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President's Corner jonahrzy@gmail.com

President's Corner

It is a great honor for me to extend my warmest greetings to all primatologists around the world who work closely with the affiliated primate societies. Together, we have continually emphasized the critical importance of protecting and conducting research on non-human primates, which face numerous threats, primarily due to habitat loss.

During this noble mission, we have lost several remarkable figures who contributed immensely to the field of primatology. Among them was Dr. Milton de Mello, one of the founding members of the Brazilian Society of Primatology, whose significant contributions helped advance Brazilian and Latin American primatology. We also mourned the passing of Dr. Marie Randrianarison, an active member of GERP Madagascar, who was dedicated not only to research but also to education. Furthermore, we lost Dr. Thelma Rowell, a pioneering primatologist renowned for her studies on social behavior and reproduction in various catarrhine monkeys. These individuals were forces to be reckoned with, and their loss has deeply affected us all. Together with their families and the IPS community, we honor their legacy and continue to mourn their departure.

Despite these tragic losses, we have strived to honor their memory by continuing our work with determination and purpose. As a member of the APS Board Committee and President of IPS, I had the privilege of attending the 3rd African Primatological Society Congress in South Africa in September 2024, alongside fellow IPS members.

The year 2024 was indeed marked by significant challenges, including the preparations for the 30th IPS Congress, which will take place in Antananarivo, Madagascar, in 2025. We are confident that this milestone event will attract wide participation. Additionally, the official Madagascar IPS website is now online, and registration for the congress is already open. A heartfelt congratulations to Trudy Turner and her team, as well as to the committee members Michele Mulholland, Josia Razafindramanana, and Sylviane Volampeno, for their exceptional efforts in making this possible.

I would like to take this opportunity to express my sincere gratitude to all IPS officers who have dedicated their time and energy with unwavering passion for the advancement of IPS and global primatology. My special thanks go to Secretary General Julio César Bicca-Marques for his tireless leadership, and to our ex-officio Past President, Karen Strier, for her continuous support, guidance, and valuable advice. I also commend the election and awards committee for their excellent teamwork in organizing the upcoming IPS election, which will be held in early 2025.

I sincerely thank all those who accepted nominations as candidates for the IPS presidency, as well as the current vice-presidents who are running for re-election, and all new candidates who have stepped forward to stand for election.

Finally, I extend my heartfelt appreciation to everyone who has consistently supported and assisted IPS in its mission. I wish great success for the 30th IPS Congress in Antananarivo, Madagascar, and I hope to see many of you there !

With my best wishes,

Jonah Ratsimbazafy
President, International Primatological Society

VP for Communication

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Greetings Primate People!

We have collated our Officer reports, and an amazing series of updates from our grantees. As grant submission season is coming up I'd like to highlight that range-country nationals have the opportunity to receive a round of pre-submission feedback on applications from the Research committee. Any questions please ask.

In news news the IPS is now on Bluesky. Please follow us there IPS-primatenews.bsky.social (or find us on Insta, Facebook, and X for all your primate news). We also have our Congress website up and running, and there'll be lots more Congress related activities as the year progresses.

There is still time (deadline Feb 10th) to nominate your fellow primatologists for either our Lifetime Achievement Award or the Outstanding Achievement Award.

And finally, please continue to send us all your updates, queries, and primate news and fun.

Hoots and best wishes for a wonderful 2025!

Cat Hobaiter
IPS VP for Communication

VP for Captive Care

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The Welfare and Captive Care committee is up and running. We are 14 members strong, with representation from eight countries and with welfare expertise across prosimian, monkey and ape species. In 2023 we restarted our Captive Care Grants program which supports welfare work in range countries and made two awards. Marina Kenyon of the Endangered Asian Species Trust received a grant for her project, "Critical care outside cage for rescued black-shanked douc (*Pygathrix nigripes*) & silvered langur (*Trachypithecus margarita & germaini)* from the illegal wildlife trade in South Vietnam." And Ana Camila Beltrán Urrego's project, "Assessment of health, gut microbiota, and fecal proteomics in Cotton-to tamarin (*Saguinus Oedipus*) family under human care after fruit reduction and inclusion of gums in the diet" at the Zoo Cali Foundation was also successfully funded. Congratulations to them both!

In 2024 the committee worked with the Officers of IPS to write a letter of support for the passage of the Captive Primate Safety Act (CPSA of 2024) in the United States which would prohibit the possession of primates by private individuals. Later in the year the committee set initial goals including sponsoring a symposium at the IPS Congress, determining whether we can create an IPS award focused on welfare, and developing virtual office hours so IPS members can discuss pressing issues with committee members. We will also investigate offering virtual instruction on animal welfare issues and we will update instructions for our grant program and its review process. Our long-term goal is to revise the IPS "International guidelines for the acquisition, care, and breeding of nonhuman primates" which were last issued in 2007. We will be busy! Please contact me if you have other thoughts about things the Welfare and Captive Care committee might address.

Mollie Bloomsmith
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Secretary General

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Since the report for the 2023 IPS Council Meeting, I have organized meetings, taken minutes, assisted the President, worked on bureaucratic matters of the Society, and corresponded with parties interested in the Society's business. The most important activities are listed below:

• I worked with President Jonah Ratsimbazafy on the agendas for the 2023 Pre-Congress Council Meeting, General Assembly, and Post-Congress Council Meeting.
• I organized the Pre- and Post-Congress Council Meetings and the General Assembly, and took the minutes.
• I discussed with the Treasurer and Vice President for Membership the details of the IPS guidelines for submitting conference bids, which will soon be available online.
• I have updated the Constitution and Bylaws with changes approved by the membership.
• I wrote letters in support of requests of Affiliate Societies approved by the Officers, including the creation of a Colobine Monkey Day proposed by Andie Ang.
• I organized Officers' meetings via Zoom to discuss varied matters.
• I have reminded all members of the IPS Council of the need to keep the Society informed of changes in the officers of their respective Affiliated Societies.
• I contacted La Societe Francophone de Primatologie to ask if they would accept the invitation to hold the 2029 IPS Congress in Strasbourg, France, as approved by the Society. La Societe Francophone de Primatologie declined the invitation. I then sent messages to all European primatological societies, except La Societe Francophone de Primatologie, inviting them to submit a bid to host the 2029 Congress. So far, no European society that has responded has accepted the invitation. The focus on Europe is due to the fact that the last IPS Congress held in Europe was in Edinburgh in 2008. After the upcoming 2025 Congress in Madagascar and the 2027 Congress in China, we will have had four Congresses in Asia, three in the Americas, and two in Africa.

I hope this report finds everyone well and that we are successful in promoting the conservation of all nonhuman primates in 2025, while helping to improve the lives of vulnerable people around the world.

Finally, I am looking forward to an outstanding IPS Congress in Antananarivo, Madagascar. I hope to see everyone either in person or virtually in August.

My best wishes to everyone,
Júlio César

VP for Research

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I am happy to report on behalf of the IPS Research Committee that we have supported 22 exciting projects in the 2023 and 2024 granting cycles. Some of these grantees, as well as those from previous cohorts, will be presenting at the IPS Congress in Madagascar, so please look out for them!

In 2023, we had 38 applications, and awarded a total of $14,970 in 10 grants, all to PhD students. Their projects are listed below, along with the country of their affiliation. * denotes range-state national.

- Silvia Carboni (Canada): "Exploring the Interplay between olfaction and microbes in two sympatric species of wild Peruvian tamarins"
- *Sebastian Garcia-Restrepo (Colombia): "Unraveling the evolutionary relationships of capuchin monkeys in the northern Andean region of South America: use of integrative taxonomy in the study of *Cebus* spp. in Colombia"

- Florence Landry (Canada): "Can infant morbidity and mortality be influenced by infant nutritional development? The case of wild olive baboons (*Papio anubis*)"
- Brynn Lowry (USA): "Integrating field and satellite data for identifying high nutrient patches and inferring primate abundance."
- *Virendra Mathur (Canada): "Spatial cognition in colobine monkeys: movement patterns of Himalayan langur *Semnopithecus schistaceus* in a montane habitat"
- Abigail Morris (USA): "Influence of vegetation composition on male and female gorilla dispersal and population genetic structure in Lac Tele"
- Jenna Owens (USA): "Characterizing directional gene flow of three primate species at The Área de Conservación Guanacaste, Costa Rica"
- *Rina Vololonirina (Madagascar): "Behavioral ecology of black and white ruffed lemur (*Varecia variegata*) population in the Vohibe Forest, Vatomandry, Atsinanana Region"
- Lauren Wiseman-Jones (USA): "The physiological and behavioral responses of Virunga mountain gorillas (*Gorilla beringei)* to social and anthropogenic stressors"
- Eric Wuesthoff (USA): "Exploring the influence of tropical forest regeneration on movement, diet, and seed dispersal in threatened lemurs in Maromizaha Protected Area, Madagascar"

In 2024, we had 37 applications, and awarded a total of $17,994 in 12 grants, including 2 MA students, 8 PhD students and one post-doc. Their projects are listed below, along with the country of their affiliation. * denotes range-state national.

- *Dishari Dasgupta (India): ""Exploring intentional gestural communication in free-ranging
  Hanuman langurs in India"
- Riley Derby (USA): "Social relationships and integration in adolescent mountain gorillas"
- Chloé Gherardi (USA): "You can't have it all: variation in fiber digestibility in wild lemurs and interactions with the gut microbiome"
- *Locadia Dzingwena (South Africa): Reproductive phenology variation of chacma baboons (*Papio ursinus*) in southern African biomes: a camera trap approach
- *Deyatima Ghosh ((India): "Close to humans: personality differences in rhesus macaques in response ot human interactions in a tropical forest in India"
- Kale Hawks (USA): "Variation in age at puberty, social behavior, and dispersal in male olive baboons (*Papio anubis*)"
- Nadine Hussein (Canada): "The nutritional ecology of mantled howler monkeys (*Alouatta palliata*) in Costa Rica"
- Mariana Matos (USA): "Intersecting human and non-human primate space use with the cross-species transmission of gastrointestinal parasites in a human-modified landscape, in Northern Madagascar"
- Kaylie McNeil (USA): "Metabolic adaptation and reproductive outcomes in wild female lowland woolly monkeys"
- Alice Michel (USA): "Long-distance communication in gorillas"
- *Eric Ndayishimiye (UK): "Physiological response to social stressors and the fitness consequences in adult male mountain gorillas (*Gorilla gorilla beringei*)"
- Sydney Self (USA): "Dispersal patterns in a rapidly increasing population of *Colobus vellerosus* living in forest fragments"

I'd like to remind IPS members that the Research Committee offers pre-submission feedback on draft proposals from nationals of range countries. We received 6 such requests in each of the two years and one of the final proposals was funded. If you know applicants who would could benefit from this service, please help us get the word out: we realize that access to mentorship varies considerably, and we have the capacity to offer more people feedback.

Also, many thanks to the Research Committee members that helped review proposals in the last two cycles. They include: Katherine Amato, Andie Ang, Fernando Campos, Rebecca Chancellor, Oscar Chaves, Bert Covert, Laurence Culot, Tony DiFiore, Sofya Dolotovskaya, Nate Dominy, Cedric Girard-Buttoz, Amanda Koerstjens, Andres Link, Suchinda Malaivijitnond, Nga Nguyen, Patrick Onyango, Lilian Cortes Ortiz, Julia Ostner, Adriadna Rangel, Onja Razafindratsima, Julie Teichroeb, Yamato Tsuji, Sarie van Belle, Eva Wikberg, and Shinya Yamamoto.

Finally, keep in mind that the next round of proposals will be due on March 1st 2025, with pre-proposals for pre-submission reads due February 1st, 2025. These are our dates every year. See the IPS web pages for application form, FAQs and sample applications that were successful.

Marina Cords, VP Research

VP for Conservation

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IPS Bulletin-Conservation Committee-Ja2025

We would like to wish you all the best for 2025! These are challenging times for people, nature and our fellow primates around the globe and your work and efforts to converse primates and their environment have never mattered more, but together we can ensure that 2025 is filled with success stories, amazing new research, outputs and initiatives yielding positive impacts and outcomes. Thanks to all for your passion, commitment ad efforts!

This is the perfect occasion for me to also express my deepest gratitude to the IPS Conservation Committee members for their commitment, time and help and previous grantees for their wonderful reports, please keep them coming!

This past year, we received a total of 29 applications for our **IPS Conservation grants**. We are delighted to have been able to award nine grants for a total value of $12,222.28; the following were the successful grantees:

- **Bradley Christin:** Primate density and hunting along a critical ecotone in the northern Republic of the Congo
- **Dessalegn Obsi Gemeda:** How does forest loss and degradation aggravate human-grivet monkey interactions in south-western Ethiopia?
- **Emilee N. Hart:** Validation of a novel field-friendly stress checker to measure fecal glucocorticoid metabolites in northern white-cheeked gibbons (*Nomascus leucogenys*)
- **Giovanna Reichert:** Assessing Natural and Anthropogenic Barriers to Gene Flow of the Critically Endangered Cotton-top Tamarin (*Saguinus oedipus*)
- **Isabela Normando Mascarenhas:** Minimizing risks associated with the reintroduction of the buffy-tufted-ear marmoset (*Callithrix aurita*) in Atlantic Forest fragments in southeastern Brazil
- **Kris Sabbi:** Pests to partners: Leveraging behavioral observation as a conservation tool at the intersection of chacma baboon socioecology and community-led forest restoration in Nature's Valley, South Africa
- **Lucilla Citon:** Surveillance of potentially zoonotic pathogens in black and gold howler monkeys (*Alouatta caraya*) in urban settlements in Northern Argentina
- **Mariana Mota:** Demographic and Ecological Census of Western Lowland Gorilla (Gorilla gorilla gorilla) and Central Chimpanzee (*Pan troglodytes troglodytes*) in Mayombe National Park, Cabinda, Angola
- **Marsya Christyanti Sibarani :** Comparing sampling methods to investigate the effect of habitat fragmentation to small ape populations in Sumatra, Indonesia

We also received three applications for the **Galante Family Winery Conservation Scholarship**. Our 2024 awardee is **Patrick Tusiime**, current Director of the Kibale Forest Schools Program in Uganda, East Africa. The scholarship of $2,500 will allow Patrick to complete his part-time MSc program in One Health at the University of Edinburgh, Scotland. This will equip him to more effectively contribute thereafter to promoting a One Health approach in the conservation of primates, especially great apes, in his home country of Uganda and critically share his new gained skills and knowledge with others. We continue to be immensely grateful to the Galante family for their continued support for this amazing scholarship which provides citizens of primate habitat countries a wonderful opportunity to enhance their training and education in conserving primates.

Major congratulations to this year's grantees; we look forward to receiving your reports which we will showcase in the IPS newsletter.

The IPS conservation committee also finalised last autumn the selection process for the **2025 IPS Pre-congress Training Program (PCTP)** which will take place at the ValBio Center in Madagascar from the 2-9th August 2025. We received a total of 83 well deserving applications. Sixteen participants were selected: 4 from Madagascar, 4 from continental Africa, 4 from Asia and 4 from Central and South America, with 13 countries represented:

- Alvine Dadjo , Cameroon
- Marcel Ngabikwiye, Rwanda
- Rassina Assane Farassi, Mozambique
- Richard Busobozi, Uganda
- Nguyen van Tay, Vietnam
- Robi Kasianus, Indonesia
- Tharuka Murthi, Sri Lanka
- Titir Debnath, India
- Kava Ginot, Madagascar
- Laura Mahatoly, Madagascar

- Njara Raharinoro, Madagascar
- Onjaniaina Mirantsoa Ramilijaona, Madagascar
- Alma Hernández Jaramillo, Columbia
- Anamélia de Souza Jesus, Brazil
- Lucero Hernani Lineros, Bolivia
- Nicolas Gorostiaga, Argentina

Do not forget to submit an application for:

1. ***the new round of IPS conservation grants (Deadline March 1st)***
2. ***the Galante Family Winery Conservation Scholarship (Deadline April 1st)***

REMINDER: We offer feedback on IPS conservation grant proposals on a case-by-case basis from nationals of primate range-state countries. For this purpose, applications must be submitted by the **1st February** for feedback; revised submissions must be resubmitted by the March 1st deadline.

Yours sincerely,
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VP for Education

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The Education Committee of IPS awards the Lawrence Jacobsen Education grant of up to $1,500 to support the development of primate conservation education programs. In addition, we award the Charles Southwick Conservation Education Commitment Award, in the amount of $2,000: $1,500 to the recipient and $500 given in the recipient's name to a project of their choosing in their community.

Lawrence Jacobsen Education grant

I am very grateful to the Education Committee members who helped review and judge the applications in 2024: Adrian Barnett, Alejandra Duarte, Carla Castro, Francine Dolins, Inza Kone, Maria Joana Ferreira da Silva, Luciana Oklander, Martin Kowalewsky, Mewa Singh, Misato Haiashi, Rachel Ikemeh Ashegbofe, Simplicious Gessa, Suchinda Malaivijitnond, Valentina Truppa, and Zarin Machanda.

Six reviewers reviewed and scored each grant out of 35. I standardized the scores and ranked applications based on their mean standardized score. Reviewers were asked to provide comments to help applicants in improving their future applications. Four applicants used the opportunity of pre-application to get feedback on their proposal before the deadline in 2024.

We received 10 applications from 3 countries across Latin America (Brazil [3 applications], Peru [2 applications], and Mexico), two African countries (Cameroon and Uganda) and two Asian Countries (Sri Lanka and Nepal). We awarded a total of US$ 6,000 across three grants, all of which included Community Conservation Initiatives in their applications.

2024 Awardees:

Emily Otali. Project: *Inspiring Future Ugandan Primate Conservationists through Career Days.* Country: Uganda

Gabriela Rezende. Project: *Teachers as sowers of the conservation of the black lion tamarin in southeast Brazil.* Country: Brazil

Bishwanath Rijal. Project: *A Conservation Education and Community Outreach Program for Safeguarding Primates and Promoting Human Health in Myagdi District, Nepal*. Country: Nepal

Charles Southwick Conservation Education Commitment Award

The Education Committee members also helped with evaluating the nomination for the Charles Southwick Conservation Education Commitment Award in 2024. We awarded Mr. Muhammad Nur US $2,000: $1500 given directly to him and $500 given in his name to a project of his choosing in his community.

The Education committee is getting ready to review the applications for the Lawrence Jacobsen Education Development Grants 2025. Details are available at the IPS website where you can find examples of past successful applications. Remember, we provide feedback on proposals from nationals of range-state countries. Applications must be submitted by February 1st for feedback, with revised submissions due by March 1st. Address questions and completed applications to me.

In addition, we are preparing ourselves for the student paper and poster competition for the 2025 meeting in Antanarivo. All students planning to attend the next Congress are encouraged to participate and to indicate their desire to participate through the abstract submission process. Within the IPS aim of increasing geographical diversity and equality we created the "Global South Student Award". Students eligible for this competition must both be a resident of and conducting research in a Global South country. Student members of IPS from the Global South are also eligible to enter the general Student Competition. We ask the student to indicate which competitions they want to enter during the abstract submission. In addition, we are preparing to offer mentorship on how to deliver oral and poster presentations for students of Global South that were pre-selected for the students' competition. Students can indicate their interest in mentorship on these tasks during the registration process.
We will follow the format used in the previous IPS meetings. We will select 10-20 finalists based on abstracts in advance of the Congress. Then, at the Congress, each competition judge will be able to review and directly compare all of the finalists. Check the Congress website for more details.

Thank you!

If any members are interested in serving on the Education Committee, as a student competition judge, a mentor, or have specific issues they would like addressed, please contact me at patrizar@usp.br

I look forward to seeing you all in Madagascar!

Patrícia Izar
VP for Education

Trea$ury Note$

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Happy New Year! 2025 promises to be an exciting year for IPS. Registration is open for our Congress in Madagascar and proposals for symposia and workshops are in. Abstracts are due by February 15. Please remember to update your membership before you register for the meetings so that you can take advantage of member rates for registration.

Last November a notice was sent to everyone on the IPS mailing list informing them of changes to our membership dues rate model. To ensure both equity and the financial health of the organization, we have moved to a model that uses the World Bank country income categories when determining dues. The model was introduced at the 2023 business meeting in Kuching and went into full effect in December, 2024. When you enter your country of residence, you will be able to choose your status—professional/faculty, contingent faculty/postdoc or student. We will be adding a retired category in the near future. The fee structure is as follows:

Country Category	Professional/ Faculty
High Income	$100
Upper Middle	$50
Other	$10

IPS has an extremely active grants program and members receive free access to the International Journal of Primatology. Your support of IPS continues to allow this to happen.

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We are also committed to never letting financial considerations be a barrier to membership. If you cannot afford the dues, please write to me.

This year, IPS has been the recipient of two significant donations. These donations have set up funds for grant support. The first was from the family and friends of Columbian primatologist, Sebastian Ramirez Amaya who died in a tragic accident while conducting field work for his PhD dissertation. Details of award eligibility can be found on the IPS website. The first award recipient was announced this year. The second major donation was from ReWild. We are particularly grateful to Russ Mittermeier who was instrumental in obtaining these funds. The funds have been used to establish the Masters and Genin African Primatology Fund. The fund honors the memories of Judith Masters and Fabien Genin who were tragically killed in South Africa. The fund is specifically designated to help African and Malagasy students and is currently accepting donations.

IPS has also received donations to support both Malagasy and African students attend the meetings in Madagascar. If you would like to receive information about eligibility for support, please get in touch with me before you register for the congress.

Looking forward to seeing you all in Madagascar.

Trudy Turner
Treasurer and VP for Membership
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(Please note this is a temporary email for Jan-Feb 2025, we'll be sharing a new one soon.)

IPS Elections Committee Report

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IPS Elections Committee Report—10 January 2025

At the request of the IPS President, Jonah Razimbafazy, I convened and coordinated the 2024-2025 IPS Elections Committee. The 2024-2025 Election Committee members (in alphabetical order) are: Rachel Ashegbofe, Nancy Caine, Marie Charpentier, Misato Hayashi, Jonah Ratzimbafazy (co-chair), Johannes Refisch, June Rubis, Christoph Schwitzer, Eleonore Setz, and Karen Strier (co-chair).

Briefly, our process followed protocol and precedent. A call for nominations was sent to all IPS members via email, with a December 20, 2024 deadline. Open positions were: President (replacing Johan Razimbafazy after completion of one non-renewable term), Vice President for Communication (replacing Cat Hobaiter after completion of her second and final term), Vice President for Education (replacing Pat Izar after completion of her second and final term), and Vice President for Student Affairs (a new position).

We received eight nominations for President, three nominations for VP for Communication, seven nominations for VP for Education, and four nominations for VP for Student Affairs. The Elections Committee members independently ranked all nominations, and I collated their ranks anonymously by summing the ranks and by tallying the percentage of Committee members who had included each nominee among their top ranks. The results of these two methods were the same. The Elections Committee then unanimously approved the final rankings and authorized me to contact the top-ranked nominees for each office, with the goal of obtaining a slate of two candidates per position.
Once the full slate was confirmed, the IPS officers unanimously endorsed the ballot, and we are now in the process of soliciting the candidate statements that will appear on the ballot sent to IPS members for voting.

The ballot will also include two continuing positions that require endorsement, one for current Vice President for Ethics, Diversity, Equity & Inclusion, to serve a second and final term, and the other for the current appointed Vice President for Scientific Programming, to serve a first elected term.

On behalf of the Elections Committee, I thank everyone who submitted nominations and all of the nominees who have agreed to run for office. I also thank the Elections Committee members for their valuable service.

Karen B. Strier, IPS Past President and co-chair, Elections Committee

IPS Captive Care Grant Report

Ana Camila Beltrán Urrego & Alessandra Melchart

Assessment of health and gut microbiota in *Saguinus oedipus* under human care after the reduction of fruits and inclusion of gums in the diet

Ana Camila Beltrán Urrego[1], Alessandra Melchert[2]

[1]Graduate Wildlife Program, São Paulo State University (UNESP)
[2] Department of Veterinary Clinics, School of Veterinary Medicine and Animal Science,Sao Paulo State University, Botucatu, Brazil

1 INTRODUCTION

The Cotton-top tamarins (*Saguinus oedipus*) are currenty classified by the IUCN as critically endangered. This species has a limited geographic distribution, being found only in the dry tropical forests of Colombia [1] One of the critical aspects to be improved in captive conditions is nutritional management, which is considered one of the key domains of animal welfare [2].

Captive diets for this species are generally characterized by high levels of fruit and sugars, low fiber content, and limited inclusion of insects and gums, which are important components of their natural diet [3]. This imbalance is closely linked to the high incidence of gastrointestinal problems such as chronic diarrhea, chronic enteritis, colon adenocarcinoma [4,5] and subsequent alterations in gut microbiota [6]. Based on these findings, it has been suggested that the feeding model for primates should shift toward a fruit-free diet with higher inclusions of fibrous elements such as vegetables, and others like gums, insects, and commercial primate-food [7].

However, few studies have integrated microbiome assessment with health parameters in fruit-free diet models, which is necessary to generate objective recommendations for dietary adjustments specific to this species.

2 PROJECT OBJECTIVES

Specific Objectives

- Assessing the health and nutritional status of animals through physical and chemical parameters before and after the dietary change.
- Determining the abundance and composition of the gut microbiota before and after the dietary transition.
- Offering baselines for transitioning Cotton-top tamarins to a low-fruit diet according with health results.

3 BRIEF OVERVIEW OF STUDY LOCATION AND METHODS

The study was conducted at the Cali Zoo (FZC) facilities, located in the Department of Valle del Cauca, in southwestern Colombia (Figure 1). It was authorized by the Ethics Committee of FMVZ – UNESP – Botucatu under Protocol CEUA 0384/2023 and approved by the Research Coordinator of the Cali Zoological Foundation in accordance with Colombian law.

Fourteen Cotton-top tamarins were evaluated (7 males and 7 females). The target diet formulation was based on the recommendations outlined by the EAZA [8]. A 5-week transition period was implemented to adapt the diet from a high-fruit diet (51% on an as-fed basis), to a low-fruit diet (10%), rich in vegetables (50%), and supplemented with gum, primate mix, and insects. Both the initial and final diets were evaluated through a bromatological analysis.

Figure 1. Illustrative location of Cali, Colombia

The animals were evaluated before, and after 4 months of receiving the new diet. The following data were collected: biweekly body weight, weekly fecal score and food consumption; physical examination data, including body condition score, coat quality, oral health, and gastrointestinal tract assessment, before and at the end of the study; hematology, serum biochemistry, abdominal ultrasound, behavioral evaluation, and gut microbiota composition along with potential metabolite production, before and at the end of the study.

4 KEY RESULTS/MAIN FINDINGS

Sugar intake was significantly lower in the final diet (p = 0.001). Also, significant increases were seen in the estimated intake of fibre and NDF (p = 0.02) which was linked to the higher proportion of vegetables. These changes in nutrient intake were related to the results seen in faecal scores, which showed a clear improvement, shifting from loose or watery stools (Score 4-6) to well-formed and consistent stools (Score 2-4) in all evaluated groups, starting from week 3 of the dietary transition.

In general, the animals maintained optimal health, with all hematological and biochemical values remaining within normal ranges during both evaluation moments. However, improvements in coat quality were noted, correlating with significant increases in average body weight, particularly in females (p = 0.01), as well as improvements in body condition score in 78% of the evaluated animals. Animals that remained in suboptimal body condition were elderly individuals with severe oral health issues, which compromised their chewing ability, making this a highly relevant factor for nutritional management planning.

Regarding the gut microbiome, there was a marked increase in species diversity in the samples collected under the low-fruit diet. Before the diet transition, the most prevalent genera were *Bifidobacterium*, *Streptococcus*, *Prevotella*, and *Blautia*. After dietary adaptation, the most abundant genera were *Bifidobacterium*, *Prevotella*, *Blautia*, *Holdemanella*, and *Lactobacillus*. Both, before diet change and after diet change analyses highlighted the dominant presence of the species *Bifidobacterium longum*, *Bifidobacterium adolescentis*, *Prevotella copri*, and *Blautia_A wexlerae*. An increase in the potential production of acetate, lactate, propionate, and butyrate was detected, which was linked to the increased presence of bacteria species capable of utilizing and fermenting fiber. There was also a general increase in the potential production of B-complex vitamins (B2, B7, B9, and B12) and vitamin K, which may offer significant health benefits to the individuals.

Additionally, we successfully developed educational materials for children focused on the species, which were shared with the Fundación Proyecto Tití in Colombia for use in their teaching programs within rural communities.

5 DISCUSSION OF RESULTS

Our findings suggest that a dietary regimen characterized by reduced sugar content and increased fiber intake may be optimal for the management of Cotton-Top tamarins under human care as dietary modification demonstrated significant health and nutritional benefits, which could potentially contribute to the sustainable maintenance of populations in conservation programs.

Furthermore, the study identified the most prevalent bacterial phyla and genera within the gut microbiota, including Bifidobacteria, which could serve as potential probiotics. These microbes may be beneficial for incorporation into comprehensive nutritional management strategies or for use in gut microbiota modulation in ex-situ settings. Nevertheless, ongoing, rigorous monitoring is required, along with further exploration into the functional roles of the intestinal microbiome.

Additionally, it is essential to complement this process with educational initiatives, as one of the key challenges identified was shifting the prevalent association between primates and fruit consumption among zoo staff and zoo visitors.

6 ACKNOWLEDGEMENTS

We thank the FZC for providing their staff and facilities, the institution's veterinary team, and the diet preparation staff. We also extend our deepest thanks to the financial support from IPS and the CAPES organization, who helped make this project a reality.

7 REFERENCE LIST

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IPS Captive Care Grant Report

Marina Kenyon

"Critical care outside cage for rescued black-shanked douc (*Pygathrix nigripes*) & silvered langur (*Trachypithecus margarita* & *germaini*) from the illegal wildlife trade in South Vietnam"

Dr Marina Kenyon.

Endangered Asian Species Trust

Captive care

1 BACKGROUND/INTRODUCTION

The Endangered Asian Species Trust works with the Vietnamese Government tin supporting law enforcement in South Vietnam, by founding the Dao Tien Endangered Primate Species Centre, in Cat Tien National Park. The specialist rescue centre receives confiscated endangered primates from the Forestry Protection Department. The centre specialises in Golden-cheeked gibbon (*Nomascus gabriellae*), Black-shanked douc (*Pygathrix nigripes*), pygmy loris (*Xanthonycticebus pygmaeus*) and Silvered langurs (*Trachypithecus spp*.).
The centre provides an International Standard of care for Vietnam and works hard to develop reintroduction protocols for primates fit for release. Additionally EAST undertakes invaluable education awareness in the buffer zone of Cat Tien National Park, Nationally and Internationally.

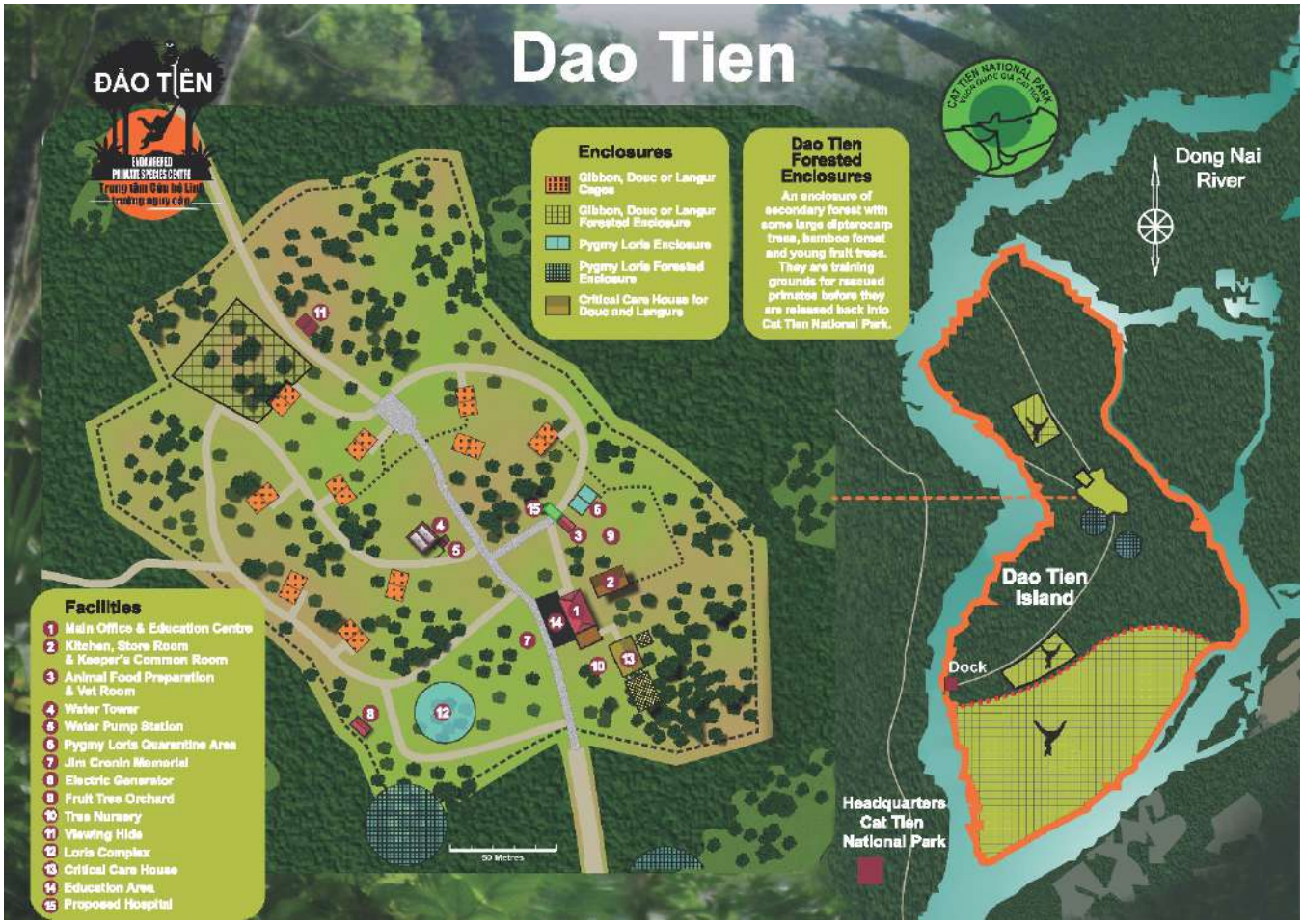
2 PROJECT OBJECTIVES/AIMS

- Build an outside caged enclosure for infant douc and silvered langurs receiving critical care after rescue from the illegal wildlife trade.
- Although the infants are taken into the trees by care givers, the outside cage means during times when they are not with carers, they have access to an enclosure for sun, view of trees and also view of the larger enclosure with older individuals, to enable recognition of their species and their wild species specific behaviours.
- Providing stimulus of the forest and other conspecifics is vital for survival of these sensitive species.

Rescued black-shanked douc from the illegal wildlife trade

3 BRIEF OVERVIEW OF STUDY LOCATION AND METHODS IF/AS RELEVANT

The Dao Tien Endangered Primate Species Centre (DTEPSC), located in Cat Tien National Park, 180 km north east from Ho Chi Minh City, in South Vietnam. The centre was founded under Decision No. 2312/QDBNN-PTNT (09/07/2008) and continued with Decision No. 1220/QD-BNN-HTQT (03/23/2021) from the Ministry Agriculture Rural Development (MARD). In collaboration with Cat Tien National Park, Dao Tien receives confiscated endangered primates from the illegal wildlife trade. Individuals are health checked and socialised with conspecifics. Candidates suitable for release are rehabilitated in forested enclosures ready for release back to the wild.



- KEY RESULTS/MAIN FINDINGS OR OUTCOMES

- In November 2024 the small caged outside area was successfully built, linked by a mesh tunnel to the intensive care bedrooms for the infant douc and silvered langurs being hand reared.

The new cage for hand reared infants

- The enclosure is made of 1 inch mesh, with a concrete edge, limiting predator access. A safety porch has been built to ensure safety.
- The enclosure is linked to the house via a mesh tunnel, with a slide, to ensure the infants are safe inside the house during the night.
- The cage is on the southern side of the house for extra sunlight and more warming sun. Also away from any other animal or human disturbance. A private place, overlooking the larger primate outside cage.

4 IMPLICATIONS OF PROJECT/DISCUSSION OF RESULTS

- This enclosure is key for sensitive douc and langur survival. Douc are one of the most sensitive and difficult primates to care for in the world. They are mentally sensitive and are known to survive better when close to conspecifics. This house enables infants too small to enter with larger individuals, be able to see them when they are in their outside enclosure.

- The cage is south facing for morning warm sun, also looking onto trees and bamboo. For infants to see the forest, something familiar, they are more likely to survive. If kept inside a critical care house all the time, more likely to lose the will to live.

- The new enclosure is planted with native douc trees, enabling access to leaf, bark and soil, all key to douc survival.

- The outside enclosure provides extra stimulus for infants, infant douc in shock post rescue from the illegal wildlife trade, if left unstimulated they will slowly fade and die.

- The house is connected via a short closed walkway to the managers accommodation, allowing safe and secure 24hr care for infants when they need it.

5 ACKNOWLEDGEMENTS

Thank you to the staff of Dao Tien for preparing the area for the build, planting the enclosure and of course their continued support of every rescued primate in need. Thank you to Cat Tien National Park for continued collaboration and support for this project. Thank you to IPS for allowing us to build this space to help care for infant douc and langurs in South Vietnam.

IPS Research Grant Report

Jack L. Richardson

SEX DIFFERENCES AND THE ROLE OF SOCIAL PLAY IN DEVELOPMENT OF WILD MOUNTAIN GORILLAS.

INTRODUCTION: Play is a developmentally plastic behaviour, performed as an affiliative interaction between individuals, without a clear direct short-term function[1]. As play may be costly, multiple hypotheses have been discussed to explain its adaptive benefit. The 'motor training hypothesis'[2] links play to the improvement of motor skills, whereas the 'social relationship hypothesis'[3] posits that play is a platform to practise behaviours and learn how to interact with other individuals. Developmental milestones to measure these outcomes are defined as "a set of behaviours, skills or abilities that are demonstrated by specified ages during infancy and early childhood in typical development"[4]. Reaching such milestones earlier shows correlation with increased time spent playing, for example documentation of younger ages of attainment of motor skills (independent travelling, carrying position), behavioural metrics (spatial independence and mating) and increased amount of cerebellar synapses[5,6].
Mountain gorillas, have an accelerated life history compared to other African apes (e.g. early age of first birth), demonstrate the highest levels of sexual dimorphism and exhibit a polygynous social structure. The social structure of gorillas drives male-male competition, which selects for the development of larger sizes and physical competition in males. Male and female primates diverge in their reproductive strategies, and as such their life-histories and developmental priorities are different[7]. Primates show differences in frequencies of play between the sexes, typically with males playing more than females. Given these physical differences and variation in male and female reproductive strategies, we expected sexual differences in the function of play should be particularly salient in mountain gorilla populations.

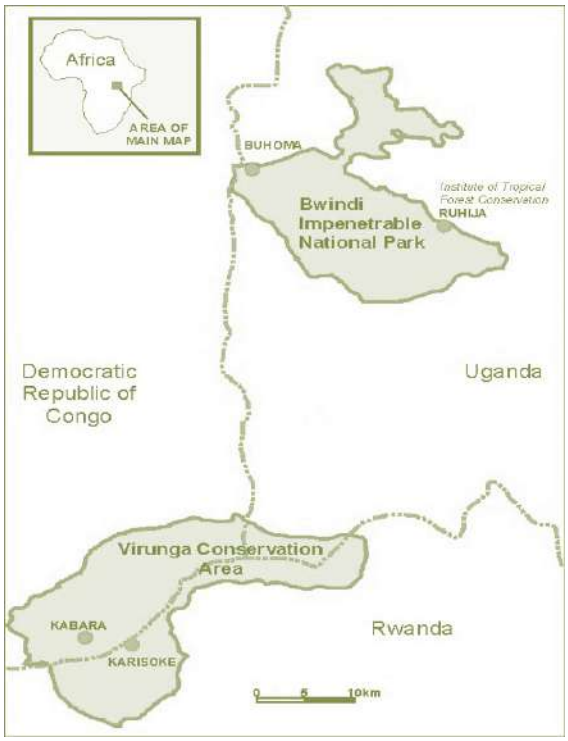


Figure 1: Map shows the two populations of mountain gorillas, Bwindi Impenetrable National Park and the Virunga Conservation Area. This study was performed in Ruhija in Bwindi.

PROJECT AIMS: This research was the first to undertake a comprehensive study of play in Bwindi mountain gorillas. This research used long-term behavioural data to analyse sex differences in play rates and measure correlations between rates of play and attainment of motor and social developmental milestones. The two specific aims were: **1**) Understand the differences between male and female play rates. **Prediction**: Male gorillas will play more than female gorillas. **2**) Test if play promotes the acquisition of motor and social skills. **Prediction**: Increase in time spent playing will be associated with earlier age at attainment of physical/motor (proximity, physical) and social (aggression, affiliation) developmental milestones.

STUDY LOCATION AND METHODS: This study was conducted on five habituated mountain gorilla (*Gorilla beringei beringei)* groups in eastern sector (Ruhija) of Bwindi Impenetrable National

Park, Uganda from 2005 until 2023 (Figure 1). A total of 33 immature individuals between the age of 0.59 (~7 months) and 8 years were included. Female mountain gorillas are categorised as adults from 8 years onwards.

Behavioural Data Collection: Focal animal sampling was conducted totalling 6,278 hours of focal time. We then calculated monthly proportion of time spent in play for each individual, for a total of 1,115 monthly data points. Which were analysed using mixed models to examine the effect of sex and age on monthly play rates.

Developmental Milestones: A total of 31 developmental milestones were extracted from the behavioural data. All milestones were categorised according to six categories: Affiliative, Agonistic, Carry, Mating, Physical, and Proximity. To analyse the relationship between developmental milestones and play rate, we used generalized linear mixed models to test correlations between variation from average play rates and variation from average milestones attainment age. These analysis were performed on all the milestones together and each category separately.

Ethical note: Data used as part of this study were collected non-invasively from nonhuman primates. Permission was granted for the study by Uganda Wildlife Authority and Uganda National Council of Science and Technology, and we adhered to all protocols and guidelines set by these institutions.

RESULTS: Aim 1 play rates*:* We found that males play more than females, specifically between the ages of 2-6 years (Figure 2). This was consistent with our prediction that males would need to play more to develop motor and social skills, because they exhibit more physical competition and sustain more social affiliative relationships than females.

Aim 2 play and developmental milestones*:* We found support for the prediction that increased play rates would correlate with earlier attainment of developmental milestones. This relationship was stronger for females than for males. Additionally, when we analysed different types of developmental metrics separately (affiliative, agonistic, physical, proximity) the correlation between play and developmental measures was only significant for agonistic and proximity milestones. For agonistic milestones females had a stronger relationship between increased play and earlier age of

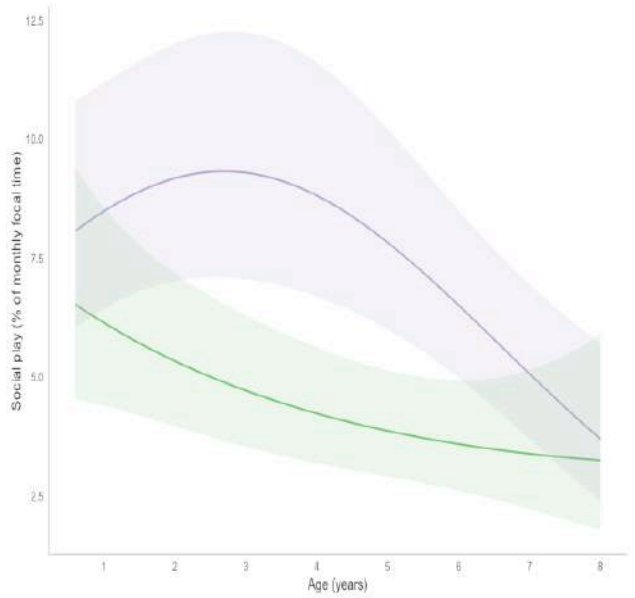


Figure 3: The effect of age and sex on social play rates, immature gorillas from ages 0.59-8 years. Shaded areas =

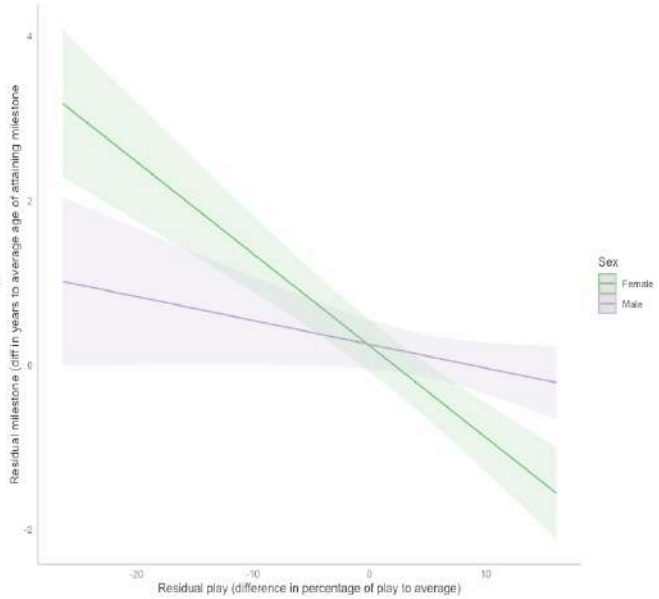


Figure 3: Relationship between residual play and sex with residual milestone age.

attainment than males, but for proximity

milestones there were no sex differences. Our analyses cannot suggest a direction of causation in our results and thus we interpreted our findings to mean that play rate ontogenies and other developmental metrics are likely linked, with individuals who play more than average for their age also reaching other developmental milestones at earlier ages.

DISCUSSION: Overall, we accomplished two major goals. 1) We presented data on social play for the Bwindi mountain gorilla population for the first time. 2) We provided a framework to measure development benefits of play and provided a better understanding of which developmental milestones can be used to support which hypotheses.

Our study provided support for both hypotheses tested (motor training hypothesis and social relationship hypothesis). Therefore, we conclude that play can be a platform for the development of both motor and social skills. We found large sex differences in play rates, in Bwindi mountain gorillas, with males playing more than females, particularly during the juvenile period (3-6 years) confirming our prediction that social play behaviour among immatures may be more relevant for male skills development than females. As such, play may serve as a platform for the behavioural and morphological dimorphism present in adult gorillas. Additionally, despite males typically playing more than females we found a stronger correlation between age of attainment of developmental milestones with rate of play in females than males. This suggests that females may be building skills sooner, particularly when they are playing more. This could also be an effect of females having a faster pace of development than males, as they disperse from their natal groups and reach reproductive age sooner. Therefore, they may need to build certain physical and social skills sooner than males.

Whilst our framework can be used for other species, it should not be assumed that social play provides the same benefits and costs as we demonstrate in gorillas. It is necessary to contextualise predictions of this framework based on the behavioural and socio-ecological variation characteristics of each study population. For example, we were less successful at differentiating between motor and social hypothesis of play, but this may be because of the social structure in mountain gorillas and predictions that both motor and social skills are more important for males. As we study social play more comprehensively in a larger range of animal species, we may come closer to understanding the functional benefits of play and why it has been conserved across the tree of life. This hypothesis driven framework made progress towards delineating patterns of play that relate to different evolutionary hypotheses. Our use of a wild population with long-term data allowed a more accurate understanding of how socio-ecological variation in a primate's habitat can affect social play behaviour.

ACKNOWLEDGEMENTS: We would like to thank the Ugandan Wildlife Authority and Ugandan National Council for Science and Technology for the permission to conduct this research in Bwindi Impenetrable National Park and for support in data collection. Funding for the data collection used in this research was provided by Max Planck Society, the United States Fish and Wildlife Service Great Ape Fund, Berggorilla Regenwald Direkthilfe, National Science Foundation (BCS 1753651), International Primatological Society, The Leakey Foundation and graduate fellowship support from The George Washington University's Columbian College. We thank Shannon McFarlin, Martha Robbins, Rachel S. Nelson, Andrew M. Robbins, Patrick J. Tkaczynski, Aaron A. Sandel and Chris Young, Alison Fletcher, Elizabeth Lonsdorf, and Francys Subiaul for help and advice on the writing and analysis. We greatly appreciate the many people that have worked at the Bwindi gorilla project since 1998, that have contributed to the long-term data collection. For a full list of people that have worked at the project see: (https://www.eva.mpg.de/primate-behavior-and-evolution/research-groups/gorilla-group/).

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IPS Research Grant Report

Tabor Whitney

Associations between ecological attributes and the gut microbiome of mantled howlers in Los Tuxtlas, Mexico

Tabor Whitney, Northwestern University

1 BACKGROUND/INTRODUCTION

The continuous conversion of natural habitats to agricultural fields and pasturelands creates fragments that isolate animals, forcing them to adjust to new environments with different resources. Studying the physiological responses of primates to environmental change is central to our understanding of primate ecology and evolution. Recently, gut microbial analysis has emerged as a new tool for examining primate responses to anthropogenic habitat change. While research has highlighted that habitat quality has impacted the gut microbiome (GM) [1–3], few have considered which aspects of habitat quality are associated with changes in the GM.

The endangered Mexican mantled howler monkey (*Alouatta palliata mexicana*) population of Los Tuxtlas, Mexico, has lost most of its original habitat. The Los Tuxtlas region is an anthropic matrix of forest remnants immersed between cities, pasture lands, and agriculture. The small fragments that comprise the region are some of the only remaining habitats for numerous plant and animal species, some unique to the area. It is projected that 14% of the current forest cover in the region will be gone by 2025 [4]. This rapid conversion of natural habitat forces howler monkeys to live in small forest fragments with different resources, and as a result, their GMs have shifted. Our results confirm howler monkey physiology varies with anthropogenic disturbance, and we determine which ecological attributes should be prioritized for future mitigation strategies.

2 PROJECT OBJECTIVES/AIMS

Aim: Which ecological attributes are associated with GM diversity and composition for the mantled howler monkey?

Objective: Anthropogenic pressures and seasonal variations create disturbances across fragmented landscapes, resulting in a mosaic of habitats with varying levels of viability for wild primate social groups. To support the long-term survival of threatened species, it is crucial to quantify the ecological attributes of habitat fragmentation and incorporate physiological responses to guide conservation strategies. Using the varying levels of habitat degradation across forest fragments in Los Tuxtlas, we assess key ecological attributes to identify the specific aspects of degradation that elicit physiological responses in a wild primate population. Additionally, the findings of this study identify high-priority forest fragments for mitigation efforts and highlight the ecological attributes that must be targeted for effective conservation. Reassessing GMs over time will enable monitoring of the efficacy of restoration and mitigation efforts on mantled howler monkey subpopulations.

3. BRIEF OVERVIEW OF STUDY LOCATION AND METHODS

Study Site: The Los Tuxtlas region is one of the last remnants of tropical forest on the coastal plain in the Gulf of Mexico. The region covers 155,122ha, and the small riparian forest fragments comprise 9% of the reserve's area and are the second-largest forest category after tropical evergreen forests [4]. These small areas are crucial for biodiversity conversation as they serve as biological and genetic corridors connecting different forest patches and are some of the only remaining habitats for numerous plant and animal species, some of which are unique to the area [5].

Figure 1: Map of study sites and the ten geographically distinct forest fragments.

Sample Collection: We collected fecal samples from 77 adult male and female mantled howler monkeys across ten distinct forest fragments.

Ethics/Permissions: Each sample was collected non-invasively. We always maintained a 6ft distance, and Northwestern's IACUC permitted us to conduct research without a permit.

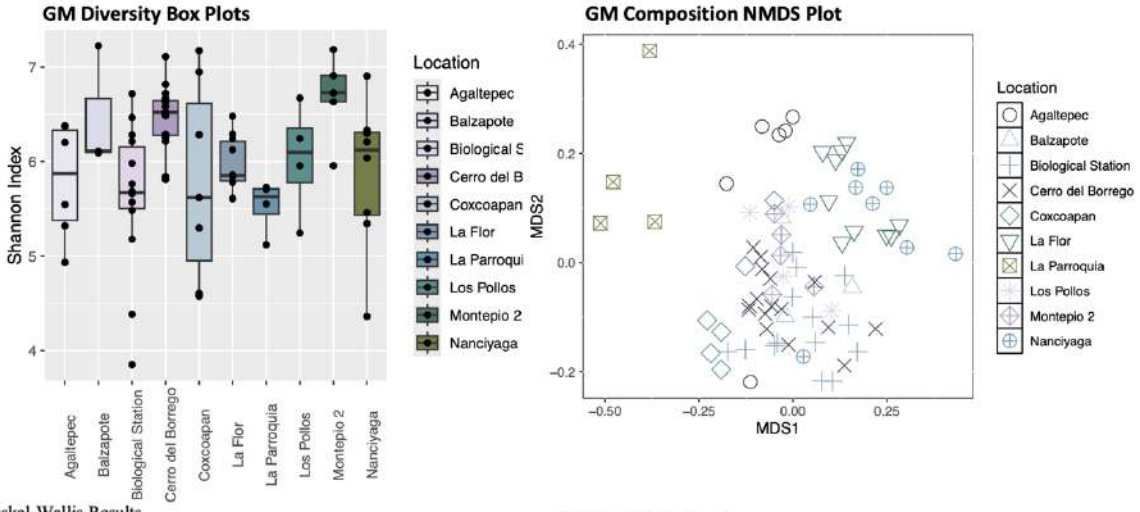
Ecological Attribute Quantification: We set ten vegetation transects in each fragment, and the ecological attributes measured include area, shape, degree of isolation, number of tree lines, proximity to human settlements, mean DBH, basal area, and tree species diversity.

GM Quantification: We amplified the V4-V5 region of the 16S rRNA gene using the primers 515F/926R [6]. We sequenced the amplicons using the Illumina MiSeq V3 platform and assembled them using QIIME2 [7–9].

*Statistical Analyses***:** To examine the influence of ecological attributes on GM diversity, we followed an information-theoretical approach and multi-model inference to generate generalized linear models (GLMs) [10]. We then calculated the relative importance of each predictor based on its Akaike weight in all models. To examine the influence of ecological attributes on GM composition, we used a permutational multivariate analysis of variance (PERMANOVA) with the *adonis2* function in R using the *vegan* package [11].

4 KEY RESULTS/MAIN FINDINGS OR OUTCOMES

GM diversity and composition significantly differ between the ten forest fragments (*Figure 2*). For which ecological attributes are associated with this difference, we found that proximity to human settlements is the only significant factor predicting GM diversity when accounting for all other ecological attributes. GM diversity and human proximity have a positive relationship (cor = 0.34, p = 0.002). Based on PERMANOVAs, the number of treelines, degree of isolation, and basal area of tree species (specifically tree species that were fruiting at the time of surveying) predict GM composition.



Kruskal-Wallis Results

Alpha-Diversity Metric	Factors	eta2[H]	P-Value
Shannon Index	Location	0.35	0.02*

PERMANOVA Results

Beta-Diversity Metric	Factor	R2	P-Value
Bray-Curtis	Location	0.21	0.03*

Figure 2: (A) There is variation in GM diversity between the ten fragment locations (p =0.02). (B) This NMDS plot highlights the clustering of howler monkeys by fragment location, indicating that those residing in the same forest have GMs that are more similar than those in other fragments (p=0.03).

5 IMPLICATIONS OF PROJECT/DISCUSSION OF RESULTS

This study shows that the endangered Mexican mantled howler monkeys' physiology shifts across forest fragments in response to ecological attributes and proximity to human settlements. Identifying which attributes drive these changes helps prioritize forest fragments and social groups for future mitigation strategies. Our findings suggest that monkeys in fragments with fewer tree lines, higher isolation, fewer fruiting food species, and closer proximity to humans experience shifts in their GM. Mitigation efforts could focus on restoring habitat corridors by planting tree lines to connect small fragments to larger forests, improving access to resources, and reducing human disturbance. While mitigation efforts and conservation action plans are in the early stages of development, our project demonstrates the power of using the GM as a non-invasive biomarker. We argue that non-invasive biomarkers are underutilized and demonstrate that their use, paired with habitat surveys, provides a quick and cost-effective way to measure the welfare of endangered species to inform conservation and management actions.

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IPS Research Grant Report

Sebastián García-Restrepo

Unraveling the evolutionary relationships of capuchin monkeys in the northern Andean region of South America: Use of integrative taxonomy in the study of *Cebus* spp. in Colombia

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1. BACKGROUND/INTRODUCTION

Integrative taxonomy considers that conceptual advances in disciplines related to the origin and evolution of species can contribute to the resolution of taxonomic uncertainties at the species level by integrating the available systematic information using a holistic approach (Schlick-Steiner et al. 2010). Its approach can be summarized in three points: a) Morphological methods fail in some cases, requiring the application of other approaches; b) Even when morphology can be successful in delimiting species, other approaches can significantly help and speed up the process; and c) The use of several disciplines helps taxonomy to go beyond the naming of species, to understand the processes that cause them (Schlick-Steiner et al. 2010). *Cebus* spp. are widely distributed primates in Colombia with five to six species currently recognized. They have been subject to multiple taxonomic changes based mainly on geographic distribution and morphological characteristics such as craniodental differences and fur coloration (García-Restrepo and Montilla 2021). However, the systematic, nomenclature, and historical geographical distribution of the taxa occurring in the northern Andes region continue to be subject to discussion (García-Restrepo and Montilla 2021). Phylogenetic analyses have suggested that *C. versicolor* and *C. cesarae* are morphologically and genetically distinct (Lima et al. 2017). Nevertheless, few samples from the Magdalena Valley and the Caribbean region have been included. In addition, *versicolor-cesarae* geographical limits are unclear, and detailed studies on their morphological distinction are also needed. Concerning *C. malitiosus* its monophyly and phylogenetic position remain unresolved (Boubli et al. 2012). Regarding *C. leucocephalus*, its distribution range in Colombia remains unclear. The validity of the subspecies of *C. capucinus* (*C. c. capucinus* and *C. c. curtus*) has been debated as the large variability of characters within the group makes the subspecific designation difficult (Hershkovitz 1949). Several authors have considered *C. capucinus* as a monotypic species due to the lack of evidence of morphological and genetic differentiation of the monkeys of Gorgona Island compared to continental locations (García-Restrepo and Montilla 2021).

Research on the number of species and subspecies, their distribution, as well as *Cebus* spp. morphological and genetic diversity, would help to prioritize conservation projects at national and international scales. Furthermore, it highlights the importance of integrating methods from different disciplines in primate systematics research in the region. Currently, our international research group is conducting studies of *Cebus* in museum collections in Colombia, Venezuela, and the United States to better understand the *Cebus* radiation in this region.

2. PROJECT OBJECTIVES/AIMS

The objectives of this study were to evaluate the systematics, ecomorphology, and biogeography of *Cebus* spp. in the northern Andes region based on morphometric analysis of the skull and mandible, phylogeographic analysis, and phylogenetic inferences from genomic data.

3. BRIEF OVERVIEW OF STUDY LOCATION

IPS funding covered part of my travel expenses to visit four museums during my doctoral research internship in the United States between August and October 2023: American Museum of Natural History, Field Museum of Natural History, Smithsonian National Museum of Natural History, and Natural History Museum of Los Angeles County. During these visits, I examined specimens from eight countries (Honduras, Nicaragua, Costa Rica, Panamá, Colombia, Venezuela, Ecuador, and Perú) to collect data and photographs for morphometric analysis and skin and tissue samples from specimens for genetic analysis. The morphometric data were processed at the Universidad de los Andes in Bogotá (Colombia), and the skin and tissue samples will be processed at UCLA (USA). No approval of research ethics committees was required to accomplish the goals of this study.

4. KEY RESULTS/MAIN FINDINGS OR OUTCOMES

I reviewed 188 specimens of *Cebus* in the four museums in the United States (in addition to 122 specimens from seven museums in Colombia) corresponding to the following taxa: *adustus* Hershkovitz, 1949*, aequatorialis* Allen, 1914, *albifrons* (Humboldt, 1812), *capucinus* (Linnaeus, 1758), *cesarae* Hershkovitz, 1949, *curtus* Bangs, 1905, *cuscinus* Thomas, 1901, *imitator* Thomas, 1903, *leucocephalus* Gray, 1865, *malitiosus* Elliot, 1909, *versicolor* Pucheran, 1845, and *yuracus* Hershkowitz, 1949. Specimens correspond to eight countries (Honduras, Nicaragua, Costa Rica, Panamá, Colombia, Venezuela, Ecuador, and Perú). The dataset includes the type specimens of *C. aequatorialis*, *C. albifrons adustus*, *C. albifrons cesarae*, *C. albifrons albifrons*, *C. albifrons yuracus*, and *C. malitiosus*; and included the types specimens of three taxa currently considered junior synonyms of other species: *C. capucinus nigripectus* (=*capucinus*), *C. capucinus limitaneus* (=*imitator*), and *C. albifrons pleei* (=*cesarae*). In addition, I took skin and tissue samples from 24 specimens of *Cebus* at the Field Museum, 27 at the American Museum of Natural History, and 15 at the Smithsonian National Museum of Natural History, which will be processed in UCLA.

The morphometric data have already been processed and analyzed in a manuscript submitted to the Journal of Mammalian Evolution in September 2024. To assess morphometric disparities and compare the results with hypotheses based on genetic evidence about *Cebus* taxonomy, we used 2D geometric morphometry on 206 adult specimens (127 males, 77 females, 2 unsexed) belonging to 12 taxa by assigning anatomical landmarks in the frontal (11), lateral (18), and occlusal (16) views of the skull, and semilandmarks to describe the mandible outline (24). Skull shape is more variable than centroid size, lateral and ventral views of males seem to be more useful for distinguishing taxa, and morphometric disparities exist across and within all three geographic groups analyzed (Central America, Andes, and Amazon). Although skull shape in *Cebus* spp. varies slightly, our results suggest differences among some taxa and highlight the utility of studying shape to complement methods that have focused on size. Although some results agree with taxonomic classifications based on molecular evidence, it is important to note that *Cebus* species cover wide geographical distributions with high inter- and intraspecific phenotypic variability and a high diversity of ecological conditions. This makes it difficult to provide species diagnoses based just on morphometric or morphological characters and suggests the need to integrate different sources of evidence to resolve uncertainties about taxonomy and evolutionary relationships in the genus.

5. ACKNOWLEDGEMENTS

To the curators and technical assistants of the 11 biological collections visited for their help in accessing the specimens.

Left) Sebastián García-Restrepo visiting the Primate type specimens' section in the Field Museum of Natural History, Chicago, August 2023. **Right**) *Cebus* primate skins showing variation in color. National Museum of Natural History, Smithsonian Institution, Washington D.C., September 2023.

Skulls of *Cebus* specimens at the Field Museum of Natural History, Chicago, August 2023.

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IPS Research Grant Report

Corinna A. Most

"Do differences in the social behavior of wild infant olive baboons (*P. anubis*) persist in the juvenile period?"

Dr. Corinna A. Most (Iowa State University)

1 BACKGROUND/INTRODUCTION

Successfully navigating complex social environments is crucial for most primates and is thought to have been a driving force in the evolution of primate cognition[1,2]. However, few studies of the evolution of primate social cognition have taken into consideration the ontogeny of sophisticated social behavior in non-human primates, and the development of social competence in non-human primates is as yet not well understood. This project collected behavioral data on wild juvenile olive baboons at the Uaso Ngiro Baboon Project (UNBP) in Laikipia, Kenya, to investigate how immature individuals acquire the necessary social skills to become successful members of their group. In particular, this study examined whether differences in early social experiences and behavior persisted in later life stages, building on 16 months of behavioral data collected on the same individuals as infants and investigating whether the differences observed when they were younger persisted at this later age. Baboons are excellent subjects for this type of study because they display great variation and flexibility in social behavior[3]. As adults, olive baboons live in large heterogeneous groups that can exceed 100 animals, and in these "troops", individuals are involved in intricate social networks of a number of long-, medium-, and short-term relationships with both kin and non-kin that can be cooperative and affiliative as well as competitive and aggressive. An individual baboon's social relations affect every single aspect of its daily life, so that there is no aspect of their experience that is not intrinsically social[4]. This longitudinal study, situated within the long-term ecological and demographic data available from the site, was thus able to provide invaluable insight into the development of social competence in a cognitively sophisticated and socially complex non-human primate.

2 PROJECT OBJECTIVES/AIMS

The data previously recorded on these individuals as infants included information on mother-infant interactions, infant social relationships, and infant social behavior (play, distress, and orientation to interactions). Our findings indicated that infants with poorly responsive mothers displayed greater social competence (as indicated by lower frequency of distress and higher frequency of play behavior and of orientation towards social interactants). These results were in contrast with the human literature, which associates greater maternal responsiveness with better developmental outcomes. Therefore, the objective of this project was to examine whether the effects of maternal responsiveness were consistent throughout several life stages or whether the expected positive effects of maternal responsiveness on the development of social behavior only became apparent at a later age. Specifically, we wanted to assess whether those individuals that had experienced poorly responsive mothers and displayed greater social competence as infants would continue exhibiting greater social competence as juveniles.

3 BRIEF OVERVIEW OF STUDY LOCATION AND METHODS IF/AS RELEVANT

This research was carried out between over the course of 10 weeks in summer 2019 at the Uaso Ngiro Baboon Project (UNBP) in the Laikipia Plateau of Kenya. This field site was established at its current location in 1984 by Dr. Shirley Strum through the translocation of three troops of baboons that she had been previously studying at a different site in Kenya[5]. There are therefore decades of long-term behavioural, demographic, and ecological data on the study troops, and the animals – while still wild – are fully habituated to researchers. I conducted focal follows of two hours each of

all the juveniles that had been part of my original research as infants, continuously recording all social interactions they were involved in. During the focal follows I also recorded every five minutes the location of the focal animal relative to the rest of the troop (central/peripheric) and the identity of all individuals within a 5m radius of the focal animal. I recorded 138 hours of observational data (an average of ~15.3 hours per individual) and further supplemented my own data with the long-term behavioral records available at the site. This project was conducted in full compliance of both Kenyan (KWS and NACOSTI) and U.S. (IACUC) animal research guidelines.

4 KEY RESULTS/MAIN FINDINGS OR OUTCOMES

Unfortunately, only nine of the infants that had been studied in 2015 and for whom there was information on maternal responsiveness were still alive in 2019. Of these, three had experienced poor maternal responsiveness. Since the sample size was fairly small, we focused on grooming, as it was one of the most common social behaviors the individuals in the study engaged in and one that the UNBP also records long-term data on. In particular, we focused on number of grooming interactions and number of grooming partners recorded for each juvenile between 2016 and 2019 (see **Table 1**). Between the observational data collected for this project and the long-term UNBP data, there were 3,315 grooming interactions that the nine individuals had been involved in between 2016 and 2019. We found significant sex differences in grooming behavior, with females engaging in more grooming interactions and having more grooming partners (MWU, for both: $z = -2.449$, $p = .016$), so the two sexes had to be examined separately. Since only one of the four female juveniles had experienced poor maternal responsiveness as an infant and she was also slightly older than the other females and already a subadult, no further statistical analyses were conducted on the females. Of the five male juveniles, two had experienced poor maternal responsiveness, so although the sample was very small it was at least possible to compare a similar number of individuals. On average, males with responsive mothers had each engaged in ~255 grooming bouts with 26 grooming partners (so ~10 grooming bouts per partner), while males with poorly responsive mothers had each engaged in 140 grooming bouts with ~32 partners (so ~4.3 grooming bouts per partner). None of the differences were statistically significant, but the strongest effect of maternal responsiveness was on the number of grooming interactions (MWU, $z = -1.732$, $p=.083$). It was hard to draw any conclusions about the role of rank for either males or females.

ID	Sex	Maternal Rank	Maternal Resp.	Grooming Interactions	Grooming Partners	Grooming Interactions/ Partner
CF	F	M	Good	425	70	6.1
FG	F	M	Good	843	77	10.9
LV	F	L	Good	517	88	5.9
BZ	M	M	Good	247	21	11.8
LW	M	M	Good	291	22	13.2
UM	M	L	Good	226	35	6.5
JR	F	M	Poor	486	63	7.7
PG	M	H	Poor	111	26	4.3
QI	M	M	Poor	169	39	4.3

Table 1. ID, sex, maternal rank, maternal responsiveness, number of grooming partners, number of grooming interactions, and number of grooming interactions per partner between 2016-2019 for the juveniles in this study.

5 IMPLICATIONS OF PROJECT/DISCUSSION OF RESULTS

Because of the small sample size, these research findings are very tentative. Nonetheless, they do suggest that variation in maternal responsiveness might have long-term effects on the development of baboon social skills. Specifically, there has been extensive research on the importance for female baboons of close and stable grooming relationships, which have been found to influence not only a female's own health and lifespan, but also the survival of her infants [e.g, 6,7]. Unfortunately, it was impossible to draw any conclusions about the effects of maternal responsiveness on the grooming behaviour of the females in our study. However, the males that experienced poor maternal responsiveness as infants went on to have more grooming partners but fewer grooming interactions and, therefore, fewer grooming interactions with each partner – i.e., they had more but weaker social bonds. This supports the prediction – based on the human developmental literature – that while infants that experience poor maternal responsiveness may initially display greater social competence (possibly as a consequence of the earlier maternal rejection experienced) they may perhaps not fully develop the sophisticated social skills required to develop and sustain strong and stable relationships. Although the importance of these close relationships has been primarily established for female baboons, the socio-cognitive skills required to maintain them are likely beneficial for both sexes. Future research will continue to expand this study by 1) increasing the sample size, 2) assessing the effects of rank on social behavior, and 3) examining other types of social behavior.

6 ACKNOWLEDGEMENTS

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IPS Research Grant Report

Megan Mah

Multimodal fruit foraging and interspecific sensory variation in wild sympatric platyrrhines

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Research Committee

1 INTRODUCTION

The senses are crucial for food evaluation, as foraging animals assess food quality based on colour, scent, softness, taste, and sound[1]. Several studies have examined the role of colour vision in food evaluation[2-6], but less is known about the use of non-visual senses. Given the evolutionary plasticity of sensory systems, variation in sensory morphology and genetics among species has been used to infer their relative reliance on different senses, including during foraging[7-10]. For example, species with relatively larger olfactory bulbs[8] or more olfactory receptor genes[10] are assumed to rely more heavily on olfaction. Such inferences have also been applied to dietary guilds (e.g., the large olfactory bulbs of frugivores are suggested to indicate that olfaction is more important for fruit foraging than for other diets[8]), and dietary specialization is expected to impact the sensory use of species, since sensory systems evolve through selection favouring individuals that are well-equipped to evaluate their foods and should be well-adapted to principal diet type[11]. Yet, little work has explored species differences in sensory behaviour during foraging, making it unclear to what extent we can infer the sensory foraging behaviour of species from their morphology, genetics, or diet.

Primates exhibit wide variation in sensory systems[1], occupy diverse dietary niches[12], and are relatively easy to observe when foraging[13], making them a well-suited taxon in which to explore interactions among sensory systems, diet, and foraging behaviour. A handful of studies suggest that primate species differ in sensory use when evaluating the same foods[14-16], which may be linked to morphological or dietary variation (e.g., *Saimiri sciureus* have more dexterous hands and use manual touch more often than *Ateles geoffroyi*[15]; frugivorous *Varecia variegata* use olfaction more often than folivorous *Propithecus coquereli*[16]). Colour vision differences due to opsin gene variation may also affect the sensory use of foraging primates. For example, dichromatic (red-green colourblind) *Cebus imitator* sniff fruits more often than trichromatic ('normal' colour vision in humans) individuals, suggesting that dichromats may rely more on non-visual senses to mitigate their colour vision deficiency[17]. However, comparative studies of primate sensory food evaluation have largely been experimental, and the paucity of work exploring sensory use among species foraging in the wild leaves research removed from natural selective pressures.

2 OBJECTIVES

Here, we asked: do primate species differ in sensory food evaluation behaviours in ways that correlate with sensory or dietary variation? We hypothesized that the sensory morphology, sensory genetics, and dietary specialization of species shape their use of senses to evaluate foods. To test our hypothesis, we observed the fruit foraging behaviour of three species of wild sympatric primates, predicting that during fruit evaluation: 1) species with higher thumb-to-index finger length ratios, indicating greater manual dexterity[18], use manual touch more often, 2) dichromatic individuals use non-visual senses more often than trichromatic individuals, and 3) more frugivorous species use sensory evaluation behaviours more often.

3 METHODS

We studied habituated Geoffroy's spider monkeys (*Ateles geoffroyi*; one social group, $n = 24$), mantled howler monkeys (*Alouatta palliata*; two social groups, $n = 12$), and white-faced capuchins (*Cebus imitator*; two social groups, $n = 44$) in Sector Santa Rosa (SSR), Área de Conservación Guanacaste,

Costa Rica. The colour vision systems of these species are well-established: *A. geoffroyi* and *C. imitator* comprise both dichromatic and trichromatic individuals, whereas *A. palliata* are trichromatic only due to an opsin gene duplication[19]. Our *A. geoffroyi* and *C. imitator* study individuals have previously been genotyped for colour vision type (i.e., dichromatic or trichromatic). We ranked the species from most to least frugivorous by conducting a literature review of their published diet compositions (proportion of monthly diet composed by fruits) and digitally measured their thumb and index finger lengths from images of their hands. Finally, we collected behavioural data from the study species in SSR from May-Aug 2021 to supplement data we previously collected between 2004-2018 in SSR and Isla Agaltepec, Mexico. When monkeys foraged on fruit, we conducted 1-10 min modified focal animal samples[13] to record fruit investigation sequences, including each fruit-directed sniff, manual touch, and bite, as well as every fruit ingestion or rejection; we sampled as many group members as possible in each fruit patch, rotating among age-sex classes.

To test for variation among the study species in amount of frugivory and thumb-to-index finger length ratio, we ran Kruskal-Wallis tests followed by Dunn post hoc tests. We filtered our behavioural data to only include investigation sequences on fruit species that all three monkey species consumed, resulting in a final dataset of 26,094 investigation sequences on 13 fruit species. Using generalized linear mixed models with Poisson distributions, we analyzed the effects of 1) monkey species and 2) colour vision type on the number of investigation sequences that included 1) sniffing (olfactory behaviour), 2) manual touching (touching behaviour), and 3) biting followed by fruit rejection (tasting behaviour); we included the total number of investigation sequences as an offset and monkey ID and fruit species as random effects. This research complied with the laws of Costa Rica, Mexico, Canada, and the United States, as well as protocols approved by the Área de Conservación Guanacaste (R-SINAC-ACG-PI-027-18; R-025-2014-OT-CONAGEBIO) and the University of Calgary's Life and Environmental Care Committee (AC19-0167).

4 RESULTS

We observed significant differences among the study species in amount of frugivory and thumb-to-index finger length ratio. Monthly fruit consumption was higher in *Ateles* than in *Cebus* and *Alouatta*, and higher in *Cebus* than in *Alouatta* (i.e., *Ateles* > *Cebus* > *Alouatta*). Thumb-to-index finger length ratio was higher in *Cebus* than in *Alouatta* and *Ateles*, and higher in *Alouatta* than in *Ateles* (i.e., *Cebus* > *Alouatta* > *Ateles*). The species also differed significantly in the number of fruit investigation sequences that included olfactory, touching, and tasting behaviours. *Ateles* sniffed fruits more often than *Alouatta* and *Cebus*, and *Alouatta* sniffed fruits more often than *Cebus* (i.e., *Ateles* > *Alouatta* > *Cebus*). *Cebus* touched fruits more often than *Ateles* and *Alouatta*, and *Ateles* touched fruits more often than *Alouatta* (*Cebus* > *Ateles* > *Alouatta*). *Cebus* and *Alouatta* bit-rejected fruits more often than *Ateles* but did not differ significantly from each other in bite-reject frequency (i.e., *Cebus* = *Alouatta* > *Ateles*). Finally, colour vision type had significant effects on use of non-visual senses, with dichromats sniffing and bite-rejecting fruits more often than trichromats. However, dichromats and trichromats did not differ significantly in frequency of manual touching behaviour.

5 DISCUSSION

There was mixed support for the sensory morphology component of our hypothesis. Consistent with our prediction, *Cebus* had the most dexterous hands and evaluated fruits using manual touch most often. However, *Ateles* touched fruits more often than *Alouatta* despite having lower manual dexterity. These results suggest that relationships between sensory morphology and sensory behaviour may be more complicated than sometimes acknowledged, and we recommend exercising caution when using morphological proxies to infer sensory reliance. Our finding that dichromatic *Ateles* and *Cebus* evaluated fruits with olfactory and tasting behaviours more often than trichromatic *Alouatta*, *Ateles*, and *Cebus* is consistent with the hypothesis that dichromats rely more heavily on non-visual senses to mitigate their reduced colour vision abilities. Finally, our results provide mixed support for the dietary specialization component of our hypothesis. As predicted, *Ateles* used olfaction to evaluate fruits more often than the less frugivorous *Alouatta* and *Cebus*, and *Ateles* and *Cebus* used manual touch more often than the less frugivorous *Alouatta*. These findings are consistent with our expectation that sensory foraging behaviour should be well-adapted to principal diet type. Unexpectedly, *Alouatta* used olfaction to evaluate fruits more often than the more frugivorous *Cebus*, and *Ateles* bit-rejected fruits the least often despite being the most frugivorous. The latter result is potentially complicated by the fact that biting also provides information about fruit hardness (i.e., it may also be a touching behaviour). Moreover, our study only included three species, and studying a greater number of primate species would be instructive for clarifying the relationships between dietary specialization and sensory use for food evaluation.

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IPS Research Grant Report

Evelina Daniela Rodrigues

Chimpanzee 'babytalk': Gestural motherese in wild chimpanzees (*Pan troglodytes*)

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1 BACKGROUND

We often adjust the way we communicate when interacting with young children: child-directed communication (also known by many different names including: 'motherese', 'parentese', 'babytalk', and 'infant-directed communication'). In humans, characteristics of this special communicative register include the use of simplified vocabulary, slower tempo, exaggeration of certain communicative features, and a higher rate of repetitions[1,2]. Although not universal[3,4], and with considerable variability in how it is expressed between cultures–including in the number and type of communicative features that are adjusted–this special register is found all over the globe. The very widespread use suggests that it may be a species-typical capacity within human communication, and could potentially be evolutionarily older than modern humans[5]. The study of child-directed communication in our closest phylogenetic relatives–chimpanzees–allows us to gain insights into its evolution.

Of the different types of communicative signals employed by chimpanzees, gestures are a promising modality, as they are already employed in highly flexible ways that appear sensitive to characteristics of the recipient[6,7]. Some evidence for immature-directed gestures has been reported in great apes[8,9]. Some adjustments described in chimpanzee mothers' communicative behaviour specifically relate to their use of gesture modalities depending on offspring age: chimpanzee mothers are less likely to produce tactile gestures, as compared to visual gestures, as their infants get older[8]. However, to date, only broader aspects of gestural communication, such as the modality used, have been investigated and the finer-grained features often explored in humans, such as communicative unit durations and rates, remain unexplored.

2 PROJECT OBJECTIVES

The aim of this study is to investigate whether wild chimpanzees accommodate their gestural communication to immature recipients. For this purpose, we focused on chimpanzee mothers and first investigated whether they adjusted their gestural communication to a recipient's age, sex, and kinship when interacting with any individual in the community. We then focused on mother-offspring pairs to analyse whether they specifically adjusted their gestural communication to the age and sex of their offspring. The focus on mother-offspring interactions allowed us to explore adaptations to more fine-grained age differences. We particularly explored: vocabulary complexity, temporal patterns, and the prominence of chimpanzee mothers' communication.

3 STUDY LOCATION AND METHODS

The dataset contains communicative gestures produced by 25 mothers from two wild and habituated chimpanzee communities: 15 females from Sonso (East African Chimpanzees; *Pan troglodytes schweinfurthii*) and 10 females from Bossou (West African chimpanzees; *Pan troglodytes verus*). To be included in our study, females had to be subadult or adult (older than 9 years) with at least one dependent offspring present in the group during the study period. We extracted archival video data from Sonso and Bossou that were included in the wider GesturalOrigins project[10]. As this project focused on gestures produced by specific females, and the

Sonso archive covered a more limited period than the Bossou archive, we collected additional data on Sonso females from November 2022 until January 2023. Regional permits from the Ugandan National Council of Science and Technology and the Ugandan Wildlife Authority were already in place for data collection. Existing data were collected with all regional and national research permits in place, and with ethical approval from the Animal Welfare and Ethics Committee of the University of St Andrews.

For this study, we focused on the following individual and relational features: signaller and recipient ID and age; recipient sex; their kinship relation (maternal kin-related, non-maternal kin related). For each communicative event, we extracted information about the goal (e.g., give me affiliation, groom me, etc.), gesture actions (e.g., Raise, Push, etc.), their sequential use (relative position in the sequence), and evidence for emphasis, persistence, and number of repetitions in gesture actions that allow repetition in their structure (e.g.: Hitting, Stomping). Our coding scheme[11] allowed us to extract detailed information about the timings of different communicative elements (e.g., sequences, gestures, gesture units, latencies, etc.).

Our study explored five gestural features: type/token ratio (the number of gesture forms performed by the individual divided by the number of gesture instances); gesture rate (the number of gestures per sequence divided by the total sequence duration); the likelihood of using repetition (either within an instance of gesturing or a repetition of the same gesture type in the sequence of gestures); gesture duration (here we considered the relevant part of the gesture, discarding the recovering phase); and the hold/repetition phase duration (for the gesture actions that have the option of then being held in place or rhythmically repeated).

4 KEY RESULTS

In total we included 1206 gesture tokens produced by chimpanzee mothers towards any individual of the community, of which 550 were directed to their offspring. Sonso mothers produced 735 tokens of 43 gesture types, of which 407 tokens of 28 gesture types were directed towards their offspring. Bossou mothers produced 471 tokens of 42 gesture types, of which 143 tokens of 26 gesture types were directed towards their offspring. We found that chimpanzee mothers adjusted the expression of their gestural communication to the age of their recipient and with their kin relationship, but not to the sex of their recipient.

When analysing the communication of chimpanzee mothers to any individual of the community, these females used shorter units, both gesture duration and hold/repetition phase duration, towards immature individuals. They also used more diverse gesture repertoires towards subadults as compared to adults. Their kinship relationship with the recipient also impacted a range of communicative features. Chimpanzee mothers used shorter gestures, communicated at a faster pace, with a more diverse set of gesture forms, and were less likely to use repetitions towards non-kin individuals.

The temporal patterns found in the analyses of mother chimpanzees' communication to any member of the community were not found in the mother-offspring subset of communications. When focusing on the subset of communication directed towards their offspring, chimpanzee mothers were still using a more diverse set of gesture forms towards their subadult offspring (as compared to juveniles) and were more likely to use repetitions towards older offspring.

5 DISCUSSION OF RESULTS

Chimpanzee mothers adjusted the expression of their gestural communication to the age of their recipient and with their kin relationship. The lack of any effect of recipient's sex on gestural expression suggests that sex may be a less relevant feature to adapt in communication. It might be, instead, more critical to adapt other social behaviour in which communication occurs – such as gregariousness. For example, a mother might impact their offspring development and opportunities to communicate by changing the number and range of available social partners through selective subgrouping[12].

The age of the recipient influenced some of the gestural features analysed in this study, but in the opposite direction to that predicted from the use of child-directed communication in human languages: mothers produced shorter gestural units towards immature individuals and were more likely to use repetitions towards older offspring. Specifically, they produced shorter gestures and shorter hold/repetition phases towards infants and juveniles as compared to towards adults and subadults. In combination, this finding suggests that mothers

were shortening the optional aspects of gesture production (holds and repetitions). Interestingly, when analysing the subset of communication between mothers and their offspring we did not find any effect of the recipient age on these temporal aspects, suggesting that kinship and recipient age may interact. It is possible that this trend only emerges in larger datasets or that it might be driven by non-kin individuals with whom the mother is likely less familiar and interacts with less frequently. Kinship seems to play an important role, with mothers communicating more clearly and more efficiently with non-kin individuals, using more types of gestures, at a faster pace, shorter gestures, and less repetition. The social dynamics of the chimpanzee groups and the need to convey clear messages to more distantly related individuals may have been prioritized through evolution, over other communicative patterns and functions.

This study shows that chimpanzees accommodate their communication when interacting with immature individuals, suggesting that this ability is evolutionarily older than human language, and was likely present in a last common ancestor. However, the way in which chimpanzees accommodate their communication differs from what has been described in human child-directed communication. These adjustments might serve to signal social closeness or manage hierarchical relationships, rather than facilitating learning alone. Future research is needed to explore additional aspects of chimpanzee communication, including its social functions and the interplay between kinship, age, and other factors in shaping gestural expressions.

6 ACKNOWLEDGEMENTS

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IPS Research Grant Report

Molly Amber Hirst

Purifying Selection and Complex Patterns of Introgression of Putative Sperm-Related Genes in a Howler Monkey Hybrid Zone

Molly Amber Hirst, University of Michigan

Note: The original goal of this IPS grant was to aid with costs associated with genotyping *Alouatta palliata, A. pigra,* and their hybrids in southern Mexico to determine levels of admixture for a sperm morphology comparison project. However, due to the COVID-19 pandemic, the blood samples required for genotyping the sampled individuals were never shipped to the US. Instead, the IPS funds were used to generate the first *A. pigra* genome assembly, which I used to complete my final dissertation research chapter[1]. This report focuses mostly on the generation of the genome assembly, not all of the downstream analyses that resulted from the generation of the assembly.

1 BACKGROUND/INTRODUCTION

The rapid development of technological and analytical approaches to generate genome assemblies for non-model organisms allows us to compare genomes of closely related species to investigate genetic mechanisms arising during and after species diverge. Here I analyzed divergence of putative sperm-related genes between *Alouatta palliata* and *A. pigra*, sister species with different mating systems that form a hybrid zone. I previously found interspecific sperm morphological differences between these species, consistent with hypothesized differences in sperm competition intensity, as well as evidence of sperm and testicle anomalies in hybrid individuals. Given that many sperm traits are heritable and affected by selection, and that reproductive isolation is strong between these species, I asked if putative sperm-related genes show patterns of introgression consistent with adaptation or reproductive isolation in the hybrid zone, and how these patterns may relate to patterns observed in other instances of primate hybridization, such as between humans and archaic hominins. To answer these questions, I selected several genes that are detrimental to sperm development or function in humans and identified differences among these putative sperm-related genes between the two *Alouatta* parental species. I recognize that these putative sperm-related genes may or may not actually contribute to the phenotypes I observed in these howler monkeys, and that this study is only beginning to explore putative sperm-related genes in this howler monkey hybrid system. I built a *de novo* genome assembly for *A. pigra* and used the publicly available genome assembly for *A. palliata* to begin exploring putative sperm-related genes by first looking at orthologs of 126 genes known to cause sperm defects in humans. I recovered exon fragments of 116 genes in both species, from which 86 had at least one base pair difference between species. I further narrowed my focus to 21 genes that were located in scaffolds for which previously generated SNP genotypes existed for admixed and non-admixed individuals to infer patterns of introgression of these genes in the hybrid zone. I found that 17 of these genes are under purifying selection, and four are under neutral evolution. Most of the SNPs associated with genomic regions near these genes show excess *A. palliata* ancestry; however, I found approximately equal instances of SNPs showing reduced and increased introgression patterns. Importantly, I found nine SNPs located between 8 to 8000 bp away from putative exonic regions of sperm-related genes, which likely reflect the pattern of introgression of those genes in the hybrid zone. Two of those genes, CATSPER3 and TEX11, show reduced introgression in the hybrid zone and have elevated expression in human testes. When comparing the set of genes to those located in known deserts of archaic ancestry in humans, I found one gene (GSTM1) located within a desert of archaic ancestry in humans that is on a genomic scaffold with an *Alouatta* SNP that shows reduced introgression. If the putative gene follows the same pattern of introgression as the SNP, it suggests that GSTM1, or genes linked to it, may be important in reproductive isolation across primates. This study has greatly contributed to the development of genomic resources, enabling a more comprehensive examination of the importance of sperm-related genes in adaptative introgression and reproductive isolation in the howler monkey

hybrid zone. To that end, further studies analyzing *Alouatta* orthologs of other known sperm-related genes in humans and other organisms are needed.

2 PROJECT OBJECTIVES/AIMS

- Build the first *A. pigra* genome assembly. This includes DNA extraction of 4 *A. pigra* individuals, choosing the highest quality extraction for the assembly, library preparation and whole-genome sequencing, *de novo* genome assembly, quality control, and general assembly statistics determination.
- Find and compare known human sperm-related gene orthologs in *A. palliata* and *A. pigra* genome assemblies.
- Determine if these gene sequences are located on genomic scaffolds previously found to be associated with reduced, increased, or neutral introgression patterns in the howler monkey hybrid zone. Test for selection. Compare nearby *Alouatta* SNPs with regions of excess or lack of archaic ancestry in humans.

3 BRIEF OVERVIEW OF STUDY LOCATION AND METHODS

I completed all research described here and in my sperm morphology studies in accordance with the University of Michigan Institutional Animal Care and Use Committee (PR00009319 and PRO00009558) and in adherence to the legal requirements in Mexico (SGPA/DGVS/03676/07, SGPA/DGVS/04949/07, SGPA/DGVS/03293/10, SGPA/DGVS/4936/19) and the United States (USFWS 08US178005/9, 12US46480A/9, 21US77243D/9). All research described here follows the American Society of Primatologists Ethical Principles for the Treatment of Non-Human Primates. As part of an ongoing study of *Alouatta* hybridization[3], our field team collected a whole blood sample preserved in lysis buffer from an adult female *Alouatta pigra* in Campeche, Mexico in 2010. I performed DNA extraction and quality control before sending the extraction to the University of Michigan's Sequencing Core for library preparation and whole genome sequencing. The prepared libraries were sequenced on a shared flow cell (3.75%) using the Illumina NovaSeq 6000 platform (Illumina, Inc.) for 300 cycles, resulting in 150 bp paired-end reads. Given that the sequencing was conducted on the Illumina platform, which generates short-reads, I chose to assemble the genome using SPAdes, a specialized short-read *de novo* genome assembler. I then used QUAST to evaluate the quality of the resulting genome assembly. To determine the depth and consistency of the assembly, I calculated its coverage using BBMap.

4 KEY RESULTS/MAIN FINDINGS OR OUTCOMES

The whole-genome sequencing of the *A. pigra* sample produced 354,394,574 reads, exhibiting a GC content of 41%. After trimming the raw reads that did not meet my filtering criteria, I retained 35
296,687,342 reads ranging in length from 50-151 bases and with a GC content of 40%. The genome assembly resulted in 139,436 contigs, encompassing a total of 2,461,750,957 base pairs. The overall GC content of the assembly is 40.71%, with 99.21% of the reads having been constructed in the assembly. Overall, the genome assembly has an N50 value of 26,980 with the largest scaffold consisting of 265,688 base pairs, and an average of 29.09X depth of coverage.

As part of the subsequent analysis, I focused on 126 human genes known to affect sperm morphology. I excluded Y chromosome genes because both *Alouatta* genomes were built with female DNA, and mitochondrial genes because they are under different selective pressures. To find the orthologs of these human genes in both *Alouatta* genome assemblies, I implemented BLAST searches. I successfully identified exon sequences from 106/113 autosomal genes and 5/5 X-linked genes in both *Alouatta* genome assemblies. There were seven genes that could not be located in the *Alouatta* genome assemblies, perhaps due to the incompleteness of both assemblies, or because they have greatly diverged from the human sequences.

I tested 21 putative sperm-related genes for modes of selection in MEGA11 using Fisher's exact test and found no evidence of positive selection in these gene coding regions, i.e., there were no statistical comparisons between the dN/dS ratios between *A. palliata* and *A. pigra* that showed a *p*-value of 0.05 or less. Despite the presence of some non-synonymous substitutions between the sequences of *A. palliata* and *A. pigra*, most of the genes I explored showed evidence of purifying selection, where $p = 1$ (N = 17 genes). The other four genes showed evidence of neutral evolution, where $0.05 < p < 1$.

I identified 52 SNPs that were identified as outliers in previous introgression analysis of this hybrid zone[2] and were located on the same genomic scaffolds as the 21 genes of interest in my analysis. I found nearly equal instances of SNPs with a positive (20/52) or negative (23/52) beta outlier, which indicates reduced and increased introgression, respectively. Most SNPs (N = 27) showed negative alpha (i.e., excess *A. palliata*) with an equal share of negative beta (i.e., increased introgression, N = 14), or positive beta (reduced introgression, N = 13). Nine SNPs were alpha, but not beta, outliers, and five were beta, but not alpha, outliers. The closest SNP to a gene sequence of interest was located 8 bp away from a sequence in the CATSPER3 gene and showed excess *A. pigra* ancestry and reduced introgression.

5 IMPLICATIONS OF PROJECT/DISCUSSION OF RESULTS

I created the first *de novo Alouatta pigra* reference genome assembly to compare putative sperm-related genes between *A. pigra* and *A. palliata* that are known to be vital for proper sperm development, function, and morphology in humans. I also leveraged existing genome-wide SNP data from the *A. pigra x A. palliata* hybrid zone to infer patterns of introgression of these genes, and human genomic maps of depleted ("deserts") and elevated archaic hominin ancestry to explore potentially common genes associated with reproductive isolation and adaptation in primates. Overall, I focused on 21 putative sperm-related genes, of which 17 show evidence of purifying selection and four are under neutral evolution. These 21 genes span 19 scaffolds in the *A. palliata* genome assembly and contain 52 SNPs. The patterns of introgression of the SNPs varies, with most showing excess *A. palliata* ancestry (31/52), and split patterns of increased and reduced introgression (23/52 and 20/52, respectively). From my howler monkey dataset, I found six SNPs associated with genes located in deserts of archaic ancestry on human chromosome 1, with most showing *A. palliata* ancestry and reduced introgression. I also found two SNPs associated with genes in regions of elevated archaic ancestry in humans; however, these SNPs show contrasting patterns of increased and decreased introgression, and only one is an alpha outlier, showing excess *A. palliata* ancestry. However, with my data it is not possible to determine if the patterns of introgression of these SNPs are related to those of the actual genes. Overall, my results reveal a complex tapestry of putative sperm-related genes, their patterns of introgression in the howler monkey hybrid zone, and their candidacy as barrier or adaptive loci. A more comprehensive examination of additional autosomal sperm-related genes, as well as whole genome resequencing of multiple parental and admixed individuals with varying hybrid indexes, should provide more clarity on the role of sperm-related genes in reproductive isolation and adaptation in the howler monkey hybrid zone.

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IPS Research Grant Report

Philippa Hammond

Remote monitoring of predation pressure and its effects on baboon demographics, ranging patterns, and nocturnal behaviours.

Philippa Hammond | University of Oxford, United Kingdom

The spread of predation risk across an environment creates a "landscape of fear" that acts as a significant driver of behaviour and evolution in prey species[1]. In primates, predation pressure is thought to influence demographic features like group size and male: female ratios. It is also thought to impact primates' movement within their landscape, their use of the ground, and their activity budgets, as well as sleep-related behaviours[2,3,4].

A convoluting issue in predation research is that predation pressure is often treated as "a homogenous evolutionary force", despite the fact that different predators pose a variety of threats to their prey[5], for example through different diel patterns and hunting techniques. Another issue is that certain aspects of predation pressure are likely to influence the behaviour of primates at the individual level, whilst others might have more of an effect at the troop or population level. The relationships between the effects of risk at different levels has been underexplored.

Project objectives

In this project, a component of my PhD research, I set out to study the issues outlined above. Following our habituation of two troops of chacma baboons in Gorongosa National Park (GNP), Mozambique, the proposed outline for this project was to study the effects of predation pressure at the troop level and population level within GNP. Originally, we intended to set up a localised camera trap grid that covered the home ranges of the two habituated troops within a broader pre-established camera trap grid covering 300km^2. This research plan had to be altered after complications caused by Cyclone Idai in March 2019, followed by delays in predator reintroductions to the park, and then the Covid-19 Pandemic. Rather than using nested camera trap grids, we used two separate camera trap grids to compare the terrestrial activity of neighbouring populations of chacma baboons.

Terrestrial activity is rare in primates, but is associated with a variety of morphological, behavioural, and cognitive traits across the order, and is a defining feature of our own human lineage. With the exception of humans, primates who engage in terrestrial behaviours rarely do so outside of daylight hours. Predation pressure is thought to have played a significant role in shaping behaviours like the selection of arboreal sleep sites, but little is known – particularly at the landscape scale – about how risk interacts with other ecological and seasonal variables to drive or inhibit terrestrial primate activity. In this project, we used camera trap data to investigate how patterns of terrestrial activity in baboons vary with tree coverage, proximity to water, season, anthropogenic variables, and the prevalence and type of apex predators in the landscape.

Study location and methods

GNP (-18.96°, 34.36°), is composed of ca. 4,000 km2 of heterogeneous habitats and is inhabited by over 200 troops of chacma baboons, amongst many other species. Whilst apex predators including lions, leopards, wild dogs, and spotted hyenas used to be widespread in the park, civil war in Mozambique between 1977 and 1992 decimated wildlife in the park and apex predators were almost completely lost from the landscape, except for a small but now growing lion population. Since 2018, both wild dogs and leopards have been successfully reintroduced to the park, but these reintroductions occurred after the data collection period for this study[3].

A 5333 km^2 buffer zone surrounds Gorongosa National Park and is designated as a mixed-use area, inhabited by 200 000 subsistence farmers. To the east of Gorongosa's park boundaries and buffer zone lies a 460 km^2 forestry concession, certified by the Forest Stewardship Council for low intensity, rotational timber harvesting.

The area is composed of predominantly tall miombo woodlands, with some variation in vegetation density between more open grasslands and closed-canopy riparian areas, but generally tree coverage is more homogenous than within the park's boundaries. The concession is inhabited by several mammalian species, with leopards as the only known apex predator in the area. The concession partly contains a human settlement, as well as several households and a sawmill[6]. This study draws on data collected from two camera trap grids, mapped in Figure 1 below.

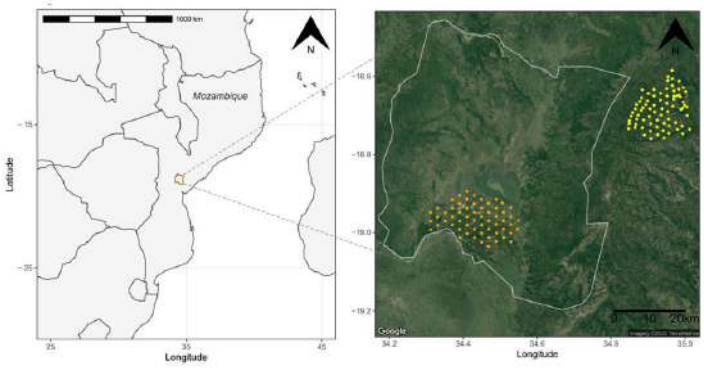

Figure 4: Map of GNP in Mozambique (on left), and the locations of two camera trap grids in relation to the park's boundaries (on right). The position of each camera station is marked in orange for the GNP grid and in yellow for the Forestry Concession (FC) grid.

We analysed camera trap detections of baboons' terrestrial activity both spatially and temporally. We used Generalised Linear Mixed Effect Models (GLMMs) to explore whether baboon activity varied with localised predator activity, ecological, seasonal, and anthropogenic variables across each of the grids. We also used temporal overlap analyses to explore variation in baboon activity across the 24-hr cycle, how these diel patterns differed across the two grids, and to what extent the two baboon populations showed temporal partitioning with their respective predator species (lions in the GNP grid and leopards in the FC grid). This work was carried out with ethical clearance from Oxford University (APA/1/5/ACER/23Jan2018) and from the Ministry of Tourism and the Gorongosa Restoration Project in Mozambique (permit numbers PNG/DSCi/C114/2018 and PNG/DSCi/C93/2018).

Findings and discussion

We had hypothesised that baboons may avoid or be more arboreal in areas where predators were more active across each grid. However, we found no evidence linking variation in terrestrial baboon activity to localised predator presence. The results from these models also indicated that baboons' terrestrial activity did not vary reliably with tree coverage, proximity to water, or proximity to anthropogenic features in the environment. Across the GNP camera trap grid, we did find season to be a significant predictor of baboon terrestrial activity, with baboons more active on the ground during the late dry season than at other times of year. Baboon detections across the grid might be lower in the wet season because baboons do not need to move around as much to find resources. Alternatively – or perhaps as an additional factor – the wet season likely coincides with greater availability of arboreal resources like fruit, whilst the dry season requires baboons to spend more time on the ground travelling to scarce water sources or foraging for grasses or underground storage organs.

From our diel analyses, our results showed that baboons across both grids were clearly diurnal, as can be seen in Figure 2. However, we found a low probability that the baboon activity patterns from each grid came from the same underlying distribution. Across FC, baboon activity was highest during the day compared to dawn and dusk periods, and no activity was detected at night. Across GNP, however, baboon activity was high during both dawn and day periods, only declining slightly at dusk, and there were even a few (n = 20) detections of baboons on the ground at night. Figure 2 clearly shows a peak in terrestrial activity detection soon after sunrise in GNP, whilst the FC distribution is centred more around the middle of the day.

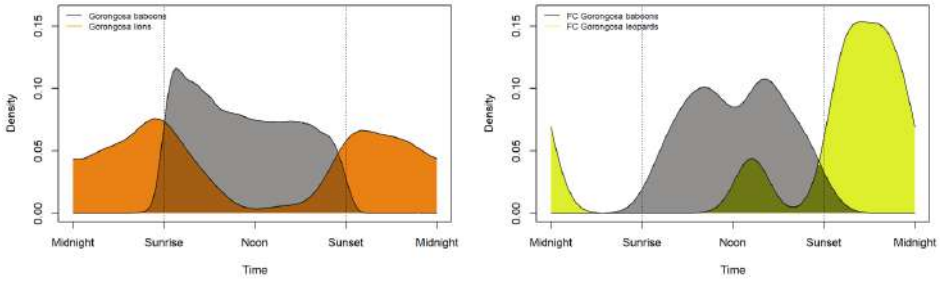

Figure 5: Graphs of the temporal overlap between detections of baboons (grey) and apex predators at each site (GNP lions in orange and FC leopards in yellow). Dotted vertical lines represent sunrise and sunset, standardised across sites and seasons.

This difference in timing of terrestrial activity across the two grids might be due to the presence of different apex predators in each landscape. Figure 2 demonstrates that both lion and leopard detections were predominantly nocturnal and that baboons show significant temporal partitioning with both predator species in their respective environments. However, baboons appear to exhibit greater temporal partitioning with leopards (their primary predator) whilst perhaps feeling safer to venture to the ground very early in the morning and even sometimes at night in the GNP environment, despite overlap with lion activity.

This study highlights the potential impact of risk variation on primate terrestriality at a landscape scale, with a particular emphasis on temporal dynamics and predator species. To build a fuller picture of these dynamics, it will be necessary to tease apart the effects and interactions of many spatial and temporal variables. For instance, whilst we found no effect of woody cover on baboon terrestriality in this study, it is likely that there are thresholds above and below which tree coverage drastically alters baboons' spatial activity patterns. And whilst each of the landscapes featured in this research was only inhabited by one apex predator, ecosystems with several carnivores will have more complex dynamics of spatial and temporal partitioning amongst members of the guild, with varying consequences for prey behaviour. Remote sensing methodologies are improving our abilities to monitor community ecology at broad scales and with unprecedented scope for longitudinal and simultaneous monitoring of species. These technologies, combined with more traditional *in situ* observational research, will facilitate more accurate and nuanced models of how risk impacts behavioural ecology and evolution in primates and other taxa.

Acknowledgements

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IPS Research Grant Report

Juliane Damm

Modulation of HPA-activation through social dynamics and behavior in wild Geoffroy's spider monkeys (*Ateles geoffroyi*)

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1 Background and Study aim

When researchers apply the traditional stress model to wild animal populations, they often argue that high levels of glucocorticoids (GC) can be used as an indicator for decreased fitness. This argument has been questioned, given that wild populations are subject to selective pressures and observable responses to environmental and social challenges are, thus, presumably adaptive (1). The allostasis model has been proposed as an alternative model allowing investigation of the interaction between the individual and the complex natural environment in an integrative manner (2). According to the allostasis model, the energetic state of the individual prior to the onset of a challenge predicts whether and to what extent the hypothalamic-pituitary-adrenal axis (HPA) will be activated and how long it would take for a prolonged activation of the HPA to lead to detrimental health effects (2). Research, particularly on wild primates, has shown that social factors (e.g., social networks, dominance rank, presence of bonding partners, frequency of affiliative behavior) can affect the individuals' HPA activation reducing overall GC levels (3).

Spider monkeys live in groups with high degrees of fission-fusion dynamics, in which group members frequently join and separate into subgroups of variable size and composition (4). This flexibility made them an ideal species for my research project. The aim of my study was to examine whether Geoffroy's spider monkeys' high degree of fission-fusion dynamics modulate HPA activation concentrations in response to ecological challenges. I focused on two environmental challenges that have previously been shown to elicit HPA activation in a variety of species (e.g., food scarcity [5]), predation risk [6]) and examined whether behavioral coping (e.g., fission into smaller subgroups, fusion into larger subgroups) during periods with these challenges had an impact on mean GC concentrations.

2 Methods

The study complied with the animal protection and wildlife research laws of Mexico (NOM-126-SEMARNAT-2000, NOM-059-SEMARNAT-2010) and the necessary research permit was granted from the Secretaría de Medio Ambiente y Recursos Naturales (SGPA/DGVS/03005/19). The study followed the Code of Best Practices for Field Primatology provided by the International Primatological Society (IPS) and the American Society of Primatologists. 40

I studied a well-habituated group of 54 wild Geoffroy´s spider monkeys (as of December 2020) living in the protected area of Otoch Ma´ax Yetel Kooh in Yucatan (Figure 1) and collected ecological, behavioral, and physiological data of all adult and subadult females and males (n=30). With the support of my research assistants, Romina Yitani, Daniele Baraldi, and Carlos Mukul, we non-invasively collected a total of 1199 fecal samples and I quantified fecal GC metabolite (fGCM) concentrations of 703 samples using a commercial cortisol enzyme immunoassay (7). We followed subgroups 4-5 days/week for ca. 8 hours per day and collected all-occurrence data of fission and fusion events (8). Because actual predation events are rarely observed in spider monkeys, perceived predation risk was quantified indirectly using the occurrences of predator elicited alarm calls (9). Eulogio and Macedonio Canul collected data on food availability by monitoring food trees on an established phenology trail once a month (10). I analyzed the data by implementing linear models and linear mixed models using the lm4 package of R (cf. 7).

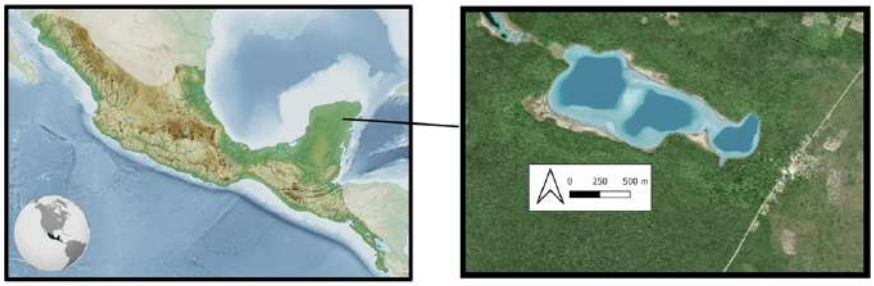

Figure 1: Location of the study area. Map on the left: modified from © Sémhur / Wikimedia Commons. Map on the right: modified from Google maps and georeferenced with QGis (version 3.28.9).

3 Main Results

Food availability and subgroup size

I found that low food availability was associated with increased fGCM concentrations in wild spider monkeys. When I focused only on periods of low food availability, I found that fGCM concentrations were lower when monkeys spent time in smaller subgroups than when they spent time in larger subgroups (Figure 2, left), indicating that fissioning into smaller subgroups may serve as behavioral coping in periods of food scarcity.

Perceived predation risk and subgroup size

I found that perceived predation risk (i.e. when alarm call rates were higher) was associated with an increase in fGCM concentration after accounting for the possible masking effect of behavioral coping (i.e. if behavioral coping is very effective, it is possible that no difference between high and low perceived predation risk can be found). When I focused only on periods of high perceived predation risk, I found that fGCM concentrations were lower when individuals spent time in larger subgroups than when they spent time in smaller subgroups (Figure 2, right), indicating that fusing into larger subgroups may serve as behavioral coping in periods of high perceived predation risk.

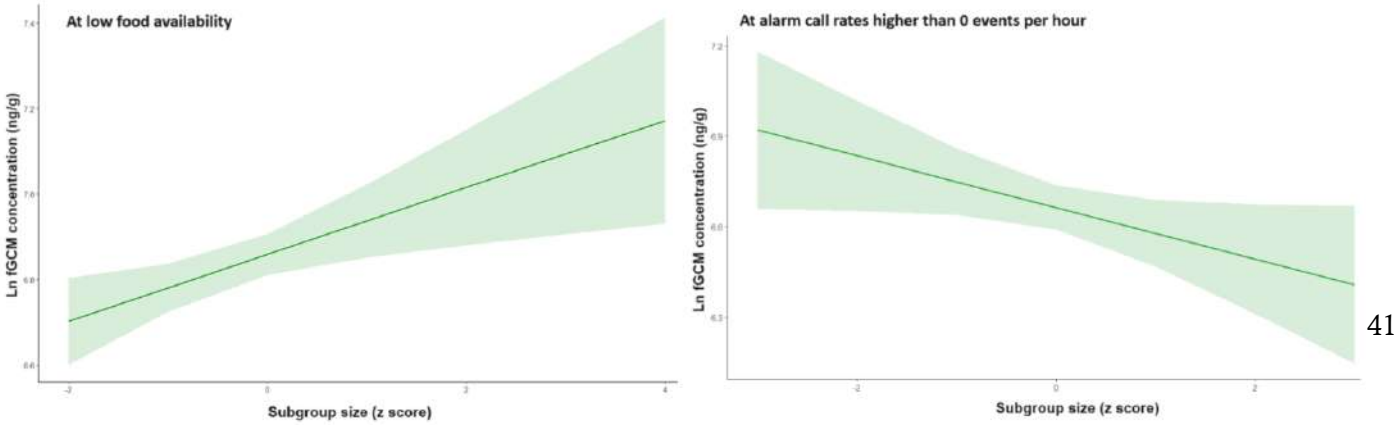

Figure 2: Graphical illustration of the predictions obtained from the linear models investigating the association between fGCM concentration of subgroup size at low food availability (left) and at high perceived predation risk (right).

4 Conclusion and Impact

The results of my study provide evidence that a high degree of fission-fusion dynamics in wild Geoffroy´s spider monkeys serve as effective behavioral coping to modulate HPA activation in response to two major ecological challenges. I found that 1) under conditions of low food availability fGCM concentrations were lower in smaller subgroups, and 2) under conditions of increased perceived predation risk fGCM concentrations were lower in larger subgroups. If predation risk is high under conditions of low food availability an increase in subgroup size is highly adaptive but comes at the cost of increased allostatic load. Increased allostatic load is adaptive when it allows for behavioral flexibility. This highlights the importance not only of assessing allostatic load but also considering how it relates to fitness (1). My study also contributes to knowledge

of spider monkey endocrinology, which will allow future research to investigate the similarities and differences in how behavioral coping strategies modulate GC physiology in sympatric but behaviorally and physiologically divergent taxa (e.g., *Ateles* and *Alouatta*; [11]) or between taxa that independently evolved convergent behavioral strategies (e.g., *Ateles* and *Pan* [12]). Lastly, my contribution to understanding the importance of behavioral coping on how individuals deal with challenges might allow for more elaborate research questions and predictions related to how the endangered spider monkeys will be able to weather anthropogenic challenges.

5 Additional outcomes

Thanks to the IPS research grant, I was able to train Daniele Baraldi, an Italian volunteer, and Carlos Canul Mukul, a young man from the local community, in physiological and behavioral sample collection in the field. I participated as a teacher in four university courses (Universidad Veracruzana in May and September 2020 and May 2024; University of Calgary in October 2022). I presented the results of the validation of the commercial enzyme immunoassay I used to quantify GCM in feces from Geoffroy´s spider monkeys at the 2023 IPS conference in Kuching and published the results of the validation study in the American Journal of Primatology in January 2024. With the collected data, I will be able to prepare at least two more publications and I plan on presenting the results at the SLAPrim (Latin American Society of Primatologists) conference in November 2024 and disseminating findings to a more general audience and specifically to the local community of Punta Laguna.

6 My heartfelt thanks go to:

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IPS Conservation, Captive Care, Education Grant Report

Clara Mariencheck

Characterizing COVID 19 genetic risk factors in non-human primates: *ACE2* variation in captive chimpanzees (*Pan troglodytes verus*)

Clara Mariencheck, PhD Candidate, The George Washington University

BACKGROUND

The respiratory virus SARS-CoV-2 that led to the COVID-19 global pandemic presents an ongoing threat to both human and nonhuman primates. The host cell receptor for this virus, ACE2, encoded by the *ACE2* gene, displays amino acid residues that are key for the virus to bind to the cell.[1] The ACE2 protein is expressed on the surface of epithelial cells in the lung and small intestine, as well as in arterial and venous endothelial cells.[1] The mammalian *ACE2* gene is highly conserved at both the peptide and DNA levels; therefore, most mammals share very similar genetic sequences and proteins for this receptor.[2] Coronaviruses and other respiratory viruses that share this receptor have been co-evolving along with the hosts for many years.[3] Catarrhines especially are at high risk of SARS-CoV-2 transmission due to their genetic similarity to humans; specifically, humans and other catarrhines share the same set of twelve amino acid residues in the protein ACE2 that are important for the virus to bind and enter the cell.[4] As our closest living relatives, chimpanzees (*Pan troglodytes*) are at high risk of transmission of respiratory viruses, such as SARS-CoV-2. Respiratory virus outbreaks are common in primate communities, such as in the wild chimpanzee populations in Gombe National Park.[5] These outbreaks are presumed to be linked to human transmission at several different research sites, including Taï National Park[6] and Kibale National Park[7] and can be fatal to the infected individuals.[8] In humans, several genetic variants have been identified that might confer some degree of resistance to infection and mortality in COVID-19 cases[9]; however, intraspecific *ACE2* variation has not been well-explored in nonhuman primates such as chimpanzees. Population-scale studies of *ACE2* variation in primates could help evaluate the risks that primates face and illuminate potential genetic adaptations that nonhuman primates may have to protect them from respiratory viruses such as SARS-CoV-2.

PROJECT AIMS

Aim 1: Assess *ACE2* variation in a panel of captive chimpanzees (*Pan troglodytes verus,* n=15) in coding regions where human variation is also present.
Aim 2: Examine identified genetic variants for functional impacts on their encoded proteins.
Aim 3: Investigate patterns of *ACE2* protein evolution across non-human primates.

STUDY POPULATION

We used blood and tissue samples stored at the George Washington University Primate Genomics Laboratory, having been collected from captive western chimpanzees (*Pan troglodytes verus*) from captive facilities across the United States over decades by collaborators (J Ely, W Hopkins, and others), including the Emory National Primate Research Center and Keeling Center for Comparative Medicine and Research[10]. This project did not involve any animal handling or contact. Our panel comprised 15 unrelated individuals, including 2 males and 13 females to account for phasing. *ACE2* is located on the X chromosome, so we had a total of 28 X chromosomes from which to amplify the *ACE2* sequence.

METHODS

DNA extraction and quantification: DNA was extracted from the 15 blood samples using the Qiagen DNeasy Blood and Tissue Kits and automated via a QIAcube robotic workstation (QIAGEN). These samples were assessed for quality and quantity by Nanodrop spectrophotometry to determine the concentration of chimpanzee DNA.[11, 12]
***ACE2* PCR and direct sequencing:** We used a primer pair to amplify exon 9 of the ACE2 molecule, which encodes a region of the binding site that encompasses most of the receptor binding motif that is essential for SARS-CoV-2 S-protein binding[13] and has numerous destabilizing or beneficial variants in human

populations.[9] We performed custom PCR reactions to amplify this exon in each individual. We assessed each PCR product for successful amplification via gel electrophoresis and Sanger sequenced it using the forward primer (Genewiz, Plainfield, N.J., USA). Each sequence trace file was visualized and analyzed using 4Peaks to review the data for the presence of heterozygosity at the base pair level. Sequence data were aligned to the chimpanzee reference sequence to identify any variants. We then analyzed nonsynonymous variants for their functional implications on the protein produced using PolyPhen-2[14] and the BLOSUM 62 matrix.[15]

Gene protein evolution analysis: We then investigated patterns of selection on the gene *ACE2* across the entire primate clade. Sequence data were mined from the NCBI database using the human genome as the reference (e.g. via BLAST). We identified sequences from 27 primate species that had enough coverage for the analysis. Sequence data were then aligned and edited using Aliview,[16] and visually surveyed for any sequence errors or alignment artifacts. Sequences were then trimmed and refined by hand to ensure an open-reading frame and to remove any artifacts that were incompatible with the program. We created a phylogenetic tree derived from user-defined consensus trees created with Bayesian inferences of the well resolved primate phylogeny by 10ktrees.[17] We estimated site-specific dN/dS ratios across the included primates using the open-source software package HyPhy[18] on the Datamonkey webserver.[19] Using the program MEME,[20] we tested for evidence of episodic positive/diversifying selection using a p-value threshold of 0.05.

RESULTS and DISCUSSION

***ACE2* variation and functional impact:** Among the 15 individuals sequenced, the SARS-CoV-2 receptor binding motif (HEMGH) was highly conserved. We found one nonsynonymous variant (T1425C) that resulted in an amino acid change (Ile421Thr) in 3 of the 15 individuals. This variant is likely functional with a PolyPhen-2 score of 0.985 (probably damaging), which indicates that this sequence change would affect the function of the encoded protein. This variant has a BLOSUM 62 score of -1, indicating that the new amino acid has significantly different chemical properties, further supporting the PolyPhen-2 prediction. All 3 individuals with this variant were female and each had a 'healthy' or normal copy of the *ACE2* gene as well. This variant could potentially affect the binding affinity of the virus to the cell as it is close to the receptor binding motif for the S protein; however, the effects of this variant on the function of ACE2 outside of a viral receptor are not known. As all the individuals with this variant were female and heterozygous for the normal version, they might have compensated for the change of function of the ACE2 protein in some of their cells. The variant could also confer a degree of resistance of some cells that express the variant protein to the SARS-CoV-2 virus. The identification of a significant variant in a relatively small panel of individuals indicates that we need to further study this gene in both captive and wild populations to fully understand both the risk that nonhuman primates face of viral transmission and how our own evolutionary history might have led humans to be especially susceptible to this virus.

Positive selection on *ACE2* in the primate lineage: We detected 11 sites with signatures of episodic positive/diversifying selection among 27 primate species (Table 1).

Table 1 Meme analysis of site-specific positive selection in *ACE2* ($p \leq 0.05$). α = synonymous substitution rate (dS); $\beta+$ = non-synonymous substitution rate for the positive/neutral evolution component; LRT = likelihood ratio test statistic

Site	α	β+	LRT	p-value
658	4.39	539.02	12.56	0
359	0	257.07	8.03	0.01
364	0	86.95	5.53	0.03
65	0	93.17	5.1	0.04
156	0.03	113.55	4.72	0.04
218	0	77.63	5.07	0.04
728	14.44	1242.74	4.67	0.04
783	7.6	599.78	5.08	0.04
267	0	3185.73	4.57	0.04
646	0	64.06	4.56	0.05
692	0	61.84	4.3	0.05

Site 364 had a variant, 364Thr, that was specific to primates with geographical ranges in the Americas, which could indicate that the variant is specific to a pathogen. Since this relatively conserved gene in mammals shows evidence of positive selection across multiple sites, along with this geographic-specific variation, lineage-specific selective pressures may have led to these distinctions. This finding is consistent with the conclusion that the common ancestor of primates in the Americas as well as that of the Philippine tarsier was likely resistant to SARS-CoV-2, whereas the common ancestor of African and Asian monkeys and apes was not.[21]

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IPS Conservation Grant Report

Abid Ali

Population Density and Estimate of Endangered Kashmir Langur (*Semnopithecus Ajax*) from the Khyber Pakhtunkhwa Province of Pakistan

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BACKGROUND

The Kashmir langur, known as *Semnopithecus ajax* (here after; *S. ajax*), distributed across the Himalayan regions of India, Nepal, parts of Bhutan, and Pakistan [1,2]. Despite its endangered status, this species lacks research attention and conservation efforts in Pakistan.[3].In Pakistan, previously the *S. ajax* is thought to occur in Azad Jammu and Kashmir (AJK) Pakistan [4]. However, there is a complete scarcity of information on langur from the Khyber Pakhtunkhwa (KPK, here after) province of Pakistan. This gap in understanding hinders effective conservation planning for the species within the country [5].

The known habitat range of the *S. ajax*, though limited to small forest patches in Pakistan, is under severe threat due to various human activities. These include illegal logging, grazing by local communities, and the growing impact of tourism and construction projects [5,6]. Additionally, the expansion of road networks and major infrastructure projects, coupled with the effects of climate change, are further diminishing the langur's habitat. These combined pressures are pushing the species perilously close to extinction in the region [7]. Therefore, an urgent need to study the population density and abundances of this primate from the province.

Here, we provide the first-ever population density in KPK province and distribution map of the species in Pakistan, helping to inform conservation planning and management efforts.

PROJECT AIMS

Our project aimed to report the first ever national wise distribution range and density of langurs in Pakistan.

1. Distribution range and Population density of the *S. ajax* in Pakistan.
2. Raise awareness about the importance of conserving the *S. ajax* and its habitat through conservation education programs.

STUDY LOCATION AND METHODS

The study area covered two major Districts from the KPK Province such as District Mansehra, and District Kohistan, both located in Western Himalayas of Pakistan.

DATA COLLECTION AND ANALYSIS

Our research in Pakistan, conducted from September 2021 to April 2023, followed all legal requirements and received approval from the KPK Wildlife department. Using a total of 51 transects distributed across seven reserve forests, we estimated the population density of *S. ajax* based on ecological niche suitability categories. Of the 51 transects surveyed, we recorded the presence of *S. ajax* in only 38. These occurrence points were used for inclusion in the modelling. Additionally, to increase the number of records, we added 196 records from independent field sampling, so that we had 234 records. Subsequently, we filtered the database to discard incorrectly geo-referenced records, as well as duplicate and/or atypical coordinates. The remaining data were loaded into QGIS® 3.28.4 software for spatial filtering i.e., 0.5 km. We used 24 variables with a resolution of 1 km² (Table 1).

Table 1. Predictor variables employed in the ecological niche modelling of *Semnopithecus ajax* in Western Himalayas of Pakistan. They are ordered by category, variable name, abbreviation, unit, variable type and sources.

Category	Variable	Abbreviation
Bioclimatic	- Annual Mean Temperature	Bio01
	- Mean Temperature Diurnal Range (Mean of monthly)	Bio02
	- Temperature Seasonality (standard deviation ×100)	Bio04
	- Temperature Annual Range	Bio07
	- Mean Temperature of Driest Quarter	Bio09
	- Annual Precipitation	Bio12
	- Precipitation Seasonality (Coefficient of Variation)	Bio15
	- Potential Evapo - Transpiration of Wettest Quarter	PET_WQ
	- Potential Evapo - Transpiration of Driest Quarter	PET_DQ
	- Arid Index Thornthwaite	AIT
	- Thermicity Index	TI
	- Growing Degrees Day 5	GDD5
	- Maximum Temperature Coldest	Max_TC
	- Mean Temperature Coldest	Mean_TC
	- Climatic Moisture Index	CMI
Topographic	- Elevation	Elev
	- Slope	Slo
Vegetation and habitat	- Forest Height	FH
	- Tree Cover	TC
	- Tree Cover Fraction	TCF
	- Normalized Difference Vegetation Index	NDVI
	- Land Cover	LC
Anthropogenic	- Human Influence Index	HII
	- Human Footprint Index	HFI

Pearson's correlation coefficient was used to check the collinearity between these 24 predictors and discarded variables with bivariate correlation higher than ±0.80. Therefore, to run the model, we used the variables: Mean_TC, PET_WQ, Bio07, CMI, LC, HFI, Slo, NDVI and fitted MaxEnt algorithm using the *Kuenm* package in the Rstudio®[8]. We considered 10 random replicates, 500 maximum iterations, 13,000 background points. The final model was evaluated by partial ROC in addition, we validated it by the area under the curve AUC.

From the modelling, we considered estimating the population density of *S. ajax* by category of ecological niche suitability to know which best adjusts to the habitat of the species. Based on the model raster projection, and considering the threshold for reclassification, we categorized the probability of suitability into three levels: 1) Null to scanty ($P < 0.10$), 2) Low to medium ($0.10 \leq P < 0.50$), and 3) high to very high ($P \geq 0.50$). With the areas delimited by category, we grouped the records by location using the 38 field records obtained for the present study. We employed Distance® 7.3 software to

select models and assess goodness of fit, enabling us to estimate distribution range, population density, and population size.

FINDINGS

The validation of the model yelded high sensitivity in a low specificity of the ROC curve, with AUC = 0.96 ± 0.01. The variables with the highest contribution of information to the model (> 10% contribution) were Mean_TC (36.3%), PET_WQ (19.7%), Bio07 (16.1%) and CMI (13.5%). The area calculated for the category of high to very high ($P \geq 0.50$) was 474.17 km² (5.81%), from low to medium ($0.10 \leq P < 0.50$) was 1099.80 km² (13.49%), and from null to scanty suitability ($P < 0.10$) was 6,581.13 km² (80.70%). With respect to the number of records (i.e., cluster or individual) of *S. ajax* by suitability category, 23 observations were in high to very high probability, 25 in low to medium probability, and 26 in null to scanty probability. Regarding the general population density (ind km^{-2}) by niche suitability category, in the area with high to very high probability ($P \geq 0.50$) was the highest density with ~ 0.21 ind km^{-2}. We estimated a population of ~ 100 individuals. Next was a density of ~ 0.11 ind km^{-2} and an estimated population of ~ 122 individuals in the area with low to medium probability ($0.10 \leq P < 0.50$). On the other hand, the area with null to low probability ($P < 0.10$), which had the largest surface area, presented the lowest density with ~ 0.01 ind km^{-2}; with a population of ~ 91 individuals of *S. ajax*

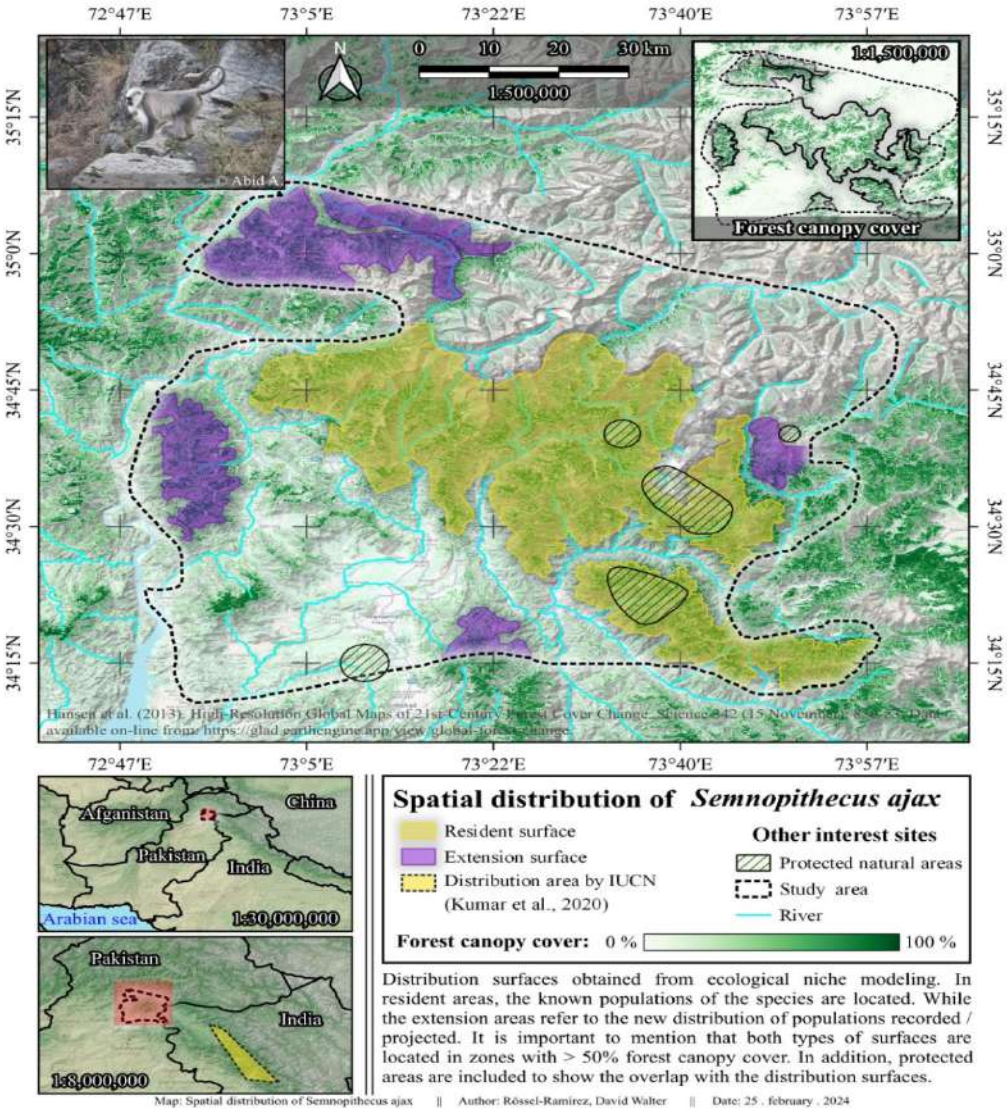

Figure 1. *Overall, the map represents the distribution range of the S. ajax across the country.*

CONSERVATION THROUGH COMMUNITY INVOLVEMENT (CCI)

We conducted about four sessions on community engagement and children's education, involving 150 community members and children's from local schools and local participant. we conducted sessions to raise awareness about the conservation status of *S. ajax* and finding ways to cope with human langur conflict. These interactive sessions utilized visual aids, storytelling, and hands-on activities to convey the crucial role of wildlife in maintaining ecological balance. Community members showed a growing inclination towards fostering coexistence with wildlife. The success of these initiatives lies in the heightened engagement and positive attitudes observed among both community members and children.

FUTURE CONCERNS

Our study on langur population density in KPK province paints a concerning picture of their endangered status. Despite being the first to report on langur populations in this region, based on preliminary data from local people, our findings reveal a troubling decline in their numbers. The expansion of agriculture, urbanization, and illegal logging are rapidly reshaping langur habitats, leading to fragmentation and loss. Langurs in Pakistan face challenges such as food scarcity and habitat degradation, exacerbated by tourism and poaching. Despite their endangered status, langurs lack adequate protection under Pakistan's conservation efforts. Urgent action is needed to address these threats and safeguard langur populations for the future.

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IPS Lawrence Jacobsen Development Grant

Lalaina Rafianinana

"Protecting Primates through education and community art in Lavavolo, Madagascar"

Conservation Fusion

Antananarivo, Madagascar

Background.

Madagascar is undeniably one of the highest priorities for biodiversity conservation due to its extreme species richness- particularly primates with 112 known species. Unfortunately, according to the IUCN, 90% of all lemur species are threatened with extinction. Education is critical to create a healthy balance between people and primates. Recognizing the urgent need for action, Conservation Fusion is engaging children and communities in education promoting a knowledge and understanding of the Madagascar's biodiversity -especially lemurs, ultimately instilling ownership and positive change.

Over the past 13 years, our team has built strong ties with the community of Lavavolo in southwest Madagascar, where four critically endangered lemur species reside. Lavavolo stands out for its dedication to wildlife protection, driven by cultural beliefs and education on biodiversity importance. However, neighboring villages pose a threat due to lack of awareness, resulting in forest habitat destruction. Support from IPS enabled capacity building among local Lavavolo leaders, empowering them to educate seven neighboring villages. Using art, education, and hands-on activities, children in Lavavolo gained knowledge about lemurs to educate peers in neighboring villages about the crucial need to protect the forest, the unique home of these lemurs.

Project Objectives

Engage children and community leaders in education and art to create conservation champions capable of expanding conservation messages and actions to peers and neighboring villages.

Specific Objectives:

Increase Knowledge of Local Lemur Species Amongst Children from Lavavolo Village:

- Conduct educational sessions to enhance understanding of local lemur species among 200 children residing in Lavavolo village.

Community Art Project:

- Facilitate a collaborative art project within the Lavavolo community to raise awareness about lemur conservation and environmental stewardship.

Environmental Education Program at the Dream School:

- Implement an environmental education curriculum to provide instruction on lemurs, one health, ecological concepts, biodiversity, and conservation principles through hands-on learning activities to reinforce classroom teachings and foster practical application of environmental knowledge.

Conservation Camp:

- Offer immersive experiences, including nature walks, wildlife observation, and interactive discussion and games to instill a sense of stewardship and responsibility towards conservation. Tree Planting - Habitat Restoration:

- Coordinate tree planting initiative within Lavavolo village to restore degraded habitats and create suitable environments for lemurs and other wildlife.

Empower Local Leaders with Community Outreach Program:

- Provide capacity-building workshops and training sessions for local leaders in Lavavolo village to equip them with the knowledge and skills necessary to conduct effective community outreach.
- Empower leaders to serve as ambassadors for lemur conservation, facilitating dialogue, and engagement with neighboring communities to promote collective action for environmental sustainability.

These specific objectives were designed to address key aspects of the conservation education program, encompassing knowledge dissemination, community engagement, and capacity building to foster long-term conservation impact in the Lavavolo village and surrounding areas.

Location

Lavavolo is a rural village comprised of approximately 77 households in close proximity to the Lavavolo Classified Forest in the Toliara region of southwestern Madagascar. The region consists primarily of dry, spiny forest and indicative of xeric conditions with little annual rainfall. The study area is situated along the southern coast, and includes one of two regions, the Mahafaly Plateau.

Key Results and Outcomes.

1. **Significant Increase in Awareness and Knowledge**: Evaluations conducted after the initial environmental education lesson in August 2023, involving 69 children, showed a remarkable 162% increase in awareness, knowledge, and intentions to support primate conservation. Children demonstrated improved understanding of various aspects related to lemurs, including habitat, behaviors, diet, and reasons for protecting them.

Enhanced Learning through Interactive Teaching Methods: The educational session utilized interactive teaching methods, such as visual aids, stuffed animal demonstrations, and student-led presentations to engage children effectively. This approach facilitated better comprehension and retention of information about lemurs and their conservation resulting in expanded knowledge base. Prior to the lesson, children could identify only 8 lemur-related ideas, including species names, habitats, and behaviors. After the session, children demonstrated a significant increase in their knowledge base, identifying a total of 21 lemur-related ideas, including all four local species names, various habitats, behaviors, diets, and actions humans can take to protect lemurs.

2. **Active Paritcipaiton and Engagement**: 83 Children actively participated in various activities, such as creating lemur puppets, reading lemur storybooks, singing songs about lemurs, and conducting drawing evaluations. These hands-on experiences fostered one-on-one engagement and enriched their learning experience.
3. **Confirmation of Understanding**: Student-led performances and drawing evaluations served as effective forms of evaluation, confirming that children had grasped the information conveyed during the lesson. This feedback mechanism ensured that the educational objectives were met and that the information was well-understood by the students.
4. **Community Engagement through Art:** The installation of a lemur-inspired mural at the Dream School fostered strong connections to nature within the Lavavolo community. Participation in the mural painting process involved community members of all ages, leading to discussions about local lemur species and exploration of biodiversity across Madagascar. The mural's strategic location ensured daily exposure to its message, serving as a constant reminder of the importance of lemur conservation and environmental stewardship.
5. **Gender Equality and Leadership Empowerment:** The selection of 10 community leaders, comprising an equal gender ratio of 5 men and 5 women, for participation in the lemur training and outreach program marked a significant shift in cultural norms. Traditionally, women in the region are not allowed to assume leadership roles. The appointed leaders demonstrated initiative and commitment by expanding the scope of outreach activities beyond the initially specified village of Itampolo to an additional 7 villages. Their proactive approach underscores the potential for inclusive leadership to drive meaningful conservation efforts and community engagement across multiple localities, thus amplifying the program's impact on lemur conservation beyond Lavavolo.

Figure 1:
Discussion: Implications of Project

The project's implications extend beyond the immediate outcomes observed within Lavavolo village, highlighting broader implications for conservation initiatives in similar contexts. By creating relevant educational tools and empowering local leaders to assume responsibility for promoting primate protection and habitat conservation, the project exemplifies a community- driven approach to environmental stewardship. This approach emphasizes the importance of collaboration, teamwork, and strong communication channels in fostering collective action towards conservation goals.

One key lesson learned is the significance of cultural sensitivity and inclusivity in conservation efforts. Effective engagement with local communities necessitates a willingness to ask questions, actively listen, and respect cultural differences and local nuances. By incorporating diverse perspectives and traditional knowledge into conservation solutions, initiatives are more likely to resonate with communities and garner long-term support.
Furthermore, the project underscores the transformative power of education in driving positive change. By increasing knowledge levels and promoting understanding of the importance of primate conservation, education serves as a catalyst for action. Empowered with knowledge, community members are better equipped to make informed decisions and take proactive steps towards protecting primates and their habitats.
In conclusion, the project's emphasis on community engagement, capacity building, and education underscores the potential for grassroots initiatives to catalyze meaningful conservation outcomes. By fostering partnerships, embracing cultural diversity, and prioritizing education, conservation efforts can inspire local ac=on and contribute to the sustainable management of natural resources for generations to come.

Acknowledgements.
Special thanks to the Lavavolo community. We also wish to acknowledge the Madagascar Biodiversity Partnership for logistical and infrastructure support. We are grateful for additional financial support from Blank Park Zoo and re:wild. Thank you to the International Primatological Society for their both financial support in addition to valuable advice and encouragement for our project proposal.

IPS Research Grant Report

Abigail Morris

Influence of vegetation composition on male and female gorilla dispersal and population genetic structure in Lac Tele, Republic of Congo

Abigail Morris, University of California, Davis

Introduction

Dispersal is an important life history trait, playing a key role in genetic and cultural evolution. This is especially true in group-living social animals, where both social and ecological factors interact. Habitat characteristics can constrain dispersal, restricting gene flow and shaping the genetic and social structure of a population. Using population genetics as a tool, this project investigates the ecological factors that shape dispersal patterns in male and female western lowland gorillas (Gorilla gorilla gorilla).

Project objectives

The goals of this pilot study were to determine the feasibility of my proposed dissertation work. This project aims to build upon our knowledge of primate dispersal, as it investigates different ecological forces (distance and environment) that drive female gorilla dispersal, and the impacts of this interaction on overall population genetic structure. Female culturally inherited preferences for a particular food plant species can reduce gene flow between habitat types, enhancing the potential for local adaptation. The Lac Télé region of Republic of Congo includes two habitat types separated by a sharp boundary, or ecotone. This ideal system allows me to independently measure the effects of distance and habitat differences on dispersal decisions. This study thus allows for more exploration into the causes and effects of cultural transmission of behavior and diet, providing a mechanism through which future projects can assess the development of local genetic and behavioral adaptations.

The overarching goal of this project is to disentangle the primary ecological drivers of gorilla dispersal decisions and evaluate the impact of these behaviors on overall population genetic structure. The information learned through this research will inform conservation efforts in the region and provide researchers with more insight on dispersal patterns and subsequent gene flow within a highly heterogeneous landscape. The aim of this pilot study was to validate methods for continued research and analysis in future field seasons. Its primary objectives are as follows:

Objective 1: Assess dietary differences between gorillas ranging in swamp and terra firma habitats to understand whether female habitat-specific dietary preference exists.

Objective 2: Distinguish between the roles of distance and habitat type on female dispersal decisions. Assess the multigenerational consequences of habitat-specific female dispersal decisions on the gorilla population genetic structure.

Study Location and Methods

The Lac Telé region of Republic of Congo includes two habitat types separated by a sharp boundary, or ecotone. On one side is an elevated terra firma forest, and on the other is a much larger swamp forest. Each habitat encompasses over 350 km^2, large enough to host numerous gorilla groups (avg. group home range: 14km^2)[1]. Plant composition differs between swamp and terra firma, and gorilla diets reflect this difference[2]. Because of the sharp ecotone, given a fixed dispersal

distance, two gorillas could end up in different habitats. This ideal system allows me to independently measure the effects of distance and habitat differences on dispersal decisions.

During the month spent in the forest, I succeeded in collecting 93 gorilla fecal samples. Each sample was stored in 98% ethanol for 24-48 hours before it was transferred onto silica. I photographed each sample and noted each sample name, along with its GPS location, age, and any special characteristics. GPS waypoints were taken at each unique site and were plotted on the map below (Figure 1). Data was collected at every nest site, in which I recorded the GPS location, number of nests, plant material used for each nest, the height of each nest, and amount of canopy cover above each nest.

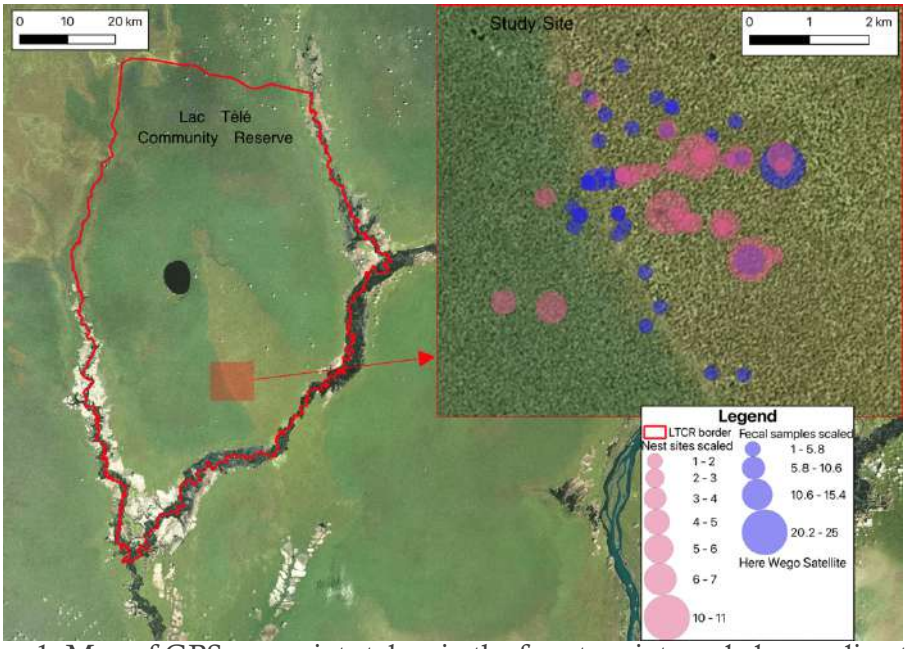

Figure 1: Map of GPS waypoints taken in the forest, points scaled according to number of nest sites and samples found in each location.

Main Findings and Next Steps

The high volume of gorilla fecal samples found and collected in the Lac Télé Community Reserve indicates a very high density of gorillas within this forest. I measured bolus diameter of each sample that fit the criteria (many samples were flattened or diarrhea, so accurate measurements were sometimes impossible to obtain). I created a histogram of these measurements (Figure 2) and compared them to published data to estimate the ages of the individuals that deposited each fecal sample.

While back in the United States, I will begin genetic extraction from the fecal samples collected in the Lac Télé Community Reserve. DNA extracts will be genotyped at 16 autosomal microsatellite loci in replicate to ensure accuracy. To complement these biparental markers, mitochondrial DNA sequences of the control region and 10 microsatellite loci of the Y chromosome (uniparental matrilineal, and holandric datasets) will also be generated. Sex of each individual will be determined with an amelogenin assay. To address habitat-specific diets, I will combine macroscopic and molecular dietary analyses as complementary and confirmatory tests, using DNA metabarcoding for the chloroplast trnL locus.

Additionally, I will apply for more research grants in order to undergo a second field season in the Lac Télé Community Reserve, during which I plan to spend 2-3 months in the forest gathering more fecal samples and thus increasing my current sample size.

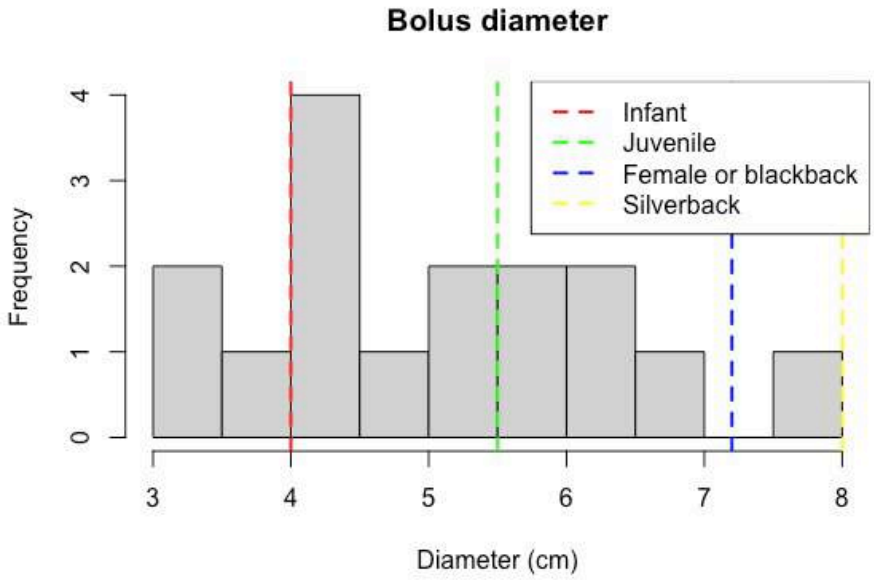

Figure 2: Histogram of bolus diameters of gorilla fecal samples. Dashed lines indicate age range estimates based on bolus diameter, as according to the literature[3].

Implications of Project

This pilot study succeeded in showing the feasibility of my overall dissertation proposal. This project will further our understanding of dispersal in social animals. Habitat-specific dispersal of females likely results in genetically distinct habitat-specific subpopulations, or ecological clusters, of gorillas. Behavioral variation within these clusters can influence genetic structure, with implications for population viability. This allows for more exploration into causes and effects of cultural transmission of behavior and diet, providing a mechanism through which future projects can assess the development of local genetic and behavioral adaptations. This research will serve as the first chapter of my thesis and will be expanded upon through two subsequent chapters focused on habitat-specific diet and anthropogenic barriers to dispersal and its impacts on population genetic structure, respectively.

Acknowledgements

The success of this pilot study would not have been possible without the aid of the IPS Research Grant. Additionally, I would like to thank my professor, Dr. Damien Caillaud, and my colleague, Alice Michel, for their help adjusting research methods in the field. Our Congolese guides proved invaluable in the forest, locating and tracking gorilla groups and assisting with data collection. I mentored a Congolese Masters student, Jeancy, whose project on effects of habitat vegetation differences on gorilla diet and nest construction will help further my own research as well as any other future projects in this region. I thank him for his dedication to this work and gorilla conservation as a whole.

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IPS Conservation Grant Report

Bradley Christin

Primate Density and Hunting Along an Ecotone in northern Republic of the Congo

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INTRODUCTION

Ecotones harbor a high diversity of animal species by providing a blend of distinctive and shared resources found in neighboring ecosystems[1,2]. However, these wildlife hotspots also attract hunters, especially in rural areas with limited livelihood options, such as the Congo Basin. The resulting variations in terrain, vegetation, and hunting pressures generate complex interactions that influence primate richness and abundance on either side of an ecotone. A prime example can be seen in the Lac Télé Community Reserve (LTCR), Republic of the Congo. Here, the abrupt transition between swamp forest and terra firma forest plays a pivotal role in shaping primate populations and their vulnerability to hunting.

The LTCR (4,400 km²) is a vital segment of the world's largest swamp forest, holding one of the highest documented densities of critically endangered western lowland gorillas (~13,221 individuals, 2.9/km²), large numbers of endangered central chimpanzees (~3,189 individuals, 0.7/km²), and an abundance of other primate species with varying levels of legal protection[3]. Primate vulnerability peaks near the swamp forest-terra firma ecotone. Here, high concentrations of animals are connected to hunters by a trail that runs north-south along the ecotone, starting from the village of Impongui at the southern end. Hunting is likely more intense in terra firma habitat because of its solid terrain[3], resulting in uneven hunting pressures and a source-sink dynamic, where primates from the swamp forest migrate to the terra firma forest. Although the 27 communities in the LTCR (~20,000 inhabitants) depends mostly on fish for protein (91%), the overexploitation of fish stocks is increasing reliance on bushmeat, heightening primate hunting pressures[4,5]. Therefore, this study aims to investigate hunting pressures and primate responses near a swamp forest-terra firma ecotone.

OBJECTIVES

This study collected baseline data on primate and wildlife densities in relation to: 1) indirect hunting indices, 2) distance from the village of Impongui, and 3) proximity to the ecotone. The findings aim to identify species most affected by hunting, assess predation risks, and reveal population dynamics and source-sink relationships in these habitats.

METHODS

This study included 16 transects, each 1.5 km long, arranged in 8 adjacent pairs meeting at the ecotone (Figure 1). Transects ran perpendicular to the swamp-terra firma ecotone, oriented south to north, with Impongui as the southern reference point. A camera trap was placed at the center of each transect in an open, visible area along the transect line.

For primate groups, I used a rangefinder and compass to measure distance, height, and angle from the transect, recording GPS coordinates, and species when possible. For indirect animal signs (e.g., tracks, dung, foraging evidence) and hunting signs (e.g., traps, cartridges), I recorded GPS locations (~5 m accuracy) and measured perpendicular distance from the transect with DBH tape. For great ape nest sites, I measured the perpendicular distance of each individual nest to the transect line. In dense vegetation, I visually estimated distances. Signs within 1 m of the transect were rounded to 0. Due to the high density of bushpig and duiker signs, data were recorded only if encounters were over 50 m apart, while all encounters for lower-density species like great apes were included.

Data were analyzed in R (version 4.3.2). The relationship between animal signs and distance to Impongui was assessed using Spearman rank correlation tests. Encounter rates between transects closer to and farther from the ecotone were compared with a paired t-test. Differences in animal sign counts between habitat types were examined using a Wilcoxon rank sum test. This study received permits from the Agence Congolaise de la Faune et des Aires Protégées, Institut National de Recherche Forestière, Wildlife Conservation Society, and La Reserve Communautaire du Lac Télé (Figure 2). My research adhered to strict ethical standards, ensuring the respectful treatment of all participants involved.

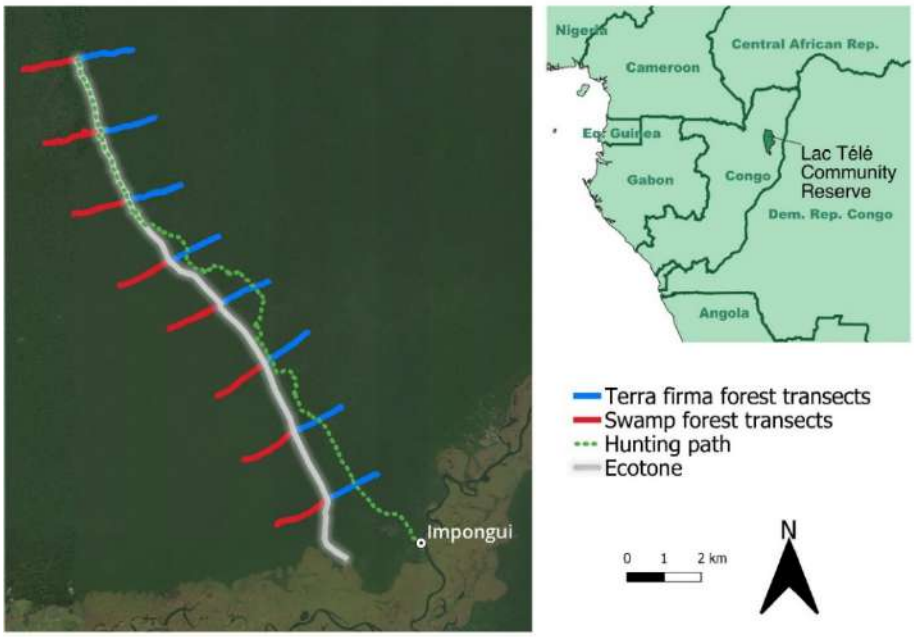

Figure 1. Map of the 16 transects carried out in this study.

Figure 2. Landscape along the Likouala-aux-herbes river in the Lac Télé Community Reserve.

KEY RESULTS AND DISCUSSION

This preliminary study marks the first effort to conduct a transect survey of all terrestrial wildlife in the LTCR. Most animal signs were from ungulates (bushpigs, duikers, and sitatungas: 50.0%) and primates (gorillas, chimpanzees, and monkeys: 37.6%), with small mammals (mongoose and allies, pangolins, African brush-tailed porcupine, rats: 8.5%) and other taxa (elephants, golden cats, crocodiles, hornbills: 3.9%) contributing less. I recorded 72 great ape nest sites (Figure 3), including 61 gorilla nest sites (259 nests) and 11 chimpanzee nest sites (40 nests). In the terra firma forest, I found 5 times as many great ape nests (11.54 nests/km) as Poulsen and Clark's 2004 study in the LTCR (2.31 nests/km). The opposite pattern was found in the swamp forest (this study: 0.91 nests/km, Poulsen and Clark, 2004: 13.06 nests/km). These differences may result from underestimation in Poulsen and Clark's terra firma transects, though transect placement was not shown in their study.

Figure 3. Gorilla nest built with a plant from the Marantaceae family.

No evidence of hunting was found on the 16 transects or hunting trail. Although four hunting camps were found in between transect lines roughly 4, 5, 9, and 14 km from Impongui, still no hunting evidence was found. This is likely because the study occurred outside peak hunting seasons (July and November), which align with high-water periods of limited fishing (Bondeko et al., 2024). Despite this, Bondeko et al. (2024) estimated that 269 hunters in the LTCR harvest 166.8 tons of bushmeat annually, likely impacting animal distribution and density.

A strong positive correlation was found between the number of great ape nests in the terra firma forest and the distance from Impongui (Spearman rank correlation test: $\rho = 0.905$, $p = 0.0046$; Figure 4). Great ape signs more than doubled between 10 km and 12 km in the terra firma forest, possibly suggesting evidence of hunting. Indeed, hunting and trapping activities are primarily concentrated within 10 km of the nearest village, as reported in Gabon[7], Equatorial Guinea[8], and Cameroon[9]. There was no significant correlation between the number of monkey signs and distance from Impongui in terra firma forest ($\rho = 0.344$, $p = 0.4029$) nor in the swamp forest ($\rho = 0.6988$, $p = 0.05379$); however, this is likely due to a small sample size.

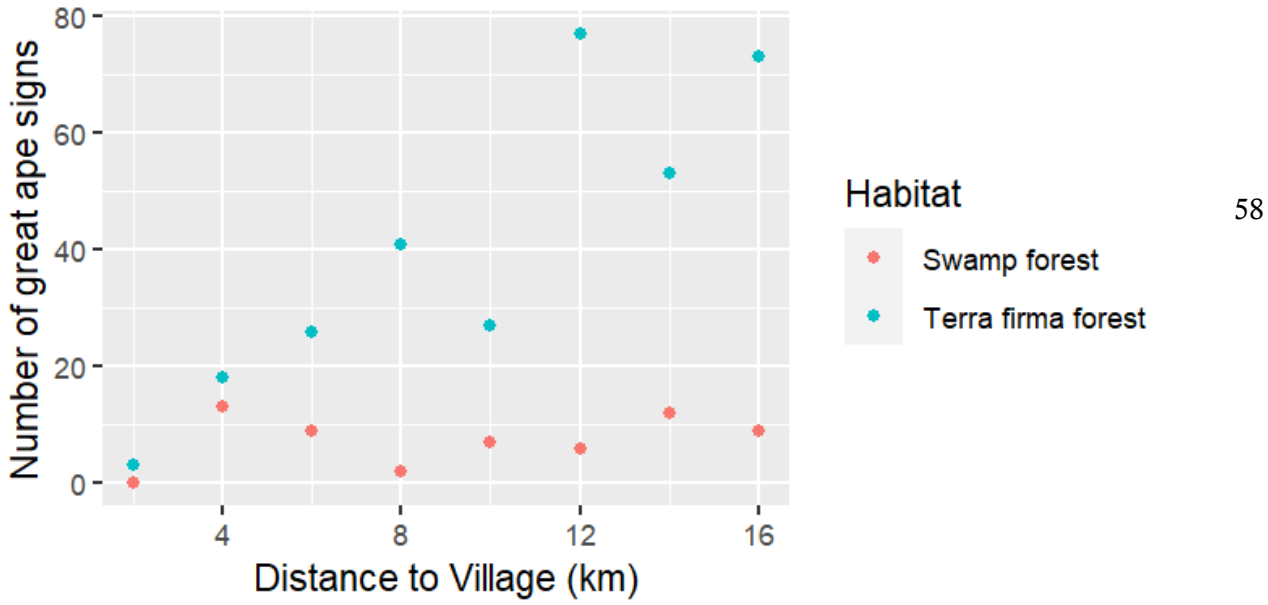

Figure 4. Relationship between the number of great ape signs in relation to the distance from the village of Impongui.

In the terra firma forest, significantly more animal signs were recorded in the half of the transect closest to the ecotone than the other half farther away (Paired t test: t = 3.5839, df = 7, p-value = 0.0089). In contrast, no significant difference was observed in the swamp forest (Paired t test: t = □ 0.41, df = 7, p = 0.6938). For gorillas, these results align with previous studies in the LTCR, where gorilla nests were rarely found within 1 km of the ecotone, with only one nesting site within 2 km[10]. Overall, wildlife encounter rates were significantly higher in terra firma forest compared to swamp forest (Wilcoxon signed rank test: V = 36, p = 0.014), with 304 signs recorded in terra firma versus 180 in swamp forest. Future research should extend transects farther from the ecotone and nearest village, include data from more villages, and use the marked-nest method to estimate great ape density more accurately.

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