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**AVALIAÇÃO DA DENSIDADE POPULACIONAL E SELEÇÃO DE
MICROHABITATS EM PASSERIFORMES FLORESTAIS NA RPPN MATA DO
CEDRO, RIO LARGO, AL, CENTRO DE ENDEMISMO PERNAMBUCO**

São Paulo- SP, 2021

Luiza Carvalho Prado

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Dissertação apresentada ao Programa de Pós-Graduação em Conservação da Fauna da Universidade Federal de São Carlos como parte dos requisitos exigidos para a obtenção do título de Mestre em Conservação da Fauna.

Orientador: Mercival Roberto Francisco

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Resumo

O Centro de Endemismo Pernambuco (CEP) é uma área de Mata Atlântica com altos níveis de biodiversidade devido a fatores biogeográficos e históricos. Localizado no Nordeste do Brasil, ao norte do Rio São Francisco, o CEP é considerado uma das regiões mais ameaçadas do planeta, sendo a área de Mata atlântica mais fragmentada e degradada de todo o bioma. O CEP abriga uma série de espécies endêmicas e ameaçadas de extinção, algumas ainda em processo de descrição. A classificação do grau de ameaça da maioria das espécies do CEP em listas de espécies ameaçadas de extinção levou em consideração apenas as distribuições geográficas e as áreas de ocorrência dos taxa. Embora estimativas de densidade e de tamanhos populacionais sejam importantes para estas classificações, estes dados são inexistentes para estes animais. Estas estimativas, quando associadas a estudos de seleção e preferência de habitat geram ainda informações capazes de melhor direcionar recursos e esforços para a conservação. Neste trabalho foram realizados censos populacionais em um dos fragmentos de maior representatividade do Estado de Alagoas, através da metodologia de *Distance sampling* por transectos lineares, de cinco passeriformes florestais, ameaçados de extinção e endêmicos do CEP: o Papa-taoca-de-pernambuco *Pyriglena pernambucensis* (Vulnerável/VU), o Chupadente-de-máscara-preta *Conopophaga melanops nigrifrons* (VU), a Choca-da-mata *Thamnophilus aethiops distans* (Em perigo/ EN), o Flaltim-marrom *Schiffornis turdina intermedia* (VU), e a Maria-de-barriga-branca *Hemitriccus griseipectus naumburgae* (VU). Apesar de quatro dos cinco táxons estarem incluídos na mesma categoria de ameaça, as densidades populacionais encontradas variaram entre 0.13 e 0.73 indivíduos por hectare, sendo a espécie com menor densidade *P. pernambucensis* e maior *H. griseipectus naumburgae*. Através de extrapolações das densidades observadas foi possível produzir estimativas de tamanho populacional mínimo para as espécies estudadas. Dois destes táxons também tiveram suas preferências de microhabitat analisadas (*P. pernambucensis* and *C. m. nigrifrons*). Para ambos, características específicas relacionadas à vegetação do sub-bosque florestal foram selecionadas, demonstrando a importância da manutenção da heterogeneidade deste tipo de ambiente.

Palavras-chave: Avifauna, Biodiversidade, Conservação, Espécies Ameaçadas, Mata Atlântica.

Abstract

The Pernambuco Endemism Center (PEC) is an area of Atlantic Forest with high biodiversity levels due to biogeographic and historical factors. Located in Northeastern Brazil, north of the São Francisco River, the PEC is considered one of the most threatened regions on earth, being the most fragmented and degraded Atlantic Forest area in the entire biome. The PEC houses a series of endemic and endangered species, some of which are still in the process of being described. The classification of the levels of threat of the animals from the PEC in red lists have been based solely on their geographic distribution and areas of occurrence, especially because data on population density and population size estimates are unavailable. These estimates, when associated with studies on habitat selection and preference, can provide conservation managers with important information, useful to decide where and how to direct resources and efforts for conservation. In this work, population censuses estimates were carried out for five passerine species in one of the most representative PEC fragments from the state of Alagoas, using Distance sampling linear transects. All of the species are threatened with extinction and endemic to the PEC: the Pernambuco Fire-eye *Pyriglena pernambucensis* (VU), the Black-cheeked Gnat-eater *Conopophaga melanops nigrifrons* (VU), the White-shouldered Antshrike *Thamnophilus aethiops distans* (EN), the Brown-winged Mourner *Schiffornis turdina intermedia* (VU), and the White-bellied Tody-tyrant *Hemitriccus griseipectus naumburgae* (VU). Although four of the five taxa are included in the same threat category, population densities varied from 0.13 to 0.73 individuals per hectare, being *P. pernambucensis* the taxa with the lowest, and *Hemitriccus g. naumburgae* the species with highest population density estimate. Through extrapolation of the observed densities, it was possible to produce minimum population sizes estimates for these birds. Two of these taxa, belonging to the forest understory insectivorous passerines, also had their microhabitat preferences analyzed. For both, microhabitat selection was based on characteristics of the forest understory vegetation, indicating the importance of microhabitat heterogeneity.

Keywords: Avifauna, Atlantic Forest, Biodiversity, Conservation, Endangered species.

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AOO	Area of Occupancy/ Área de Ocupação
CEP	Centro de Endemismo Pernambuco
EN	Em Perigo de extinção
ICMBio	Instituto Chico Mendes de Conservação da Biodiversidade
IUCN	<i>International Union for Conservation of Nature</i>
DS	<i>Distance Sampling</i>
MMA	Ministério do Meio Ambiente
ONGs	Organizações Não Governamentais
PEC	Pernambuco Endemism Centre
VU	Vulnerável

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1. Introdução:

1.1. A Mata Atlântica e o Centro de Endemismo Pernambuco

A Mata Atlântica é um dos principais biomas de ocorrência em território brasileiro, distribuindo-se por toda a região leste do país e estendendo-se, ainda, por território Argentino e Paraguai (MMA, S/A). Este bioma vem sendo amplamente modificado ao longo da história e após mais de 500 anos de exploração, estimativas indicam que restam apenas entre 11.4 a 16% dos 150 milhões hectares originais de mata (Ribeiro et al, 2009). Dados como estes configuram a Mata Atlântica como um dos biomas que mais sofreu e ainda sofre destruição e fragmentação a nível mundial, e por consequência, com maior número de espécies ameaçadas de extinção (ICMBio, 2018). Apesar disso, a Mata atlântica ainda se mantém como uma das áreas de maior diversidade biológica do planeta (Marsden et al, 2005; Tabarelli et al, 2005), sendo, no Brasil, o bioma com a segunda maior diversidade de espécies, atrás apenas da Amazônia (ICMBio, 2018).

A grande diversidade biológica na Mata Atlântica pode ser explicada pela combinação de múltiplos fatores como: as amplas variações latitudinais (6 a 32° S) e altitudinais (nível do mar até 1.700 m) ao longo de seu território, que implicam na formação de variadas paisagens com múltiplas graduações climáticas e formações vegetais, propiciando um ambiente capaz de abrigar uma grande diversidade de espécies tanto de fauna quanto de flora (Silva et al, 2004). As peculiaridades deste bioma favoreceram, então, a existência de altas taxas de endemismo ao longo de sua área de ocorrência, classificando, biogeograficamente, algumas de suas regiões como centros de endemismo, onde diversas espécies encontram ambientes específicos ideais para sua sobrevivência (Silva et al, 2004). Esses aspectos caracterizam a floresta atlântica como um *hotspot*, tornando-a alvo prioritário, a nível mundial, de diversos programas de conservação (Myers et al, 2000; Tabarelli et al, 2005; Tabarelli & Roda, 2005).

O Centro de Endemismo Pernambuco (CEP) é um dos quatro centros de endemismo atualmente reconhecidos em território brasileiro, e faz referência a área de Mata Atlântica no Nordeste do território brasileiro, ao norte do Rio São Francisco, distribuída entre os Estados de Alagoas, Pernambuco, Paraíba e Rio Grande do Norte (Silva et al, 2004; Figura 1). No passado, o CEP reunia cerca de 56.000 km² de áreas de mata, porém, estima-se que atualmente cerca de apenas 2% desta área original ainda seja composta por florestas, o que caracteriza o CEP como um dos ambientes a nível mundial que mais sofreu degradação ao longo do tempo, estando entre as regiões mais ameaçados do mundo (Coimbra-filho & Câmara, 1996; Lima & Capobianco 1997; Ranta et al, 1998; Silva & Tabarelli, 2001).

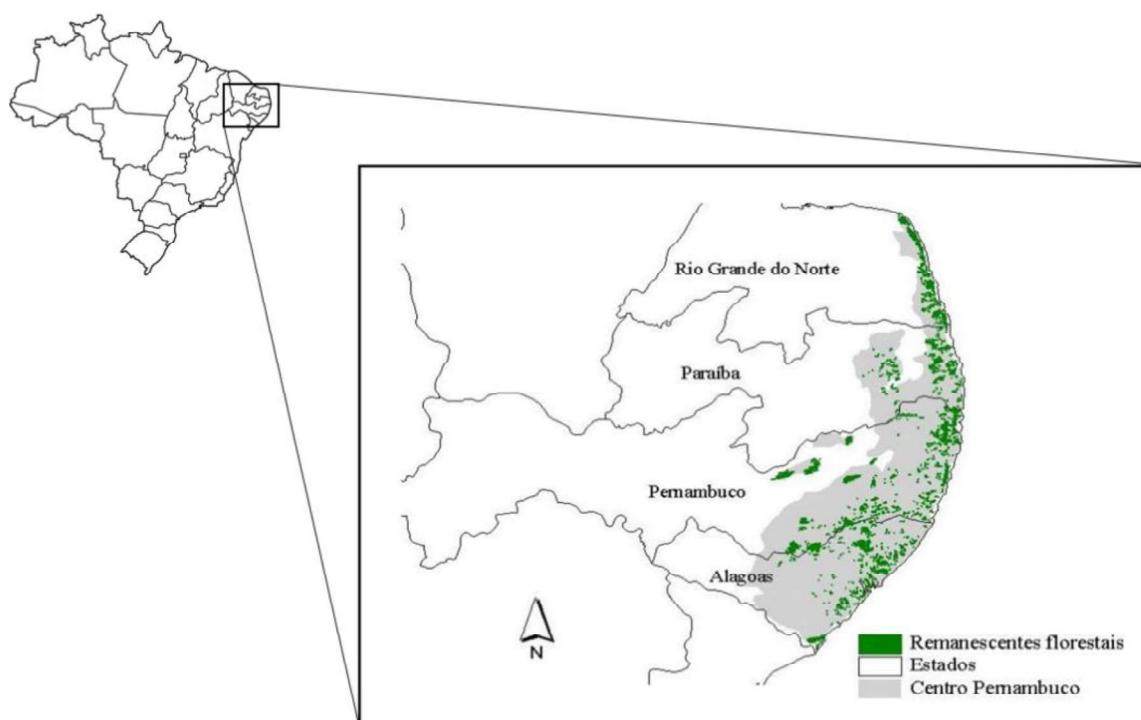


Figura 1. Localização do Centro de Endemismo Pernambuco, ilustrando sua distribuição original (cinza) e remanescentes florestais (verde). Figura modificada de Neto e Tabarelli (2002).

O pouco que ainda sobrou das áreas de mata do CEP encontra-se distribuído entre os 4 Estados citados na forma de fragmentos florestais dos mais variados tamanhos, compondo uma verdadeira paisagem de mosaicos (Neto & Tabarelli, 2002; Figura 1). As causas associadas ao

grande desmatamento e fragmentação da região são originárias desde a colonização do Brasil, no entanto foram acentuadas entre as décadas de 1970 e 80, através da implementação por parte do Governo Federal do programa Proálcool, que buscava uma alternativa mais acessível ao uso do petróleo como fonte de combustível, e maior parte das florestas da região foram substituídas por áreas de cultivo de cana-de-açúcar (Silveira et al, 2003; Pereira et al, 2014). A iniciativa perdura até os dias de hoje, de modo que a maior parte das áreas de mata remanescentes da região está direta ou indiretamente ligada a propriedades privadas, especialmente envolvidas com a produção industrial de cana-de-açúcar a nível nacional e internacional

Ainda assim, o CEP é capaz de abrigar 435 espécies de aves, sendo 18 espécies e 21 subespécies endêmicas à região (Silveira et al, 2004; Pereira et al, 2014; Barnett & Buzzetti, 2014, ICMBio, 2018b) e apresenta novas espécies ainda sendo descritas (Silveira et al, em preparação; Dickens et al, 2021). No entanto, os processos aos quais o CEP foi submetido possuem impactos significativamente negativos na fauna local, tendo causando a extinção de quatro espécies de aves endêmicas nas últimas décadas (*Pauxi mitu*, *Glaucidium mooreorum*, *Cichlocolaptes mazarbarnetti* e *Philydor novaesi*) (Silva et al, 2004; Roda et al, 2011; Pereira et al, 2014) e tornando o CEP a região com o maior número de espécies de aves ameaçadas de extinção em todo o Brasil, de acordo com a última Lista Vermelha de Fauna Brasileira Ameaçada de extinção publicada pelo ICMBio (2018a e b). Como fator agravante, tem-se ainda cerca de 65% dessas espécies com ocorrência exclusiva em ambientes florestais (Roda, 2003), apontando a dependência de diversas espécies a áreas que se apresentam cada vez mais escassas e degradadas na região.

Esses fatos demonstram a urgência do desenvolvimento de pesquisas focadas no CEP e sua biodiversidade, visando não só impedir novas extinções, mas também identificar, listar, classificar e compreender necessidades ambientais básicas de todas as espécies de ocorrência

da área, evitando que outras espécies sejam perdidas, e permitindo que ações para a conservação das espécies endêmicas do CEP sejam propostas de forma adequada.

1.2. Censos populacionais e espécies ameaçadas de extinção

Para o grupo das aves, trabalhos de alta qualidade tem sido produzidos, apresentando bons levantamentos nos fragmentos de maiores dimensões, principalmente nos estados de Alagoas, Pernambuco e Paraíba (e.g. Roda et al, 2011; Pereira et al, 2014). Com isso, a listagem das espécies de ocorrência, incluindo espécies ameaçadas, já é relativamente bem conhecida e áreas prioritárias para conservação já foram identificadas (Pereira et al, 2016; BirdLife International, 2021). No entanto, diversos fragmentos nos Estados de Alagoas e Rio Grande do Norte ainda não foram sequer inventariados, e consequentemente dados associados a áreas de ocorrência, estimativas populacionais e fatores que determinam a permanência e diversidade de espécies na região continuam poucos conhecidos.

O status de conservação o qual a grande maioria das espécies de ave da região está classificada, assim como maior parte da fauna brasileira, está associado ao critério B utilizado pela IUCN e ICMBio para classificação do risco relativo de extinção das espécies presentes na lista vermelha, que é referente à distribuição geográfica (Anexo 1). Isso demonstra que a classificação atual de muitas das espécies presentes no CEP não leva em consideração fatores como o tamanho das populações e a densidade populacional em suas respectivas áreas de ocorrência, bem como flutuações desses valores, que também são critérios de avaliação utilizados pela IUCN e ICMBio (Critérios A C e D) (ICMBio, 2018a; IUCN, 2019) (Anexo 1). No contexto do CEP, onde a ação antrópica ainda é intensa e a degradação e fragmentação da área são evidentes, é esperado que haja flutuações nas populações, inclusive com populações apresentando declínio contínuo há décadas. Esses dados demográficos, ainda pouco conhecidos

para muitas das espécies da fauna brasileira no geral, além de serem um recurso que permite a avaliação do estado de conservação de uma espécie, possibilitando sua categorização dentro dos critérios da IUCN e ICMBio, se analisados de forma contínua, também permitem monitorar o tamanho das populações ao longo do tempo, comparar populações da mesma espécie e espécies diferentes dentro das mesmas áreas e até áreas diferentes, bem como averiguar a viabilidade das populações no futuro, além de permitir a mensuração de impactos antrópicos, como poluição, caça e perda de habitat (Bibby et al, 2000).

Existem diferentes metodologias descritas para a obtenção de dados relacionados a estimativas populacionais de animais vida livre (Bailey, 1951; Mares et al, 1981; Buckland et al, 1993; Buckland et al, 2001; Matter et al, 2010). A escolha da metodologia a ser empregada deve ser baseada em fatores diretamente relacionadas aos hábitos da espécie (ou espécies) alvo do estudo, tipo de ambiente onde é encontrada e experiência do pesquisador. O processo de determinação da densidade de uma população e a seu respectivo tamanho populacional podem parecer simples, mas contar organismos pode ser altamente complexo. O grande desafio é garantir que os dados coletados em condições adversas do campo se enquadrem em exigências mínimas das análises estatísticas, de modo que é imprescindível também, que as análises levem em consideração as dificuldades enfrentadas pelos pesquisadores para uma estimativa fidedigna (Bibby et al, 2000).

Distance sampling (DS) é uma das principais metodologias utilizadas para a produção de dados relacionados à abundância de populações. O DS engloba uma série de métodos baseados na detecção de objetos-alvo ao longo de trilhas (transectos lineares) ou pontos fixos aleatoriamente distribuídos dentro da(s) área(s) amostradas e possui 3 premissas chave: (a) os objetos de estudo são identificados com convicção, (b) os objetos de estudo são detectados em seu ponto inicial (antes de se mover em relação à presença do observador), (c) as distâncias são

mensuradas com precisão, mas que podem não ser tão rígidas em alguns casos (Buckland et al, 1993; Buckland et al, 2001; Buckland et al, 2008). O observador é então responsável por registrar as distâncias perpendiculares entre ele e o(s) objeto(s) de estudo e através de uma série de cálculos e modelos, é capaz de encontrar uma probabilidade de detecção que produzirá uma estimativa de densidade.

Sua relativa simplicidade e capacidade de produzir dados de alta confiabilidade, permitem que seja empregada em diferentes ambientes e com diversas espécies, inclusive em situações desafiadoras, sendo a principal metodologia empregada para realização de estudos de estimativas populacionais em espécies selvagens no mundo todo (Buckland & Anderson, 2004; Thomas et al, 2010). Para aves em ambientes florestais, o DS demonstra bons resultados (Gale et al, 2009; Raman, 2003; Cornils et al, 2014), no entanto, ainda há poucos estudos realizados com espécies de aves brasileiras utilizando essa metodologia (Bernardo et al, 2011; Tonetti & Pizo, 2016; Alves et al, 2017).

1.3. A seleção de microhabitats e a conservação de espécies ameaçadas

Associado à perspectiva demográfica de uma espécie, a compreensão de aspectos relacionados às suas necessidades e preferências básicas de alimentação e habitat também possuem grande influência na compreensão da dinâmica de uma população, uma vez que esta pode variar de acordo com a perda ou manutenção destes requisitos, que são fundamentais para sua sobrevivência (Bolger et al, 1991). Estudos sobre seleção de habitat podem variar entre diferentes níveis de abordagem em relação a categorias de habitat (florestas, campos abertos, áreas urbanas, por exemplo) ou em relação a características dentro de um habitat específico (presença de cursos d'água, distância entre fontes de água, densidade de serrapilheira, densidade de arbustos, abertura e altura do dossel, entre outros). Desta forma, as variáveis

abordadas em estudos de seleção podem ser mais gerais (macro), mais específicas (micro), ou a combinação de ambos (Manly et al, 2002).

Para as espécies endêmicas do CEP, que são altamente carentes em dados científicos e residem em um ambiente altamente fragmentado e constantemente exposto à degradação, estudar e compreender seus aspectos demográficos e requerimentos básicos de habitat pode evitar uma série de novas extinções precoces na região. Elucidar estes fatores possibilita não só identificar locais mais adequados e esperados para a ocorrência de espécies, mas também o reconhecimento das espécies como especialistas ou generalistas e permite, ainda, associar estas informações às principais ameaças enfrentadas pelas espécies, possibilitando também a proposição de estratégias mais direcionadas para sua conservação (Schochat & Tsurim, 2004; Marsden et al, 2005; Kaneage, 2009; Namgay & Wangchuk, 2016) e sendo o ponto de partida para estudos mais aprofundados (Manly et al, 2002).

2. Objetivos Gerais

Este trabalho teve como objetivos estudar a seleção de microhabitats para duas espécies de passeriformes endêmicos do CEP, bem como conduzir censos populacionais em cinco espécies, gerando informações relevantes para a conservação das mesmas, incluindo a contribuição na atualização do status de ameaça dos taxa analisados.

3. Capítulo 1: Conservation status and key microhabitat parameters for two endangered tropical forest understory insectivorous passerines from the Pernambuco Endemism Center

Abstract

The Pernambuco Center of Endemism (PEC) is the most fragmented and degraded of the Atlantic Forest regions, being considered as hotspot within a hotspot. Recent bird extinctions and the great number of endangered taxa have called the attention of conservation practitioners all over the world to this area. Among the most vulnerable groups of birds are the insectivorous passerines specialized in thriving in forest understory, yet empirical information on demography and habitat requirements are unavailable for most taxa. Here we provide censuses estimates and microhabitat selection information for two endangered forest understory insectivorous passerines endemic to the PEC, the Pernambuco Fire-eye, *Pyriglena pernambucensis*, and the Black-cheeked Gnateater, *Conopophaga melanops nigrifrons*. Distance-sampling estimates resulted in population densities of 0.15 and 0.35 individuals/ha, respectively, in one of the most representatives Atlantic Forest fragments from PEC. Extrapolations of population densities to the areas of known occurrence of these taxa resulted in minimum population estimates of 3586 individuals for the Pernambuco Fire-eye, and 8366 individuals for the Black-cheeked Gnateater. Although these are underestimates, they were enough to discard the possibilities of these species to change their status from Vulnerable to Endangered. Microhabitat preference analyses revealed that both species selected sites with denser forest understory vegetation, with the presence of lianas, which however, were not associated to recently created forest gaps, demonstrating that the maintenance of habitat heterogeneity can be important for these birds. In face of the ongoing threats to the PEC areas, our data will serve to parameterize other studies and may contribute to practical conservation policies.

Key Words: birds, population census, distance sampling, microhabitat selection

3.1. Introduction

The Brazilian Atlantic Forest is one of the main biodiversity hotspots on earth (Myers et al 2000). After more than 500 years of exploitation, only about 11.4 to 16% of the original area is left, and around 97% of the remnants are smaller than 250 ha and are exposed to intense anthropogenic pressures (Ribeiro et al 2009). Within this megadiverse biome, the Pernambuco Endemism Center (hereafter PEC) is of special concern, being considered as a hotspot within a hotspot (Tabarelli et al 2006, Pontes et al 2016). The PEC was a portion of 56.000 km² of the Atlantic Forest distributed northern from São Francisco River, in the states of Paraíba, Pernambuco, Alagoas and Rio Grande do Norte, northeastern Brazil. This is the mostly degraded of the Atlantic Forest regions, with only approximately 2% of the original forests

remaining (Pereira et al 2016, Garbino et al 2018), and especially during the decades of 1970 and 1980 the forest remnants suffered intense logging to supply the boilers of the sugar-cane and ethanol local industries with timber. Three bird species from PEC were recently recognized as globally extinct (ICMBio 2018); some were likely extinct before being described to science (Silveira et al 2003, Lees & Pimm 2015), and many others are on the verge of extinction (Pereira et al 2014, 2016, ICMBio 2018), most with no conservation actions proposed.

In this scenario of intense degradation, studies on bird population demography are important because they can be the first step for estimating local and global population sizes, which in turn can parameterize population viability analyses, and are elementary for the assessment of the actual conservation status of the target species (Sinclair et al 2006, Cornils et al 2015, Tonetti & Pizo 2016, Machado et al 2019). Further, studies on microhabitat selection can contribute to the understanding of local extinctions, and can indicate habitat characteristics that must be preserved or restored for the conservation of certain species (Sinclair et al 2006, Vié et al 2009, Cornils et al 2015, Tonetti & Pizo 2016).

In the Neotropics, the guild of the forest understory insectivore passerines is of interest to conservation practitioners (Powell et al 2015). These birds are often unwilling to display long flights and are reluctant to cross open areas (Oliveira-Jr et al 2011), which favors the isolation of small populations in fragments of habitats (Camargo et al 2015). For these reasons, they are vulnerable to microhabitat changes caused by anthropogenic activities, including logging and fragmentation (Powell et al 2015), and they become more exposed to inbreeding and to the negative demographic impacts of population isolation (Camargo et al 2015). Although they are not game birds and are not target to illegal trapping, two of the three bird species from PEC that were recently uplisted as extinct are forest understory insectivorous passerines, the Cryptic Treehunter (*Cichlocolaptes mazarnanetti*), and the Alagoas Foliage-gleaner (*Philydor novaesi*), and of the 13 other globally endangered taxa, most are forest understory insectivore passerines (Pereira et al 2014, ICMBio 2018), which makes the understanding of their demographic aspects and of their habitat requirements of broad theoretical and practical interest.

Here we provide distance sampling-based population density estimates, and microhabitat selection information for two endangered forest understory insectivorous passerines from PEC, the Pernambuco Fire-eye, *Pyriglena pernambucensis*, and the Black-cheeked Gnat-eater, *Conopophaga melanops nigrifrons*, from one of the most representative Atlantic Forest remnants from Alagoas state, northeastern Brazil. Further, we extrapolated

population density values to areas with reliable records of these species to obtain the first minimum global population size estimates. To our knowledge, this is the first empirical study to address demographic aspects and microhabitat requirements for endangered bird species from this important global hotspot.

3.2. Methods

3.2.1. Study area

Field work was carried out at RPPN Mata do Cedro ($9^{\circ}31'23.82''S$; $35^{\circ}55'6.53''W$), a 978-ha privat Atlantic Forest conservation unit surrounded by sugar cane plantations, located at the municipality of Rio Largo, state of Alagoas, northeastern Brazil (Fig. 1). Vegetation is characterized as open ombrophilous forest (Roda & Santos 2005), and like most of the PEC fragments it has been through selective logging during the decades of 1970 and 1980, and today forest tracts in middle to late generation stages can be observed, with emergent trees already noticeable (Roda & Santos 2005, Pereira et al 2014, 2016). The climate is AS' according to the classification of Köppen: tropical with a well-defined dry season (October through January), and a long rainy season concentrated during the autumn and winter. Average annual precipitation varies from 1600–1700 mm, and average minimum and maximum temperatures vary from 21–22°C, and 30–31°C, respectively (Roda & Santos 2005, Barros et al 2012).

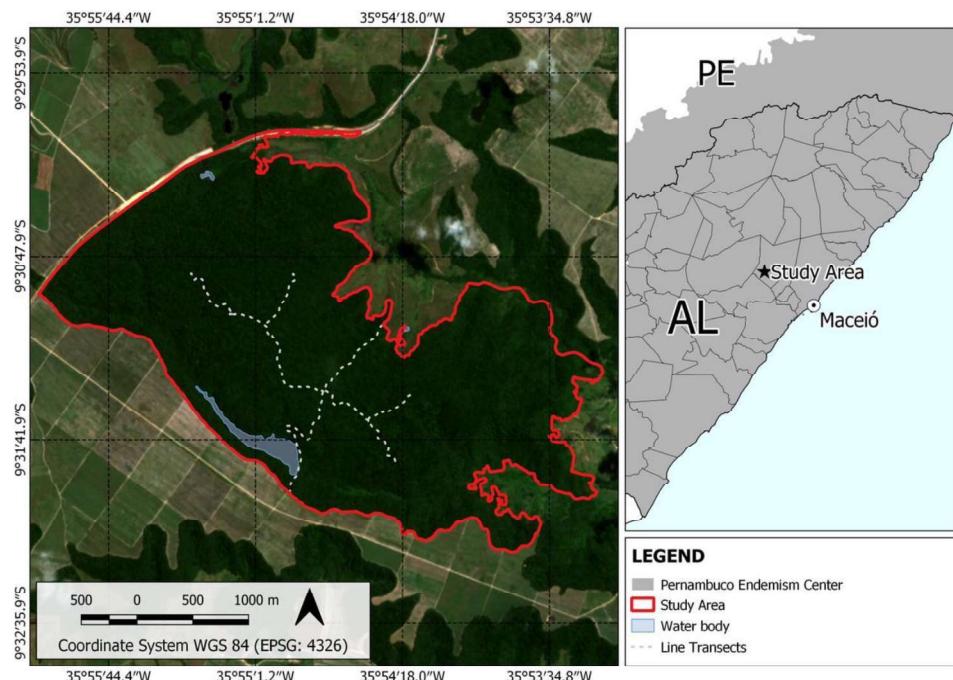


Figure 1. Study area, RPPN Mata do Cedro, in the state of Alagoas, northeastern Brazil. Dotted lines depict the transects used in Distance Sampling analyses.

3.2.2. Study species

Previously classified as a subspecies of *Pyriglena leuconota*, the Pernambuco Fire-eye, *P. pernambucensis* (Thamnophilidae) was recently recognized as a full-species by the Brazilian Committee of Ornithological Records (CBRO) (Piacentini et al 2015). This species is endemic to the PEC, occurring in the states of Alagoas, Pernambuco and Paraíba. It is forest-dependent and inhabits the low forest strata, including borders and areas in low regeneration stages, but it is reluctant to displace between fragments (ICMBio 2018). It forages on arthropods often on the leaf litter, and it is among the typical ant-follower birds (Zimmer & Isler 2020).

The Black-cheeked Gnat-eater, *Conopophaga melanops nigrifrons* (Conopophagidae), is the subspecies of the widely distributed *C. melanops* restricted to the PEC, in the states of Alagoas, Pernambuco, and Paraíba (Whitney & de Juana 2020). It is remarkably sedentary and dependent on well-preserved forests, although it can be seen near borders and regenerating areas (ICMBio 2018). Like the Pernambuco Fire-eye, the Black-cheeked Gnat-eater also forage on forest floor and it can follow army-ants to capture arthropods (Whitney & de Juana 2020). Although it has been long classified as a subspecies, taxonomic reviews are advised to confirm if it could be a full species (ICMBio 2018).

3.2.3. Population density estimates

Population densities of the Pernambuco Fire-eye and of the Black-cheeked Gnat-eater were estimated using line-transects distance sampling approach (Buckland et al 1993, 2001). Seven line-transects with lengths varying from 0.5 to 2.17 km were distributed in the interior of the forest fragment, totaling 6.36 km. Two of the transects were in pre-existent, 1 to 2 m wide trails, being one still used only for vigilance, and the other was abandoned at least 20 years ago. The other transects were established in forest interior without disturbing the vegetation. As vegetation in the pre-existing trails was in late regeneration stage, they were not treated as different habitats for statistical analyses.

Data were collected from October 2019 to January 2020, which corresponds to the summer in the region. This is the season in which the days are longer and birds were proved to vocalize intensely during this period. Each transect was walked 10 times with at least 10 days intervals, at an average speed of 1.5-2 km/h, always during the first hours after sunrise (4:30–9:00 am). While walking transects, the birds were detected either visually or audibly and the perpendicular distance between the initial detection point of each individual and the transect line was measured with a laser measuring tool (Stanley tlm100 – 30 mt) to improve data accuracy. Data of all individuals, independently of gender and age, were collected without truncation distance by a single observer (LCP) trained in Pernambuco Fire-eye and Black-cheeked Gnat-eater identification, and both sides of each transect were considered.

3.2.4. Microhabitat selection

To address microhabitat selection in the two studied taxa, we compared a set of parameters between sites in which birds were observed, and sites chosen at random in the study area. Specifically, we aimed to address microhabitat selection during the period of resources scarcity (the dry season). This is because the availability of certain microhabitats may be more important for bird's survival during this period than during the rainy season, in which arthropods are theoretically more abundant (Devries & Walla 2001, Coutinho-Silva et al 2017). Then, microhabitat data collection also occurred from October 2019 to February 2020, which corresponds to the dry season in the region (summer).

Random sites were generated throughout the whole forest fragment by QGIS v. 3.8.1 (QGIS 2020). These random points were accessed by following the shortest routes created by a GPS (Garmin eTrex® 20x), which could start from forest border or from one of the trails. Sites used by birds were located when observers were walking these routes, to ensure that they were also searched at random across the whole area. Random points and locations where individuals were first observed were adopted as the centroids of 5 m radius plots. Flying individuals were not considered, meaning that most birds were foraging or defending territories, and when birds were in pairs or small groups, only the location of the first observed individual was considered. At “Use” and “random” plots we measured the following biotic microhabitat parameters: canopy openness, canopy height, amount of leaf litter, number of trees or saplings with diameter at breast height (DBH) < 10 cm, number of trees with DBH > 10 cm,

presence/absence of lianas, vegetation density 0.5 m above ground, vegetation density 1.5 m above ground, and biomass. Further, as abiotic parameters, we estimated altitude and declivity.

Canopy cover was estimated by only one of the researchers (LCP) at the center of the plots using a spherical densiometer (Convex Model-A, Forest Suppliers, Inc., Jackson, MS, USA). The equipment was positioned in the four cardinal directions and the number of illuminated grids was averaged to estimate canopy openness following the manufacturer instructions (Lemmon 1957). Canopy height was estimated by a trained observer (LCP) and the distance of reference points were measured with the laser measuring tool (Stanley tlm 100 – 30 mt) to improve measurement accuracy. The amount of leaf litter was estimated by its height in cm in eight different points: in the four cardinal directions at the center of the plots, and at the four cardinal directions 1 m from the center of the plots. Then, the values were averaged to obtain the estimation of leaf litter in the plot. The number of trees or saplings with DBH > and < 10 cm at 5 m radius were estimated by direct counting, while lianas and fallen branches/trees were considered as dummy variables (presence/absence). Vegetation density at 0.5 and 1.5 m above ground were measured with a 5 m metal rod subdivided into 50 intervals of 10 cm. Then, vegetation density was estimated as the total number of touches by vegetation in the four cardinal directions. Biomass at “use” and “random” plots was assessed using remote sensing through a NDVI (Normalized Difference Vegetation Index) layer derived from a Sentinel-2 Level-1C (L1C) MS satellite scene (dated from March 19th, 2019). NDVI is widely used to describe habitat productivity and biomass availability (Riedel et al, 2005; Hansen et al, 2009). The NDVI layer was generated using the band equation $\left(\frac{NIR-red}{NIR+red}\right)$ from Lange et al (2017). Altitude and declivity layers were obtained from the Brazilian TOPODATA project (Instituto Nacional de Pesquisas Espaciais – INPE; <http://www.webmapit.com.br/inpe/topodata/>). Values of NDVI, altitude and declivity at “use” and “random” locations were extracted using ‘*raster*’ (Hijmans et al, 2013) and ‘*rgdal*’ (Bivand et al, 2015) packages in R (R Core Team, 2020).

3.2.5. Statistical analyses

Population density estimates were obtained by the model selection procedure implemented in the software Distance 7.0 (Thomas et al 2010). In this approach, a set of pre-defined key detection functions (uniform, hazard-rate, half-normal, and negative exponential key functions) are used to model how the probability of detection decreases with increasing perpendicular

distances, which is then used to obtain corrected population density estimates (Buckland et al 1993, 2001, Thomas et al 2010). Each detection function was run with cosine, hermite polynomial, and simple polynomial adjustments, resulting in 12 different models. The efficiency of each model to fit the data was assessed by i) Akaike's Information Criteria (AIC), ii) the goodness-of-fit test of Kolmogorov-Smirnov, and iii) by visual inspection of quantil-quantil plots (Q-q plots). Due to the small sample sizes expected for endangered species restricted to small areas, the different transects were considered as a unique longer transect for the statistical analyses, and all the records obtained in transect replicates were pooled together, which permits a more accurate modeling (see also Bernardo et al 2011). Then, the final population density estimate was subdivided by 10 (the number of replicates) to report the actual number of individuals per hectare. Variation Coefficient and lower and upper 95% confidence intervals associated to population density estimates were obtained by the analytical approach, using default parameters settings.

To test habitat preferences, “use” and “random” plots were compared using Generalized Linear Models (GLM) with binomial distribution and logit-link function. All of the non-categorical explanatory variables were standardized using z-score and tested for correlations (Pearson's $r \geq 0.6$). Model parameter estimates, their standard errors (SE), and values of z-tests used to verify the levels of significance of each variable within the models are presented. All of the statistical procedures were conducted using the software R (R Core Team, 2020).

3.2.6. Conservation status assessment

Both the Pernambuco Fire-eye and the Black-cheeked Gnat-eater were classified as “vulnerable” in the Brazilian Red List (ICMBio 2018). The Brazilian taxa assessment committee adopts the classification criteria of IUCN (see IUCN 2019), and these taxa were currently classified as vulnerable based on an estimated Area of Occupancy (AOO) $< 2000 \text{ km}^2$, which is still declining and is highly fragmented (ICMBio 2018, IUCN 2019), and demographic information was not used due to the lack of empirical information. Then, we performed a literature review on bird surveys carried in PEC fragments and we multiplied our population density estimates by the size of the areas with reliable records of these taxa to obtain rough minimum population size estimates. As surveys were already conducted in the largest fragments (see Results below), these extrapolations are valuable mainly to address the criteria C2a(i) from

IUCN guidelines for taxa assessment (IUCN 2019), which regards to the maximum numbers of individuals that each independent area can hold.

3.3. Results

3.3.1. Population density estimates

For the Pernambuco Fire-eye we recorded 83 perpendicular distances, varying from 0.5 to 65 m (25.20 ± 17.49). The detection function Uniform Cosine presented the lower ΔAIC , a low CV, and the less significant goodness-of-fit test (Table 1). Further, Q-q plot showed no remarkable deviations and the graphic of the detection probability vs perpendicular distances (Fig. 2) provided visual evidences for a good model fit. Using this selected model, the corrected population density (subdivided by 10) in the study area was 0.15 individuals/ha, and the estimated population size for the study areas was 147 individuals.

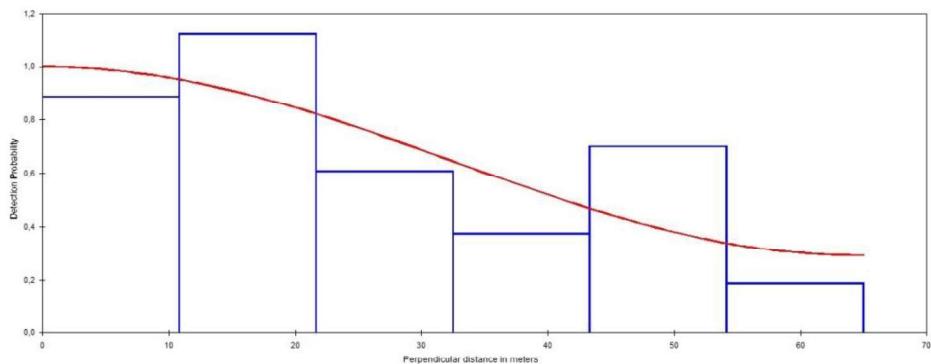


Figure 2. Detection probabilities of the Pernambuco Fire-eye at different perpendicular distances, and adjustment of the Uniform Cosine detection function.

For the Black-cheeked Gnateater we recorded 167 perpendicular distances varying from 0.5 to 80 m (22.84 ± 15.61). For this taxon, the three Half-Normal detection functions presented $\Delta AIC = 0$, but goodness-of-fit tests were marginally significant for these models. Then, we opted for choosing Hazard-Rate Cosine to estimate population density, as all of the Hazard-Rate models had only slightly bigger ΔAIC values (0.32) and they presented the less significant goodness-of-fit probabilities (Table 1). The Q-q plot and the graphic of the detection probability vs perpendicular distances also confirmed the visual fit of this model (Fig. 3). Following this

model, the corrected population density was 0.35 individuals/ha, with an estimated population size for the study area of 342 individuals.

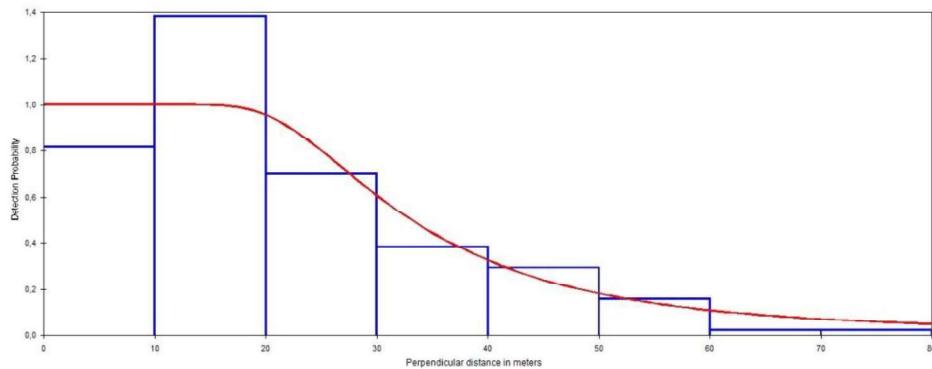


Figure 3. Detection probabilities of the Black-cheeked Gnateater at different perpendicular distances, and adjustment of the Uniform Cosine detection function.

Table 1. Model selection results based on distance sampling data used to estimate population densities (D) for two endangered species from the Pernambuco Center of Endemism (PEC): the Pernambuco Fire-eye (*Pyriglena pernambucensis*), and the Black-cheeked Gnateater (*Conopophaga melanops nigrifrons*). The best-fitted models were selected using AIC Criteria, and the relative importance of each model was evaluated by ΔAIC . For each model, estimates of lower (LCL) and upper 95% confidence limits (UCL), variation coefficient (CV), and probability of the Kolmogorov-Smirnov goodness-of-fit test (P) are presented. These estimates were obtained by pooling together 10 transect replicates, without correction.

	AIC	ΔAIC	D	LCL	UCL	CV	P
Pernambuco Fire-eye							
Uniform Cosine	681.98	0.00	1.55	1.30	1.86	0.09	0.839
Uniform Polynomial	683.25	1.28	1.37	1.22	1.53	0.06	0.270
Uniform Hermite	683.25	1.28	1.37	1.22	1.53	0.06	0.270
Half-Normal Cosine	682.06	0.08	1.51	1.25	1.83	0.10	0.716
Half-Normal Polynomial	682.06	0.08	1.51	1.25	1.83	0.10	0.716
Half-Normal Hermite	682.06	0.08	1.51	1.25	1.83	0.10	0.716
Hazard-Rate Cosine	683.48	1.50	1.67	1.10	2.54	0.21	0.689
Hazard-Rate Polynomial	683.48	1.50	1.67	1.10	2.54	0.21	0.689
Hazard-Rate Hermite	683.48	1.50	1.67	1.10	2.54	0.21	0.689
Neg. Exponential Cosine	682.20	0.22	1.86	1.36	2.54	0.16	0.705
Neg. Exponential Polynomial	682.20	0.22	1.86	1.36	2.54	0.16	0.705
Neg. Exponential Hermite	682.20	0.22	1.86	1.36	2.54	0.16	0.705
Black-cheeked Gnateater							
Uniform Cosine	1352.94	1.28	3.68	3.23	4.19	0.07	0.112
Uniform Polynomial	1353.36	1.70	3.28	3.08	3.48	0.03	0.036
Uniform Hermite	1386.24	34.57	2.44	2.39	2.48	0.01	0.000
Half-Normal Cosine	1351.66	0.00	3.73	3.32	4.18	0.06	0.093
Half-Normal Polynomial	1351.66	0.00	3.73	3.32	4.18	0.06	0.093
Half-Normal Hermite	1351.66	0.00	3.73	3.32	4.18	0.06	0.093
Hazard-Rate Cosine	1351.98	0.32	3.47	3.00	4.02	0.07	0.208
Hazard-Rate Polynomial	1351.98	0.32	3.47	3.00	4.02	0.07	0.208
Hazard-Rate Hermite	1351.98	0.32	3.47	3.00	4.02	0.07	0.208

Neg. Exponential Cosine	1354.68	3.02	3.72	2.90	4.77	0.13	0.140
Neg. Exponential Polynomial	1354.83	3.16	3.57	2.77	4.60	0.13	0.160
Neg. Exponential Hermite	1354.83	3.16	3.57	2.77	4.60	0.13	0.160

3.3.2. Microhabitat Selection

In total, we collected microhabitat parameters from 28 detection plots for the Pernambuco Fire-eye, 34 plots for the Black-cheeked Gnateater, and from 54 random plots. Values or frequencies of the parameters are shown in Table 2. For both species, the z-score transformed numerical variables altitude and declivity were correlated (Tables S1 and S2). Then, the GLM analyses were partitioned to address these parameters in separate models (see Table 3). For the Pernambuco Fire-eye, in the model including altitude, vegetation density 1.5 m above ground was positively correlated to species presence, and in the model containing declivity, vegetation densities at both 1.5 m and 50 cm above ground presented significant positive correlations. Notably, the presence of trees with DBH \leq 10 cm presented marginal negative correlations in both models (Table 3). For the Black-cheeked Gnateater, in both models partitions the presence of lianas and vegetation density 1.5 m above ground were positively correlated to the probability of occurrence of the taxa (Table 3).

Table 2. Values or frequencies of 12 environmental parameters used to investigate microhabitat selection by the Pernambuco Fire-eye, *Pyriglena pernambucensis*, and by the Black-cheeked Gnateater, *Conopophaga melanops nigrifrons*. The addressed parameters were: canopy opening (CO), canopy height (CH), presence/absence of lianas, presence/absence of fallen trees or branches (FT), number of trees with diameter at breast height > 10 cm (DBH > 10 cm) and \leq 10 cm (DBH \leq 10 cm), vegetation density at 1.5 m above ground (VD 1.5 m), vegetation density 50 cm above ground (VC 50 cm), amount of leaf litter (LL), altitude, declivity, and biomass (NDVI).

	Pernambuco Fire-eye	Black-cheeked Gnateater	Random points
CO	15.45 ± 7.08 (3.9 – 30.69)	11.41 ± 5.01 (4.16 – 25.22)	13.71 ± 8.50 (3.9 – 44.46)
CH	17.43 ± 6.18 (3 – 30)	15.85 ± 5.74 (5 – 35)	17.28 ± 5.53 (2 – 27)
Lianas	4 Yes 24 No	11 Yes 23 No	4 Yes 50 No
FT	21 Yes 7 No	17 Yes 17 No	29 Yes 25 No
DBH > 10 cm	8.09 ± 3.79 (1 – 21)	6.96 ± 2.38 (3 – 12)	7.47 ± 3.08 (3 – 16)
DBH \leq 10 cm	36.30 ± 17.29 (15 – 117)	27.9 ± 13.76 (7 – 61)	30.0 ± 14.76 (10 – 68)
VD 1.5 m	3.12 ± 1.14 (0.75 – 6.5)	4.45 ± 2.16 (1.5 – 11)	4.27 ± 2.8 (1.25 – 18.5)
VD 50 cm	1.29 ± 0.63 (0.25 – 3)	1.52 ± 0.92 (0 – 4)	1.38 ± 0.84 (0.25 – 4)
LL	3.28 ± 1.28	3.84 ± 1.18	3.78 ± 1.28

	(1.19 – 7.69)	(2.06 – 5.87)	(2 – 6.12)
Altitude	134.1 ± 24.18 (41.67 – 159.91)	140.5 ± 15.79 (92.05 – 157.77)	134.34 ± 22.0 (85.3 – 159.2)
Declivity	11.71 ± 9.83 (1.40 – 34.17)	14.48 ± 11.24 (1.54 – 34.8)	14.24 ± 11.47 (1.22 – 40.40)
NDVI	0.76 ± 0.06 (0.50 – 0.80)	0.78 ± 0.02 (0.73 – 0.81)	0.78 ± 0.03 (0.63 – 0.83)

Table 3. Results of GLM modeling with binomial distribution used to infer abut microhabitat parameters selected by the Pernambuco Fire-eye, *Pyriglena pernambucensis* (Thamnophilidae), and by the Black-cheeked Gnat-eater, *Conopophaga melanops nigrifrons* (Conopophagidae). Model parameters were canopy opening (CO), canopy height (CH), presence/absence of lianas, presence/absence of fallen trees or branches (FT), number of trees with diameter at breast height > 10 cm (DBH > 10 cm) and ≤ 10 cm (DBH ≤ 10 cm), vegetation density at 1.5 m above ground (VD 1.5 m), vegetation density 50 cm above ground (VC 50 cm), amount of leaf litter (LL), altitude, declivity, and biomass (NDVI). As for both species the parameters altitude and declivity were correlated, the analyses were partitioned into two blocks to consider these two variables in independent models (Partition 1 and Partition 2). The estimated parameter value (Estimate), standard error (SE), value of the z-test used to test whether the parameter has differed significantly from zero, and its level of significance (P) are presented for each model parameter.

	Estimate	SE	z-value	P
Pernambuco Fire-eye				
<i>Partition 1</i>				
(Intercept)	-1.26	0.58	-2.17	0.030
CO	0.15	0.42	0.36	0.716
CH	-0.07	0.33	-0.21	0.830
Lianas	1.72	0.97	1.76	0.077
FT	0.15	0.74	0.21	0.836
DBH > 10 cm	-0.05	0.37	-0.13	0.900
DBH ≤ 10 cm	-0.95	0.49	-1.93	0.054
VD 1.5 m	1.20	0.42	2.87	0.004
VD 50 cm	0.62	0.34	1.81	0.070
LL	0.47	0.31	1.54	0.124
Altitude	0.94	0.50	1.89	0.059
NDVI	-0.58	0.48	-1.22	0.223
<i>Partition 2</i>				
(Intercept)	-1.27	0.58	-2.17	0.030
CO	-0.03	0.38	-0.09	0.930
CH	-0.09	0.33	-0.28	0.778
Lianas	1.66	1.02	1.63	0.104
FT	0.30	0.73	0.42	0.676
DBH > 10 cm	-0.15	0.38	-0.38	0.701
DBH ≤ 10 cm	-0.96	0.49	-1.95	0.050
VD 1.5 m	1.14	0.40	2.82	0.005
VD 50 cm	0.72	0.35	2.06	0.039
LL	0.53	0.31	1.69	0.090
Declivity	-0.66	0.39	-1.69	0.090
NDVI	-0.60	0.47	-1.27	0.203

Black-cheeked Gnat-eater				
<i>Partition 1</i>				
(Intercept)	-0.89	0.45	-1.96	0.050
CO	-0.29	0.32	-0.92	0.358
CH	0.36	0.33	1.08	0.281
Lianas	1.74	0.78	2.23	0.026
FT	0.13	0.58	0.23	0.818
DBH > 10 cm	0.08	0.30	0.28	0.780
DBH ≤ 10 cm	-0.52	0.32	-1.65	0.099
VD 1.5 m	1.06	0.50	2.13	0.033
VD 50 cm	0.20	0.28	0.71	0.478
LL	0.26	0.27	0.98	0.326
Altitude	0.20	0.29	0.68	0.498
NDVI	0.20	0.30	0.65	0.516
<i>Partition 2</i>				
(Intercept)	-0.87	0.45	-1.93	0.053
CO	-0.27	0.32	-0.86	0.391
CH	0.37	0.34	1.10	0.271
Lianas	1.72	0.78	2.20	0.027
FT	0.11	0.57	0.19	0.847
DBH > 10 cm	0.09	0.30	0.30	0.766
DBH ≤ 10 cm	-0.54	0.31	-1.71	0.087
VD 1.5 m	1.02	0.49	2.10	0.035
VD 50 cm	0.21	0.28	0.75	0.452
LL	0.28	0.27	1.03	0.303
Declivity	-0.15	0.28	-0.54	0.590
NDVI	0.19	0.30	0.63	0.530

3.3.3. Conservation status assessments

In our literature review we found 39 areas from the PEC with bird surveys published in the scientific literature, and the Pernambuco Fire-eye and the Black-cheeked Gnat-eater were recorded in all of them. These areas varied from 25 to 4469 ha, and together they summed 23,904 ha. The extrapolating our population density estimates to this area resulted in a minimum population size of 3586 individuals for the Pernambuco Fire-eye, and 8366 individuals for the Black-cheeked Gnat-eater.

3.4. Discussion

3.4.1. Population density

Population density is one of the main parameters explaining rarity, and rare species have increased chances to become endangered (Goerck 1997, Birskis-Barros et al 2019). However, population density estimates based on distance sampling methods for both endangered and non-endangered birds from neotropical forests are scarce, which limits the possibilities of

comparisons. The population density of the Black-cheeked Gnateater (0.35 individuals/ha) being more than twice as big as the density found for the Pernambuco Fire-eye (0.15 individuals/ha) confirms our prediction that bird species from the PEC included in the Brazilian Red List based on habitat parameters alone can have highly divergent populations densities and estimated population sizes (see discussion bellow), and consequently different risks of extinction even when presenting very similar geographic distributions. For the near threatened Southern Bristle-Tyrant, *Phylloscartes eximius* (Tyrannidae), from the Atlantic Forest of southeastern Brazil, estimated population density was 0.13 individuals/ha, which was close to the value we obtained for the Pernambuco Fire-eye, and it was considered very low when compared to unpublished data on another conger (Tonetti & Pizo 2016). For the endangered Black-cheeked Ant-tanager, *Habia atrimaxillaris* (Thraupidae) from tropical forests from Costa Rica, population densities varied from 0.24 to 0.27 individuals/ha (Cornils et al 2015), also evidencing that population density of the Pernambuco Fire-eye was relatively low and of the Black-cheeked Gnateater was relatively high.

3.4.2. Microhabitat selection

The microhabitat parameters correlated to the presence of these two insectivorous forest understory passersines were associated to characteristics of forest understory vegetation, rather than to characteristics of forest canopy, leaf litter, declivity, altitude, or local biomass concentration. Both species selected at least one parameter related to vegetation density. For the Pernambuco Fire-eye, the selection for sites with increased vegetation density at both 1.5 m and 50 cm above ground, associated with a lower concentration of plants with DBH \leq 10 cm and the presence of lianas being close to significance in model partition 1 ($P = 0.077$), suggest that this species selected sites that were in earlier regeneration stages in relation to the random plots, where sunlight penetration may have permitted herbaceous vegetation to grow on forest floor. Although the presence of the Black-cheeked Gnateater also seemed to be related to regenerating areas, they proved to choose more mature sites, with concentration of lianas and denser understory vegetation, but in places where herbaceous vegetation no longer existed. In tropical forests, areas with denser vegetation often arise in forest gaps, i.e. due to the fall of large trees. The presence of forest gaps has been demonstrated to affect the frequencies of many bird groups in tropical forests (Banks-Leite & Cintra 2008). However, the lack of correlation

between canopy opening and the presence of our studied taxa suggest that they have not searched for recently formed gaps.

For forest understory insectivorous birds, the selection for microhabitats with denser vegetation can be related to food abundance, as a number of studies have demonstrated that in tropical forests, the production of young leaves can attract more herbivorous insects, which consequently can attract more insectivorous birds (Richard & Windsor 2007). It is worth noting that our study was conducted during the dry season, in which insects' abundance is often reduced (Devries & Walla 2001, Coutinho-Silva et al 2017), suggesting that the selected microhabitats may be important to increase foraging efficiency during this period of food scarcity.

Our data reinforce the previous findings that microhabitats in neotropical forests are heterogeneous and that different bird species can show preferences for specific habitat parameters (Cintra & Cancelli 2008). It suggests that the maintenance of habitat heterogeneity is an important goal for the conservation of PEC bird communities. We are unaware, however, if the logging activities that occurred in the past have contributed to increase the number of habitats selected by the Pernambuco Fire-eye and by the Black-cheeked Gnat-eater. If this is true, their population densities may be temporarily inflated and will tend to decrease as forest fragments reach more advanced regeneration stages, and it should be monitored in the future.

3.4.3. Conservation status assessment

Our global population estimative are certainly underestimations of the real population sizes of the studied species because there are many other Atlantic Forest fragments in the PEC that were not surveyed or do not have any related scientific publication. However, these calculations were enough to show that, according to the population size IUCN criteria (C criteria), none of the two taxa we addressed were eligible to be uplisted to Endangered, as it would require a global population smaller than 2500 mature individuals. Our data also permitted to infer about the C2a(i) criteria, which regards to the numbers of individuals present within subpopulations, and is highly applicable to species inhabiting fragmented habitats like the PEC. According to this criteria, vulnerable taxa are distributed in populations presenting 250 to 1000 individuals, while endangered taxa should have no populations with more than 250 individuals. As we confirmed the presence of the Pernambuco Fire-eye and of the Black-cheeked Gnat-eater in the biggest

remaining Atlantic Forest fragments from the PEC, we could make inferences about this criterion with a certain level of precision. Based on our population density estimate, an area of approximately 1,600 ha would be needed to retain a population of 250 individuals of the Pernambuco Fire-eye, the species with lower population density, and in our literature review we found at least two areas above this size with reliable records of our study species, suggesting that the “Endangered” category is not applicable based on this criterion. However, none of the areas would be capable to maintain more than 1000 individuals of the Pernambuco Fire-eye, suggesting that this species have no chances to change its category to Least Concern in the future.

Although tropical forest understory insectivorous birds are of especial concern to conservation practitioners, empirical data on their microhabitat requirements and censuses estimates for the most vulnerable taxa are still scarce. In regions like the PEC, already highly fragmented and deteriorated, the endangered taxa will likely persist only with intensive management actions, meaning that knowledge on habitat requirements and census estimates of the most vulnerable taxa are urgently needed. Our study is among the very few providing these types of information for birds of this guild in the neotropics, and will serve as comparative data for other studies and may contribute to practical conservation policies.

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3.6. Supplementary Material

Table S1. Pearson r correlation coefficients for 12 microhabitat parameters obtained in points of occurrence of the Pernambuco Fire-eye, and at random points distributed throughout the study area. The addressed variables were: Canopy opening (CO); Canopy height (CH); Tangles of lianas (L); Fallen branches or trees (FT); numbers of plants with DBH > 10 (DBH>); numbers of plants with DBH ≤ 10 (DBH≤); vegetation density at 1.5 m height (VD 1.5); vegetation density at 50 cm height (VD 50); amount of lie litter (LL), altitude (Alt); declivity (DCL), and biomass (NDVI).

	CO	CH	L	FT	DBH>	DBH≤	VD 1.5	VD 50	LL	Alt	DCL	NDVI
CO	1											
CH	-0.01	1										
Lianas	-0.20	-0.05	1									
FT	0.28	0.05	-0.24	1								
DBH > 10	-0.27	-0.24	0.15	-0.26	1							
DBH ≤ 10	-0.22	-0.19	0.25	-0.34	0.17	1						
VD 1.5 m	0.16	-0.00	-0.04	0.14	-0.32	0.01	1					
VD 50 cm	0.14	0.01	-0.01	0.03	-0.09	0.03	-0.10	1				
LL	-0.22	-0.02	0.05	0.11	0.10	-0.05	0.01	-0.05	1			
Altitude	-0.50	0.07	0.16	0.10	0.15	0.10	-0.01	0.01	0.30	1		
Declivity	0.28	0.08	-0.24	0.01	-0.25	-0.29	-0.04	0.04	-0.15	-0.68	1	
NDVI	-0.29	0.09	0.17	-0.16	0.24	0.36	-0.16	-0.10	0.15	0.35	-0.22	1

Table S2. Pearson r correlation coefficients for 12 microhabitat parameters obtained in points of occurrence of the Black-cheeked Gnateater, and at random points distributed throughout the study area. The addressed variables were: Canopy opening (CO); Canopy height (CH); Tangles of lianas (L); Fallen branches or trees (FT); numbers of plants with DBH > 10 (DBH>); numbers of plants with DBH ≤ 10 (DBH≤); vegetation density at 1.5 m height (VD 1.5); vegetation density at 50 cm height (VD 50); amount of lie litter (LL), altitude (Alt); declivity (DCL), and biomass (NDVI).

	CO	CH	L	FT	DBH>	DBH≤	VD 1.5	VD 50	LL	Alt	DCL	NDVI
CO	1											
CH	0.09	1										
Lianas	-0.25	-0.39	1									
FT	0.21	0.13	-0.29	1								
DBH > 10	-0.14	-0.29	0.06	-0.12	1							
DBH ≤ 10	-0.11	-0.16	0.06	-0.15	0.15	1						

VD 1.5 m	-0.12	-0.10	0.25	-0.08	-0.23	-0.05	1					
VD 50 cm	0.11	0.25	-0.14	0.28	0.02	0.08	-0.09	1				
LL	-0.25	-0.02	-0.05	0.11	0.03	-0.09	0.14	0.03	1			
Altitude	-0.01	-0.03	-0.16	-0.05	-0.00	-0.07	-0.07	0.03	-0.01	1		
Declivity	0.06	0.18	0.10	0.07	0.09	-0.07	0.01	-0.00	0.04	-0.73	1	
NDVI	0.00	-0.12	0.06	-0.18	-0.05	-0.04	0.04	-0.11	0.03	0.20	-0.24	1

4. Capítulo 2: Population density and conservation status of three endangered bird species from the Pernambuco Center of Endemism, northeastern Brazil

Abstract

Determining the relative risks of extinction of declining taxa is important to delineate conservation priorities and to guide the investments in conservation. Although Red Lists are predicted to provide these parameters to conservation managers, many taxa are assessed only based on habitat characteristics due to the lack of information. Brazil concentrates the greatest number of endangered avian taxa on earth, yet demographic information is lacking for most of them. Here we present distance-sampling population density estimates for three endangered tropical forest taxa endemic to the Pernambuco Center of Endemism (PEC), the most critically disturbed Atlantic Forest region. The analyzed taxa were the White-shouldered Antshrike *Thamnophilus aethiops distans* (EN), the Brown-winged Mourner *Schiffornis turdina intermedia* (VU), and the White-bellied Tody-tyrant *Hemitriccus griseipectus naumburgae* (VU). The estimated numbers of individuals/ha were highly variable, being 0.21, 0.14, and 0.73, respectively. Using the criteria C2a(i) from IUCN, which regards to the number of mature animals in individual subpopulations, we estimated the area needed to retain populations of 250 individuals, bellow which a taxon is considered as Endangered, and 1,000 individuals, bellow which a taxon is listed as Vulnerable. With this extrapolation we showed that none of the fragments remaining in the PEC could hold more than 1,000 individuals of the White-shouldered Antshrike and of the Brown-winged Mourner, in such a way that these taxa may not reach the Least Concern category unless large forest tracts are restored or reconnected. These findings corroborated the need to include these two taxa in the Brazilian Red List, which was previously done based solely on habitat information. On the other hand, the fragments of the CEP are enough to maintain large populations of the White-bellied Tody-tyrant, corroborating the premise that taxa classified in similar threat categories based on habitat characteristics alone can have highly divergent population sizes and consequently, divergent risks of extinction. It reinforces the fact that conservation planning can be biased in the absence of demographic information.

Key words: Aves, census, distance sampling, tropical forest, Atlantic Forest

4.1. Introduction

Determining the relative risks of extinction of declining taxa is essential to indicate conservation priorities, and to optimize the investments in conservation (Rodrigues et al 2006, Bennun et al 2018). Red lists are predicted to provide this information for conservation managers, but a species's assessment rely on a set of demographic information that are often unavailable (Bachman et al 2019, Santini et al 2019). Then, criteria related to habitat conservation and distribution, such as habitat extension, reduction tendencies, and levels of fragmentation have been the only parameters used for the categorization of many species (see ICMBio 2018, Santini et al 2019). Although these criteria have been sufficient to include a relevant number of taxa in red lists, the application of demographic criteria is still important for at least three main reasons: first, species vary naturally in population densities within target habitats (Gottschalk & Huettmann 2011), in such a way that in a same geographic region different species can have highly divergent population sizes; second, species sharing the same endangered habitats can respond differently to the effects of habitat disturbances, meaning that their risks of extinction are not uniform (Powell et al 2015), and third, the determinants of the demographic parameters of many taxa may not be related only to habitat amount and quality, but also to other anthropogenic effects such as poaching and trapping (Bernardo et al 2011; Alves et al 2017). Then, censuses work are essential for the proper assessment of the conservation status of endangered taxa, and their publications as scientific articles with detailed methodological descriptions permit the reproducibility of the methods for monitoring future population tendencies (Alves et al 2017, Tonetti & Pizo 2017).

Brazil is one of the richest countries in the world in number of bird species, but it is also the country that concentrates the greatest number of endangered bird taxa (BirdLife 2021). Species inhabiting the Atlantic Forest are of special concern because 120 (51%) of the 234 Brazilian endangered taxa, including subspecies, are resident in this biome (ICMBio 2018). Although the destruction and unsustainable exploitation of the Atlantic Forest has the potential for causing a catastrophic wave of global bird extinctions, censuses estimates are available only for a very few taxa (see Alves et al 2017, Tonetti & Pizo 2017). As censuses data often provide the first basis for local and global population sizes estimates, this lack of information is likely leading to misclassifications of taxa in red lists, and it has hampered the investigation of population tendencies through time (Alves et al 2017).

The Pernambuco Center of Endemism (hereafter PEC) is the portion of the Atlantic Forest distributed in northeastern Brazil, northern from São Francisco River, in the states of

Alagoas, Pernambuco, Paraíba and Rio Grande do Norte. While it shelters a unique biota, with elevated levels of endemism (Tabarelli et al 2006, Pontes et al 2016), this is the most fragmented and degraded of the Atlantic Forest regions, with only about 2-6% of the forested areas remaining, scattered in small fragments (Pereira et al 2016, Garbino et al 2018). This region has alarmed conservation practitioners because three bird species endemics from the CEP were recently recognized as globally extinct: the Pernambuco Pygmy-owl (*Glaucidium mooreorum*), the Cryptic Treethunter (*Cichlocolaptes mazarnanetti*), and the Alagoas Foliage-gleaner (*Philydor novaesi*), and many other are on the verge of extinction (Pereira et al 2014, ICMBio 2018). Paradoxically, demographic information needed for the accurate assessment of the taxa in red lists, and to give raise to conservation plans are unavailable for most taxa.

Here we provide distance sampling-based population density estimates for three endangered bird taxa endemic to the PEC, the White-shouldered Antshrike *Thamnophilus aethiops distans* (EN), the Brown-winged Mourner *Schiffornis turdina intermedia* (VU), and the White-bellied Tody-tyrant *Hemitriccus griseipectus naumburgae* (VU), from one of the most representatives remaining Atlantic Forest fragments from the state of Alagoas, Brazil. Our population densities estimate, multiplied by estimates of the Areas of Occupancy (AOO) previously provided for these taxa by the evaluation committee of the Brazilian Red List of endangered taxa (ICMBio 2018), resulted in the first rough global population size estimates for these taxa. To our knowledge, this is the first population density estimate for an endangered bird species of PEC, and one of the few population assessments based on census data for Atlantic Forest birds.

4.2. Material and Methods

4.2.1. Study area

Censuses estimates were performed in a 978-ha privat Atlantic Forest conservation unit (RPPN Mata do Cedro; 9°31'23.82"S; 35°55'6.53"O), at the municipality of Rio Largo, Alagoas state, northeastern Brazil (Fig. 1). The area is surrounded by sugar cane plantations and the vegetation is classified as open ombrophilous forest (Roda & Santos 2005). Despite selective logging occurred in the decades of 1970 and 1980 in most PEC fragments, the area currently presents tracts of habitats in middle and late regeneration stages, with emergent trees already present (Roda & Santos 2005, Pereira et al. 2014, 2016). The climate is AS', following Köppen classification: tropical with a well-defined dry season from October through January, and a long rainy season corresponding to the months of the autumn and winter. Average annual rainfall is

1600–1700 mm, and average minimum and maximum temperatures range from 21–22°C, and 30–31°C, respectively (Roda & Santos 2005, Barros et al. 2012).

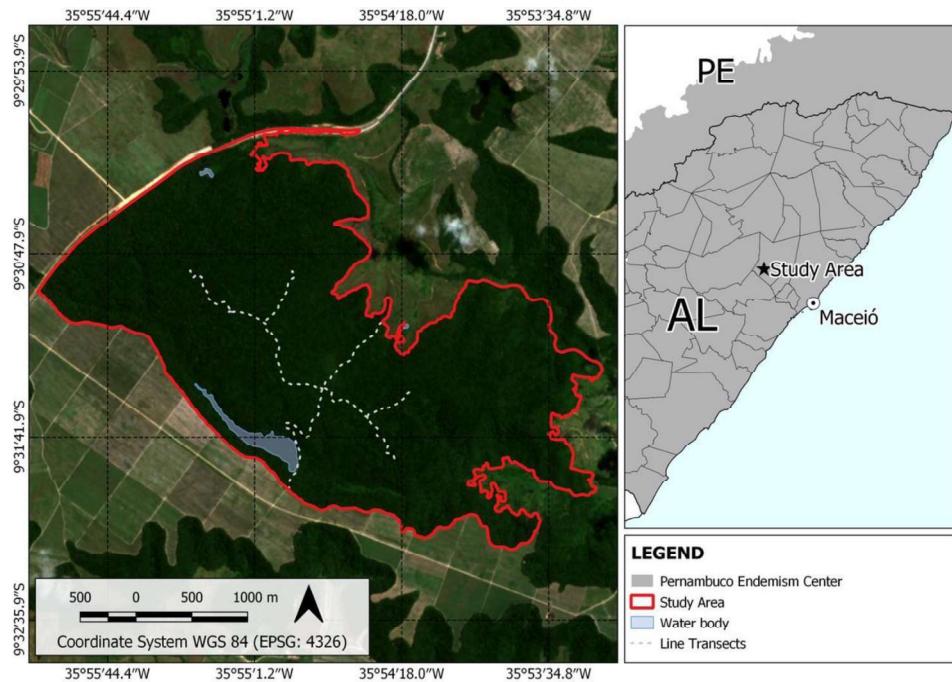


Figure 1. RPPN Mata do Cedro, located at the municipality of Rio Largo, Alagoas state, northeastern Brazil. Dotted lines represent the transects used in Distance Sampling surveys.

4.2.2. Study species

The White-shouldered Antshrike *Thamnophilus aetiops distans* (Thamnophilidae) is an insectivorous passerine that occur in the understory of mature or secondary forests in late generation stages (ICMBio 2018). It forages near the ground and can follow army-ants to capture invertebrates, not rarely being a component of mixed-species flocks (Zimmer & Isler 2020). The species is known to occur in only 25 forest fragments from Alagoas, Pernambuco, and Paraíba states and it was classified as endangered (EN) due to an estimated occupancy area of only 244 km², (24.400 ha) that is highly fragmented and still experience severe threats (ICMBio 2018).

The White-bellied Tody-tyrant *Hemitriccus griseipectus naumburgae* (Tyrannidae) is a small flycatcher that occur in the understory and midstory of forested areas, where they are commonly seen capturing insects (Schulenberg 2020). Considering that the entire PEC is estimated to preserve 2.200 km² of Atlantic Forests (ICMBio 2018), and that the White-bellied Tody-tyrant avoid small fragments, the occupancy area of the species was estimated to be about

2000 km² by the Red List committee, reason why it was classified as Vulnerable (VU) (ICMBio 2018).

The Brown-winged Mourner *Schiffornis turdina intermedia* (Tityridae) is also restricted to the forested areas from the PEC. It inhabits forest mid-strata and feed on fruits and insects (Snow & Kirwan 2020). It was classified as Vulnerable for the same reason of the White-bellied Tody-tyrant. Even in the absence of censuses, the population was estimated to be about 10.000 individuals, distributed in highly fragmented areas that cannot hold more than 1000 individuals each, which, however, is still to be confirmed.

All of these subspecies are endemic to the PEC; are geographically isolated from other subspecies and present diagnostic characteristics that led them to be classified as different forms (Schulenberg 2020, Snow & Kirwan 2020, Zimmer & Isler 2020). Taxonomic reviews, however, are still needed to confirm if they could receive the status of full species (ICMBio 2018).

4.2.3. Population density estimates

To estimate population densities of the three taxa, we used a line-transect distance sampling approach (Buckland et al. 1993, 2001). We established seven line-transects, with lengths varying from 0.5 to 2.17 km, that were distributed in forest interior and totaled 6.36 km (Fig. 1).

Field work was performed from October 2019 to January 2020. This is the summer in the region, when the days are longer and birds vocalize intensely. Transects were walked 10 times by a single trained observer (LCP), with at least 10 days intervals, from 4:30–9:00 am (first hours after sunrise), at an average speed of 1.5-2 km/h. For each bird detected visually or audibly, the perpendicular distance between the initial detection point and the transect line was sampled using a laser measuring tool (Stanley tlm100 – 30 mt) to improve data accuracy. We did not establish truncation distances, we sampled both sides of the transects, and individuals of all ages and gender were considered.

4.2.4. Statistical analyses

Population densities were estimated using the model selection procedure of the software Distance 7.0 (Thomas et al. 2010), by which pre-defined detection functions (uniform, hazard-rate, half-normal, and negative exponential key functions) are applied to model how detection probability decreases across perpendicular distance classes, generating corrected population density estimates (Buckland et al. 1993, 2001, Thomas et al. 2010). Since each of the four detection functions can be associated to cosine, hermite polynomial, and simple polynomial adjustments, we exploited a total of 12 different models for each taxa. To select the model that best fitted the data, we used Akaike's Information Criteria (AIC), the goodness-of-fit test of Kolmogorov-Smirnov, and the visual analyses of quantil-quantil plots (Q-q plots). To reach the minimum number of 50 records necessary for Distance efficient modeling (), the set of transects was treated as an unique longer transect in the statistical analyses, and the records obtained in transect replicates were pooled together (see also Bernardo et al. 2011). Then, the obtained number of individuals per hectare was subdivided by the number of replicates ($n = 10$) to generate the actual population density estimate. We used the analytical approach, with default parameters settings, to estimate Variation Coefficients and lower and upper 95% confidence intervals associated to population density data.

4.3. Results

In total, considering the 10 transect replicates, we obtained 105 records for the White-shouldered Antshrike, with perpendicular distances varying from 2.1 to 70 m (25.33 ± 18.01); 110 records for the Brown-winged Mourner, with perpendicular distances varying from 0.5 to 80 m (32.37 ± 16.57), and 330 records for the White-bellied Tody-tyrant, with perpendicular distances varying from 1.0 to 70 m (20.16 ± 13.07).

For the White-shouldered Antshrike, although the key detection function with the lower ΔAIC value was Uniform Cosine, the Kolmogorov-Sminov goodness-of-fit test was significant for this model, indicating a poor model fit, which was corroborated by a relatively high variation coefficient (Table 1). On the other hand, the Hazard-Rate detection functions were the only presenting non-significant goodness-of-fit tests, had only slightly bigger AIC values compared to the best model based on ΔAIC (Uniform Cosine), and presented the lower confidence intervals (Fig. 2). Further, the Q-q plots had no evidences for relevant deviations. As all of the Hazard-Rate models returned identical results, independently of the three types of adjustments (cosine, Hermite polynomial, and simple polynomial) (Table 1), we considered the Hazard-Rate Cosine to obtain population density estimates.

Again, for the Brown-winged Mourner, the key detection function with lower ΔAIC was Uniform Cosine, and its adherence to the data was confirmed by the non-significant goodness-of-fit test (Table 1). Although the three combinations of the Hazard-Rate model also had non-significant goodness-of-fit tests and very similar Q-q plots compared to the Uniform Cosine, the graphic depicting the detection probabilities distributed according to perpendicular distances visually evidenced the best fit of the Uniform Cosine model (Fig. 3).

For the White-bellied Tody-tyrant the three combinations of the Hazard-Rate detection function returned the same results, being the models with the lower ΔAIC , and also the only models with non-significant goodness-of-fit tests (Table 1). The Q-q plots evidenced no remarkable deviations and the graphic of detection probabilities versus perpendicular distances also provided a visual evidence that this was the best fitted model (Fig. 4).

In Table 1 we present population density estimates (D) obtained by pooling together the 10 transect replicates. After correcting the data (subdividing by 10), the resulted numbers of individuals per hectare estimated with the selected models were 0.21 for the White-shouldered Antshrike, 0.14 for the Brown-winged Mourner, and 0.73 for the White-bellied Tody-tyrant. Then, the expected number of individuals for the three taxa in our study area of 978 ha would be 205, 137, and 714, respectively.

Table 1. Model selection results based on distance sampling data used to estimate population densities (D) for three endangered taxa from the Pernambuco Center of Endemism (PEC): White-shouldered Antshrike (*Thamnophilus aethiops distans*), Brown-winged Mourner (*Schiffornis turdina intermedia*), and White-bellied Tody-tyrant (*Hemitriccus griseipectus naumburgae*). The best-fitted models were selected using AIC Criteria, and the relative importance of each model was evaluated by ΔAIC . For each model, estimates of lower (LCL) and upper 95% confidence limits (UCL), variation coefficient (CV), and probability of the Kolmogorov-Smirnov goodness-of-fit test (P) are presented. These estimates were obtained by pooling together 10 transect replicates, without correction.

	AIC	ΔAIC	D	LCL	UCL	CV	P
White-shouldered Antshrike							
Uniform Cosine	862.41	0.00	2.39	1.90	3.01	0.12	0.008
Uniform Polynomial	862.98	0.57	2.36	1.92	2.89	0.10	0.010
Uniform Hermite	864.83	2.42	1.99	1.63	2.38	0.09	0.019
Half-Normal Cosine	868.79	6.38	1.95	1.69	2.24	0.07	0.009
Half-Normal Polynomial	868.79	6.38	1.95	1.69	2.24	0.07	0.009
Half-Normal Hermite	868.79	6.38	1.95	1.69	2.24	0.07	0.009
Hazard-Rate Cosine	863.13	0.72	2.06	1.60	2.64	1.28	0.070
Hazard-Rate Polynomial	863.13	0.72	2.06	1.60	2.64	1.28	0.070
Hazard-Rate Hermite	863.13	0.72	2.06	1.60	2.64	1.28	0.070
Neg. Exponential Cosine	864.41	2.00	2.39	1.90	3.01	0.12	0.008
Neg. Exponential Polynomial	869.58	7.17	2.49	1.89	3.27	0.14	0.026
Neg. Exponential Hermite	869.58	7.17	2.49	1.89	3.27	0.14	0.026

Brown-winged Mourner							
Uniform Cosine	930.24	0.00	1.30	0.90	1.88	0.19	0.098
Uniform Polynomial	939.18	8.94	1.58	1.50	1.66	0.02	0.011
Uniform Hermite	939.18	8.94	1.58	1.50	1.66	0.02	0.011
Half-Normal Cosine	937.07	6.83	1.49	0.83	2.65	0.30	0.027
Half-Normal Polynomial	930.76	0.51	1.51	1.16	1.96	0.13	0.021
Half-Normal Hermite	939.14	8.90	1.62	1.15	2.26	0.17	0.006
Hazard-Rate Cosine	932.20	1.96	1.36	1.28	1.46	0.03	0.085
Hazard-Rate Polynomial	932.20	1.96	1.36	1.28	1.46	0.03	0.085
Hazard-Rate Hermite	932.20	1.96	1.36	1.28	1.46	0.03	0.085
Neg. Exponential Cosine	937.45	7.21	1.48	0.95	2.29	0.22	0.030
Neg. Exponential Polynomial	941.18	10.94	1.58	1.50	1.66	0.02	0.011
Neg. Exponential Hermite	941.18	10.94	1.58	1.50	1.66	0.02	0.011
White-bellied Tody-tyrant							
Uniform Cosine	2572.44	7.40	7.33	6.19	8.68	0.09	0.023
Uniform Polynomial	2576.96	11.92	8.24	7.56	8.99	0.04	0.027
Uniform Hermite	-	-	-	-	-	-	-
Half-Normal Cosine	2571.80	6.76	7.45	6.24	8.89	0.09	0.045
Half-Normal Polynomial	2576.36	11.32	8.49	7.80	9.24	0.04	0.012
Half-Normal Hermite	2573.14	8.10	7.41	6.18	8.87	0.09	0.035
Hazard-Rate Cosine	2565.04	0.00	7.26	6.69	7.87	0.04	0.231
Hazard-Rate Polynomial	2565.04	0.00	7.26	6.69	7.87	0.04	0.231
Hazard-Rate Hermite	2565.04	0.00	7.26	6.69	7.87	0.04	0.231
Neg. Exponential Cosine	2574.44	9.40	7.33	6.19	8.68	0.09	0.023
Neg. Exponential Polynomial	2578.96	13.92	8.24	7.56	8.99	0.04	0.027
Neg. Exponential Hermite	2581.78	16.74	8.05	6.63	9.79	0.10	0.004

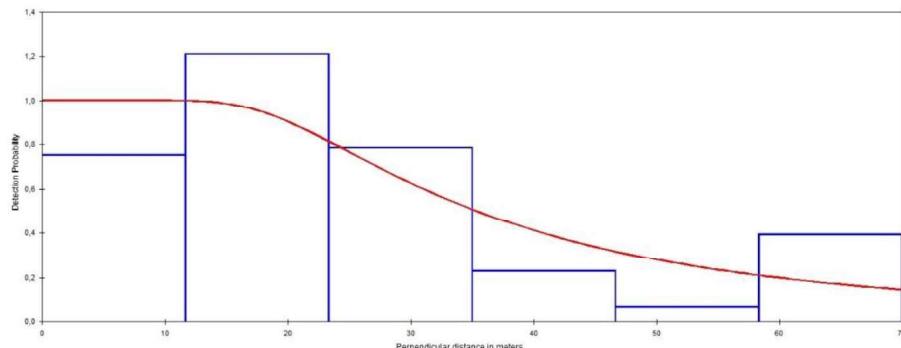


Figure 2. Detection probabilities of the White-shouldered Antshrike at different perpendicular distances, and adjustment of the Hazard-Rate Cosine detection function.

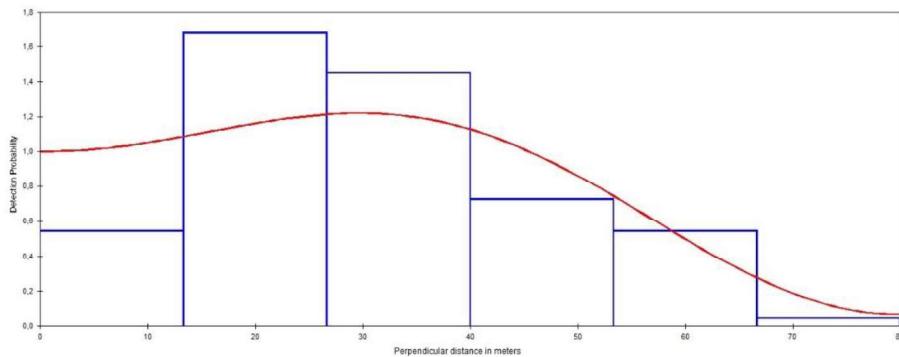


Figure 3. Detection probabilities of the Brown-winged Mourner at different perpendicular distances, and adjustment of the Uniform Cosine detection function.

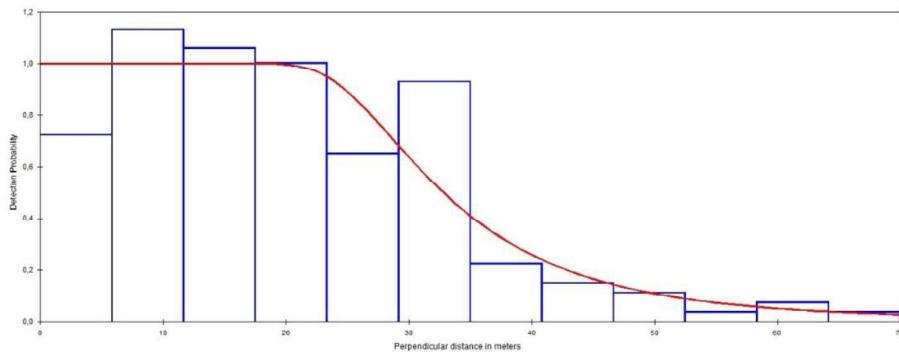


Figure 4. Detection probabilities of the White-bellied Tody-tyrant at different perpendicular distances, and adjustment of the Hazard-Rate Cosine detection function.

4.4.Discussion

During the elaboration of the Brazilian Red List of Endangered Bird Species, specialists composing the assessment committee estimated the Area of Occupancy (AOO) (IUCN 2019) for some of the taxa from the PEC. For the White-shouldered Antshrike, AOO was calculated by the summation of 4 km² grids in which the species was known to occur, which resulted in an area of approximately 244 km². Despite the lack of censuses information, this taxa was listed as Endangered (EN) based on the criteria B2ab(ii,iii) from IUCN (ICMBio 2018, IUCN 2019), which means that AOO is < 500 km². For the Brown-winged Mourner, AOO was not provided, and despite the lack of censuses data, the global population was estimated in 10.000 individuals, with no subpopulations presenting more than 1.000 reproductive animals. Then, this taxon was listed as Vulnerable (VU) based on criteria C2a(i), which means that the number of mature individuals in each population is <1.000 (IUCN 2019). The White-bellied Tody-tyrant, on the other hand, was considered as a more frequent taxa, for which AOO was assumed to be an

approximation of the total amount of Atlantic Forest in the CEP fragments ($\sim 2.000 \text{ km}^2$). Then, the latter was listed as Vulnerable by the IUCN criteria B2ab(iii), i.e. AOO is less than 2,000 km^2 , and the areas are fragmented and still declining (ICMBio 2018, IUCN 2019).

For the White-shouldered Antshrike, the multiplication of the estimated AOO by the population density we found resulted in a global population of 5124 individuals, which according to the IUCN population size criteria (criteria C) would place this taxon in the Vulnerable category (population of mature individuals between 2500 and 10,000) (IUCN 2019). Considering the criteria C2a(i), which regards to the number of mature animals in individual subpopulations, a taxon may be considered as endangered when individual populations hold 50 to 249 mature individuals, and Vulnerable when individual subpopulations hold from 250 to 1000 individuals (IUCN 2019). Using our population density estimate, an area of 1190 ha would be needed for keeping a population of 250 individuals, and an area of 4762 ha would be necessary for 1000 individuals. At least 10 Atlantic Forest areas from the CEP have between 1000 and 2000 ha, many with records of the White-shouldered Antshrike, meaning that including this taxon in the category Endangered based on C2a(i) is not advised. Areas with more than 4000 ha, however, are no longer available, in such a way that this taxon cannot reach the Least Concern category considering the C2a(i) criteria, unless large tracts of habitats are restored or reconnected. Then, based on the demographic criteria provided here, this taxon would reach the Vulnerable category, meaning that its current Endangered status based on its limited AOO must prevail.

The Brown-winged Mourner presented the lower population density estimate (0.14 individuals/ha). With such a low population density, an area of approximately 7143 ha would be necessary for keeping a population of 1000 individuals, and an area of 1786 ha would be needed for keeping 250 animals. Areas of 7000 ha do not exist in the PEC, but a few current populations may have between 250 and 500 animals, which corroborates the decision of the Brazilian Red List committee to include this taxon in the Vulnerable category due to the limited sizes of the individual populations. Like the White-shouldered Antshrike, the Brown-winged Mourner variety endemic to the PEC is impeded to reach the Least Concern category due to the small sizes of the forest fragments, unless large forest tracts are restored or reconnected.

Of the three addressed taxa, the White-bellied Tody-tyrant was the one of less conservation concerns, under a demographic perspective. Considering the C2a(i) criteria, the number of 1000 birds per subpopulation needed to reach the Least Concern category could be reached in areas above 1370 ha, which are scarce, but still occur in the PEC. Further, if the

AOO estimate of 2,000 km² presented in Brazilian Red List for this taxon is true, the global population of the White-bellied Tody-tyrant could be about 146,000 individuals, which is highly above the 10,000 individuals necessary to reach the Least Concern category based on the population size criteria of IUCN (C criteria) (IUCN 2019).

In summary, extrapolations based on our population density estimates confirmed the vulnerability of two of the three analyzed taxa also by using demographic information, being the limited sizes of subpopulations the aspect of main demographic relevance for the assessment of the birds from the PEC. Our findings also corroborated our premise that demographic information is important to indicate conservation priorities and to optimize the investments in conservation (Rodrigues et al 2006, Bennun et al 2018), as taxa listed in the same categories based on habitat parameters only, can have highly variable population sizes and divergent risks of extinction.

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5. Discussão:

Densidade e tamanho populacional global de um táxon são alguns dos principais parâmetros capazes de explicar raridade, de modo que táxons raros são mais propensos a estarem ameaçados de extinção e, em muitos casos sem intervenções, se tornarem extintos (Goerck 1997, Birskis-Barros et al 2019). No entanto, estimativas populacionais baseadas em censos coletadas a campo, através de metodologias como *Distance Sampling*, são escassas para espécies de aves neotropicais tanto classificadas como ameaçadas, quanto não-ameaçadas. A carência deste tipo de dados limita a possibilidade de comparação dos tamanhos populacionais entre táxons e a compreensão das dinâmicas de suas populações ao longo do tempo.

Neste trabalho, cinco táxons de aves passeriformes florestais ameaçado de extinção e endêmicos de uma das regiões mais ameaçadas das Américas tiveram suas densidades populacionais estimadas em um fragmento florestal de grande importância na região. A partir desses dados, novas abordagens voltadas para estimativas populacionais, área de distribuição e ocorrência poderão ser empregadas, permitindo a obtenção e análise de informações antes não disponíveis.

Dentre os cinco táxons estudados, quatro estão classificados como vulnerável (VU) na lista vermelha de fauna brasileira ameaçada de extinção (*P. pernambucensis*, *C. melanops nigrifrons*, *S. turdina intermadia* e *H. griseipectus naumburgae*) um como em perigo de extinção (EN) (*Thamnophilus aethiops distans*). Apesar dos quatro táxons inseridos na mesma categoria, as densidades populacionais encontradas no presente estudo foram, de fato, diferentes e consequentemente espera-se que as populações apresentem populações globais de tamanhos também diversos. As variações entre densidade e tamanho populacional encontradas, confirmam nossa hipótese de que espécies incluídas nas mesmas categorias da Lista vermelha de fauna Brasileira Ameaçada de Extinção, baseado somente em parâmetros de distribuição e

ocorrência podem apresentar estimativas populacionais bastante divergentes e consequentemente riscos de extinção distintos, mesmo quando ocorrem nas mesmas áreas.

Outros estudos com passeriformes neotropicais apresentaram resultados similares. Para a espécie Quase Ameaçada (NT) *Phylloscartes eximius* (Tyrannidae), natural da Mata Atlântica do Sudeste Brasileiro, a densidade populacional estimada foi de 0.13 i/ha, o que é bem próximo do valor encontrado para a Papa-taoca-de-Pernambuco e Flautim-marrom, e foi considerado um valor baixo quando comparado com outros dados não publicados (Tonetti & Pizo 2016). Para a espécie ameaçada *Habia atrimaxillaris* (Thraupidae) (EN), endêmica das florestas tropicais da Costa Rica, foram estimadas densidade populacionais variando entre 0.24 e 0.27 i/ha (Cornils et al 2015), evidenciando densidades populacionais relativamente baixas para *P. pernambucensis* e *S. turdina intermedia*.

Apesar de ser o táxon com grau de ameaça mais elevado (EN), a Choca-lisa (*T. a. distans*) apresentou densidade populacional maior que *P. pernambucensis* e *S. turdina intermedia*. Este fato, deve ser visto como positivo para o táxon, visto que a multiplicação de sua área de ocupação (AOO) estimada pelo ICMBio (ICMBio, 2018) pela densidade encontrada por nós resultaria em um tamanho populacional global de 5124 indivíduos, o que, de acordo com o critério C da IUCN (tamanho populacional) classificaria a espécie em um grau mais baixo de ameaça (de Em Perigo para Vulnerável), pois sua população de indivíduos maduros sexualmente estaria entre 2500 e 10.000 indivíduos (IUCN 2019). Considerando o critério C2a(i), que diz respeito ao número de indivíduos sexualmente maduros em cada subpopulação, um táxon pode ser considerado Em Perigo, quando cada subpopulação abriga entre 50 e 249 indivíduos em maturidade sexual e Vulnerável, quando cada subpopulação abriga de 250 a 1000 indivíduos (IUCN 2019). Com os dados de densidade estimadas para este táxon, seria necessária uma área de 1190 hectares para manter uma população de Choca-lisa com 250 indivíduos sexualmente maduros, e uma área de 4762 hectares para 1000 indivíduos. No CEP,

cerca de apenas 10 áreas possuem áreas equivalentes a 1000 e 2000 hectares, sendo algumas delas com registro de ocorrência de *T. aethiops distans*, o que significa que este táxon não deve ser classificado como Em Perigo baseado na categoria C2a(i). No entanto, áreas com mais de 4000 hectares não podem mais ser encontradas na região do CEP, o que justifica este táxon a não ser inserido na categoria Pouco Preocupante, de acordo com o mesmo critério, a não ser que grandes áreas florestais do CEP fossem reconectadas ou restauradas. Desta forma, com base em critérios demográficos aqui apresentados, este táxon alcançaria a categoria Vulnerável, no entanto, a categoria Em Perigo baseada na limitada AOO deverá prevalecer e permanecer.

Já o Flautim-marrom (*S. turdina intermedia*) demonstrou-se como o táxon de maior preocupação sob uma perspectiva demográfica. Para este táxon, com densidade populacional estimada de 0.14 i/ha, seria necessária uma área de 7143 hectares para abrigar uma população de 1000 indivíduos, e uma área de 1786 hectares para manter uma população mínima de 250. Como áreas maiores de 7000 ha não existem mais na região do CEP, e poucas estão próximas de 1000 ha, é justificável a classificação deste táxon determinada pelo comitê da Lista Vermelha Brasileira como VU. Desta forma, assim como a Choca-lisa, o flautim-marrom também não deverá mudar de categoria para pouco preocupante devido ao tamanho dos fragmentos que ocorre, a não ser que esses fragmentos fossem conectados ou restaurados.

A Papa-taoca-de-Pernambuco (*P. pernambucensis*) foi a segunda espécie com menor densidade populacional estimada com 0.15 i/ha. Baseado nesta estimativa, seria necessária uma área de 1600 ha para manter uma população de 250 indivíduos. Em nossa revisão de literatura foram identificadas pelo menos duas áreas com tamanho acima do necessário, com registros confiáveis da espécie, o que sugere que alterar sua categoria de VU para EN não é aplicável de acordo com o critério C. No entanto, nenhuma das áreas registradas seria capaz de manter uma subpopulação com mais de 1000 indivíduos, sugerindo que esta espécie não deverá migrar para categorias inferiores no futuro.

A Chupa-dente-de-máscara-preta (*Conopophaga melanops nigrifrons*), apresentou resultados razoáveis em relação a sua densidade populacional (0.35 i/ha), sendo necessária uma área de aproximadamente 715 hectares para uma população de 250 indivíduos, e 2860 ha para 1000 indivíduos. Associando-se o valor encontrado de densidade para este táxon à sua AOO sugerida no livro vermelho (inferior a 2000 km²), esperar-se-ia uma população global total de cerca de 70000 indivíduos, número acima do esperado para uma espécie classificada como pouco preocupante.

Já a Maria-de barriga-branca (*H. g. naumburgae*), a partir dos resultados obtidos foi considerada a espécie menos ameaçada, mesmo que classificada na mesma categoria das outras três. Considerando o critério C2a(i), o número total necessário de 1000 indivíduos por subpopulação, necessários para que uma espécie seja classificada como pouco preocupante, poderia ser atingido em uma área de pelo menos 1370 ha. Áreas deste tamanho são escassas na região do CEP, mas ainda existem. Adicionalmente, se a estimativa total de área de ocorrência (AOO) para este táxon for real (cerca de 2000 km²), a estimativa populacional global para este táxon é de 146.000 indivíduos, número bem acima do necessário para a classificação de uma espécie na categoria Pouco Preocupante (LC), baseado no critério voltado para tamanho populacional da IUCN (critério C) (IUCN 2019).

Certamente os táxons aqui estudados apresentam variações de densidade populacional entre suas áreas de ocupação, uma vez que esta sofrerá influência não apenas de fatores bióticos e intrínsecos de cada táxon, mas também de fatores abióticos, bem como da ação antrópica em cada área. Porém, mesmo com essas variações, nossas estimativas globais muito provavelmente subestimam o tamanho real das populações estudadas, uma vez que ainda existem diversos fragmentos do CEP não inventariados ou que não possuem nenhuma publicação científica correlata.

Sendo assim, estes cálculos são o suficiente para demonstrar que, de acordo com o critério focado em estimativas populacionais da IUCN (critério C), nenhum destes táxons é elegível para ser elevados a categorias de maior grau de ameaça, o que se demonstra positivo para as espécies. Os dados obtidos também nos permitem fazer inferências sobre o critério C2a (i), em relação ao número de indivíduos em cada subpopulação reconhecida, que é altamente aplicável a espécies de ocorrência em áreas altamente fragmentadas, como é o caso do CEP.

Em suma, extrapolações baseadas na densidade populacional estimadas no presente trabalho confirmam a vulnerabilidade de 4 dos 5 táxons analisados através de informações demográficas, sendo o tamanho estimado para as subpopulações o principal aspecto limitante para avaliação do grau de risco de extinção das espécies de aves do CEP. Os achados aqui apresentados corroboram com nossa premissa de que dados demográficos são importantes para indicar espécies prioritárias para conservação e otimizar o direcionamento dos recursos disponíveis para esses fins, visto que são escassos (Rodrigues et al 2006, Bennun et al 2018).

Para as espécies endêmicas ao CEP, algumas ações já foram indicadas, e estão principalmente associadas não apenas à manutenção e conservação dos remanescentes ainda existentes (Pereira et al, 2016; Melo et al, 2018), mas também à reconexão de fragmentos vizinhos através de corredores ecológicos e reflorestamento de áreas degradadas (Silva & Tabarelli, 2000; Barnett et al, 2005), a determinação de IBAs (Important Bird Areas) (Barnett et al, 2005; Pereira et al, 2016), e a realização de pesquisas científicas visando listar e identificar espécies e registrar seus locais de ocorrência (Pereira et al, 2014). Outra ação eficaz que vem sendo empregada com o auxílio de ONGs é a conversão de remanescentes em áreas protegidas, principalmente RPPNs (Reservas Particulares do Patrimônio Natural), uma vez que grande parte dessas áreas está inserida em propriedades particulares associadas à monocultura de cana-de-açúcar (Tabarelli & Roda, 2005).

Em relação aos táxons que ainda tiveram suas preferências de microhabitat estudadas (*P. pernambucensis* e *C. melanops nigrifrons*), é importante destacar que passeriformes tropicais insetívoros de sub-bosque são de grande interesse conservacionista pois são altamente vulneráveis à perda e fragmentação de habitat, estando entre os primeiros grupos a se extinguirem em áreas florestais perturbadas e o último a colonizar áreas de regeneração florestais (Peh et al., 2005; Pavlacky et al., 2015), uma vez que são espécies sedentárias e tendem a não cruzar áreas abertas (Powell et al., 2013, 2015a). Adicionalmente, compõem uma guilda de grande importância ecológica, controlando populações de insetos herbívoros que quando em desequilíbrio, podem ser altamente nocivos ao ecossistema (Şekercioğlu, 2006; Powell et al., 2015b; Nyfeller et al., 2018). Em regiões como o CEP, altamente fragmentadas e deterioradas, táxons ameaçados de extinção necessitam de ações intensas de manejo e proteção para sobreviver e persistir, o que significa que não apenas dados sobre estimativas populacionais, mas também em relação aos requerimentos básicos de habitat são extremamente necessários para ações viáveis e direcionadas aos táxons mais vulneráveis.

O presente estudo está entre os poucos fornecendo este tipo de informação para aves desta guilda na região neotropical, e poderá servir como dados comparativos para estudos futuros e contribuir para políticas públicas práticas direcionadas. Os parâmetros de microhabitat correlacionados às preferências dos dois táxons de passeriformes insetívoros de sub-bosque estudados estão mais diretamente associados a características da vegetação do sub-bosque do que a características do dossel, serrapilheira, declividade, altitude ou concentração de biomassa. Ambas espécies selecionaram pelo menos um parâmetro relacionado a densidade da vegetação.

Para *P. pernambucensis*, a seleção por locais com elevada densidade da vegetação tanto a 1.5m quanto a 0.5m de altura associado com a densidade mais baixa de árvores com DHB \leq 10 cm e a presença de cipós com valor próximo ao significativo no modelo 1 ($P= 0.077$) sugerem que esta espécie seleciona locais em estágio inicial de regeneração, quando

comparados aos pontos aleatórios coletados, onde a alta incidência de luz solar favorece o crescimento de vegetações herbáceas no solo da mata. Apesar da presença de *C. melanops nigrifrons* também parecer estar relacionada a áreas em regeneração, este táxon demonstrou escolha por locais mais maduros, com concentração de cipós e sub-bosque mais denso, mas onde plantas herbáceas já não estão mais presentes.

Em florestas tropicais, áreas com a vegetação mais densa podem surgir de clareiras, devido à queda de grandes árvores por exemplo. A presença dessas clareiras tem forte influência na presença de diversos grupos de aves tropicais (Banks-Leite & Cintra 2008). No entanto, a não correlação entre os táxons alvos de estudo e a abertura do dossel sugere que estes táxons não buscam por clareiras recém formadas.

Para Passeriformes insetívoros de sub-bosque, a seleção de microhabitat com vegetação mais densa pode estar relacionada a disponibilidade de alimento, como já foi demonstrado em estudos em florestas tropicais, em que a presença de folhas jovens pode atrair insetos herbívoros, o que consequentemente irá atrair aves insetívoras (Richard & Windsor 2007). É importante destacar que este estudo foi desenvolvido na estação seca da região, em que a abundância de insetos pode estar reduzida (Devries & Walla 2001, Coutinho-Silva et al 2017), o que permite sugerir que os microhabitat selecionados podem ser de grande importância para a eficiência do forrageamento em períodos de escassez de alimentos.

Os resultados aqui apresentados reiteram estudos anteriores sobre a heterogeneidade de microhabitat em florestas neotropicais e sobre as especificidades que diferentes espécies de aves podem demonstrar em relação à um mesmo habitat (Cintra & Cancelli 2008). Isto sugere que a manutenção da heterogeneidade do habitat é uma grande meta a ser alcançada para possibilitar a conservação de comunidades de aves do CEP. Porém, não é sabido se as atividades de extrativismos que ocorreram no passado contribuíram ou não para o aumento dos tipos de habitat selecionados pelos táxons estudados. Caso tenham, a densidade populacional destes

podem estar temporariamente aumentadas e deverão decrescer enquanto os fragmentos florestais alcançam estágios de regeneração mais avançados, ressaltando a importância do monitoramento destas populações.

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5. Anexos:

Anexo 1: Resumo dos critérios utilizados pelo método IUCN para categorização de risco de extinção (ICMBio, 2018a).

A. Redução da População (Declínio medido ao longo de 10 anos ou 3 gerações, o que for mais longo)			
	Criticamente em Perigo	Em Perigo	Vulnerável
A1	≥ 90%	≥ 70%	≥ 50%
A2, A3 e A4	≥ 80%	≥ 50%	≥ 30%
A1 Redução da população observada, estimada, inferida ou suspeitada de ter ocorrido no passado, sendo as causas da redução claramente reversíveis E compreendidas OU não ter cessado. A2 Redução da população observada, estimada, inferida ou suspeitada de ter ocorrido no passado, sendo que as causas da redução podem não ter cessado OU não ser compreendidas OU não ser reversíveis. A3 Redução da população projetada ou suspeitada de ocorrer no futuro (até um máximo de 100 anos). A4 Redução da população observada, estimada, inferida, projetada ou suspeitada, sendo que o período de tempo deve incluir tanto o passado quanto o futuro (até um máximo de 100 anos), e as causas da redução podem não ter cessado OU não ser compreendidas OU não ser reversíveis.			(a) observação direta; (b) índice de abundância apropriado para o táxon; (c) declínio na área de ocupação, extensão de ocorrência e/ou qualidade do habitat; (d) níveis reais ou potenciais de exploração; (e) efeitos de táxons introduzidos, hibridação, patógenos, poluentes, competidores ou parasitas.
B. Distribuição geográfica restrita e apresentando fragmentação, declínio ou flutuações			
	Criticamente em Perigo	Em Perigo	Vulnerável
B1 Extensão de ocorrência	< 100 km ²	< 5.000 km ²	< 20.000 km ²
B2 Área de ocupação	< 10 km ²	< 500 km ²	< 2.000 km ²
E pelo menos 2 dos seguintes itens: (a) População severamente fragmentada, OU número de localizações = 1 ≤ 5 ≤ 10 (b) declínio continuado em um ou mais dos itens: (i) extensão de ocorrência; (ii) área de ocupação; (iii) área, extensão e/ou qualidade do habitat; (iv) número de localizações ou subpopulações; (v) número de indivíduos maduros. (c) flutuações extremas em qualquer um dos itens: (i) extensão de ocorrência; (ii) área de ocupação; (iii) número de localizações ou subpopulações; (iv) número de indivíduos maduros.			
C. Tamanho da população pequeno e com declínio			
	Criticamente em Perigo	Em Perigo	Vulnerável
Número de indivíduos maduros	< 250	< 2.500	< 10.000
E C1 ou C2			
C1 Um declínio continuado observado, estimado ou projetado de pelo menos (até um máximo de 100 anos no futuro): 25% em 3 anos ou 1 geração 20% em 5 anos ou 2 gerações 10% em 10 anos ou 3 gerações			
C2 Um declínio continuado observado, estimado, projetado ou inferido E pelo menos uma das 3 condições: (i) número de indivíduos maduros em cada subpopulação: ≤ 50 ≤ 250 ≤ 1.000 (a) (ii) ou % indivíduos em uma única subpopulação 90–100% 95–100% 100%			
(b) flutuações extremas no número de indivíduos maduros			
D. População muito pequena ou distribuição muito restrita			
	Criticamente em Perigo	Em Perigo	Vulnerável
D Número de indivíduos maduros	< 50	< 250	D1. < 1.000
D2 Área de ocupação restrita ou número de localizações, sob uma ameaça futura plausível de levar o táxon à condição de CR ou EX em curto prazo.	-	-	D2. Tipicamente AOO < 20 km ² ou Número de localizações ≤ 5
E. Análises quantitativas			
	Criticamente em Perigo	Em Perigo	Vulnerável
Indicando que a probabilidade de extinção na natureza é de:	≥ 50% em 10 anos ou 3 gerações	≥ 20% em 20 anos ou 5 gerações	≥ 10% em 100 anos