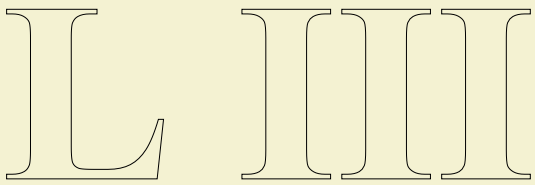
LEOPOLD III-FONDS VOOR NATUURONDERZOEK EN NATUURBEHOUD

FONDS LEOPOLD III POUR L'EXPLORATION ET LA CONSERVATION DE LA NATURE



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1. Toelagen voor veldwerk buiten Europa
Subsides pour missions de terrain hors de l'Europe

Voor het dienstjaar 2023 heeft het Leopold III-Fonds acht toelagen verleend voor terreinzendingen buiten Europa. De ingekorte versies van de zendingsverslagen worden hierna weergegeven.

Au cours de l'exercice 2023, le Fonds Léopold III a octroyé huit subsides pour des missions de terrain hors de l'Europe. Les versions raccourcies des rapports de mission sont reprises ci-après.

1.1. BAYAT, Manon (PhD student Un. Libre de Bruxelles) & DELLEUZE, Mélanie (Doctorat en cotutelle Un. Libre de Bruxelles et Université de Chili)

Plasticité trophique et variabilité du microbiome digestif d'oursins du genre *Abatus*.
Mission au Chili, 1 – 29 avril 2023.

1. Introduction, cadre et rappel des objectifs

Compte tenu de la vitesse et de l'ampleur des changements climatiques, notamment dans l'Océan Austral, des conséquences majeures sont attendues sur les écosystèmes. En milieu marin côtier, ces changements mènent à des modifications dans la structure des communautés benthiques, dans l'abondance et la disponibilité de la nourriture, ou encore dans les interactions proies-prédateurs. De ce fait, les chaines alimentaires de ces communautés et les maillons qui les caractérisent en seront perturbés.

Étudier les flux d'énergie des organismes via deux processus inhérents à ces flux, c'est-à-dire l'étude de leur plasticité trophique (dont le régime alimentaire) et l'étude de leur microbiome digestif (paramètre important pour la santé et adaptation de l'hôte, notamment via la digestion et assimilation des nutriments) est aujourd'hui important dans un cadre de changements globaux et d'adaptation à ces changements, et attire de plus en plus l'intérêt de la communauté scientifique.

La péninsule Antarctique, les îles subantarctiques et la Patagonie, sont encore peu considérées pour l'étude des flux d'énergie. Ces régions peuvent cependant être considérées comme de véritables « laboratoires naturels » dans ce contexte de changements environnementaux rapides car elles subissent peu de pressions anthropiques d'autres natures (e.g. urbanisation, pollution des eaux). Elles sont néanmoins en première ligne en ce qui concerne les changements climatiques et notamment l'augmentation des températures.

Élargir l'étude trophique et microbienne des échinides dans ces régions, tel que l'avait initiée Mélanie lors de son mémoire, est donc une bonne opportunité pour améliorer nos connaissances sur les effets de variations environnementales sur les communautés benthiques côtières.
Ce projet a donc pour but de poursuivre les études de caractérisation du microbiome intestinal (et de ses variations) d'oursins du genre *Abatus* échantillonnés au Chili (réalisées par Mélanie dans le cadre de son mémoire), mais également d'initier l'investigation de leur plasticité trophique. *In fine*, une estimation des flux énergétiques inhérents à ces oursins, et leur potentielles variations dans un contexte de changements climatiques sera établie (similaire à un projet de recherche déjà initié par Manon, en Antarctique).

Ce présent projet, pour lequel la demande de subside avait fait objet, ne concernait qu'une partie de nos thèses respectives, c'est-à-dire la partie réalisée en Patagonie. Celui-ci s'inscrit cependant dans une étude plus large, incluant des collaborations avec l'Université de Magellan à Punta Arenas et l'Université du Chili à Santiago.

Il possède quatre axes de recherches :

1. Investiguer la plasticité trophique d'oursins du genre *Abatus,*
2. Estimer la variabilité taxonomique et fonctionnelle du microbiome chez *Abatus*,
3. Intégrer des données environnementales récoltées lors de la collecte d'oursins dans des régions contrastées, incluant la Péninsule Antarctique, les îles subantarctiques, et la Patagonie afin d'étudier les liens unissant la plasticité trophique, le microbiome intestinal et les conditions environnementales chez *Abatus*,
4. Investiguer en laboratoire (aquarium) les facteurs expliquant la variabilité du microbiome, tels que la température, permettant *in fine* de prédire les réponses du microbiome face à des perturbations.

2. Matériel et méthodes

Étude observationnelle

L'expédition a eu lieu à proximité de l'Université de Magellan à Punta Arenas (Chili), où le matériel nécessaire au traitement des échantillons (dissection-préservation-extraction d'ADN) a été mis à disposition au sein du laboratoire de biologie moléculaire du LmAS (Laboratoire des écosystèmes marins antarctiques et subantarctiques), représenté par la docteur Karin Gérard et le docteur Andrés Mansilla.

L'échantillonnage a eu lieu dans la Baie Possession en Patagonie atlantique (Fig. 1), où le site se caractérise par des forts courants de marée semi-diurnes et des vents puissants. Au total 80 individus adultes *Abatus cavernosus* (Figure 2) ont été échantillonnés à pied à marée basse. Les individus ont été

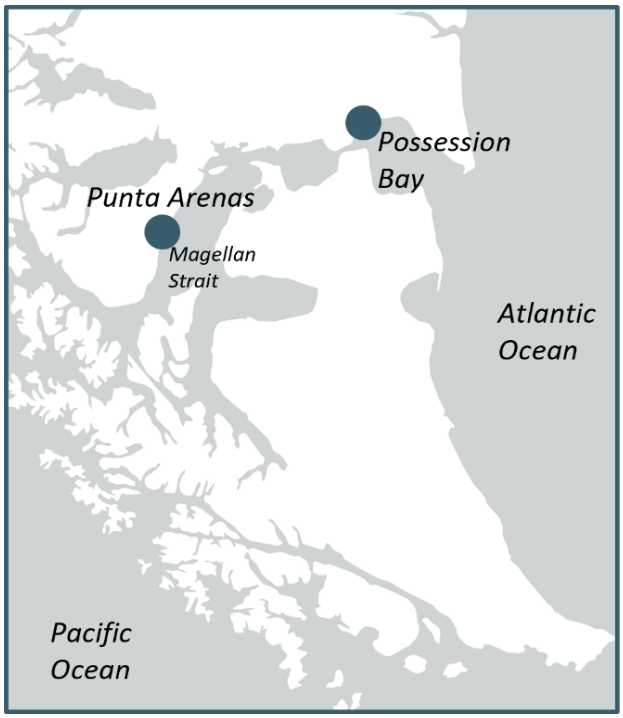

Fig. 1. Localisation du site d'échantillonnage de *A. cavernosus* dans la Baie Possession en Patagonie Atlantique

immédiatement stockés dans un réservoir contenant de l'eau de mer et du sédiment *in situ* à 4ºC jusqu'à leur dissection. Du sédiment superficiel et de l'eau de mer (15L) provenant de l'environnement immédiat des oursins ont également été collectés et stockés à 4ºC jusqu'à leur traitement en laboratoire.

Fig. 2. Représentation *d'Abatus cavernosus*

Une fois dans les dépendances du laboratoire de biologie moléculaire du LmAS (UMag), entre 2 et 12 heures après l'échantillonnage, 20 individus ont été sexés, mesurés et disséqués dans des conditions stériles afin de collecter les pelotes digestives (*i.e.*, agrégation sphérique de digesta des oursins), l'épithélium intestinal, les gonades et les piquants. Les échantillons de pelotes digestives et d'épithélium intestinal ont été préservés dans du RNAlater tandis que les échantillons de gonades et de piquants ont été préservés dans l'éthanol. Le sédiment collecté sur le terrain a été homogénéisé et séparé en 6 réplicats. L'eau de mer (2,5 L) a été filtrée en suivant une filtration directe avec un filtre de 3µm, total de 5 réplicats. Tous les échantillons ont été conservés à -20ºC jusqu'à l'extraction d'ADN.

Les extractions d'ADN et les PCRs amplifiant la région V4-V5 du gène ribosomal 16S, utilisée pour la partie microbiome, des échantillons de pelotes digestives et d'épithélium intestinal ont été réalisées sur place, où le taux de réussite de ces amplifications est de quasi 100%.

Les PCRs amplifiant un fragment du gène codant pour une sous-unité de la cytochrome oxidase (COI), utilisée pour estimer le régime alimentaire, sont en cours de réalisation dans notre laboratoire à l'ULB. Les mesures d'isotopes stables, permettant l'étude de la niche trophique, se feront dans les prochains mois à l'Université de Liège.

Étude expérimentale

La partie expérimentale de ce projet vise à étudier comment le microbiote intestinal d'*Abatus cavernosus* répond aux variations de température, tant d'un point de vue taxonomique comme fonctionnel. Le design expérimental comprend 5 traitements de températures : 6ºC (température contrôle, moyenne annuelle du site), 10ºC (température moyenne en été), 14ºC, 18ºC et 22ºC (températures plus élevées, représentant des températures de vagues de chaleur). Chaque traitement est constitué de 10 réplicas individuels dans des aquariums contenant de l'eau de mer du détroit de Magellan et du sédiment du site d'échantillonnage.

Le reste des individus collectés (n=60) dans la Baie Possession ont été mis dans des aquariums à 6ºC dans les dépendances du Centre IDEAL à Punta Arenas. Les individus ont été acclimatés pendant deux semaines à 6ºC, ensuite la température a été progressivement augmenté (4ºC/jour), une fois atteinte la température de chaque traitement les individus ont été placé dans des aquariums individuels afin de les maintenir à température constante pendant 6 jours (Figure 3). L'échantillonnage des individus s'est

effectué lors du 6ème jour d'exposition à la température de chaque traitement et après 6 jours de résilience à 6ºC (Figure 3). Avant leurs dissections, les individus ont été mis dans des chambres respirométriques pour mesurer leur consommation d'oxygène et avoir accès à l'état physiologique de l'hôte. Comme pour l'échantillonnage *in situ* les individus ont été sexés, mesurés, pesés et disséqués dans des conditions stériles afin de collecter les pelotes digestives, l'épithélium intestinal, les gonades et les piquants. Ces échantillons vont être conservés à -80ºC pour mieux préserver l'ARN. Dû à des complications logistiques, cette expérience est toujours en cours de réalisation par Mélanie dans le cadre de sa thèse.

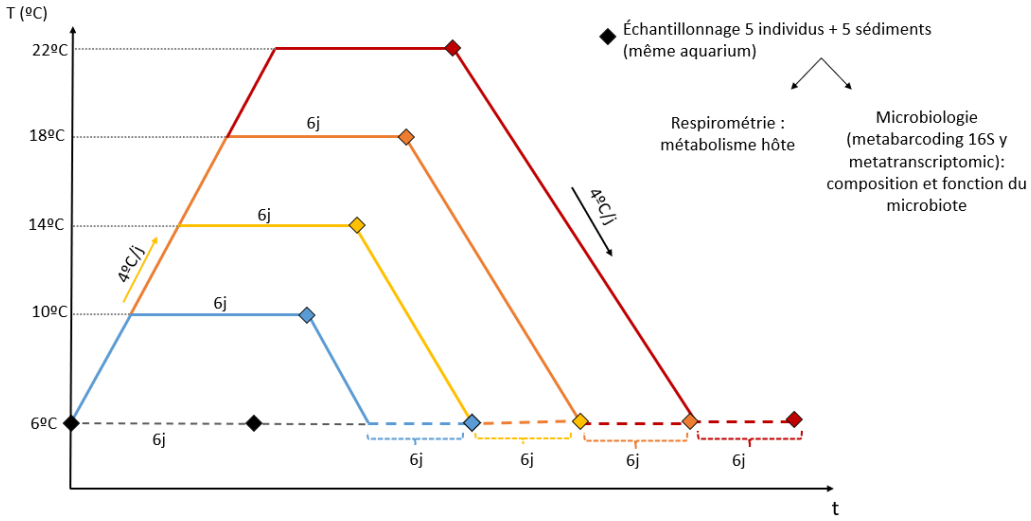

Fig. 3. Schèma du design expérimental de l'étude expérimentale menée chez *A. cavernosus*.

Utilisation des résultats et Perspectives

Les analyses génétiques et d'isotopie n'étant pas encore finalisées, les premiers résultats ne seront obtenus que dans quelques semaines à quelques mois.

Les résultats des analyses génétiques (metabarcoding 16S) réalisées sur les pelotes digestives et l'épithélium intestinal permettront d'obtenir des premières connaissances sur le microbiome propre à chacune de ces structures, et ses potentielles variations.

Les résultats du metabarcoding utilisant le marqueur COI permettront d'obtenir un aperçu du régime alimentaire de ces oursins, et les isotopes stables permettront quant à eux d'estimer leur niche trophique. Considérés ensemble, ces différents résultats permettront d'étudier les potentielles corrélations entre le régime alimentaire, la niche trophique, le microbiome et les conditions environnementales de l'habitat des oursins.

L'étude expérimentale étant toujours en cours, l'analyse de ces échantillons ne se fera que lors du retour de Mélanie au laboratoire d'Ecologie Moléculaire de L'Université du Chili. L'analyse de ces échantillons comprends l'extraction d'ADN, et l'amplification par PCR du gène ribosomial 16S, complété par une approche de métatranscriptomique qui doit encore être mis au point pour avoir accès à la composition fonctionnelle.

Ces différents résultats seront analysés dans le cadre de nos thèses respectives et seront inclus dans nos publications futures discutant de la plasticité trophique de différentes espèces d'échinides subantarctiques, de la variabilité du microbiote selon les espèces et l'environnement et de la variabilité du microbiote en fonction de la température.

1.2. DE KESEL, Wim (PhD student UAntwerpen)
Arbovirus sylvatic cycle in mosquitoes and rodents in an urban-rural environment in Morogoro, Tanzania.
Mission to Tanzania, 1 May – 5 June 2023.

Introduction
Over the past decades, the number of arthropod-borne virus (arboviruses) outbreaks increased worldwide. To date, little is known about the sylvatic cycle of these viruses, especially in Africa. We are interested in the arthropod borne viruses (arboviruses) that are transmitted via mosquitoes by blood feeding on mammalian hosts. Our research hypothesizes that small mammals, such as rodents, could function as an important amplifying host for arboviruses. Previous studies have already found that small mammals can harbor arboviruses so the question arises what role they might play in the sylvatic cycle, both in anthropogenic and natural habitats.

To answer this question, we set out to perform a field sampling of both mosquitoes and rodents in an urban rural gradient in Morogoro, Tanzania. This mission was performed in collaboration with the Institute of Pest Management (IPM) at the Sokoine University of Agriculture (SUA). This due to our longstanding relation and collaboration with IPM, they are also very experienced in rodent capturing and handling.

The objectives of this field mission were to: establish the natural feeding behavior of hematophagous arthropods and their role as arbovirus vectors in a human dominated landscape; characterize arbovirus diversity in rodents in the same human dominated landscape. We captured almost 5898 mosquitoes of which almost 65% were female, of those little over 7% had a visible bloodmeal or eggs in their abdomen.

We estimate the overall species diversity to be at least seven distinct species belonging to four genera. For the rodents we captured ten individuals with three distinct species each belonging to a different genus. Additionally, four shrews were captured. We are currently still in the process of analyzing the data and the molecular work to verify the morphological identifications and the screening of bloodmeals and arboviruses is planned in the coming months.

Progress of mission
The sampling was performed in the municipality of Morogoro on four locations, each location was chosen based on its position according to an urban-rural gradient. All locations were sampled at the same time and traps were set for four consecutive days.

Rodents were trapped using Sherman Life traps baited with a mixture of peanut butter and wheat flower of which a small quantity (pea size ball) was added in each trap. The rodent traps were placed in straight lines of 20 traps with approximately five meters between individual traps. Due to other ongoing projects the number of available traps was not consistent for each trapping session. The number of available traps varied between 80 and 200 traps.

Mosquitoes were captured using BG-Pro traps, which are battery power suction traps, baited with a sugar-yeast mixture that produces carbon dioxide. Traps were set either in the morning or in the afternoon of the first day, the second and third day the traps were emptied and rebaited around the same time and the fourth day traps were emptied.

All captured animals were brought back to the lab at the IPM for processing. Rodents were weighted, measured, sexed and then humanly euthanized via inhalation of dimethyl ether. Once euthanized the specimen was dissected and the following tissue samples were taken: whole blood on filter paper, serum (extracted from whole blood), feces, lung, liver, kidney, spleen, eye, and the complete digestive tract. For each rodent all the following collection information was recorded: date, location code, identification

number, genus, species, sex, sexual condition, weight, head-body length, tail length, ear length, hindfoot length, storage number of filter paper and remarks. All rodent samples were stored in a -20°C freezer at the IPM and are current being stored at the Univ. of Antwerp in a -80°C freezer in the EVECO lab.

The captured mosquitoes were either euthanized via inhalation of dimethyl ether or via placing the capture bag in a -20°C freezer for 10 minutes. Afterwards, mosquitoes were morphologically identified under a stereo microscope to sex, genus and if possible, to a species level.

Female mosquitoes were stored in individual wells in 200µL of DNA/RNA shield within a 96 well plate. Males were stored dry in an Eppendorf tube with maximum 20 individuals from the same genus/species, from the same location from the same day. For every mosquito, all collection information was recorded: date, location code, trap number, mosquito number, sex, genus, species, blood fed (Y/N/Eggs), storage identification and remarks. All mosquitoes were stored in a -20°C freezer and are currently stored in a -80°C freezer at the University of Antwerp in the EVECO lab.

Results

We had a total of five trapping periods, one period was a one day trapping (24h) the other four periods were four days of trapping (three consecutive nights, 3x 24h). This accounts to 312 hours of trapping rodents and mosquitoes.

In total we captured ten rodents and four *Crocidura spp*. over the entire trapping period (Fig. 1). The captured rodent species were: two *Cricetomys ansorgei*, two *Mus musculus* and six *Mastomys natalensis*. We were not able to morphologically identified the *Crocidura spp*.to a species level on location. For all species we captured equal amounts of male and female individuals, except for *Mus musculus* were both individuals were female.

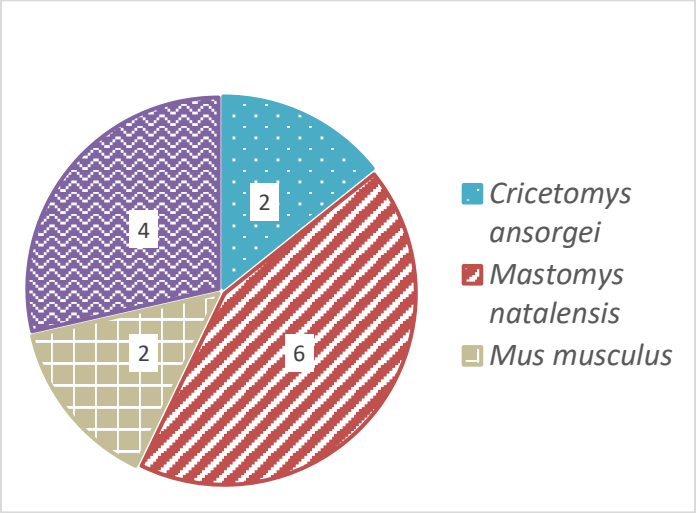

Fig. 1: Number of caught rodent and shrews species.

We captured 5898 mosquitoes, belonging to five genera: 344 *Aedes,* nine *Anopheles*, 5465 *Culex*, 28 *Mansonia*, six *Toxorhynchites* and 46 unknown specimens (Fig. 2).

Morphological identification to the species level was not always possible, but we estimate a minimum of seven distinct species. 3869 specimens were identified as female, 1961 as male and 68 as unknown. Of those females, 176 had a bloodmeal and 100 had visible eggs in their abdomen. In the coming months, a selection of specimens will be DNA-barcoded to confirm the species identifications, afterwards pools of females from the same species will be made to screen for the presence of arthropod borne viruses.

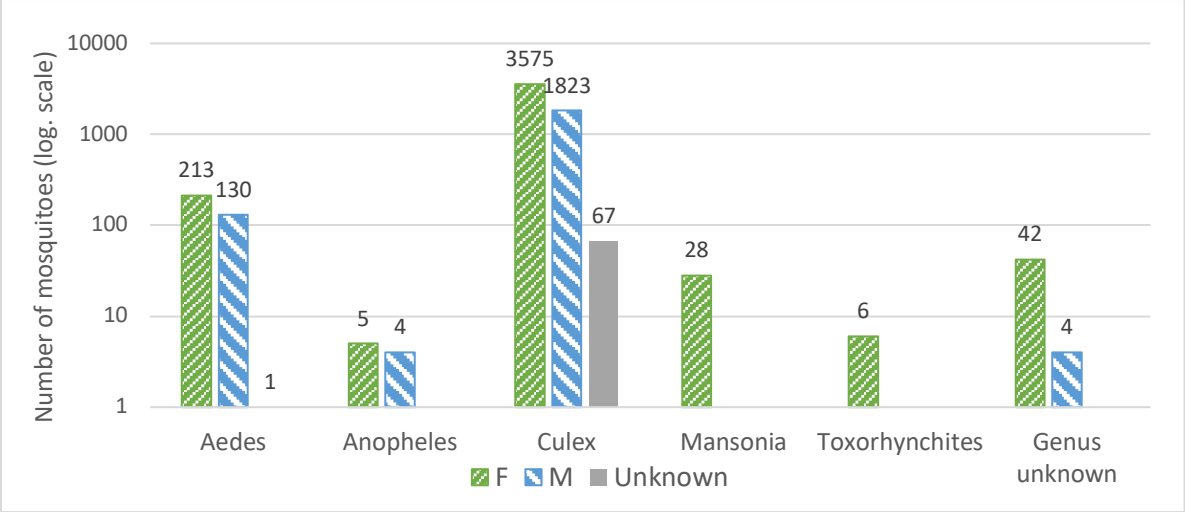

Fig. 2: Number of caught mosquitoes per genus according to sex.
Y-axis is in logarithmic scale to visualize the number between genera.

In terms of sampling success, we captured on average one small mammal per trapping day on all four sites combined. For the mosquitoes, the average trapping success was almost 454 mosquitoes per day on all four sites combined.

Gained perspectives

During this fieldwork we noticed that rodent trapping during the end of the rainy season is exceedingly difficult. The rodent population is then exceptionally low since there is not yet the lush green vegetation on which the rodents thrive. Due to the small population size and the limited number of traps we were unable to capture the envisioned 400 rodents. However, I made arrangements that I can use the blood samples that our local colleagues collected from the other rodent experiments.

For the mosquito sampling we experienced that setting up and maintaining the traps is quite simple and easily explainable to non-academic people. The only concern is that the traps are best placed in an area which is somehow protected or shielded from the public and that they are best not placed in direct line of site. The reason for this is that the traps contain a battery which is expensive and loosing this would result in a non-functioning trap.

Storing the collected mosquitoes before sorting them is best done in a fridge but not longer than three days, since we noticed that mosquito abdomens started to turn greenish after 2-3 days. This discoloration made morphological identification harder and could influence the molecular analysis later, since this discoloration is due to bacteria.

If samples are not being processed within 3 days, they are best stored in a -20°C freezer. In the beginning we stored our unprocessed mosquitoes immediately in the freezer, but we quickly realized that if the specimen had liquid (blood or eggs) in the abdomen, this liquid expanded due to the freezing process. This again influenced the morphological identifications and could also lead to contaminations if blood from abdomens would touch other mosquitoes.

The sorting of mosquitoes was a very labor-intensive process and required a constant focused attention. For future field sampling I would recommend having multiple stereo microscopes so that multiple people can work at the same time and share the workload.

We now have also realized that to perform the detailed morphological identifications we need optimal microscopes in a perfect working condition, which is most often not available in Africa. Secondly you would also need a clear identification key of potential mosquitoes which might occur in the region to successfully perform the morphological identifications. These identification keys are quite often not

available or are very general for larger areas such as Africa, as was the case with the keys we used. Therefore, we are compiling a pictorial key of the mosquitoes we captured in that region. Of course, this key will not be as detailed as official keys, but it will give a visual reference combined with a molecular identification for future mosquito studies in the same region.

Expected publications

We are currently in the process of drafting a paper on the presence of arbovirus antibodies in the African Multimammate mouse (*Mastomys natalensis*) from the same town and area as where we now sampled. Depending on the progress of the workflow it might be possible to include the limited samples we have collected now.

Beside that we will try to publish a paper on the mosquito diversity in the different areas that we sampled and try to find links between this diversity and the level of human presence. Either combined in this paper or as a second publication we will analyze the mosquitoes for the presence of arboviruses and the blood host they used. The aim of that second publication is to investigate the main blood hosts and arboviruses in different mosquitoes and how this might vary in different areas of human dominance.

We are also in the process of setting up a long-term capturing of mosquitoes in the same and surrounding areas of Morogoro Tanzania. We are so far planning to continue sampling for a year with 1 week of sampling per month.

All publications following this long-term sampling will also credit the Leopold III fund, since it is thanks to this fund that we were able to purchase the traps and the batteries.

Estimated period for publications are:

- Arbovirus serostatus of *Mastomys natalensis* in Morogoro, Tanzania (potentially samples of this mission might be included) ➔ end 2023
- Mosquito diversity in Morogoro, Tanzania ➔ end spring 2024
- Arbovirus presence in mosquitoes from Morogoro, Tanzania ➔ end spring 2024
- Mosquito diversity throughout the year ➔ end 2024

Conclusion

The field mission was very successful in terms of capturing mosquitoes, we did not envision we would capture such quantities of mosquitoes. This is beneficial for further analysis due to the large sample size but also warrants extra attention to the selection of samples to be screened. Since it is not possible to screen each sample individually due to the screening costs.

Unfortunately, the amount of rodents capture was very low and was a set back during the field work. Luckily there are other rodent projects ongoing from which it is possible to utilize blood samples for our screening. Concrete agreements still have to be made but we don't foresee any problems with exchanging data and samples.

In general this was a labor intensive field mission but very satisfying in terms of data collection and establishing new relations that benefit this PhD project and other future projects.

1.3. DE WEERDT, Joëlle (doctorante ULB)
What can whale songs tell us about population connectivity?
Mission to Nicaragua, 1 August – 30 September 2023.

Waters along the coast of Central America present a unique place in the world where the migratory routes of northern and southern hemisphere humpback whale populations overlap spatially.

Interestingly, song transmission between the northern and southern hemisphere humpback whale populations has never been examined and understanding the potential exchange between both populations could be the key to understand song propagation in the world ocean basins.

Humpback whales have a strong acoustic behavior and males produce songs during breeding season. Songs are culturally transmitted and are shared by individuals of the same regions, but songs can evolve through time. We wish to investigate population connectivity by studying the acoustics of both the northern and southern hemisphere populations breeding off the Pacific coast of Nicaragua.

Project background
Nicaragua is a developing country of Central America bordered by both the Caribbean Sea and the Pacific Ocean. Human activities along coastal areas put increasing pressure on coastal and marine ecosystems through the tourism industry (cruise ships and whale watching tour operators), fishing activities, and the construction of megaprojects, such as an interoceanic canal across Nicaragua, connecting the Caribbean Sea and the Pacific Ocean.

Unfortunately, there is a lack of knowledge on the presence and ecology of cetacean species along the Pacific coast of Nicaragua, which impedes decision makers to understand the impact of these potential pressures on the presence, behavior and habitat use of cetaceans.

Generating knowledge on the different cetacean species from the Pacific will be therefore essential for future environmental impact assessment of anthropogenic activities. For that reason, a study was initiated in 2016 by the applicant Joëlle De Weerdt and her non-profit organization ELI-S (https://eli-s.com) to understand species diversity, habitat use and distribution of cetaceans during the dry season (December to April).

The objectives are to:

(i) understand the impact of ocean upwelling on cetacean community structure;

(ii) characterize the humpback whale population migrating from the US West Coast to Nicaragua in terms of spatio-temporal distribution, habitat use and group types; and

(iii) assess migratory destinations of humpback whales from both the northern and southern hemisphere.

To achieve these objectives, systematic boat-based surveys are conducted in southern Nicaragua (near the proposed Nicaragua Canal and regional tourism development). Once a group of cetaceans is encountered, geographical position, behavior and photo-identification data are taken and environmental conditions such as Beaufort Sea state, swell height, visibility, precipitation are assessed.

Photo-identification (Photo ID) allows the identification of individuals by using natural marks on the ventral side of photographed flukes (humpback whales) (Katona *et al*., 1979) and the left and right side of the dorsal fin (dolphins). Ongoing and anticipated applications of this research could include, in the perspective of conservation biology, the establishment of marine protected areas, as well as an increase in awareness of local communities (including boat operators, fishermen, tourist-oriented business owners, residents and visitors) through workshops and trainings on responsible whale watching practices. This will potentially offer an economical solution to the financial pressure that exists in an impoverished area where fish populations are declining drastically mainly due to industrial (over)fishing.

Objective of the mission

The objective is to investigate population connectivity by studying the acoustics of both the northern and southern hemisphere populations breeding off the Pacific coast of Nicaragua. To reach this objective we will compare song phrases of both populations and search for correspondences.

Material and methods

The project implies field work between January and April (funded by other grants) and August and September 2023 through boat-based surveys during the wet season (August – October 2023), which covers the peak of humpback whale breeding season from the southern hemisphere humpback whale population (Figure 1).

During the surveys, a directional hydrophone (H2a-XLR) connected to a recorder (Tascam DR-05V2) was deployed (= hydrophone dips) to determine presence/absence of singers every 30 minutes. Whenever a singer was detected, and if their song was audible above the background noise, a recording was made at a sampling rate of 92 kHz and 24 bits resolution for as long as the song was detectable.

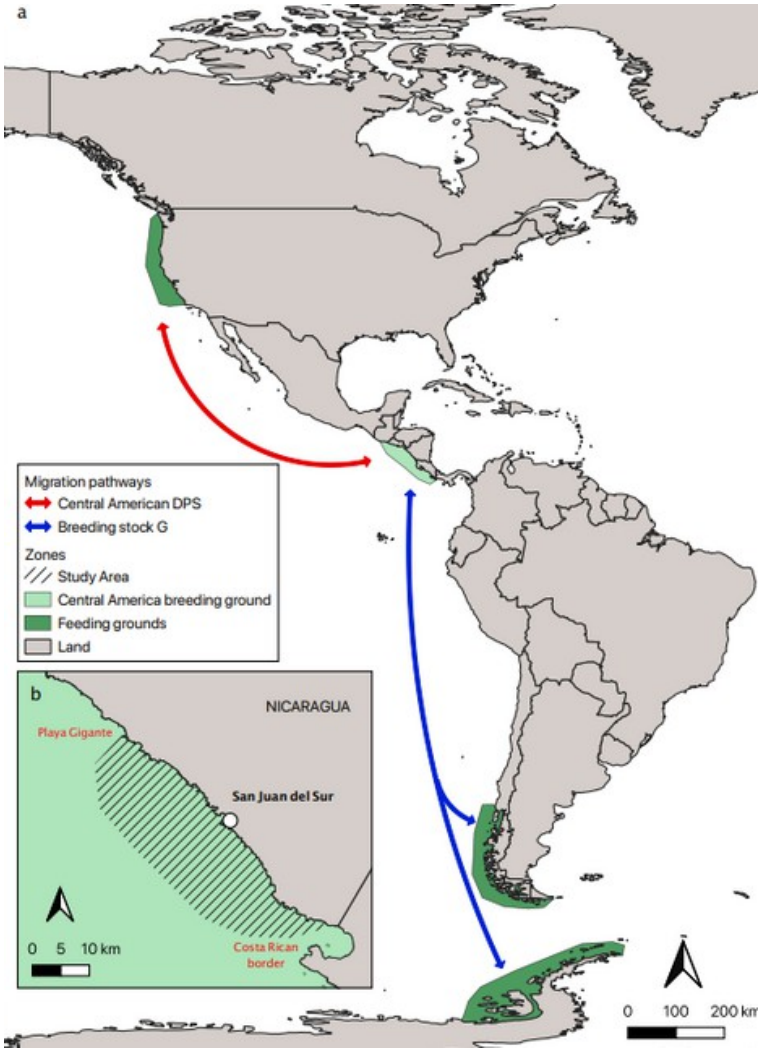

Fig. 1: a) Conceptual frame of the research indicating the migratory path of the northern (Central America DPS) and the southern (Breeding Stock G) hemisphere population. b) Study site for acoustical sampling, San Juan del Sur, Nicaragua. Figure from Laetitia Mitchell Master thesis 2021.

To be able to describe and analyze the sounds in humpback whales songs, we can visualize spectrograms. Spectrograms are visual representations of the frequencies of sounds. The frequency is on the vertical axis (y-axis) and the time is on the horizontal axis (x-axis). The visualization of the songs on Raven Pro 1.6 (Cornell Lab of Ornithology) were all done by selecting the same specific parameters.

Results

During the first season, a total of 36 days of boat trips was organized covering 164 hours of boat time and 1,777 km between January and April 2023. A total of 24 days of boat based surveys were organized covering 128 hours of boat time and 1,336 km between August and September 2023 (Figure 2). Ten songs were recorded between January and April 2023, and 10 songs were recorded between August and September 2023. A total of 303 hydrophone dips were made with 168 between August and September 2023 (Table 1).

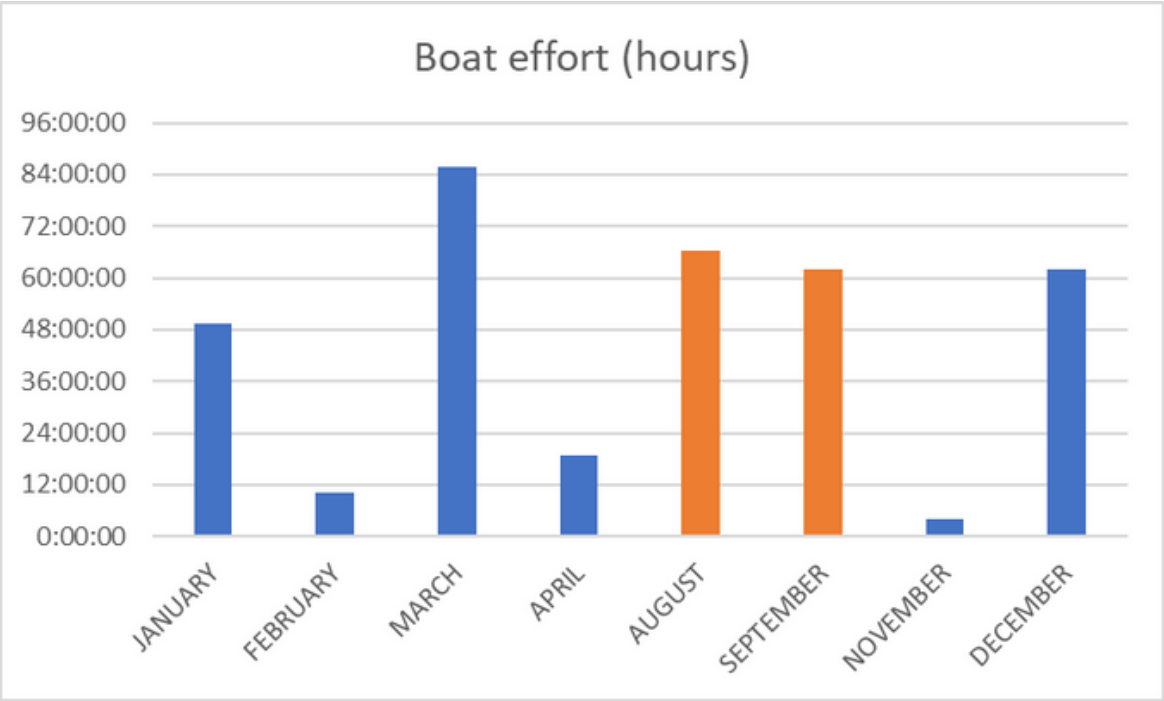

Fig. 2. Monthly research effort (hours) of the boat-based surveys in San Juan del Sur, Nicaragua.

Month	Total	Hydrophone dips
JANUARY	49:18:00	**58**
FEBRUARY	10:13:00	**2**
MARCH	85:47:00	**54**
APRIL	18:52:00	**21**
AUGUST	66:13:00	**87**
SEPTEMBER	61:56:00	**81**
NOVEMBER	3:54:00	**0**
DECEMBER	62:10:00	**67**

Table 1 : Number of hydrophone dips made every month during boat-based surveys.

Preliminary results

We have currently analyzed 7 songs out of the 10 recorded during the wet season (August to September) and have identified 7 phrases so far.

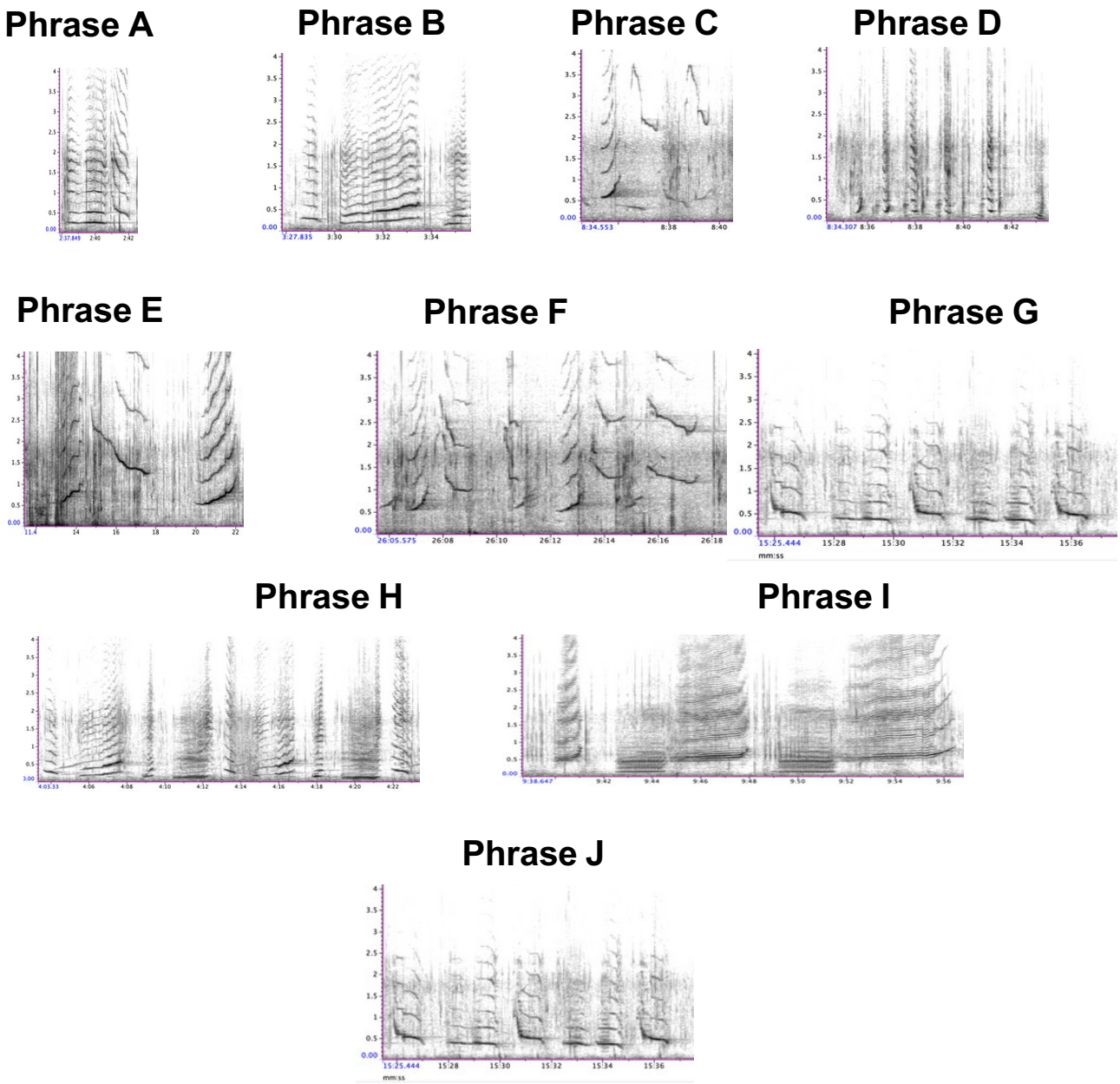

Fig. 3: Monthly research effort (hours) of the boat-based surveys in San Juan del Sur, Nicaragua.

Perspectives

In the next months we wish to further analyze the four remaining songs of the southern hemisphere humpback whale population and compare them with the songs characterized for the northern hemisphere population (Figure 4). Songs will be characterized as part of an ongoing master thesis project. Characterisation of songs includes the analysis of their units, phrases and theme structure for both populations and will be compared amongst populations in the next months. This song comparison will allow us to determine the presence of population connectivity.

A first characterisation of themes was produced for the northern hemisphere whale song recorded in March 2023 in Nicargua through a colaboration with Jim Darling from Whale Trust (Hawaii). A publication on population connectivity including data from 2021, 2022 and 2023 is anticipated to be redacted and submitted by end of 2024.

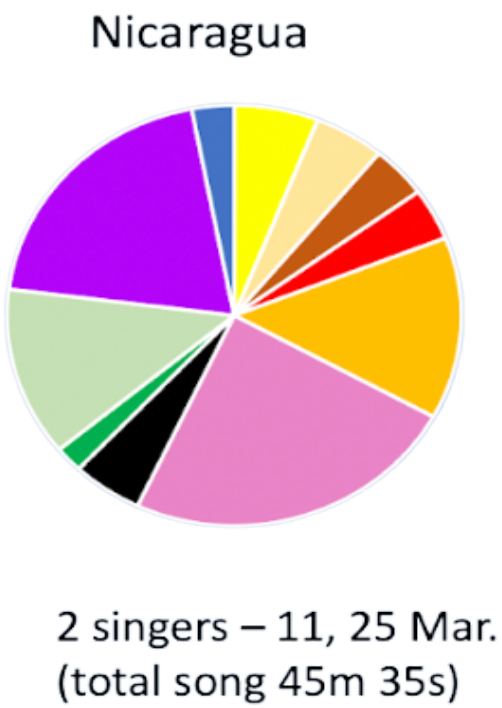

Fig. 4: Song structure of the Central American DPS (northern hemisphere) based on theme proportions identified in two songs recorded in March 2023 in Nicaragua. Courtesy of Jim Darling. Colors indicate proportions of different themes identified in two songs.

1.4. DRÖGE, Saskia (PhD student KU Leuven)
Environmental outcomes of voluntary sustainability standards (VSS) in the cocoa sector in Luwu Timur, Sulawesi Selatan, Indonesia.
Mission to Indonesia, 1 July – 28 October 2023.

1. Introduction
The world population continues to grow and is predicted to reach 9.7 billion in 2050[1]. Likewise, the demand for agricultural products is predicted to increase by 50%. Land-use change like deforestation and agricultural intensification are main drivers of biodiversity loss[2,3]. Voluntary Sustainability Standards (VSS) try to reduce the environmental impact of agricultural production by defining criteria for producers, for example, no deforestation for plantation establishment. The number of VSS has increased rapidly within the last years but studies on VSS effectiveness for cocoa *(Theobroma cacao)* and in Asia remain rare[4]. The aim of this mission and research was to close this knowledge gap and to gain better understanding of the effectiveness of VSS for sustainable cocoa production in Indonesia, i.e., if VSS lead to environmental benefits and how the design of VSS shape environmental outcomes.

The specific research questions were:
I) do certified and non-certified cocoa plantations in Luwu Timur, Sulawesi Selatan, Indonesia differ in biodiversity?
II) which VSS criteria cause differences in environmental outcomes among VSS?
III) do trade-offs among environmental and socioeconomics outcomes of VSS exist?

2. Organisation and general course of the mission
The research and mission was done in collaboration with IPB University, Bogor, Indonesia and is part of research project *'Voluntary Sustainability Standards (VSS) as a governance mechanism for*

sustainable global food and wood systems - VSS4f/wOOD'[5] at KU Leuven, Belgium. The field team consisted of two researchers, one ornithologist and one field assistant. As researchers, Saskia Dröge joined from KU Leuven and Muhammad Justi Makmun Jusrin joined from IPB University. Justi is a third year master student in Tropical Ecology. He flew the drone during field work and will analyse the drone data for his master thesis. Dadang Dwi Putra joined the team as local guide and ornithologist. He is based in Palu, Sulawesi, and has several years of experience working with different research projects in Indonesia. Hajar supported the field team as local guide, introducing the research project to farmers and village administratives, translating conversations and organising, for example, accommodation. All team members were involved in measuring vegetation structure on cocoa plantations.

The mission took place between July 1st and October 28th, 2023. The process of getting a research permit and research visa took a long time and could only be completed after arrival (standard procedure). Saskia Dröge arrived in Bogor on July 2nd and the completion of all necessary documents took about three weeks with support of Ibu Uceu from the International Collaboration Office at IPB. For completion, we had to visit the Immigration Office in Bogor four times to receive the necessary temporary stay permit as well as the National Police in Jakarta for a travel permit. All documents needed to be shown at regional government offices in Sulawesi (i.a., district administration in Malili, Kecamatan administration in Tomoni and Burau) prior to field work. The data collection eventually started on July 24th after all administration was complete. We revisited farmers who had already participated in a socioeconomic survey on VSS by PhD student Janne Bemelmanns in December 2022. Her socioeconomic survey included 600 cocoa farmers in 30 villages in the districts Luwu, Luwu Utara and Luwu Timur in Sulawesi (20 farmers per village, 10 villages per district). We restricted our research area to the district Luwu Timur and targeted a subset of farmers from the socioeconomic survey.

We set up a base camp in the city Tomoni in Luwu Timur to store equipment, tools and samples (Fig. I, Appendix). For the data collection, we searched for accommodation more close to the villages to reduce travel time and facilitate data collection. In Rante Mario and in Lauwo, we could find accommodation with the head of the village and sub-village. Forest cover in the Luwu Timur was very low as most of the forest was lost during the cocoa boom in the 1980s and 1990s and most of the remaining tree cover is now rrestricted to higher elevation and slopes in the north of Luwu Timur. The selection of forest plots with similar conditions as cocoa plantations (same altitude, low or no slope) was hence not possible. The biggest challenge we faced during the mission was to find suitable cocoa plots to include in our environmental assessment. For being able to do a trade-off analysis with socioeconomic data on VSS later, we were bound to select farmers and plots within the socioeconomic survey of Janne Bemelmanns. For each village, we had a list of farmers and their cocoa plots from this survey. We made the experience that it is easiest to ask the village head or the head of the farmer association to show us the cocoa plots from the list. Meeting each farmer proved to be difficult as farmers went out to their plantations early morning and returned late afternoon. We oftentimes met farmers in plantations when being showed around and could then talk to farmers verifying, for example, their certification status and other information on plantations we already had.

We experienced that many cocoa plots were converted to oil palm between the socioeconomic survey in December 2022 and the start of the environmental survey in July 2023 or that farmers were planning conversion. Farmers reported cocoa plantations in general to have low productivity and fertiliser to be unavailable or expensive so that switching to, for example, oil palm was financially more attractive. Other cocoa plantations were replanted with young cocoa trees and were thus not suitable for the assessment. In the villages Rante Mario, Cendana and Batu Putih, Balai Kembang and Margolembo, many cocoa plantations were on slopes. We had to exclude plantations on slopes because we could not collect drone data for slopes above 25 degrees as the drone could not follow the terrain. For the vegetation measurements, we used a 40 x 40 m plot design requiring plantations to be larger than 0.16 ha. Overall, many cocoa plantations in the research area are small (< 0.5 ha) and heterogeneous and we could not fit our 40 x 40 m plot inside. Cocoa farmers in the research area are typical smallholder farmers farming one or two cocoa plots and in total oftentimes less than one hectare. The search for suitable cocoa plots was thus the most time intensive factor during the mission. With all the limitations and after having visited all ten villages included in the previous socioeconomic survey in Luwu Timur, we were

able to select 62 cocoa plantations for the environmental assessment. 31 of the plantations were certified under the Rainforest Alliance certification scheme with Mars being the certificate holder of 16 plantations and Cargill holding the certificate for 15 plantations. Mars and Cargill initiated the certification and farmers do not need to pay to become certified. To become certified, plantations are visited by Mars, Cargill and/or Koltiva staff (certifying body). After the initial audit, plantations are usually visited (audited) once per year to ensure compliance with VSS criteria. Farmers may receive premiums for selling certified cacao, for example, in cash from Cargill and in kind (agricultural products like fertiliser) from Mars. An analysis of the implementation of the Rainforest Alliance scheme and its socioeconomic outcomes based on the previous survey will be done by PhD researchers Janne Bemelmans and Charline Depoorter within the research project VSS4f/wOOD.

Rainforest Alliance was founded in 1987. Since then, the VSS has updated its standard with the most recent version being from 2020. In 2018, it merged with UTZ. The environmental criteria of the latest Rainforest Alliance standard, among others, are:

- A minimum of 15% native vegetation cover for shade tolerant crops like cocoa
- Certified plantations not being on land deforested after 2014
- No hunting or collection of threatened animals and plants
- Protection of water bodies using buffer zones
- Using integrated pest management and reduction of agrochemical use to a minimum necessary

1. Material and methods

During the field mission between July and October 2023, we collected environmental data in 62 cocoa plantations in Luwu Timur, Sulawesi Selatan, Indonesia (Fig. I). 31 of the plantations were certified under Rainforest Alliance. We established a 40 x 40 m plot (0.16 ha) in each plantations to measure the vegetation structure: we measured the height and the girth at 30 cm height of all cocoa trees *(Theobroma cacao)* in a 20 x 20 m subplot and the height and girth of all shade trees (including coconut) in the entire 40 x 40 m plot. We decided to measure the girth of the cocoa trees at this height because cocoa trees branch already at low height in the research area due to their management. Cocoa trees are usually planted at equal space of 3 to 4 m. We also noted the cacao variant used and estimated the age of the cocoa trees. The age of the cocoa tree base might be older than the top branches because side grafting is a common practice to rejuvenate trees. We used the girth and height of the cocoa and shade trees to calculate the basal area of cocoa and shade trees per hectare and the above ground carbon stock for each plantation. For carbon stock estimations, we also collected drone imagery using a multispectral drone at different flight heights (60 m; 80 m; 100 m) and we will compare results to the standard measurement. The analysis of the drone imagery will be the main focus of Justi's master thesis.

We measured canopy closure, ground cover and took soil samples at the plot center and at 10 m distance from the plot center in each cardinal direction (hence 5 points per plot). The canopy closure was measured using a spherical densitometer as well as hemispherical photographs. The hemispherical photographs were later analyzed in the software Gap Light Analyzer to estimate the canopy openness and, in turn, canopy closure. For the ground cover, we estimated the ground covered by herbaceous plants as well as leaf litter. We took soil samples of the top soil layer (0-15 cm) to assess organic carbon, total nitrogen, total phosphorus, pH and soil texture. Soil analysis will be done at the lab PT. Biodiversitas Bioteknologi Indonesia in Bogor.

In each plot, we used standard point counts to estimate bird species richness. We recorded all birds seen and heard for 20 min after sunrise between 6 and 8 am and in the afternoon before sunset between 4 and 6 pm. We furthermore collected acoustic data using autonomous sound recorders (AudioMoths) recording at least 4 consecutive days in each plot.

We use the bird species richness as an indicator for biodiversity of plantations. To determine if it differs between certified and non-certified cocoa plantations, we will use a generalized linear mixed model. We will include the bird species richness as dependent variable and certification, cocoa basal

area, shade tree basal area, shade tree species richness, tree cover remaining in 200 m buffer around plantation as independent variables (fixed factors). To account for variation caused by location of the plantation, we will include the village where the plantation was located as random factor. The tree cover around plantations was calculated in Google Earth Engine using the NASA GEDI Global Forest Canopy Height 2019 data with 30 m spatial resolution[10]. We filtered for pixels with canopy height above 15 m and calculated the pixel area in 200 m buffer around each plantation.

2. Results

We collected environmental data for 62 cocoa plantations in Luwu Timur, Sulawesi Selatan, Indonesia. 31 of plantations were certified under the Rainforest Alliance scheme and 31 were not certified. Plantations were located in 6 villages with each village differing slightly in characteristics. For example, in Sumber Alam many cocoa plantations had papaya *(Carica papaya)* and pepper (*Piper nigrum)* intercropped with cocoa and in Lauwo many plantations were on land previously used for rice cultivation thus showing more wet soils. The research area in Luwu Timur in general had little residual forest. The landscape is dominated by agricultural use with large oil palm plantations as well as nickel mining by foreign investors. Remaining forests are found in less accessible slopes in the mountains.

Preliminary results show that cocoa basal area varied from 2.21 to 15.31 m^2 per hectare (average 6.22 m^2), shade tree basal area varied from 0 to 7.87 m^2 per hectare (average 1.89 m^2). Certified and non-certified plantations did not differ significantly in basal area of shade trees and cocoa trees. Canopy closure measured with the spherical densitometer varied from 0 to 96.1% (average 47.6%) and did not differ significantly for certified and non-certified plantations. Most shade was typically produced by cocoa trees themselves and some plantations had no shade trees within the 40 x 40 m plot. We observed that oftentimes the borders of the plantations are lined with trees producing most shade. Due to this heterogeneity in plantations it is difficult to assess the Rainforest Alliance criterion of at least 15% native vegetation cover. In total, 22 shade tree species were present in plantations with durian *(Durio zibethinus)* being most commonly used. Shade tree species are typically selected by their purpose, for example, durian for income through the selling of the fruits.

The total size of cocoa plantations varied from 0.21 to 2.5 ha (average 0.77 ha) and did not differ significantly between certified and non-certified plantations. The cocoa variants used by farmers were 25, 45 and a local variant. The variant 45 is used because it has large beans but some farmers reported the variant to produce only few pods per year. Cocoa productivity was in general low, around 447.8 kg dry beans per hectare. All farmers used fertiliser in their plantations but affordable, subsidised fertiliser is limited and farmers reported the low soil fertility to strongly limit cocoa yields. All cocoa plantations were affected by diseases. Black pod *(Phytophthora spp.)* and Vascular Streak Dieback (VSD, *Oncobasidium theobromae*) were present on nearly all plantations and cocoa trees showed leaf necrosis and dead branches. Some plantations also had cocoa pods attacked by squirrels or rats, cocoa pod borer *(Conopomorpha cramerella)* or Helopeltis *(Helopeltis spp.)*. Although farmers know that removal of cocoa pods infected by black pod is advised, farmers do not always remove infected pods or leave them on the ground potentially infecting more cocoa pods in the plantation. Due to low productivity and difficulties to, for example, get fertiliser, we saw many farmers converting their cocoa plantations to, for example, oil palm and maize.

In total, we observed 71 bird species in the cocoa plantations (5488 observations, 7792 individuals). 32% of observations were visual while the majority were aural detections (68%). We observed one eagle which we could not identify and hence did not include in the analysis (1 observation). The Sooty-headed bulbul *(Pycnonotus aurigaster)*, the Collared kingfisher *(Todiramphus chloris)*, the Golden-bellied gerygone *(Gerygone sulphurea)*, the Brown-throated sunbird *(Anthreptes malacensis)* and the White-shouldered triller *(Lalage sueurii)* were the most commonly detected bird species which represented 52% of observations and were present in and around 93.5% of plantations. All of the five common species are not threatened, generalist species with low to medium forest dependency, and indicate a clear agricultural landscape. Of the 71 detected bird species, three are endemic to Indonesia (5% of observations) and 17 are endemic to Sulawesi (6% of observations). The

Pale-bellied Myna *(Acridotheres cinereus)* is classified as vulnerable, and the Black-headed Kingfisher *(Actenoides capucinus)* is classified as near threatened by IUCN but both species were rare (< 0.01% of observations). We observed bird capturing with nets and glue throughout the study area and within cocoa plantations. 35 of the 71 bird species have a medium to high forest dependency (2721 observations, 49.6% of observations). Preliminary results show that bird diversity among certified and non-certified cocoa plantations did not differ significantly. We will analyse the bird data still in more detail, for example, if there are more endemic bird species in certified plantations. The sound recordings and also the soil samples still need to be analysed. All reported results are preliminary results so far. Once results are finalised and published, we will send the journal articles to the King Leopold III Fund for Nature Exploration and Conservation.

3. Perspectives

The research is part of the research project VSS4f/wOOD at KU Leuven, Belgium, and the project is ongoing till January 2024. A future collaboration of KU Leuven and IPB University and the continuation of the project is desired by the professors involved in the project. A proposal for a subsequent research project on VSS will be written if a suitable call for proposals will be available. Justi is planning to continue his studies after graduation from his master in Tropical Ecology from IPB and will write an application for a PhD grant. Furthermore, Justi recently founded an NGO (Hutan.In) working on forest conservation, research and other topics like plastic waste recycling. With the help of Saskia, he will apply for funding for a project on plastic waste recycling in 2024.

4. Destination of material collected

The collected data on vegetation structure and the data of bird point counts was digitised and saved in a cloud and is accessible to researchers at KU Leuven, Belgium, and IPB University, Bogor, Indonesia. Theacoustic data and the drone imagery is stored on two sets of hard drives with one set being available at KU Leuven and the other set remaining at IPB University.

Appendix

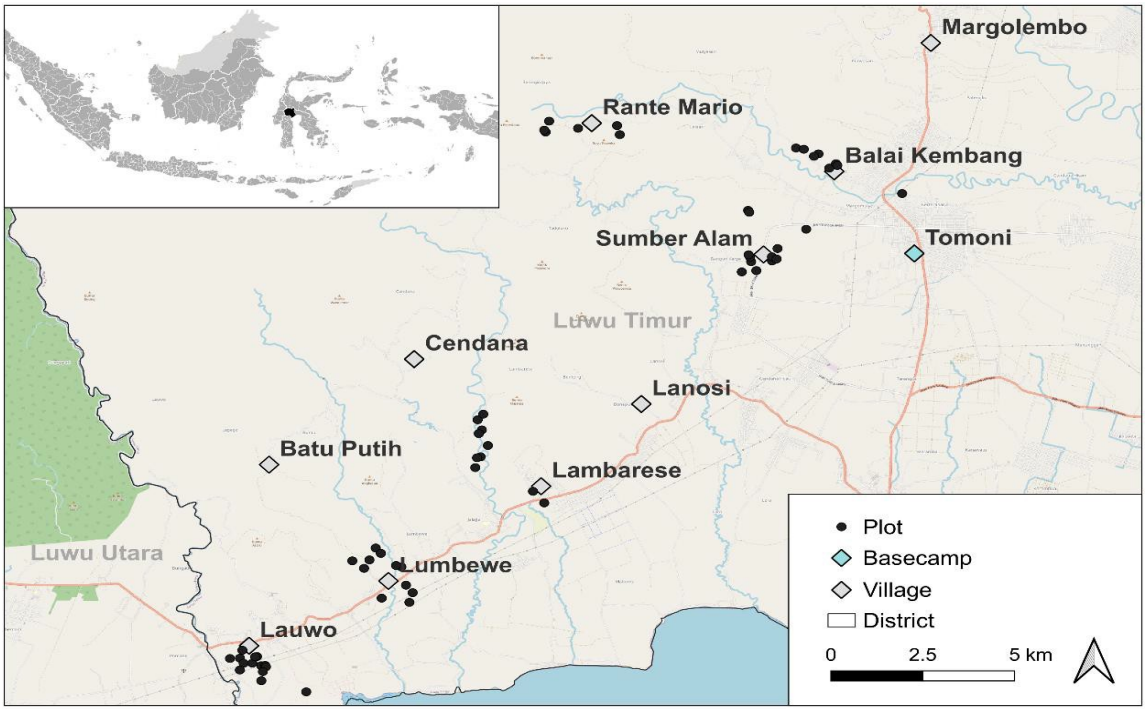

Fig. 1. Map of the research area in Luwu Timur, Sulawesi, Indonesia. The small map shows the location of Luwu Timur in Indonesia. The shown villages were previously sampled for a socioeconomic survey on VSS effectiveness in December 2022. Revisiting the same villages, we selected 62 cocoa plantations (31 certified; 31 non-certified) in six villages.

Fig. 2. Cocoa plantation in Lauwo, Luwu Timur, Sulawesi, Indonesia, with young oil palms. The cocoa plantation will be converted into oil palm and cocoa trees will be cut once the oil palms become bigger. Conversion of cocoa plantations into oil palm was present throughout all villages visited in Luwu Timur.

1.5. JOSSART, Quentin (postdoc, scientific collaborator ULB)
Biodiversity of intertidal communities from the Falkland Islands.
Mission to the Falkland Islands, 8 – 22 April 2023.

La mission s'est déroulée sur quinze jours du 8 au 22 avril 2023 aux îles Malouines (Falkland Islands). Ces îles australes représentent un cadre d'étude idéal du fait de leur grande diversité d'habitats et de leur localisation géographique au confluent de plusieurs courants océaniques. Dans le cadre d'une collaboration avec le « South Atlantic Environmental Research Institute (SAERI) » (Dr Narissa Bax), le travail de terrain a principalement consisté en l'échantillonnage et la mesure d'abondance *in situ* via une approche par quadrats (25 cm de côté, Figure 1).

Fig. 1 – Quadrat (25cm X 25cm) utilisé lors de l'échantillonnage. Un total de douze échantillonnages a pu être réalisé dans l'est des Malouines, majoritairement en zones rocheuses (8 échantillonnages) mais aussi en zones sableuses (4 échantillonnages). Les sites investigués (plusieurs échantillonnages par site) ont été les suivants : Salvador, Cape Pembroke (Figure 2), Mare Harbour, Murrel, Volunteer Point et Surf Bay.

Fig. 2 - Site de collecte de Cape Pembroke (East Falklands)

Au total, plusieurs centaines d'échantillons ont été collectés, révélant une importante diversité d'invertébrés marins. Un inventaire préliminaire de la faune a été réalisée à partir des identifications sur le terrain. Les taxons les plus emblématiques/indicateurs répertoriés sont les suivants : *Acanthocyclus albatrossis* (fighting crab) - *Anasterias antarctica* (common sea star, Figure 3) - *Antholoba achates* (flower-lobed anemone) - *Asterina fimbriata* (fringed bat star) - *Austrocidaris sp* (pink pencil urchin) - *Bunodactis octoradiata* (red anemone) - *Doris kerguelensis* (small bumped nudibranch) - *Fissurela crassa* (thick key-hole limpet) - *Halicarcinus platanus* (flat-backed crab) - *Labidiaster radiatus* (sunstar) - *Mytilus edulis* (blue mussel) - *Nacella magellanica* (Magellanic copper limpet) - *Notochthamalus scabrosus* (rough barnacle) - *Odontaster sp.* (brushed bat star) - *Pareuthria plumbea* (leaden whelk) -

Patagonotothen sp. (rockcod) - *Plaxiphora aurata* (hairy chiton) - *Phyllodocidae* (leaf worm) - *Polyzoa sp.* (dog ear tunicate) - *Psolus sp.* (plated sea cucumber) - *Siphonaria lessoni* (Lesson's false limpet) - *Sphaeromatidea* (isopod) - *Spirobinae* (Spiralled tubeworm) - *Thelepus sp.* (spaghetti worm) - *Tonica lebruni* (pink spotted chiton).

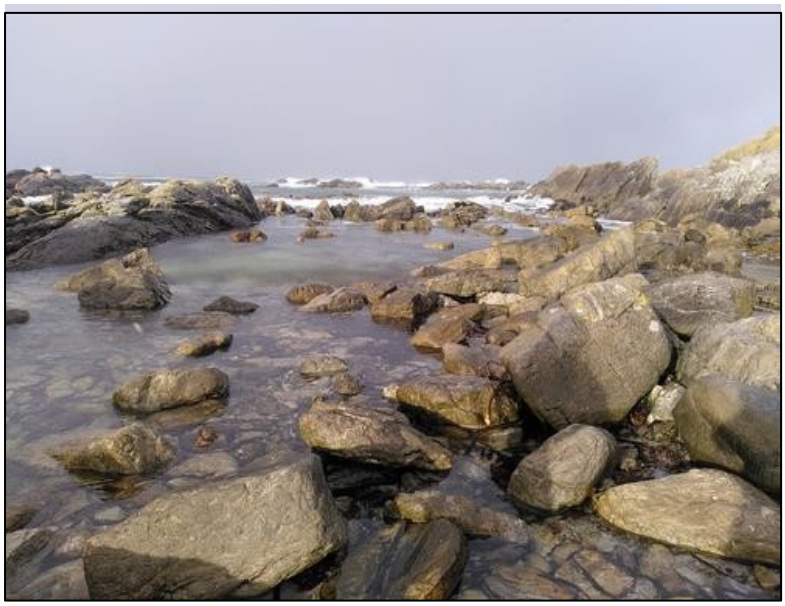

Fig. 3 - *Anasterias antarctica*, une espèce fréquemment observée lors de l'échantillonnage.

Les échantillons collectés seront prochainement triés et identifiés morphologiquement en microscopie et à l'aide de guides/clés de référence. Des photos et/ou des spécimens seront également envoyés à des experts taxonomiques afin de confirmer ou d'affiner les identifications. Dans une deuxième partie, la caractérisation génétique de ces macro-invertébrés sera réalisée, en utilisant la région du code-barre du gène mitochondrial de la cytochrome c oxydase I (COI). En combinant les données morphologiques et génétiques, une identification finale, au niveau taxonomique le plus précis possible, sera obtenue. Ces identifications intégratives seront combinées avec les données d'abondance obtenues sur le terrain, après quoi, des analyses statistiques univariées et multivariées seront réalisées. Les données obtenues permettront de mettre en lumière la diversité, la composition et la structure de ces communautés, ainsi que leur intérêt en termes de conservation (partage de données avec le département Environnement du gouvernent malouin).

1.6. LARRIDON, Isabel (PhD, Priority Leader, RBG Kew) **& SIMÕES, Ana Rita** (PhD, Taxonomist Africa, RBG Kew)
Flora van Afrikaanse graslanden met focus op Cyperaceae, Asteraceae en Convolvulaceae.
Zending naar Zambia, 30 april – 18 mei 2023

1. Algemeen kader
In het kader van twee lopende onderzoeksprojecten werd een veldexpeditie naar het noorden van Zambia uitgevoerd. Deze projecten zijn een doctoraatstudie gericht op het ontwarren van de evolutionaire relaties in het genus *Bulbostylis* (Cyperaceae) in Afrika, en een internationaal onderzoeksproject met als doel de generische classificatie van de tribus Vernonieae (Asteraceae) in het oostelijk halfrond. Als gevolg hiervan was het doel van de veldexpeditie het verzamelen van herbariumspecimens en bladstalen op silicagel om morfologisch en moleculair onderzoek uit te voeren om de diversiteit, evolutie en conservatiestatus van deze plantengroepen beter te begrijpen. Hieronder wordt elk van de twee projecten verder toegelicht.

Soorten van het genus *Bulbostylis* zijn te herkennen aan plukjes lange witte haren aan de mond van de bladscheden. Wereldwijd omvat het genus 227 soorten waarvan c. 127 inheems zijn in Afrika en 47 soorten voorkomen in Zambia (POWO, 2023). Het doel van deze doctoraatsstudie is om een duidelijk begrip te krijgen van de diversiteit, evolutie en conservatie van *Bulbostylis* in Afrika door een integratieve taxonomische benadering te gebruiken die meerdere gegevensbronnen combineert, waaronder fylogenetica, (micro)morfologie, ecologie en biogeografie. Daarnaast willen we, gebruikmakend van nieuwe mogelijkheden op het gebied van machine learning, nieuwe methodes testen voor geautomatiseerde soortidentificatie. Een van de hoofdstukken van deze thesis zal bestaan uit de taxonomische behandeling van *Bulbostylis* in Centraal-Afrika voor de Flore d'Afrique Centrale uitgegeven door de Plantentuin Meise. Alhoewel het noorden van Zambia niet valt binnen Centraal-Afrika, bied de geplande veldreis een veilige optie om veel van de *Bulbostylis* soorten uit Centraal-Afrika te besturen in veld. Isabel Larridon is hoofdpromotor van de doctoraatstudent Jérémie Morel.

Bijna één op de tien soorten bloemplanten behoort tot de zonnebloemfamilie (Asteraceae). Classificatie van Asteraceae wordt bemoeilijkt door het grote aantal soorten (meer dan 33.000) en genera (ca. 1.800). Een van de grootste tribus in deze familie, Vernonieae, is zo onhandelbaar gebleken voor taxonomische revisie dat ze vaak de 'evil tribe' wordt genoemd. Historisch gezien werden meer dan 1.000 van de ca. 1.500 soorten in Vernonieae geplaatst in het genus *Vernonia* Schreb. Dit genus is onlangs heromschreven en bevat nu slechts 21 soorten uit Amerika, wat leidde tot de noodzakelijke herclassificatie van ca. 1.000 soorten van *Vernonia* naar andere genera. Veel van dit werk is voltooid voor het westelijk halfrond, maar tot heden zijn er weinig studies uitgevoerd voor de meer dan 700 soorten Vernonieae uit het oostelijk halfrond. Er zijn momenteel minstens nog 200 soorten geplaatst in *Vernonia*, maar vanwege de slechte generieke afbakening in deze tribus is het onduidelijk tot welk genus ze nu behoren. In het lopende project wordt de generische classificatie van Vernonieae in het oostelijk halfrond aangepakt met behulp van nieuwe benaderingen in vijf doelstellingen: (1) kritieke bemonsteringslacunes overwinnen die integratieve taxonomie van Vernonieae verhinderen door

gedetailleerde herbariumstudie en veldexpeditieis; (2) een robuust evolutionair kader ontwikkelen op basis van fylogenomica en een reeks macro- en micromorfologische karakters verkennen om taxonomische revisie te vergemakkelijken; (3) een systematische studie uitvoeren die fylogenomische en morfologische gegevens combineert om de generieke plaatsing van alle soorten Vernonieae in het oostelijk halfrond op te lossen; (4) taxonomische informatie verspreiden via de Global Compositae Database; en (5) training geven aan de volgende generatie in plantensystematiek en taxonomische expertise in Asteraceae. Beide aanvragers maken deel uit van het leidinggevende team van dit project: Isabel Larridon as co-PI and Ana Rita Simoes als onderzoeker naar micromorfologische kenmerken om de genera van elkaar te onderscheiden.

De expeditie naar Zambia werd georganiseerd in samenwerking met de University of Zambia in Lusaka, en met de Royal Botanic Gardens Kew. Een van de belangrijkste samenwerkingen verliep met Dr. David Chuba, Lecturer aan de University of Zambia in Lusaka.

2. Originaliteit en toegevoegde waarde van het project

Argumenten om het veldwerk in het kader van deze projecten uit te voeren waren:
De meeste herbariumspecimens die beschikbaar zijn voor het genus *Bulbostylis* en de tribus Vernonieae werden verzameld c. 50 jaar geleden. Recente herbariumspecimens en silicagelstaaltjes voor DNA-extractie van hoge kwaliteit zijn over het algemeen niet beschikbaar voor deze plantengroepen. Gebrek aan gegevens heeft het bestuderen van hun evolutionaire en biogeografische geschiedenis belemmerd. Bovendien blijft de afbakening van soorten onzeker zonder een gedetailleerd op DNA gebaseerd onderzoek.

Wereldwijd omvat het genus *Bulbostylis* 227 soorten waarvan c. 127 zijn inheems in Afrika, waarvan 47 in Zambia (POWO, 2023). Voor *Vernonia* zijn de meeste soorten op het westelijk halfrond al verplaatst om genera te scheiden. echter c. Er blijven 240 Afrikaanse soorten in *Vernonia*, waarvan 47 in Zambia voorkomen (POWO, 2023). Zambia bevat een unieke combinatie van soorten die ook voorkomen in Oost-, Centraal- en Zuidelijk Afrika; daarom laat inzamelen in Zambia toe om een breed scala aan soorten te verzamelen en basisinformatie te verschaffen voor toekomstige studies, niet alleen in Zambia maar ook in andere Afrikaanse regio's. Er zijn meer herbariumspecimens van hoge kwaliteit nodig om (micro)morfologische kenmerken te onderzoeken om taxonomische revisie te vergemakkelijken. De veldexpeditie zal (1) het aantal herbariumspecimens van hoge kwaliteit vergroten om een morfologische studie mogelijk te maken; en (2) het genereren van DNA-gegevens mogelijk maken voor een aanzienlijk aantal van hun soorten.

3. Doelstellingen van de zending

a) Inzamelen van complete en goed bewaarde en gemonteerde herbariumspecimens als voucher voor de ingezamelde data en stalen.
b) Inzamelen van data bijhoordend tot de herbariumspecimens (geografische coordinaten, hoogte, morfologie, fenologie, taxonomische geschiedenis, lokale namen, habitat- en vegetatiegegevens en andere ecologisch relevante data, ...).
c) Bladstalen op silicagel voor DNA extractie. Het DNA uit de bladeren zal gebruikt worden om met behulp van de high-throughput sequencing techniek targeted sequencing fylogenomische data te genereren voor het bestuderen van verwantschapsrelaties.
d) Pollenstalen voor micromorfologische studie van Vernonieae soorten.
e) Foto’s van de plantensoorten in habitat die zullen verspreid worden via iNaturalist voor het ondersteunen van geautomatiseerde soortsidentificatie in het veld.

4. Voorbereiding en reeds lopende activiteiten

De selectie van de collectielocaties gebeurde aan de hand van de informatie op de etiketten van herbariumspecimens en online databases.

Hoewel vers en silicagel gedroogd materiaal het beste uitgangspunt vormen voor het bekomen van bruikbaar DNA voor fylogenetische studies is DNA extractie uit goed geconserveerd herbariummateriaal mogelijk. Dit betekent dat materiaal van een groot aantal soorten beschikbaar is in

de ons omliggende herbaria: de collecties van de Universiteit Gent (GENT), de Plantentuin Meise (BR), de Nationale Plantentuin te Meise (BR), het herbarium van het Muséum National d' Histoire Naturelle te Parijs (P) en het Kew Herbarium (K). Extracties uit herbariummateriaal hebben echter een wisselend succes afhankelijk van de droogtechniek en de nabehandeling die de specimens kregen. Ook zijn sommige soorten in herbaria enkel vertegenwoordigd als zeer oude collecties of door typemateriaal. Om dit probleem op te vangen is het collectioneren van vers materiaal absoluut noodzakelijk.

De moleculaire techniek waarvan gebruik gemaakt zal worden voor dit project (targeted sequencing) werd reeds getest op >300 Cyperaceae soorten uit bijna alle genera van Cyperaceae (Larridon et al., 2021), en ook al met succes is gebruikt in studies op Convolvulaceae (Simões et al., 2022). Voor Vernonieae is ook reeds een studie gaande die deze techniek aanwend, en stalen die tijdens deze expeditie in Zambia zullen verzameld worden zullen bijdragen tot dit onderzoek. Onderzoeksfinanciering voor het moleculaire labowerk is reeds beschikbaar (doctoraatsfinanciering Jérémie Morel, onderzoeksproject Vernonieae, etc.).

Als voorbereiding op de expeditie werden alle praktische regelingen getroffen voor de reis naar Zambia en de logistiek ter plaatse, alsook de nodige vergunningen voor het inzamelen en uitvoeren van planten voor wetenschappelijk onderzoek, dit alles in samenspraak met de University of Zambia.

5. Studiegebied en verloop van de expeditie
De veldexpeditie vond plaats in de Mutinondo Wilderness Area, een beschermd gebied in het noorden van Zambia van 10.500 hectare waar eerder meer dan 1700 plantensoorten zijn geïdentificeerd. De redenen om voor deze locatie te kiezen zijn: (1) rijkdom aan soorten van de beoogde plantengroepen, (2) nabijheid van de Democratische Republiek Congo wat een belangrijk studiegebied is in de twee gekoppelde projecten (bijv. bijdrage aan Flore d'Afrique Centrale); en (3) het vermogen om veldwerk uit te voeren in een veilige omgeving voor doctoraatstudent Jérémie Morel.

Bij aanvang van de expeditie in Zambia werd een dag voorzien in Lusaka zodat praktische regelingen kunnen worden getroffen met de medewerkers van de University of Zambia in verband met het inzamelen van het plantenmateriaal. Op het einde van de expeditie werden een 3-tal dagen voorzien om te netwerken met de collega's van de University of Zambia (UNZA), de exportpermits te regelen met verantwoordelijken van UNZA en het bevoegde ministerie, het UNZA herbarium te bezoeken en om gastlessen te geven ter plaatse.

De gastlessen gingen door op woensdag 17 mei:
- Dr. Isabel Larridon: Accelerating Taxonomy and Tropical Important Plant Areas
- Dr. Benoit Loeuille: An overview of Vernonieae (Composieae/Asteraceae)
- Doctoraatstudent Jérémie Morel: The genus *Bulbostylis* (Cyperaceae) in Africa: an integrative approach

Data en planning:
De veldexpeditie vond plaats van 30 april tot 18 mei 2023.
- 30 april 2023: vlucht naar en regeling logistiek in Lusaka, Zambia
- 1 mei 2023: afspraak met Dr. David Chuba in de University of Zambia om de onderzoekspermit op te halen
- 2-14 mei 2023: verplaatsing naar en expeditie in de Mutinondo Wilderness Area, Zambia
- 15-17 mei 2023: bezoek University of Zambia in Lusuka voor het geven van een gastlezingen en onderzoek in het herbarium, en het regelen van de exportpermits
- 18 mei 2023: vlucht terug.

6. Methoden en uitgevoerde activiteiten
Om een moleculair onderzoek te kunnen uitvoeren, werden er enerzijds jonge bladeren ingezameld op silicagel en anderzijds herbarium vouchers ingezameld om een morfologische representatie te hebben ter validatie van de soort.

Er werden drie tot vijf jonge bladeren per soort ingezameld. Deze bladstalen werden in theezakjes gestoken opgeborgen en genummerd. Het genummerde staal werd in een Zip loc zakje met silicagel bewaard. Dit zorgt ervoor dat de bladstalen op een tweetal dagen droog zijn. Door het snelle droogproces, heeft het DNA minder tijd om te degraderen en hebben we een goede DNA kwaliteit voor verdere moleculaire analyses.

Om te kunnen voorleggen dat de stalen daadwerkelijk afkomstig zijn van de vermelde soorten, werd er per soort een herbarium voucher ingezameld. Hiervoor werden enkele individuen met bladeren en bloemen en/of vruchten gedroogd. Een duplicaat wordt gedeponeerd in het UZL herbarium van de University of Zambia, een duplicaat in het NDO herbarium in Kitwe van de Forestry Ministry die de onderzoekspermit leveren, en bijkomende duplicaten in GENT, K, P en/of BR.

Data bijhorend tot de herbariumspecimens (geografische coördinaten, hoogte, morfologie, fenologie, taxonomische geschiedenis, lokale namen, habitat- en vegetatiegegevens en andere ecologisch relevante data, ...) werden genoteerd en opgenomen in databases en vermeld op de etiketten van de herbariumspecimens.

Foto's van de habitus en het habitat van de ingezamelde planten zullen verspreid worden via iNaturalist voor het ondersteunen van geautomatiseerde soortsidentificatie in het veld.

7. Resultaten

Kwaliteitsvol plantenmateriaal werd verzameld voor het lopend onderzoek (zie de lijst toegevoegd als Appendix). Eveneens werden in het gebied een aantal soorten gedocumenteerd die er niet eerder werden aangetroffen aangezien het grotendeels onderbemonsterd is. Het verdere onderzoek op het ingezamelde plantenmateriaal gebeurt na terugkeer uit Zambia grotendeels aan de UGENT en aan de Royal Botanic Gardens Kew.

8. Gebruik van de resultaten en vooruitzichten (publicaties inbegrepen)

Het ingezamelde plantenmateriaal zal dienen als basis voor verder moleculair, biogeografisch en systematisch onderzoek op Cyperaceae en Asteraceae. Het plantenmateriaal en de data ingezameld tijdens deze expeditie en de observaties in het veld een bijdrage leveren aan meerdere wetenschappelijke publicaties.

Het verzamelde plantmateriaal zal bijdragen aan het produceren van de volgende resultaten tijdens het doctoraatsonderzoek over *Bulbostylis*:

1. Een nieuwe op DNA gebaseerde infragenerische classificatie van *Bulbostylis*;
2. Een gedateerde fylogenie die voorouderlijke karakterstaten en de biogeografische en diversificatiegeschiedenis van *Bulbostylis* reconstrueert;
3. Een geïntegreerde taxonomische behandeling van *Bulbostylis* in Afrika, inclusief een identificatiesleutel, beschrijvingen, illustraties, verspreidingskaarten en beoordelingen van de IUCN Rode Lijst.

Het zal ook bijdragen aan de verwachte resultaten voor het Vernonieae-project:

1. Een fylogenomische studie van Vernonieae in het oostelijk halfrond;
2. Een integrerend taxonomisch kader voor een herziene classificatie van Vernonieae op genusniveau in het oostelijk halfrond.

1.7. MEEUS, Sofie & GROOM, Quentin (Agentschap Plantentuin Meise)
Building knowledge on invasive non-native species in Diego Garcia.
Mission to Diego Garcia Island, Pacific Ocean, 12 – 30 May 2023.

1. Introduction

Background and goals of the mission

Diego Garcia is a remote atoll in the Indian Ocean with Ramsar status and spans an area of 30 km². This crudely 'V' shaped atoll stretches over 64 km with a large internal lagoon. Its highest point, only a few meters above sea level, comprises ancient coral rubble and sand. The climate, characterized as humid-tropical, remains fairly constant throughout the year, influenced by the surrounding ocean. While atolls, in general, are relatively species-poor, their ecosystems cover only a small area globally and are highly threatened by invasive species and sea level rise.

The island currently has no permanent residents, hosting only UK and US military personnel, along with civilian contractors primarily from the Philippines. Before the construction of the military base, Diego Garcia was sparsely populated, featuring coconut plantations as the main agricultural crop. Although these plantations remain, they now pose a conservation challenge as they are weedy and dominate the vegetation. The natural vegetation consists of tropical forest, with species such as *Barringtonia asiatica* and *Pisonia grandis*, accompanied by an understory of shrubs, ferns, and grasses.

The island faces challenges from impactful invasive species, including rats, donkeys, cane toads, and various pantropical weeds. The delicate balance of the ecosystem is at risk, emphasizing the urgency of understanding and addressing the threats posed by invasive species.
The mission had multiple goals centered around the themes of conservation, invasive species, and specimen collection for the herbarium. The explicit objectives were:

1. Documenting all vascular plant species on Diego Garcia, including native, introduced, or cultivated species.
2. Conducting surveys in all habitats and areas of the island to ensure completeness and evaluate the widespread distribution of each species.
3. Collecting high-quality herbarium specimens from the flora to serve as voucher specimens and enrich the BR herbarium at Meise Botanic Garden.
4. Providing expert advice to local conservationists on identifying species that may require control measures.

2. Organization and overview of the mission

We collaborate with scientists from various institutions such as the UK Centre for Ecology and Hydrology, the Natural History Museum, London and INBO. Unfortunately, due to logistical challenges, these partners were unable to join the expedition. The planned team had to be adjusted, and the mission proceeded in May with only Dr. Quentin Groom and Dr. Sofie Meeus from Meise Botanic Garden focusing on the creation of a complete inventory of vascular plants. On site we received support from our local partner, host, and contact in Diego Garcia, Lhemar Antipasado, who serves as a Natural Resource Specialist and Conservation Program Manager at PWD Environmental. A positive consequence of not travelling with the others, though, was that we were less dependent on the planning of the team members.

The primary challenge was accessing Diego Garcia, as tourism is prohibited due to the U.S. navy support facility. Flights, organized by the military and scheduled weekly from Bahrain, occasionally face delays or cancellations. Fortunately, our flights proceeded relatively smoothly except for a 24-hour delay on the last day.

An unforeseen challenge was posed by cyclone Fabien, which reached category 3 strength near Diego Garcia, resulting in substantial rainfall throughout the initial week of our expedition, adversely affecting the efficiency of our fieldwork.

The reception we got from the military, civilians and fellow scientists on the island was excellent. We were helped a lot despite the cyclone during one week and visiting dignitaries.

3. Observations and collections

We collected a total of 114 specimens in duplicate. These specimens, combined with records from our expedition and data from previous expeditions (1883-2022), are currently being used as evidence to compile a complete list of the flora of the Chagos Archipelago.

We recorded 988 observations on iNaturalist, encompassing 165 species of vascular plants encountered during our expedition. Each observation is meticulously documented with photographs, contributing open, community-curated data to enhance our understanding of the island's flora.

These results not only enrich our herbarium collection at Meise Botanic Garden but also contribute essential data to ongoing research projects, including the compilation of distribution maps and red-listing assessments for the Chagos Archipelago. The substantial addition of new material to the BR herbarium is a notable outcome, given the absence of plant material from the Chagos Islands before this trip. The BR herbarium now stands as a repository for valuable specimens from the Chagos Archipelago, second only to the Kew Herbarium.

4. Perspectives

This mission aligns with an enduring collaboration between the UK Centre of Ecology and Hydrology (UK) and INBO (Belgium), with a specific emphasis on invasive alien species. This collaboration has manifested in initiatives like the COST Action Alien-CSI and projects such as TrIAS (Belspo). Meise Botanic Garden is currently leading in the development of a Horizon Europe project application centered on invasive alien species. This application involves the same esteemed partners, recognized authorities in the field of invasion biology.

5. Location of specimens and doubles

Collected specimens are deposited at Meise Botanic Garden and duplicates donated to Royal Botanic Gardens, Kew and Singapore Botanic Garden.

6. Title of publications in progress and/or planned

We are pleased to announce the forthcoming publication, currently in progress, intended for submission to the Biodiversity Data Journal. The title of the publication is:

"The Flora of the Chagos Archipelago: A Conservation Checklist of Native and Introduced Plant Species"

Authors include:

- Sara Bárrios, Royal Botanic Gardens Kew, Richmond, London, GB
- Colin Clubbe, Royal Botanic Gardens Kew, Richmond, London, GB
- Danielle Frohlich, SWCA Environmental Consultants, 1200 Ala Moana Blvd, Suite 380, Honolulu, HI 96814 (dfrohlich@swca.com)
- Quentin Groom, Meise Botanic Garden, Meise, Belgium
- Sofie Meeus, Meise Botanic Garden, Meise, Belgium
- Jodey Peyton, UK Centre for Ecology & Hydrology, Benson Lane, Crowmarsh Gifford, Wallingford, UK, OX10 8BB

This collaborative effort, made possible through the invaluable support of the Leopold III Fund, brings together expertise from Royal Botanic Gardens Kew, Meise Botanic Garden, SWCA Environmental Consultants, and the UK Centre for Ecology & Hydrology, reflecting a comprehensive approach to documenting and conserving the rich biodiversity of the Chagos Archipelago.

7. Conclusion

The mission contributes to long-term conservation goals, providing essential data for the territory's Conservation Management Plan and supporting biodiversity research.

1.8. MIRONOVA, Polina (Research group Mycology, Ghent University)
Laboulbeniales (Ascomycota) from Mozambique.
Mission to Mozambique, 3 – 19 March 2023.

Introduction and objectives
Tropical regions are seen as biodiversity hubs for many groups of organisms (Pianka, 1966; Hillebrand, 2004; Mittelbach *et al*., 2007). However, there is a knowledge gap on the extent of fungal diversity in the tropics which leaves the latitudinal gradient hypothesis for the fungal kingdom a question mark. Some studies have shown that some fungal groups do not follow the latitudinal gradient hypothesis (Tederesoo *et al*., 2014; Větrovský *et al*., 2019), while other groups have been found to be more hyperdiverse in the tropics (Arnold *et al*., 2000). We hypothesize that microfungi follow the latitudinal gradient hypothesis (LGH), and are thus more diverse in the tropics than in temperate regions.

The objective of this trip was to expand upon the list of known arthropod-associated fungal (Laboulbeniales) species in Africa and the knowledge of their global distribution. The data collected on this trip will contribute to microfungal ecological, evolutionary, and systematic research.

Similar standardized fieldwork, led by M. Catherine Aime (Purdue University) and Danny Haelewaters (Ghent University), is planned to take place in all four hemispheres. The collected data and material from these field trips will allow us to compare neo- and paleotropical microfungal diversity with neo- and paleotemperate diversity. To date, fieldwork has been completed in North Carolina (summer 2022), Brunei (October 2022) and Mozambique (March 2023).

Organisation of trip and participants
I joined Dr. Danny Haelewaters (DH) (mycologist) and Kirk Silas (bat specialist) on this trip. The expedition was overall successful, but some days were affected by the rains from cyclone Freddy.

Materials and methods
Collecting arthropods - Different entomological techniques were used to sample arthropods from the soil/leaf litter, those attracted to bait (rotting chicken), and flying taxa, along a transect surrounding the plots which were 10m x 100m in dimension.

Four Malaise traps were set up at every 25m and were collected after 24hrs. Winkler bags were used to collect leaf litter/soil arthropods, the litter was picked up at 25m and 75m, put in a Winkler bag and the arthropods that had fallen into the ethanol were collected after 72hrs. Ten baited pitfall traps were placed every 10m and were collected and reset in a different plot after 24hrs. Additionally, a litter reducer was used (12.5m, 37.5m, 62.5m, and 87.5m) to collect litter-dwelling arthropods and an umbrella was used as a beating sheet (every 10m) to collect arthropods from shrubs and tree foliage. All arthropods were put in 99% ethanol.

Catching bats and isolating bat flies – Bat surveys were done twice in each plot during the evening using 6 m and 12 m ground mist-nets (36mm mesh, 4 shelves, Avinet, Portland, Maine). The mist-nets were placed so that at least one pole was within 10 m of a plot, over trails that bats use as flight paths. Surveys started at sunset, until midnight. The opened nets were checked for bats every 20 minutes. Bats were disentangled from the nets and placed into cotton bags until they could be processed. Each bat was screened for ectoparasites (ticks, fleas, mites, bat flies) and these were collected with forceps and put into ethanol tubes. Forearm length and weight were also measured for each bat.

Results and perspectives
Around 2463 specimens of arthropods were collected comprising different orders: Hymenoptera (ants), Blattodea (termites and cockroaches), Diptera, Coleoptera (ground beetles, scarab beetles, etc), Orthoptera, Mantodea, Diplopoda, etc. These specimens will be screened individually, looking for Laboulbeniales thalli under a stereomicroscope. Some thalli will be taken off the host using a fine entomological pin and prepared on a slide to identify and study the morphology. Other thalli will be used for DNA extraction.

During our stay in Gorongosa National Park, we screened part of the entomological collection of EO Wilson Laboratory. 16 out of 900 screened arthropods were infected with Laboulbeniales thalli, likely from the genus *Laboulbenia*. Microscopic slides of the thalli were prepared by DH.

We caught seven bat species in total, five species from mistnetting (*Chaerephon pumilus, Neoromicia nanus, Pseudoromicia rendalli, Scotophilus viridis and Triaenops afer,*) and two species from collecting them with a hand net (*Hipposideros caffer, Rhinolophus lobatus*) (Figure 1). We found bat flies on two species of bats and mites on *Neoromicia nanus* (Table 1). These arthropods will also be screened for Laboulbeniales thalli.

Bat species	Bat flies	Other ectoparasites
Chaerephon pumilus		
Hipposideros caffer		
Neoromicia nanus		x
Pseudoromicia rendalli		
Rhinolophus lobatus	x	
Scotophilus viridis		
Triaenops afer	x	

Table 1. Ectoparasites collected from caught bat species

Fig. 1. A) *Rhinolophus lobatus* male with orange underarm hair. B) *Rhinolophus lobatus* orange morphotype. C) *Triaenops afer* D) *Hipposideros caffer* E) *Neoromicia nanus* F) *Chaerephon pumilus* G) *Pseudoromicia rendalli*

Destination collected material

We received research permits and export permits from Gorongosa National Park administration signed by Marc Stalmans (Director of Scientific Services). Based on previous experiences with lost mail packages to Mozambique slides will be deposited in the mycology Herbarium at Gent University (GENT).

Conclusion

An estimated amount of 2463 specimens of arthropods were collected using entomological trapping methods and from bats. These specimens will be screened individually in the upcoming six months and which will lead to an inventory of Laboulbeniales species in Gorongosa National Park. Furthermore, the collected specimens provide an important data point in the paleotropical region which will allow us to compare microfungal diversity in different biogeographic realms.

2. Varia - Divers

2.1. Evenementen – Evénéments

- 31.08.2023 Voordracht: “Een nieuwe soort cicade lijkt op een Triceratops.”
  Twee entomologen van het KBIN hebben een nieuwe soort dwergcicade (lengte 7-8 mm) uit Australië beschreven met een opvallende kop: zij heeft drie “horens”. De soort wordt beschreven in het *Belgian Journal of Entomology*, 137. Diverse expedities werden gecofinancierd door het Leopold III-Fonds.

2.2. Ontvangen boeken en documentatie - Livres et documents reçus

2.3. Wetenschappelijke publicaties voortvloeiend uit terreinzendingen financieel gesteund door het Leopold III-Fonds
Publications scientifiques issues de missions de terrain cofinancées par le Fonds Léopold III

Het aantal wetenschappelijke publicaties verwezenlijkt met financiële steun van het Leopold III-Fonds bedraagt meer dan 1.600. De publicaties verschenen in **2023** worden hierna vermeld.

Le nombre des publications scientifiques réalisées avec l’appui financier du Fonds Léopold III s’élève à plus de 1.600. Celles publiées en **2023** sont mentionnées ci-dessous.

2.3.1. Publicaties als gevolg van het Biologisch Station Koning Leopold III op het eiland Laing in Papoea-Nieuw-Guinea
Publications suite à la Station biologique Roi Léopold III à l'île de Laing en Papouasie Nouvelle-Guinée

Geen titels ontvangen – Aucun titre reçu

2.3.2. Publicaties als gevolg van andere terreinzendingen
Publications suite à d'autres missions de terrain

Baudoux C., Biwolé A., Hardy O. J., Webber B. L., Heuret P. 2023. Can the competition dynamics of non-native invaders be reconstructed to reveal historical impact? The case of *Cecropia peltate* and *Musanga cecropioides* (Urticaceae) in Cameroon. *Biological Invasions.* https://doi.org/10.1007/s10530-023-03175-4

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Rafael, J.A. & Pollet, M., 2023. First record of Pipunculidae (Diptera) from Martinique, with a description of *Eudorylas dumbardoni* sp. nov. *Sudies on Neotropical Fauna and Environment.* https://doi.org/10.1080/01650521.2023.2266171

Raick, X., Di Iorio, L., Lecchini, D., Bolgan, M. & Parmentier, E., 2023. "To Be, or Not to Be": Critical Assessment of the Use of α-Acoustic Diversity Indices to Evaluate the Richness and Abundance of Coastal Marine Fish Sounds. *Journal of Ecoacoustics,* 7 (1): 1-25, figs 1-5. doi/10.35995/jea7010001

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