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Larissa Gomes de Jesus

Laterality in Psittaciformes: evaluation of behavioral and temperament aspects

Juiz de Fora
2024

Larissa Gomes de Jesus

Laterality in Psittaciformes: evaluation of behavioral and temperament aspects

Dissertação apresentada ao Programa de Pós-graduação em Biodiversidade e Conservação da Natureza da Universidade Federal de Juiz de Fora como requisito parcial à obtenção do título de Mestre Ciências Biológicas. Área de concentração: Biodiversidade e conservação da Natureza

Orientadora: Doutora Aline Cristina Sant'Anna

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BANCA EXAMINADORA

Doutora Aline Cristina Sant'Anna - Orientadora
Universidade Federal de Juiz de Fora

Profa. Dra. Selene Siqueira da Cunha Nogueira
Universidade Estadual de Santa Cruz

Profa. Dra. Lilian Tonelli Manica
Universidade Federal do Paraná

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RESUMO

A lateralidade é caracterizada como uso assimétrico dos hemisférios cerebrais e estruturas bilaterais. Em psittacíformes, pode ser expressa pela preferência de uso da visão monocular ou uso de um pé específico para alimentação. O objetivo dessa dissertação foi avaliar a lateralidade em Psittacíformes neotropicais e relacioná-la ao temperamento, emocionalidade e interações sociais através de uma revisão bibliográfica e aplicação de metodologias estabelecidas. Os objetivos específicos dessa dissertação foram: reunir informações sobre lateralidade em Psittacíformes através de uma revisão de escopo (RS) e análises bibliométricas (Capítulo 1); identificar a lateralidade em três espécies de psitacídeos (*Psittacara leucophthalmus*, *Primolius maraca*, *Pionius maximiliani*) e relacionar o fenômeno a aspectos do temperamento e sociabilidade (Capítulo 2). No Capítulo 1, 20 publicações foram incluídas na RS sendo que 40% delas foram realizadas na Austrália, 35% nos Estados Unidos da América e 5% no Brasil. A lateralidade foi estudada em 31 gêneros de Psittacíformes, dos quais 77% ocorrem na Oceania, 16% na América e 6% na África. A predominância de uso do pé para alimentação foi avaliada em 80% dos estudos, relatando preferência média de 61% pelo pé esquerdo (variando de 5% a 100%). A preferência ocular foi avaliada em 20% dos estudos. O tópico mais investigado foi a relação entre lateralidade e cognição (20%). No Capítulo 2, os resultados apontaram a presença da lateralidade nas três espécies analisadas, ressaltando a preferência pelo olho direito. Para avaliar a força da lateralização, utilizamos o índice de lateralidade (LI), que varia de -1 (totalmente destro) a 1 (totalmente canhoto). Para a preferência por um membro pra alimentação e apoio unipodal, não houve uma direção predominante. Correlações significativas entre as preferências visuais e as dimensões do temperamento foram observadas (atividade, ousadia, ansiedade e proximidade com o ser humano). Quanto maior a atividade, menor o LI total e LI no teste de seixos e sementes, ou seja, maior a preferência pelo olho direito. As aves mais ousadas e mais ansiosas tiveram menor LI no teste de reação à pessoa, o que também

resulta em preferência pelo olho direito. A última correlação representa uma maior evitação ao humano e maior o uso do olho direito (menor LI no teste do novo objeto). Não foram encontradas correlações significativas entre a lateralidade e as interações sociais positivas e negativas. Os resultados dessa dissertação acrescentam dados à literatura científica sobre a lateralidade em psitacíformes, além de revelar o panorama geral sobre o tema. Além disso, nossos achados reiteram os benefícios da relação entre lateralidade e as diferenças individuais para o bem-estar animal, possibilitando uma forma não invasiva de identificar as aves mais propensas ao estresse, medo e agressividade. Assim, é possível criar estratégias personalizadas para manejo de animais em cativeiro, levando em consideração as respostas emocionais individuais. Esse foi o primeiro estudo a identificar e relacionar a lateralidade com dimensões do temperamento em Psittacíformes.

Palavras-chave: assimetrias laterais, cognição, papagaios, personalidade.

ABSTRACT

Laterality is characterized by the asymmetrical use of cerebral hemispheres or bilateral structures. In psittacids, it can be expressed through the preference for using monocular vision or a specific foot for feeding. The objective of this dissertation was to evaluate laterality in Psittaciformes and relate it to temperament, emotionality, and social aspects by reviewing the scientific literature and applying established methodologies to identify this feature in Neotropical Psittaciformes. The specific objectives of this dissertation were: to gather information on laterality in Psittaciformes through a scoping review (SR) and bibliometric analyses (Chapter 1); to identify laterality in three species of psittacids (*Psittacara leucophthalmus*, *Primolius maracana*, *Pionus maximiliani*) and to relate the phenomenon to aspects of temperament and sociability (Chapter 2). In Chapter 1, 20 publications were included in the SR, with 40% conducted in Australia, 35% in the United States, and 5% in Brazil. Laterality was studied in 31 genera of Psittaciformes, of which 77.4% occur in Oceania, 16.1% in America, and 6.4% in Africa. The preference for using a specific foot for feeding was evaluated in 80% of the studies, reporting an average left-foot preference of 61.1% (ranging from 5.0% to 100%). Eye preference was evaluated in 20% of the studies. The most investigated topic was the relationship between laterality and cognition (20.0%). In Chapter 2, the results indicated the presence of laterality in the three analyzed species, highlighting a preference for the right eye. The strength of laterality was analyzed by laterality index which ranges from -1 (totally right-eyed) to 1 (totally left-eyed). For footedness and unipedal support, there was no predominant direction. Significant correlations between visual preferences and temperament dimensions were observed (activity, boldness, anxiety, and closeness to humans). The higher the activity, the lower the total LI and LI in the pebble and seed test, indicating a greater preference for the right eye. The bolder and more anxious birds had a lower LI in the reaction to person test, also resulting in a preference for the right eye. Greater avoidance of humans correlated with increased right eye use (lower LI in the novel object test). No significant

correlations were found between laterality and positive or negative social interactions. The results of this dissertation add data to the scientific literature on laterality in Psittaciformes and provide an overview of the topic. Additionally, our findings reinforce the benefits of understanding the relationship between laterality and individual differences for animal welfare, offering a non-invasive method to identify birds more prone to stress, fear, and aggression. This enables the elaboration of personalized management strategies for captive animals, considering individual emotional responses. This was the first study to identify and relate laterality with temperament dimensions in Psittaciformes.

Keywords: cognition; lateral asymmetries; parrots; personality.

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LIST OF ABBREVIATIONS AND ACRONYMS

SR: Scoping Review

L: Left Footed

R: Right Footed

NL: Non-Lateralized

PD: Predominant Direction

LI: Laterality Index

DP: Directional Preferences

OUT: Outcome

ASAS: Area de Soltura de Animais Silvestres

CETAS: Centro de Triagem de Animais Silvestres

IBAMA: Instituto Brasileiro do Meio Ambiente

IEF: Instituto Estadual de Florestas

NO: Novel object

RP: Reaction to person

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1 GENERAL INTRODUCTION

Laterality, or the specialization of the brain hemispheres to process information differently and asymmetric behaviors (Vallortigara; Rogers, 2005; Rogers, 2010), is a feature widespread among animals such as bees (Anfora et al., 2010), amphibians (Keuroghlian-Eaton et al., 2024), pigs (Goursot et al., 2018), birds (Franklin; Lima, 2001), and humans (MacNeilage et al., 2009). Therefore, hemispheric lateralization could be a common feature of the brain and its evolutionary history (Anfora et al., 2010). Among the advantages of having a lateralized brain is the potential increase in neural capacity by allowing each hemisphere to specialize in specific tasks, thus avoiding redundant functions while maintaining the same brain volume (Vallortigara; Rogers, 2005; Kaplan; Rogers, 2021). However, laterality could have disadvantages for survival due to predictable behaviors; predators can anticipate the direction of escape of their prey and adjust their hunting strategies to fit the prey's behavior (Rogers, 2000).

Evidence suggests that the avian brain is as lateralized as the human brain (Rogers, 2008). Testing the monocular visual field is a noninvasive alternative to identify hemispheric asymmetries. A study conducted with chicks incubated in dark and light conditions revealed that light-incubated chicks were capable of developing functional laterality by using the right eye to identify seeds and avoid distracting targets, and the left eye to analyze a model predator (Dharmaretnam; Rogers, 2005). From this information, it is possible to infer hemispheric specialization. The left hemisphere (right eye) is responsible for focused attention and categorizing different stimuli, not easily distracted, and controlling routine behaviors. In contrast, the right hemisphere (left eye) is responsible for global attention (easily distracted), detecting and responding to threatening stimuli such as predators, and controlling emergency responses such as escape, fear, and aggression (Rogers, 2008; 2010).

Emotional responses are also lateralized between the hemispheres. The affective and motivational hypotheses suggest that positive emotions or approach motivations are processed by the left hemisphere, while negative emotions or withdrawal motivations are processed by the right hemisphere (Rogers, 2010; Goursot et al., 2021). Another hypothesis proposes that the behavioral activation system, which is involved with reward sensitivity and regulates approach behaviors, is regulated by the left hemisphere, while the behavioral inhibition system, which regulates approach-avoidance behaviors, is processed by the right hemisphere (Gable et al., 2018; Goursot et al., 2021). The right hemisphere is also involved in physiological stress responses such as heart rate and the hypothalamic-pituitary-adrenal axis (Rogers, 2010; Goursot et al., 2021).

The different functions of the hemispheres can lead to differential reactions to environmental stimuli, which may be expressed as individuals' behavioral differences consistent over time, or temperament (Réale et al., 2007; Rogers, 2009). Studies indicate that right-biased pigs are bolder and more sociable (Goursot et al., 2019). The relationship between laterality and boldness was also reported for cichlids (*Archocentrus nigrofasciatus*), where strongly lateralized individuals were bolder and explored novel environments more (Reddon; Hurd, 2008). While laterality and temperament have been studied in numerous species, Psittaciformes remain largely unexplored in this regard. Therefore, understanding the relationship between laterality and temperament in Psittaciformes is important for explaining differences in individual behaviors, with potential positive and negative biases.

Among birds, laterality in the Psittaciformes group has been studied for many years, with the first record dating back to the early 20th century (Friedman; Davis, 1938). It can be expressed as foot preferences when manipulating food items and eye preferences when viewing certain scenes (Magat; Brown, 2009). Phylogenetic analyses indicate the existence of a strongly lateralized ancestor with a large body size that fed on big seeds. As body size decreased, so did

laterality, shifting their feeding habits to smaller seeds that did not require manipulation (Brown; Magat, 2011b).

Therefore, understanding lateral preferences could be an important tool for animal welfare, as it can help identify individuals prone to greater vulnerability to stress and more reactive to novel stimuli (Rogers, 2009; Goursot et al., 2019). Furthermore, studies relating to laterality, affective styles, and temperament can guide more individualized welfare strategies for captive animals. Recognizing differences in their daily routines can enable the identification of those who are more resilient, reactive, aggressive, or fearful (Goursot et al., 2021). It could be particularly helpful when managing these animals in captivity facilities.

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2 GENERAL OBJECTIVES

The objective of this dissertation was to evaluate laterality in Psittaciformes and relate it to temperament, emotionality, and social aspects by reviewing the scientific literature and applying established methodologies to identify this feature in Neotropical Psittaciformes.

2.1 Specific objectives

Chapter 1: gather scientific evidence regarding laterality in this taxonomic group, through a SR and to provide a summarization of the various mechanisms (cognitive, motor, visual, and ontogenetic) through which lateral preferences are expressed within the Psittaciformes.

Chapter 2: to evaluate whether three neotropical psittacid species (*Psittacara leucophthalmus*, *Pionus maximiliani*, and *Primolius maracana*) exhibit laterality in behavior such as footedness, eye preferences and unipedal support, and to correlate the bird's laterality to their temperament and social interactions.

3 CHAPTER 1

Cognitive and behavioral aspects of laterality in Psittaciformes: A Scoping Review

Larissa Gomes^a, Maria G. Marçal-Pedroza^a, Gabriela Ramos^a, Maria Eugênia Andrighetto Canozzi^b,
Aline Cristina Sant'Anna^c

^a Postgraduation Program in Biodiversity and Nature Conservancy, Federal University of Juiz de Fora, 36.036-330, Juiz de Fora, MG, Brazil.

^b Instituto Nacional de Investigación Agropecuaria (INIA), Estación Experimental La Estanzuela, Ruta 50 km 11, Colonia 70000, Uruguay.

^c Department of Animal Science at School of Agricultural and Veterinarian Sciences of the São Paulo State University (Unesp), 14.884-900, Jaboticabal, SP, Brazil

Resumo

A lateralidade é definida como assimetrias de estruturas bilaterais ou viéses comportamentais. A lateralidade se manifesta como a preferência por um membro específico (predominância de uso do pé ou mão) ou por um campo monocular de visão (preferência ocular). Este estudo teve como objetivo reunir informações sobre lateralidade em Psittaciformes utilizando revisão de escopo (RS) e análises bibliométricas. A pesquisa incluiu quatro bases de dados (Web of Science, APA, Scopus e CABI). Consideramos como critérios de inclusão publicações revisadas por pares e anais de conferências. A busca inicial resultou em 90 citações, mas apenas 20 publicações estavam relacionadas ao tema de interesse e foram incluídas na RS. Entre elas, 40% foram realizadas na Austrália, 35% nos EUA e 5% no Brasil. A lateralidade foi estudada em 31 gêneros de Psittaciformes, dos quais 77% ocorrem na Oceania, 16% na América e 6% na África. A predominância de uso do pé foi avaliada em 80,0% dos estudos, relatando uma preferência média de 61.1% (variando de 5,0% a 100%) pelo pé esquerdo. A preferência ocular

foi avaliada em 20% dos estudos. O tópico mais investigado foi a relação entre lateralidade e cognição (20%), seguido da relação entre lateralidade e aspectos filogenéticos, tamanho do cérebro, tamanho lexical e anatomia da retina. A RS revelou novas oportunidades para expandir a pesquisa sobre aspectos da lateralidade, como assimetrias específicas de contexto e avaliações em espécies neotropicais.

Palavras-chave: Assimetrias laterais; comportamento; footedness; papagaios; preferências laterais.

Abstract

Laterality is defined as asymmetries of bilateral structures or behavioral biases. Laterality manifests as the preference for a specific limb (footedness) or for a monocular field of vision (eye preference). This study aimed to gather information about laterality in Psittaciformes using scoping review (SR) and bibliometric analyses. The search included four databases (Web of Science, APA, Scopus, and CABI). We considered peer-reviewed publications and conference proceedings as inclusion criteria. The initial search resulted in 90 citations, but only 20 publications were related to the topic of interest and included in the SR. Among them, 40.0% were conducted in Australia, 35.0% in the US, and 5.0% in Brazil. Laterality was studied in 31 Psittaciformes genera, 77.4% of them occur in Oceania, 16.1% in America, and 6.4% in Africa. Footedness was assessed in 80.0% of the studies, reporting prevalences of left-foot preference at 61.1% on average (ranging from 5.0% to 100%). Eye preference was evaluated in 20.0% of the studies. The most investigated topic was the relationship between laterality and cognition (20.0%), followed by the relationship between laterality and phylogenetic aspects, brain size, lexical size, and retinal anatomy. The SR revealed new opportunities for expanding research on

aspects of laterality, such as context-specific asymmetries and evaluations in Neotropical species.

Keywords: Behavior; footedness; lateral asymmetry; lateral preference; parrots.

1 Introduction

Laterality is defined as the presence of asymmetries in bilateral structures or behavioral biases (Vallortigara; Rogers, 2005). Traditionally, laterality was believed to be a characteristic exclusive to humans. However, it is now known that laterality is widespread across various species, including chordates such as fish and amphibians (Vallortigara; Rogers, 2005). This fact suggests that hemispheric lateralization may be a common feature of brains with diverse structures and evolutionary history (Anfora et al., 2010).

There are numerous examples of how laterality can be expressed in both vertebrate and invertebrate animals. For instance, bees are capable of learning and associating odors by extending their proboscis, with their learning enhanced when they use the right antenna (Anfora et al., 2010). Anuran amphibians exhibit faster reactions when a predator is in their left monocular field of vision, indicating that fear and predator evasion responses are processed in the right side of their brain (Lippolis et al., 2002). Squids and fish show a preference for swimming counterclockwise when preying on shrimps (Karim et al., 2016). Additionally, flies, ants, spiders, and snails demonstrate behavioral asymmetries, such as a preference for a specific eye or asymmetries in courtship behavior (Rogers, 2012). These examples highlight the diverse ways in which laterality manifests across various taxa.

Brain asymmetries have potential to increase neural capacity (Kaplan; Rogers, 2021). As one hemisphere becomes specialized in a specific task, the other hemisphere is available to learn and perform other functions preventing interference from the contralateral hemisphere and avoiding redundant functions (Vallortigara; Rogers, 2005). Also, it allows a greater range of functions to be performed while maintaining the same brain volume, as larger brains are more energetically costly (Wiper, 2017). By specializing and dividing tasks between hemispheres, organisms can optimize their cognitive abilities and efficiency (Kaplan; Rogers, 2021).

However, depending on the context, laterality may have disadvantages, for example in predator-prey interactions. Predators can learn and exploit the lateral biases of their prey, making them more susceptible to predation (Vallortigara; Rogers, 2005). If prey consistently shows a biased preference for certain directions or exhibits predictable lateral behaviors, predators can anticipate and adjust their hunting strategies accordingly (Vallortigara; Rogers, 2005). The poeciliid fish (*Brachyrhaphis episcopi*) is an example, as it loses potential prey by swimming toward the favored side instead of following the food (Brown et al., 2004; Wiper, 2017).

The order Psittaciformes (Class Aves) comprises three families: Strigopidae (four species of New Zealand parrots), Cacatuidae (22 species from Oceania), Psittaculidae (202 species from Africa, Asia and Oceania), and Psittacidae (177 species of America and African parrots) (Birds of the World, 2022). In this group, for example, the first evidence of laterality dates to the first half of the 20th century (Friedman; Davis, 1938). Since then, studies revealed that parrots exhibit laterality through their preference for one side of monocular vision, which indicates the corresponding contralateral hemisphere's involvement (Brown; Magat, 2011a). They also demonstrate laterality through the use of their feet to manipulate food items (Rogers, 1980). In psittacids, laterality seems to result in differences in information processing. The left hemisphere is associated with roles such as foraging, discerning and manipulating food items, attention, and recognition of vocalizations, while the right hemisphere is related to controlling sexual behavior, spatial recognition, and reactions to predators (Vallortigara; Rogers, 2005). Thus, these functional asymmetries in the parrot brain contribute to their specialized cognitive abilities and behavioral responses.

Previous narrative reviews on laterality included information about parrots. Harris (1989) exclusively focused on footedness in parrots. Cohen (2012) reviewed asymmetries of neurobehaviors in humans and other vertebrates, including information about parrots. Rogers

(2017) also conducted a broader review on the strength of lateralization in relation to behavior and cognitive performance in vertebrates, which included data about parrots. Currently, there is no comprehensive summary of scientific data about laterality in parrots. This gap can be better addressed through a scoping review (SR), which would provide a detailed overview of the methodologies used and a systematic synthesis of the existing knowledge. Conducting a SR would enable us to point out gaps in the topic of interest and guide future studies. Based on the previous evidence of laterality in Psittaciformes, this study aimed to gather scientific evidence regarding laterality in this taxonomic group, through a SR and to provide a summarization of the various mechanisms (cognitive, motor, visual, and ontogenetic) through which lateral preferences are expressed within the Psittaciformes.

2 Material and methods

2.1 Search protocol

Ethical approval was not necessary because this is a theoretical study. The SR followed the PRISMA protocol (Page et al., 2021). The search strategy was based up on the PICO terms: Population, Intervention, Comparison, and Outcome (Brown et al., 2006). The population of interest was parrot OR parakeet OR psittacine OR psittacidae OR psittaciformes OR psittacids. The intervention was laterality OR “lateral asymmetry”. The comparison was “foot preference” OR “eye preference” OR footedness OR eyedness OR “ocular dominance”. The outcomes were left OR right OR ambidextrous OR non-lateralized.

Our search was conducted in November of 2022 and updated in February 2024, through four databases (CABI, Web of Science, Scopus, and APA). The search included peer-reviewed papers and conferenced proceedings published from 1938 to 2024.

2.2 Inclusion criteria and screening process

The inclusion and exclusion criteria were established through consensus among four of the co-authors (LG, MGMP, GR, ACSA). Publications that solely focused on laterality or parrots (but not both) and those that referred to lateral differences unrelated to cerebral hemispheric lateralization were excluded. Publications that specifically investigated laterality (footedness or eye preference) in parrots, whether correlated with other behavioral characteristics or not, were included. Only publications written in English, Portuguese, or Spanish were considered acceptable for inclusion in the SR. No additional restrictions were imposed concerning the publication year, sample sizes, or the quality of the journals. All the selected publications (**Table 1**) reported the presence of laterality in Psittaciformes. We evaluated parameters such as footedness and eye preference to enhance our research.

The search produced the following results: CABI (5), Web of Science (42), Scopus (39), and APA (4). All the results (n=90) were imported into the EndNote Web software for organization and removal of duplicate references (n=54). The remaining publications underwent six stages of triage (**Figure 1**): Step 1-: titles and abstracts were evaluated to identify and remove citations that were not relevant to the SR; step 2-:the full text of the publications was read and assessed to identify and remove publications that did not fit this SR; and step 3- reference lists of the literature reviews published by Harris (1989), Cohen (2012), and Rogers (2017) were checked to identify relevant publications that may not have been found in our initial SR; step 4- publications obtained from the literature reviews were evaluated to determine if they were available and eligible and those not eligible were excluded based on the criteria described below; step 5- publications from the literature review were evaluated to identify and

remove publications that did not fit the SR; step 6- all eligible publications were read, and relevant information was extracted.

Table 1: Publications included in the SR, along with the source, type of laterality studied, correlations, and genres.

Author	Source	Laterality Type	Correlate traits	Genus
Friedman and Davis, (1938)	The Auk	Footedness	Not applicable	<i>Amazona; Coracopsis; Psittacula; Ara; Aratinga; Tanygnathus; Brotogeris</i>
McNeil et al., (1970)	Ibis	Footedness	Total length of homologue legs	<i>Aratinga</i>
Rogers, (1980)	Bird Behavior	Footedness	Not Applicable	<i>Cacatua; Calyptorhynchus; Platycercus; Callocephalon</i>
Snyder et al., (1996)	TENNET VII	Footedness	Age, sex, legband, handedness of owner	<i>Cacatua; Psittacus; Poicephalus; Amazona; Ara; Anodorhynchus</i>
Snyder and Harris, (1997)	Neuropsychologia	Footedness	Lexical size	<i>Psittacus</i>
Snyder and Harris, (1998)	TENNET VIII	Footedness	Lexical size	<i>Psittacus</i>
Snyder and Bonner, (2001)	TENNET XI	Footedness	Ontogenesis	<i>Psittacus</i>
Magat and Brown, (2009)	Proceedings of the Royal Society Biological Sciences	Footedness and eye preference	Cognition and problem-solving	<i>Nymphicus; Melopsittacus; Eolophus; Callocephalon; Calyptorhynchus; Cacatua; Alisterus; Polytelis</i>
Brown and Magat (2011,a)	Biology letters	Footedness and eye preference	Body size, primary diet	<i>Neopsephotus; Melopsittacus; Nymphicus; Platycercus ; Cacatua; Alisterus; Glossopsitta; Trichoglossus; Psephotus; Calyptorhynchus ; Aprosmictus; Polytelis; Neophema;</i>

Brown and Magat, (2011,b)	Behavioral Ecology	Footedness	Phylogenetic	<i>Cacatua; Callocephalon; Calyptorhynchus; Nymphicus; Probosciger; Eclectus; Alisterus; Aprosmictus; Polytelis; Neophema; Platycercus; Barnardius; Purpureicephalus; Psephotus; Glossopsitta; Psitteuteles; Trichoglossus; Melopsittacus; Neopsephotus</i>
Randler et al., (2011)	Laterality	Footedness	Not applicable	<i>Psittacula</i>
Schiffner and Srinivasan, (2013)	PLOS One	Footedness	Social behavior	<i>Melopsittacus</i>
Coimbra et al., (2014)	The Journal of Comparative Neurology	Footedness and eye preference	Anatomy of the ocular retina	<i>Calyptorhynchus; Cacatua; Nymphicus; Eolophus</i>
Cussen and Mench, (2014)	Animal Cognition	Footedness	Sex, foraging, cognitive flexibility, and long-term memory	<i>Amazona</i>
Duggan et al., (2016)	South Australian Ornithologist	Footedness	Social and aggression behaviors	<i>Melopsittacus</i>
d'Antonio-Bertagnolli and Anderson, (2018)	Laterality	Footedness	Cognition behaviors and solving tasks	<i>Melopsittacus</i>
Berg et al., (2020)	Evolutionary Ecology	Footedness	Latitudinal habitat	<i>Platycercus</i>

Godinho et al., (2020)	Animal Cognition	Footedness and Eye preference	Cognition behaviors and solving tasks	<i>Amazona</i>
Kaplan and Rogers, (2021)	Symmetry	Footedness	Brain size	<i>Eolophus; Cacatua; Calyptorhynchus; Callocephalon; Platycercus, Melopsittacus; Nymphicus; Alisterus; Eclactus; Neophema; Aprosmictus; Barnardius; Purpureicephalus; Psephotus; Glossopsitta; Psitteuleles; Trichoglossus; Neopsephotus</i>
Regaiolli et al., (2021)	Applied Animal Behavior Science	Footedness	Cognition and problem-solving	<i>Ara</i>

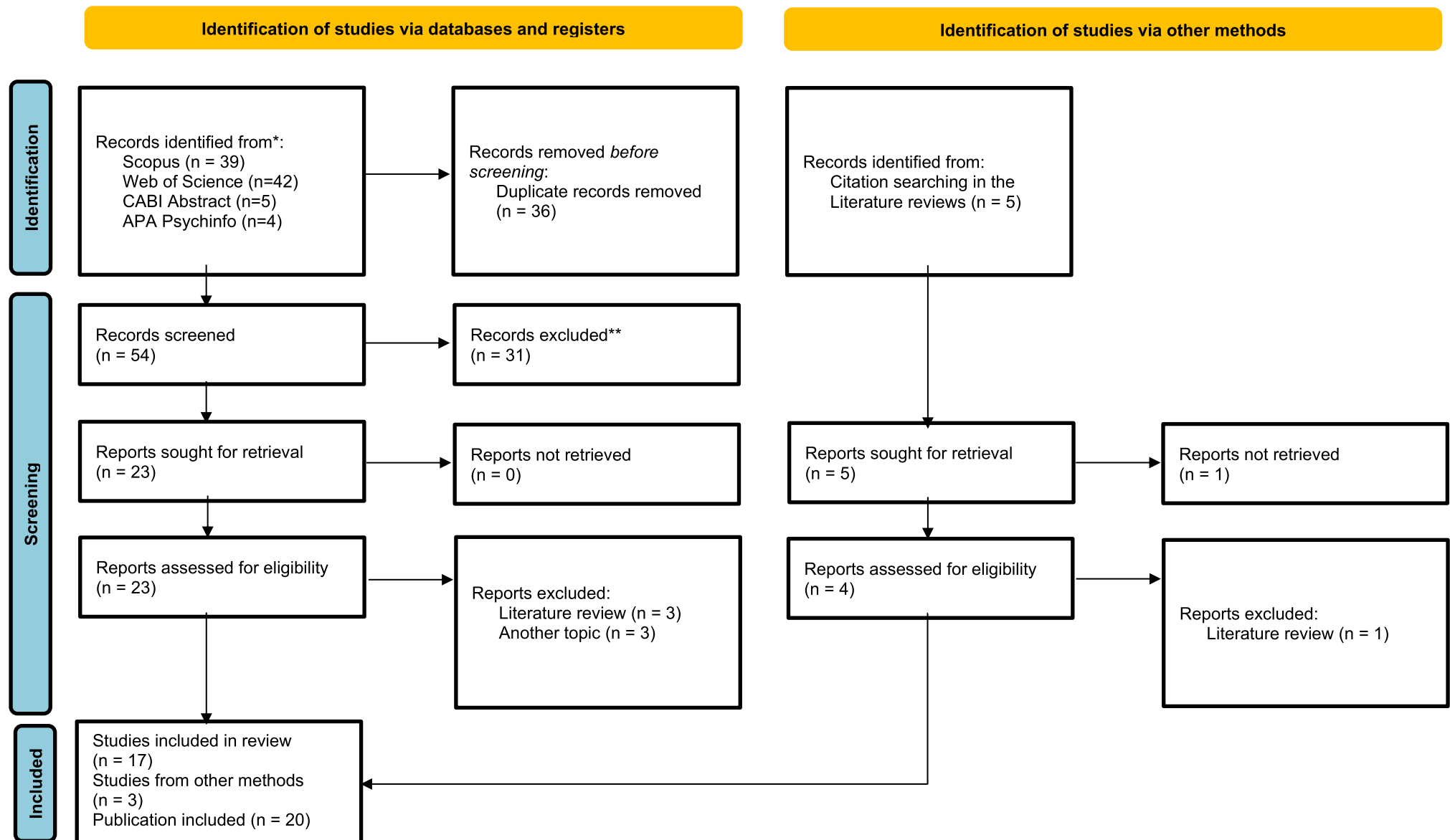


Figure 1: Prisma Flow Diagram (Page et al., 2021), contained the number of publications resulting from the search protocol and the number of publications filtered on each stage of the screening processes from the SR.

2.3 Data extraction and analyses

The publications were read and screened independently by three co-authors (LG, MGMP, and GR). The extracted information were: journal name, publication type, study design, the location of the studies, the genus of the parrots examined, number of specimens and whether the individuals were from captive aviaries or wild animals, definition of laterality, the type of laterality studied, the methodology used to measure laterality, how many times it was measured (repetitions), the percentage of left-footed individuals, the percentage of left-eyed individuals, any correlated traits investigated and interest outcome.

Reliability of the search terms used in this SR was assessed by a word cloud (WordCloud R Package; Fellows, 2018). To build the word cloud, we used words of the title and the keywords of all the included publications.

Non-parametric analyses are often preferred in laterality research because the behavioral outcomes in laterality studies usually consists of categorical variables (e.g., ‘left’, ‘right’, ‘non-lateralized’ or ‘ambidextrous’), which does not fit the assumptions of parametric tests. The non-parametric analyses provide a more conservative and appropriate statistical analysis due to the nature of the data (McGrew; Marchant, 1998). In this study we summarized the quantitative data (quantitative synthesis) only for footedness, as only three publications used eye preference as a measure of laterality. In the present study, the following analyses were reported, using the numerical data included in the publications when available:

- To evaluate the percentage of outcomes (OUT = [\sum study * species studied in each publication]) with evidence of laterality, we defined a cut-off point where laterality was characterized as ‘moderate’ if the study reported more than 55% of lateral bias for the referred species (Wiper, 2017), and ‘strong’ laterality when more than 80% lateral bias was reported. In the present study, the total number of outcomes was OUT = 108.
- The consistency of evidence was evaluated according to Eq.1, where the ‘number of outcomes that found the evidence’ included those outcomes in which at least moderate laterality was reported, regardless of the direction (left or right).

$$\text{Consistency of evidence}^* = \frac{\text{number of outcomes that found the evidence}}{\text{total number of outcomes that evaluated the evidence}} \times 100 \text{ (Eq.1)}$$

- To evaluate the predominant direction (PD) of laterality, we first calculated the number of outcomes showing evidence of laterality to left (L), right (R), or non-lateralized (NL), using moderate laterality (above 55% of lateral bias) as the cut-off point. It resulted in L = 59 outcomes, R = 26 outcomes, and NL = 23 outcomes. This data was used in equation Eq.2 (England et al., 2020; Harrison et al., 2014). If PD values were higher than 0, the results indicate predominant direction to left and if PD values were smaller than 0, the results indicated a predominant direction to the right or non-lateralized.

$$\text{PD}^* = \frac{\sum(L) - \sum(R+NL)}{\text{total number of outcomes that evaluated the evidence}} \text{ (Eq.2)}$$

- The Laterality Index (LI) is an equation frequently used to describe quantitatively the direction and intensity of lateral bias: $LI = (R-L)/(R+L)$ (Hopkins, 1999). The resulting values range from -1.0 to 1.0 with the absolute value reflecting the strength of limb preferences. There is a second LI equation that focuses on directional preferences $DP =$

$R/ L+R \times 100$ (Hopkings et. al, 2016; Wiper, 2017). The results from the second equation had a cut-off value of 50% which means that values above 50% indicates rightward preference and values below 50% indicates a leftward preference (Wiper, 2017). The LI was reported in four of the publications included in this SR.

- Other statistical methods can be used to evaluate laterality such as z-score which helps to determine if the laterality observed is beyond what is expected by chance (Hopkings,1999). The z-score was reported in a single publication, as reported in the results section.

3 Results and Discussion

3.1 General aspects

The search retrieved 20 publications able to be included in the SR (**Table 1**). In the word cloud (**Figure 2**), the most frequent term was lateralization (11.22%), followed by laterality and parrots (9.18%), foot and cognition (6.12%), and handedness, preference, asymmetry, and brain (5.10%). Most of these terms were present in the search strategy. Through the word cloud, we established the specific aspects of the publications that will be discussed, that are footedness, correlation of footedness with cognition, phylogeny, behavior, eye preference, and ontogeny.

Among the included studies, 40.0% (8/20) were conducted in Australia, 35.0% (7/20) in the US, and 5.0% (1/20) in Brazil. Most of the studies included multiple species of different genera. The Psittaciformes genera examined were predominantly (77.4%; 24/31) from Oceania (**Figure 3**). The studies on laterality in parrots were primarily concentrated in Australia, with a focus on taxonomic genera occurring in Oceania. This result is probably related to the great biodiversity of parrots on this continent (Forshaw, 2010). According to this

author, the Neotropics also have numerous parrot's species. Nevertheless, there were few studies (16.1%; 5/31) about this topic in Neotropical species.

Regarding the sample size, the average number of specimens studied was 22.7 birds, ranging from 1 to 458 (**Table 2**). In most of the studies, the methods for assessing laterality were by using repeated measures over time, repeating, on average, 13.5 times, ranging from 1 to 155 replications, with most of them (45.0%; 9/20) having 10 to 20 replications. Some studies did not use repetitions (Randler et al., 2011) and five of them did not inform it (McNeil et al., 1971; Rogers, 1980; Berg et al., 2020; Godinho et al., 2020; Kaplan; Rogers, 2021).

Footedness was evaluated in 80.0% (16/20) of the publications, while eye preference (eyeness) was assessed in 15.0% (3/20). The consistency of evidence for laterality was present in 78.0% of the publications and strong laterality in 48.0% of the outcomes. In the studies assessing footedness, the left lateral bias reported was 61.1%, on average, ranging from 5.0% to 100% of lateral bias to left. The predominant direction (PD) of laterality was 0.092 indicating a predominant direction to left.

In four publications the laterality index (LI) was calculated at individual level (Duggan et al, 2016; d'Antonio-Bertagnolli; Anderson, 2018; Godinho et al, 2020, Regaiolli et al., 2021). The first one Duggan et al. (2016), analyzed Budgerigars (*Melopsittacus undulatus*) and revealed a mean LI for unipedal support of -0.12 (ranging from -0.63 to 0.57) and for scratching side -0.02 (ranging from -0.38 to 0.47). In d'Antonio-Bertagnolli and Anderson (2018), for the same species the mean LI for unipedal support was 0.11 (ranging from -1 to 1) and for preening side was 0.08 (from -0.11 to 0.24). For the Blue-fronted Amazon parrots (*Amazona aestiva*), Godinho et al. (2020) reported a mean LI for footedness of 0.01 (from -1 to 1). In Regaiolli et al. (2021), three species of macaws were analyzed (*Ara chloropterus*, *Ara ararauna*, *Ara macao*) and the mean LI for footedness were -0.26 (from -0.58 to 0.90). In this publication, the z-score was also used to classify the macaws as left or right-footed, ranging

from -4.75 to 6.77. For right-footed macaws the z-score was > 1.96 , left-footed had z-score < -1.96 and in ambidextrous (non-lateralized) individuals it was $-1.96 < z\text{-score} < 1.96$.

Considering the context of research, 65.0% (13/20) of the publications were carried out with captive specimens, 20.0% (4/20) mixed wild and captive individuals, and only 5.0% (1/20) were exclusively with animals in the natural environment. The most studied topic was the relationship between laterality and cognition (25.0%; 5/20). This was followed by investigations into the correlation between laterality and phylogenetic aspects, brain size, lexical size, and retina anatomy.



Figure 2: Word cloud using the most cited and frequent words in the title and keywords of the 20 publications included in the SR. The larger words were used more frequently.

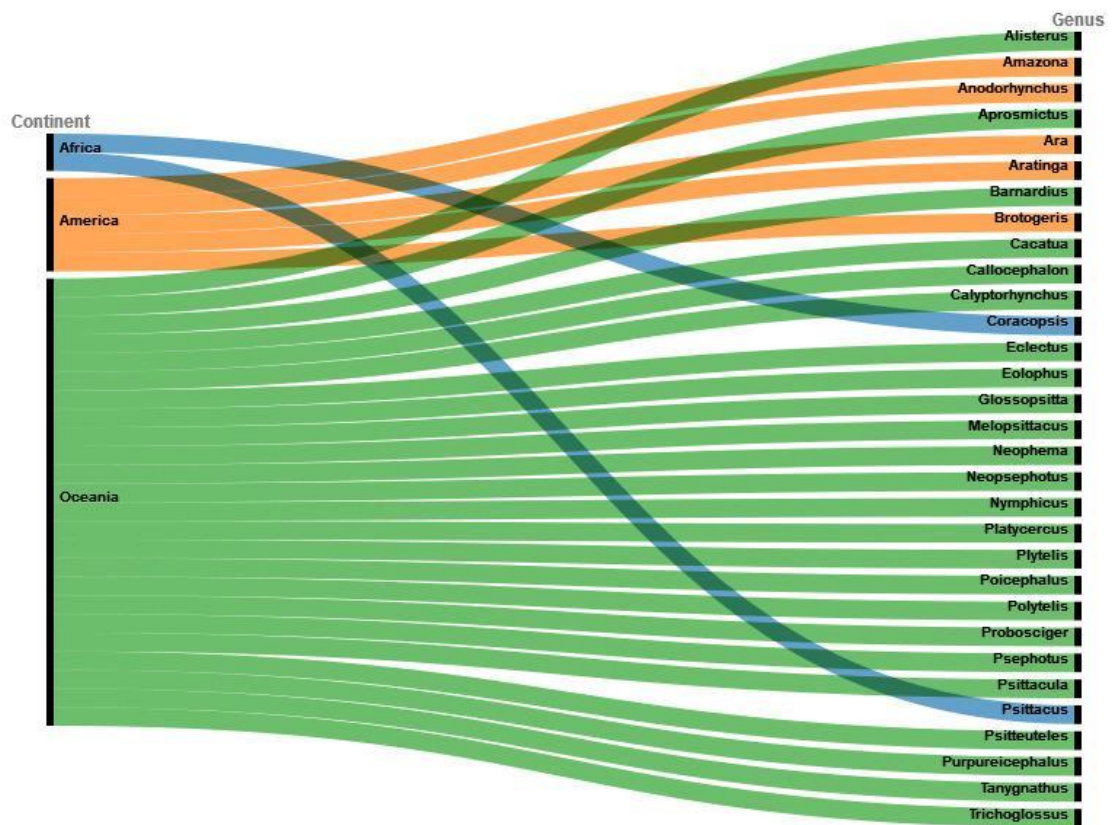


Figure 3: Relationship between the most studied genera in the publications included in the SR and their respective continents.

Table 2: The main methods used for identifying laterality in the publications included in the SR.

Methodology	Laterality type	Repetitions	Sample size	Articles that applied the methodology
Footedness	Footedness	3-155	1-522	Brown and Magat, 2011a; Randler et al. 2011; d'Antonio-Bertagnolli and Anderson, 2017; Berg et al. 2020; Kaplan and Rogers, 2021; Regaiolli et al., 2021; Snyder et al. 1996; Snyder et al.,1997; Snyder et al.,1998; Snyder and Bonner, 2001; Schiffner and Srinivasan, 2013; Cussen and Mench, 2014; Duggan et al., 2016; Rogers, 1980; Friedman and Davis, 1938; McNeil et al.,1970
Eyeness	Footedness and eyeness	10	10-20	Brown and Magat, 2011b
Observing laterality in natural behavior	Footedness	8-20	8-12	d'Antonio-Bertagnolli and Anderson, 2017; Schiffner and Srinivasan, 2013; Duggan et al., 2016
Dig discrimination	Footedness	18	11	d'Antonio-Bertagnolli and Anderson, 2017
Tool use problem	Footedness	18	11	d'Antonio-Bertagnolli and Anderson, 2017
Pebble-and-seed	Footedness and eyeness	10	5-41	Magat and Brown,2009; Godinho et al.,2020
Multi-access-box	Footedness and eyeness	Not informed	41	Godinho et al.,2020
String-pulling task	Footedness	10	7	Magat and Brown,2009; Regaiolli et al.,2021
The Hamilton Search task (HST)	Footedness	24	13	Cussen and Mench, 2014
Questionnaire	Footedness	5	91	Snyder and Harris,1997

Score based in the bodies position	Footedness	155	3	Snyder and Bonner,2001
Choice between two perches	Footedness and landing positions	20	12	Schiffner and Srinivasan,2013
Free landing on a single, long perch	Footedness and landing positions	20	12	Schiffner and Srinivasan,2013
Free landing on a single, long perch, two birds	Footedness and landing positions	13	12	Schiffner and Srinivasan,2013
Landing on a single, axially oriented single perch	Footedness and landing positions	20	12	Schiffner and Srinivasan,2013
Foot choice when climbing onto a perch	Footedness and landing positions	20	12	Schiffner and Srinivasan,2013

3.2 Behavioral tests used in the research of laterality in Psittaciformes

The included publications used different behavioral tests for evaluating laterality and for exploring their relationships with cognition or other behavioral traits. Here we briefly describe each behavioral test and more information about which publications applied these methodologies can be found in **Table 2**.

Footedness: In this test, a slice of fruit or a seed is placed in front of the parrot and the foot used to grasp the food is recorded by counting how many times the parrot uses each foot to manipulate food items (Friedman; Davis,1938).

Eye Preference: Eye preference is usually assessed by counting the number of times the parrot uses its dominant eye to fixate on a food item, based on the orientation of its head (Magat; Brown,2009).

Observation of laterality during the expression of natural behaviors: Using focal observations, the foot (left or right) used for the following behaviors is recorded: foot support (perching on one foot), scratching foot (using one foot to scratch the head), preening side (nibbling the feathers), and stretching side (Duggan et al., 2016).

Dig discrimination: Dig discrimination is a test used to investigate cognitive abilities of parrots (d'Antonio-Bertagnolli; Anderson, 2017). Laterality is also assessed by observing the freely occurring behaviors during the test, following the same methodology described above (applied in Duggan et al., 2016). Initially, the parrot is exposed to a five-day excavation process. The apparatus consists of a wooden support with two holes containing plastic cups positioned at the parrot's chest level. Training begins with only the reward, the millet seed, until 3 minutes pass, or the parrot consumes all the seeds. In the second training session, sand is added with a partially exposed piece of millet seeds. In the third and final trial, the millet seeds are completely covered with sand, requiring the parrot to dig to reach and consume seeds. After training, the actual test begins: two cups of different colors containing a mixture of sand and seeds are presented to the parrots, but only one contains the millet seed reward. The goal is for the parrot to choose and dig the correct cup to find the reward.

Tool use problem: This test consists of placing a piece of millet seed halfway through a tube, just beyond the reach of the parrot's beak. To solve this problem, parrots must demonstrate the insight to use a coffee-rod stirrer as a tool to remove the seed from the tube. This test

investigates the cognitive ability to solve a tool-use task. Laterality can also be recorded by observing lateral biases in freely occurring behaviors during the test (d'Antonio-Bertagnolli; Anderson, 2017).

Pebble-and-seed: The pebble-and-seed discrimination test is a method to analyze parrots' eye preferences and cognitive performance in a context similar to foraging. In this task, the parrot is required to peck at thirty-five seeds among fifty small pebbles of similar size and color placed in a tray within a controlled home cage, ensuring each parrot is tested individualized. To motivate foraging behavior, the birds are not fed prior to the task. Discrimination performance is calculated as the number of consumed seeds divided by the total number of pecks. Laterality was previously investigated by examining their foot and eye preferences while manipulating food (Magat; Brown, 2009).

Multi-access-box: Parrots need to use their feet or beak to solve the tasks. On face 1 of a box, there is a window that can be opened by pulling a horizontal "button". On face 2, a rope needs to be pulled to obtain the food. On face 3, the individual has to place a small ball into a hole in a tube, so that it would roll and hit the food, making it available. On face 4, it is necessary to insert a stick into a hole to push the food out. The multi-access box is a cognitive task that presents ecologically relevant challenges similar to those parrots might encounter in the wild. Laterality is investigated by observing foot preferences during food handling and counting behaviors involving left or right foot use during the experimental sessions (Godinho et al., 2020).

String-pulling task: Two strings are presented to the birds, with only one containing a reward, like a tied peanut. The birds must manipulate the correct string with their feet, pulling it to reach the food. At a higher level of difficulty, the strings can be switched. This method tests parrots' ability to obtain an item suspended from a string. Laterality is identified, and the pattern of foot preference while manipulating the string is related to individual success (Magat; Brown, 2009).

The Hamilton Search Task (HST): This task consists of three phases that assess spatial memory (phase 1) and cognitive flexibility (phases 2 and 3). In phase 1, the reward placement is pseudorandomized, and it is expected that the parrots acquire a pseudorandom search learning set during the five days of testing. In phase 2, the reward location is fixed and determined by the least-preferred location on the first phase. In phase 3, the rewarded location is the same as in the second phase, but the subjects are allowed to make only a single choice. Laterality is prior identified by recording foot preferences when manipulating food reward (Cussen; Mench, 2014).

Questionnaire: This method is appropriate for companion parrots. A questionnaire with 10 items and a standard method for testing foot preference in their pets is given to the guardian. They were asked to count the number of separate words in their pets lexicons of human speech sounds (Snyder; Harris, 1997).

Score based on body positions: Lateral bias in head or body posture, foot use, or grooming activity is evaluated. Each animal receives a score according to whether there is a left-sided bias (-1), midline body/head positioning (0), or a right-sided bias (+ 1) (Snyder; Bonner, 2001).

Choice between two perches: Two perches are offered to the birds, one to the left and the other to the right of their flight path. Their landing choices are recorded (Schiffner; Srinivasan, 2013).

Free landing on a single, long perch: The birds are free to land anywhere along the length of the perch, and their landing positions are recorded as left, right or central segment of the perch (Schiffner; Srinivasan, 2013). A variation of this test involves two birds, both are released simultaneously and can land anywhere along the perch. Both landing positions are recorded to determine whether the birds retain their take-off configuration or swap positions (Schiffner; Srinivasan, 2013).

Landing on a single, axially oriented perch: A single perch, placed at the far end of the tunnel and oriented perpendicular to the back wall, is offered to the birds. This experiment investigates

the side bias of the birds, determining whether they land on the perch by approaching from the left or right side (Schiffner; Srinivasan, 2013).

Foot choice when climbing onto a perch: An experimenter, standing directly in front of the bird, induces the bird to climb onto a smaller perch, with 60 cm long. To initiate the climb with a specific foot, the perch is gently pressed against the bird's chest. The foot (left or right) used to initiate the climb is recorded (Schiffner; Srinivasan, 2013).

3.3 Principal topics investigated

3.3.1 Footedness as an aspect of lateralization

Through footedness, it is possible to assess the lateralization of cerebral functions in birds that use their feet for food manipulation (Rogers, 1980). Since the beginning of this research field, footedness has been the principal aspect used to evaluate laterality in parrots. One of the first studies on limb preferences in Psittaciformes already suggested a predominance of left-handed animals within this taxonomic group (Friedmann; Davis, 1938). This preference remained consistent over time, with Rogers (1980) demonstrating the same result when reanalyzing data from the earlier study. However, it is important to highlight that, in the wild, it is not always possible to use the preferred limb due to environmental conditions, such as the location of food or perch, which can interfere with food manipulation. Therefore, studies of footedness with animals in aviaries are more common (Randler et al., 2011). This was also demonstrated in our SR, in which most of the studies were conducted with captive specimens.

To assess limb preference, behavioral observations during feeding have been a simple approach adopted in various studies. This methodology can be applied to correlate footedness with other aspects, e.g., the patterns and strength of foot preferences were accessed and determined by footedness and then correlated to ecological aspects, so it would be possible to estimate a phylogenetic relationship between the species (Brown; Magat, 2011). These

authors found out that across Australian parrots, the strength and direction of laterality are related to phylogeny, and the strength of laterality is also related to ecological factors. Observing the feeding activities in natural environments by ring-necked parakeets (*Psittacula krameri*), Randler et al. (2011) mentioned that there is a foot preference in both population and individual levels and an insignificant difference between left and right footed.

It is also important to highlight the reports of predominance of ambidexterity or non-lateralization (i.e., both feet were evenly used) in some taxa of parrots. For instance, when subspecies of *Platycercus elegans* (*P. e. elegans*, *P. e. faveolus*, and *P. e. adelaidae*) were compared, ambidexterity was predominant in those with variable and intermediate plumage coloration (Berg et al., 2020). A right-footed preference was observed at the population level in all the subspecies among non-ambidextrous individuals (Berg et al., 2020). In one specific subspecies, *P. e. adelaidae*, footedness was related to latitude and longitude, with the proportion of right-footed individuals being higher in cooler, wetter, and more densely vegetated southerly populations (Berg et al., 2020).

Lateralization may vary depending on the task at both individual and population levels. In Budgerigars (*Melopsittacus undulatus*) researchers identified which foot the parakeet used to make the first contact with the perch, revealing a variation in their choice of perch, landing location, and direction of approach while landing (Schiffner; Srinivasan, 2013). The lateralization at individual level in parakeets was also observed in behaviors such as unipedal foot support and side preference for scratching (Duggan et al. 2016; d'Antonio-Bertagnolli, 2018). These behavioral asymmetries become evident in parrots at approximately four months old, when foot preferences can be rated in activities like preening and foot handling (Snyder; Bonner, 2001).

A significant association between right footedness and larger lexicon in both Amazon (*Amazona sp.*) and African Grey parrots (*Psittacus*) was mentioned by Snyder and

Harris (1997). According to them, the right-side bias represents a behavior index of contralateral hemispheric specialization, i.e., the study proposed that vocal recall is partially related to categorizing properties of the left hemisphere. Another relation between footedness and neural functions was mentioned by Kaplan and Rogers (2021), who proposed that individuals with larger brain mass would have stronger foot preferences and left footedness is correlated positively with the size of the nidopallium relative to the whole brain.

A great part of the studies added in the SR used footedness to assess laterality. However, most of the research was conducted with Australian parrots, and then, only a part of the Psittaciformes fauna was represented, making it difficult to extrapolate these limb preferences to the entire clade.

3.3.2 Eye preference as an aspect of lateralization

There were few studies assessing lateral bias based on eye preference (Brown; Magat, 2011 a; Brown; Magat, 2011 b; Coimbra et al., 2014). Generally, parrots use their preferred monocular field of vision to analyze food items and then employ the corresponding foot to manipulate them, indicating a strong relationship between ocular preference and foot preference (Brown; Magat, 2011 a). Individuals with pronounced ocular asymmetries achieved greater success in cognitive tests (Magat; Brown, 2009), although some species challenged this pattern (Brown; Magat, 2011 a).

It has also been shown that topographic architecture of the retina may reflect microhabitat preferences and varying degrees of lateralized visual behaviors that aid in locating and manipulating food items in cockatoos (Coimbra et al., 2014). In lateralized species of cockatoos, a higher density of ganglion cells in the perifoveal region of the left retina is linked to coordinated actions between the left eye and left foot during feeding. In contrast, non-lateralized species have similar densities of ganglion cells in both the left and right retinas,

associated with the absence of preference for a specific eye in locating food items. The presence of equivalent densities of ganglion cells in the dorsotemporal area of both retinas suggests a role in binocular orientation during practical activities (Coimbra et al., 2014).

The SR reveals a scarcity of publications focusing on monocular laterality, highlighting it as an important yet poorly understood topic. The scarce available literature does not indicate a clear tendency for a preference for the right or left eye, unlike the better-documented preference towards left observed with footedness, leaving this question open.

3.3.3 Relationships between laterality and cognition

In our search, five studies correlated laterality with cognition (Magat; Brown, 2009; Cussen; Mench, 2014; d'Antonio-Bertagnolli; Anderson, 2018; Godinho et al., 2020; Regaiolli et al., 2021). In general, these studies hypothesize that these asymmetries promote an increase in the cognitive capacity of animals, leading to better performance in cognitive tests, and that the intensity of laterality has a greater influence on cognitive performance than the direction itself (Magat; Brown, 2009). The methodologies used in these studies focused on foraging abilities (Table 2), such as the pebble and seed test (Magat; Brown, 2009; Godinho et al., 2020) and string-pulling test (Magat; Brown, 2009; Regaiolli et al., 2021).

In a more complex experiment, multiple-access box was provided to the parrots and they needed to use their feet or beak to solve the tasks in each face of the box, that had four possible solutions (Godinho et al., 2020). Eleven parrots solved at least one face of the boxes, and no differences were found between right- and left-handed individuals or between males and females. In Australian Budgerigars, cognitive abilities were assessed through the 'tool use problem' and with a 'dig task' (d'Antonio-Bertagnolli; Anderson, 2018). The results showed an association between rightward preening side and greater success in the dig task. Lastly, 'The

Hamilton Search Task' (Cussen; Mench, 2014), previously used only in humans or primates (described in Ha et al., 2011), was adapted for Psittaciformes.

Vocal capacity and vocal repertoire can be used to predict cognitive capacity in parrots. It has been reported an association between the lexicon repertoire and foot preference, what suggests a possible higher cognitive function in lateralized parrots (Snyder; Harris, 1997). The vocabulary size depends primarily on the parrot's capacity to learn, rehearse and memorize novel vocal calls, while the motor asymmetries are related to hemispheric asymmetries, and the researchers proposed the involvement of these features in lexical size (Snyder; Harris, 1997).

In summary, the relationship between laterality and cognitive capacities confirms once more the advantages of having an asymmetric brain. The publications evaluated evince that lateralized individuals would have greater results in solving multiples cognitive problems. Future research should focus on other cognitive aspects not investigated yet, such as the cognitive and judgement bias.

3.3.4 Phylogeny of laterality

This topic was assessed in only one study (Brown; Magat, 2011 b), that investigated the pattern and strength of lateralization in Australian parrots and to what extent laterality is constrained by phylogenetic relationships. The Australian Psittaciformes phylogeny suggests that the distribution of foot preferences indicates a divergence in the evolution of laterality within the group. Phylogenetic analyses have shown the presence of a lateralized ancestor, with a larger body size and adapted to feed on large seeds or pods, which required manipulation skills. The reduction of laterality in Australian Psittaciformes species is accompanied by a decrease in body size and a shift in feeding habits, consuming smaller seeds and flowers (Brown; Magat, 2011 b).

In the Cacatuidae family, which retained the ancestral pattern with a large body size, most taxa became left-footed. On the other hand, for Psittaculine, the expression of laterality evolved from a left-footed ancestral to a right-foot preference in most individuals. As for the Loriinae, birds with small bodies, they did not display laterality and feed on small seeds and flowers (Brown; Magat, 2011 b). Therefore, natural selection maintains, modifies, or even eliminates limb preference and laterality based on the dietary and ecological characteristics of the species.

Few studies investigated the phylogenetic bases of laterality, despite it being postulated in the literature as an ancestral condition, widespread since the early chordates, suggesting that laterality is a common feature of the brain with different structures and evolutionary histories (Valortigara; Rogers, 2005; Anfora, 2010). In other animals such as fish from the Belontiidae family, phylogenetic analyses have revealed low variance in eye preference, suggesting a significantly reduced ancestral condition of cerebral lateralization in this clade (Clotfelter; Kuperberg, 2006). Phylogenetic analysis involving laterality for the remaining Psittaciformes is not known. The lack of published information in the scientific literature leaves us without an understanding of the ancestral relationships associated with laterality and its role in the overall evolution of the group.

3.3.5 Laterality and the expression of natural behaviors

The three studies exploring the relationship between laterality and natural behaviors were conducted with the same species, the Australian Budgerigar (*Melopsitacus undulatus*) (Schiffner; Srinivasan, 2013; Duggan et al., 2016; d'Antonio-Bertagnolli; Anderson, 2018). Schiffner and Srinivasan (2013) suggested that behavioral laterality is complex and multifaceted, extending beyond a preference for one side of the body. These findings indicate that Budgerigars' lateral preferences in different tasks are independent of each other, reflecting

a highly task-specific lateralization. On the other hand, d'Antonio Bertagnoli and Anderson (2018) suggested that laterality is a stable and widespread characteristic among individuals of this species. They propose that, at least for the four specific behaviors evaluated (unipedal support, scratching foot, preening side, and stretching side), the sampled animals exhibited consistent lateral preferences. Additionally, Duggan et al. (2016) addressed social behaviors to relate lateral preferences to pair bonds and aggressiveness, without a significant correlation between these behaviors. This was attributed to the fact that the animals involved in the study had not yet established a clear hierarchy. Lastly, laterality can result in individual differences in activity levels and impact on overall performance during active tasks. In other words, lateral behavioral preferences may be intrinsically linked to physiological and temperament differences (d'Antonio-Bertagnoli; Anderson, 2018).

The relations between laterality and behavioral aspects were investigated for a few species and using a limited amount of specific behavioral categories, not focusing on stable interindividual differences in these behaviors (i.e., animal personality or temperament). Evidence from other taxonomic groups, e.g., domestic cats (McDowell et al., 2016), domestic dogs (Schneider et al., 2013), and donkeys (Díaz et al., 2021), suggest a possible relationship between laterality and temperament. Another important aspect to be investigated is the relationship between emotionality and laterality, as differences in emotional processing in the two hemispheres are reported. It was proposed that the regulation of approach-avoidance conflicts (i.e., the Behavioral Inhibition System or BIS) is processed by the right hemisphere, while sensitivity to reward (i.e., the Behavioral Activation System or BAS) is processed by the left hemisphere (Gable et al., 2018; Goursot et al., 2021). The study of emotional lateralization holds promise for understanding how affective states, personality and lateral biases are integrated.

3.3.6 Ontogenesis of laterality

Although the relevance of ontogenetic development of laterality, since it could help unravel its evolutionary foundations, only a single study on the subject has been included in this SR (Snyder; Bonner, 2001). This study was a pilot observational trial, involving three Congo African grey infants (*Psittacus erithacus*) and investigated the postnatal development of motor asymmetries. After 24 days of birth, the chicks began to exhibit leftward biases and after four months, this pattern persisted. For Psittaciformes, expressing motor biases from the early days of life would be disadvantageous before they achieve thermoregulatory self-sufficiency and autonomous motor control. During this period, they are exclusively dependent on the mother, whose role is to carry the chicks to the nest's center (Snyder and Bonner, 2001).

The ontogeny of laterality in birds is dependent on genetic and environmental factors. The first one affects only the position of the embryonic body, while the resulting difference between left and right visual stimulation shapes the visual pathways in a lateralized manner (Gunturkun; Ocklenburg, 2017). The significance of non-genetic factors is evident in visual asymmetries in birds, where genes only influence the position of the embryonic body, while the resulting difference between left and right visual stimulation shapes the visual pathways in a lateralized way. During the incubation period, the embryo assumes an asymmetric position inside the egg. As a result, the right eye receives light stimulation because it is located adjacent to the shell, while the left eye is deprived of light. Therefore, the right eye of the embryo is repeatedly stimulated by light (Gunturkun; Ocklenburg, 2017). The curved and lateralized position of the bird embryo is most likely mediated by the Nodal cascade, as the processes underlying the asymmetric positioning of the viscera are always accompanied by the embryo's torsion and a turn of the head to the right (Ramsdell; Yost, 1998). Thus, it is proposed that normal rearing conditions correspond to right-eye stimulation, resulting in left-hemisphere superiority for visual object discrimination. This population bias is not genetically determined

by factors within the visual system but by the lateralized environmental factor of light input, which results from the genetically determined body position.

The existing study on the ontogeny of laterality in parrots (Snyder; Bonner, 2001) estimates a time of 24 days for the onset of asymmetry expression, but this time was based on only a few individuals of a single species of African avian fauna (*Psittacus erithacus*), which prevents us from extrapolating these conclusions to neotropical and Australian species. Aspects of embryonic asymmetry were not emphasized, in other words, the ontogenetic process of laterality described in birds, in general, may not work the same way in Psittaciformes, and their peculiarities are not understood yet.

4 Conclusion

Throughout this SR, the existing scientific literature on laterality in Psittaciformes evidenced a preference for the left foot, but a lack of sufficient evidence for a stable preference for a monocular side of vision. The SR also highlighted the main methodologies used for identifying the motor asymmetries in parrots. The most investigated topic in the literature available is the relationship between laterality and cognition. Ontogenetic and phylogenetic aspects were poorly exploited in the publications, and it represents an opportunity for further research including a wide range of species. Few studies were conducted with Latin American taxa, while species from other continents have data dating back 30 years. There is also a gap in data related to context-specific expression of lateral biases. It was also scarce studies evaluating the associations between laterality with other aspects of parrots' mental life, such as emotionality and stable inter-individual differences (i.e., temperament). Studies on other taxonomic groups proposed that laterality may be associated with the temperament of each individual, which should be a promising research question for parrots. Future research should also involve both basic and applied aspects, enabling a better understanding of laterality and its relationships with other evolutive and ecological phenomena in the lives of Psittaciformes.

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4. CHAPTER 2

Laterality in Neotropical Psittacids (Aves, Psittacidae): Evaluation of Its Relationship with Temperament and Social Interactions

Larissa Gomes ^a, Gustavo Nunes ^a, Maria Eduarda Caçador Branco ^a, Aline Cristina Sant'Anna ^b

^a Postgraduation Program in Biodiversity and Nature Conservancy, Federal University of Juiz de Fora, 36.036-330, Juiz de Fora, MG, Brazil.

^b Department of Animal Science at School of Agricultural and Veterinarian Sciences of the São Paulo State University (Unesp), 14.884-900, Jaboticabal, SP, Brazil

Resumo

A lateralidade é caracterizada como assimetrias em estruturas bilaterais. Em psitacídeos, pode ser expressa pela preferência de uso da visão monocular ou uso de um pé específico para alimentação. O objetivo desse trabalho foi identificar a lateralidade em três espécies de psitacídeos e relacionar o fenômeno a aspectos do temperamento e sociabilidade. Analisamos 50 indivíduos das seguintes espécies: *Psittacara leucophthalmus* ($n=15$), *Primolius maraca* ($n=12$), *Pionius maximiliani* ($n=23$), em parceria com o CETAS Juiz de Fora. Para avaliar a lateralidade utilizamos como indicadores os comportamentos de preferência por um membro específico para manipulação de alimentos, pé de descanso e preferência visual através do campo de visão monocular utilizado durante os testes de seixo e sementes, novo objeto e reação à pessoa. O teste de seixo e sementes avalia a capacidade cognitiva e de forrageamento das aves, ao estimulá-las a identificar e consumir as sementes em meio aos seixos de tamanho e cores similares. Por fim o temperamento das aves foi avaliado segundo suas respostas individuais nos testes do novo objeto e reação à pessoa. Após a coleta dos dados, quantificamos as frequências de uso de cada um dos membros e calculamos o índice de lateralidade (LI) através da equação

$LI = (L-R) / (L+R)$, no qual L representa o uso do lado esquerdo e R do direito. Coeficientes de correlação de Spearman foram calculados para avaliar as relações entre a lateralidade e os aspectos comportamentais investigados (temperamento e comportamento social). Os resultados apontaram a presença da lateralidade nas três espécies analisadas, ressaltando a preferência pelo olho direito. Para a preferência de membro e apoio unipodal, não houve uma direção predominante. Correlações significativas entre as preferências visuais e as dimensões do temperamento foram observadas (atividade, ousadia, ansiedade e proximidade com o ser humano). Quanto maior a atividade, menor o LI total e LI no teste de seixos e sementes, ou seja, maior a preferência pelo olho direito. As aves mais ousadas e mais ansiosas tiveram menor LI no teste de reação à pessoa, o que também resulta em preferências pelo olho direito. Maior evitação ao humano, maior o uso do olho direito (menor LI no teste do novo objeto). Não foram encontradas correlações significativas entre a lateralidade e as interações sociais positivas e negativas. Nosso trabalho trouxe contribuições para a literatura científica identificando a expressão comportamental de lateralidade em espécies neotropicais, as quais possuem poucos dados a respeito do tema. Além disso, esse foi o primeiro trabalho evidenciando a relação entre o temperamento e a lateralidade em psitacídeos.

Palavras-chave: assimetrias comportamentais; cognição; temperamento; Psittacidae.

Abstract

Laterality is defined as asymmetries in bilateral structures. In parrots it can be expressed by the preference for using the monocular visual field or a specific foot for feeding, also known as footedness. The aim of this study was to identify laterality in three Psittacine species and examine its relationship with aspects of their temperament and sociability. We analyzed 50 individuals of the following species: *Psittacara leucophthalmus* ($n=15$), *Primolius maraca*

($n=12$), *Pionius maximiliani* ($n=23$), in partnership with CETAS- Wild Animal Rehabilitation Center of Juiz de Fora. To evaluate laterality, we used the following indicators: footedness, unipedal support while resting, and eye preferences through the monocular visual field during the tests: pebble and seed, novel object test, and reaction to person. The pebble and seed test evaluate cognitive capacities and foraging skills of birds by encouraging them to identify and consume seeds among pebbles of similar size and color. The temperament was evaluated according to the birds' individual responses in the novel object and reaction to person tests. After data collection, we quantified the frequency of use of each foot and eye and calculated the laterality index (LI) using the equation $LI = (L-R)/(L+R)$, where L represent the left side use and R represents the right. Spearman correlation coefficients were calculated to assess the relationships between laterality and the behavioral aspects investigated (temperament and social behavior). The results indicated the presence of laterality in the three bird species, highlighting the right eye preference and no predominant direction was found for footedness and unipedal support. Significant correlations between eye preference and the four temperament dimensions (activity, boldness, anxiety and proximity to humans) were found. The greater the activity, bigger the total ocular LI and the LI in the pebble and seed test towards right. Bolder and more anxious birds had lower LI in the human reaction test, thus right eye preferences. Avoidance to humans was related to lower LI in the novel object test, therefore right eye preferences. No significant correlations were found between laterality and positive and negative interactions. Our study contributed to the scientific literature by identifying laterality in neotropical species, which have limited data on the subject. Additionally, this was the first work to demonstrate the relationship between temperament and laterality in psittacids.

Keywords: behavioral asymmetries; cognition; temperament; Psittacidae.

1 Introduction

Differential processing of neural inputs and control of outputs by the right and left sides of the brain is known as functional asymmetry or lateralization (Rogers, 2008). Lateralization of the cerebral hemispheres is a characteristic present in the chordates and can result in asymmetries in the use of structures and bilateral behaviors (Vallortigara; Rogers, 2005). A lateralized brain can result in an increase in neural capacity, allowing for complementary and parallel processing of information in the hemispheres (Kaplan; Rogers, 2021). Laterality can be quantified by assessing foot or eye preference (Magat; Brown, 2009), because the stimulus is often processed in one of the cerebral hemispheres, generally opposite to the chosen side (Rogers, 2008).

Psittaciformes are known for their high cognitive capacity (Magat; Brown, 2009), and there is evidence that laterality can specialize each hemisphere in these animals. The left hemisphere predominates in behaviors such as foraging, discernment and manipulation of food items, attention, and recognition of vocalizations. Conversely, the right hemisphere predominates in the control of sexual behaviors, spatial recognition, and reactions to predators (Vallortigara; Rogers, 2005).

In Psittaciformes, laterality can be exhibited through the preference for a specific foot used to manipulate food items when eating. This preference has already been documented in several species (Berg et al., 2020). Foot preference can be assessed at both individual and population levels (Duggan et al., 2016), but environmental, behavioral, and ecological variables can also influence its expression (Berg et al., 2020). Evidence suggest that this group had a strongly lateralized ancestor with a larger body size that fed on larger seeds (Brown; Magat, 2011b). The loss of laterality was associated with a decrease in body size, followed by a shift in feeding habits to smaller seeds that did not require manipulation (Brown; Magat, 2011b).

Laterality can also be exhibited through the preference for a specific side of the monocular visual field (Brown; Magat, 2011a). This behavior is widely expressed in psittacids, as they analyze food with their preferred visual field and manipulate it with the corresponding foot. An asymmetry in their visual system can increase the speed at which they recognize objects (Rogers, 2017). More lateralized birds tend to better distinguish their food, providing adaptive advantages at both individual and population levels. For example, in the *Cacatuidae* family, significant morphological specificities have been found in the retinal ganglion cells of the left eye of left-handed cockatoos, characterized by an increased density (Coimbra et al., 2014).

In the last 10 years, studies on laterality in psittacids have focused on the correlation between brain asymmetries and cognitive aspects (Cussen; Mench, 2014; D'antonio-Bertagnolli; Anderson, 2017; Godinho et al., 2020; Regaiolli et al., 2021). Behavioral indicators of hemispherical specialization, such as foot and eye preference, contribute to an increase in these birds' cognitive capacity (Magat; Brown, 2009), and provide advantages for foraging, helping them distinguish food in the environment and solve problems (Godinho et al., 2020). Laterality intensity could influence more than just direction; it may enhance the ability to coordinate tasks involving the foot and beak to extract seeds from pods and manipulate objects (Magat; Brown, 2009; Regaiolli et al., 2021). Thus, a better understanding of visual asymmetry and its relationship with foot preference can indicate an aptitude for the release of captive psittacids, as laterality can improve their foraging ability and increase their chances of survival after release.

Temperament refers to behavioral differences that are consistent over time and context (Réale et al., 2007). Previous studies suggest that laterality can be related to temperament. For example, cats with a limb preference (motor laterality) exhibited stronger temperament traits such as confidence and affection (McDowell et al., 2016). Individuals with right-side

preferences tend to be bolder, more sociable, and display more exploration in novel environments (Braccini; Caine, 2009; Rogers, 2010; Goursot et al., 2019). Lateral preferences indicate asymmetric use of one hemisphere, with each hemisphere specializing in different functions (Vallortigara; Rogers, 2005), such as the processing of emotions. The left hemisphere is more associated with positive (or approach) emotions, while the right hemisphere is associated with negative (or withdrawal) emotions (Davidson, 1992; Leliveld et al., 2013). Therefore, an individual's preference for one side of the body (consequently, dominance of one hemisphere) can lead to different responses to environmental stimuli and novel situations, expressed through consistent temperament profiles (Rogers, 2009; Goursot et al., 2019).

Studies on laterality in psittacids are still scarce and focus on only a few species, leaving gaps to be filled in this research field. No studies have assessed laterality in the White-eyed Parakeet (*Psittacara leucophthalmus*), the Scaly-headed Parrot (*Pionus maximiliani*), or the Blue-winged Macaw (*Primolius maracana*). Additionally, the relationship between laterality and temperament in these birds has not been investigated yet.

The aim of this study was to evaluate whether three neotropical psittacid species (*Psittacara leucophthalmus*, *Pionus maximiliani*, and *Primolius maracana*) exhibit laterality in behavior such as footedness, eye preferences and unipedal support, and then to correlate the bird's laterality to their temperament and social interactions. We expect that the three species of psittacids will exhibit laterality for the behaviors at either the individual or population level. Based on previous studies, we anticipate a significant correlation between laterality and temperament, with individuals showing right-side preferences being bolder. Finally, we also expect a significant relationship between laterality and social interactions, with positive interactions related to right-side preferences and negative interactions to the left side.

2 Methodology

2.1 Study area and animals

This study was approved by the Ethics Committee on Animal Use of the Universidade Federal de Juiz de Fora (Protocol no. 008/2023), the Instituto Brasileiro do Meio Ambiente e dos Recursos Renováveis - IBAMA (no. 02015.000580/2023-04), and the Instituto Estadual de Florestas - IEF (no. 2100.01.0004298/2023-69). The study was conducted in a Wild Animal Release Area (ASAS) in Santana do Deserto, Minas Gerais, Brazil, in partnership with the Wild Animal Rehabilitation Center (CETAS) of Juiz de Fora, Minas Gerais, Brazil. A total of 50 adult birds were studied: 15 (9 males and 6 females) White-eyed Parakeets (*Psittacara leucophthalmus*), 12 (7 males and 5 females) Blue-winged Macaws (*Primolius maracana*), and 23 (12 males and 6 females) Scaly-headed Parrots (*Pionus maximiliani*). The birds were kept in captivity under the supervision of IBAMA and IEF for approximately 8 months. Data collection was conducted in two aviaries: aviary 1, where the tests were conducted (8.5 m length x 7.0 m width x 3.0 m height) and the aviary 2, where the birds were housed (12.9 m length x 7.0 m width x 3.0 m height).

2.2 Data collection

2.2.1 Footedness

To investigate whether there is laterality in foot preference, we observed the animals in the aviary 2 during feeding time, and we recorded which foot was used to manipulate the food (**Figure 1**), following Magat and Brown (2009). The birds were first identified by their leg band number (CETAS - IBAMA/IEF) and then marked with non-toxic ink (Walmur Instrumentos Veterinários Ltda[®]). The aviary was enriched before the beginning of the observations with branches and leaves from trees such as *Bauhinia forficata*, *Tibouchina granulosa*, *Congea sp*, *Erythrina mulungu*, *Handroanthus albus*, and fruits from *Syagrus romanzoffiana*, *Inga laurina*,

blackberries, in addition to their regular food items (sunflower seeds and commercial fruits - banana, apple, grape, mango, guava, watermelon, squash, beetroot, cabbage, etc.). The observations were conducted continuously twice a day, from 10 AM to 12 PM and from 2 PM to 4 PM over 5 consecutive days. Two trained observers were responsible for recording which foot the birds used to manipulate fruits, seeds, and leaves. In this part of the study, 14 White-eyed Parakeets, 17 Scaly-headed Parrots, and 4 Blue-winged Macaw were assessed.



Figure 1: Methods used to assess laterality: **A** Footedness behavior in the Blue-winged Macaw; **B** Tray contained the 50 pebble and 35 seeds for the pebble and seed test; **C** A Scaly-headed Parrot during the pebble and seed test; **D** Observation of unipedal support in the Scaly-headed Parrot. Source: a) Paula Neto (2024); b) Larissa Gomes (2023); c) Maria Eduarda Branco (2023); d) Gustavo Nunes (2023).

2.2.2 Pebble and seed test

To identify the relationship between eye preference, foraging ability, and cognition, we used the pebble and seed test (Magat; Brown, 2009; Godinho et al., 2020). This test assesses the animal's capacity to distinguish seeds from pebbles, a concern that can occur during foraging. During this test, we also recorded eye preference whenever the bird looked at the pebbles and seeds. The test was conducted individually in a cage (1.17 m length x 0.55 m width x 0.50 m height) placed inside the aviary 1 and lasted for at least 5 minutes. Before the test began, the animals were left alone for 5 minutes to habituate. In the test, a tray (40 cm length x 30 cm width) containing 35 sunflower seeds, and 50 similar pebbles was presented to the birds inside the cage, and the time was initiated from there (**Figure 1**). All tests were filmed with the camera placed on a tripod in front of the cage, and the observer was hidden by a camouflage cape, so they were not in the bird's visual field. If the bird initiated contact with the tray from the fourth minute onward, we added 3 minutes to the test. If there was no contact at all during the first 5 minutes, we added 5 minutes, totaling 10 minutes. We assessed eye preference by counting the number of times the animal used each monocular visual field side (right or left), the number of pecks at the pebbles and seeds, and the total amount of seeds eaten. All pecks at the pebbles or seeds were counted, as well as the total number of seeds consumed (eaten by the birds). Test success rate was evaluated by dividing the number of seeds consumed by the total number of pebbles. Recordings were done continuously throughout the whole test using focal observations from the videos. Each test was repeated 3 times for each animal with one week apart. To stimulate the animal's interactions with the test, we initiated it at 06:30 AM, before feeding time.

2.2.3 Unipedal support

To investigate whether there is laterality in foot preference for unipedal support (when the bird stands or perches on one leg while the other is tucked up close to the body when resting), two observers, with an interobserver agreement above 80%, continuously recorded every instance of unipedal support behavior during observations in the aviary 2 conducted from 10 AM to 12 PM and from 2 PM to 4 PM over 5 consecutive days. This category was analyzed by the percentage of observation time (**Figure 1**).

2.2.4 Social interactions

We continuously recorded social interactions during behavioral observations conducted twice a day (from 10 AM to 12 PM and from 2 PM to 4 PM) over 5 consecutive days in the aviary 2. Data was expressed as frequency (occurrences/minute). We documented negative social interactions such as pecking (when one bird pecks another), kicking (when a bird uses its foot to kick another), and attempts to peck and kick (without physical contact). The birds involved in these negative interactions were categorized as winners (those displacing another bird) and losers (the displaced bird) (Ramos et al., 2021). Positive social interactions included attempts to copulate and co-feeding (when one bird feeds another). Subsequently, these data were correlated with the individuals' laterality index obtained from the eye preference, footedness, and unipedal support.

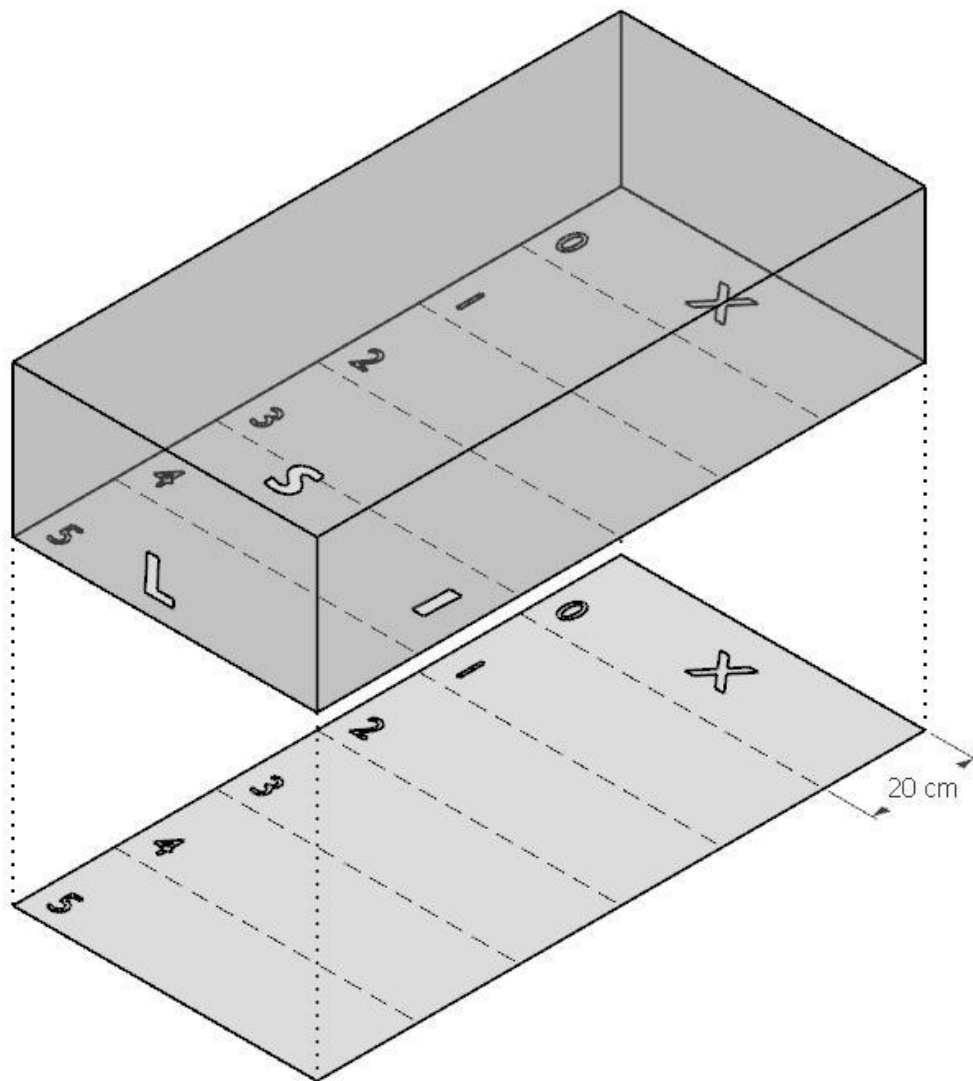
2.2.5 Temperament

To assess temperament, we used the novel object (NO) and reaction to person tests (RP), (**Figure 2**) adapted from Ramos et al. (2021). The animals were tested individually in the same cage from the pebble and seed test, divided by a red line into five 20 cm quadrants. The zero quadrant represents the place where the novel object or person stands. The first quadrant was

the closest to the stimulus (a novel object or person), and the fifth was the farthest. The birds could also be positioned on the superior, lateral, or inferior bars of the cage (**Figure 3**). Before the beginning of the tests each bird was left alone in the cage for 5 minutes to habituate. The first test was the novel object, and it was recorded each time that the bird interacts with the object. The reaction to a person test was conducted after a 3-minute interval following the end of the novel object test. The person stood in front of the grid and placed the hand inside the cage and it was recorded each time that the bird attempts to pick the hand. Each test lasted 5 minutes, and behaviors were recorded using focal sampling and instantaneous recordings at 10-second intervals. The category behaviors were rest, movement, attention, inactivity, locomotion, excitement, preening, interaction to environment, interaction to the novel object, interaction to person, latency to touch the novel object (in seconds), latency to try touch the person (in seconds), touch into the novel object (number of occurrences), attempts to touch the person (number of occurrences), vocalizations (number of occurrences) (see Supplementary Material S1 for full descriptions). We registered laterality through eye preference by counting which monocular visual field side was most used to look at the novel object or the person. Each test was repeated 3 times, with 47-day intervals between the first and second repetition and 22-day between the second and third repetition.



Figure 2: Scaly-headed Parrots during temperament tests: **A** first test with a hat as novel object; **B** second test with a ball as novel object; **C** third test with a basket as novel object; **D** reaction to person test. Source: Maria Eduarda Branco (2023).



Source: Larissa Gomes (2024).

Figure 3: Model cage to exemplify the locations that the birds could occupy during the pebble and seed and temperament test. Numbers 0-5 represent the quadrants, each divided into 20 cm sections. The X marks the location of the novel object. In the reaction to person test, the person stood in front of the X and placed their hand inside the cage. Abbreviations: S: superior location; L: lateral location; I: inferior location.

2.3 Data analysis

Statistical analyses were done using nonparametric statistics (Hopkings, 1999; McGrew; Marchant, 1997). The softwares Jamovi (JAMOVI VERSION 2.5, 2024) and SAS (SAS/STAT® 15.3) were used. Laterality was measured by observing asymmetric behaviors (eye preference and footedness), and a minimum of 10 lateralized behaviors records were required for inclusion in the analysis. Animals that did not meet this criterion in a respective test were excluded from its data analysis. This allowed us to determine how many times an animal looked with the right or left eye and how many times it used each foot (left or right) to manipulate food items and for unipedal support. Subsequently, a chi-square goodness-of-fit test was performed to test whether the proportion of right and left sides differed (deviated from 50%, $p \leq 0.05$). For p -values ≤ 0.05 in chi-square test, the individual was considered lateralized (1) and $p > 0.05$ as non-lateralized (0). This categorization was used for further analyses.

To determine the laterality index (LI), we used the calculation: $LI = (L - R) / (L + R)$, where L stands for the left and R stands for the right (Hopkings, 1999; Wiper, 2017). Positive LI results indicate a preference for the left, while negative results indicate a preference for the right, with values ranging from -1 to 1. Laterality expressed as LI from eye preference in the pebbles and seed, novel object, and reaction to person tests were compared using Kruskal-Wallis to evaluate if there were differences in laterality among the tests. From this analysis, we concluded that it would be possible to calculate a general laterality index for eye preferences, as there were no significant differences between tests. Therefore, we established a general eye preference for each animal, calculating the LI for the data of all tree tests summed.

In the pebbles and seed test, the animals' rate of success was calculated by dividing the number of seeds consumed (eaten by the birds) by the number of pebbles (Magat; Brown, 2009; Godinho et al., 2009). Then, a Mann-Whitney U analysis was used to evaluate whether laterality contributed for the animals' success in the test. The dependent variable was the rate of success,

and the independent variable was the laterality, categorized as: 0 = non-lateralized and 1 = lateralized (based on the results of the chi-square test, regardless of side).

To assess temperament, a factor analysis using multivariate methods of data dimensionality reduction was conducted to identify the main temperament dimensions, following Ramos et al. (2021). Variables with loadings ≥ 0.40 were considered main contribution to the factors. Each animal's individual factor score was then correlated with the laterality index, success, and seeds consumed in the pebbles and seed test. LI was also correlated with the frequency of positive and negative social interactions, as well as with success and seeds consumed in the pebbles and seed test, using Spearman's correlation coefficient.

3 Results

3.1 Laterality

We registered eye asymmetry in the pebbles and seed test, the novel object test, and the reaction to person test (**Table 1**). In the pebbles and seed test, 46.66% (14/30) of the birds preferred the right eye, 3.33% (1/30) preferred the left eye, and 50.0% (15/30) were non-lateralized. The average Laterality Index (LI) was -0.26, also revealing a preference for the right eye. In the novel object test, 31.57% of the birds preferred the right eye, while 68.42% (26/38) were non-lateralized. The average LI was -0.26. In the third test, reaction to person, 22.22% (10/45) preferred the right eye, 6.66% (3/45) preferred the left eye, and 71.11% were non-lateralized. In this test, the average LI was -0.16. Thus, it was clear that right laterality in terms of ocular preference was evident.

From these data, the Kruskal-Wallis's test was applied to assess whether there was a difference in the direction of laterality depending on the type of test. No significant difference was found ($X^2 = 4.72$; $df = 2$; $p = 0.09$) in laterality between the tests; therefore, it was possible to consider one general variable, total ocular LI, for eye preference (**Table 2**). Of the 50 animals

analyzed (15 White-eyed Parakeet, 12 Blue-winged Macaw, 23 Scaly-headed Parrot), 54% (27/50) preferred the right eye, 2% (1/50) preferred the left eye, and 44% (22/50) were non-lateralized. The average LI was -0.28. Separately, 53.33% (8/15) of White-eyed parakeet preferred the right eye, 6.66% (1/15) preferred the left eye, and 40% (6/15) were non-lateralized, with an average LI of -0.42. Among Blue-winged macaw, 66.66% (8/12) preferred the right eye, while 33.33% (4/12) were non-lateralized, with an average LI of -0.25. Finally, 48% (11/23) of Scaly-headed parrot preferred the right eye, and 52% (12/23) were non-lateralized, with an average LI of -0.21.

Footedness was initially evaluated in 35 animals (14 white-eyed parakeet, 4 blue-winged macaw, and 17 scaly-headed parrot); however, in only 24 individuals (9 white-eyed parakeet, 4 blue-winged macaw and 11 scaly-headed parrot) was it possible to record the minimum number of 10 food manipulation behaviors needed for data analysis. Of these, 33.33% (8/24) were right-footed, 37.5% (9/24) were left-footed, and 29.16% (7/24) were non-lateralized. The average LI for footedness was -0.04 (**Table 3**). Thus, there was no evident predominance of one side, with approximately one-third of individuals in each condition and an LI close to zero.

Evaluating the three species separately, 22.22% (2/9) of white-eyed parakeet preferred the right foot, 11.11% (1/9) preferred the left foot, and 66.66% (6/9) were non-lateralized. The mean LI was -0.05. For blue-winged macaw, 75% (3/4) were left-footed, and 25% (1/4) were non-lateralized, with an average LI of 0.33. For scaly-headed parrot, 54.54% (6/11) were right-footed and 45.45% (5/11) were left-footed, with an average LI of -0.17. Thus, at the individual level, laterality was present within species, but there was no predominant direction. Among the blue-winged macaw, the sample size was insufficient to conclude a preference for the left foot. Regarding unipedal support, 35 birds were observed (14 white-eyed parakeet, 4 blue-winged macaw, and 17 scaly-headed parrot), but only 11 scaly-headed parrot had sufficient records for

analysis. Of these parrots, 27.27% (3/11) used the right foot for support, 27.27% (3/11) used the left foot for support, and 45.45% (5/11) were non lateralized, with an average LI of -0.01 (**Table 4**). In this analysis, a predominant direction was also not identified.

Table 1: Eye preferences for the white-eyed parakeet (*Psittacara leucophthalmus*), blue-winged macaw (*Primolius maracana*) and scaly-headed parrot (*Pionus maximiliani*) in the following tests: pebble and seed, novel object and reaction to person. We included in table only birds that totaled the minimum of 10 records of asymmetric use of monocular visual field. Laterality index indicates the strength of the asymmetries and ranges from -1 (totally right-eyed) to 1 (totally left-eyed). Abbreviations: R: right; L: left; NL: non-lateralized; LI: Laterality index; NO: novel object test; RP: reaction to person test. Results from chi-square tests (p-values), where * for $p < 0.05$ and ** $p < 0.01$.

Species	Identification band	p-value	LI pebble and seed	Laterality direction pebble and seed	p-value	LI NO	Laterality direction NO	p-value	LI RP	Laterality direction RP
<i>P.leucophthalmus</i>	2008	0.03*	-0.64	R	0.04*	-0.50	R	0.35	-0.17	NL
<i>P.leucophthalmus</i>	2009	-	-	-	0.05*	-0.54	R	0.07	-0.47	NL
<i>P.leucophthalmus</i>	2168	-	-	-	-	-	-	0.03*	-0.64	R
<i>P.leucophthalmus</i>	2381	0.00**	-0.46	R	0.60	0.09	NL	0.04*	0.30	L
<i>P.leucophthalmus</i>	2396	0.00**	0.46	L	0.66	-0.09	NL	0.74	0.05	NL
<i>P.leucophthalmus</i>	7257	-	-	-	0.68	-0.08	NL	0.63	-0.08	NL
<i>P.leucophthalmus</i>	7267	-	-	-	-	-	-	0.36	-0.27	NL
<i>P.leucophthalmus</i>	7275	-	-	-	-	-	-	0.40	-0.23	NL
<i>P.leucophthalmus</i>	7277	-	-	-	-	-	-	0.00**	-1.00	R
<i>P.leucophthalmus</i>	7281	-	-	-	-	-	-	0.38	-0.15	NL
<i>P.leucophthalmus</i>	7283	-	-	-	-	-	-	0.13	-0.38	NL
<i>P.leucophthalmus</i>	7293	0.10	-0.16	NL	0.69	-0.08	NL	0.85	-0.03	NL
<i>P.leucophthalmus</i>	7927	0.22	-0.18	NL	0.73	0.06	NL	0.52	-0.08	NL
<i>P.leucophthalmus</i>	7931	0.00**	-0.56	R	0.62	-0.08	NL	0.00**	0.63	L
<i>P.leucophthalmus</i>	10511	0.02*	-0.60	R	0.00**	-0.69	R	1.00	0.00	NL
<i>P. maracana</i>	31	-	-	-	0.00*	-1.00	R	0.39	0.10	NL
<i>P. maracana</i>	65	-	-	-	0.19	-0.21	NL	0.84	-0.04	NL
<i>P. maracana</i>	104	0.00**	-0.43	R	0.06	-0.25	R	0.38	-0.15	NL
<i>P. maracana</i>	110	0.00**	-0.49	R	0.10	-0.33	NL	0.59	0.10	NL
<i>P. maracana</i>	183	0.46	-0.18	NL	0.04*	-0.50	R	0.20	-0.20	NL

<i>P. maracana</i>	4331	-	-	-	0.56	-0.11	NL	0.02*	0.32	L
<i>P. maracana</i>	8266	-	-	-	0.00**	-0.53	R	0.01*	-0.52	R
<i>P. maracana</i>	8267	-	-	-	0.00**	-0.66	R		-0.02	NL
<i>P. maracana</i>	8341	-	-	-	-	-	-	0.33	-0.16	NL
<i>P. maracana</i>	8978	-	-	-	0.01*	-0.56	R	0.04*	-0.35	R
<i>P. maracana</i>	8990	-	-	-	0.00**	-0.58	R	0.54	0.10	NL
<i>P. maracana</i>	9087	0.00**	-0.61	R	0.72	-0.06	NL	0.02*	-0.39	R
<i>P. maximiliani</i>	39	0.21	0.19	NL	1.00	0.00	NL	-	-	-
<i>P. maximiliani</i>	63	-	-	-	0.126	-0.33	NL	0.08	-0.50	NL
<i>P. maximiliani</i>	64	0.69	-0.05	NL	0.196	0.33	NL	1.00	0.00	NL
<i>P. maximiliani</i>	77	0.06	-0.21	NL	0.274	-0.17	NL	0.05*	-0.33	R
<i>P. maximiliani</i>	4232	0.16	-0.11	NL	0.264	0.14	NL	0.07	0.20	NL
<i>P. maximiliani</i>	4234	0.00**	-0.39	R	0.449	-0.14	NL	0.00**	-0.53	R
<i>P. maximiliani</i>	4303	-	-	-	0.00**	-0.70	R	0.8	0.06	NL
<i>P. maximiliani</i>	4294	0.41	-0.17	NL	-	-	-	0.7	-0.07	NL
<i>P. maximiliani</i>	4334	0.42	0.09	NL	-	-	-			
<i>P. maximiliani</i>	4498	0.42	-0.11	NL	0.25	-0.33	NL	0.08	-0.50	NL
<i>P. maximiliani</i>	8241	0.71	0.05	NL	0.40	-0.23	NL	0.24	0.23	NL
<i>P. maximiliani</i>	8259	0.09	-0.20	NL	-	-	-	-	-	-
<i>P. maximiliani</i>	8268	0.00**	-1.0	R	0.00**	-0.82	R	0.55	-0.12	NL
<i>P. maximiliani</i>	8275	0.27	0.24	NL	-	-	-	-	-	-
<i>P. maximiliani</i>	8287	1.00	0.00	NL	0.64	-0.11	NL	0.53	0.13	NL
<i>P. maximiliani</i>	8328	0.07	-0.47	NL	0.13	0.26	NL	0.11	0.29	NL
<i>P. maximiliani</i>	8352	0.00**	-0.46	R	0.67	-0.06	NL	0.00**	-0.60	R
<i>P. maximiliani</i>	8353	0.00**	-0.31	R	0.24	-0.23	NL	0.19	-0.19	NL
<i>P. maximiliani</i>	8354	0.05	-0.25	R	0.13	-0.45	NL	0.00**	-1.00	R
<i>P. maximiliani</i>	8358	0.00**	-0.41	R	0.25	-0.19	NL	0.03*	-0.30	R
<i>P. maximiliani</i>	8359	0.00**	-0.32	R	0.34	-0.22	NL	0.16	-0.38	NL
<i>P. maximiliani</i>	8993	-	-	-	0.64	0.11	NL	0.87	-0.02	NL

Table 2: Total eye preferences for the white-eyed parakeet (*P.leucophthalmus*), blue-winged macaw (*P. maracana*) and scaly-headed parrot (*P.maximiliani*), after the junction of the tests: pebble and seed, novel object and reaction to person. We included in table only birds that totaled the minimum of 10 records of asymmetric use of monocular visual field. Laterality index indicates the strength of the asymmetries and ranges from -1 (totally right-eyed) to 1 (totally left-eyed). Results from chi-square tests (p-values), where * for $p < 0.05$ and ** $p < 0.01$. Abbreviations: R: right; L: left; NL: non-lateralized; LI: laterality index. R% represents the percentage of use of the right eye and L% the percentage of use of the left eye.

Species	Identification band	R (%)	L (%)	p-value	LI	Laterality direction
<i>P.leucophthalmus</i>	2008	67.90	32.10	0.01*	-0.36	R
<i>P.leucophthalmus</i>	2009	100	0	0.00**	-1.0	R
<i>P.leucophthalmus</i>	2168	100	0	0.00**	-1.0	R
<i>P.leucophthalmus</i>	2381	59	41	0.02*	-0.18	R
<i>P.leucophthalmus</i>	2396	34.90	65.10	0.00**	0.30	L
<i>P.leucophthalmus</i>	7257	55.40	44.60	0.46	-0.11	NL
<i>P.leucophthalmus</i>	7267	100	0	0.00**	-1.0	R
	7275	61.50	38.50	0.58	-0.23	NL
<i>P.leucophthalmus</i>	7277	100	0	0.00**	-1.0	R
<i>P.leucophthalmus</i>	7281	57.60	42.40	0.49	-0.15	NL
<i>P.leucophthalmus</i>	7283	100	0	0.00**	-1.0	R
<i>P.leucophthalmus</i>	7293	56	44	0.14	-0.12	NL
<i>P.leucophthalmus</i>	7927	53.80	46.20	0.40	-0.08	NL
<i>P.leucophthalmus</i>	7931	55.50	44.50	0.11	-0.11	NL
<i>P.leucophthalmus</i>	10511	35.90	34.10	0.01*	-0.32	R
<i>P. maracana</i>	31	68.10	31.90	0.00**	-0.36	R
<i>P. maracana</i>	65	57.10	42.90	0.31	-0.14	NL
<i>P. maracana</i>	104	66.80	33.20	0.00**	-0.34	R
<i>P. maracana</i>	110	63.30	36.70	0.01*	-0.27	R
<i>P. maracana</i>	183	63	37.00	0.03*	-0.26	R
<i>P. maracana</i>	4331	41.60	58.40	0.17	0.17	NL
<i>P. maracana</i>	8266	76.30	23.70	0.00**	-0.53	R
<i>P. maracana</i>	8267	62.50	37.50	0.03*	-0.25	R
<i>P. maracana</i>	8341	51.10	48.90	1.00	-0.02	NL
<i>P. maracana</i>	8978	71.40	28.60	0.00**	-0.43	R
<i>P. maracana</i>	8990	57.60	42.40	0.27	-0.15	NL
<i>P. maracana</i>	9087	70.90	29.10	0.00**	-0.42	R
<i>P. maximiliani</i>	39	47.50	52.50	0.80	0.05	NL
<i>P. maximiliani</i>	63	69.70	30.30	0.04*	-0.39	R
<i>P. maximiliani</i>	64	48.90	51.10	0.92	0.02	NL
<i>P. maximiliani</i>	77	61.30	38.70	0.01*	-0.23	R

<i>P. maximiliani</i>	4232	48.60	51.40	0.68	0.03	NL
<i>P. maximiliani</i>	4234	68.40	31.60	0.00**	-0.37	R
<i>P. maximiliani</i>	4294	57.40	42.60	0.31	-0.15	NL
<i>P. maximiliani</i>	4303	76.10	23.90	0.00**	-0.52	R
<i>P. maximiliani</i>	4334	45.50	54.50	0.49	0.09	NL
<i>P. maximiliani</i>	4498	60	40.00	0.09	-0.20	NL
<i>P. maximiliani</i>	8241	74.10	52.90	0.62	0.06	NL
<i>P. maximiliani</i>	8259	61.10	38.90	0.08	-0.22	NL
<i>P. maximiliani</i>	8268	73.90	26.10	0.00**	-0.48	R
<i>P. maximiliani</i>	8275	56.30	43.80	0.60	-0.13	NL
<i>P. maximiliani</i>	8287	49.20	50.80	1.00	0.02	NL
<i>P. maximiliani</i>	8328	43.20	56.80	0.27	0.14	NL
<i>P. maximiliani</i>	8352	67.70	32.30	0.00**	-0.35	R
<i>P. maximiliani</i>	8353	62.80	37.20	0.00**	-0.26	R
<i>P. maximiliani</i>	8354	68.70	31.30	0.00**	-0.37	R
<i>P. maximiliani</i>	8358	67.20	32.80	0.00**	-0.34	R
<i>P. maximiliani</i>	8359	65.50	34.50	0.00**	-0.31	R
<i>P. maximiliani</i>	8980	100	0	0.00**	-1.00	R
<i>P. maximiliani</i>	8993	49.20	50.80	1.00	0.02	NL

Table 3: Footedness results for the white-eyed parakeet (*P.leucophthalmus*), blue-winged macaw (*P. maracana*) and scaly-headed parrot (*P.maximiliani*). We included in table only birds that totaled the minimum of 10 records of asymmetric foot use. Laterality index indicates the strength of the asymmetries and ranges from -1 (totally right-footed) to 1 (totally left-footed). Results from chi-square tests (p-values), where * for $p < 0.05$ and ** $p < 0.01$. Abbreviations: R: right; L: left; NL: non-lateralized; LI: laterality index. R% represents the percentage of use of the right foot and L% the percentage of use of the left foot.

Species	Identification band	R (%)	L (%)	p-value	LI	Laterality direction
<i>P. leucophthalmus</i>	2168	46.15	53.85	0.78	0.08	NL
<i>P. leucophthalmus</i>	2396	47.06	52.94	0.8	0.06	NL
<i>P. leucophthalmus</i>	7257	72.22	27.78	0.00**	-0.44	R
<i>P. leucophthalmus</i>	7267	52	48	0.84	-0.04	NL
<i>P. leucophthalmus</i>	7277	41.03	58.97	0.26	0.18	NL
<i>P. leucophthalmus</i>	7281	41.67	58.33	0.56	0.17	NL
<i>P. leucophthalmus</i>	7283	26.39	73.61	0.00**	0.47	L
<i>P. leucophthalmus</i>	7927	52.63	47.37	0.69	-0.05	NL

<i>P. leucophthalmus</i>	10511	91.11	8.89	0.00**	-0.82	R
<i>P. maracana</i>	104	52.46	47.54	0.50	-0.05	NL
<i>P. maracana</i>	110	18	82	0.00**	0.64	L
<i>P. maracana</i>	183	24.24	75.76	0.00**	0.52	L
<i>P. maracana</i>	9087	39.90	60.10	0.00**	0.20	L
<i>P. maximiliani</i>	39	15.79	84.21	0.00**	0.68	L
<i>P. maximiliani</i>	64	100	0	0.00**	-1.0	R
<i>P. maximiliani</i>	4232	100	0	0.00**	-1.0	R
<i>P. maximiliani</i>	4234	100	0	0.00**	-1.0	R
<i>P. maximiliani</i>	4294	5.26	94.74	0.00**	0.89	L
<i>P. maximiliani</i>	4498	100	0	0.00**	-1.0	R
<i>P. maximiliani</i>	8241	28.57	71.43	0.01**	0.43	L
<i>P. maximiliani</i>	8352	16.67	83.33	0.00**	0.66	L
<i>P. maximiliani</i>	8353	98.41	1.59	0.00**	-0.97	R
<i>P. maximiliani</i>	8354	80.87	19.13	0.00**	-0.62	R
<i>P. maximiliani</i>	8359	0	100	0.00**	1.0	L

Table 4: Unipedal support results for the scaly-headed parrot (*P. maximiliani*) the only specie that totalized the minimum of 10 records of asymmetric foot use. Laterality index indicates the strength of the asymmetries and ranges from -1 (totally right-footed) to 1 (totally left-footed). Results from chi-square tests (p-values), where * for $p < 0.05$ and ** $p < 0.01$. Abbreviations: R: right; L: left; NL: non-lateralized; LI: laterality index. R% represents the percentage of use of the right foot and L% the percentage of use of the left foot.

Species	Identification band	R (%)	L (%)	p-value	LI	Laterality direction
<i>P. maximiliani</i>	64	76.60	23.40	0.00**	-0.53	D
<i>P. maximiliani</i>	77	71.43	28.57	0.04*	-0.43	D
<i>P. maximiliani</i>	4294	60	40	0.37	-0.20	NL
<i>P. maximiliani</i>	4334	7.32	92.68	0.00**	0.85	E
<i>P. maximiliani</i>	8241	78.57	21.43	0.03*	-0.57	D
<i>P. maximiliani</i>	8259	26.92	73.08	0.01*	0.46	E
<i>P. maximiliani</i>	8268	57.14	42.86	0.51	-0.14	NL
<i>P. maximiliani</i>	8275	23.26	76.74	0.00**	0.53	E
<i>P. maximiliani</i>	8328	37.50	62.50	0.31	0.25	NL
<i>P. maximiliani</i>	8352	73.33	26.67	0.07	-0.47	NL
<i>P. maximiliani</i>	8353	46.15	53.85	0.78	0.08	NL

3.2 Pebble and Seed Test

Individuals of the three species could differentiate the pebbles from the seeds, with an average success rate of 29%. For the white-eyed parakeet, the average success rate was 34% (ranging from 16% to 84%), for the blue-winged macaw it was 10% (ranging from 4% to 30%), and for the scaly-headed parrot it was 31% (ranging from 2% to 76%).

Laterality did not influence the success rate (Mann-Whitney $U = 97$, $p = 0.545$). Through correlation analyses (Spearman's rho, $p < 0.05$), significant positive correlations were found between the success rate and total ocular LI, LI in the pebble and seed test, and seeds consumed. This indicates that the higher the success rate, the more intense the LI for eye preferences (higher scores in LI represent laterality towards the right) and the more seeds were consumed (**Table 5**). Seeds consumed also had significant positive correlations with total ocular LI and pebble and seed LI, indicating that the more seeds consumed, the stronger the LI for eye preferences. Seed pecking was positively correlated with pebble and seed LI, seed consumed, and success rate, indicating that more seed pecks resulted in a higher LI for eye preferences in the pebble and seed test, more seeds consumed, and a higher success rate. Pebble picking was also positively correlated with LI for eye preferences in the pebble and seed test, seed consumed, and success, indicating that more pebble picks resulted in higher LI values, more seeds consumed, and a higher success rate (**Table 6**).

Table 5: Significant correlations between variables from the pebbles and seed test and laterality index from pebble and seed and total ocular index. The cut point for Spearman's Rho correlation was $p < 0.05$. The symbol * represents $p < 0.05$ and ** $p < 0.001$. Abbreviations: LI: laterality index.

Variable 1	Variable 2	Spearman's Rho	p-value
Success	LI total ocular	0.32	0.04*
Success	LI pebble and seed	0.35	0.04*
Success	Seed consumed	1.00	0.00**
Seed consumed	LI total ocular	0.33	0.04*
Seed consumed	LI pebble and seed	0.35	0.04*
Seed peck	LI pebble and seed	0.37	0.03*
Pebble peck	LI pebble and seed	0.38	0.02*

Table 6: Significant correlations between temperament factors and laterality index from pebble and seed, novel object and reaction to person tests and total ocular index, and pebble and seed variables; consumption, success, seed and pebble pecks. The cut point for Spearman's Rho correlation was $p < 0.05$. The symbol * represents $p < 0.05$ and ** $p < 0.001$. Abbreviations: LI: laterality index; RP: reaction to person and NO: novel object.

Variable 1	Variable 2	Spearman's Rho	p-value
Activity	LI total ocular	-0.38	0.01*
Activity	LI pebble and seed	-0.59	0.00**
Activity	Seed consumed	-0.72	0.00**
Activity	Success	-0.72	0.00**
Boldness	LI RP	-0.37	0.01*
Anxiety	LI RP	-0.38	0.01*
Closeness to person	LI NO	0.34	0.02*

3.3 Correlations of Laterality with Temperament

Through factor analysis, the first five factors were retained and presented a cumulative variance of 67.48%. The first factor, Activity, explained 23.10% of the data variance. Variables with a positive load on factor 1 characterized active animals: excitement_{RP} (0.48), excitement_{NO} (0.55), movement_{RP} (0.51), locomotion_{RP} (0.55), top location_{RP} (0.57), top location_{NO}

(0.63), lateral location_{RP} (0.68), locomotion_{NO} (0.70), movement_{NO} (0.70), lateral location_{NO} (0.71). Variables with a negative load represented inactive animals: inferior location_{NO} (-0.81), inferior location_{RP} (-0.75), rest_{NO} (-0.70), rest_{RP} (-0.51), attention_{NO} (-0.47). The second factor, defined as Boldness, explained 17.83% of the data variance. Positive loads characterized bold animals: inferior location_{RP} (0.40), movement_{NO} (0.44), locomotion_{NO} (0.44), quadrant 1_{RP} (0.50), interactions with the novel object_{NO} (0.51), quadrant 1_{NO} (0.56), locomotion_{RP} (0.67), movement_{RP} (0.71). Negative variables represented shy animals: rest_{RP} (-0.71), latency_{NO} (-0.55), quadrant 5_{NO} (-0.50), attention_{NO} (-0.46), latency to touch the person_{RP} (-0.46), rest_{NO} (-0.44).

The third factor, characterized as Anxiety, explained 10.73% of the data variance. Positive loads represented more anxious animals: movement_{NO} (0.41), locomotion_{NO} (0.41), and latency to touch the person_{RP} (0.44). Variables with negative loads characterized less anxious animals: excitement_{RP} (-0.66), quadrant 1_{RP} (-0.61), vocalizations_{NO} (-0.59), excitement_{NO} (-0.56), vocalization_{RP} (-0.50) and rest_{NO} (-0.41). The fourth factor, which explained 8.61% of the data variance, characterized Neophilia and Neophobia. Variables with high positive loads represented neophilic animals: quadrant 1_{NO} (0.52), interactions with the novel object_{NO} (0.63) and touches on the novel object_{NO} (0.67). Negative loads represented neophobic animals: latency to touch the novel object_{NO} (-0.52). The final factor, Closeness to Humans, explained 7.19% of the data variance. Positive variables represented animals that were closer to humans: attention_{RP} (0.49), touches on the person_{RP} (0.54) and interaction with the person_{RP} (0.57). Negative variables represented animals that avoided humans: quadrant 5_{NO} (-0.47), preening_{NO} (-0.46) and quadrant 5_{RP} (-0.40).

Regarding the correlations between laterality and temperament, the first factor, activity, was negatively correlated with the total ocular LI, pebbles and seed LI, consumption, and success rate (**Table 6**). This means that more active birds had stronger right eye preference

(lower LI), consumed fewer seeds, and had a lower success rate. The second factor, boldness, was negatively correlated with LI in the reaction to person test, indicating that bolder birds preferred using their right eye. The third factor, anxiety, had a negative correlation with LI in the reaction to person test. More anxious birds had a stronger preference for using their right eye in this test. The last factor, closeness to humans, was positively correlated with LI in the novel object test, so birds with lower values for this factor also had lower LI, indicating a preference for using their right eye.

3.4 Laterality and social interactions

After observing and recording the positive and negative social interactions of the birds, it was possible to perform a correlation analysis to investigate whether a relationship existed between laterality and sociability in these psittacids. However, no significant correlations were found ($p > 0.05$) between positive and negative social interactions and the laterality index for eye preferences, footedness, and unipedal support.

4 Discussion

In the present study, we reported the presence of laterality in three of Neotropical psittacids species: white-eyed parakeet, blue-winged macaw and scaly-headed parrot. Eye preferences were registered in 28 of the 50 birds analyzed, with a clear predominance of the right eye (27 birds). Footedness was present in 17 of the 24 birds; however, there was no predominant direction. This lack of a preferred side was also observed in unipedal support, as 6 of the 11 scaly-headed parrots demonstrated laterality without a preferred side. Significant correlations were identified between eye preference and the temperament dimensions of activity, boldness, anxiety, and closeness to humans. Finally, the results did not show a significant correlation between laterality and positive or negative social interactions within the analyzed species. Therefore, we partially confirmed our hypothesis that the analyzed psittacids

would exhibit laterality and that there would be a relationship with temperament. However, the absence of a significant relationship between laterality and social interactions was not expected.

The preference for using the right eye among the birds evaluated during the tests represents a behavioral index of hemispheric lateralization, indicating dominance of the left cerebral hemisphere. This hemisphere is responsible for recognizing objects and categorizing information, such as differentiating seeds from pebbles (Rogers; Anson, 1979; Rogers, 2017). It has been suggested that individuals who rely more on the left hemisphere during foraging place less load on the right hemisphere, which remains more alert to external threats. This allows the animal to perform both tasks (foraging and vigilance) simultaneously and more efficiently (Rogers et al., 2004; Rogers, 2008). Although the tests in the present study did not include a model of predator, the presence of an unknown observer may have been perceived as a potential threat, possibly dividing the birds' attention during the tests also seen in Magat; Brown, 2009.

The percentage of left- and right-footed psittacids in the present study was similar (nine left-footed and eight right-footed), making it impossible to support the predominance of one side and suggesting laterality at individual level, but not population-level. In the blue-fronted Amazon parrot (*Amazona aestiva*), similar proportions were found, with five left-footed parrots and six right-footed (Godinho et al., 2020). However, in the literature for Psittaciformes, left-footedness tend to be more common, as seen in the orange-winged Amazon parrot (*Amazona amazonica*) (Cussen; Mench, 2014) and several Australian Psittaciformes (Magat; Brown, 2009; Brown; Magat, 2011a; Brown; Magat, 2011b). Previous studies have shown a correlation between eye and foot preferences, with the same side being preferred for both behaviors. However, in some birds, as observed in the present study, different directions were found. For example, in the Cacatuidae family, a strong preference for the left foot to manipulate food was found, but the right eye was preferred to analyze it (Magat; Brown, 2009).

We identified laterality in natural behaviors such as unipedal support in six individuals of scaly-headed parrots. However, no predominant direction was observed: half were left-footed, and the other half were right-footed. These findings suggest laterality at the individual level, but not population-level, similar to what has been observed in budgerigars (*Melopsittacus undulatus*) (Duggan et al., 2016; d'Antonio-Bertagnolli; Anderson, 2018). In these studies, laterality was identified in two individuals with a significant unipedal preference for the left foot. For white-eyed parakeet and blue-winged macaw, the minimum number of unipedal support records was not reached, possibly due to the temperament of these birds, which become more agitated and skittish in the presence of an observer, resulting in less time spent resting during observation. According to the literature on psittacid temperament, bolder grey parrots (*Psittacus erithacus erithacus*) and white-eyed parakeet tend to move more in the presence of an observer and, when exposed to stressful stimuli, tend to take more risks and exhibit high levels of activity (van Zeeland et al., 2013; Ramos et al., 2021).

The results of the pebble and seed test revealed a variation in success rate among the three species, and laterality did not influence success, which contrasts with findings in the literature where left-footed blue-fronted parrots had better performance in this test than right-footed ones (Godinho et al., 2020). However, the success rate was positively correlated with the pebble and seed, total ocular LI, and seed consumed. Absolute values of LI indicate the strength of the asymmetries (Rogers, 2017), so the higher the success rate, the stronger the eye preferences and the more seeds were consumed. This finding corroborates what Magat and Brown (2009) proposed, suggesting that the direction of laterality has little influence on success, with the strength of lateralization being the main predictor of performance. In a similar experiment, strongly lateralized pigeons (*Columba livia*) had more success in foraging than non-lateralized ones (Güntürkün et al., 2000). A positive correlation was found between seed picking, seed consumed, and LI in the pebble and seed test, demonstrating that stronger

laterality (in this study, higher absolute LI values indicate right-side preferences) led to more seed picking and consumption. Similarly, pigeons and chicks, those with stronger right-eye asymmetries had better performance in avoiding pebbles and greater success (Rogers, 2017).

Another relationship observed was the negative correlation between the first temperament factor, activity, with the pebble and seed laterality index, seed consumed, and success. This means that higher activity is associated with stronger right laterality (lower LI), fewer seeds consumed, and consequently a lower success rate. Higher activity values correspond to more excitable animals, that display more movements (van Zeeland et al., 2013; Ramos et al., 2021). This could result in lower interaction with the test, which requires the animal to stand still and be attentive to the tray.

The second factor, boldness, was negatively correlated with the reaction to person LI. Therefore, the bolder the birds (those who moved more and approached novel objects and people), the greater the intensity of right-eye laterality. Previous research revealed similar results. For example, in pigs, laterality direction played an important role in temperament, with right-lateralized individuals being bolder and more exploratory in novel contexts (Goursot et al., 2019). The strength of laterality was also related to boldness (Reddon; Hurd, 2009; Goursot et al., 2019). In fish, for instance, strongly lateralized individuals explored their environment more thoroughly and were bolder in new environments (Reddon; Hurd, 2009). Furthermore, previous studies suggested that individuals with left hemispheric dominance were bolder and more exploratory than those with right hemispheric dominance (Goursot et al., 2021).

The third dimension, anxiety, was negatively correlated with the reaction to person laterality index. This indicates that anxious birds moved more, had higher latency to approach the person, and showed stronger right eye preference. Previous studies on this temperament dimension suggest that anxiety is related to fear and vigilance behaviors (Paulino et al., 2018; Coutant et al., 2018). Emotion processing differs between the cerebral hemispheres

(MacNeilage et al., 2009), with each hemisphere responsible for motor control of the opposite side of the body (Rogers et al., 2013). Literature suggests that positive emotions or approach motivations are processed by the left hemisphere (right eye), while the right hemisphere (left eye) processes negative emotions or withdrawal motivations (Goursot et al., 2021). Therefore, the correlation identified in the present study differs from previous research, as it was expected that anxiety would be processed by the right hemisphere (related to left eye preference). However, studies with other bird species, such as dark-eyed juncos (*Junco hyemalis*), showed results similar to ours, with a dominance of the left hemisphere (and right eye) when birds observed a model predator (a frightening stimulus) (Franklin; Lima, 2001).

The fifth dimension, closeness to humans, had positive correlations with novel object LI. Birds with higher avoidance of humans (staying distant from the person during the test) had higher LI values, indicating a preference for the right eye. Studies on hemispheric asymmetries indicate that the right hemisphere (left eye) is involved in the physiological response to stress, engaging the pituitary, hypothalamus, and adrenal axis (Rogers, 2010; Ocklenburg et al., 2016; Goursot et al., 2021), and controls reactive behaviors, while the left hemisphere (right eye) controls proactive behaviors (Rogers, 2009, 2010; Goursot et al., 2021). Therefore, our results do not support these proposals since the birds analyzed exhibited right laterality (dominance of the left hemisphere). Since there was no preference for the left eye in the novel object LI, it is possible to infer that weaker LI towards the right indicates greater proximity to humans.

No significant correlations were found between positive and negative social interactions and laterality. In budgerigars, the absence of a significant relationship between laterality and aggressive social interactions was attributed to the non-establishment of hierarchy among individuals, as it was a new colony (Duggan et al., 2016). Similarly, in the present study, which was conducted in a release area where individuals frequently arrive for rehabilitation and leave for reintroduction, the hierarchy between birds may not be fully established. Our results differ

from what was found in the literature. For example, in other bird species, such as Caribbean flamingos (*Phoenicopterus ruber*), a preference for resting the neck on the left side is associated with a higher likelihood of involvement in aggressive encounters (Anderson et al., 2010). In domestic pigs, a preference for the right side was associated with a greater number of vocalizations during an open field test, indicating that right-handed pigs are more sociable than left-handed ones (Goursot et al., 2019).

As an implication of the present study, laterality is a valuable indicator of hemispheric asymmetries that contribute to animal welfare, as it can help identify individuals that may be more vulnerable to stress, aggression, and reactivity to novel situations (Rogers, 2009; Found; St. Clair, 2016; Goursot et al., 2019). Through this understanding, personalized management strategies can be created, such as handling the animal from the correct side in response to fearful situations (Leliveld et al., 2013), in zoos and rehabilitation centers like CETAS. Furthermore, the relationship between laterality and temperament enable to identify bolder, more exploratory, and less aggressive individuals (Goursot et al., 2021). This consideration is important in release programs, as selecting birds with different temperament traits could improve the survival chances of the group (Paulino et al., 2018).

It is important to highlight the weaknesses of the present study. For pebble and seed, footedness and unipedal support analyses, only four individuals of blue-winged macaw were available to participate on the tests. Thus, we cannot consider the results as a pattern of the specie, since it could be individual bias. The difference in sample size occurred due to the unpredictability of the birds arrival at the wildlife rehabilitation center studied, what could be a reality for researches involving wild specimens kept under human care. Despite this limitation, the results obtained are promising and can be valuable in helping to design future studies on this topic.

5 Conclusion

In the present study, we identified laterality in the three analyzed species: white-eyed parakeet, blue-winged macaw, and scaly-headed parrot, through right-eye preferences. The birds were lateralized for footedness and unipedal support; however, there was no predominant direction for these behaviors. Our findings contribute to the knowledge of lateral preferences in Neotropical Psittacidae species, which have little representation in the scientific literature. Furthermore, the strength of laterality showed a significant correlation with the success rate in the pebble and seed test, corroborating the hypothesis that the intensity of laterality has more influence on cognitive capacity than the direction. For the first time in Psittacidae, significant correlations were identified between right-eye preferences and the temperament dimensions activity, boldness, anxiety, and closeness to humans. Our results also suggest that emotional valence and laterality are related, and identifying the direction of side preferences could indicate the emotional responses of the birds (e.g., whether they are more reactive, aggressive, or fearful). Therefore, we highlight the importance of studies on laterality, temperament, and emotional asymmetries for animal welfare, as they help create more individualized welfare strategies. For future research, we suggest ontogenetic analyses of laterality in Psittaciformes and phylogenetic studies to understand its influence on Neotropical species. The analyses of footedness and unipedal support in blue-winged macaws had limitations, such as an insufficient number of individuals, so it would be beneficial to replicate this study with a larger sample size.

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5 GENERAL CONCLUSION

Through this dissertation, it was possible to obtain an overview of the data present in the literature on laterality in psittaciformes, identifying the most studied species, the main methodologies, and the topics correlated with laterality. In addition, we present an empirical study with three Neotropical species (*Psittacara leucophthalmus*, *Primolius maracana*, *Pionus maximiliani*), correlating laterality with the temperament and social interactions of these birds.

In Chapter 1, the results obtained from the scoping review revealed a general preference for the left foot. There was insufficient evidence to establish a preference for either eye. The most investigated topics were the relationship between laterality and cognition. Additionally, less explored topics were identified, such as phylogenetic and ontogenetic aspects, along with a lack of studies involving Latin American species.

In Chapter 2, we investigated laterality in three species present in the Brazilian fauna: *Psittacara leucophthalmus*, *Primolius maracana*, and *Pionus maximiliani*. The results indicated a preference for the right eye and individual-level laterality for footedness and unipedal support behaviors. Through the pebble and seed test, it was identified that these birds could distinguish the seeds from the pebbles, with the strength of laterality acting as a more important factor than the direction itself. Four temperament factors—activity, boldness, anxiety, and closeness to humans—were correlated with right eye laterality. Finally, positive and negative social interactions did not show significant correlations with laterality.

Finally, we emphasize the importance of this issues for animal welfare science, as identifying lateral preferences is an easy and non-invasive way to identify birds that are more predisposed to stress, fear, and aggression. This would allow managers to develop individualized management strategies, considering individual differences and emotional responses of each animal under human care.

Appendices

Appendices 1. Categories recorded in the two temperament tests: novel object (NO) and reaction to person (RP), for the three species studied: *Pionus maximiliani*, *Primolius maracana*, and *Psittacara leucophthalmus*.

Placement categories	
Place	Place where the animal is: top grid, bottom grid, or side grid.
Quadrant	Quadrant where the animal is. The animal can be at the 1 st (0-20 cm), 2 nd (21-40 cm), 3 rd (41-60 cm), 4 th (61-80 cm), or 5 th (81-117 cm) quadrant. The distances of the quadrants were given in relation to the position of the stimulus in the tests.
Activity categories	
States	
Rest	The animal remains at the same spot. It can stand using both feet, just one foot or its beak while other body parts remain still or agitated. The animal may be spinning without changing places. There is no locomotion of the animal.
Movement	The animal changes its position, moving from one spot to another. It can move by flight, walking, or using its beak to climb the cage bars, branches, and perches.
Attention	The animal is attentive to the novel object or person and external stimuli (such as observers, other birds in the aviary, predators flying over the aviary, dogs, and keepers). The animal stays alert. The bird can quickly move his head in different directions or remain with his head still with its neck stretched, while focusing on the stimulus. One or both eyes open. The animal can move its feet without leaving the spot. Not accounted for when the bird is expressing another behavior.
Inactivity	The animal rests. It can have its feathers ruffled, one foot tucked, or its head turned back, tucked between its wings. The animal sleeps, not attentive to any stimuli, whether from the test or the environment, and has both eyes closed.
Locomotion	The animal moves from one spot to another, walking, or flying. It can use its beak to help in locomotion while walking on the cage grid, perches, or on the ground.
Excitement	The animal moves any part of its body but stays at the same spot, swinging, turning, or flapping its wings. It can use the beak as a support while shaking its body. It can remain with its feet still and its body slightly lowered while its wings tremble. The animal raises and lowers its body quickly.

Preening	The animal adjusts its feathers using its beak, stretches its wings, stretches its legs, ruffles its feathers, yawns, scratches itself, or cleans its beak. In the aviary, the bird can take a bath.
Environment interaction	The animal interacts with the cage, pecking at the paint used to mark the quadrants of the enclosure, the grid, or the cloth used to cover the sides of the cage. It can interact with elements of the environment that were picked up by the beak (“chewing”). In the aviary, it can interact with environmental enrichments (perches and leaves) and other objects in the enclosure.
Novel object interaction	The animal interacts with the novel object by pecking it or touching it with the foot.
Person interaction	The animal interacts with the person by attempting to peck them or touch them with the foot.
Feeding	The animal eats fruits or sunflower seeds from feeders or the ground. The animal can feed on enrichments, but only if they are fruits or flowers.
Allopreening	One animal cleans and preens the feathers of another. Not accounted as a positive social interaction.
Others	The animal does not perform any of the activities above. The animal interacts positively or negatively with another animal. Allofeeding. Attempted copulation. The animal performs stereotypical behaviors.
Unipedal Support	The animal remains in a resting posture, using one of its feet to support itself, while the other is tucked close to the body.
Events	
Latency to touch the novel object	Time (in seconds) to touch the novel object for the first time.
Latency to try to touch the person	Time (in seconds) to try to touch the person's hand for the first time.
Touch in the novel object	Number of touches in the new object, either with the beak or feet.
Attempts to touch the person	Number of attempts to try to touch the person's hand, either with the beak or feet.
Vocalizations	The animal vocalizes, including human vocalizations (whistles, songs, etc.)
Abnormal behavior	The animal exhibits stereotypical behavior or any other behavior that is not like the species.
Aggressive conduct	The animal displays aggressive behavior, ruffling its feathers, opening its wings and beak advancing towards the object/person, and/or retreating.

Reaction to attempts to capture	The animal runs away from the person (1), remains still in the same spot (2), or tries to attack the person (3) when being captured at the end of the reaction to person test.
Positive social interactions	The animal tries to mate with another or allofeeds.
Negative social interactions	The animal pecks and kicks or threatens to peck and kick another individual. There is usually a winner and a loser in the interaction. The one who loses leaves the spot, and the other who wins stays in the same place.
Human vocalizations	The animal emits vocalizations of human nature, such as whistles, songs, and words.

Annex



MINISTÉRIO DA EDUCAÇÃO
UNIVERSIDADE FEDERAL DE JUIZ DE FORA
CERTIFICADO

Certificamos que a proposta intitulada "**Lateralidade, temperamento e comportamento em cativo em Psitacídeos**" protocolo número 008/2023, sob responsabilidade de **Aline Cristina Sant'Anna, Larissa Gomes de Jesus, Gustavo Nunes de Almeida, Gabriela de Araújo Porto Ramos e Maria Eduarda Caçador Branco** – que envolve a utilização de animais pertencentes ao filo *Chordata*, subfilo *Vertebra* (exceto homem), para fins de pesquisa científica - está de acordo com os preceitos da Lei 11.794 de 8 de outubro de 2008, com o Decreto 6.899 de 15 de Julho de 2009, bem como normas editadas pelo Conselho de Controle de Experimentação Animal (CONCEA), e foi **APROVADA** pela Comissão de Ética no Uso de Animais da Universidade Federal de Juiz de Fora na reunião de 07/03/2023.

We certify that the proposal "**Lateralidade, temperamento e comportamento em cativo em Psitacídeos**", protocol number 008/2023, under the responsibility of **Aline Cristina Sant'Anna, Larissa Gomes de Jesus, Gustavo Nunes de Almeida, Gabriela de Araújo Porto Ramos e Maria Eduarda Caçador Branco** – which involves the use of animals belonging to the phylum *Chordata*, subphylum *Vertebra* (except human beings), for scientific research purposes – is in accordance with Law 11.794 of October 8, 2008, and Decree 6899 of July 15, 2009, as well as with the rules issued by the National Council for Control of Animal Experimentation (CONCEA), and was **APPROVED** by the Ethic Committee of Animal Use of the Federal University of Juiz de Fora/MG in the meeting of 03/07/2023.

Finalidade	() Ensino (x) Pesquisa Científica
Vigência da autorização	08/05/2023 a 08/05/2026
Médico Veterinário responsável pela pesquisa ART e vigência	Laura Silva de Oliveira CRMV-MG 16939
Espécie/linhagem/raça	Maritaca (<i>Psittacara leucophthalmus</i>), Maracanã (<i>Primolius maracanã</i>), Maitaca (<i>Pionius maximiliani</i>), Tuim (<i>Forpus xanthopterygius</i>), Papagaios (<i>Amazona</i> sp.) e Araras (<i>Ara ararua</i> , <i>Ara chloropterus</i>)
Nº de animais	45, divididos da seguintes forma: - 15 Maritacas (<i>Psittacara leucophthalmus</i>) - 15 Maracanã (<i>Primolius maracanã</i>) - 15 Maitaca (<i>Pionius maximiliani</i>) - 10 Tuim (<i>Forpus xanthopterygius</i>)

	- 15 Papagaios (Amazona sp., n = 15) - 5 Araras (Ara araruana e Ara chloropterus)
Peso/Idade	Variável
Sexo	Variável
Procedência dos animais	Centro de Triagem de Animais Silvestres (CETAS, Juiz de Fora, MG)
Local de manutenção dos animais	Centro de Triagem de Animais Silvestres (CETAS, Juiz de Fora, MG)

APROVEITAMOS A OPORTUNIDADE PARA INFORMAR QUE:

- 1) O projeto/treinamento de pesquisa deve ser desenvolvido conforme delineado no processo aprovado;
- 2) A CEUA/UFJF deve ser informada de todos os fatos relevantes que alterem o curso normal do projeto/treinamento. É papel do pesquisador responsável assegurar medidas imediatas adequadas frente a e evento não previstos.
- 3) Eventuais modificações ou emendas ao processo devem ser apresentadas à CEUA/UFJF de forma clara e sucinta, identificando a parte a ser modificada e suas justificativas.
 - No caso de treinamento, esta aprovação tem validade de 12 meses a partir da data de aprovação pela CEUA/UFJF e para cada evento realizado nesse período, o pesquisador responsável deverá apresentar um relatório das atividades realizadas;
 - No caso de projeto de pesquisa, esta aprovação tem validade de acordo com o cronograma proposto no protocolo. Os relatórios deverão ser enviados a cada 12 (doze meses), a partir da data de aprovação da CEUA/UFJF.
- 4) Havendo interesse na renovação do projeto, a solicitação deverá ser protocolada até o último dia de validade da atual proposta. Após esta data uma nova proposta deverá ser encaminhada.

Atenciosamente,

Coordenação da CEUA/UFJF



Documento assinado eletronicamente por **Ana Eliza Andreazzi, Professor(a)**, em 21/03/2023, às 09:29, conforme horário oficial de Brasília, com fundamento no § 3º do art. 4º do [Decreto nº 10.543, de 13 de novembro de 2020](#).



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SUPERINTENDÊNCIA DO IBAMA NO ESTADO DE MINAS GERAIS

OFÍCIO Nº 189/2023/SUPES-MG

Belo Horizonte/MG, na data da assinatura digital.

À Sra.

MARIA AMÉLIA DE CONI E MOURA MATTOS LINS

Diretora Geral

INSTITUTO ESTADUAL DE FLORESTA-IEF/MG

E.mail: dg.ief@meioambiente.mg.gov.br

Assunto: Autorização para a realização da Pesquisa "Lateralidade, temperamento e comportamento em cativeiro em Psitacídeos".

Referência: Caso responda este Ofício, indicar expressamente o Processo nº 02015.000580/2023-04.

Com nossos cumprimentos, faço referência ao Projeto de Pesquisa "Lateralidade, temperamento e comportamento em cativeiro em Psitacídeos", que envolve o estudo com animais que dão entrada no Cetas do IBAMA em Juiz de Fora, protocolado sob número SEI 15933510 anexo.

Após análise do referido Projeto, conforme Parecer Técnico nº 1/2023-Cetas-JUIZ DE FORA-MG/Ditec-MG/Supes-MG (15933661) anexo, informo estar de acordo com a realização do projeto conforme apresentado e encaminhado para sua apreciação, considerando que trata-se de gestão compartilhada do Cetas entre o Ibama e o IEF/MG.

ANEXO:

- Projeto de Pesquisa (15933510);
- Parecer Técnico nº 1/2023-Cetas-JUIZ DE FORA-MG/Ditec-MG/Supes-MG (15933661).

Atenciosamente,

Pedro Paulo Ribeiro Mendes de Assis Fonseca
SUPERINTENDENTE SUBSTITUTO DO IBAMA-MG



Documento assinado eletronicamente por **PEDRO PAULO RIBEIRO MENDES DE ASSIS FONSECA**, **Superintendente Substituto**, em 02/06/2023, às 15:26, conforme horário oficial de Brasília, com fundamento no art. 6º, § 1º, do [Decreto nº 8.539, de 8 de outubro de 2015](#).



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ESTADO DE MINAS GERAIS
 INSTITUTO ESTADUAL DE FLORESTAS
 Gerência de Conservação e Restauração de Fauna Silvestre Terrestre

AUTORIZAÇÃO

Referência: Processo nº 2100.01.0079445/2021-55

AUTORIZAÇÃO Nº 68003390	PERÍODO DE VALIDADE 19/06/2023 A 19/06/2024	PROCESSO IEF Nº 2100.01.0004298/2023-69
TÍTULO DO PROJETO: "Lateralidade, temperamento e comportamento em cativeiro em Psitacídeos."		
INSTITUIÇÃO: Universidade Federal de Juiz de Fora ENDEREÇO: Rua José Lourenço Kelmer, s/n - São Pedro, Juiz de Fora - MG		
PROFESSOR/PESQUISADOR COORDENADOR DAS ATIVIDADES: NOME: Drª Aline Cristina Sant'Anna CPF: 324.097.638-20 NACIONALIDADE: Brasileira		
EQUIPE TÉCNICA (NOME/CPF/REGISTRO DE CLASSE): Larissa Gomes de Jesus (mestranda) - UFJF Gustavo Nunes de Almeida (graduando em Ciências Biológicas) - UFJF Maria Eduarda Caçador Branco (graduanda em Ciências Biológicas) - UFJF Gabriela de Araújo Porto Ramos (doutoranda) - UFJF		
AUXILIARES DE CAMPO (NOME/CPF OU IDENTIDADE): Não se aplica.		
LOCAL(IS) ONDE AS ATIVIDADES SERÃO EXECUTADAS: Centro de Triagem e Reabilitação de Animais Silvestres – CETRAS/IEF – Juiz de Fora MUNICÍPIO: Juiz de Fora UF: Minas Gerais Áreas de Soltura de Animais Silvestres (ASAS – IBAMA/IEF), MUNICÍPIO: Santana do Deserto MUNICÍPIO: Bias Fortes. UF: Minas Gerais		
TÁXONS A SEREM ESTUDADOS: Aves - Psitacídeos TIPO DE ATIVIDADE: Observação e aplicação de testes não invasivos nos animais.		
DESCRIÇÃO DAS ATIVIDADES: Será realizada avaliação da lateralidade por meio de filmagens dos animais no momento da alimentação a fim de identificar qual membro é utilizado para manipular o alimento. Para as análises da relação das preferências visuais (<i>eyednees</i>) e habilidade de forrageamento, será realizado o teste de seixo e sementes, com os animais mantidos individualizados, distribuindo em uma bandeja de 30 cm x 40 cm, 35 sementes em meio a 50 pedregulhos de tamanho e cor similares às sementes. Os animais serão avaliados por 5 minutos, sendo filmados durante esse período. Para avaliação do temperamento em cativeiro serão aplicados dois testes para avaliar o temperamento para espécimes de maitacas e maracanãs, o teste do novo objeto e teste de reação à pessoa desconhecida. Para avaliação dos comportamentos em cativeiro serão realizadas apenas para maitacas e maracanãs no viveiro de experimentação da área ASAS, com os animais de cada espécie mantidos em dois grupos separados. A etapa do monitoramento será realizada apenas com as maitacas por meio de colocação de radiocolar.		

CRONOGRAMA DE ATIVIDADES (ATIVIDADES COM MÊS E ANO DE INÍCIO E FIM):

Revisão de literatura: segundo semestre de 2023, ano de 2024 e primeiro trimestre de 2025.
 Coleta de dados sobre lateralidade: Terceiro trimestre de 2023.
 Coleta de dados sobre temperamento: terceiro e quarto trimestre de 2023.
 Coleta de dados sobre comportamento: segundo semestre de 2023 e primeiro trimestre de 2024
 Monitoramento pós soltura em maitacas: Quarto trimestre de 2023 e primeiro trimestre de 2024.
 Processamento e análise dos dados: Segundo e terceiro trimestre de 2024.
 Redação e submissão de resumos e artigos: segundo, terceiro e quarto trimestre de 2024.
 Relatório final para IBAMA e IEF: primeiro trimestre de 2025.

MATERIAIS E MÉTODOS:*Coletas de dados***a) Avaliação da lateralidade**

Para as análises de preferência de uso de membro (*footedness*), serão realizadas filmagens dos animais no momento da alimentação a fim de identificar qual membro é utilizado para manipular o alimento, seguindo a metodologia proposta por Magat e Brown 188 (2009). Os indivíduos participantes do estudo serão identificados pelo número da anilha CETAS - IBAMA/IEF e por meio de tinta marcadora atóxica e separados em um viveiro para a realização das filmagens. No momento da marcação individual com tinta todos os indivíduos incluídos no projeto serão pesados. As avaliações ocorrerão no período da manhã, concomitantemente com o horário da oferta de alimento pela equipe do CETAS, que ocorre às 07 horas. Serão oferecidas bandejas de sementes, ração para psitacídeos e frutas, colocada próximo a poleiros. Para que os animais consigam acessar o alimento, eles precisarão se segurar no poleiro com um pé e com o outro manipular os itens alimentares. Os experimentos serão filmados para posterior análise dos vídeos. Os testes serão replicados por três semanas, nas segundas, quartas e sextas, totalizando nove avaliações por espécie.

Para as análises da relação das preferências visuais (*eyednees*) e habilidade de forrageamento, será realizado o teste de seixo e sementes. Esse teste consiste em analisar a preferência ocular ao visualizar as sementes, além de permitir avaliar a capacidade do psitacídeo em distinguir o alimento em meio a pedregulhos, um problema que pode vir a acontecer durante o forrageamento. Os testes serão realizados com os animais mantidos individualizados. Irar-se distribuir em uma bandeja de 30 cm x 40 cm, 35 sementes em meio a 50 pedregulhos de tamanho e cor similares às sementes. Os animais serão avaliados por 5 minutos, sendo filmados durante esse período. A partir da análise dos vídeos será contabilizada a preferência ocular, além de número de bicadas e quantidade de sementes consumidas. O sucesso na resolução do teste será contabilizado pela quantidade de sementes consumidas dividida pela quantidade total de seixos. Os testes serão repetidos por três vezes com cada animal, sendo comparado o desempenho dos animais ao longo dos testes para analisar se a lateralidade se manteve. Para estimular a participação dos animais, os testes serão realizados antes do horário da alimentação. Para analisar-se a lateralidade na expressão de comportamentos naturais como uso do pé de repouso, serão feitas filmagens dos animais no poleiro, para análise das assimetrias no apoio unipodal. Com essas imagens também será possível analisar a lateralidade no contexto das interações sociais dos animais no viveiro. Os comportamentos serão analisados através de observações de 07:00 às 09:00 durante cinco dias consecutivos e registraremos as seguintes categorias: repouso, manutenção das penas, movimentação, exploração, alimentação e alolimpeza (analisadas por % do tempo de observação) e interações sociais positivas e negativas (em ocorrências/minuto).

b) Avaliação do temperamento em cativeiro

Serão aplicados dois testes para avaliar o temperamento para espécimes de maitacas e maracanãs, o teste do novo objeto e teste de reação à pessoa desconhecida. Eles serão feitos individualmente no viveiro de experimentação da área ASAS. Cada teste terá duração de 5 minutos e as categorias comportamentais serão registradas através do método de amostragem focal com intervalos amostrais de 20 segundos. As categorias de comportamento registradas serão: alerta, inativo, movimentação, manutenção, interação com o ambiente, vocalização, latência para tocar no novo objeto, número de toques no novo objeto e distância de fuga da pessoa desconhecida. Cada teste será repetido 3 vezes com um intervalo de 10 a 15 dias entre as repetições.

c) Avaliação dos comportamentos em cativeiro

As observações comportamentais também serão realizadas apenas para maitacas e maracanãs no viveiro de experimentação da área ASAS, com os animais de cada espécie mantidos em dois grupos separados. Cada grupo será transferido do viveiro de manutenção para o viveiro de experimentação três dias antes do início das observações. Elas serão realizadas por 5 dias por grupo, no período da manhã, de 07:00 às 09:00, e no período da tarde, de 15:00 às 17:00. Os comportamentos serão registrados por um único observador através do método de amostragem focal com intervalos amostrais de 5 minutos. As categorias comportamentais registradas serão: repouso, manutenção, movimentação, alimentação, interação com o ambiente, alolimpeza, vocalização, vocalização humana, comportamento anormal, interação social negativa (chute, bicada, ameaça de chute e ameaça de bicada) e positiva (alolimpeza).

d) Soltura e monitoramento pós-soltura

A etapa do monitoramento será realizada apenas com as maitacas. Os rádio-colares serão distribuídos entre os indivíduos com escores mais significativos do traço de temperamento avaliado. Primeiro serão colocados colares falsos de tamanho e peso iguais aos colares verdadeiros, por 15 dias, para que as maitacas se habituem com o objeto. Um dia antes da soltura, os colares falsos serão substituídos pelos verdadeiros. A tecnologia de rádio telemetria auxilia no encontro dos animais através da transmissão e recepção de sinais provenientes dos colares, técnica bastante utilizada em estudos de monitoramento após reintrodução (Allard et al., 2019; Bernardo et al., 2011; Bremner-Harrison et al., 2004; de Milliano et al., 2016; Haage et al., 2017; Lopes et al., 2017; Pratalongo, 2004; Sinn et al., 2014). No dia da soltura, a porta do viveiro será aberta e as maitacas poderão sair por conta própria, configurando uma soltura branda, método relacionado com maiores taxas de sucesso em programas de translocação (Resende et al., 2021). A porta ficará aberta até que o último indivíduo deixe o viveiro e depois disso será fechada. Alimentação suplementar será fornecida próxima aos viveiros durante os cinco primeiros meses (ou até que as aves estejam sendo avistadas consumindo o alimento fornecido), para que elas tenham uma fonte de alimento conhecida e segura, enquanto se familiarizam com o novo

ambiente. Essa técnica é um fator comum em reintroduções de sucesso de psitacídeos (White et al., 2012). Imediatamente após a abertura dos viveiros, serão quantificados: o tempo que cada ave permaneceu no viveiro após a abertura do portão; a ordem de saída de cada ave do viveiro; retornos para o viveiro após saírem; o consumo da alimentação suplementar por animal e o tempo de consumo da alimentação suplementar por animal. Para acompanhar as aves que se dispersarem da área de soltura, a equipe sairá em dois turnos por dia, às 05:00 e às 15:00, em busca das aves e serão registradas: a sobrevivência; as coordenadas do local em que o animal se encontra; o comportamento dos indivíduos na natureza e se os indivíduos soltos estão ou não na presença de outros coespecíficos nativos.

Análises dos dados

Para determinar a lateralidade em cada um dos testes (*footedness*, membro de apoio e *eyedness*) será utilizada a fórmula $LI = [(L - R) / (L + R)]$, na qual LI representa o índice de lateralidade, L utilização do membro ou olho esquerdo e R utilização do membro ou olho direito. Valores positivos da LI representam preferência para a esquerda e valores negativos preferência para a direita, tendo resultados variando de -1 a 1. Também será feito o teste focado em um lado para confirmar a lateralidade direcional $LI = [(R / L + R) \times 100]$, em que valores obtidos maiores de 50% indicam lateralidade para a direita e menores que 50% lateralidade para a esquerda, quanto mais distante da média maior a força da lateralidade (Wiper, 2017).

Posteriormente, Modelos Lineares Generalizados (GLM) serão aplicados para avaliar as relações da espécie e da massa corporal com a lateralidade de cada teste. Os modelos irão incluir as variáveis indicativas de lateralidade de cada teste (L e LI), o efeito fixo de espécie, além da covariável massa corporal (em g).

Para avaliação do temperamento, primeiramente será aplicada uma análise de fatores para redução da dimensionalidade dos dados e obtenção das dimensões principais do temperamento, conforme descrito em Ramos et al., 2021. Em seguida, os escores dos animais em cada um dos fatores será correlacionado com as variáveis indicativas de lateralidade e do comportamento em cativeiro, por meio de testes de correlação de Pearson.

Por fim, serão utilizados testes de associação (correlação de Pearson ou Spearman, teste de qui-quadrado em tabela de contingência) para relacionar a lateralidade, o temperamento e os comportamentos em cativeiro com as variáveis obtidas a partir do monitoramento pós-soltura.

DESTINO DO MATERIAL BIOLÓGICO COLETADO:

Não se aplica.

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- O pesquisador deverá encaminhar à Gerência de Conservação e Restauração de Fauna Silvestre Terrestre do IEF, relatório final das atividades conforme estabelecido na Portaria IEF nº 148/2013, informando o número do processo IEF mencionado nesta autorização.
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- O titular desta autorização ou os membros da equipe técnica que estiverem exercendo as atividades no Cetras deverão estar sempre acompanhados desta autorização para apresentá-la, quando solicitado.

SEI/GOVMG - 68003390 - Autorização

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Belo Horizonte, 19 de junho de 2023.



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