance of an individual dying before producing offspring or if it maximizes lifetime reproductive success. However, breeding at an early age can be risky if survival of the offspring or of the adults is reduced due to inexperience. Numerous previous studies have shown support for the idea that reproductive success increases with age, due to higher breeding experience of older birds (Nol & Smith 1987).

Nevertheless, our understanding of the causes of variation in reproductive success with age remains incomplete. It some cases, it is apparent that a positive relationship between reproductive success and age is due to age-related experience in such tasks as foraging (Förschler & Kalko 2006) and the care of eggs and young (Robertson & Rendell 2001). Lower reproductive success may also change with age due to senility and lower fertility as a bird ages (Partridge 1989). Additionally, as the value of current reproduction increases relative to that of future reproduction (as should be the case with older *vs.* younger birds), the stress response may be attenuated, with older birds exhibiting higher stress-induced prolactin levels ensuring that reproduction is not suspended compared to younger birds (Angelier *et al.* 2007).

The genus *Turdus* is one of the largest avian genera on the planet, comprising 85 species (Gill & Donsker 2016). Substantial information on how to estimate the age of *Turdus* thrushes is already available, facilitating research on their age-specific behaviors. *Turdus nigriceps* and *T. rufiventris* acquire definitive adult plumage after 14 months and *T. amaurochalinus* do so in at least 16 months, although they do not necessarily begin to breed at that age (Howell *et al.* 2003). Molt cycle information is lacking for *T. chiguanco*; nevertheless, subadults of all the species we studied are in their formative plumage in spring, which is similar to adult plumage except that it is more opaque and heavily worn. They may also exhibit some retained juvenile body feathers and wing coverts (*e.g.*, Ortiz *et al.* 2017).

Previous research has shown that subadult *Turdus* thrushes that are one year old may have a well-developed cloacal protuberance, which raises the possibility that these thrushes reproduce before acquiring their adult plumage (Capllonch 2015). Given the gap in information on age-related breeding effort in Neotropical passerines, we evaluated the incidence of breeding activity of subadult *Turdus* thrushes in northwestern Argentina.

METHODS

We captured thrushes using mist nets during their breeding season, from September-February 2015, 2016, 2017, and January-February 2018 at sites located within four major ecosystem types of Tucumán province, Argentina: Montane Chaco Woodland, Temperate Cloud Forest, Subtropical Montane Forest and Lowland Foothill Forest (Cabrera 1976). The sites we worked at are: 1) Ticucho, 26°31'S; 64°53'W, 600 m a.s.l.; this site is found in Montane Chaco Woodland, and common trees here are Schinopsis marginata, Acacia caven, Prosopis nigra, Geoffroea decorticans, Ceiba insignis as well as several types of cactus (e.g., Thrichocereus terscheckii, Opuntia sp.); 2) Botanical Garden, Reserva de Horco Molle, 26°55'S; 65°05'W, 550 m a.s.l.; this site is located in Lowland Foothill Forest, with common plants being Heliocarpus popayanensis, Tecoma stans, Juglans australis, Anadenanthera colubrine, Cedrela lilloi, Parapiptadenia excelsa and Allophyllus edulis; 3) El Rulo, Yerba Buena, 1000 m a.s.l.; this site is in Subtropical Montane Forest and is primarily composed of Cinnamomum porphyrium, Cupania vernalis, Myrsine

laetevirens, Myrsine ferruginea, Eugenia mato, Myrcianthes mato, Eugenia pungens, Eugenia seudo-mato, and Prunus tucumanensis; 4) Quebrada del Río Blanquito, town of Tafí del Valle, 26°52'S; 65°41''W, 2400 m elevation; this site is in Temperate Cloud forest and primarily composed of Alnus acuminata with scattered Sambucus peruviana, Rosa rubiginosa; 5) Rancho del Hombre Solo, El Infiernillo, km 78 on Route 307, 2800 m elevation; this site is located 20 km north of the town of Tafí del Valle (26°49'S; 65°43'W) and is primarily composed of grasses (Festuca hieronymeus) and patches of bushes (Iochroma australe, Baccharis sp. and Satureja parvifolia).

Data collection

We classified a thrush as a juvenile (*i.e.*, hatched during the current season), as a subadult (*i.e.*, hatched during the previous season) or as an adult (*i.e.*, hatched at least two seasons prior to capture) using the amount of skull ossification, the color and texture of the gape (*i.e.*, soft, swollen and yellow in juveniles) and plumage/molt characteristics (*e.g.*, shine, wear, retained juvenile feathers or recently molted; Wolfe *et al.* 2009, Ortiz *et al.* 2017).

Turdus can be reliably classified as subadults because they retain the juvenile wing coverts, which exhibit buffy tips, and exhibit swollen gapes for several months during the first year of life (*e.g.*, Ortiz *et al.* 2017). After breeding and before migrating in fall, they molt body feathers, remiges and rectrices, molting into their basic definitive plumage, which is much brighter and without buffy tips (Capllonch 2015). For example, *T. amaurochalinus* begins molting immediately after breeding in late summer or in early autumn and continues molt during fall migration (Soria *et al.* 2008).

We classified a thrush as being in breeding condition by the presence of a swollen cloacal protuberance and a developed brood patch (Ralph *et al.* 1996) and also determined sex by the presence of a swollen cloacal protuberance (males) or developed brood patch (females; Ralph *et al.* 1996). For *T. nigriceps*, we also used sexual dimorphism in plumage as an indicator of sex (Narosky & Yzurieta 2010).

Nest monitoring

We conducted nest searches by walking along paths, paying particular attention to forks in branches of trees and shrubs, which are typical locations where thrushes place their cup nests. We also observed the behavior of adult thrushes to find nests, such as escape movements from the nest when we approached. Females will also make a distinct "cluck" call when an observer is near the nest, which helped detect the presence of an active nest. When nests have nestlings, both parents are often near the nest and make distinct vocalizations, which aids in finding the nest. We checked nest contents using a ladder or with a video camera attached to a long pole. We classified a nest as successful if it fledged at least one nestling. We classified a nest as failed if there were missing eggs (*i.e.*, which were present in a previous visit to the nest) or when nestlings that were too young to fledge were missing. If a nest failed, it was checked at least once more to determine if it was re-used. If a nestling fledged from a nest, the nest was checked again ten days later to determine if the nest was re-used.

RESULTS

We caught a total of 515 *Turdus* thrushes from four species (*T. nigriceps*, *T. chiguanco*, *T. amaurochalinus* and *T. rufiventris*), 126 of which were in breeding condition, accounting for 24.5% of all thrushes captured. Forty thrushes had a brood patch (31.7% of those in breeding condition), of which 11 were subadults (8.7%). Eighty-six thrushes (68.3%) had a swollen cloacal protuberance, 18 of which were subadults (14.3%).

We monitored 130 nests, 38 of which belonged to *T. amaurochalinus*, 12 to *T. nigriceps*, 38 of *T. chiguanco*, and 42 to *T. rufiventris* (Table 1). None of these nests belonged to a subadult male, three belonged to a subadult female, and the rest belonged to adults. Success rates of nests belonging to adults (*i.e.*, excluding the three nests belonging to a subadult female) ranged between 28.6 and 50% (Table 1). In all three cases in which the nest belonged to a subadult female, their social mate was an adult male. The first belonged to a *T. nigriceps* subadult female, which was captured next to the nest on 08 November 2016 in El Rulo, San Javier. This nest was successful, fledging three nestlings; the nest was not re-used. The second was a nest of a *T. amaurochalinus* subadult female, which was captured leaving the nest in Ticucho on 18 November

2017. The eggs were predated and the female did not reuse the nest. The third was located in the basin of the Blanquito River near Tafí del Valle and which belonged to a *T. nigriceps* subadult female that was captured on 03 Jan 2018 in a mist net placed near the nest. The three nestlings of that nest were killed in an intense storm, likely due to excessive water in the nest, which was placed in a cavity. The nest was not re-used.

DISCUSSION

We found that subadult *Turdus* thrushes attempt to breed, although our results suggest that only a small proportion of subadult *Turdus* thrushes do so. We only found three nests belonging to subadults, such that we cannot yet make definitive conclusions regarding the probability of success between adults and subadults; nevertheless, subadults of both sexes were found in breeding condition. Although we did not find any nests belonging to subadult males, the males we sampled that had a swollen cloacal protuberance may have pursued extra-pair copulations.

Previous research has shown that adults may benefit from holding territories in higher quality habitat with better nest sites and by having more experience with breeding and foraging, allowing them to maintain a proper energy balance, which is key to physiologically preparing for reproduction (Robertson & Rendell 2001). In contrast, subadult thrushes may use their first year to obtain the necessary skills necessary to successfully breed, such as nest building and foraging. Notably, we found several adult nests abandoned before laying eggs, whereas all nests of subadults had eggs. Further research comparing nest outcomes between adults and subadults at different stages of the nesting cycle may shed light on how these and other bird species develop the skills necessary to nest successfully.

Further studies on age-related breeding effort

Adult nests	Abandoned before laying	Failed at egg stage	Failed at nestling stage	Successful	Total nests	% Successful
T. amaurochalinus	1	14	6	16	37	43.2
T. nigriceps	0	5	0	5	10	50.0
T. chiguanco	4	13	9	12	38	31.6
T. rufiventris	1	24	5	12	42	28.6
Total	6	56	20	45	127	
Subadult nests						
T. amaurochalinus	0	1	0	0	1	0.0
T. nigriceps	0	0	1	1	2	50.0
Total	0	1	1	1	3	

Table 1. Nest outcomes for four species of *Turdus* thrushes monitored in Tucumán province, Argentina.

in Neotropical passerines not only offers a better understanding of the poorly-understood first year of life of Neotropical birds, it can also provide novel insights into the drivers of population dynamics within and across species, since a trade-off may exist between investing in reproduction early in life and fitness later in life (reviewed by Fay et al. 2016). For example, female Willow tits (Parus montanus) that breed every year of their first five years of life have a lower probability of survival later in life than females that skip breeding during at least one of their first five years of life (Orell & Belda 2002). Such a relationship between fitness and investment in breeding early in life may vary between populations, with the age at which individuals within a population first attempt to breed often being related to the environmental conditions experienced by each population (Fay et al. 2016). Thus, research on the age at which birds first breed contributes to a deeper understanding of avian life-history strategies and population dynamics, and is a subject for which we still have many gaps to fill.

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Reproductive behavior of White-tailed Kites (*Elanus leucurus*) in the Pampas of Argentina

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ABSTRACT: The White-tailed Kite (*Elanus leucurus*) is a raptor that has been less studied in the Neotropics than in the Nearctic region, especially in relation to its reproductive behavior. In this study, we report information about the phenology and activity patterns of this raptor at the Pampas of Argentina. We found that White-tailed Kites have a prolonged breeding season, from October to May. Time-activity budgets of mating pairs indicated a pronounced division of roles in parental care between sexes. Females devoted most of their time to nest construction, incubation and chick care (80% of total time) and males to food provisioning and vigilance (70% of total time). We registered 11 cases of prey transfer from the male to the female. In four cases the transfer occurred in flight and in the remaining seven cases while individuals were perched. Our results agree with general patterns on breeding behavior of White-tailed Kites from North America, suggesting a consistent behavioral pattern throughout the species' distribution.

KEY-WORDS: breeding, parental care, raptor, sex roles, time-activity budgets.

INTRODUCTION

The White-tailed Kite (*Elanus leucurus*) is a mediumsized raptor widely distributed across the Americas (43°N–46°S), from the southeastern United States to the Patagonia in Argentina and Chile (Figueroa *et al.* 2006, Thiollay 2019). This species inhabits open habitats such as wetlands, grasslands and savannas (Fjeldså & Krabbe 1990, Dunk 1995), but it is also commonly found in modified environments such as green areas of cities and villages, agroecosystems, farmlands, and garbage dumps (Ferguson-Lees & Christie 2001, Thiollay 2019). Kites are usually conspicuous during their foraging activities, due to their white coloration and because they search for prey performing showy flights at medium or low altitude (Mendelsohn & Jaksic 1989, Dunk 1995).

The feeding ecology is the best known aspect of White-tailed Kite's ecology. Most studies performed along the species' distribution range agree that this kite is a specialist predator of small mammals, particularly rodents (USA: Hawbecker 1940, Bond 1942, Dixon *et al.* 1957, Waian & Stendell 1970, Stendell & Myers 1973; Chile: Meserve 1977, Schlatter *et al.* 1980, González-Acuña *et al.* 2009; Argentina: Leveau *et al.* 2002, Sarasola *et al.* 2007, 2010, Baladrón *et al.* 2018; Brazil: Scheibler 2004). This species is characterized as an active-search predator that uses hovering as its main hunting technique (Watson 1940, Warner & Rudd 1975, Mendelsohn & Jaksic 1989, Dunk 1995, Baladrón et al. 2018). The information available about reproductive behavior of the White-tailed Kite is scarce and mostly comes from North America (e.g., Hawbecker 1940 & 1942, Watson 1940). The breeding season covers spring, summer and autumn in its northern range. This prolonged breeding season is related to the occurrence of double broods, especially in years of high food abundance (Hawbecker 1940, Dixon et al. 1957, Dunk 1995). There is little information about parental care, some authors arguing that both parents take part similarly in the incubation and nest construction activities (Watson 1940, Hawbecker 1942), while others indicate that females perform most of these tasks (Dixon et al. 1957). The male provides food to the female and chicks throughout the season (Hawbecker 1940, Watson 1940, Warner & Rudd 1975). In its southern range, the information available on reproductive behavior is scarce and, in most cases, anecdotal (Fraga 1984, Jaksic et al. 1987), although it is estimated that the breeding season would also be prolonged in this part of kites' distribution (de la Peña 2016).

In the framework of a broader study on foraging ecology of the White-tailed Kite in the Pampas of Argentina (see Baladrón *et al.* 2018), we registered different activities of kites related to reproductive behavior. In this study, we compiled and analyzed the information collected during such sampling, emphasizing on reproductive phenology, parental care, and prey transfer activity.

METHODS

The study was carried out in the southeast of Buenos Aires province, Argentina (37°32'S–37°55'S; 57°19'W– 57°30'W), which belongs to the Pampas region (Cabrera 1971). The landscape in this region is dominated by grasslands, although the original community of grasses has been heavily modified by livestock grazing and agriculture (Soriano *et al.* 1991, Bilenca & Mińarro 2004). The study area is characterized by its heterogeneity, including a diverse array of natural environments, such as grasslands, marshes, coastal dunes, and native woodlands, and modified environments, such as grazing fields, croplands, and urban areas (Isacch *et al.* 2016).

We identified the territories of two pairs of kites, from which we obtained data on phenology and behavioral patterns in different years between 2006 and 2008. One of the groups was located in a woodland area dominated by the native tree known as Tala (Celtis ehrenbergiana) in Nahuel-Rucá ranch (37°37'10"S; 57°25'20"W, hereafter "tala pair"). The activity of the tala pair was registered during four observation days (26 April 2006: 146 min; 27 April 2006: 279 min; 04 May 2006: 256 min; 21 October 2007: 82 min). The other group was located in a periurban area (Parque Lago village; 37°45'40"S; 57°27'15"W, hereafter "periurban pair"). The activity of the periurban pair was also registered during four observation days (27 April 2007: 160 min; 22 May 2007: 260 min; 17 January 2008: 88 min; 04 April 2008: 312 min). Using 10×50 binoculars, we recorded the behavior of kites individually from a fixed position (continuous recording method; Martin & Bateson 1993, Gaibani & Csermely 2007). From the total sampling time, we quantified only the reproductive activities of parents in the vicinity of the nest. We assigned sexes to individuals based mainly on their behavior, following the behavioral patterns reported for North America (Watson 1940, Hawbecker 1942, Dixon et al. 1957). Although the White-tailed Kite shows some sexual dimorphism (females are slightly larger and have darker backs than males; Dunk 1995), these features are of a comparative nature (Hawbecker 1942). Thus, we used external characters to differentiate sexes only when both individuals were seen together. In all cases, our assumptions of sex identification (either based on behavior or dimorphism) were positively confirmed during copulation events. All samplings were conducted under good weather conditions (no extreme wind, no rain nor fog).

We classified the activities of each individual in one of five categories: 1) vigilance: the individual watches over the nest from the air or from a perch; 2) perching: the individual remains inactive or performs maintenance activities (cleaning, grooming); 3) nest construction: the individual carries materials or builds the nest; 4) courtship: interactions between male and female related to courtship (*i.e.*, vocalizations, material transfer, prey transfer, copulation); 5) in the nest: the individual stays in the nest; we assumed that the main behavior during this activity was incubation or chicks' care (depending on the stage of the breeding cycle). We calculated timeactivity budgets for each sex as the proportion of time that each individual devoted to each activity respect to the total time registered each day. Thus, we obtained daily budgets for each individual, and then we calculated the mean values and standard errors of each activity for each sex. In addition, we described in a detailed manner the events of prey transfer from the male to the female and quantified the time that kites employed in prey handling and consumption.

RESULTS & DISCUSSION

In the southeastern Pampas of Argentina, the Whitetailed Kite showed a prolonged breeding season. The starting date of the breeding season was quite variable. We registered copulation events as early as 21 October (tala pair, 2007) and as late as 27 April (periurban pair, 2007). However, we also registered the tala pair making courtship displays and building the nest as late as 04 May 2006. Such delayed starting date would correspond to a second nesting attempt. Double-brooding is rare in accipitrids, however when the abundance of rodents is high and prolonged it is known that the White-tailed Kite, the Common Black-shouldered Kite (E. axillaris), and the Black-winged Kite (E. caeruleus) may raise two broods in the same breeding season (Mendelsohn 1983, Johnsgard 2009, Thiollay 2019). This behavior has also been recorded for cooperative breeders, such as Harris's Hawks (Parabuteo unicinctus), for which second brood attempts will depend on the presence of helpers at the nest (Johnsgard 2009, Thiollay 2019). The final of the breeding season was also variable. We registered fledglings in nest areas as early as 02 February (tala pair, 2007) and as late as 04 April (periurban pair, 2008). Thus, the duration of the breeding season included spring, summer and autumn, which is in accordance with previous reports from North America (Hawbecker 1940, 1942, Dixon et al. 1957).

Nests were located in the upper part of trees. The tala pair placed its nest at ~7.5 m of height in a tala, which was located at the edge of the woodland (Fig. 1). This nest was used by kites in previous and later years to our sampling period (A.V.B. pers. obs.). The periurban pair placed its nest at ~12 m of height in a *Eucalyptus* spp. In the same site, we registered another nest (which was abandoned early in the season) at ~4 m in a *Pinus* spp. These results agree with those from California, where this

kite shows little selectivity in the type and height of trees used for nesting (Dixon *et al.* 1957).

We registered a total of 550 min of White-tailed Kites' reproductive activities. Mating pairs showed a pronounced division of roles in parental care between sexes, which was revealed by different activity budgets between males and females (Fig. 2). From total time devoted by kites to nest construction, approximately 98% of time corresponded to females. Previous reports showed mixed results about the participation of sexes in nest construction. In California, for example, Watson (1940) and Hawbecker (1940) reported that both sexes build the nest, but later Dixon et al. (1957) indicated that this task is almost exclusively done by the female, which is in agreement with our results. However, since both pairs nested in previously built nests, in our study we did not record the complete process of nest construction. Many species of accipitrids use old nests, to which they add new material (Saggese & de Lucca 2001, de la Peña 2016), which may be a strategy to save time and energy at the beginning of the season (Thiollay 2019). Thus, the nest building activity that we observe was likely related to nest "maintenance", a task that is performed almost exclusively by the female in most accipitrids, although the male may bring much of the material to her (Rettig 1978, Salvador-Jr. et al. 2008, de Lucca & Saggese 2012). In later stages of the breeding cycle, females stood guarding near or into the nest and males devoted most of their time to vigilance and prey provisioning (Fig. 2). These results indicate that incubation and direct feeding of chicks would be done almost totally by the female. On the other hand, we found that the prey provisioning was performed in total by the male, which is in agreement with previous reports for California (Watson 1940). This pattern of role partitioning between sexes, with the male being responsible for most of the hunting and the female



Figure 1. Location of one of the nests of White-tailed Kites (*Elanus leucurus*) in the upper part of a Tala (*Celtis ehrenbergiana*) in the edge of a Tala woodland (**A**). Male watching the territory perched near the nest (**B**) and hunting in a neighboring patch (**C**). Photo author: M.S.B. (**A** & **B**); N. Chiaradia (**C**).

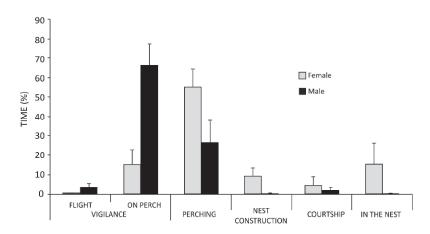


Figure 2. Time-activity budgets of male and female White-tailed Kites (*Elanus leucurus*) in the Pampas of Argentina. Values are expressed as percentages (mean and standard error).

for most of the parental care in the nest (*i.e.*, incubation, chicks care, and nest maintenance), is observed in most accipitrids, except in vultures (Newton 1979, Collopy 1984, Pavez 2001, Salvador-Jr. *et al.* 2008, Thiollay 2019).

We registered 11 events of prey transfer from the male to the female. All events began with the male arriving to the nest area carrying a prey in its talons and emitting whistling calls. Watson (1940) reported that the male approaches to the nesting area emitting vocalizations in order to annunciate his arriving and to coordinate the transfer. Then, the male hovered over the nesting area holding the prey with his feet downwards. We witnessed four cases of aerial transfer, in which the female leaves her perch and fluttered along the male and, when she was close enough, she positioned behind the male to take the prey with her claws. In the remaining seven cases, the transference occurred in perch. The male arrived carrying the prey and perched next to the female (or the female flies over and perched next to him), and then she took the prey from the side. Watson (1940) reported that both transfer methods were used in the same frequency by kites in California. Aerial food exchange seems to be a common practice among accipitrids, particularly harriers (Circus) and kites (Elanus). Although this behavior is clearly linked to sex role division (Watson 1977), the incidence of aerial transfer may respond to differential female requirements (e.g., hunger; Dixon et al. 1957), habitat characteristics (e.g., areas of dense vegetal cover; Watson 1977), and predation risk (e.g., for ground nesting species; Negro & Galván 2018). More studies are needed to elucidate the biological significance of this behavior.

Frequently, after prey transfer, individuals vocalized and made courtship displays, opening the wings and moving the tail. Once the female took the prey, she briefly handled it (range = 4-38 s), and lasted an average of 7.4 min to ate the prey (range = 1.15-16.7 min). The male stay perched patrolling the area while the female was eating (7 of 11 events). In the remaining cases the male flew away and resumed foraging. After eating, the female stood in the site in five cases and flew away with the male in two cases. In the remaining four cases, she breaks twigs from the tree, and carries it to the nest in her bill. The behavior of taking twigs and build the nest in response to food transfer seems also to be usual for this species in California (Watson 1940, Dixon et al. 1957). It was frequent that, while the female was eating, she was harassed by potential cleptoparasites, especially Chimango Caracaras (Milvago chimango) and, in a lesser extent, Southern Caracaras (Caracara plancus) (Baladrón & Pretelli 2013). In most cases, intruders were chased and escorted out of the territory by the male. This agonistic relationship with caracaras may be the counterpart of that reported between kites and ravens (Corvus spp.) in North

America (Dixon et al. 1957).

Studies on raptor behavior are scarce in the literature, especially in the Neotropical region. In the case of the White-tailed Kite, there is an asymmetry between the information available for North and South America (Hawbecker 1940 & 1942, Watson 1940, Warner & Rudd 1975, Mendelsohn & Jaksic 1989, Baladrón *et al.* 2018). Although based on sporadic records and a modest time of observation, our study show that in the Pampas the White-tailed Kite have a prolonged breeding season and show a pronounced division of roles in parental care between sexes. Our findings agree with previous reports for northern populations (Dunk 1995), and suggest a consistent behavioral pattern throughout species' distribution.

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Southeastern Brazilian tyrannulets as flower watchers

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ABSTRACT: Several species of insect-eating birds occasionally visit flowering trees or shrubs to feed on arthropods at flowers instead of nectar or petals. In southeastern Brazil, the Planalto Tyrannulet *Phyllomyias fasciatus* (10.3 g) and the Gray-headed Tody-Flycatcher *Todirostrum poliocephalum* (7 g) often watch flowers to get insects and can visit flowering trees for hours or for several days. We describe the foraging behavior of these two tyrannulets at flowering trees and also report observations on several other species, mostly tyrant-flycatchers and tanagers. As an opportunistic foraging strategy, flower watching can be expected to be more common among small, canopy or edge birds that sally or hover-glean to catch small insects on or near foliage.

KEY-WORDS: foraging behavior, insects, Phyllomyias fasciatus, tanagers, Todirostrum poliocephalum, tyrant-flycatchers.

INTRODUCTION

Birds often visit flowers for nectar or to eat petals (Willis 2002). Insects attracted to flowers provide another type of food, and certain birds watch flowers to catch them. Pereyra (1941) noted many birds eating insects in flowering corn fields, and suggested that such birds as the Spectacled Tyrant Hymenops perspicillatus could help pollinate the crop. Beehler (1980) reported several species catching insects in flowering trees in New Guinea, and the Green Honeycreeper Chlorophanes spiza in Panama (as described earlier by Snow & Snow 1971). Stiles et al. (1989) reported the Tropical Kingbird Tyrannus melancholicus getting butterflies near flowers, and stated that the Black-headed Tody-Flycatcher Todirostrum nigriceps often forages in flowering trees. Tampson (1990) noted the Cattle Tyrant Machetornis rixosa catching insects in palm flowers. Dobbs & Greeney (2006) observed that nearly 1% of insects captured by the Rufous-breasted Flycatcher Leptopogon rufipectus are taken on flowers.

Here, we discuss flower watching in southeastern Brazil by the Gray-headed Tody-Flycatcher *Todirostrum poliocephalum*, a close relative of *T. nigriceps*, and the Planalto Tyrannulet *Phyllomyias fasciatus*. These two tyrannulets occasionally visit flowers for hours, or for several days. We also report some observations for the Bran-colored Flycatcher *Myiophobus fasciatus* and others.

METHODS

Most observations were at the Santa Lúcia Biological Station (19°58'S; 40°32'W, 650 m a.s.l.) in the Santa Teresa mountain range of central Espírito Santo state, Brazil. Flowering trees were mainly cultivated avocados *Persea americana* (Lauraceae) and Japanese Plums *Eriobotrya japonica* (Rosaceae) by the laboratory and several native yellow-flowered *Senna multijuga* var. *lindleyana* (Fabaceae) along the entry road, 500 m upriver. Observations were made with 8×30 and 10×42 binoculars from an appropriate distance so as not to disturb the birds.

G.A.B. studied insectivorous and omnivorous birds foraging at flowers of three neighboring avocados on 04, 05 and 09 September 1994, for a total of 1405 min. For each bird visitation, data recorded included species, number of individuals, arrival and departure time, foraging maneuvers and substrate of captures. Whenever it was not possible to record an individual's arrival or departure time, we used the average visit length calculated for the species. Foraging maneuvers were classified according to Remsen & Robinson (1990) into eight categories. These categories were then combined with substrates of capture to generate composite codes representing foraging tactics (e.g., SH/F indicates a sally-hover to capture an insect on a flower). E.O.W. studied P. fasciatus, M. fasciatus and other species at flowers mainly April-May 1996. We also report observations on tyrant-flycatchers and other passerines

² In memoriam.

made elsewhere in southeastern Brazil. Scientific names of birds follow Piacentini *et al.* (2015). Bird body masses are from Wilman *et al.* (2014).

RESULTS

In September 1994, twelve species foraged on insects attracted to avocado flowers at Santa Lúcia (Table 1). Ten others visited flowers exclusively for nectar (*Thalurania glaucopis*, *Coereba flaveola* and *Dacnis cayana*) or perched/foraged away from flowers (Cranioleuca pallida, Xenops rutilans, Piprites chloris, Camptostoma obsoletum, Lathrotriccus euleri, Euphonia violacea and Tangara cayana). Todirostrum poliocephalum, P. fasciatus and three tanagers visited the trees in more than one day and made considerably more visits or spent considerably more time foraging at flowers than other species (Table 1). Todirostrum poliocephalum spent the largest amount of time on trees and made long visits concentrated in the second half of the morning, when it was a regular and constant visitor. Typically, one bird of a pair that held a territory in the area would forage for long periods in the avocados, sometimes accompanied by its mate. Aerial maneuvers (short sally-strikes and, to a lesser extent, sallypounces and leaps) were used to capture prey mainly on substrates other than air, and about 65% of captures were on or very close to flowers (Table 2).

Phyllomyias fasciatus was the most frequent species in the avocados. It made medium-length regular visits

throughout the morning and early afternoon (Table 1). Usually only one or two birds foraged in the trees at the same time, but up to four were present on some occasions. Prey were mostly captured in the air or at flowers with sally-strikes and, less frequently, sally-hover and sally-pounce maneuvers. Nearly 62% of insect-catching attempts were at or near flowers (Table 2). This species showed the greatest range of foraging maneuvers among all visitors.

The avocado flowers were visited earlier by a *T. poliocephalum* on 03 September 1994, with short strikes under leaves near the flowers. We have often seen it sallying under leaves away from flowers, at dense vine tangles in woods or at edges. E.O.W. also noted visits by two *P. fasciatus*, which sallied to the air or leaves near flowers at 10–12 h. Later, two birds were sallying in a small flowering tree (not identified) in the woods downriver. We have seen it sallying away from flowers, or getting *Myrsine* fruit or *Alchornea* arils, on other occasions.

On 24 April 1996, 16:22–16:28 h, one *P. fasciatus* sallied for insects in the *Senna*. On the 25 April, between 9–10 h, two were now and then near Japanese Plum flowers by the lab, between visits by tanagers and relatives that poked their bills deep into the flowers (*D. cayana, Tangara seledon* and *Schistochlamys ruficapillus*). A *T. poliocephalum* worked briefly near one of the *P. fasciatus*, but soon left. On the 26 April, E.O.W. watched at the *Senna* from 15:24 h to dark at 17:30 h. The main visitor was a *M. fasciatus*, but two *P. fasciatus* worked 16:00 h in the crowns, sallying to catch insects on flowers several times.

	No. of visits/day			Total No.	Maan lanadh	Total time	Average No.
Species	04 Sept	05 Sept	09 Sept	of visits	Mean length of visits	spent on trees	of individuals per visit
Todirostrum poliocephalum	-	3	8	11	26.3	289	1.1
Phyllomyias fasciatus	-	8	19	27	9.2	249	1.2
Saltator maximus	-	1	7	8	8.9	71	1.0
Tangara ornata	-	3	17	20	2.8	56	1.3
Tangara sayaca	-	1	4	5	5.0	25	1.6
Tolmomyias sulphurescens	-	-	6	6	3.2*	19.5*	+1.0
Hemitriccus nidipendulus	2	-	1	3	3.0	9	1.0
Pachyramphus castaneus	-	1	2	3	1.7	5	1.0
Pachyramphus viridis	-	-	2	2	1.2	2.5	1.0
Contopus cinereus	-	-	2	2	0.7	1.5	1.0
Piaya cayana	-	-	1	1	1.0	1	1.0
Hylophilus poicilotis	-	1	-	1	1.0	1	1.0
Total	2	18	69	89	-	-	-

Table 1. Number of visits on each day of observation, total number of visits, mean length of visits (min), total time spent on trees (min) and average number of individuals per visit for birds recorded foraging on insects attracted to avocado flowers at Santa Lúcia, Espírito Santo state, Brazil, in September 1994.

*possibly overestimated because the length of some visits could not be determined.

Maneuver/	Phyllomyias	Todirostrum	Tangara	Tangara	Saltator
substrate	fasciatus	poliocephalum	ornata	sayaca	maximus
SS/A (SS/A*)	49 (26)	7 (5)	-	3 (2)	2
SS/F	26	18	2	-	2
SS/LL	2	3	-	-	-
SS/UL	3	-	-	-	-
SS/Fo	5	-	-	-	1
SS/B	2	4	-	-	-
SP	-	-	1	-	1
SP/F	6	5	-	-	-
SP/Fo	1	-	-	-	-
SH	1	-	-	-	-
SH/F	6	-	-	-	-
FCh	-	-	1	1	-
Le	-	-	3	-	-
Le/A	2	-	-	-	-
Le/B (Le/B*)	1 (1)	2	-	-	-
Le/F	2	2	-	1	-
Le/LL	-	2	-	-	-
Le/Fo	-	2	-	-	-
Lu	2	-	1	1	-
Lu/F	-	-	-	1	-
RO	-	1	-	-	-
RO/F	1	1	-	-	-
RO/A*	1	-	1	-	-
RO/UP	-	1	-	-	-
RU/F	1	-	-	-	-
RU/A*	-	-	1	-	-
n	111	48	10	7	6

On 14 May, 15:54 h, two *P. fasciatus* were back to the *Senna* and worked near flowers, at times pecking deep inside, 16:00–16:10 h. After 5 min in the low bushes below, one returned for 3 min to sally to air or leaves near flowers, and again at 16:21 h for several sallies to the air, then dropping to a *Cecropia* below. A third bird and the two moved into the *Senna* 16:24 h to 16:32 h, then wandered off. At 16:42 h, a *P. fasciatus* returned for six more short sallies to the air, moving off by 16:50 h.

On 19 May, there were several visits by *P. fasciatus*. Between 14:38–14:51 h one did ten air sallies and three pecks into flowers before preening, then five other sallies (all next to flowers or leaves). At 15:44 h, two were in the *Senna* trees, sallying four times before moving to a *Myrsine* bush with tiny flowers. Between 16:01–16:04 h, two *P. fasciatus* returned, sallying eight times or more to the air by flowers. Between 16:10–16:34 h one worked the flowers, preening and regurgitating two seeds on a twig and defecating another, before 32 short sallies to near flowers 16:19–16:32 h. Between 16:36–16:38 h, one returned for five short sallies, then to a *Cecropia* off east. Two and then three birds continued to sally in the area 16:40 h on, using other trees and a telephone wire, but between 17:00–17:10 h some sallies were again near flowers, after which birds wandered off for the night.

On most days, the *M. fasciatus* worked low bushes under or near the *Senna*. On 26 April, however, one sallied in the midlevels of the *Senna* at 15:46 h, returning 15:52 h after some long calls and sallying to near flowers. It fled if people or bicycles passed, but returned to catch insects with sallies to flowers, the air, or foliage; at 16:23 h it dropped to bushes, sallied to the ground in the road a few times, and did not return to the flowers. The next morning, 08:05–08:15 h, the *M. fasciatus* was less timid and sallied to or near flowers up one tree several times.

Elsewhere, E.O.W. noted *P. fasciatus, M. fasciatus* and six other small flycatchers sallying or hovering for insects near flowers of trees or bushes in southeastern Brazil (Table 3). Late on 15 August 2002, fallen flowers of *Tabebuia chrysotricha* (Bignoniaceae) on the lawn of the UNESP Campus (22°23'S; 47°33'W, 620 m a.s.l.), Rio Claro, São Paulo state, attracted insect-eating birds such as the Great Kiskadee *Pitangus sulphuratus*, *M. rixosa*, woodpeckers, wrens and others, while flowers in the trees nearby attracted the Yellow-bellied Elaenia *Elaenia* *flavogaster* and the White-crested Tyrannulet Serpophaga subcristata.

In southern Brazil, G.A.B. noticed two other tyrant-flycatchers watching flowers of bushes to catch insects (Table 3). In addition, in the Pampas grasslands around Lavras do Sul (30°48'S; 53°54'W, 315 m a.s.l.), 05-08 January 2018, at least three species of tyrantflycatchers were plucking soldier beetles Chauliognathus flavipes (Cantharidae) from the umbellate flower stalks of Eryngium chamissonis (Urb., 1879) (Apiaceae) in densely vegetated upland swales dominated by this spiny sedge. These polymorphic, soft-elytra beetles are distinctly colored with yellow and black and gather by the thousands on the upright inflorescences of E. chamissonis, which rise up to 1.5 m above the vegetative stratum in midsummer to form a temporary emergent layer of flowering shoots. Birds seen capturing the beetles in a more or less systematic way included a family group of Yellow-browed Tyrants Satrapa icterophrys on the 04 January, plus a solitary individual on the 06 January, and two M. fasciatus on the 05 and 08 January, at four different locations. One

Table 3. Observations of tyrant-flycatchers (Rhynchocyclidae and Tyrannidae) foraging on insects attracted by flowers insoutheastern Brazil.

Species	Plant (family)	Locality (coordinates, altitude, a.s.l.)*	Date (observer)**
<i>Phylloscartes ventralis</i> Mottle-cheeked Tyrannulet	<i>Rubus rosifolius</i> (Rosaceae)	Mariana Pimentel, RS (30°19'S; 51°36'W, 230 m)	18 July 1997 (1)
<i>Tolmomyias sulphurescens</i> Yellow-olive Flycatcher	<i>Ceiba speciosa</i> (Malvaceae)	near Broa Reservoir, SP (22º08'S; 47º52'W, 740 m)	25 March 1984 (2)
<i>Todirostrum cinereum</i> Common Tody- Flycatcher	Inga sp. (Fabaceae)	Sooretama, ES (19°03'S; 40°09'W, 85 m)	27 December 1992 (2)
<i>Phyllomyias fasciatus</i> Planalto Tyrannulet	<i>Vochysia</i> sp. (Vochysiaceae)	Rio do Cipó, MG (18°40'; 43°59', 570 m)	20 December 1997 (2)
	unidentified bush	Campos do Jordão, SP (22°38'S; 45°26'W, 1600 m)	11 August 2001 (2)
<i>Phyllomyias griseocapilla</i> Gray-capped Tyrannulet	unidentified flowers	Augusto Ruschi Biological Reserve, ES (19°54'S; 40°33'W, 850 m)	September 2002 (2)
<i>Culicivora caudacuta</i> Sharp-tailed Tyrant	tiny flowers of bushes	Broa prairie, SP (22°14'S; 47°52'W, 715 m)	14 June 2002 (2)
<i>Myiarchus swainsoni</i> Swainson's Flycatcher	unidentified yellow flowers	Intervales, SP (24°16'S; 48°25'W, 830 m)	04 March 1988 (2)
<i>Myiarchus ferox</i> Short-crested Flycatcher	<i>Croton floribundus</i> (Euphorbiaceae)	Fazenda São José, SP (22°21'S; 47°29'W, 650 m)	12 January 1992 (2)
<i>Myiophobus fasciatus</i> Bran-colored Flycatcher	<i>Inga</i> sp. (Fabaceae)	Fazenda São José, SP	09 September 2001 (2)
<i>Muscipipra vetula</i> Shear- tailed Gray Tyrant	<i>Tecoma stans</i> (Bignoniaceae)	Monte Alverne, RS (29°34'S; 52°22'W, 350 m) SP – São Paulo, RS – Rio Grande do Sul	23 September 1995 (1)

*Acronyms of states: ES – Espírito Santo, MG – Minas Gerais, SP – São Paulo, RS – Rio Grande do Sul.

**Observers: 1: G.A. Bencke, 2: E.O. Willis.

Highland Elaenia *Elaenia obscura* also briefly fed on the insects on the first day. Beetles were picked or snapped off flower heads and outermost peduncles mostly with glean, reach and leap maneuvers, and the birds seemed to make little effort to catch them, since prey was everywhere and almost stationary.

Other species

On 27 April 1996, a tanager flock visited Senna trees by the lab of the Museu de Biologia Mello Leitão (19°58'S; 40°36'W, 680 m a.s.l.), Santa Teresa, sallying to air or leaves for insects near or far from flowers (Tangara palmarum, T. ornata, T. sayaca, and T. cayana). A female Barred Antshrike Thamnophilus doliatus pecked an insect from a Bauhinia flower (Fabaceae) at Barão Geraldo District (22°50'S; 47°05'W, 620 m a.s.l.), Campinas, São Paulo state, on 06 June 1998 (E.O.W.). On 15 October 1995, a male Hepatic Tanager Piranga flava worked the crown of a flowering Phytolacca dioica tree (Phytolaccaceae) for about 25 min at Itati (29°23'S; 50°11'W, 230 m a.s.l.), Rio Grande do Sul state, hopping and jumping along branches to catch insects on or near flowers with short sallies, flutter-chases and one reach-out; the bird often watched closely the movements of flying insects around flowers before attempting to capture its prey (G.A.B.).

DISCUSSION

The temporary association of insect-eating birds with flowering trees or shrubs is scarcely documented in the literature. We expect this opportunistic behavior to be more common among small-bodied, edge or canopydwelling birds that sally or hover-glean to catch small insects on or near foliage, because *i*) insects attracted to flowers are usually small (mostly hymenopterans, flies and beetles, but also butterflies and moths; Willmer 2011), making them non-rewarding prey for larger birds; *ii*) plant species showing massive flowering (*i.e.*, producing large numbers of exposed flowers to attract relatively nonspecialized pollinators) predominate in the upper strata of tropical forests and along borders (Janzen 1975, Baker *et al.* 1983); and *iii*) flowers are rarely accessible directly to perching birds in the New World (Cronk & Ojeda 2008).

In southeastern Brazil, *P. fasciatus* (10.3 g) often watches flowers and sallies for or pecks insects, *T. poliocephalum* (7 g) and *M. fasciatus* (9.9 g) less. As illustrated here, many other birds (mass range 6.3-102 g) visit flowering trees to get insects, but they move through the trees and do not obviously watch flowers as do the tyrannulets above, or perhaps do it for shorter periods (*e.g., Piranga flava*). Wholly insectivorous birds such as *T. poliocephalum* may forage in flowering trees for longer periods as compared to similarly sized species that also feed on fruits, presumably because they are able to find most of their food items in the flowers and/or foliage, and also because arthropods at flowers are a rapidly renewing resource (Beehler 1980). In contrast, *P. fasciatus* and several tanagers make more frequent but shorter visits to flowering trees, possibly because insects are only part of their diet. Individuals of *P. fasciatus* observed at Santa Lúcia often regurgitated mistletoe seeds upon arriving at the flowering trees, indicating they had been feeding on fruit shortly before.

Several other Tyrannoidea visit eucalyptus or other flowers, but more study is needed to establish whether arthropods, nectar, or pollen are used. We did not confirm any activity that could have pollinated flowers, except when one bird put its head in a flower. Flight near flowers could have picked up some pollen, but tanagers and others that visit flowers directly are more likely to pollinate.

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Nesting information for Tropeiro Seedeater (Sporophila beltoni), an endemic songbird from southern Brazil

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ABSTRACT: We present the first nesting information for the Tropeiro Seedeater (*Sporophila beltoni*), a newly described and poorly known Neotropical passerine endemic to southern Brazil. We observed a novel male biased behavior for nest site selection in the Tropeiro Seedeater based on seven events of pre-nesting display courtship. We describe the nest, eggs, nestlings, and fledglings based on 133 nests found over four breeding seasons (2007 to 2011). The nest is a cup-shaped structure made with dry grass inflorescences and spider webs. It is placed in low, forked branches of substrate shrubs and contains multiple attachment points. The eggs are typically white with dark spots or stripes, and are pyriform in shape. Average egg dimensions are 18.2 × 13.2 mm. Nestlings fledge after 10 days. We provide the key information for distinguishing the nest, eggs, nestlings and fledglings of *S. beltoni* from other sympatric *Sporophila* species.

KEY-WORDS: austral migrant, breeding biology, fledgling description, nest site selection, nestlings.

INTRODUCTION

The Tropeiro Seedeater, *Sporophila beltoni*, is the rarest and perhaps the most threatened of the 35 species of Neotropical seedeaters. It breeds in a limited zone from upland shrub-grasslands of southern Brazil and migrates north to its wintering grounds in the Cerrado Biome. This species was recently split from the Plumbeous Seedeater (*Sporophila plumbea*) which occurs in Cerrado savanna (Repenning & Fontana 2013). Its distinguishing features are the bluish-gray plumage and bright-yellow bill in males. The Tropeiro Seedeater has a restricted and fragmented distribution, and has experienced population declines due to habitat loss and pressure from bird trappers (Repenning *et al.* 2010). The species is classified as "Vulnerable" globally (BirdLife International 2017).

Information about nesting biology of *Sporophila* species has been increasing, now including details about several species such as *S. caerulescens* (Francisco 2006), *S. hypoxantha* (Facchinetti *et al.* 2008, Bichinski 2011, Franz & Fontana 2013), *S. cf. hypochroma* (Roda & López-Lanús 2008), *S. collaris* (Facchinetti *et al.* 2008), *S. leucoptera* (Francisco 2009), *S. melanogaster* (Rovedder & Fontana 2012, Fontana & Repenning 2014) and *S. lineola* (Oliveira *et al.* 2010). These information may

provide the basis for comparative analyses of nesting behavior and provide the necessary background to assess the evolutionary relationships within the *Sporophila* genus.

Basic information such as description of nests, eggs and nestlings are prevalent in studies about reproductive biology of the group. The behavior involving parental care and, to a lesser extent, estimates of survival and reproductive success have also been determined (Facchinetti *et al.* 2008, Franz & Fontana 2013, Repenning & Fontana 2016). However, one poorly known aspect concerning the *Sporophila* genus is its premating behavior and the role of males and females in the selection of the nest site (Rising *et al.* 2012). Here, we studied the breeding biology of the Tropeiro Seedeater for 4 years and present the first description of its pre-nesting behavior, nest, eggs and nestlings/fledglings.

METHODS

We carried out the study in the southern Brazilian uplandgrasslands of south Santa Catarina and northeastern Rio Grande do Sul states (28°26'S; 50°24'W), during 4 breeding seasons (from October to March, 2007– 2011). The study areas comprise valley shrub-grasslands on very steep terrain, at elevations from 700 to 950 m a.s.l. (Repenning *et al.* 2010). We followed adult birds as soon as they arrived in the breeding areas beginning in late October. We then searched for nests by observing breeding evidences of paired birds. We evaluated nest building by focal observation with a telescope (20×60 mm), or made video recordings. We video-recorded 14 nests during nest building (34 h). We measured the nests and support plants with an analog caliper (0.01 mm) and a ruler (0.5 mm). We collected the plants used as nest support and as nest materials for identification of species. Scientific botanical names follow The Plant List (2013).

We weighed the eggs when they were reasonably freshly-laid (*i.e.*, 1–3 days from lay). We weighed nestlings (n = 74) daily with a digital balance (0.01 g), and measured eggs with caliper (0.01 mm). We determined the relative mass of eggs by dividing mean egg mass by mean female body mass. For monitoring nestlings/fledglings we banded them using aluminum bands provided by the Brazilian Banding Agency (CEMAVE/ICMBio) and with a combination of color bands. We used the Smith's (1975) color catalog to standardize the description of the color of eggs and nestlings.

RESULTS

Selection of the nest site

We observed males performing a secretive and peculiar display courtship during nest site choice by females. Nest sites were located in a particular habitat type, in general a patch of shrubs among rocks in dry soils in steep places. In seven independent cases observed, the males performed a typical short flight within a bush, calling very specific song notes and exposing the white wing speculum (*i.e.*, vibrating wings like an on/off signal) while they were followed by the female. Each male visited suitable forks in the shrubs making movements similar to those executed by females when shaping the chamber of the nest. Soon after that the female follows the male and check each bush, testing the places indicated by male with a brooding posture. In all the times observed the females allocated their nests in the fork of bush visited/indicated by male and tested previously. Each event observed included multiple male suggestions of potential nesting places within the breeding territory, however the nest attempts were always in the same patches of bushes each annual cycle. We recorded about 30 males that returned to the same breeding territory that changed their nesting sites by only a few meters in successive years, most of them within the same vegetation patch. Nesting attempts were usually with different females across breeding seasons. Six reproductive males observed performing nest site

display were males with final plumage and only one had a formative drab plumage (first year) male.

Nest building and nest materials

The nest is a low cup (sensu Simon & Pacheco 2005), located in a fork of a bush (sensu Simon & Pacheco 2005), made with dry grass inflorescences and attached with spider webs to the substrate through multiple points (sensu Hansell 2000). Construction of the nest takes 5 days on average (range 4–10 days, n = 15 nests) and is only performed by the female (Fig. 1). We identified 16 species of shrubs used for nesting in the following families: Asteraceae (12), Myrtaceae (2), Escalloniaceae (1) and Anacardiaceae (1) (Fig. 1). Fibers used to build nests were inflorescences from the genera Eragrostis and Panicum. Eragrostis airoides and E. polytricha were also present in all nest linings of the incubation chambers of nests. Inflorescences such as *Calamagrostis viridiflavescens*, Briza spp., Paspalum spp., Eustachys uliginosa, and lichen (Usnea sulcata) appeared in low frequency in the composition of the nests, and were used in the outer edge of some nests only. Fibers were tied up with spider webs. We recorded three events of females collecting webs from ground funnel webs of the spider Aglaoctenus lagotis (Lycosidae) (Fig. 1). We summarize the morphometric parameters of the nests in Table 1.

Eggs and nestlings

The eggs typically have a whitish background, ranging from pale cream tones (Pale Cream Horn) to shades of turquoise (Turquoise Green; Smith 1975). The spots or strips are grayish, black, beige, brown or burgundy, and may be of an intense shade or pale. The shape of the eggs ranged from pyriform (predominant) to oval (Fig. 1). The mean morphometric parameters of eggs were: length = 18.2 ± 0.14 mm (range 17.2-19.3 mm, n = 26); width = 13.2 ± 0.08 mm (range 12.7-14.1 mm, n = 26 eggs) and mass 1.46 g ± 0.03 g (range 1.29-1.64 g, n = 16 eggs). The relative mass of eggs was on average 12.2% of the female body mass.

Newly hatched nestlings had a body mass average of 1.29 g and when they left the nest they had a body mass average of 9.03 g (Fig. 2). In the first two days the nestlings were quiet and have thin, transparent, pinkish skin with tufts of grayish-white down sparsely covering the head, back, wings and flanks. The bill tip is yellowish with yellowish commissure, and a pinkish-orange lining of the mouth. At day 2 the pins of remiges become visible. Nestlings' eyes open on day 4, when the remiges (pin feathers) and contour feathers start to emerge. The growth of body neossoptiles occurs in the following order: coronal/occipital, dorsal, pelvic and femoral. The feathers sheaths disintegrate on days 6 to 8, ending the pin feather

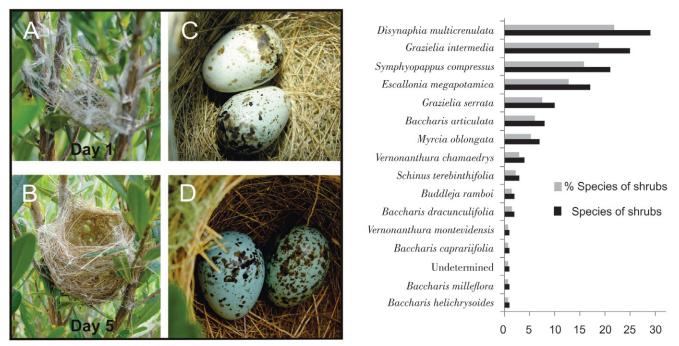


Figure 1. Nest of Tropeiro Seedeater *Sporophila beltoni* in the initial (**A**) and final phases (**B**) of construction (days 1 and 5, respectively). Eggs showing variation in background color, white/pale (**C**) and turquoise (**D**) and spotting patterns. On the right is a list of species of shrubs that were used as substrates for nests of the Tropeiro Seedeater (n = 133 nests).

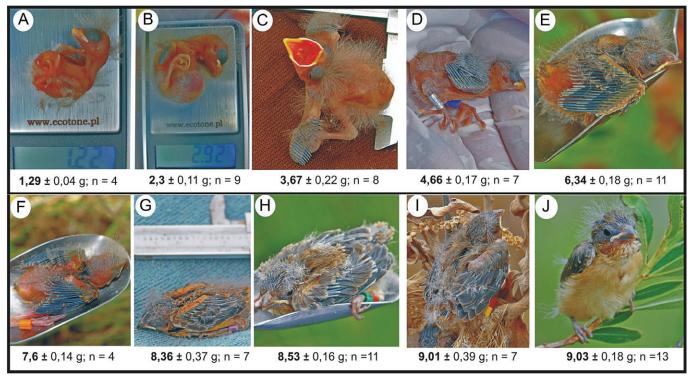


Figure 2. Daily growth (mean ± SE body mass) of nestlings and fledglings of the Tropeiro Seedeater, Sporophila beltoni.

stage. Around day 10 the nestlings' plumages are poorly complete and with short tails when they leave the nests (Fig. 2). Between 10 and 13 days fledglings have limited flight. The general color of covert feathers and remiges are a drab brown and the culmen is Sepia/Brown. Feeding independence of fledglings occurs around day 50 when they exhibit the complete juvenile plumage or starting the pre-formative molting.

DISCUSSION

Selection of the nest site

We revealed a novel behavior that involves the selection of nest sites in the Tropeiro Seedeater. The repeated pre-nesting male behavior suggests a strong male bias in indicating the site for nest-building to the female,

although the male abstains completely from the nest building phase. This breeding behavior has not been described for any other species of Neotropical seedeater, or for other passerines in the region. In monogamous species the selection of the nest site often results from a subtle interplay between the two sexes where male and female inspect potential nest sites together (Collias & Collias 1984). Analogous behavior has been documented for Australian grass-finches (Estrildidae), in which the male leads his mate and indicates the potential nest places. After finding a suitable fork of a branch he attracts the female's attention to it by hopping back and forth, uttering special calls. However, the final choice is then made by the female (Immelmann 1965). Far beyond philopatry the results presented here shows fidelity of specific places for nesting within the same territories each season. Oldest males were tenacious to same breeding territories which contributes for stability on nest site year after year. Hence, we emphasize that nesting success from previous breeding season(s) may be the ultimate factor shaping this rare pre-nesting behavior (Payne 1979, Repenning & Fontana 2016).

Nest, eggs and nestlings

The nest of S. beltoni is similar to that of another syntopic Sporophila species. However, our results provide sufficient details for reliable diagnosis when compared to congeners' nests. In contrast to nests of S. caerulescens (Francisco 2006), the nest of S. beltoni is slightly larger (deeper, with inner diameter largest and thicker edge). Also, S. beltoni do not nest in trees and do not use herbal root fibers in the incubation chamber, which are common features for S. caerulescens nests. Sporophila beltoni nests have greater inner diameter and depth, on average, compared with nests of other Capuchino species (e.g., S. melanogaster, S. hypoxantha and S. pileata) and nests are always placed at greater heights. The adhesion of the nest to the substrate in S. beltoni differ from other Capuchino species which can be classified as bottom multiple vertical (sensu Simon & Pacheco 2005). Additionally, the wall of S. beltoni nests is thinner and less dense than that of nests of sympatric Capuchinos' species. Sporophila beltoni do not nest on clumps of grasses while Capuchinos nesting on clumps of grasses or on mixed clumps and sub-bushes has been described (Bichinski 2011, Rovedder & Fontana 2012, Franz & Fontana 2013).

The color pattern of the eggs of *S. beltoni* varies, and they appears almost indistinguishable from eggs of other *Sporophila* spp. based on color and shape. However, the species has some of the largest eggs among the *Sporophila* for which dimensional size data are available. They are, on average, smaller only than eggs of *S. leucoptera* and are equivalent to the size of those of the allopatric *S.* *collaris* (disregarding *Oryzoborus*). The relative mass of the eggs is within the range of the values reported for other congeners. However, the relative mass of the eggs is substantially greater than has been determined for other temperate-zone passerines (4.5 to 7.7% of the female body mass) (Welty & Baptista 1988).

Juveniles of Sporophila can be cryptic in areas where two or more sympatric species occurs. Sporophila beltoni juveniles are similar to those of S. hypoxantha, S. melanogaster and S. pileata, but have larger body size (length and mass) and lack the tawny/buff colored plumage usually present in Capuchino juveniles. When the first pre-formative molt starts in S. beltoni they then show no white wing speculum, which is present in Capuchinos. The nestling/fledgling of S. caerulescens, might overlap in measurements and plumage color with S. beltoni nestling/ fledgling. However, while they complete the pre-juvenal molt S. beltoni retains the brown plumage while S. *caerulescens* acquires an olive drab plumage (M.R. unpub. data). Finally, we suggest that the description of Tropeiro Seedeater associated to illustration in details of the nestlings until they fledge can provide useful parameters for estimating the nestling's age in other breeding biology studies of Neotropical seedeaters.

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Nest and nestling description of *Automolus rufipileatus* from Brazil

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ABSTRACT: We describe here the first documented nest and nestling of the Chestnut-crowned Foliage-gleaner (*Automolus rufipileatus*) in Brazil. They were recorded in the municipality of Vitória do Xingu, state of Pará, at Amazonian domains. As other members of the genus *Automolus*, the nest of *A. rufipileatus* was in a steep clay bank on the edge of a small stream, comprising an inclined entrance tunnel to access two distinct chambers. The nest can be classified as a cavity with inclined tunnel. When discovered, the nest housed a nestling in advanced stage of development; therefore, it was not possible to describe the eggs, incubation period and nestling's initial development. We encourage additional studies on Chestnut-crowned Foliage-gleaner reproductive biology.

KEY-WORDS: Amazonian Forest, Furnariidae, life history, reproductive biology, Xingu River.

Nowadays, there are 35 genera and 105 species of Furnariidae birds recognized in Brazil (Piacentini *et al.* 2015). The family presents a great diversity of architectural nests with more than 20 distinct forms of construction, ranging from tangles of sticks, as in genera *Synallaxis* and *Phacellodomus*, to constructions in steep banks, as in the genus *Automolus* (Zyskowski & Prum 1999). Nine species of *Automolus* are known to Brazil (Piacentini *et al.* 2015), *A. rufipileatus, A. melanopezus, A. cervicalis, A. subulatus, A. ochrholaemus, A. infuscatus, A. paraensis, A. lammi* and *A. leucophthalmus.* However, there is a lack of information about the behavior, territory, and, specially, reproductive biology of these species, except for *A. ochrolaemus* (Van Tyne 1926, Skutch 1952), *A. leucophthalmus* (Marini *et al.* 2007) and *A. paraensis* (del Hoyo *et al.* 2018).

Based on previous studies, the nests found for all species of *Automolus* genus exhibit the same pattern: they are cavities in the soil, usually in steep banks bordering small streams (Van Tyne 1926, Marini *et al.* 2007). Because of this reproductive behavior, *Automolus* species are commonly known as "*barranqueiros*" in Brazil, which means, in a free translation, those who use steep banks for nesting. Specifically, Londoño (2014) reported photographs of *A. rufipileatus* eggs and nests. However, Londoño (2014) did not describe the dimensions of the nest and eggs, or the environment surrounding the nest and, probably for this reason, Remsen-Jr. (2018)

considered that this species' nest was not described. From Londoño (2014) it is possible to check (through the investigation of photographs) that the nest follows the same pattern described in the present study, occurring in burrows in soil, and the eggs are white and ovoid.

Automolus rufipileatus is a resident species and it is distributed across South America, including Bolivia, Colombia, Ecuador, Peru, Guyana, French Guiana, Suriname, Venezuela, and Brazil (Birdlife International 2017, Remsen-Jr. 2018). Records in Brazil are exclusive to the Amazonian domain, where two subspecies are recognized: *A. r. consobrinus* and *A. r. rufipileatus*. The latter occurs south of the Amazon River (Remsen-Jr. 2018) and in the Volta Grande do Xingu, Pará state.

Life history of the species is virtually unknown, with little information regarding its habitat and food. According to Remsen-Jr. (2018), the species is insectivorous and forages by turning over dried leaves and litter; they can be seen in pairs and eventually in mixed flocks. They inhabit wetlands, alluvial forests and *terra firme* environments with presence of bamboo (Sick 1997, Remsen-Jr. 2018). This work aims to present information concerning the reproductive biology of *A. rufipileatus*, through description of a nest and a nestling observed in the municipality of Vitória do Xingu (Volta Grande do Xingu), state of Pará.

On 17 February 2015, during the rainy season, a nest

of Chestnut-crowned Foliage-gleaner was found 80 cm above the ground inside the soil walls of a steep bank of a small stream "*igarape*" in *terra firme* forest (Fig. 1A), with Açai Palm Trees (*Euterpe oleracea*) bordering the stream (3°19'54''S; 52°16'29''W, 213 m a.s.l). The nest was housing a single nestling, which was in a pinfeather stage. The nest was composed of an inclined-up access tunnel, with dimensions 7 cm height, 6 cm width and 20 cm depth, measured with a ruler (Fig. 1B & C), conducting to two chambers in a line sequence. The first chamber was wider and flattened $(9 \times 15 \times 10 \text{ cm} \text{ approximately} - \text{chamber 1})$, and the second chamber was smaller and basket-shape $(6 \times 9 \times 8 \text{ cm} \text{ approximately} - \text{chamber 2};$ Fig. 1C). Because of the difficulty in measuring the cavities due to the format and dimensions of the access tunnel, the measures of the chambers were estimated. Both chambers were lined with rootlets, but chamber 2, which was housing the nestling, had a thicker layer suggesting it was the place where eggs were incubated. We found fecal bags excreted by the chick in chamber 1 (Fig. 2A) and it

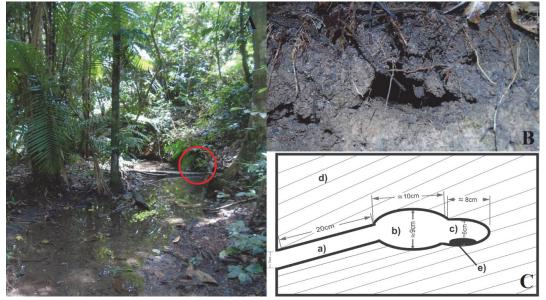


Figure 1. *Terra firme* forest where the nest of *Automolus rufipileatus* was found (**A**). Entrance of the access tunnel of *A. rufipileatus* nest (**B**). Sketch of *A. rufipileatus* nest, in a longitudinal cut (**C**), showing **a**) access tunnel; **b**) chamber one; **c**) chamber two; **d**) soil texture surrounding the nest; and **e**) rootlets thicker layer in basket-shape inside chamber two.



Figure 2. The interior of an *Automolus rufipileatus* nest: in the foreground the chamber one with nest garbage (**A**). Nestling inside the chamber two in the first visit (**B**). Nestling in the third visit (**C**).

is plausible to assume that these fecal bags were moved from chamber 2 to chamber 1 by adults. Unfortunately, the fecal bag removal event was not observed over 10 h of observations. However, three fecal bags were removed between our first (17 February) and second (18 February) visits to the nest.

The single nestling found in the nest during the first visit presented few down feathers and reddish-brown emerged pinfeathers of developing wings and tail feathers (Fig. 2B). In the third visit (24 February), we observed that the nestling had already developed the uniform reddish-brown plumage, coating practically the whole body (Fig. 2C), and it was ready to leave the nest. During this visit, we collected biometrical data (wing = 6.1 cm, tail = 4.2 cm, tarsus = 2.2 cm, beak length = 1.7 cm, total length = 14.2 cm) and banded the nestling, which had a darkish rufescent-brown plumage. In the fourth visit (27 February), the nestling was not found inside the nest and it was not observed again. In two visits, we saw an adult in the vicinity of the nest a few times, and on all occasions only one adult was seen at a time, always in a silent and discreet approach.

The nest of the Chestnut-crowned Foliage-gleaner follows the same pattern observed for other species of the genus, in which nests are built in steep bank, with cavityshaped tunnels, bordering water bodies (Van Tyne 1926, Skutch 1952, Marini *et al.* 2007). The classification of the nest of *A. rufipileatus*, based on Simon & Pacheco (2005), is cavity/with-tunnel/simple/platform, although the nest described here is not effectively contemplated by this classification because it presents two chambers after the access tunnel. Remsen-Jr. (2018) reported two males in breeding condition in February in Venezuela, the same period of reproduction confirmed in this study. Both studies registered the same reproductive period for the species.

Our study reports some new information on the life history of the Chestnut-crowned Foliage-gleaner and the first documented nest of the species in Brazil. We believe that the records presented here may help other ornithologists with the discovery of other nests of Chestnut-crowned Foliage-gleaner, in earlier stages of development (building the nest, posture and hatching eggs), therefore, leading to an increase in our knowledge on the natural history and reproductive biology of this species. Such information may be key for conservation actions, especially for Amazonian species, which are constantly threatened by increasing deforestation.

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Bird species that occupy river edge in continuous forest tend to be less sensitive to forest fragmentation

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ABSTRACT: Along a distance gradient from a given river, two types of habitat can be recognized: natural river edge and forest interior, each one with its own vegetation characteristics and dynamics. In a continuous area of the Brazilian Atlantic Forest, we investigated (1) if bird communities are different between a riverbank of a small stream and an inland forest habitat; (2) if the species of the river edge habitat are the ones that persist in the most in forest fragments after deforestation of a continuous forest; (3) if the river edge habitat species are those that are less sensitive to forest fragmentation. It is expected that there are differences in the bird communities and the occupancy of some species between the two habitats. We allocated 16 sampling points in each of the habitats and sampled the birds by point counts with a short radius of 30 m. Results suggest that there is a significant difference between the composition of the bird communities of the river edge and forest interior habitats, although the species richness is similar. Six species were more likely to occupy the river edge and 14 species had a greater probability of occupancy in the forest interior. Species associated with the river edge habitat (15 species) tend not to be sensitive to forest fragmentation (12 species). In this study, we demonstrated that river-border species of continuous forest areas form a significant part of the bird communities that persist in small forest fragments, with intense edge effect. This shows that not all forest edge species are the result of the colonization from open areas. Congruently, species that occupy the most distant areas from the river vegetation in a continuous forest are those more sensitive to forest fragmentation.

KEY-WORDS: Atlantic Rainforest, bird sensitivity, forest interior, natural edge, probability of occupancy.

INTRODUCTION

The Brazilian Atlantic Forest Biome, a tropical forest strip that stretches along 3300 km of the Brazilian coast beside inland areas in Argentina and Paraguay, has thousands of endemic species (more than 650 species of vertebrates and 8000 species of plants) and is considered one of the key biodiversity hotspots in the world (Tabarelli et al. 2010, Mittermeier et al. 2011). The seasonal-semideciduous forest (SF), a type of forest in the Atlantic Forest Biome, extends through the center-south of the country interior, between 200 and 800 m of altitude and could be considered an ecoregion; there, approximately 220 tree species occur, 10% of which are endemic to this forest type (Morellato & Haddad 2000, Oliveira-Filho & Fontes 2000, Scheer & Blum 2011, Anjos et al. 2018). Locally in SF, many rivers, large and small, and streams flow from upland areas to the lowland areas, as is common along the Atlantic Forest. A distinct riparian environment with a unique vegetative formation characterizes those lowland river edges, which is the focus of the present study. This lowland river edges forests usually has a much less dense canopy with few emerging trees while the under and midstory have a higher density of smaller

tree species; also, it is common to have the fall of trees and consequently the creation of clearings that allow the occupation of bamboo species (Bianchini *et al.* 2001, Anjos *et al.* 2007).

This lowland riparian environment constitutes a transition between the river and the associated upland forest, marking a natural border or ecotone boundary. A forest ecotone is a consequence of the meeting of distinct natural plant communities, which, in turn, influences the diversity of wild animals across the landscape, dependent on distance from a rivers' edge and the characteristic transition in topography, plant community, hydrological regimes, and soil types (Naiman et al. 1993, Shirley 2005). Considering several taxonomic groups, some studies suggest greater species richness in riparian environments compared to distinct forest (Naiman et al. 1993), others found greater richness in non-riparian environments (McGaragal & McComb 1992, Peres 1997) and some found no difference between these two habitat types (Gomez & Anthony 1998, Rykken et al. 2007).

On birds, several studies indicated the great importance of the riparian environment (natural river edges) as a uniquely sustaining habitat with relatively more species than associated upland forest areas (Woinarski

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et al. 2000, Kajtoch et al. 2007, Dominguez-López & Ortega-Álvarez 2014, Rannestad et al. 2015, Berduc et al. 2015, Sekercioglu et al. 2015, Gomez et al. 2016). In a tropical rainforest in Hong Kong, a higher number of individuals and bird species were recorded in the riparian environment, when compared to an area 100-250 m distant from the river due to the high availability of adult aquatic insects, which confirms the response of birds to river proximity (Chan et al. 2008). In a 656 ha fragment of the Atlantic Forest in northern Paraná state, with little topographic variation (~150 m altitude), Anjos et al. (2007) showed that 43% of all bird species were associated with riparian forest due to differences in vegetation; they sampled 81 species and found that 19 and 45 species were unique to the non-riparian and riparian habitat, respectively. Another study conducted in the same site showed that the difference between the bird communities is mainly due to the presence of bamboo in the riparian forest (Chusquea sp., Willrich et al. 2016). Therefore, riparian bird species may comprise a significant proportion of the overall local forest bird richness and with particular traits associated to that vegetation closer to rivers.

Riparian forest could be in some extension comparable to edges of forest fragments, since both are ecotones. We argue that edge birds of riparian forest should be more adapted to the edge of forest fragments, while birds inhabiting the interior of the forest avoid those created habitats after deforestation (Gimenes & Anjos 2003, Hansbauer et al. 2008). In this study, we tested if it is possible that birds that originally live in the riparian environment in the continuous forest could be more tolerant to edges that appear after forest fragmentation. To do this, the first objective of this study was to verify if bird communities are different between a natural river edge habitat and a forest interior. For this, we investigated the richness and composition of the bird communities. The hypothesis is that the richness and composition of the bird communities of the two habitats are different due to the difference of resources found in both habitats, e.g., the availability of adult aquatic insects in the river natural edge habitat and several resources (both animal and vegetal), associated to the river edge vegetation. The second objective of this study was to compare the occupancy of the bird species between the two habitats. In this case, we evaluated the occupancy probability of the bird species according to the different habitat types. The occupancy of some bird species is expected to be different between the two types of habitat, due to differences in vegetation. The third objective of this study was to verify if the river natural edge habitat species are less sensitive to forest fragmentation. In order to do this, we investigated the association between the number of bird species closely related with the river natural edge habitat and their sensitivity to forest fragmentation, which was previously

determined for the bird species of SF (see Anjos 2006, Anjos *et al.* 2011). The hypothesis is that the natural river edge habitat species are the ones that persist in most of forest fragments in relation to the forest interior, after the deforestation of a continuous forest, that is, those species are less sensitive to forest fragmentation. The reason for this hypothesis is that the vegetation of river edges presents low trees and bushy entanglement in the lower stratum, phyto-physiognomy that resembles edges of forest fragments.

METHODS

Study area

The study was developed in the Iguaçu National Park (INP), in the municipality of Céu Azul, Paraná ($25^{\circ}09'12''S$; $53^{\circ}50'42''W$, Fig. 1). INP was created in 1939 and its total area is 185,262.2 ha; it is a fully protected Conservation Unit whose predominant vegetation is SF (ICMBio 2014). In reality, INP is home to the country's largest continuous SF area. The INP climate, according to the classification of Köppen, is of type *Cfa* subtropical humid or mesothermic with hot summer, with average temperatures between 15 and 25 °C and rainfall above 900 mm, also distributed throughout the year (Melo *et al.* 2006).

SF is related, in virtually the whole area of occurrence, to a climate of two well defined seasons - one rainy and one dry (Veloso *et al.* 1991). The vegetation is dense and presents a great variety of vegetal species, constituted by arboreal elements (perennial or deciduous), as well as shrub, lianas and epiphytes. Among tree species that are associated with SF are: Assai Palm (*Euterpe edulis* Mart.), Peroba (*Aspidosperma polyneuron* Müll.Arg.), Brazilian Rosewood (*Aniba rosaeodora* Ducke), Alecrin (*Holocalyx balansae* Micheli), Angico-cedro (*Parapiptadenia rigida* (Benth.) Brenan) and Argentine Cedar (*Cedrela fissilis* Vell., Guimaráes *et al.* 2003).

Sampling areas

We sampled two habitats and denominated them as "River Edge" and "Forest Interior". The "River Edge" (RE; 25°09'43''S; 53°49'39''W) is located on the border of a tributary of the Azul River, and the "Forest Interior" (FI; 25°09'28''S; 53°50'09''W) is located at 300 m of RE, at higher altitude (565 m a.s.l.) and at 470 m of forest edge (Fig. 2 in Appendix I). The tributary of the Azul River is a small one, with 8 to 15 m width in the section studied. In each habitat, 16 points were established (Fig. 1). The shortest distance between RE and FI sampling points was 80 m.

In RE, points were allocated in four tracks (REA,

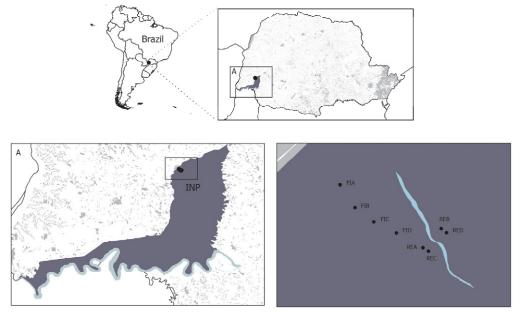


Figure 1. Location of Paraná state in South America and the region of the study in the Iguaçu National Park (INP), western Paraná, southern Brazil. The black dots indicate the location of the sample units (REA, REB, REC and RED in RE and FIA, FIB, FIC and FID in FI).

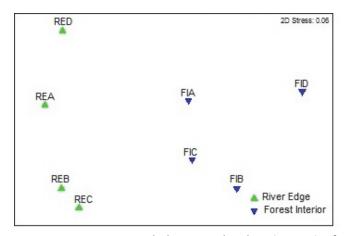


Figure 2. Non-metric multidimensional scaling (NMDS) of bird communities in eight different sample units (REA, REB, REC, RED, FIA, FIB, FIC, and FID) occurring in two habitats ("River Edge" and "Forest Interior") at INP.

REB, REC and RED), perpendicular to the river. The trails in this habitat were at least 300 m apart (Fig. 2 In Appendix I). In FI, the points were allocated on a trail of 1200 m located inside the forest. The 16 points located in this track were subdivided into four blocks of four points (blocks FIA, FIB, FIC, FID, Fig. 3 in Appendix I).

In RE habitat we observe lower trees, with a height of 8 to 15 m, having less plant species richness. Also, the species are adapted to periodic flooding which supports high humidity (ICMBio 1999). In the transition from RE habitat to IF habitat, vegetation changes and trees become taller. In the FI habitat there is a greater richness of plant species; there are large trees, with maximum heights of 35 m in the emerging layer, and it is common to find in the best-preserved parts, trunk with diameters at breast height (DBH) of more than 1 m (ICMBio 1999).

Bird sampling

We used the point count method with radius of 30 m (Bibby *et al.* 1993) in bird sampling. This method is very effective in studies of avian habitat relationships (Anjos *et al.* 2010). Sampling was carried out during the breeding season of January and February of 2013, in which birds are more likely to be detected by the observer through their vocalizations.

Each set of four points in both habitats was considered as a sample unit, which was sampled in one day. These sample units were named REA, REB, REC and RED in RE and FIA, FIB, FIC and FID in FI. On each day the points of each sample unit were sampled consecutively 1, 2, 3 and 4, and then again reversing the sequence, 4, 3, 2 and 1. For example, on track A in RE, on one day the sampling sequence of the points was REA1, REA2, REA3, REA4, REA4, REA3, REA2, REA1. The following day the sampling sequence of the points was reversed: REA4, REA3, REA2, REA1, REA1, REA2, REA3, REA4. We sampled each sampling unit for two days, and we sampled each sampling point four times (as if they had been sampled on four mornings). Studies performed in the Atlantic Forest using the point count method demonstrated that 3 to 5 days of sampling are sufficient to detect more than 90% of the species recorded in a sample area (Anjos 2007, Cavarzere et al. 2013). Sampling began shortly after sunrise, when daytime birds start to vocalize, and ended 2.5 h after sampling at the first point, under favorable climatic conditions. We sampled each point for 10 min with a 10 min interval between points. According to Anjos *et al.* (2010), if the observer is interested in assessing differences between numbers of bird species at different locations, the time of 10 min is sufficient and in SF, 96% of the species are recorded during this time interval.

Statistical analysis

We estimated the total richness of the bird species using the non-parametric $Chao_2$ species estimator (Herzog *et al.* 2002). We evaluated the influence of space (spatial autocorrelation) on the composition of bird species through the Mantel test (1000 permutations). The geographic distance matrix was obtained by the Euclidean distance on the geographical coordinates of the sample units. We obtained the similarity matrix of the species composition using the distance of Bray Curtis on the abundance of the species in each sample unit. We performed the analysis in R software (R Development Core Team 2015), using the package "vegan" (Legendre & Legendre 1998, Oksanen *et al.* 2016).

We estimated the relative abundance for a single species in a habitat (RE or FI), called the Index of Point Abundance (IPA), by dividing its contact number by the total number of points sampled in each site (Bibby *et al.* 1993). To avoid double counting for the same individuals' precautions were taken particularly for those highly mobile species by adopting a field form that is divided into different quadrants as suggested by Vielliard & Silva (1990). Thus, a number of contacts of 30 for a given species resulted in an IPA equal to 0.468 (30 contacts divided by 64 points sampled).

To verify differences in bird species composition between the two habitats we used a permutational analysis of variance (PERMANOVA) (Anderson 2001). This analysis was performed in R software (R Development Core Team 2015), using the packages "vegan" (Oksanen *et al.* 2016) and "BiodiversityR" (Kindt & Coe 2005).

We used non-metric multidimensional scaling (NMDS) to visualize the similarity in community composition between the two habitats (Clarke 1993). Data was transformed through weighting dispersion to reduce the contribution of high abundance species in the similarity. Similarities of Bray-Curtis were used to construct the distance matrices between sample units. This analysis was performed using software PRIMER v. 6 (Clarke & Gorley 2006).

For each bird species recorded in both habitats, we calculated the probability of occupancy in the different habitats using single-season occupancy modeling (MacKenzie *et al.* 2002). These models involve the estimation of two parameters: the occupancy (Y), which

is the probability of the species being present in one place, and the probability of detection (p).

Single season occupancy modeling requires multiple visits to sampling units during a season in which species can be detected. This model assumes that during these visits no individual enters or leaves the population (closed model). At each visit the observer detects the presence ("1") or absence ("0") of the species of interest. The absence may be a real absence of the species or a failure to detect the species. This type of modeling adjusts the variation in probability of detection while estimating the probability of occupancy of bird species. By incorporating the probability of detection into the models, the imperfect detection is considered and the bias in the parameter estimation is reduced (MacKenzie *et al.* 2006).

Since each point was sampled four times, in each sampling we recorded if each species of bird was detected or not. Thus, the detection history for each bird species in each area was obtained. The detection history was then used to estimate the probability of occupancy of the species.

For this model, the estimated parameters (occupancy and detectability) may be a function of covariates. The PRESENCE software (Hines 2006) recognizes two types of covariates, (1) site-specific covariates, which are constant for the site within the same season, e.g., habitat type, fragment size, or generalized weather patterns, such as drought or El Niño; and (2) sampling-occasion covariates, which may vary at each sampling, such as temperature, precipitation, time of day or observer (Hines 2006). In order to verify the probability of occupation of the bird species in each of the habitats, in the present study the first type of covariant was used. Thus, we tested whether the probability of occupancy of each species of bird occurred as a function of habitat. We ran occupancy models which assumed that the occupancy and detection of the species were constant - null models (e.g., same probability of occurrence among all points sampled), models that assumed that the probability of species occupancy was in function of the covariant habitat, models that assumed that species detection was a function of covariant habitat and models who assumed that the probability of species occupancy and species detection was in function of the covariant habitat.

We used the Akaike Information Criterion (AIC, Burnham & Anderson 2002) for small sample sizes (AICc), to select the most parsimonious model. The best models were those with lower AICc values and higher AICc weights; the closer to 1 the AICc weight value, the greater the likelihood of the model being chosen as the best (Burnham & Anderson 2002). This analysis was performed using PRESENCE 9.0 software (Hines 2006).

We used a contingency table to investigate the relationship between the number of birds associated

with RE and IF habitats with their sensitivity to forest fragmentation (sensitive and non-sensitive). The bird species' level of sensitivity used was presented in Anjos (2006) and Anjos et al. (2011). Anjos (2006) determined the sensitivity of the birds to forest fragmentation in SF based on samplings carried out in 14 forest fragments of different sizes and degrees of isolation. Species were considered sensitive if they occurred only in control fragments or in large, non-isolated fragments; species not sensitive to forest fragmentation were those that occurred in all fragments, including the smallest and most isolated ones (Anjos 2006). In the present study we used two criteria to determine if a given species of bird was associated with one of the habitats, RE or FI: 1) the species should be exclusive to one habitat and with at least three contacts during the total sampling period; or 2) the species should have a higher probability of occupancy in one of the habitats. The software Past 3.0 was used to calculate the contingency table (Hammer et al. 2001).

The taxonomy and nomenclature followed American Ornithologists' Union - South American Classification Committee Checklist for South American Birds (SACC; Remsen-Jr. *et al.* 2016).

RESULTS

We recorded a total of 80 species of birds in both habitats, similar to the estimated richness (Chao₁, 84 ± 4 species). We detected no autocorrelation between the geographical distances of the sample units and the species composition (Mantel r = 0.063, P = 0.320). We recorded 65 bird species in RE and 68 in FI. The number of species estimated by Chao₁ for RE was 70 ± 4 species and the estimated number for FI was 79 ± 6 species. Therefore, we found no difference between the estimated richness of the two habitats. Twelve species were recorded only in RE and 15 were exclusive to FI (Appendix II).

The composition of bird communities differed between habitats (PERMANOVA, Pseudo-F = 6.785, P < 0.010). In concordance with this result, the ordering of the NMDS showed that the RE and FI sample units differ in composition and abundance of bird species, as they were grouped separately (Fig. 2).

Among the 53 species that occurred both in RE and FI, the habitat type influenced the probability of occupancy of 20 species (Appendix III). Six species had a higher probability of occupancy in RE (*Melanerpes flavifrons*, *Xiphocolaptes albicollis*, *Capsiempis flaveola*, *Platyrinchus mystaceus*, *Sirystes sibilator*, *Saltator similis*) and 14 species showed a higher probability of occupation in FI (*Crypturellus obsoletus*, *Trogon rufus*, *Pteroglossus castanotis*, *Hypoedaleus guttatus*, *Dysithamnus mentalis*, *Conopophaga lineata*, *Grallaria varia*, *Chamaeza* campanisona, Dendrocolaptes platyrostris, Leptopogon amaurocephalus, Schiffornis virescens, Cyanocorax chrysops, Trichothraupis melanops, Basileuterus culicivorus).

Considering the set of species analyzed, 15 bird species were more associated to RE habitat, of which three are sensitive and 12 are not sensitive to forest fragmentation. Twenty-three bird species were more associated to FI habitat, of which 14 are sensitive and 9 are not sensitive to forest fragmentation (Appendix IV). Thus, the data suggest that species of birds susceptible to habitat fragmentation were those associated with the FI habitat while those that are not sensitive to fragmentation were those associated with RE habitat ($\chi^2 = 6.13$; P = 0.013).

DISCUSSION

We found a significant difference between the composition of the bird communities of RE and FI habitats, although the species richness was similar. The difference in the composition was due to several exclusive species in each habitat and to several species that occurred in both habitats but which showed greater occupancy in only one habitat. Species associated with the RE habitat tend not to be sensitive to forest fragmentation. In the study by Anjos (2006) on the sensitivity of birds to forest fragmentation, species that present tolerance to edges showed low sensitivity to fragmentation. The results of the present study indicate that of the total species associated to the RE habitat, only 20% are sensitive to forest fragmentation, while 61% of the species associated to the FI habitat are sensitive to forest fragmentation (Appendix IV).

A large number of physical and biological processes occur from the edge of a fragment because of the influence of the matrix habitat (Laurance et al. 2011). This influence on physical and biological processes occurs up to 200-500 m from the border into the fragment (Laurance et al. 2011). Therefore, small and/or very elongated fragments are "all edge", that is, without an interior free of edge effects. These processes can affect forest bird species. Species associated with FI, such as Micrastur semitorquatus, Automolus leucophtalmus, Grallaria varia and Schiffornis virescens do not occur in fragments smaller than 60 ha; on the other hand, of the 15 species associated with RE, about 67% persist in small forest fragments of 11 and 25 ha (Anjos 2001). We should highlight that non-forest colonizer bird species occur in the edge of forest fragments. Those are species from open and/or Cerrado areas, such as Rupornis magnirostris, Colaptes melanochloros, Melanerpes candidus, Patagioenas maculosa and Myiarchus swainsoni (Anjos 2001, Baptista et al. 2016, Bierregaard et al. 2016, Joseph 2016, Winkler et al. 2016). Thus, the composition

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of birds of the edge of a forest fragment should originate mainly from those living on river banks in continuous forest combined with those colonizers from open areas. However, some species of interior forest can also persist in the fragments. Anjos (2001) studied the bird community in small forest fragments (56, 25 and 11 ha in size). Based on the present study and in Anjos *et al.* (2007), we found that the majority of the bird species that live in these small fragments are species that inhabit river bank and/or are colonizing species: 80% in FA, 82% in FB and 84% in FC.

River's edge birds are less sensitive to forest fragmentation

Lindsey et al.

Fragmentation and habitat degradation cause changes in the forest edge, such as increased temperature and light intensity. In Neotropical forests, birds that live on riverbanks in a continuous forest and occupy the edge of the remaining habitat after fragmentation and birds from open areas should select similar abiotic conditions such as air temperature, spatial variation of solar radiation, humidity and wind speed. On the other hand, forest species, such as understory insectivorous birds, select microhabitats with different abiotic characteristics and do not occupy the edge of the small forest fragments or fragments considered "all border" (Pollock et al. 2015, Stratford & Stouffer 2015). In fact, birds associated with darker microhabitats are more sensitive to forest edge than birds that use brighter microhabitats (Patten & Smith-Patten 2012). However, the forest interior is also home to several sensitive species to forest fragmentation, which are not particularly associated to forest understory, such as Pionopsitta pileata, one of the most threatened species of psittacines due to the massive destruction of their habitat (Sigrist 2013), which occurred exclusively in the FI habitat. It is important to point out that in RE there were also species that were exclusive of that habitat and are sensitive to forest fragmentation, such as Coccyzus melacoryphus, Hylopezus nattereri and Tityra cayana. The reason of these species' sensitivity may be related to the vegetation structure, or even the lower humidity of the edges of a fragment compared to the river's edge (Pollock et al. 2015).

When comparing the results of the present study with those obtained in the interior of São Paulo state by Cândido-Jr. (2000), who compared the avifauna between the edge of the fragment and the forest interior, similarities are found. Three species were most associated with the forest interior in both studies: *A. leucophtalmus*, *D. platyrostris* and *T. melanops*. However, one species, *Tachyphonus coronatus*, presented different results: in the present study it was associated to the RE habitat and in the cited study it was associated to the forest interior. This species inhabits the edge of the forest, *capoeiras*, parks, gardens and adapts well to a variety of edges, disturbed habitats and secondary forests (Sick 1997, Hilty 2016); more than 60% of the diet of this species is composed by invertebrates (Wilman *et al.* 2014). Populations at the edge of their geographical distribution are generally smaller than those closest to the center of the geographical distribution (Holt *et al.* 2005). The INP is situated on the southern edge of the geographical distribution of *T. coronatus.* It is possible that the population of *T. coronatus* in the INP is smaller than the population of the species at the site studied by Cândido-Jr. (2000). Perhaps in INP the individuals of this species were concentrated in the habitat RE due to greater availability of some type of resource or even by competition and the presence of predators.

In this study we demonstrated that river-edge bird species of a continuous area of forest form a significant part of the bird communities that persist in small forest fragments, with intense edge effect. This shows that not all the forest edge species are the result of colonization from open habitats. As expected, species that occupy the most remote areas of the river vegetation in a continuous forest are those most sensitive to forest fragmentation.

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APPENDIX I

Figure 1. Sites sampled in the INP: A) "River Edge", on the edge of a tributary of the Azul River, and B) "Forest Interior".

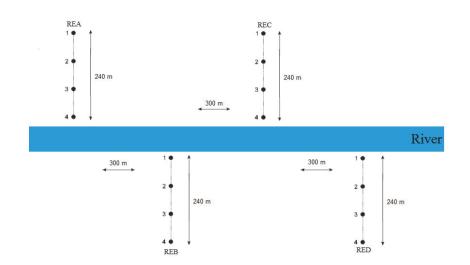


Figure 2. The four points of each of the trails (REA, REB, REC and RED) in the River Edge habitat sampled at INP.

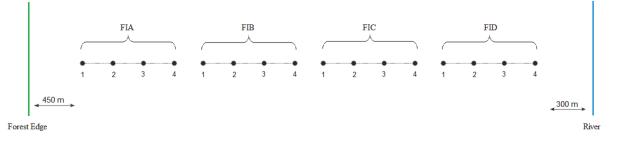


Figure 3. The four blocks (FIA, FIB, FIC and FID) of four points located on the trail of FI habitat at INP.

APPENDIX II

Families and bird species sampled in RE and FI habitats at INP. Taxonomy follows American Ornithologists' Union - South American Classification Committee Checklist for South American Birds (Remsen-Jr. *et al.* 2016).

	Presence				
Bird species	RE	FI			
TINAMIDAE					
Crypturellus obsoletus	Х	Х			
Crypturellus parvirostris		Х			
Crypturellus tataupa	Х	Х			
COLUMBIDAE					
Patagioenas picazuro	Х	Х			
Geotrygon montana	Х				
Leptotila verreauxi	Х				
CUCULIDAE					
Piaya cayana	Х	Х			
Coccyzus melacoryphus	Х				
TROCHILIDAE					
Phaethornis pretrei		Х			

Bird species	Presence				
	RE	FI			
TROGONIDAE					
Trogon surrucura	Х	Х			
Trogon rufus	Х	Х			
MOMOTIDAE					
Baryphthengus ruficapillus	Х	Х			
RAMPHASTIDAE					
Ramphastos dicolorus	Х	Х			
Selenidera maculirostris	Х	Х			
Pteroglossus castanotis	Х	Х			
PICIDAE					
Picumnus temminckii		Х			
Melanerpes flavifrons	Х	Х			
Colaptes melanochloros	Х				
Dryocopus lineatus	Х	Х			
Campephilus robustus	Х	Х			
FALCONIDAE					
Micrastur semitorquatus		Х			
Milvago chimachima		Х			
PSITTACIDAE					
Pionopsitta pileata		Х			
Pionus maximiliani	Х	Х			
Pyrrhura frontalis	Х	Х			
Psittacara leucophthalmus	Х	Х			
THAMNOPHILIDAE					
Hypoedaleus guttatus	Х	Х			
Mackenziaena severa	Х	Х			
Thamnophilus caerulescens	Х	Х			
Dysithamnus mentalis	Х	Х			
Herpsilochmus rufimarginatus	Х	Х			
Drymophila rubricollis		Х			
Drymophila malura	Х	Х			
Pyriglena leucoptera	X	X			
CONOPOPHAGIDAE					
Conopophaga lineata	Х	Х			
GRALLARIIDAE					
Grallaria varia	Х	Х			
Hylopezus nattereri	X	~~			
RHINOCRYPTIDAE					
Eleoscytalopus indigoticus	Х	Х			
FORMICARIIDAE		~~			
Chamaeza campanisona	Х	Х			
Chamaeza meruloides	X	X			

Bird species	Presence				
bhu species	RE	FI			
FURNARIIDAE					
Sittasomus griseicapillus	Х	Х			
Dendrocincla fuliginosa	Х	Х			
Dendrocolaptes platyrostris	Х	Х			
Xiphocolaptes albicollis	Х	Х			
Xiphorhynchus fuscus		Х			
Lochmias nematura	Х				
Anabacerthia lichtensteini	Х	Х			
Automolus leucophthalmus		Х			
Synallaxis ruficapilla	Х	Х			
TYRANNIDAE					
Myiopagis caniceps	Х	Х			
Camptostoma obsoletum	Х	Х			
Capsiempis flaveola	Х	Х			
Leptopogon amaurocephalus	Х	Х			
Hemitriccus diops	Х				
Poecilotriccus plumbeiceps	Х	Х			
Tolmomyias sulphurescens	Х				
Platyrinchus mystaceus	Х	Х			
Lathrotriccus euleri	Х	Х			
Pitangus sulphuratus		Х			
Myiodynastes maculatus	Х				
Megarynchus pitangua	X	Х			
Sirystes sibilator	X	Х			
TITYRIDAE					
Tityra cayana	Х				
Schiffornis virescens	X	Х			
INCERTAE SEDIS					
Piprites chloris		Х			
CORVIDAE					
Cyanocorax chrysops	Х	Х			
TURDIDAE	25				
Turdus leucomelas	Х	Х			
THRAUPIDAE	25				
Cissopis leverianus		Х			
Trichothraupis melanops	Х	X			
Tachyphonus coronatus	X	21			
Dacnis cayana	45	Х			
Hemithraupis guira	Х	23			
Conirostrum speciosum	X	Х			
INCERTAE SEDIS	Λ				
Saltator similis	Х	Х			

D: 1 ·	Presence				
Bird species	RE	FI			
CARDINALIDAE					
Habia rubica		Х			
PARULIDAE					
Setophaga pitiayumi	Х	Х			
Myiothlypis leucoblephara	Х	Х			
Basileuterus culicivorus	Х	Х			
ICTERIDAE					
Cacicus haemorrhous	Х	Х			
Cacicus haemorrhous	Х	Х			
FRINGILLIDAE					
Euphonia pectoralis		Х			

APPENDIX III

Best models tested for occupancy probability as a function of the different habitat types (RE and FI) at INP, for the bird species that occurred in both habitats. Occupancy (Ψ); Probability of detection (p); Difference between the AICc models (Δ AICc). Taxonomy follows American Ornithologists' Union - South American Classification Committee Checklist for South American Birds (Remsen-Jr. *et al.* 2016).

Bird species	Model	ΔAICc	AICc Weight	
TINAMIDAE				
Crypturellus obsoletus	Ψ(habitat),p(.)	0.00	0.40	
Crypturellus tataupa	$\Psi(.),p(habitat)$	0.00	0.357	
COLUMBIDAE				
Patagioenas picazuro	Ψ(.),p(.)	0.00	0.386	
CUCULIDAE				
Piaya cayana	Ψ(.),p(.)	0.00	0.362	
TROGONIDAE				
Trogon surrucura	$\Psi(.),p(habitat)$	0.00	0.317	
Trogon rufus	Ψ(habitat),p(.)	0.00	0.448	
MOMOTIDAE				
Baryphthengus ruficapillus	$\Psi(.),p(habitat)$	0.00	0.404	
RAMPHASTIDAE				
Ramphastos dicolorus	Ψ(.),p(.)	0.00	0.459	
Selenidera maculirostris	Ψ(.),p(.)	0.00	0.389	
Pteroglossus castanotis	Ψ(habitat),p(.)	0.00	0.523	
PICIDAE				
Melanerpes flavifrons	Ψ(habitat),p(.)	0.00	0.369	
Dryocopus lineatus	Ψ(.),p(.)	0.00	0.402	
Campephilus robustus	Ψ(.),p(.)	0.00	0.358	
	$\Psi(.),p(habitat)$	1.34	0.183	
	Ψ (habitat),p(habitat)	2.57	0.099	

Bird species	Model	ΔAICc	AICc Weight
PSITTACIDAE			
Pionus maximiliani	Ψ(.),p(.)	0.00	0.518
	$\Psi(.),p(habitat)$	2.48	0.187
	Ψ (habitat),p(habitat)	4.59	0.034
Pyrrhura frontalis	Ψ(.),p(.)	0.00	0.323
Psittacara leucophthalmus	Ψ(.),p(.)	0.00	0.298
THAMNOPHILIDAE			
Hypoedaleus guttatus	Ψ(habitat),p(.)	0.00	0.486
Mackenziaena severa	Ψ(.),p(.)	0.00	0.417
Thamnophilus caerulescens	Ψ(.),p(.)	0.00	0.358
Dysithamnus mentalis	Ψ(habitat),p(.)	0.00	0.617
Herpsilochmus rufimarginatus	Ψ(.),p(.)	0.00	0.502
Drymophila malura	Ψ(.),p(.)	0.00	0.293
Pyriglena leucoptera	Ψ(.),p(.)	0.00	0.284
CONOPOPHAGIDAE			
Conopophaga lineata	Ψ(habitat),p(.)	0.00	0.482
GRALLARIIDAE			
Grallaria varia	Ψ(habitat),p(.)	0.00	0.439
RHINOCRYPTIDAE			
Eleoscytalopus indigoticus	$\Psi(.),p(habitat)$	0.00	0.270
FORMICARIIDAE			
Chamaeza campanisona	Ψ(habitat),p(.)	0.00	0.593
Chamaeza meruloides	Ψ(.),p(.)	0.00	0.3586
FURNARIIDAE			
Sittasomus griseicapillus	$\Psi(.),p(habitat)$	0.00	0.486
Dendrocincla fuliginosa	Ψ(.),p(.)	0.00	0.376
Dendrocolaptes platyrostris	Ψ(habitat),p(.)	0.00	0.461
Xiphocolaptes albicollis	Ψ(habitat),p(.)	0.00	0.360
Anabacerthia lichtensteini	Ψ(.),p(.)	0.00	0.441
Synallaxis ruficapilla	Ψ(.),p(.)	0.00	0.485
TYRANNIDAE			
Myiopagis caniceps	Ψ(.),p(.)	0.00	0.450
Camptostoma obsoletum	Ψ(.),p(.)	0.00	0.407
Capsiempis flaveola	Ψ (habitat),p(.)	0.00	0.468
Leptopogon amaurocephalus	Ψ(habitat),p(.)	0.00	0.468
Poecilotriccus plumbeiceps	$\Psi(.),p(habitat)$	0.00	0.348
Platyrinchus mystaceus	Ψ (habitat),p(.)	0.00	0.389
Lathrotriccus euleri	$\Psi(.),p(habitat)$	0.00	0.474
Megarynchus pitangua	$\Psi(.),p(habitat)$	0.00	0.347
Sirystes sibilator	Ψ(habitat),p(.)	0.00	0.506
TITYRIDAE			
Schiffornis virescens	Ψ(habitat),p(.)	0.00	0.640

Bird species	Model	ΔAICc	AICc Weight
CORVIDAE			
Cyanocorax chrysops	Ψ(habitat),p(.)	0.00	0.556
TURDIDAE			
Turdus leucomelas	$\Psi(.),p(habitat)$	0.00	0.468
THRAUPIDAE			
Trichothraupis melanops	Ψ(habitat),p(.)	0.00	0.349
Conirostrum speciosum	Ψ(.),p(.)	0.00	0.358
INCERTAE SEDIS			
Saltator similis	Ψ(habitat),p(.)	0.00	0.317
PARULIDAE			
Setophaga pitiayumi	Ψ(.),p(.)	0.00	0.412
Myiothlypis leucoblephara	$\Psi(.),p(habitat)$	0.00	0.338
Basileuterus culicivorus	Ψ(habitat),p(.)	0.00	0.480
ICTERIDAE			
Cacicus haemorrhous	Ψ (habitat),p(habitat)	0.00	0.480

APPENDIX IV

Bird species associated to river edge (RE) and forest interior (FI) habitats in the present study with their respective sensitivity to forest fragmentation (sensitive and non-sensitive) according to Anjos (2006) and Anjos *et al.* (2011).

Bird species	Sensitive	Non-sensitive
FI		
Crypturellus obsoletus		Х
Trogon rufus	Х	
Pteroglossus castanotis	Х	
Hypoedaleus guttatus		Х
Dysithamnus mentalis		Х
Conopophaga lineata		Х
Grallaria varia	Х	
Chamaeza campanisona	Х	
Dendrocolaptes platyrostris		Х
Leptopogon amaurocephalus		Х
Schiffornis virescens	Х	
Cyanocorax chrysops	Х	
Trichothraupis melanops	Х	
Basileuterus culicivorus		Х
Phaethornis pretrei	Х	
Xiphorhynchus fuscus	Х	
Automolus leucophthalmus		Х
Piprites chloris	Х	
Dacnis cayana	Х	
Pionopsitta pileata	Х	
Micrastur semitorquatus	Х	

Bird species	Sensitive	Non-sensitive
Euphonia pectoralis	Х	
Picumnus temmincki		Х
RE		
Melanerpes flavifrons	Х	
Xiphocolaptes albicollis		Х
Capsiempis flaveola		Х
Platyrinchus mystaceus		Х
Sirystes sibilator	Х	
Saltator similis		Х
Geotrygon montana		Х
Colaptes melanochloros		Х
Hylopezus nattereri		Х
Lochmias nematura		Х
Hemitriccus diops		Х
Tolmomyias sulphurescens		Х
Tityra cayana	Х	
Tachyphonus coronatus		Х
Hemithraupis guira		Х

Hyacinth Macaw (Anodorhynchus hyacinthinus) nests in a mosaic of protected areas in Carajás and surrounding areas, state of Pará, Brazil

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ABSTRACT: Natural history studies can provide information that can be used in species conservation and management. The present study provides information about the nests and nest sites of Hyacinth Macaws (*Anodorhynchus hyacinthinus*) in a mosaic of protected areas in Carajás and surrounding areas in the Amazon region of the state of Pará, Brazil. Data were collected during four expeditions conducted in 2007, 2008, 2013 and 2014. A total of 66 tree cavities were monitored, and 28 of these were used by Hyacinth Macaws. In addition, 38 breeding events were recorded (some nests were active in multiple seasons), and 17 eggs and 33 nestlings were observed. Environmental variables for the 24 cavities that were actively used by Hyacinth Macaws were evaluated. Most of the nest cavities were located in *Sterculia* sp. trees (86.6%) and in open areas (*e.g.*, near pastures). Nesting tree and cavity measurements were variable, but the mean diameter at breast height of the trees containing Hyacinth Macaw nest cavities was larger than that reported from other regions, such as the Pantanal. The present study describes the unique ecology and life history of Hyacinth Macaws in Carajás, for which information is scarce. This information will facilitate the conservation and management of Hyacinth Macaws and can be used locally for environmental education.

KEY-WORDS: breeding ecology, endangered bird, hollow-nesting bird, Psittacidae, Sterculia sp.

INTRODUCTION

The Hyacinth Macaw Anodorhynchus hyacinthinus is the largest member of the Psittacidae and has suffered major population declines over the last 30 years (BirdLife International 2016). The species is classified as "Endangered" in CITES Appendix I and as "Vulnerable" by the IUCN (BirdLife International 2018), owing to illegal trade, local hunting, and habitat loss (Mittermeier et al. 1990, Guedes 1993, Snyder et al. 2000). The species was once widely distributed in Brazil, yet most of the remaining individuals occur in three major regions. Current populations can be found in the eastern Amazon (Tocantins and Pará states), in central Brazil (Maranhão, Piauí, Bahia, Tocantins, Goiás, Mato Grosso, and Minas Gerais states), and in the Pantanal (Mato Grosso and Mato Grosso do Sul states; BirdLife International 2016); the latter region contains the largest remaining population (Munn et al. 1987, Guedes et al. 2008, Pivatto et al. 2008, BirdLife International 2016).

Although long-term studies of Hyacinth Macaws have been conducted in the Pantanal region (Guedes 1993, Antas 2004, Guedes 2009), little is known about the population status, breeding ecology, or life-history of the species in other locations (Snyder *et al.* 2000, Dornas *et al.* 2013). Vegetation structure and composition differ in the three main areas where Hyacinth Macaws occur, and consequently, resource availability varies (Presti *et al.* 2015). Furthermore, given the current trends of land use in the Amazon Forest, with high rates of deforestation and habitat loss, it is necessary to understand the ecological requirements of Hyacinth Macaw in the Amazon region.

One region in the Amazon, Serra dos Carajás, is a complex of protected areas in the locality between the Xingu and Tocantins Rivers that harbors a permanent breeding population of Hyacinth Macaws, which may be the largest in the Amazon region (Presti *et al.* 2009, Rosa 2016). The region is mostly covered by tropical rainforest, with dense populations of large trees, including *Sterculia* spp., *Euxylophora paraensis*, Brazil Nut Trees,

Bertholletia excelsa, and *Parkia* spp, that are important for the nesting of the local Hyacinth Macaw population (Presti *et al.* 2009, Rosa 2016). However, despite the importance of Serra dos Carajás to the maintenance of Hyacinth Macaw populations, little is known about its ecological requirements in the region (Presti *et al.* 2009). Accordingly, the aim of the present study was to describe the ecology and life-history of Hyacinth Macaws in the mosaic of protected areas in the Amazon region of Carajás, state of Pará, Brazil.

METHODS

Study area

We conducted surveys in the mosaic of protected areas in Carajás and the surrounding private properties, southeast of Pará (06°00'S; 50°30'W, Fig. 1). Carajás contains six protected areas: three National Forests (FLONA), one indigenous reserve, one Environmental Protected Area (APA), and one Biological Reserve, which together form a large forest fragment of ~12,000 km² (Martins *et al.* 2012). The vegetation in the region can be divided in two main categories: dense and open tropical rainforest (known locally as "Floresta Ombrófila Densa" and "Floresta Ombrófila Aberta"), which covers the majority of the area, and Savanna (metalophylic vegetation, known locally as *canga*; Martins *et al.* 2012). We monitored three

sites within the region, site 1: Rio Itacaiúnas; site 2: Canaã dos Carajás; and site 3: FLONA Itacaiúnas.

Site 1 was located between FLONA Carajás and FLONA Tapirapé-Aquiri. There were six vegetation types in the area: a) dense tropical rainforest, which features epiphytes and arboreal strata reaching 35 m, as well as highly dense understory strata; b) lowland open tropical rainforest; c) open tropical rainforest, which is characterized by the presence of palm trees, including Babassu Palm (*Orbignya phalerata*); d) open tropical forest with lianas; e) riparian forest, which features fast-growing vegetation, few canopy trees, intermediate strata with palm trees, and a high density of woody and herbaceous lianas; and f) savannas (ICMBio 2009).

Site 2 was located in the southernmost part of FLONA Carajás and the surrounding properties. Five municipalities are adjacent to the mosaic of protected areas in Carajás: Marabá, Parauapebas, Canaã dos Carajás, Água Azul do Norte, and São Félix do Xingu. The main economic activities of the region are livestock rearing and agriculture (Palheta-da-Silva 2004, Coelho *et al.* 2008), and the major threats to the protected areas are deforestation, wildfires, and mining. Although this area was extensively cleared for livestock and agriculture, the site contained large flocks of Hyacinth Macaws (Presti *et al.* 2009).

Site 3 was located in the northern area of FLONA Itacaiúnas and the surrounding properties. The site was highly modified and included many private properties

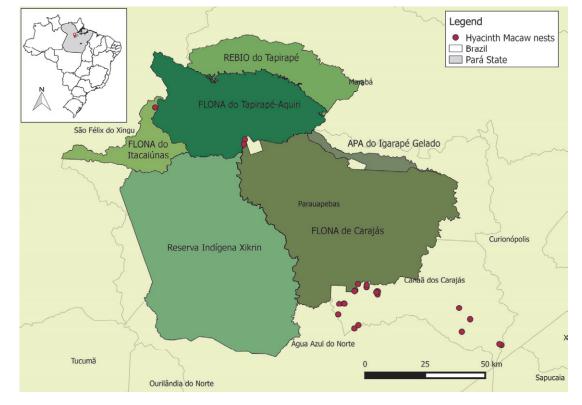


Figure 1. Hyacinth Macaw *Anodorhynchus hyacinthinus* nests monitored in a mosaic of protected areas and surroundings. Green areas represent the six protected areas in Carajás.

and pastures, and the FLONA contained large numbers of cattle. Natural vegetation cover was only observed on slopes and uneven terrain (G.F.S., pers. obs.).

Data collection

We conducted four approximately 30-day-surveys in the study area, in 2007, 2008, 2013, and 2014. We monitored each site from one up to four times between September and October of each year, resulting in a total of 458 h of fieldwork (visits at site 1 in 2013 and 2014 was 96 h; at site 2 in 2007, 2008, 2013 and 2014 was 338 h; and at site 3 in 2013 was 24 h). Expeditions depended on availability of boats, 4WD vehicles, and authorized teams. As such, site 2 that included several farms and readily accessible roads, was visited more frequently than the other two sites.

Bird sampling

Fieldwork involved searching for and describing cavities used by Hyacinth Macaws. We used tracks, creeks, and roads inside the properties to approach potential nesting trees, and information from local inhabitants was also important in locating nests. Only cavities that contained macaws or signs of the presence of Hyacinth Macaw, such as bite marks or remnant pieces of food, feathers, or feces, were monitored (Guedes 1993, Guedes & Seixas 2002). We classified the cavities as active (containing eggs or nestlings), inactive (without breeding activity at the time of the survey, but active in other seasons, either before or after the current season), or potential (without breeding activity, but with signs of Macaw presence).

We monitored cavities that contained stingless bees (*Trigona* sp.) but we did not monitor cavities that contained Honey Bees (*Apis mellifera*) or were otherwise inaccessible, due to structural issues (main trunk broken), even if the cavities had been previously active or Hyacinth Macaws were observed nearby. Some nest cavities were used for more than one season, and each observation of breeding activity was considered and counted as a breeding event.

We accessed cavities using climbing and rappelling techniques (Guedes 1993, Guedes & Seixas 2002). If nesting trees and cavities were active during at least one breeding season, 10 environmental variables were measured for each nesting tree and nest cavity (Table 1). We counted the number of eggs and nestlings. We collected environmental variables for 24 nesting trees, except for diameter at breast height (DBH), which was measured for 20 nesting trees.

Data analysis

Descriptive statistics (mean and standard deviation, SD) were calculated for the environmental variables collected

Table 1. Nest tree and cavity variables measured for trees containing Hyacinth Macaw nests at three sites in the mosaic of protected areas in Carajás, Pará state, Brazil.

Variable		Description
Nest tree	Species	Tree species in which nest cavity was located.
	Tree height	Height of nesting tree (m), measured from the entrance of the nest to the ground using a measuring tape; the entrance to the top of the tree was estimated visually. Both measurements were summed.
	Cavity height	Height of the cavity entrance (m), measured from the entrance of the nest to the ground using a measuring tape.
	DBH	Diameter at breast height (cm), measured as the circumference of the tree at breast height using a measuring tape.
	Location	Location of the nesting tree: open area, edge of the forest, or forest interior (> 5 m from the edge, Guedes 1993).
Nest cavity	Entrance width	Entrance width (cm), measured from lateral edges of the cavity with a ruler.
	Entrance height	Entrance height (cm), measured from the top edge to the bottom edge of the cavity using a ruler.
	Cavity width	Width of the cavity (cm), measured from the entrance to the back wall of the cavity using a ruler.
	Cavity depth	Depth of the cavity (cm), measured from the bottom edge of the entrance to the cavity floor using a ruler. This measurement was not taken for all nests.
	Origin	Most likely origin of cavity: broken branch, fungal action, or other birds (<i>e.g.</i> , members of the Picidae family).

at the active nest cavities. The relationship between tree and cavity height was evaluated using Pearson's correlation. All statistical analyses were performed using R version 3.1.1 (R Core Team 2014).

RESULTS

We monitored a total of 66 potential cavities in 2007, 2008, 2013, and 2014. Of these, 28 were actively used by Hyacinth Macaws (Fig. 1). Eight cavities were active more than once, with six used for two breeding seasons

and two used for three breeding seasons, and the other 20 nests were only used for a single breeding season. In total, 38 breeding events were recorded (Table 2).

We recorded cavities in 10 tree species, whereas breeding events were only recorded in seven (Table 3). At site 1, we registered nest cavities in five tree species, with breeding events only observed in *Cariniana* sp. and *Parkia* sp. At site 2, we registered nest cavities in six tree species, mostly (86.6%) in *Sterculia* sp. (likely *S. pruriens*), and all the cavities recorded at site 3 were found in *B. excelsa* (Table 3).

Most of the nests (87.5%, n = 21) were located in

Table 2. Nest cavities, breeding events, eggs, and nestlings of Hyacinth Macaws in the mosaic of protected areas in Carajás, Pará state, Brazil, registered during surveys in 2007, 2008, 2013 and 2014. Cavities: total number of cavities monitored; inaccessible: number of cavities with Hyacinth Macaws nearby but which could not be monitored due to the presence of Honey Bees (*Apis mellifera*) or due to the main trunk being broken; stingless bees: number of cavities with stingless bees; Other species: number of other bird species breeding in or using the cavity; *n* active cavities: cavities with eggs or nestlings; and breeding events/eggs/nestlings: number of breeding events, eggs, and nestlings per year/survey and total. (-) Not monitored in that season.

Site (effort)	Caritin	Inaccessible Stingles	Stingless	ngless Other <i>n</i> activ	<i>n</i> active	Breeding events/eggs/nestlings				s
	Cavities	(trunk/ <i>Apis</i>)	bees	species	cavities	Total	2007	2008	2013	2014
Site 1 - Rio Itacaiúnas (2013 - 2014, 96 h)	17	0/0	0	0	2	2/2/1	-	-	1/0/1	1/2/0
Site 2 – Canaá dos Carajás (2007 to 2014, 338 h)	45	8/2	2	1	25	35/15/30	9/0/9	12/4/12	7/2/6	7/9/3
Site 3 – FLONA Itacaiúnas (2013, 24 h)	4	3/0	0	0	1	1/0/2	-	-	1/0/2	-
Total	66	13	2	1	28	38/17/33	9/0/9	12/4/12	9/2/9	8/11/3

Table 3. Tree species containing Hyacinth Macaw cavities (active, inactive, and potential; see Methods for details) in the
mosaic of protected areas in Carajás, Pará state, Brazil.

Species	Local name	Site 1	Site 2	Site 3	No. cavities	No. breeding events
Bagassa guianensis	Tatajuba	1	-	-	1	0
Bertholletia excelsa	Castanheira-do-Pará	5	-	4	9	1
<i>Cariniana</i> sp.	Estopeira	1	-	-	1	1
Ceiba pentandra	Sumaúma		1	-	1	1
Euxylophora paraensis	Amarelão	8	-	-	8	0
Helicostylis tomentosa	Inharé	-	1	-	1	0
Parkia aff. gigantocarpa	Fava-grande	-	1	-	1	1
<i>Parkia</i> sp.	Faveiro	2	1	-	3	1
Schizolobium sp.	Paricá	-	2	-	2	1
<i>Sterculia</i> sp.	Axixá	-	39	-	39	32
Total		17	45	4	66	38

open areas, and the remaining 4.2% (n = 1) and 8.3% (n = 2) of the nests were located at the forest edge or in the forest interior, respectively. At site 2, which contained most of the monitored cavities, 95.2% (n = 20) of the cavities were located in open areas (*i.e.*, pasture). In addition, most of the cavities (70.9%, n = 17) had originated from broken branches, and the remaining cavities (29.1%, n = 7) originated from the effects of fungi or bird activity. At site 2, 76.1% of nest cavities originated from broken branches.

We collected environmental variables for 24 of the 28 nest cavities. Tree height ranged from 17 to 38 m, and cavity height ranged from 7.5 to 28.8 m, whereas entrance width varied between 10 and 51 cm and entrance height varied between 17 and 137 cm (Table 4). Cavity height was positively correlated with tree height (r = 0.57, P = 0.010) and DBH (r = 0.48, P = 0.048), and entrance width was correlated with entrance height (r = 0.53, P = 0.010).

A total of 17 eggs (four in 2008, two in 2013, and 11 in 2014) and 33 nestlings (9 in 2007, 12 in 2008, 9 in 2013, and 3 in 2014) were observed during the study. The median clutch size was 2 eggs per nest, and the number of nestlings varied from 1 to 2 per nest. Furthermore, competition for cavities was observed in 4 situations and involved 5 additional species, namely Honey Bees, stingless bees (*Trigona* sp.), Red and Green Macaws (*Ara chloroptera*), Scarlet Macaws (*Ara macao*), and Collared Forest Falcons (*Micrastur semitorquatus*).

DISCUSSION

In the present study in the Carajás region, the nest cavities of Hyacinth Macaws were predominantly located in *Sterculia* sp. trees. This finding agrees with a previous report by Presti *et al.* (2009), who found that *S. pruriens* was the preferred tree species for Hyacinth Macaws nesting in Canaá dos Carajás. In the Pantanal region, most Hyacinth Macaw nests are found in Panama Trees (*S. apetala*; 94% in south Pantanal, 91% in north

Pantanal, and 86% in the Poconé sub-region; Guedes 1993, Pinho 1998, Pinho & Nogueira 2003, Antas et al. 2010). Although other species, including E. paraensis and *B. excelsa*, also exhibited indications of being used by Hyacinth Macaws, there were no or few breeding events recorded in these species. However, other tree species have been reported to contain Hyacinth Macaw nest cavities at other locations. For example, in Pantanal, Hyacinth Macaw nest cavities were recorded in Albizia niopioides, Albizia inundata, Pacara Eearpod Trees Enterolobium contortisiliquum, and Vitex cymosa. In a central region of Brazil, Hyacinth Macaws were found to occasionally use nest cavities in palm trees, including Moriche Palm Mauritia vinifera (Munn et al. 1987, Antas et al. 2010). Because Hyacinth Macaws depend on softwood trees and pre-existing cavities to breed, the presence of trees with cavities suitable for breeding is vital for the persistence of Hyacinth Macaw populations (Presti et al. 2009). Additional studies are needed to evaluate the availability of nesting trees and nest cavities in these regions.

Most nest cavities we recorded in our study were located in open areas, frequently in grazed areas. This has also been reported for Hyacinth Macaws in the Carajás region (Presti et al. 2009). Hyacinth Macaws may choose to use open areas to optimize a variety of factors, such as visibility, food accessibility, and ease of mobility, since Hyacinth Macaws are relatively large-bodied (Pinho & Nogueira 2003, Presti et al. 2009). Nonetheless, forested environments seem to be more suitable for Macaws than cleared areas (Conrado 2015). Hyacinth Macaws may tolerate a certain level of landscape degradation at locations near forested areas and may use cleared areas for breeding activities since cleared areas have improved visibility (Guedes 1993, Conrado 2015). The forested areas in Carajás play an important role in maintaining the Hyacinth Macaw population in the area, as these areas provide breeding and feeding resources, thereby allowing the macaws to use the surrounding areas, including degraded, grazed, and cleared areas (Conrado 2015). Therefore, the conservation of Carajás protected areas is vital to the conservation of the local Hyacinth Macaw population.

Table 4. Characteristics of active Hyacinth Macaw nests (n = 24) in the mosaic of protected areas in Carajás, Pará state, Brazil.

Variable	Mean ± SD	Min	Max
Tree height (m)	25.9 ± 5.7	17.0	38.0
Cavity height (m)	16.8 ± 4.9	7.5	28.8
Entrance width (cm)	20.9 ± 10.4	10.0	51.0
Entrance height (cm)	48.1 ± 35.7	17.0	137.0
Cavity width (cm)	61.2 ± 21.9	33.0	110.0
Cavity depth (cm)	34.8 ± 53.8	0.0	270.0
Diameter at breast height (cm)	366.1 ± 243.8	205.0	995.0

Most of the cavities documented in Carajás originated from broken branches. This has also been reported for Hyacinth Macaws in north Pantanal (81% of cavities SESC RPPN [Private Natural Heritage Reserve] and Pirizal, in Poconé sub-region). However, the trend was not observed in south Pantanal (Nhecolândia), where fungi, together with termites and ants, were responsible for the formation of 56% of cavities (Guedes 1993, Pinho & Nogueira 2003, Antas et al. 2010). The availability of cavities is greater in old-growth forests (Mannan et al. 1980), and the greater cavity availability may allow Macaws to use cavities produced directly by decay processes and mechanical damage and to avoid infected cavities (Cockle et al. 2012). The conservation of well-structured forests with high cavity availability is crucial for the persistence of Hyacinth Macaws in the wild (Cockle et al. 2012).

The mean height of nesting trees was higher in Carajás than that reported for other regions (north Pantanal SESC: 18.7 ± 3.3 m; Poconé sub-region: 16.3 ± 4.3 m, and south Pantanal/Nhecolândia: 14.3 ± 2.0 m; Guedes 1993, Pinho & Nogueira 2003, Antas *et al.* 2010). The same pattern was observed for mean cavity height, which was higher in Carajás than in north Pantanal SESC (9.6 ± 2.2 m), the Poconé sub-region (5.8 ± 6.9 m), and south Pantanal/Nhecolândia (7.9 ± 2.0 m, Guedes 1993, Antas *et al.* 2010). Because cavity height was positively correlated with tree height in the present study, the higher mean heights of nesting trees and cavities are likely a consequence of the structure of the Carajás forest itself, which includes dense and tall vegetation (Antas *et al.* 2010, Martins *et al.* 2012).

Nest cavity features may have significant effects on nest success and survival (Britt 2011). Cavities maintain a microclimate and protect eggs and chicks from extreme weather and predation (Britt 2011). DBH was greater in Carajás than other regions, likely due to forest structure. Hyacinth Macaws will use any available cavities that are large enough to hold nestlings. *Sterculia apetala* trees, for instance, must attain a DBH of 60 cm or greater in order to shelter Hyacinth Macaw nestlings (Santos-Jr. *et al.* 2007). Although the physical structure of nest cavities is important, other factors, such as the availability of food and water, and predator detection, may also influence the use of specific trees by Hyacinth Macaws (Sedgwick & Knopf 1992, Antas *et al.* 2010).

Some nest cavities were active during more than one breeding season in Carajás. Similarly, 30% of the cavities in south Pantanal and Nhecolândia were used more than once (Guedes 2004). This could indicate fidelity to the breeding site. Because of their size, large Macaws may have fewer suitable available cavities and exhibit moderate nest-site fidelity (Britt 2011). Further studies that mark individual Hyacinth Macaws should be conducted to evaluate the nest fidelity of the species.

Competition for cavities was observed in Carajás, as previously reported for other sites in Pantanal (Guedes 1993, Pinho & Nogueira 2003, Guedes 2009, Antas et al. 2010). Interspecific cavity competitors may be twice as likely to cause nest failure, when compared to predation (Britt 2011). Although Rio Itacaiúnas (site 1) is a preserved area, deforestation can become an issue in other sites, since it may reduce the number of cavities in the area. Secondary cavity-nesters, such as Hyacinth Macaws, depend on pre-existing cavities, and a limitation in this resource can negatively influence populations (Cornelius et al. 2008). Hyacinth Macaws may share cavities with stingless bees (Trigona sp.), but never with Honey Bees, which are aggressive and do not tolerate the presence of Macaws in their cavities. Stingless bees, however, may also become competitors over time, as the hive grows and makes the cavity unsuitable for Macaws (Guedes 1993 & 2004, Santos-Jr. et al. 2007).

Most of the surveys in the present study were conducted in October. However, the nest cavities were observed with eggs and nestlings in different stages of development, which indicated asynchrony in the breeding activity of Hyacinth Macaws. This finding is in agreement with previous studies in Carajás (Presti *et al.* 2009), but not in the Pantanal, in which a synchronous breeding pattern was observed (Guedes 1993 & 2009). The Psittacidae family is characterized by a marked hatching asynchrony (Vigo *et al.* 2011) and environmental factors, such as rain, temperature, and food resource availability may contribute to the asynchrony observed in Carajás (Presti *et al.* 2009) but not in other regions.

The clutch size observed in the present study was similar to that reported from the Pantanal region (Guedes 1993, Antas *et al.* 2010), where most nests contain two eggs and one chick. Factors, such as food availability, cavity availability, the ability of male macaws to feed nestlings, and competition may influence clutch size (Guedes 1993, Renton 2004, Djerdali *et al.* 2008, Antas *et al.* 2010). Furthermore, long-lived species, such as Hyacinth Macaws, generally experience higher adult survival rates and have smaller clutch sizes (Britt 2011).

The present study provides relevant ecological information about Hyacinth Macaws in the Amazon region, for which information of the species is scarce. Surveys in this region are financially and logistically challenging. Therefore, the data presented here represents an important contribution and is expected to facilitate conservation efforts. Understanding key ecological factors, such as nesting tree species, nest distribution, and nest characteristics, has implications for the conservation of the species. Knowing which tree species to preserve and where to best preserve them may be vital to the conservation of Hyacinth Macaws in the region. In addition, the information provided by the present study may contribute to the development of environmental education activities, which can promote a better understanding of the ecological needs of the Hyacinth Macaw and, thereby, promote its conservation.

The presence of Hyacinth Macaw breeding cavities in the mosaic of protected areas in Carajás and the surrounding areas and the characteristics of these cavities are important for our understanding of the ecological requirements of Hyacinth Macaws in the Amazon region. Hyacinth Macaws have a highly specialized niche, and the limited availability of suitable cavities within breeding sites may reduce local populations to critical levels, thereby affecting their local persistence (Guedes 1993, Johnson et al. 1997, Guedes & Seixas 2002). Because habitat degradation is a major threat to many animal species, including Hyacinth Macaws (Johnson et al. 1997), land use in Carajás represents a major threat to the viability of the local Hyacinth Macaw population. As such, basic ecological information, such as that presented here, will facilitate local conservation efforts.

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First observations of the diet of the Pearl Kite (Gampsonyx swainsonii magnus) in southwestern Ecuador

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ABSTRACT: We describe the diet of Pearl Kite (*Gampsonyx swainsonii magnus*) by pellet analysis collected in an active nest in southwest Ecuador in May 2018. Reptiles were the most consumed taxonomic group both in frequency of occurrence (FO = 46.4%) and in biomass (B = 59.8%), followed by birds (FO = 33.9% and B = 38.3%) and insects (FO = 19.6% and B = 1.8%). Our results showed Pearl Kite as a generalist raptor but with a higher consumption of reptiles, which is according with previous studies in other areas where other subspecies inhabit, despite this subspecies showed a higher consumption of birds. Despite the low sample size, this study is interesting as it is the first one on the diet of the Pearl Kite for Ecuador and also for this subspecies. Basic studies on the trophic ecology of Neotropical raptors such as this are needed. For this reason we encourage further studies to fill existing gaps in knowledge and improve effective long-term conservation strategies.

KEY-WORDS: pellets, prey, raptor, reptiles, trophic ecology, tropical dry forest.

INTRODUCTION

Knowing the trophic ecology of a species is important not only to understand the ecology of the species itself, but also to understand the ecology of the community (Lewis et al. 2004). In addition to helping researchers understand trophic niches and how they relate to community structure, studying raptor diets can provide valuable information about prey distribution, abundance, behaviour, and vulnerability (Geng et al. 2009). Neotropical region is one of the areas with the highest diversity of raptors worldwide (Ferguson-Lees & Christie 2001), however the knowledge about many aspects of the ecology of this group is scarce (Freile et al. 2014, Monsalvo et al. 2018), even for other biological aspects. One of them is the Pearl Kite (Gampsonyx swainsonii), a small raptor that inhabits dry forests and arid regions of Central and South America (van Dort et al. 2010). Three subspecies are recognized, G. s. leonae distributed in southwestern El Salvador, south Honduras, west Nicaragua, Costa Rica and Panama, north and east Colombia through Venezuela and Trinidad to Guyana and Suriname, south to northeastern Ecuador and Brazil north of Amazon River; G. s. magnus distributed in west Ecuador and northwestern Peru; *G. s. swainsonii* distributed in Brazil south of Amazon River to east Peru, north and east Bolivia, Paraguay and N Argentina (Bierregaard-Jr. & Kirwan 2018). Unlike many other Neotropical raptors, Pearl Kite is favoured by deforestation (Ferguson-Lees & Christie 2001) and, consequently, it is expanding its range of distribution in recent decades, mainly in Central America, southeast of Brazil and Ecuador (Bierregaard-Jr. & Kirwan 2018).

Despite being a fairly common raptor in most areas where it lives, it is surprising the lack of detailed studies on its trophic ecology. This raptor is considered a specialist in the capture of small lizards (Ferguson-Lees & Christie 2001), most of these data had been gathered in field observations or partial diet studies carried out in Central America or northern South America, and are restricted to the subspecies *G. s. swainsonii* and *G. s. leonae* (Martínez 1998, Ffrench 2012, Koski *et al.* 2015, Martínez-Araya & Gastezzi-Arias 2016, Pineda *et al.* 2016, Araya-Céspedes & Carvajal-Sánchez 2017, Herrera & Acosta-Burgos 2018, Bierregaard-Jr. & Kirwan 2018). As long as we know, nothing has been published to date about the diet of *G. s. magnus*.

In this survey we describe the diet of the Pearl Kite

Gampsonyx s. magnus in southwestern Ecuador, based on the analysis of pellets collected from a single active nest.

METHODS

On May 2018, we located an active nest of Pearl Kite in the municipality of Zapotillo, province of Loja, southwest of Ecuador (4°19'35''S; 80°13'20''W, 244 m a.s.l.). The breeding territory was located in a semideciduous dry tropical forest, characterized by discontinuous, natural or induced glades, and the presence of isolated trees (Cueva & Chalán 2010). The nest was a basket made of loose twigs about 25 cm in diameter at a height of 4 m in a Barbasco (*Piscidia carthagenensis*), and in which there were two fledglings (Fig. 1).

To determine the diet, pellets were collected from the ground under the active nest. Pellets were analysed in the laboratory of the University of Loja (Ecuador). Each pellet was measured with a digital calliper (\pm 0.01 mm) and weighed with a precision balance Sartorius LA-230P (\pm 0.01 g). The identification of prey remains found in each pellet followed Orihuela-Torres *et al.* (2017).

The number of prey was calculated as the number of individuals based on the presence of unique structures, such as elytra of arthropods, jaws of reptiles, feathers and bones of birds, while other parts were not used to avoid double counting (Manning & Jones 1990). To calculate the biomass of birds not identified, we rely on the size of the bones that were of a medium-sized bird. For each prey and groups consumed, frequency of occurrence (FO) was calculated as the number of individuals of each kind of prey divided by the total number of prey (Formoso et al. 2012, Orihuela-Torres et al. 2017), and biomass consumed (B) multiplying the average body mass of each prey species by the number of total individuals consumed (Orihuela-Torres et al. 2018). Both indexes were shown as a percentage to facilitate comparison. For the calculation of biomass, we used the average mass of each prey species from Instituto Nacional de Biodiversidad (INABIO) collection in Quito, Ecuador (https:// www.inabioecuador.bio/ufl/collections/). The content obtained as remains of birds, insects and reptiles were quite fragmented, therefore it could not be deposited in collections and receive a voucher number. However, this material may be used for teaching purposes and will also be provided to interested researchers.

RESULTS

A total of 25 pellets were collected and showed the following measures: length (mean = 20.67 mm, SD = 4.76 mm, range = 15.83-29.27 mm), width (mean = 11.82 mm, SD = 3.12 mm, range = 8.5-17.64 mm), mass (mean = 0.44 g, SD = 0.26 g, range = 0.2-1.2 g). We found 56 prey from at least seven species belonging to three different classes: birds (2 spp. at least), reptiles (3 spp.), and insects (1 family). Prey mass varied from 1 g (Carabidae) to 30 g (*Iguana iguana* juvenile). Reptiles were



Figure 1. Nest of Pearl Kite (Gampsonyx swainsonii magnus) with one of the two fledglings, in Zapotillo, southwestern Ecuador.

the main prey both in frequency of occurrence (46.4%) and in biomass consumed (59.8%, Fig. 2). Second in rank were birds (FO: 33.9% and B: 38.3%), and then insects (FO: 19.6%, and B: 1.8%). At the species level, Knobbed Pacific Iguana (*Microlophus occipitalis*) was the most abundant prey both in frequency of occurrence (39.3%), as in biomass consumed (47.3%). Although most birds could not be identified at species level, they appeared in a large percentage of the pellets (76%). Carabids also showed a high frequency of occurrence (19.6%), in 32% of pellets, however their contribution in biomass was scarce (1.8%) (Table 1).

DISCUSSION

The diet of Pearl Kite in Zapotillo (SW Ecuador) corresponding to the subspecies *Gampsonyx s. magnus*, was in accordance with previous occasional data recorded for the other two Pearl Kite subspecies, placing this raptors as generalist, but consuming mainly reptiles (Martínez 1998, Koski *et al.* 2015, Martínez-Araya & Gastezzi-Arias 2016, Pineda *et al.* 2016, Araya-Céspedes & Carvajal-Sánchez 2017, Herrera & Acosta-Burgos 2018). The consumption of birds in our study seems much more common than previously reported (*e.g.*, Bierregaard-Jr. & Kirwan 2018). Several hypothesis could explain this results, *i.e.*, plasticity in foraging behaviour

and prey selection due to a higher local prey abundance (Morrison *et al.* 2008), or adaptation of this subspecies to consume more birds than the others subspecies. But due to the small sample size of our study we cannot support statistically any conclusion.

In this sense the composition of the diet would confirm the preference of this raptor for foraging in open habitats where it would find a higher abundance of reptiles such as *M. occipitalis* (Chávez-Villavicencio *et al.* 2018). Likewise, this statement is supported by the presence of birds related to open habitats and bushes such as the Saffron Finch (*Sicalis flaveola*) (Ridgely & Greendfield 2001).

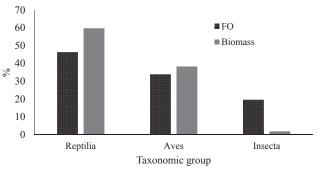


Figure 2. Frequency of occurrence (FO) and biomass consumed (B) in percentage of the diet of Pearl Kite (*Gampsonyx swainsonii magnus*) in a breeding territory of Zapotillo, southwestern Ecuador.

Table 1. Diet composition of the Pearl Kite (*Gampsonyx swainsonii magnus*) in a breeding territory of Zapotillo, southwestern Ecuador. The number of individuals consumed of each species is shown (n ind.), frequency of occurrence (FO) in percentage (%), the total biomass consumed (Biomass, in g) and the percentage (Biomass, in %).

Class/Order/Family/Species	<i>n</i> ind.	FO (%)	Biomass (g)	Biomass (%)
Reptilia	26	46.4	362	59.8
Squamata				
Iguanidae				
Microlophus occipitalis	22	39.3	286	47.3
<i>Iguana iguana</i> (juvenile)	1	1.8	30	5.0
Polychrus femoralis	1	1.8	22	3.6
Phyllodactylidae				
Phyllodactylus reissii	2	3.6	24	4.0
Insecta	11	19.6	11	1.8
Coleoptera				
Carabidae	11	19.6	11	1.8
Birds	19	33.9	232	38.3
Passeriformes				
Emberizidae				
Sicalis flaveola	2	3.6	28	4.6
Medium-sized birds	17	30.4	204	33.7
Total	56	100	605	100

Raptors are very important components in the ecosystems, since they can be used as bioindicators or key species for community monitoring, in addition to providing important ecosystem services (Donázar et al. 2016). Currently there are large gaps in knowledge of basic aspects of raptor ecology in the Neotropics (Buechley et al. 2019) such as the composition of diet, phenology and reproduction, home range or even population sizes. Basic information like this study is necessary to complete these gaps, but since the diet varies depending on factors such as sex, age, habitat and season (Beeston et al. 2005) it is necessary that this knowledge be completed throughout the distribution area, as well as replicated during different seasons and by age classes. By improving our knowledge of the trophic ecology of these magnificent birds at the top of the food chain, we will be able to propose effective long-term conservation strategies.

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A comparison of bird communities in natural and revegetated grasslands in south Brazil

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ABSTRACT: Natural grasslands are declining due to loss, fragmentation and degradation, resulting in the decline of grasslandassociated bird species. The Pampas Biome in south Brazil is not exception to this worldwide trend, facing the expansion of croplands and afforestation with exotic tree plantations for cellulose production. To cope with the continuous degradation and loss of grasslands, restoration is an important conservation strategy, but basic information regarding the response of the fauna to restoration practices in southeastern South America grasslands is lacking. Here we compared the structure of bird communities in natural grasslands and revegetated grasslands after mining by planting native and exotic grasses. We sampled birds using 5-min point counts with unlimited radius in three replicates of each habitat (natural and revegetated grasslands; average size 22.2 ± 2.3 ha). We also compared the vegetation density between the two habitat types. The structure of bird communities at natural and revegetated areas, and also a distinct species composition. Ten of the 11 grassland species that were associated to one of the two habitat types occurred more frequently in natural grasslands, which had higher vegetation density than revegetated areas. Even a decade after the beginning of the restoration process, revegetated areas did not resemble natural grasslands in bird species richness, abundance, and composition. These results differed from another study conducted in the Brazilian Pampas in which native plant species were used to actively restore a grassland, we encourage greater representation of native plant species in restoration projects.

KEY-WORDS: exotic grasses, habitat restoration, mining, Pampas Biome.

INTRODUCTION

Recently we saw an upsurge of calls for grassland conservation in face of the many threats to grasslands, including afforestation and invasion by exotic plants (Parr *et al.* 2014, Bond 2016). In the Pampas Biome of southern Brazil, for instance, approximately 60% (104,553 km²) of former grassland area had been destroyed by 2002, mostly due to its conversion to arable fields or afforestation with exotic trees (Andrade *et al.* 2015). This makes the Pampas the second Brazilian biome regarding the relative magnitude of land use changes, getting behind only to the Atlantic Forest (Overbeck *et al.* 2013).

To confront the continuous degradation and loss of grasslands, or any other vegetation type, ecological restoration is an important strategy. However, research and practice of restoration of tropical grassy biomes has traditionally fallen behind other vegetation types, such as forests (Overbeck *et al.* 2013). In addition to technical issues for proper grassland restoration (*e.g.*, availability of seeds of native grassland species), basic information regarding the response of the fauna to restoration practices is lacking. Some pending questions about the conservation value of restored grasslands are, for example, threatened animals. Birds, for instance, can be divided into different categories of dependence on grasslands, with grassland-restricted species in general among the most threatened species (Azpiroz & Blake 2009, Azpiroz *et al.* 2012): Do such bird species use grasslands restored by planting mostly exotic grasses? In North America we know that grasslands planted mostly with non-native grasses on reclaimed mines supported a community of bird species typical of natural grasslands (Scott *et al.* 2002).

Here we compared the structure of bird communities in natural grasslands and grasslands revegetated after mining with the planting of native and exotic species. More specifically, we investigated how the species richness, abundance and composition of the bird communities at revegetated areas with such a mixture of native and exotic species (but with a predominance of the latter) compare to natural grasslands. Our ultimate goal is to evaluate the efficacy of the restoration procedures currently used by mining companies from the bird's point of view. Such companies follow the Brazilian legislation that permits the use of exotic plant species in restoration (for more details see Normative Instruction ICMBio 2014).

METHODS

Study areas

This study was carried out in areas of *Companhia Riograndense de Mineração* (CRM), at Candiota region in the state of Rio Grande do Sul, south Brazil (31°33'S; 53°40'W). This region is largely occupied by openpit coal mining areas, revegetated areas, and natural grasslands (Fig. 1). According to the Köppen (1948) classification, the climate in the area is *Cfa*, with cold winter, hot summer, and rainfall distributed over the year but more pronounced between July and October. The average relative humidity is 73% in summer and 83% in winter. Average annual rainfall is around 1400 mm.

From 9 to 13 years before this study, active restoration techniques were performed by CRM in which soil from areas that would be mined later was deposited on mined areas after the reconfiguration of the topography.

Fertilizers such as triple superphosphate (NPK) and potassium chloride were added, a mix of mostly exotic (*Lolium multiflorum*, *Urochloa decumbens*, *Chloris gayana*, *Cynodon dactylon*, *Trifolium repens*) and one native grass species (*Paspalum notatum*), were sowed, and again the fertilizer (NPK) and urea were added. Natural grasslands were not actively managed, but were under fire and ungulate grazing, common and part of the evolutionary history of natural Pampas grasslands (Pillar & Velez 2010). Areas with revegetated and natural grasslands had similar sizes, ranging from 20 to 25 ha.

Bird and vegetation sampling

We sampled birds from May to December 2006 using 5-min unlimited point counts (Bibby et al. 1992) carried out from early to mid-morning (06:30-10:00 h) and late afternoon (16:00-17:30 h) in three replicates of two habitat types (natural and revegetated grasslands). Only birds seen or heard inside the sampled areas of natural and revegetated grasslands were considered. The average distance between sampling areas was c. 1.5 km. In each area we sampled eight points distant 200 m from each other in each season of the year, totaling 32 points per area and 96 per habitat. The locations of sampling points were not fixed but randomized at each season using xy coordinates (maintaining, however, the 200 m minimum distance between points). The scientific nomenclature and taxonomic ordering of birds follow Piacentini et al. (2015).

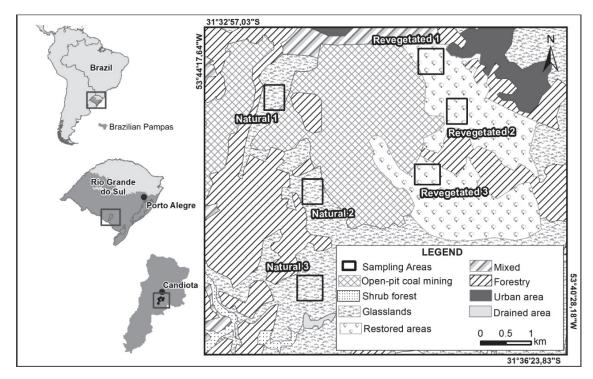


Figure 1. Map of the study site showing the location of the natural and revegetated grassland areas in southern Brazil.

At each sampling area we set two parallel transects of 150 m length each and separated 150 m from each other to assess the vertical structure of the vegetation. At each 5 m interval we counted the number of times the vegetation touched a 2-m graded rod (maximum height of vegetation) in four height classes (0–50, 51–100, 101– 150, and 151–200 cm). A quantification of the vertical structure was then given by the density of vegetation at different height classes.

Data analyses

Bird species richness was compared between natural and revegetated grasslands in two ways. Firstly, we did an analysis of rarefaction based on individuals (i.e., number of records) implemented with EstimateS® version 9.1 (Colwell 2013). This is a non-biased way of comparing the richness of species between areas, as it is not influenced by variations in the density of individuals among areas (Colwell & Coddington 1994, Krebs 1999, Gotelli & Colwell 2001). In addition, due to possible spatial dependence among samples, we compared bird species richness and number of records through a hierarchical mixed model test (nested ANOVA) using the function "lme" of the package "nlme" in R software (Oksanen et al. 2011, McDonald 2014). Sampling points were treated as random variables within each fixed treatment (Natural vs. Revegetated).

Following Azpiroz *et al.* (2012), we classified bird species according to their association to grasslands in southeastern South America in the following categories: (1) grassland-restricted species, *i.e.*, species that do not use alternative habitats, (2) species that extensively use grassland habitats, but other habitats as well, and (3) species that make extensive use of grassland habitats only in certain subregions of the southeast South American grasslands.

We calculated the species diversity for each habitat type using the Shannon-Wienner index (log [x]) (Magurran 1988). To test if bird species used more frequently any of the two habitats, we performed G tests for the species with 10 or more records. These tests contrasted the frequencies of records at natural and revegetated grasslands with the expected frequencies based on equal number of records at each habitat.

We performed group analysis to test for possible differences in the composition of bird communities between natural and revegetated grasslands using the Multi-Response Permutation Procedures (MRPP) method with Euclidean distances (Zimmerman *et al.* 1985). This method makes it possible to evaluate the dissimilarity between groups of samples. If the mean dissimilarities of the species composition observed is less than the dissimilarity between randomized groups (999 randomizations) based on the actual distribution of the observed data, the species composition is different. The change-corrected within-group agreement (A) provides and effect size of the dissimilarity between groups, ranging from < 0 to 1. The smaller is A the greater the heterogeneity between groups, while if A = 1 groups are identical. We tested for correlations in the spatial distance (Euclidean distance) and similarity in species composition (Bray-Curtis distance) between the studied areas using a Mantel test (Quinn & Keough 2002). We performed all these analyses with the "vegan" package in R software (Oksanen *et al.* 2011, R Core Team 2017).

To test for differences in vegetation density between natural and revegetated grasslands, we used a resampling technique performed with the Resampling Stats® program (Simon 1997, Blank *et al.* 2001) in which the mean between-habitat difference for each vegetation height class was compared with the mean differences obtained from 10,000 randomizations of the data, accepting as significant observed differences that lied within the 5% frequency distribution of the randomized differences.

RESULTS

We made 2298 records (1459 in natural, and 839 in revegetated grasslands) of 49 bird species (21 families, 42 species in natural, and 35 in revegetated grasslands; Appendix I). The cumulative number of bird species stabilized in both habitats, indicating that we sampled most of the species in the studied areas (Fig. 2). Rarefying down the number of records to 800 in both habitats, we got 40 species in natural and 35 species in revegetated grasslands, with non-overlapping confidence intervals indicating different species richness (Fig. 2).

Most species (28 species) were not associated with grasslands, while 13 species make extensive use of grasslands (category 2 of Azpiroz *et al.* 2012), and 8 species use grasslands only in certain regions (category 3).

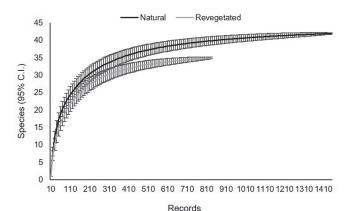


Figure 2. Rarefaction curve based on the number of bird records and their respective confidence intervals (95%) in natural and revegetated grasslands in southern Brazil.

No grassland-restricted species (category 1) was recorded (Appendix 1). The representativeness of each category of grassland association did not differ between habitats (G test: $\chi^2 = 1.080$, df = 2, P = 0.580). Among the 30 species with 10 or more records, 14 used natural grasslands more frequently than expected by chance, and only *Colaptes campestris* was associated with revegetated grasslands (Appendix I). Considering only grassland-associated birds (categories 2 and 3), 11 out of 14 species were associated to a habitat type, once again all but *C. campestris* used more frequently natural grasslands (Appendix I).

The species with the highest number of records in both habitats were Zonotrichia capensis, Sicalis luteola, Ammodramus humeralis, and Embernagra platensis, together accounting for 51% of the total number of records (Appendix I). Natural grasslands had greater diversity than revegetated areas (H' = 2.986 and 2.625, respectively), a difference mirrored by the species richness (*F* = 6.240, *P* < 0.050), and bird abundance (*F* = 19.508, P < 0.001; Fig. 3). Natural and revegetated grasslands also differed in species composition (MRPP: observed delta = 10.45, expected delta = 10.54, A = 0.007, P = 0.019). There was no correlation between the distance separating the studied areas and the pairwise dissimilarity in species composition (Mantel r = 0.198, P = 0.374, 719 permutations), indicating that species composition was not related to spatial relationships among areas.

Natural grasslands areas had higher vegetation densities at height classes 0–50 cm (mean difference = 6.28, P = 0.040), 51–100 cm (3.43, P = 0.001), and 101–150 cm (4.35, P = 0.001), but not at 151–200 cm (1.41, P = 0.150) in which a few plants were recorded at both habitats (Fig. 4).

DISCUSSION

The structure of bird communities at natural and revegetated grassland areas differed, with natural grasslands presenting higher species richness and abundance than revegetated areas, and also a distinct species composition. In addition, most of the grasslandassociated birds occurred more frequently at natural grasslands. Differences in vegetation structure between natural and revegetated grasslands is a factor to explain such differences, since the composition of bird communities in southern Brazilian grasslands (and grasslands in other regions; Hovick et al. 2015) is strongly influenced by the spatial heterogeneity of vegetation, that is, by structural changes in vegetation mostly caused in the region by disturbances like fire and grazing (Bencke 2009, Dias et al. 2014). The high sensitivity of birds to vegetation structure was also observed by Fontana et al. (2016) who found greater species richness of birds in

general, and grassland-associated species in particular, in natural grasslands compared to "improved" grasslands, *i.e.*, natural grasslands managed with the addition of fertilizers and exotic species, demonstrating the importance of natural areas for grassland birds (see also Silva *et al.* 2015).

Together with the lower density of vegetation in revegetated areas, the low number of plant species sowed, most of them exotics, in the restoration process is an additional factor that possibly contributed to the lower diversity of birds in revegetated grasslands. In comparison, natural grasslands are composed by a much diverse plant community (Menezes *et al.* 2018), which naturally promotes spatial heterogeneity. In the sole comparable study on the recovery of a bird community in actively restored grassland in southeastern South America, Silva (2019) found different composition, but similar bird species richness and abundance between a 3-yr old grassland restored with native plants and a natural grassland area. Limited as the comparison with

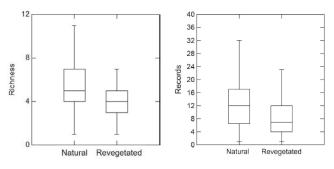


Figure 3. Boxplots showing the median (horizontal line), 25% – 75% quartiles (box upper and lower limits), and maximum and minimum values (indicated by the vertical bars) of the species richness and number of birds recorded at natural and revegetated grasslands in southern Brazil.

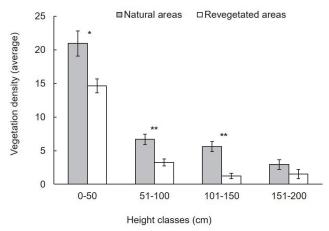


Figure 4. Vegetation density at different height classes in natural and revegetated grasslands as denoted by the number of touches of the vegetation in a 2 m graded rod. Bars indicate standard errors. Between-habitat differences are indicated by *(P < 0.050) and **(P < 0.010).

this single study might be, we expected similar results for our much older (9–13 years) revegetated areas. That our revegetated areas had smaller bird species richness and abundance than natural grassland areas is indicative that the predominance of exotic grasses in the seed mixtures used in the restoration process is inadequate for the recovery of grassland bird communities. Nonetheless, the species richness we recorded in natural grasslands (42 species) is within the range found by Silva (2019, 30– 46 species), while the richness in our restored sites (35 species) did not greatly differ from her active restoration (30 species).

Apart from the apparent low quality of revegetated areas, the fact that we have not recorded grasslandrestricted birds, that are usually more sensitive to habitat quality (Azpiroz & Blake 2009), may have to do with the landscape context of our natural and revegetated areas, surrounded by exotic monocultures of grasses (*Urochloa* sp.) and trees (*Pinus* sp., *Acacia* spp., *Eucalyptus* spp.), agriculture, and extensive livestock farming. However, as restoration of grasslands still faces many technical problems, even the small, isolated grassland remnants remaining are worth conserving (Bond & Parr 2010). As we shown here, they were preferred over revegetated areas by most grassland-associated birds.

Even though revegetated areas did not represent highquality habitats for several birds, they served as refuges for many species that do not tolerate strongly altered habitats as occur in the matrix surrounding our study areas. What remains to be learned is if revegetated areas offer structural conditions that allow the reproduction of these species, since grassland birds select breeding and nesting habitats with very specific characteristics (Cody 1985). While the reproductive success of birds in North American grasslands recovered after coal mining was comparable to that of natural habitats, indicating that revegetated areas do not necessarily represent reproductive traps for birds (Galligan *et al.* 2006), the daily survival rates of birds reared in planted grasslands was lower compared to natural grasslands (Fisher & Davis 2011).

In sum, we found that a decade after the use of predominantly exotic plants to restore grasslands on reclaimed mined areas in the Pampas of south Brazil resulted in a vegetation that was used by several grasslandassociated bird species, but bird species richness, abundance, and composition did not resemble natural grasslands. Our results differed from the only other comparable study conducted in southeastern South America grasslands that, however, used native plant species in the active restoration (Silva 2019). Therefore, until we have additional studies addressing the use of exotic grasses for the recovery of bird communities in the realm of southeastern South America grasslands, we encourage greater representation of native plant species in restoration projects, stimulating policies to overcome the technical difficulties of making available seeds of native species for restoration purposes.

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APPENDIX I

Bird species recorded in natural grasslands and grasslands revegetated after mining in south Brazil.

Family	Grassland	Number	<i>P</i> value ^b	
Species	specialization ^a	Natural	Revegetated	1 ² value ³
Tinamidae				
Rhynchotus rufescens	2	69	25	0.009
Nothura maculosa	sa 2 66		31	0.010
Anatidae				
Amazonetta brasiliensis	-	0	2	
Accipitridae				
Elanus leucurus	3	1	0	
Rupornis magnirostris	-	0	1	
Charadriidae				
Vanellus chilensis	2	2	11	0.055
Columbidae				
Columbina picui	-	8	3	
Leptotila verreauxi	-	0	1	

Family	Grassland	Number	<i>P</i> value ^b		
Species	specialization ^a	Natural	Revegetated	tated P values	
Cuculidae					
Tapera naevia	-	8	8		
Guira guira	-	8	4		
Picidae					
Colaptes campestris	3	7	40	0.002	
Veniliornis spilogaster	-	1	0		
Cariamidae					
Cariama cristata	2	0	6		
Thamnophilidae					
Thamnophilus caerulescens	-	6	0		
Thamnophilus ruficapillus	-	27	12	0.080	
Furnariidae					
Synallaxis cinerascens	-	6	7	0.84	
Synallaxis spixi	-	13	4	0.08	
Furnarius rufus	3	21	6	0.03	
Anumbius annumbi	3	19	0	< 0.001	
Phacellodomus striaticollis	3	72	1	< 0.001	
Tyrannidae					
Camptostoma obsoletum	-	11	11		
Serpophaga subcristata	-	18	20	0.73	
Pitangus sulphuratus	-	14	25	0.20	
Xolmis cinereus	2	6	0		
Tyrannus savana	3	5	2		
Vireonidae					
Cyclarhis gujanensis	-	3	0		
Hirundinidae					
Pygochelidon cyanoleuca	_	4	0		
Froglodytidae					
Troglodytes musculus	-	13	27	0.11	
Turdidae		-			
Turdus rufiventris	-	8	5	0.55	
Turdus amaurochalinus	-	1	0		
Motacillidae					
Anthus lutescens	2	0	7		
Passerellidae					
Zonotrichia capensis	-	224	164	0.03	
Ammodramus humeralis	2	135	122	0.55	
Parulidae					
Geothlypis aequinoctialis	-	50	16	0.002	
Icteridae		-			
Chrysomus ruficapillus	-	5	0		
Pseudoleistes virescens	2	19	0	< 0.001	

Family	Grassland		Number of records			
Species	specialization ^a	Natural	Revegetated	<i>P</i> value ^b		
Sturnella superciliaris	2	0	6			
Agelaioides badius	-	8	0			
Molothrus bonariensis	3	62	0	< 0.001		
Thraupidae						
Microspingus cabanisi	-	3	0			
Poospiza nigrorufa	3	23	2	0.007		
Sicalis flaveola	-	73	4	< 0.001		
Sicalis luteola	2	154 179		0.33		
Sporophila caerulescens	-	2	8	0.15		
Volatinia jacarina	2	0	7			
Donacospiza albifrons	2	87	11	< 0.001		
Embernagra platensis	ra platensis 2		58	< 0.001		
Paroaria coronata	-	2	0			
Fringillidae						
Spinus magellanicus	-	56	3	< 0.001		

^a Association to grasslands in southeastern South America according to Azpiroz *et al.* (2012): (1) grassland-restricted species, *i.e.*, species that do not use alternative habitats, (2) species that use extensively grassland habitats, but other habitats as well, and (3) species that make extensive use of grassland habitats only in certain subregions of the southeastern South American grasslands. A hyphen denotes species not associated to grasslands. ^b *P* values for G tests contrasted the frequencies of records at natural and revegetated grasslands with the expected frequencies based on equal number of records at each habitat. Only species with ten or more records were tested.

Novel microsatellites for *Cypseloides fumigatus*, cross-amplifiable in *Streptoprocne zonaris*

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ABSTRACT: Based on microsatellite prospection, we isolated and characterized 21 microsatellite markers for the Sooty Swift (*Cypseloides fumigatus*) and tested the cross-amplification in the White-collared Swift (*Streptoprocne zonaris*). Both species are New World species included in the Apodidae family. From these 21, only 13 loci were polymorphic in the Sooty Swift, and their levels of polymorphism were surprisingly low compared to related species. Cross-amplification in the White-collared Swift was successful for 11 loci of the 13 polymorphic found for the Sooty Swift, but seven were monomorphic and four were biallelic. The microsatellites described here could be useful in future genetic population studies for Sooty Swifts and related species.

KEY-WORDS: Cypseloidinae, Sooty Swift, Ultraconserved elements (UCEs), White-collared Swifts.

Sooty Swifts, Cypseloides fumigatus, and White-collared Swifts, Streptoprocne zonaris, are New World apodid species. Whereas the White-collared Swift has a wide distribution, ranging from southern USA to southwestern Argentina, the Sooty Swift ranges from Argentina to Bolivia, Brazil and Paraguay (Chantler 1999). Both species present highly aerial behavior and are frequently seeing foraging and nesting together (Marín & Stiles 1992, Pearman et al. 2010, Biancalana et al. 2012, Biancalana 2014 & 2015). They nest in colonies, usually next to waterfalls and wet caves. Like other swift species they are philopatric, returning to use the same nest site over several years (Marín & Stiles 1992, Collins & Foerster 1995, Biancalana, pers. obs.). Here, we describe and characterize 13 novel polymorphic microsatellites for the Sooty Swift. In addition, we cross-amplified 11 of the 13 loci in the White-collared Swift from which seven were monomorphic and four were biallelic.

Microsatellite prospection was based on off-target sequences obtained through sequence capture and nextgeneration sequencing of Ultraconserved Elements (UCEs), as described in Amaral *et al.* (2015). Genomic DNA was obtained from a muscle sample from a Sooty Swift individual deposited at the *Laboratório de Genética e Evolução Molecular de Aves* (Universidade de São Paulo USP, Brazil – LGEMA #11411), collected at Ortigueira, Paraná state (24°12'S; 50°55'W) using the Qiagen DNeasy kit (Valencia, CA) with an RNAse treatment. Sequencing was performed at Rapid Genomics (Gainesville, FL, USA). The contigs obtained were screened for perfect di-, tri-, tetra-, penta-, and hexa-nucleotide with at least five repeats using QDD (Meglécz *et al.* 2010). QDD and Primer3 (Koressaar & Remm 2007, Untergasser *et al.* 2012) were used to design primers with default parameters and minimum fragment length of 100 bp. Each forward primer was designed with a M13 sequence (CACGACGTTGTAAAACGAC) added to its 5' end in order to pair with a third fluorescently labeled *primer*, according to the universal labeling method described by Boutin-Ganache *et al.* (2001).

For the characterization of the prospected microsatellites, 34 samples of Sooty Swifts were collected: 19 at Intervales State Park (ISP, Ribeirão Grande, São Paulo state, Brazil) and 15 at Sussuapara Canyon (SC, Ponte Alta do Tocantins, Tocantins state, Brazil). Both sites are known to have nests of the species and are monitored since 2010 (SC) and 2012 (ISP). To check the cross-amplification for White-collared Swifts, 10 adult samples were collected at ISP. Adults were mist netted and nestlings were captured in their nests and returned after sampling. Blood samples were collected from the brachial vein using microcapillary tubes and stored in absolute ethanol under room temperature. Genomic DNA was extracted from whole blood with a salt protocol adapted

from Aljanabi & Martinez (1997).

PCRs were carried out in volumes of 12 µl containing 1.5 µl of extracted DNA (30–50 ng/µl), 0.2 mM of dNTPs, 1 × PCR buffer, 3 pmol of the reverse primer, 1 pmol of forward primer, 2 pmol of FAM/HEX M13 primer, 2.5 mM of MgCl₂ and 0.5 U of Taq Polymerase (Sinapse, Inc.). Thermocycling conditions consisted of 95° (5 min), 35 cycles at 94°C (30 s), $T_A^{\circ}C$ (30 s), 72°C (30 s) with a final extension at 72°C (10 min). The optimal annealing temperature for each primer pair was determined using a temperature gradient cycle from 56 to 64 °C with a 2 °C difference between steps. PCR products were visualized on a 1.5% agarose gel using a 100 bp ladder. Successful PCR products were genotyped on a ABI 3730 (Applied Biosystems) automated sequencer and analyzed with GeneMarker 2.7.0 (Softgenetics).

We used GenAlEx 6.5 (Peakall & Smouse 2012) to estimate the number of alleles, and expected and observed heterozygosities. GENEPOP 4.2 (Raymond & Rousset 1995, Rousset 2008) was used to search for deviations from Hardy-Weinberg and linkage equilibrium. Benjamini & Yekutieli (2001) correction was applied to adjust the critical values for multiple comparisons. The search for null alleles and the estimation of their frequencies was done using MICRO-CHECKER 2.2.3 (van Oosterhout *et al.* 2004).

A total of 423 microsatellites were prospected for the Sooty Swift. From these, 138 (32.6%) were linked to UCEs regions and were discarded from primer design as they may be under purifying selection (Harvey et al. 2016) and are possibly monomorphic. From the remaining 285 microsatellites, we designed primers for 21 perfect loci (Table 1). Most were dinucleotides (81.0%), followed by trinucleotides (14.3%) and one pentanucleotide (4.8%). From the 21 loci tested in the 34 samples of Sooty Swifts, 16 were successfully amplified in Sooty Swifts (Table 1). Although the loci Cyps23 and Cyps34 successfully amplified, they did not produce consistent genotypes due to an excess of stutter bands. Thirteen loci were polymorphic with the number of alleles ranging from 2 to 8 (mean \pm SD: 3.3 \pm 0.43, Table 1). Observed heterozygosities ranged from 0.03 to 0.56 (mean $Ho \pm$ SD: 0.16 ± 0.05) and expected heterozygosities from 0.03 to 0.67 (mean $He \pm$ SD: 0.25 \pm 0.06, Table 1). These heterozygosities were low when compared to that of phylogenetically close species, though the microsatellites used in the reference literature where not the same as those designed and tested in this study (Lance et al. 2009, González et al. 2010, Oyler-McCance et al. 2011, Gutiérrez-Rodriguez et al. 2013, Sanvicente et al. 2016; Table 2). Several attempts of amplification using two hummingbird microsatellites developed in other studies, Acya3-3 and Hxan07 (Gutiérrez-Rodriguez et al. 2013, Sanvicente et al. 2016), were done by RNB, with different

settings of temperature and thermocycling conditions, but resulted in no amplifications for Sooty and Whitecollared Swifts.

The test on Hardy-Weinberg equilibrium (HWE) for each locus revealed deviations in 5 loci after Benjamini & Yekutieli (2001) correction (Cyps8, Cyps14, Cyps26, Cyps33, and Cyps35 - Table 1). The following loci had evidence of null alleles (with their respective frequencies): Cyps8 (0.237), Cyps14 (0.281), Cyps24 (0.156), Cyps26 (0.310), and Cyps33 (0.279). Among 91 paired loci, four cases of linkage disequilibrium were detected for the loci Cyps8 and Cyps12, Cyps12 and Cyps25, Cyps14 and Cyps26, and Cyps24 and Cyps35 [P < 0.01after Benjamini & Yekutieli (2001) correction]. The deviations to HWE might be caused by the presence of null alleles in some loci (Cyps8, Cyps14, Cyps26, Cyps33 and Cyps36), or due to population substructure, since samples were collected in different localities. The cross-amplification of the prospected loci in Whitecollared Swifts was successful for 11 of them. However, the loci were either monomorphic or biallelic (Table 1). Locus Cyps9, that did not amplify in Sooty Swifts, was successfully amplified for White-collared Swifts.

The low polymorphism found in Sooty Swift microsatellites may result from many factors, ranging from loci linked to conserved regions to ecological and historical factors. Milot et al. (2007) suggested, for example, that some life history traits associated with demographic patterns may result in a small effective population size, that in long periods of time can result in loss of genetic diversity. This means that not only bottlenecks might be the main cause for low genetic variability in birds (Amos & Harwood 1998). Because Sooty Swifts usually establishes small populations (ranging from two to ten individuals in general), exhibits philopatry, presents a long breeding period and raises just one chick per year (Biancalana et al. 2012, Biancalana 2015), the population might be suffering from inbreeding and/or might naturally have low genetic variability. Alternatively, population size variation due to historical factors - as climate change in the past - could also explain low genetic variation. Additional studies will be important to test alternative scenarios related to the low nuclear variation found here. The microsatellites described here will be useful to explore both ecology and evolution of Sooty Swift and closely related species.

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We are grateful to R.G. Lima for collecting the specimen used to prospect the microsatellites, and C.Y. Miyaki for loaning the tissue sample under her care at LGEMA (*Laboratório de Genética e Evolução Molecular de Aves*, **Table 1.** Characterization of 21 microsatellite loci isolated from Cypseloides fumigatus and their cross-amplification inStreptoprocne zonaris

	Primer sequence (5'-3')	GenBank	Repeat	Cypseloides fumigatus						Sta	Streptoprocne zonaris		
Locus		accession No.	motif	Т _а (°С)	n_a	Size range (bp)	H	H	<i>P</i> -value	T _a (°C)	n_a	Size range (bp)	
Cyps3	F: TGCCCAGGGCTCTAAAAGTA	MF568530	(AG) ₅	58	2	282–286	0.032	0.032	-	NSA	-	-	
	R: GCCACAATAGCAGCACAGAA												
Cyps8	F: GGCTTGACCATGAGAACCAT	MF568531	(CT) ₅	60	3	107-129	0.029	0.189	0.0001*	60	1	121	
	R: CAACATTGTCCCTGTGATCG												
Cyps9	F: GGTGATGTCATTTCCCCTCT	MF579401	(AGC) ₅	NSA	-	-	-	-	-	52	2	76–97	
	R: TTAGAAAGTGCCAGAGAAGTATCA												
Cyps12	F: GAGGCTGCAGAAAAGCTGTC	MF579402	$(AG)_5$	58	3	179–193	0.118	0.112	1	58	2	179–181	
	R: ACCCTGCTGTTCAAGGTGTT												
Cyps14	F: AGGGGTGGAGATCAGACTCA	MF579403	(AC) ₅	58	3	128-142	0.125	0.420	0.0000*	NSA	-	-	
	R: AGTCCCTTTCTTCCCCTCTG												
Cyps20	F: CATGGCTTCCTCCTTTCTGT	MF579404	(TG) ₅	58	3	102-130	0.212	0.195	1	58	2	109–131	
	R: TGGGATGACTTGTTTCTCCTG												
Cyps22	F: CCCTCGTGACCATTTTCTGT	MF579405	(CT) ₅	58	2	203-207	0.065	0.062	1	58	1	203	
	R: GGTCACACAGAGGGGAAAAA												
Cyps23	F: CGGCTAAACTGCAAGGAAAA	MF579406	(GA) ₉	62	-	-	-	-	-	62	-	-	
	R: CCTATGGGCTGCTCTGCTAC		,										
Cyps24	F: GACAGAAGCCTTTCCAGTGC	MF579407	(CA) ₅	64	4	200-208	0.138	0.219	0.0239	64	-	-	
	R: TGAGACCGGAGCTGTCTTTT												
Cyps25	F: CATCTCCCAGGTGTTTTCGT	MF579408	(AG) ₅	56	3	232-240	0.067	0.065	1	56	2	228-232	
	R: AGTTGGGAAAAGAGCACAGC												
Cyps26	F: AGGAAAGAGCCCTCTGCAAT	MF579409	(TC) ₅	56	8	129–173	0.200	0.610	0.0000*	56	1	149	
71	R: TGGGGAGCAGAAGTAGCTGT		\$ 75										
Cvps27	F: AAATGCTGGCAAAGGTCTTG	MF579410	(TG) ₅	NSA	-	-	-	-	-	NSA	-	_	
71	R: CCGTGTCCCTCACTCAGACT		× 75										
Cvps28	F: CAAACATCTGCACCCCTTTT	MF579411	(GT) ₅	56	1	153	-	-	-	60	1	151	
-) F	R: CTGACACTCGGCACAGACAT		()5	20		- , , ,							
Cyps30	F: GATTCAATGGAGTAAATGGGTAG	MF579412	(AAT) ₅	56	4	229-241	0.071	0.103	0.0506*	NSA	-	_	
C) pos o	R: TGAAGGTCTAAAGCCTCCTCAG		(1211)5	20	-	22, 211	0.071	01105	0.0900	11011			
Cyps31	F: GCGATAATGGGTGGACACTT	MF579413	(TA) ₅	NSA	-	-	_	-	-	NSA	-	_	
0) pos r	R: GATCGCTCCTCCAAAATGTG		(11)5	1 (011						11011			
Cype32	F: GGAGTAGGAGCAGCACAAGC	MF579414	(GAG) ₅	NSA	_	_	_	_		NSA	_	_	
Cyp352	R: ATCAGACACTGAGGCCATCC	1011)/) 11 1	(0/10)5	1,071						1071			
Cyrpc33	F: TATTTCTTTTGGGGGTGCTG	MF579415	(TG) ₅	62	3	148–156	0.033	0 235	0.0000*	NSA			
CypsJJ	R: CACACTGTCAACCCACCTTG	IVII)/)+I)	(10) ₅	02	5	140-190	0.055	0.255	0.0000	140/1	-	-	
Cure 3/	F: GTCTGGGAACTGTCCCCTTT	MF579416	(TG) ₆	60						60	1	161	
Суряј4	R: AGACTGGGACCCAAGGATG	WII ⁻)/9410	(10) ₆	00	-	-	-	-	-	00	1	101	
Curra ²⁵		ME570/17	(CT)	50	4	1/0 155	0.560	0 671	0.0050*	50	1	1/0	
Cypsoo	F: GGCCAGTATTAATGAAGCAGATG	MF579417	(CT) ₉	58	4	149–155	0.560	0.671	0.0059*	58	1	149	
Carr -21	R: GCCTGTGGGGGCATTAAAGAT	ME570/10	$(C \Lambda)$	50	2	152 156	0 5 2 2	0 (20	0 2020	50	1	150	
Cyps36	F: GGGATGCCTACAGTGAAAGG	MF579418	$(GA)_5$	58	3	152–156	0.533	0.638	0.3839	58	1	152	
C 27	R: TTCTTGCCAGCAACTTTGAA	MEGGO (10		NTC 4						NTO 4			
Cyps37		MF579419	(AAAAC) ₈	INSA	-	-	-	-	-	NSA	-	-	
	R: CCAGCCACACCTTTTACTGC												

Forward (F) and reverse (R) primer sequence, T_a annealing temperature, NSA no successful amplification, n_a number of alleles, observed (H_o) and expected (H_c) heterozygosity, *P*-value of the Hardy-Weinberg equilibrium test. * Significant values, considering Benjamini & Yekutieli (2001) correction (P < 0.015).

	1 71	5 8		1		0 1
Family	Species	Number of loci	n _a	H。	H _e	Reference
Apodidae	Cypseloides fumigatus	13	2-8	0.156	0.254	This study
Trochilidae	Hylocharis xantusii	16	3–10	0.68	0.7	Sanvicente et al. 2016
Trochilidae	Hylocharis leucotis	14	2-8	0.35	0.41	Sanvicente et al. 2016
Trochilidae	Calypte costae	14	2–6	0.49	0.53	Sanvicente et al. 2016
Trochilidae	Campylopterus curvipennis	10	2–13	0.529	0.610	González et al. 2010
Trochilidae	Amazilia cyanocephala	10	2–13	-	-	Gutiérrez-Rodriguez et al. 2013
Trochilidae	Selasphorus platycercus	10	2–16	-	-	Oyler-McCance et al. 2011
Trochilidae	Trochilus spp.	15	2–10	-	-	Lance <i>et al.</i> 2009

Table 2. Comparison of the number of alleles ranges, mean observed (H_o) and expected (H_e) heterozygosities between microsatellite loci developed for *Cypseloides fumigatus* and that developed for some hummingbird species.

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A case of beak deformity in the Shiny Cowbird *Molothrus bonariensis* and a review on beak deformities in wild birds in Brazil

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ABSTRACT: Beak deformities in wild birds are rarely reported. Here, I described a case of beak deformity in Shiny Cowbird, *Molothrus bonariensis*, and also provide a review on beak deformities recorded in wild birds in Brazil. In October 2016, I observed a *M. bonariensis* with a grossly elongated maxilla in the east region of the Mato Grosso state, Brazil. The literature review revealed 60 records of beak deformities in the Brazilian avifauna. The most common types of deformity were crossed maxillae or mandibles (38%), probable accidental injuries (27%), and elongations (15%). Deformities were reported in 35 bird species of 22 families. The most affected species was *Ramphastos toco* with 21 records. The number of published reports from Brazil was low overall, and are not related as an epizootic episode recorded in some bird communities.

KEY-WORDS: avian keratin disorder, beak abnormality, bill deformity, crossed beak, elongated beak.

Beak deformities are rarely reported in wild birds, which may be due to the fact that they are infrequent (Pomeroy 1962, Craves 1994). Beak deformities may be either permanent or temporary, and are caused by a range of factors (Pomeroy 1962). The main causes are genetic mutations, injuries (*e.g.*, collisions with windows), diseases, nutritional deficiencies, contact with chemical pollutants (*e.g.*, agricultural pesticides), problems during incubation, and the inadequate wear of the rhinotheca (Pomeroy 1962, Craves 1994).

Most cases of beak deformity are records of isolated cases (Pomeroy 1962, Craves 1994). One notable exception is the case of the wild birds in Alaska, USA, in particular since the 1990s. In this region, beak deformities have been recorded in more than 2500 birds of 30 species (Handel et al. 2010, Van Hemert & Handel 2010). Recurrent sightings of birds with deformed beaks were also reported in the United Kingdom in subsequent years (Harrison 2011) and in the Patagonia, Argentina (Gorosito et al. 2016). Following these epizootic episodes, considerable research efforts have been invested in an attempt to identify the possible causes of these deformities, known as avian keratin disorder (Handel et al. 2010, Van Hemert & Handel 2010, Handel & Van Hemert 2015). The most recent evidence indicates that the deformities observed in Alaska are associated with a viral infection (Zylberberg et al. 2018). However, it is still unclear whether this virus is involved in the epizootic episodes recorded in other regions of the world.

In Brazil, despite its extensive geographical area, there has been little research on the occurrence of beak deformities in wild birds, and few cases have been reported (Sazima *et al.* 2016, Souza *et al.* 2016). In the present study, I described, most probably, the first report of a case of a beak deformity in a Shiny Cowbird *Molothrus bonariensis* for Brazil. I also present a review on the published records of beak deformities in Brazil, based on a comprehensive literature search.

I used the bibliography identified in Google Scholar (https://scholar.google.com) to make a compilation on beak deformities cases in wild birds in Brazil, and other specific bibliographic sources not indexed as scientific journals of restricted circulation to Brazil and technical books. The literature search was performed using multiple combinations of the words in English and Portuguese: avian keratin disorder, beak abnormality, beak deformity, beak deformities, bill deformity, crossed beak, elongated beak, bird, wild bird and Brazil.

On the afternoon of 11 October 2016, I briefly sighted a female *M. bonariensis* with a heavily deformed beak on the campus of Mato Grosso State University (14°41'25''S; 52°20'55''W), located within the Bacaba Municipal Park in Nova Xavantina, Mato Grosso state, Brazil. I sighted the female again at the same site on the following day, at around 15:00 h, local time. This individual was part of a flock of five *M. bonariensis*. On this occasion, I could confirm that the bird had a grossly elongated maxilla (Fig. 1). In addition to being elongated (approximately three times longer than the normal length of the beak), the maxilla was quite curved downward, and had some wear at the extremity. I also observed a contrast in the coloration of the beak, which was greyish black (the standard color) at the base and brown in the elongated portion of the rhinotheca. Despite this deformity, the bird appeared to be healthy, with well-groomed plumage, and similar in size to other adult females. During the short period that I was able to monitor the bird (-4 min), it captured food in a distinct manner in comparison with the other members of the flock. I observed the bird tilting its head to one right side and using the base of its beak to catch insects (apparently ants) in the grass, while the other members foraged normally. Individual adaptations in feeding behavior have been observed in birds of other species with deformed beaks (Pomeroy 1962, Van Hemert *et al.* 2012).

I found 60 recorded cases of beak deformities in wild birds for Brazil from 11 publications (Table 1). Deformities were recorded in 35 bird species, belonging to 22 families, of which 11 families were Passeriformes. Ramphastidae and Thraupidae had the highest number of species with deformities, with 4 each, followed by Thamnophilidae, with 3. The species with the most records were the Toco Toucan *Ramphastos toco* (n = 21) and the Picazuro Pigeon *Patagioenas picazuro* (n = 3). With the

Table 1. Cases of beak deformities in wild birds registered in Brazil.

Taxon	Type of deformity	Municipality-state	Date	Source
Sulidae				
Sula dactylatra	Curved beak	Mostardas-RS	28 February 2006	Franz <i>et al</i> . 2008
Sula leucogaster	Unspecified	Paraná state	1995	Straube 1996
Ardeidae				
Ardea alba	Shortening of beak - accidental	Araxá-MG	16 March 2013	Souza <i>et al</i> . 2016
Cathartidae				
Coragyps atratus	Short and deformed upper mandible; lower mandible with fractured extremity - accidental	Colinas do Tocantins-TO	27 December 2010	Souza <i>et al.</i> 2016
Rallidae				
Aramides saracura	Crossed upper mandible	Palmeira das Missões-RS	13 May 2016	Santos <i>et al.</i> 2018
Columbidae				
Patagioenas picazuro	Upper mandible crossed	Luz-MG	May–June 2012	Vitorino & Souza 2013
	Upper mandible strongly twisted to the left	Campinas-SP	09 August 2011	Sazima <i>et al</i> . 2016
	Upper mandible strongly twisted to the left	Campinas-SP	28 April/03 May 2015	Sazima <i>et al</i> . 2016
Trochilidae				
Clytolaema rubricauda	Atrophy of upper mandible	Quatro Barras-PR	28 May 1987	Straube 1996
	Lower mandible distinctly curved to the left	Rio de Janeiro-RJ	23–24 April 2015	Sazima <i>et al</i> . 2016
Amazilia versicolor	Atrophy of lower mandible	Morretes-PR	16 November 1986	Straube 1996
Galbulidae				
Galbula ruficauda	Crossed upper and lower mandible	Itiquira-MT	13 February 2013	Souza <i>et al</i> . 2016
	Crossed upper and lower mandible	Goiânia-GO	03 October 2015	Souza <i>et al.</i> 2016
Ramphastidae				
Ramphastos toco	Malformation at the extremity of the upper mandible	Brasília-DF	08 September 2007	Souza <i>et al.</i> 2016

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Taxon	Type of deformity	Municipality-state	Date	Source
	Crossed upper and lower mandible	Batayporá-MS	26 July 2009	Souza <i>et al</i> . 2016
	Absence of the upper mandible tip - accidental	Corumbá-MS	05 September 2009	Souza <i>et al</i> . 2016
	Upper mandible crossed	Poços de Caldas-MG	06 November 2009	Souza <i>et al.</i> 2016
	Fissure at the tip of the upper mandible - accidental	Caetanópolis-MG	07 January 2010	Souza <i>et al</i> . 2016
	Absence of the lower mandible tip - accidental	Goiânia-GO	01 April 2010	Souza <i>et al</i> . 2016
	Upper mandible crossed	Uberaba-MG	04 June 2010	Souza <i>et al</i> . 2016
	Absence of the upper mandible tip - accidental	Peruíbe-SP	30 April 2011	Souza <i>et al.</i> 2016
	Absence of the lower mandible tip - accidental	São Roque de Minas-MG	10 June 2011	Souza <i>et al.</i> 2016
	Lower mandible elongated; absence of the upper mandible tip	Araxá-MG	06 April 2012	Souza <i>et al</i> . 2016
	Upper mandible crossed	Joanópolis-SP	30 April 2013	Souza <i>et al</i> . 2016
	Absence of approximately half of the upper mandible - accidental	Bonito-MS	12 December 2013	Souza <i>et al</i> . 2016
	Part of deformed upper mandible - accidental	Mundo Novo-MS	24 March 2014	Souza <i>et al</i> . (2016)
	Part of deformed upper mandible - seems to burnt	Três Corações-MG	28 June 2014	Souza <i>et al</i> . 2016
	Absence of the upper mandible tip - accidental	Mineiros-GO	30 December 2014	Souza <i>et al</i> . 2016
	Absence of approximately half of the lower mandible - accidental	Araçatuba-SP	17 February 2015	Souza <i>et al</i> . 2016
	Absence of the upper mandible tip - accidental	Campo Grande-MS	02 May 2015	Souza <i>et al</i> . 2016
	Absence of the upper mandible tip - accidental	Niquelândia-GO	04 July 2015	Souza <i>et al</i> . 2016
	Perforation of the upper mandible - accidental	Natalândia-MG	30 December 2015	Souza <i>et al</i> . 2016
	Malformation at the extremity of the upper mandible	Araçoiaba da Serra-SP	08 February 2016	Souza <i>et al</i> . 2016
	Malformation at the extremity of the upper mandible	Campo Belo-MG	13 March 2013	Rezende 2013
Ramphastos tucanus	Absence of the upper mandible tip - accidental	Comodoro-MT	06 November 2011	Souza <i>et al</i> . (2016)
Ramphastos dicolorus	Absence of the lower mandible tip - accidental	Mairinque-SP	25 May 2016	Souza <i>et al</i> . 2016
Pteroglossus aracari	Upper mandible crossed	Porciúncula-RJ	21 June 2013	Souza <i>et al</i> . 2016
Picidae				
Melanerpes candidus	Upper mandible elongated; absence of the upper mandible tip	Sacramento-MG	31 May 2014	Souza <i>et al</i> . 2016

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Taxon	Type of deformity	Municipality-state	Date	Source
Falconidae				
Falco femoralis	Upper mandible elongated; crossed lower mandible	Goiânia-GO	25 August 2013	Souza <i>et al</i> . 2016
Psittacidae				
Thectocercus acuticaudatus	Mandible vestigial	Canudos-BA	06 April 2016	Souza <i>et al.</i> 2016
Myiopsitta monachus	Upper mandible elongated	São Roque-SP	23 May 2011	Souza <i>et al.</i> 2016
	Upper mandible elongated	Poconé-MT	07 June 2011	Souza <i>et al.</i> 2016
Thamnophilidae				
Thamnophilus torquatus	Upper mandible crossed	Santo Antônio do Monte- MG	26 May 2012	Souza <i>et al.</i> 2016
Pyriglena leucoptera	Upper mandible crossed	São Miguel Arcanjo-SP	18 March 2011	Gallo-Ortiz 2011
Percnostola rufifrons	Lower mandible elongated	Curuá-PA	22 February 2012	Souza <i>et al</i> . 2016
Furnariidae				
Furnarius rufus	Upper mandible strongly twisted to the right	Campinas-SP	08 July 2010	Sazima <i>et al</i> . 2016
Pipridae				
Ilicura militaris	Upper mandible crossed	Nova Lima-MG	15 October 2015	Souza <i>et al.</i> 2016
Tityridae				
Pachyramphus polychopterus	Beak more short and curved to the side	Bocaiúva-MG	24 November 1998	Vasconcelos & Rodrigues 2006
Corvidae				
Cyanocorax cristatellus	Upper mandible crossed	Alto Paraíso de Goiás-GO	24 November 2012	Darosci 2017
Troglodytidae				
Cyphorhinus arada	Extremity of beak side- facing	Unspecified	Unspecified	Sick 1997
Turdidae				
Turdus leucomelas	Unspecified	Bocaiúva-MG	Unspecified	Vasconcelos & Rodrigues 2006
Turdus rufiventris	Upper mandible elongated	São Paulo-SP	14 September 2013	Souza <i>et al</i> . 2016
Mimidae				
Mimus saturninus	Lower mandible deflected to left side	Engenheiro Coelho-SP	15 June 2011	Souza <i>et al.</i> 2016
Icteridae				
Cacicus haemorrhous	Crossed upper and lower mandible	Aracruz-ES	31 August 2013	Souza <i>et al.</i> 2016
Chrysomus ruficapillus	Upper mandible crossed	Santo Antônio do Monte- MG	23 February 2014	Souza <i>et al.</i> 2016
Thraupidae				
Tangara sayaca	Upper mandible elongated	Piraju-SP	17 July 2010	Souza <i>et al.</i> 2016
Tangara palmarum	Fissure of the lower mandible	Porto Velho-RO	03 October 2012	Souza <i>et al.</i> 2016
Tangara ornata	Upper mandible crossed	Ubatuba-SP	16 December 2010	Souza <i>et al.</i> 2016
Sporophila nigricollis	Fissure in rhinotheca	Conceição do Mato Dentro- MG	24 November 2013	Souza <i>et al.</i> 2016
Fringillidae				
Euphonia violacea	Upper mandible elongated	Ubatuba-SP	02 May 2016	Souza <i>et al</i> . 2016

exception of a chick Brown Booby *Sula leucogaster* and a juvenile Masked Booby *Sula dactylatra*, all birds affected by deformities were adults.

Based on the published photographs and the case descriptions, I was able to determine that 38% of the cases of beak deformity involved crossed beaks, due to some type of deviation of the maxilla or mandible. A further 27% of the cases appeared to be the result of accidental injuries. This type of deformity was found primarily in three species of toucans, which can be explained by the large size of the beak of these species. First, in theory, the large beak of toucans probably becomes more vulnerable to accidental fractures. Second, a large, colorful beak provides more visual conspicuity, which facilitates field registration in relation to the other bird species. Birds with elongated beaks corresponded to 15% of the records. Altogether, other types of deformity contributed with 17% of the records, while the deformity was not described specifically in the cases of Brown Booby S. leucogaster and Pale-breasted Thrush Turdus leucomelas (Table 1).

Despite the large number of icterid species found in Brazil, beak deformities have been recorded in only three taxa, including the present case. Recently, a case of crossed beak was recorded for *M. bonariensis* in Argentina (Bianchini & Arenas 2018). I evaluated the photographic record of this case and observed that, in addition to the crossed beak, at least the upper mandible was also elongated. Apparently, this record and that of the present study are the only two records of beak deformity in *M. bonariensis*. In both cases, the elongation of the upper mandible and the difference in color of the elongated part of the beak is notable. Considering only the two isolated cases, the similarities found do not reveal much at the moment, but can serve as a basis for possible future observations.

As in most of cases recorded in Brazil, it was not possible to monitor the specimen over a long period to determine whether the deformity was permanent or temporary, nor to collect it for analysis. At first, it seems unlikely that this case of beak elongation, or any of the others recorded in Brazil, has an etiological origin similar to the alarming cases of beak deformity recorded in Alaska (Handel et al. 2010, Van Hemert & Handel 2010). In fact, the number of published reports from Brazil were low overall, and the events did not appear to be related to epizootic outbreaks. However, beak deformities are more common than suggested by the literature, given that not all cases are reported in scientific publications (Vasconcelos & Rodrigues 2006). Moreover, without systematized research efforts on the subject this issue cannot be elucidated. Laboratory analyses, the long-term monitoring of affected individuals, even in a small number of cases, and formal publication of records may help to better clarify the causes and consequence of beak deformities in birds. These approaches will contribute to the detection of possible threats to the wild bird fauna.

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Figure 1. Female Shiny Cowbird *Molothrus bonariensis* with beak elongated (**A**) and comparison with a male with normal beak (**B**) in Nova Xavantina, Mato Grosso state, Brazil. Photo author: K.N. Purificação.

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Semipalmated Sandpiper *Calidris pusilla* in Brazil: occurrence away from the coast and a new record for the central-west region

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ABSTRACT: The Semipalmated Sandpiper, *Calidris pusilla*, is a Western Hemisphere migrant shorebird for which Brazil forms an internationally important contranuptial area. In Brazil, the species main contranuptial areas is along the Atlantic Ocean coast, in the north and northeast regions. In addition to these primary contranuptial areas, there are also records of vagrants widely distributed across Brazil. Here, we review the occurrence of vagrants of this species in Brazil, and document a new record of *C. pusilla* for the central-west region and a first occurrence for the state of Goiás.

KEY-WORDS: geographical distribution, Nearctic migrant, shorebird, state of Goiás, vagrant.

The Semipalmated Sandpiper *Calidris pusilla* (Linnaeus, 1766) is a migratory shorebird species that breeds in the Arctic and sub-Arctic regions of Alaska and Canada (Andres *et al.* 2012, IUCN 2019). Every year, as the northern autumn approaches, Arctic populations fly from 3000 to 4000 km to South America (Hicklin & Gratto-Trevor 2010).

In Brazil, *C. pusilla* occurs during all months of the year, but with very few records during the boreal winter (eBird 2019, GBIF 2019, SNA 2019, WikiAves 2019). This species uses the Atlantic route, occurring from the coastal zone of the state of Amapá to the state of Rio Grande do Sul, where the main contranuptial areas with population concentrations of this bird are located along of the Reentrâncias Paraenses and Maranhenses and in the states of Amapá and Pernambuco (Rodrigues 2007, Carvalho & Rodrigues 2011, Rodrigues *et al.* 2015, SNA 2019, GBIF 2019). In the southeast and south regions, only a small population of this species is observed (Harrington *et al.* 1986, Resende *et al.* 1989, Barbieri *et al.* 2013, eBird 2019, GBIF 2019, WikiAves 2019).

In addition to the primary contranuptial areas mentioned above, there are also records of vagrants widely distributed inland in Brazil (Table 1). Based on the available information, five previous records of *C. pusilla* were found for the central-west region, of which three were obtained in the municipality of Poconé, state of Mato Grosso (Cintra 2011, Levatich & Padilha 2019) and two in the municipality of Corumbá, state of Mato Grosso do Sul (Serrano 2010, Tubelis & Tomas 2003). However, there is no evidence that these records have been correctly identified, as individuals appear not to have been collected and sent to a scientific collection, nor are images available to validate records.

On 11, 12 and 13 November 2018, an adult specimen of C. pusilla was observed in an area denominated "Lago da Piscicultura Frutos D'Água" in the municipality of Goiânia, state of Goiás, Brazil (16°34'25''S; 49°18'48''W, 719 m a.s.l.). During three consecutive days, C. pusilla was sighted in the late afternoons, foraging most of the time along with a maximum of four White-rumped Sandpipers Calidris fuscicollis (Vieillot, 1819) and with two Solitary Sandpipers Tringa solitaria Wilson, 1813 (Fig. 1). Very similar to the congeneric C. mauri and C. minutilla, which were not present at the site, the C. pusilla individual was safely identified through high-quality photographic records. Compared to C. minutilla, its size was larger, with gray upper plumage and black legs, thick beak with absence of slight (Barnett et al. 2004, Lees et al. 2013). In contrast to C. mauri, the head was smaller and less frontal in shape, the beak had a thick tip and the center of the chest was not striated (Barnett et al. 2004, eBird 2019). Digital vouchers were uploaded to WikiAves (WA3179032).

Table 1. Review of the occurrence of vagrant *Calidris pusilla* in Brazil. Location: state, municipality, place descriptionand geographical coordinates - latitude (S), longitude (W). Verifiable evidences: P – photograph, S – preserved specimen,R – recovery of marked individual, V – visually observed; - no specimen available. n = Number of individuals. Specimens:number of tumble and their respective collections (CLO:EBIRD – eBird, Cornell Lab of Ornithology; UNICAMP:FNJV– Fonoteca Neotropical "Jacques Vielliard"; WA – WikiAves).

Location	Date	Verifiable evidences	n	Specimens	Reference
Amazonas, Manaus, little lake in Careiro	29 March 1987	V	1	-	Stotz <i>et al.</i> 1992
Amazonas, Iranduba, Marchantaria Island	16 February 1989	V	8	-	Stotz <i>et al.</i> 1992
Pará, Terra Santa, Mexiana Island (1º57'S; 56º21'W)	-	-	1	-	Hellmayr & Conover 1948 <i>apud</i> Serrano 2010
Tocantins, Itacajá, near the municipality of Recursolândia	31 October 2000	V, R	1	-	CEMAVE 2007 <i>apud</i> Serrano 2010
Maranhão, Timon, crown of the Paranaíba River	2 November 2017	V, P	2	WA2765260	Galvão 2017
Ceará, São Benedito, Ibiapaba (4º00'S; 41º00'W)	08 January 1929	-	1	-	Hellmayr & Conover 1948 <i>apud</i> Serrano 2010
Ceará, Sobral, Lagoa das Marrecas (3°41'15.8''S; 40°19'51.3''W)	13 October 2018	V	11	CLO:EBIRD:OBS666659024	Levatich & Padilha 2019
Pernambuco, Petrolina (9°24'18.1''S; 40°32'10.0''W)	15 February 2003	V	1	CLO:EBIRD:OBS72752146	Levatich & Padilha 2019
Pernambuco, Serra Talhada, Açude do Saco	08 December 1973	S	1	UNICAMP:FNJV:000000656	Toledo 2019
Bahia, Juazeiro, Caatinga Forest east (9°30'52.4''S; 40°07'23.6''W)	15 February 2003	V	2	CLO:EBIRD:OBS471407770	Levatich & Padilha 2019
Bahia, Mata de São João, Sapiranga Reserve (12°34'04.6''S; 38°02'14.8''W)	25 January 2013	V	1	CLO:EBIRD:OBS499778311	Levatich & Padilha 2019
Bahia, Santa Rita de Cássia, Preto River (11º00'S; 44º32'W)	04 May 1910	-	1	-	Hellmayr & Conover 1948 <i>apud</i> Serrano 2010
Santa Catarina, Santa Rosa de Lima, Pousada Doce Encanto (28°02'00.2''S; 49°09'00.9''W)	18 November 2017	V	1	CLO:EBIRD:OBS550251697	Levatich & Padilha 2019
Mato Grosso, Poconé, Pantanal of Poconé	between 1982 and 1986	-	-	-	Cintra 2011
Mato Grosso, Poconé, Pousada Curicaca (16°30'16.0''S; 56°40'30.7''W)	11 September 2013	V	1	CLO:EBIRD:OBS224967965	Levatich & Padilha 2019
Mato Grosso, Poconé, Porto Jofre (17°21'52.3''S; 56°46'30.4''W)	04 September 2017	V	1	CLO:EBIRD:OBS668324309	Levatich & Padilha 2019
Mato Grosso do Sul, Corumbá, Nhecolândia in Campinas Farm	October 1988 and October 1989	V	-	-	Serrano 2010
Mato Grosso do Sul, Corumbá, Nhumirim Farm (18°59'S; 56°39''W)	between 1991 and 1992	V	-	-	Tubelis & Tomas 2003



Figure 1. (A) Adult of *Calidris pusilla* foraging next to adult individual of *Tringa solitaria*, on 11 November 2018; (B) *C. pusilla* foraging next to an individual of *Calidris fuscicollis*, on 12 November 2018; (C) *C. pusilla* observed on 13 November 2018. All records made in Goiânia, Goiás state, Brazil. Photo author: J.A. Oliveira.

We reviewed the records of vagrants of *C. pusilla* in Brazil and found five previous observations for central Brazil, but there are not enough evidences to confirm the correctness of the identification of these records (Table 1). Thus, here we document a new occurrence of *C. pusilla* for the central-west region and a first record for the state of Goiás.

A bird of international conservation concern, *C. pusilla* is classified as "Near Threatened", due to ongoing population declines (IUCN 2019) linked to several physiological, ecological and human-related challenges, such as: species needs to perform periodic scales for resting and foraging; good habitat conditions in both breeding and contranuptial areas; availability of food resources; competition in foraging environment; predation pressure; human disturbances (Skagen 2006, Burger *et al.* 2007, IUCN 2019).

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