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THE NEWSLETTER OF THE

ORTHOPTERISTS' SOCIETY

President's Message

ear Society members, Our 14th International Congress of Orthopterology in Mérida, Yucatán is only a few weeks away!

There is a very full program with 5 Plenaries, 10 Symposia (72 presentations), 2 Workshops, Meetings by the Mexican SENASICA and American continent GICSV locust management organisations, as well as oral presentations and posters under a number of subject headings. The scientific committee has done a commendable job in keeping similar subjects apart, but with so many interesting topics to hear and discuss, I am sure I will have to make difficult choices in what to take part in. The fact that there are so many promising presentations is a credit not only to the wide range of endeavors that our members are involved in, but even more so to Mario Poot and the organizing committee for their tireless efforts in ensuring that the 14th International Congress of Orthopterology will be a resounding success.

Thanks to the legacy left us by a former President, Ted Cohn, we have been able to provide substantial funds for travel and accommodation to our Congress: for Plenary Speakers, Symposium Organisers, Board members, and a number of students and young orthopterists. This is in addition to the Prizes and Awards that will be presented at the Congress in the Yucatán.

By DAVID HUNTER President

davidmhunter100@gmail.com



And of course, the Yucatán is a wonderful place for our Congress. As we did in Morocco, we plan to organize some day trips for accompanying persons during the Congress and a number of us, including myself, plan to take part in the post-congress tour. The tour will go out from Merida to a different region each day: to Uxmal with its pyramids, palaces, and "pelota" ball court; to the cenotes that served water sources for the Mayans, and to the biosphere reserve at Celestún. This support is all part of ensuring that as many of us as possible will be able to take advantage of the opportunity to get together in the most beautiful and interesting setting of Mérida and the Yucatán.

¡Bienvenido a Yucatán!

This will be my last President's Message, as during the congress I will

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be passing the "President's Hat" over to Axel Hochkirch, who I am certain will continue the success of our soci-

ety! Once again, enjoy another excellent *Metaleptea*, and thank you to Hojun Song, Derek A. Woller, and everyone who has contributed!

The 14th International Congress of Orthopterology: October 2023!

By MARIO A. POOT-PECH President, Organazing Committee mpootpech@gmail.com

he International Congress of Orthopterology draws near, and the history and flavor of the Mayan culture is already in the air. We are hosting events

that will be a mix of flavors: symposia, meetings, forums, cultural, and archaeological events. The Congress will be in the Hotel "El Conquistador" in Mérida City, Yucatán State, México. We have put together an excellent congress program which includes five plenary presentations, 10 member organized symposia, five contributed paper sessions, two workshops, and three focused meetings. The detailed program can be found as a PDF at https://ico2023mexico.com/program/.



OSF successfully migrated to the new platform TaxonWorks





ear Orthoptera Researchers, We are pleased to inform you about the

release of a major new version of the **Orthoptera Species File (OSF)**, during the first week of September 2023. This update marks a pivotal moment as OSF migrates to the TaxonWorks platform, aiming to provide a more streamlined and accessible experience for users.

About OSF:

OSF has been a reliable and comprehensive online database, serving as a reference for the taxonomic information and classification of the Orthoptera order. This resource has been valuable for researchers, policymakers, environmental managers, and the wider public in understanding the world's Orthoptera species.

What is New in the Release?:

The OSF team has been hard at work, meticulously migrating the database to the innovative TaxonWorks platform. Two products have been developed and supported by the **Species File Group**: 1) **TaxonWorks**, the software used for curating and accessing different filters and functionalities of the database, and 2) TaxonPages, the product to display the data to the public sourced from TaxonWorks' API.

This transition brings several enhancements to OSF:

• Enhanced Accessibility: The

By MARIA MARTA CIGLIANO

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> new public view is designed to be user-friendly from PC and mobile devices, making it easier for researchers at all levels to access the Orthoptera taxonomic information.

- Advanced Search and Analysis: TaxonWorks brings cutting-edge search and analysis capabilities, allowing researchers to explore deep into the taxonomy, morphology, and distribution of Orthoptera species.
- Efficient Workflows: Taxon-Works introduces digital tools that redefine how researchers interact with OSF, with streamlined workflows for capturing and analyzing Orthoptera diversity data.
- Collaboration and Data Sharing: The enhanced platform

fosters collaboration among researchers, encouraging the exchange of insights and data to drive the field forward.

- **Modern Data Management:** The transition to TaxonWorks ensures that OSF remains current and relevant in the ever-evolving digital landscape.
- **Continued Reliability:** OSF's reputation as a trustworthy resource for Orthoptera taxonomy remains unchanged, ensuring researchers have access to accurate and authoritative information.

Discover More:

We invite you to discover the new version of **OSF public view** where you will find resources to help you navigate the changes and take advan-





tage of the enhanced features. We also invite you to join our community via the Species File Group's regular meetings to learn more, which occur on the first Tuesday of each month - find more about it on the OSF homepage. We appreciate your ongoing support for OSF and your dedication to advancing Orthoptera research. As we move into this new phase, we are committed to maintaining the reliability and utility that you have come to expect from OSF.

Update on the Singing Insects of North America (SINA) Website

By TERESA YAWN Editor/Webmaster, SINA tmd@teresamariedreams.com

pecies distribution maps are an important feature of SINA (Singing Insects of North America). The maps are GIF or JPG images with dots super-

imposed on a map of most of North America. The dots represent county records where identified species have been found. Recently, a SINA user pointed out to me that, as they are, these maps are not useful for people who are blind or visually impaired. To address this, I am adding a section titled "Summary of Species' Distribution," and another section titled "List of States and County Records" to all SINA species' map pages. The summary gives a short description of the species' overall distribution. The list of records includes the states in which the species was found and the county records for each state. The states are organized according to geographic region. Both sections are located at

the bottom of the page. An icon at the bottom right corner of the map image says "MAP INFO" and links to the sections. I will update the summary and list of records when I add new records to the maps. These new sections will provide easy access to map data and are compatible with screen readers. You can see an example of these additions on the *Hapithus agitator*

map page.

Other projects I am working on include adding popovers and adjusting SINA for smaller screens. The popovers are for words on SINA that link to definitions on the SINA glossary page. If the definition has an associated illustration, a link goes to another page with the illustration. By using popovers, you will not need to navigate off the page you are reading. Words with popovers are highlighted in light blue. If you hover over or tap on the word, a popover will open, revealing the definition and an illustration, if there is one. The glossary will still be a part of SINA. You can see an example of a popover for the word "length" on *Cyphoderris*



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buckelli's **species page** and a popover with an illustration for the term "epistomal suture" on the Family Prophalangopsidae **page**.

Additionally, if you look at *C. buck-elli*'s species page on your cell phone or tablet, you will notice the content does not display as well as when viewed on a desktop computer. I have viewed this page on many types of smaller screens, and usually, the navi-

gation buttons are too small, the headings are very large, the font sizes are variable, and the audio bars are small. SINA was designed before small screens became so prevalent, which is why the website displays poorly on them. I am working on SINA's coding to make the website a more enjoyable experience on smaller screens, such as providing consistent formatting, properly sized buttons, and readily visible images and text. I want to ensure that all SINA users can easily navigate the website and access the information therein. And I would like the experience to be intuitive and user-friendly. Please continue sending me updates on SINA species, information on new species, and suggestions on how I can make SINA work better for you.

Updates from the Global Locust Initiative

By MIRA RIES¹ & RICK OVERSON² ¹Project Coordinator, GLI, miraries@asu.edu ²Co-director, GLI, roverso@asu.edu Arizona State University, USA

ike many of you, the Global Locust Initiative (GLI) team is preparing to attend the 14th International Congress of Orthopterology (ICO) in Mérida, Yucatán, México next month. We are looking forward to seeing many of the faces from our Global Locust Network, meeting new ones, and participating in several symposia. A tremendous thank you to the ICO organizers for their hard work to bring this congress together!

Together with Orthoptera Species File (OSF), the GLI is also hosting a workshop called **Advancements** on digital tools for the orthopterist community: Introduction to a new version of Orthoptera Species File and to HopperWiki.

OSF will present their new platform, TaxonWorks (TW), which integrates the best features of Species File Software with several new ones designed to help with the multiple tasks of taxonomists and other biodiversity scientists. The GLI will present our latest project, **HopperWiki**, currently available in beta mode. HopperWiki is set to serve as a centralized hub, allowing the global community to both access and contribute valuable information on locusts and grasshoppers, all presented in a user-friendly format and available in any language through Google Translate. We see the project as an opportunity for the global locust community to come together to create a shared resource written by a diversity of voices. A portion of the workshop will include a live demo and tour of the wiki followed by a discussion on how everyone can get involved and contribute to the project. We hope to see you there!

Regional Reports - What's happening around the world?

North America

By KATHLEEN KING Regional Representative USDA-APHIS-PPQ, Wyoming, USA butterfliekisses2u@hotmail.com

his year, the annual Entomological Society of America (ESA) meeting will be in National Harbor, Maryland from November 5-8. There will be lots of in-person activities at the meeting, as well as some online and on-demand content for those who can only attend remotely. For more information, visit the **ESA meeting** website.

The annual ESA branch meetings will be coming up in spring 2024:

- Eastern Branch, March 9-12, 2024, Morgantown, West Virginia, USA
- International Branch, April 8-10, 2024, Virtual
- North Central Branch, March 24-27, 2024, Fort Collins, Colorado, USA
- Pacific Branch, April 14-17, 2024, Waikoloa Beach, Hawaii, USA
- Southeastern Branch, March 17-20, 2024, Augusta, Georgia, USA

 Southwestern Branch, April 21-24, 2024, Albuquerque, New Mexico, USA

For more detailed information, please visit the **ESA branch meeting** website.

Theodore J. Cohn Research Fund Reports

How to make yourself heard: examining the biomechanics behind producing louder calling songs in *Acheta domesticus*

By MACKENZIE FARRELL

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n many cricket species, males produce calling songs to attract female mates from a distance (Alexander, 1961). The major purpose of calling songs is species recognition and conspecific mate attraction. Once males are in close contact with an individual female, they switch to producing courtship songs which involve a more detailed display of acoustic, visual and chemical stimuli. These courtship song characteristics may allow females to make detailed assessments of male quality (Alexander, 1961). However, the first interaction a female has with a potential mate is through the reception of a calling song. As we all know, first impressions matter! Producing an attractive calling song is essential for mating success, yet the biomechanics behind attractive calling song properties is not well understood.

Calling songs exhibit inter-individual variation in temporal features and overall song intensity (Bertram et al., 2011), and females are known to discriminate between males based on calling song features (Walker, 1957; Popov & Shuvalov, 1977; Hedrick, 1986; Stout & McGhee, 1988). The sound intensity of the calling song is an important feature as calling songs are used primarily to attract conspecific females from long distances, and louder songs can be detected at greater distances. Previous studies have shown that the intensity of calling songs is important for mate attraction in the wild (Nandi & Balakrishnan, 2013) and in laboratory settings (Pacheco & Bertram, 2014). As a result

of female preference for higher sound intensities, male cricket traits associated with producing louder calling songs may be subject to sexual selection.

Thanks to the Theodore J. Cohn Research Fund from The Orthopterists' Society, I was able to investigate the morphological and behavioral traits associated with producing high-intensity calling songs in order to determine the source of variation in calling song amplitude for the house cricket *Acheta domesticus*. Addressing the question of how males produce louder calling songs links studies on biomechanics, acoustics, and sexual selection, giving a more comprehensive understanding of mate attraction for *Acheta domesticus*.

Morphology

For the purposes of this study, commercial populations of Acheta domesticus were purchased from The Bug Company in Ham Lake, Minnesota. In order to identify the source of variation in sound intensity of the calling song I first examined the relationship between cricket morphology and sound intensity. To do this, I compared the average and maximum sound intensity of a calling male with key morphological traits (mass, pronotum width, size of key wing resonant structures, and the spacing of file teeth). Male crickets were isolated in individual containers, and their calling songs were recorded using either a Sennheiser microphone (MKH 8020, Germany) connected to a Zoom F4 Pro digital Audio Recorder, or using a ¹/₄ inch B&K microphone (Type



Figure 1. Cricket wing structures. For each cricket, the harp and mirror area were measured to estimate the total resonance area of the wing. Furthermore, the length of the file and the spacing between file teeth was also recorded.

4189, Denmark) interfaced with a B&K microphone pre-amplifier (Type 1708, Denmark). Absolute sound pressure levels (dB SPL) of cricket songs were determined at a distance of 10 cm from the source using two approaches. For the majority of crickets, the sound pressure level was calculated based on the sensitivity of the Sennheiser microphone (MKH 8020, Germany). For the remainder of the recordings, the dB SPL was determined by reading values from the B&K sound level meter (LCFeq) equipped with a ¹/₄ inch Type 4189 microphone.

After obtaining acoustic recordings, measurements of key morphological features were taken for each cricket. Mass and pronotum width were used as measures of cricket size and body condition. Each cricket was massed using a Sartorius digital scale (EN-TRIS1241-1S, Germany). Crickets were then photographed alongside a measurement scale, which allowed for measurements of the pronotum width



Figure 2. Set-up for synchronized audio and video recordings Isolated male crickets were housed in circular containers with a mesh top. The dB SPL and audio of the cricket calling song was recorded from 10cm above the cricket. The video camera was level with the cricket and angled to obtain a head-on view of the singing cricket. An IR light was placed behind the cricket for optimal camera lighting.

via analyses in ImageJ. Cricket wings were removed and photographed using a dissecting microscope (Olympus SZX2-ILLT) equipped with a camera that interfaced with CellSens (v2.3). These images were taken with a calibrated field of view, which allowed for measurements of the harp and mirror area, the file length, and the spacing between file teeth on each wing (Figure 1).

In agreement with previous research on Acheta domesticus (Gray 1997) there was a significant tendency for male size to predict the sound intensity of the calling song. Crickets with greater mass (r(34) = 0.45, p = 0.005)and larger pronotum width (r(35) =0.39, p = 0.017) performed calling songs at higher sound intensities. Furthermore, the intensity of the calling song does predict the male's body condition (r(34) = 0.45, p = 0.006). Body condition was estimated using Peig and Green's (2009) scaled mass index. This index uses Standard Major Axis Regression which better represents the relationship between mass and size when compared to Ordinary Least Squares Regression (Kelly, et al. 2014). Body condition provides a better estimate of male health and mating quality than mass or pronotum width independently. Therefore,

female selection for louder calling songs may be the first step to evaluating the quality of a potential mate.

Although the size of the cricket is related to the sound intensity of the calling song, the size of key wing resonant areas were not. Neither the right wing harp area, right wing mirror area, left wing harp area, left wing mirror

area, or all resonant areas combined (r(24) = -0.01, p = 0.95) were significantly correlated with the sound intensity of calling songs. Furthermore, crickets with wing damage (naturally acquired prior to recordings) did not sing at significantly different sound intensities than crickets with intact wings (t(20.1) = 0.52, p = 0.60). In the Pacific field cricket, Teleogryllus oceanicus, evolutionary changes in wing structure are related to changes in the acoustic properties of the calling song (Tinghitella, et al. 2021; Tinghitella, et al. 2018; Zuk, et al. 2006). However, it may be the case in A. domesticus that there is not enough variation in wing morphology for the size of resonant structures to be a limiting factor on sound intensity.

Motivation

Since morphology was not a limiting factor of calling song intensity, I next wanted to examine how a cricket's motivation to sing affected song production. Using the level of male-male competition as a proxy for motivation to find a mate, I housed crickets in three separate 16x10" habitats with varying levels of mate competition. The first group consisted of 100 males, the second group had 75 males and 25 females, and the third group had 50 males and 50 females. The level of mate competition did not affect crickets' motivation to sing, as estimated by the number of males from each treatment group who sang per minute (F(2,27) = 0.362, p =0.699). The level of mate competition also did not affect the sound intensity of the calling song (F(2, 9) = 1.8, p)= 0.2). It may be the case that for a commercial population, even when temporarily exposed to varying levels of mate competition, this does not provide enough motivation to significantly alter singing behavior. Housing crickets in these treatment groups for multiple generations may be beneficial for further research on the role of male-male competition on calling song properties.

Biomechanics

The third component of this research project involved examining stridulation biomechanics for Acheta domesticus. I recorded synchronized audio and high-speed videos of cricket stridulation to establish the relationship between the rate of wing closure and the amplitude of sound produced. Developing the method for motion tracking and synchronized sound production was a large component of this project (Figure 2). Isolated male crickets were housed in circular containers with a mesh top. The SPL and audio of the cricket calling song was recorded from 10cm above the cricket using a B&K sound level meter (LCFeq) equipped with the ¹/₄ inch Type 4189 microphone, and a ¹/₄ inch B&K microphone (Type 4189, Denmark) interfaced with a B&K microphone pre-amplifier (Type 1708, Denmark). High speed video recordings were taken using a Chronos High Speed Camera (CH14-1.0-16M, 1069fps. The audio and video were synchronized using custom software developed by Dr. Norman Lee in MATLAB (R2019a, Massachusetts) and a National Instruments Data Acquisition system (NI USB-6221, Hungary). Then using Tracker (v6.1.2) the horizontal and vertical velocity of the



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Figure 3. Velocity of wing closure predicts sound amplitude. In an eight second recording of an individual male the amplitude of each sound pulse was significantly correlated with the speed of wing closure. (top) Faster speeds of wing closure were associated with louder sound pulses (amplitude = speed*(0.0072) + 0.014). (bottom) The top panel includes an oscillogram with two sound pulses. The next two panels describe synchronized velocity of wing motion in the horizontal and vertical directions. A velocity >0 indicates the wings are moving toward each other for wing closure, a velocity < 0 indicates the wings are opening away from each other. A faster velocity of wing closure in the horizontal direction (along the direction of the file) is associated with greater sound intensities.

0.10

Time (s)

right wing plectrum was calculated for all 8700 frames. The wing velocity was then compared with an oscillogram of the calling song using RStudio (v2022.12.0+353).

0.05

1.0 -

0.9

0.8

0.7

0.6

0.5

0

-1

100

0

-100 100

0

-100

amplitude

velocity (mm/s) velocity (mm/s)

0.017

Horizontal Motion

Vertical Motion

0.00

0.018

0.019

0.020

Time of Wing Closure (ms)

0.021

0.022

0.20

Maximum Relative Amplitude of Sound Pulse

Although morphology is not a limiting factor of sound intensity, behavioral factors can influence sound production. An analysis of wing motion during stridulation synchronized with acoustic recordings demonstrates that louder pulses are produced by faster velocities of wing closure (Figure 3). Additional data collection and statistical analyses for this component of the project are still ongoing, but further research into the mechanics of stridulation and energy input is essential to better understand how male crickets produce louder calling songs.

0.15

Acknowledgements

I am very grateful to the Orthopterists' Society for the financial support through the Theodore J. Cohn Research Fund. I would also like to thank Dr. Norman Lee for his continued guidance and support throughout this project.

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 Phenotypic covariance structure and its divergence for acoustic mate attraction

Mate recognition in sympatric species of endemic New Zealand alpine grasshoppers

By MARI NAKANO

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some high-elevation New Zealand habitats (>1200 m above sea level: m.a.s.l.), the endemic alpine grasshoppers (Orthoptera: Acrididae), Brachaspis nivalis, Sigaus australis, and Paprides nitidus (Fig. 1) co-occur in high density without hybridization (Watson 1970). This is possible because sympatric species possess sophisticated communication mechanisms for successful mate recognition. Unlike some acridid species (Song et al. 2020), New Zealand alpine grasshoppers do not actively sing during mating season (Watson 1970) and show color patterns that match their habitat (i.e., camouflage). Therefore, it is unlikely that they use acoustic or visual signals to locate and select mates. However, as conspicuous antennal movements have been observed in male grasshoppers when pursuing females (Watson 1970) and chemical sensitive sensilla (sensory organs) have been observed on their antennae (Nakano et al. 2023), it is probable that these grasshoppers use chemical signals to locate and discriminate their mates and food plants.

Insect-derived chemical signals include volatiles and cuticular hydrocarbons (CHCs). CHCs are often referred to as contact-chemicals or short-range-olfactory in contrast to volatiles which are considered as long-distance-olfactory signaling (Nakano et al. 2022). This is due to the higher molecular weight and lower volatility of hydrocarbons compared to volatiles. CHCs typically comprise 20 to 40+ carbon atoms comprising alkanes, sometimes including double bond(s) (e.g., alkenes and alkadienes) and/or methyl branches (Blomquist et al. 2018; Gibbs and Rajpurohit 2010;



Figure 1. Grasshopper coloration of three alpine New Zealand species helps with camouflage: (A) *Brachaspis nivalis* male, (B) *Paprides nivalis* male, and (C) *Sigaus australis* female.

Menzel et al. 2017). Grasshoppers use both volatiles and CHCs to communicate. Flighted species that can disperse long distances (e.g., locusts, >100 km a day) rely on long-distance volatiles while species that do not travel long distances (e.g., *Chorthippus* grasshoppers, <3 m: Bailey et al. 2003; Ortego et al. 2021; Tim and Hill 2004; Weyer et al. 2012) are reliant more on CHCs. New Zealand alpine grasshoppers are flightless and are therefore likely to rely on CHC for intraspecific communication.

The purpose of this research is to elucidate the chemical signaling system in sympatric alpine grasshopper species by analyzing their CHCs and volatiles. I speculate that the grasshoppers have species- and sex-specific chemical profiles and show higher olfactory responses to the smells of their conspecific females than to heterospecifics. This research provides a better understanding of communication systems in the Acrididae and adds support towards explaining the maintenance of their taxonomic diversity.

Methods

Adult grasshoppers of *B. nivalis*, *S. australis*, and *P. nitidus* were collected at Broken River Ski Field (-43.125929, 171.687432), South Island, New Zealand, during their active summer season (February), with approval from the operators of the ski field and Department of Conservation (authorization number: 97397-FLO). To collect cuticular hydrocarbons (CHCs) (n=7 for each sex per species), live specimens were transported to the laboratory at Massey University (Palmerston North, New Zealand), and kept at 4 °C with their food plant (Celmisia spectabilis and Celmisia lyallii) with natural light prior to CHC collection. Grasshoppers were frozen for 1 day at -20 °C and each submerged in 1 mL of hexane containing 10 ng/µL of nonyl acetate as an internal standard (IS). To collect volatiles, full-body headspace collections were performed (n= 4 for each sex per species) for three timeframes over nine hours (07:00-10:00, 11:00-14:00, 16:00–19:00) with ambient light and temperature at the ski field. These time frames were used to observe changes in chemical profiles over time (if any) when the grasshoppers are active. CHC and volatile samples were analyzed using a Gas Chromatograph - Mass Spectrometer (GCMS-QP2010, Shimadzu Corporation, Kyoto, Japan).

Electroantennogram (EAG) analysis was performed with synthetic oleamide (No. O2136, Sigma-Aldrich) at Plant and Food Research, Lincoln. Ten microliters of oleamide dissolved in 95 % ethanol was placed on piece







of filter paper, and the ethanol allowed to evaporate for 10 seconds in a fume hood before being loaded into a Pasteur pipette (Sigma-Aldrich). A range of oleamide concentrations used were: 0.1 mg/mL, 1mg/mL and 5.99 mg/mL. EAG responses to stimuli were recorded from lower to higher doses, and a blank air puff and solvent control (95 % ethanol) were used at the beginning of the experiment to compare the response between these controls and the test stimuli (oleamide).

Results and Discussion

Fatty amides (oleamide and octadecanamide), *n*-alkanes, and methylbranched alkanes with chain lengths between C25 and C37 were present in cuticular extracts of four endemic New Zealand alpine grasshopper species. Analysis of the head-space collection revealed that no volatile compounds were consistently emitted by the grasshoppers, but terpenoids and green leaf volatiles were detected from some individuals. These compounds are probably derived from plants.

n-C31 and methyl-branched C33 and C34 were abundant in *B. nivalis* (peaks 8, 11–15 in Fig. 2). *Sigaus australis* and *P. nitidus* had similar CHC profiles but 11MeC35 in *P. nitidus* (peak 17 in Fig. 2) dimethyl-C35 in *S. australis* (peak 18 in Fig. 2) and were observed in higher quantities than in other species. Fatty amides were present in relatively high quantities in extractions from the female grasshopper cuticles but not in males (peaks 1–4 in Fig. 2). These species- and sex-specific abundance of particular compounds may indicate that New Zealand alpine grasshoppers use this information from CHC profiles as contact or shortdistance cues for differentiating males from females and recognizing potential mates.

Electroantennogram (EAG) responses to oleamide (dissolved in 95 % ethanol) were similar to those of 95 % ethanol in B. nivalis, S. australis and P. nitidus (Fig. 3), indicating no olfactory response to oleamide. The intensity of EAG responses to oleamide at all dosages and 95 % ethanol were two to three times more than that of the air puff. These results showed that oleamide did not elicit an electroantennogram response by males or females of B. nivalis, S. australis and P. nitidus despite its abundance within female CHCs. This observation suggests that these grasshoppers might not use olfaction to perceive oleamide, but direct contact (gustation) instead may be involved in the recognition of oleamide. Further studies involving behavioral and electrophysiological analyses are required to fully understand the sexual communication systems in New Zealand grasshoppers.

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Figure 3. Example electroantennogram responses to air puff (1), 95 % ethanol (2), oleamide at 0.1 mg/mL (3), 1 mg/mL (4) and 5.99 mg/mL (5) of 95 % ethanol in: (A) *Brachaspis nivalis* male, (B) *Sigaus australis* female, and (C) *Paprides nitidus* male.

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Extreme environments impacting the evolution of reproductive traits in the lava cricket

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urvival of organisms in extreme environments has fascinated scientists and laymen alike, with studies attempting to understand the limitations of life and

the kinds of adaptations that are needed to survive in extreme habitats. For example, freeze-tolerance has evolved multiple times, allowing insects to survive cold temperatures (Sinclair et al., 2003). Likewise, adaptations altering hemolymph physiology and homeostasis allow numerous aquatic insects to survive in extremely acidic conditions (pH \sim 3-4 for the lesser waterboatman, Corixa punctata, and the chalk-fronted corporal, Libellula julia; Morris 1989; Rockwood and Coler 1991). For persistence in otherwise inhospitable environments adaptations to reproduction are essential, yet we know comparatively less about how this function evolves in extreme conditions.

In the Hawaiian Islands, the endemic lava crickets in the genus Caconemobius (Orthoptera: Trigonidiidae, subfamily Nemobiinae) are an apt example of extremophilic organisms (Fig. 1). They are often the first multicellular colonizer of barren lava fields (Howarth & Mull, 1992; Otte, 1994) and previous work in our lab shows they persist on stillsparsely vegetated flows up to 220 years old (Heinen-Kay et al., 2021). These crickets have physiological adaptations that seem to facilitate their survival on the large expanses of bare rock. Lava crickets have increased resistance to desiccation and are only active on the surface at night (Ahearn & Howarth, 1982), presumably to cope with the dry climate and daytime surface temperatures over 50°C.



Figure 1. Caconemobius fori ('ūhini nēnē pele in Hawaiian), the endemic Hawaiian lava cricket. (NPS Photo/D. Boyle)

While the physiological adaptations that enable survival in this challenging environment have received some



Figure 2. Working in the Hawai'i Volcanoes National Park. Photo by John Rotenberry.

prior research attention, the behaviors and physiological traits involved in lava cricket reproduction remain unclear.

Lava crickets face challenges to reproduction distinct from other cricket species in their family. Lava crickets are wingless, a common adaptation seen in island insects (Howarth & Mull, 1992; Leihy & Chown, 2020; Wagner & Liebherr, 1992) and, thus, cannot use stridulation to attract conspecifics. Lava crickets are in the subfamily Nemobiinae (Orthoptera:Trigonidiidae) and most members perform a nuptial gift-giving behavior termed spur-chewing. Females will consume hemolymph from a specialized tibial spur on the male, of up to 8% of the male's body mass (Fedorka & Mousseau, 2002; Fulton, 1931; Mays, 1971) which increases the female's reproductive output (DiRienzo & Marshall, 2013). This may not be possible in the hot, dry, environment of the lava flows where conserving body fluid is imperative. Faced with infrequent nutrient availability, obtaining sufficient resources to allocate to offspring (which can be instrumental in determining offspring survival (read Roff 1993 and references therein) may be limiting for lava cricket persistence. In this case, allocating those few resources to a

few large offspring or many variable offspring could both potentially allow for population persistence. To better understand how lava cricket reproductive traits may enable their survival in the barren lava flows of Hawaii, the money provided by the Theodore J. Cohn Research fund enabled me to collect crickets from the Hawaii Volcanoes National Park on the Big Island (Fig. 2) and looked at reproductive output of adult females by quantifying the number and size of eggs they possessed. We also looked for evidence to support the spurchewing behavior by examining male tibial spur structure.

We found that these females possessed few and large eggs relative to their body size, with averages of 3-4 eggs per individual compared to other crickets in the Nemobiine family, such as the Allard's ground cricket of a similar size that we collected on the University of Minnesota campus, where wild-caught females possessed averages of 36 eggs. We did not find any evidence for the specialized male tibial spur previously reported in other Nemobiinae species. This suggests that the males may not perform the gift-giving behavior. This may represent a high cost of losing hemolymph in a challenging environment, thus leading to the loss of this

behavior. However, it is unknown where in the Nemobiinae lineage that the spur-chewing behavior appeared, necessitating the need for comprehensive phylogenetic mapping of the Nemobiine family to aid in determining the prevalence of this behaviour. We also found that lava crickets possess long ovipositors relative to their body size compared to other Hawaiian nemobilines, such as the cave-adapted Caconemobius species, suggesting that these crickets have to lay eggs deeper in the substrate to avoid drying out, similar to ovipositors seen in mainland crickets.

While trapping these crickets in the field we often captured them in groups rather than as single individuals, which may point towards these crickets residing in groups in the field (Poisson dispersion parameter = 5.7). Crickets are typically not social creatures, however, cave crickets have been observed congregating around plant roots and food sources (personal communication from Megan Porter). While being able to locate food is of incredible importance in the field, locating and choosing between conspecifics of the opposite sex is also imperative to maintain populations. Given their incredibly long antennae, it stands to reason that lava crickets are using both chemosensory and mechanosensory cues to navigate their environments. Future research with these crickets revolves around trying to understand whether chemosensory cues play a role in locating conspecifics, specifically in betweensex interactions.

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Amazonian Proscopiidae: natural history, biology, rearing, and collection of four taxa in three municipalities of Amazonas

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roscopiidae is a family of grasshoppers with few specialists, and since its description in 1838, there have been few studies on their natural history. Aguilar (1974) published "Los palitos vivientes de Lima," which discusses the natural history of Anchotatus peruvianus Brunner von Wattenwyl, 1890. There are also some studies on rearing, ecology, and embryonic development of Stiphra robusta Mello--Leitão, 1939, motivated by its economic importance as a pest species in various plantations, such as eucalyptus and cashew trees in northeastern Brazil (Moraes et al. 1983; Flechtmann & Ottati 1997; Sergio Vicentini 1999; Lima & Andrade, 2002).

Anchotatus peruvianus is from Lima, Peru, in the region of Lomas de Atocongo, an area characterized by hills, defined seasons, low-statured trees, and abundant ground vegetation, similar to the habitat where S. robusta lives. Stiphra robusta is a species that is exclusively found in northeastern Brazil, in a dry and arid climate, but with low ground vege-



Figure 1. Map with collection points sampled during study.

tation and small trees. Thus, so far, Proscopiidae have been described as grasshoppers that inhabit open spaces, such as savannas and plains, where specimens are collected on the ground or on many species of plants, including herbs, shrubs, or trees.

However, this description is far from my reality. I study Proscopiidae in the Brazilian Amazon, in a region where trees can reach over 50 meters in height, forming dense and humid forests. From the beginning of my academic career, I felt the lack of studies describing behavior or any

information that would facilitate the collection of these individuals here in the Amazon.

Dr. Alba Bentos-Pereira had a hypothesis that Proscopiidae in the Amazon are arboreal (personal communication). However, this theory could never be explored due to her research being conducted with material that did not explicitly specify the stratum in which the specimen was collected. From this, my desire to undertake a study that answers where and when these grasshoppers can be found in the forest emerged.



Figure 2. Males of the four different taxa studied during this project. A - Possible new genus, B - Pseudoproscopia amedegnatoi, C - Milenascopia bilineata, D - Carbonellis sp.



Figure 3. Photo of the couple from Balbina, Presidente Figueiredo, found on the Flickr website in 2013. Photo: Pedro Ivo.

In this way, the Theodore J. Cohn Research Fund gave me the opportunity to run a project where I would conduct monitoring in the Adolpho Ducke Reserve, a study site with approximately 4 km² of Amazon rainforest located in the city of Manaus. The project aimed to collect biological and behavioral information about these grasshoppers in their natural habitat, conduct captive breeding to gather supplementary data, and correlate the species collected with environmental variables from the reserve that are available online.

Unfortunately, it was not possible to implement the project in the Adolpho

Ducke Reserve due to the difficulty of finding Proscopiidae in the region. During three months of searching in the area, my team and I did not find any of these grasshoppers. Therefore, we chose to adapt the project to three new locations: the municipality of Iranduba, Manaus (at the Federal University of Amazonas - UFAM), and Presidente Figueiredo (in the village of Balbina), both cities in the state of Amazonas (Fig. 1).

The choice of the new locations brought a significant benefit to the project as it allowed us to collect data on the biology and behavior of twice as many taxa as the initial project. This is because the Adolpho Ducke Reserve only has records of *Pseudoproscopia* and *Carbonellis* (Fig. 2B, D), and in addition to these two, we found *Milenascopia* and a possible new genus (Fig. 2A, C). However, it was not possible to correlate the collected species with environmental variables since the new collection areas do not yet have data on environmental variables.

The points in Iranduba and Manaus were chosen because they were locations where I knew for certain that I would find Proscopiidae and Balbina was selected for a special reason. In 2019, I came across a photo from 2013 on **Flickr** by the photographer Pedro Ivo (see Fig. 3) of a couple with a coloration that had never been seen in the region. I suspected it might be a new taxon due to the lack of a similar description. Therefore, I seized the opportunity to explore the area and determine the identification of these grasshoppers.

The monitoring was conducted from September 2022 to August 2023. For the initial monitoring, we proposed a methodology that involved delineating an area of 250 meters throughout an entire year. We conducted inspections for 1 hour every 2 hours, during a total 24 hours a period, to observe the Proscopiidae. As a result, the monitoring took place at even hours, at 00:00, 02:00, 04:00, 06:00, 08:00, 10:00, 12:00, 14:00, 16:00, 18:00, 20:00, and 22:00, covering a complete day of observation.

This methodology was only possible to carry out in Iranduba because Proscopiidae individuals there live at heights close to the ground, which increases detectability. This fact is not true in other locations. In Balbina, for example, it took us 4 months to find the first male (I won't be able to describe the happiness that was, but there was even a promise to stop eating chocolate). All males found were coming down from higher stratas, which suggests that individuals do indeed live in the canopy, a stratum to which we did not have access.



Figure 4. Breeding cages with males and females separated.

In addition to the monitoring, we also collected some specimens for captive breeding. All of them were raised in my home for a year, where they received natural light and were exposed to the natural humidity of the city. Initially, I attempted to breed them in a laboratory, but the lower temperatures and dehumidifiers certainly had a negative impact on the breeding. Currently, I still have 14 Proscopiidae individuals in captivity, and they live in cages I constructed myself, measuring at least 67 cm in length, 35 cm in width, and 46 cm in height (Fig. 4).

Through this monitoring and breeding, we were able to discover that:

- All of them have generalist tendencies because they were fed with cashew tree, mango, and starfruit leaves during breeding. In contrast, in nature, they were observed feeding on plants from different families.
- They are diurnal, highly active

during the day, and that is when they feed the most. In all three monitoring points, it was observed that during the night, they choose a resting spot, except when they are mating. For these grasshoppers, it doesn't matter if it is day or night, it is always time for romance. • Mating

can last up to 14 hours, with males remaining on top of the female for many hours afterwards. This behavior may be due to the fact that males are highly territorial when it comes to females. When males are housed together without females, they can coexist in the same

cage. However, when a female is introduced to the enclosure, they engage in relentless fighting. They begin to pursue and kick each other while simultaneously moving towards the female. Males that manage to mate with females do not let go of their manly duties, even if they have to hang upside down while being challenged by another male. Females, on the other hand, do not display territorial behavior.

I have never been able to get a female to lay eggs in a substrate. We used soil, sand, and sawdust, and this may be related to a theory proposed by Dr. Bentos-Pereira that the presence of fine denticulation on the edges valves of some species may indicate a tendency towards endophytic oviposition (Bentos-Pereira, 2006).

- Males and females mimic a swaying branch in the wind to better camouflage themselves in the environment, similar to Phasmida. However, they exhibit a differential behavior, which is taking advantage of windy moments to move more quickly. We observed that when the vegetation is still (no wind) the Proscopiidae move with great caution and slowly. However, when the wind starts to move the vegetation, the Proscopiidae begin to run. The moment the vegetation returns to a resting state, these insects also reduce their activity.
- During mating, males in nature often spread their hind legs and also sway, mimicking a branch in the wind. Nymphs exhibit a behavior of remaining still (even if they were about to be or were, in fact, touched) and camouflaged, hoping to be invisible, similar to adult females. In contrast, the males tend to display more evasive behavior. Rarely have I observed them jumping; usually, they throw themselves to fall into the leaf litter and disappear among the leaves.
- These grasshoppers are definitely arboreal. During our monitoring, we observed the Proscopiidae up to a height of 10 meters. However, in the collection at INPA (National Institute for Amazon Research), we found a female of



of the ovipositor Figure 5. Pair of Carbonellis in copulation.



Figure 6. Popularization of science during Environmental Awareness Week with the breeding of Proscopiidae conducted at the Bosque da Ciência (Science Park) at the National Institute for Amazon Research - INPA. Photos: Raphael Heleodoro.

the same species from Balbina (possible new genus) collected in a malaise trap at a height of 32 meters, and a female of an unknown taxon that was also collected in a malaise trap at a height of 40 meters.

- This information can also be confirmed through a vegetation suppression survey that happened in the city of Manaus. During this survey, it was possible to find on several fallen trees, ranging from 20 to 30 meters in height, up to 8 individuals of *Pseudoproscopia* living together. This is very different from the numbers found during the monitoring where we could only search where our eyes could reach, and most of the time, we only found one or two individuals per monitoring session.
- These grasshoppers can be found throughout the year. Balbina and Manaus were the places where we

found the fewest Proscopiidae and the frequency was very similar in both locations, with at least one individual found in all collections. In Iranduba, the minimum sighting number was of 4 individuals in January 2023, and the maximum was 15 individuals in June.

In addition to all the above, much more data were collected that will be properly elaborated on in future articles. These data will be presented in graphs in order to illustrate our monitoring methodology applied in Iranduba.

This project has not only formed

the basis for an upcoming natural history article, but will also contribute to future research, including the description of the new genus, which will be completed after the results of the phylogenetic analysis I am conducting. It has also led to the discovery that Carbonellis includes a species belonging to another genus, based on male genitalia and female morphology, which was better understood during the breeding process (Fig. 5). Additionally, a study on the spermathecae of females from the same population will be conducted to determine whether they can truly be used to differentiate species within a genus.

Furthermore, this project has been showcased in science events at the National Institute for Amazon Research during Environmental Awareness Week, where the breeding program was brought to the public in order to educate people about the existence of these grasshoppers and the differences between them and Phasmida (Fig. 6).

With this project, we hope that more research on Proscopiidae will emerge, especially in the fields of ecology and behavior, and that it will serve to attract and assist future scientists. In conclusion, I would like to express my gratitude to several individuals, without whom this project could not have been carried out. Among them are: Fernando Domenico, Daniela Santos, Raphael Heleodoro, Matheus Bento, João Rafael, Renato Azevedo, Anthony Ferreira, Márcio Oliveira, Eduarda Viegas, Natália Reategui, Daniel Lima, Michaela Fialho, Juliana Queiroz, Camila Silva, Michelle Queiros, the Rebio Uatumã team, and all those who, in some way, contributed to the progress of the project.

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Sky-islands under threat: an evolutionary study of the endemic bushcrickets of the genus *Parnassiana*

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reece may be famous for its sea and many islands, but it is also a mountainous country with long mountain ranges and a large num-

ber of high-altitude summits, many reaching above 2000 m. These are often isolated and host a plethora of endemic species, forming sky islands and presenting a great natural laboratory for evolutionary studies (Dodge, 1943). This mosaic of biogeographical islands has led to a large number of endemic species and to the characterization of the Southern Balkan Peninsula as a Palaearctic biodiversity hotspot (Hewitt, 2000; Hughes and Woodward, 2017; Kenyeres et al., 2009; Tzedakis, 1993).

An inhabitant endemic to the sky islands of Greece is the bush-cricket genus *Parnassiana* of the family Tettigoniidae (example species in Fig. 1).



Figure 2. The distribution of the genus Parnassiana in the southern Balkans



Figure 1. Females of *Parnassiana tymphrestos* (left) and *Parnassiana coracis* (right), two endemic species from Greece

It is a micropterous genus with low dispersal abilities which, combined with a number of factors, such as its requirements for specific mountainous climate and the complex geological history and geomorphology of Greece, results in a large number of isolated populations with small distribution ranges (Willemse and Willemse, 2008). It is restricted to the high altitudes (usually above 1600 m) of the isolated mountain summits

of the Pindos range in Greece and its extensions in the Peloponnese and Evvoia island (Fig. 2) (Willemse et al., 2018). Some populations from Albanian mountains have also been suspected of belonging in Parnassiana. Currently Parnassiana consists of 13 known species and a number of populations with unclear taxonomic status that may represent taxa new to science (Fig. 3) (Willemse and Willemse, 2008; Willemse et al., 2018).

Our study aimed to clarify the taxonomic status of the *Parnassi*-

ana taxa and populations, and to infer their phylogenetic relationships using morphological studies and molecular analyses. The clarification of the taxonomy, distribution, and evolutionary history of Parnassiana will additionally inform current efforts to study the ecology and conservation status of the genus and of the orthopteran communities of Greek mountains, whose ecosystems are currently under threat due to climate change, changes in grazing regime (traditional goat and sheep grazing, which conserved and maintained these subalpine ecosystems in the past, is being abandoned or replaced by destructive cow grazing), as well as the planned construction of large-scale wind farms (RAE, 2023).

Parnassiana specimens were collected in two field trips during the summers of 2021 and 2022 in the mountains of the Pindos range, Evvoia island, the Peloponnese, and Albania, and stored in absolute ethanol. Collected specimens were morphologically examined and genomic DNA was extracted from hind legs.

The morphological examination of the material took place in the IBER-BAS laboratory using a stereoscope with an attached camera. Male and female genitalia were extracted when necessary, photographed and compared with literature (Heller and Willemse, 1989; Massa and Fontana,

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Figure 3. (a) Female habitus and (b) male titillators of *Parnassiana* specimens from Agrafa mts, that possibly represent a species new to science

2011; Willemse, 1984; Willemse and Willemse, 1987) and the Orthoptera Species File (Cigliano et al., 2021). To construct the phylogenetic relationships of the taxa we amplified one nuclear (ITS1-5.8S-ITS2) and two mitochondrial (NADH2, COI) molecular markers, representing populations from 19 Greek and Albanian mountains, as well as some representatives from other genera of Platycleidini. Phylogenetic trees of the concatenated genetic markers were created by using BI and ML analyses, and showed strong support. Here, the tree of the concatenated NADH2 and COI is presented, as ITS showed poor phylogenetic resolution and was excluded from the analysis (Fig. 4).

Our findings from the combined morphological and molecular study showed that many populations have intermediate characters to the described species, while others proved to differ from the previously described taxa and probably warrant the description of new species or subspecies.

The *Parnassiana* taxa from Greece included in the molecular analysis

form a monophyletic clade that is clearly separated into two branches: the *Parnassiana* populations of Peloponnese and the populations of mainland Greece.

As shown in Figure 4, there are multiple populations with unclear taxonomic status. Most notably, the populations from the mountains of Voutsikaki, Agrafa, and Avgo each most likely represent new species for science. Avgo mountain, in particular, hosts two distinct taxa, one shared with the mountain of Agrafa and one closely

related to the population from Voutsikaki (not shown on the concatenated phylogenetic tree). These cannot be related to currently described species,

EgiuliaeFres Egiuliae6WReth



These findings indicate the existence of at least four new species, including the non-*Parnassiana* populations from Albania, whose generic status is yet to be revised. The complicated patterns, intermediate populations and low genetic distances between the *Parnassiana* taxa also reflect a case of ongoing speciation.

Our work will continue with a detailed morphological revision and description of the new taxa. Next, additional analyses will be conducted, aiming at revealing the evolutionary history and biogeography of the genus, as well as outlining current and future trends.

Acknowledgements

We are grateful to the Theodore J. Cohn Research Fund for covering the expenses of the fieldwork and a significant part of the Sanger sequencing costs. This study would not have been possible without this help!



Figure 4. Phylogenetic tree of the genus *Parnassiana* inferred from concatenated NADH2 and COI sequences with BI analysis. Each species is represented with a different color

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Discovery of a hybrid zone in Hawaiian crickets opens windows to understand their speciation

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arwin described the process of the origin of species as the "mystery of mysteries." 150 years after his seminal contributions to the field

of evolutionary biology, the study of the origin of species, or speciation, is still an active area of research. A lot of work has been done describing reproductive isolating barriers in many species groups, but understanding the genetic architecture of these barriers have been lacking [1]. Questions on the number, identity, and effect sizes of the genes involved in speciation remain and will give us a holistic picture of the mechanisms involved [2]. The advent of new sequencing technologies and the decreasing cost of sequencing have made it easier to pursue such questions and speciation genetics has been a burgeoning field.

The Hawaiian flightless swordtail crickets of the genus *Laupala* provide a tractable system to answer questions on the genetics of speciation. The group has radiated to 38 species across the Hawaiian archipelago, resulting in the fastest known speciation rates in invertebrates [3]. Different species are ecologically and morphologically cryptic, but they differ in their sexual signals: male songs and cuticular hydrocarbons (CHC, non-volatile carbon compounds present on insect cuticles). In Laupala, and crickets in general, males sing to court females and different species sing distinctively. The songs of each species have different pulse rates (some are slow and some fast singers) and evidence-to-date shows that females strongly prefer conspecific songs over heterospecific songs [4]. Male singing and female phonotaxis are important initiating



Figure 1. Landscape of Palikea Peak. Inset: Collection sites: Pig wallow and Summit Area are on Palikea Peak. Kipahulu and Makapipi are separate sites where *L.orientalis* is found.

steps for the courtship rituals of these crickets and the divergence of these

traits serve as important pre-mating barriers to reproduction [5]. This makes male song and female prefer-



Figure 2. A Laupala cricket in the wild.

ence important traits to understand speciation in this group. Multiple quantitative trait locus (QTL) studies have shown that these traits are due to many genes of small effect, and some genes underlying male song and female preference are co-localized in the genome [6], [7]. QTL fine mapping and differential expression studies have given us some promising candidate genes, which are involved in neurotransmission and modulation [8], [9].

Quantitative genetics methods have provided important insights into the genes and genetics underlying speciation phenotypes in Laupala crickets. I want to complement these studies using a population genomics approach, harnessing the power of genome scans and FST outlier analyses. These methods help identify loci in the genome under divergent selection, which is a key step in understanding the process of speciation [10]. However, genomic differentiation studies often reveal large stretches of the genome that are different, thus making the search for speciation genes difficult. However, comparing populations with ongoing gene flow makes this process easier because these "genomic islands of speciation" stand out in contrast to the rest of the genome, which is homogenized due to gene flow. This is where hybrid zones come to the picture.

Hybrid zones are defined as areas where populations of different genetic ancestries meet and mate, producing offspring of mixed ancestries. Since genetic ancestry is not visible to the naked eye, we often rely on phenotypes to characterize populations. Comparing populations with different phenotypes near a hybrid zone should help us discover these genomic islands of speciation in the face of ongoing gene flow. With all this in mind, we set out to characterize the first-ever discovered hybrid zone in *Laupala* crickets in East Maui, Hawaii.

The Theodore J. Cohn Research Fund awarded by the Orthopterists' Society funded my collection field trip to this field site in East Maui for a week. We hiked the ~ 2250 ft Palikea Peak and collected crickets at different transects. Figure 1 shows the field site, and the inset shows the location in Maui. Figure 2 shows a Laupala cricket in the wild. All crickets were brought back to Cornell University and were reared until maturity. Upon maturity, I recorded the songs of all the male crickets and analyzed their songs in Raven to calculate the pulse rate. Figure 3 shows the spectrogram of the two parental species. We also collected crickets from another field site called Makapipi, where one of the parental species of the putative hybrids occur. Figure 4 shows the distribution of pulse rates at different transects at Palikea Peak and at Makapipi.

The pulse rate variation frpm one site (Pig Wallow) was higher than any population of *Laupala* crickets. Based on what we know about the pulse rate variation and the genetics behind it, we hypothesize that hybridization is going on between L.makaio and L.orientalis. Why do we think so? Because we know from many interspecies crosses in the lab that F1 hybrids have intermediate phenotype and F2 intercross or backcrosses have a wide spread of their pulse rate phenotype [11]. Among all the other cricket species occurring around Palikea Peak, these two species are found in close geographical proximity. L.makaio occurs at the highest transect (Summit area) on Palikea Peak and L.orientalis has been historically found at a nearby site Kipahulu [12]. The pulse rates of both the species (Figure 3) also correspond with a possible hybridization scenario.

Analysis of pulse rate data shows we discovered some phenotypic hybrids between two species. DNA has been extracted from the collected crickets and are being sent for genotyping-by-sequencing (GBS) to study the population genomics of the hybrid zone and identify regions in the genome under selection using genome scans. I hypothesize that a lot of these genes will be related to song and CHC phenotypes. Discovery of the first-ever hybrid zone in this system opens the window to study speciation genetics and geography of speciation in greater detail.

Acknowledgements

I thank the Orthopterists' Society's Theodore J. Cohn Research Fund for funding the field trip that made this research possible. I thank my advisor Dr. Kerry Shaw and other members of the Shaw lab for assistance in fieldwork and animal care.



Figure 3. A spectrogram showing the difference in pulse rates of the two species: L.makaio and L.orientalis.

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Figure 4. Histograms showing the distribution of pulse rates of crickets collected at different transects in Palikea Peak (a-e) and Makapipi (f).

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Orthoptera Species File Grant Reports Vanishing coastal forests of East Africa

ver the past few years, our team has been engaged in surveying coastal forests to document the Ensifera and Caelifera fauna. Our efforts have been centered around the Tanzanian coastline, with a particular focus on repeated collection trips to various locations, including Kilulu Forest, Fish Eagle Point coastal shrub forest, and Kwale sacred forest, as well as nearby island forests as well as Gendagenda, Pugu Hills, and Kazimzumbwi forests (Fig. 1).

The comprehensive list of Kazimzumbwi forests was initially provided by Hemp in 2017, accompanied by the description of seven new Orthoptera species, featuring two previously unknown genera. Notably, the genus *Pseudotomias* was introduced by Hemp in 2016, originating from Kazimzumbwi and the East Usambara Mountains. This work shed light on the patterns of speciation between West and East Africa on a large scale. These findings underscore the remarkable diversity harbored within these coastal forests, emphasizing that numerous uniden-Figure 1. Map of screened coastal forests. Source of map: Google Earth.

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Figure 3. View along a dirt road through Zaraninge forest.



Figure 4. A wide aisle cut through pristine forest.

tified taxa across various arthropod groups are still awaiting discovery and formal description.

In July 2023, we conducted our inaugural expedition to Zaraninge forest, made possible by a grant from the Orthoptera Species File (OSF). The objective was to capture photographs of rare species within Tanzania and Kenya for inclusion in the OSF. Zaraninge forest is located within Saadani National Park, situated to the south of Pangani. While the stretch of coastline extending from the Kenyan border down to Dar es Salaam exhibits only a handful of small, partially disrupted forest patches, Zaraninge forest remarkably retains a substantial area of relatively untouched terrain. Portions of the forest boast tree canopies reaching heights of 20 to 25 meters, as depicted in Figures 2 and 3.

Regrettably, a significant setback occurred this year with the clearance of a wide corridor for a power line that traverses the most pristine

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and intact section of the forest. This unfortunate development underscores the prevailing priority of commercial interests over conservation considerations, as illustrated in Figure 4. Additionally, the nearby villages

are experiencing rapid growth, raising concerns about the potential escalation of illegal logging activities that could further degrade this last bastion of untouched coastal forest between the northern Kenyan border and Dar es Salaam in the south.

Results

The initial survey conducted on the Orthoptera fauna yielded the

identification of 19 Caelifera and 20 Ensifera species. Notably, all of these species are restricted to forest habitats, with some having been exclusively documented within the confines of the Kazimzumbwi forest reserve and the Pugu Hills forest until now (as outlined in Table 1). However, this



paradigm has shifted as the Zaraninge forest reserve has now also become a recorded habitat for *Chortoscirtes puguensis*, *Afrophisis kisarawe*, *Pseudotomias kisarawe*, *Euschmidtia viridifasciata*, and *Rhainopomma wapugu*.

During our exploration, we encountered a previously unknown species of *Gonatoxia* (distinct from all other recognized *Gonatoxia* species by its red abdomen - Fig. 5). Additionally, a new *Stenoschmidtia* species (Fig. 6) was captured within the bounds of Zaraninge. Detailed descriptions of these newfound species will be written at a later point, pending further surveys and the accumulation of additional pertinent information.

Discussion

In addition to the Kazimzumbwi forest reserve and the Pugu Hills forest, Zaraninge stands as the sole remaining forested area along the



Figure 5. Contrary to the common name "Yellow Surprise," given to the genus *Gonatoxia* by Hemp (2021) this is the first species with a red abdomen. A few specimens were found in pristine forest.

Table 1. List of Orthoptera species from Zaraninge Forest Reserve. 1. Species probably endemic to the area or with one knownlocality only 2. Coastal taxa with restricted occurrence 3. Widespread species.

Таха	Habitat	Status
Ensifera		
Conocephalinae/ Agraeciini		
Afroagraecia sp.	Understory vegetation	1
Conocephalinae/Conocephalini		
Chortoscirtes puguensis Hemp, 2010	Forest edge	2
Meconematinae		
Afrophisis kisarawe Hemp, 2013	Tree layer	2
Phaneropterinae		
Catoptropterxy aurita Huxley, 1970	Tree layer	3
Debrona cervina Walker, 1870	Tree layer	3
Diogena fausta (Burmeister, 1938)	Tree layer	3
Dioncomena ornata Brunner von Wattenwyl, 1878	Herb and bush layer on	3
	clearings and along forest edge	
Eulioptera monticola Ragge, 1980	Tree layer	2
Eurycorypha sp.	Tree layer	2
Gonatoxia sp.	Tree layer	1
Parapyrrhicia acutilobata Ragge, 1980	Tree layer	2
Phaneroptera sparsa Stål, 1857	Tree and bush layer	3
Plangia satiscaerulea Hemp, 2015	Tree layer	3
Poecilogramma striatifemur Karsch, 1887	Bush layer, forest edge	3
Pseudophyllinae		
Acauloplax exigua Karsch, 1891	Tree layer	3
Cymatomera denticollis Schaum, 1853	Tree layer	3
Pseudotomias kisarawe Hemp, 2016	Tree layer	2
Zabalius ophthalmicus (Walker, 1869)	Tree layer	3
Gryllacridinae		
Gryllacridinae sp.	Understory vegetation	?
Stictogryllacris vosseleri (Griffini, 1908)	Understory vegetation	3
Caelifera / Acridomorpha		
Eumastacoidea/ Euschmidtiidae		
Euschmidtia viridifasciata Descamps, 1973	Tree and bush layer	2
Plagiotriptus hippiscus (Gerstaecker, 1869)	Tree and bush layer	2
Stenoschmidtia taurus (Rehn & Rehn, 1945)	Tree and bush layer, forest egde	
Stenoschmidtia sp.	Tree and bush layer, forest egde	1
Thericleidae/Plagiotriptinae		
Cymatopsygma flabelliferum Karsch, 1896	Tree and bush layer, forest edge	2

Таха	Habitat	Status
Acridoidea		
Lentulidae		
Mecostibus minor (Bruner, 1910)	Tree and bush layer	2
Mecostibus leprosus Karsch, 1896	Tree and bush layer	2
Rhainopomma wapugu Jago, 1981	Herb layer	2
Pyrgomorphidae		
Phyteumas olivaceus (Karsch, 1896)	Tree and bush layer	2
Acrididae		
Acridinae		
Gymnobothrus temporalis flexuosus (Schulthess,	Understory vegetation,	3
1898)	clearings and forest edges	
Catantopinae		
Abisares viridipennis (Burmeister, 1838)	Tree and bush layer	3
Catantops momboensis Sjöstedt, 1931	Understory vegetation	3
Eupropacris cylindricollis (Schaum, 1853)	Understory vegetation, on herbs	3
	and bushes on clearings and	
	along forest edges	
Phaeocatantops decoratus (Gerstaecker, 1869)	Understory vegetation	3
Pseudophialosphera tectifera (Ramme, 1929)	Forest floor	2
Stenocrobylus cervinus Gerstaecker, 1869	Understory vegetation	3
Tangana asymmetrica Ramme, 1929	Forest floor	2
Eyprepocnemidinae		
Paraprocticus pendulus (Karsch, 1896)	Forest floor, along forest edge,	3
	in dense bushland	
Hemiacridinae		
Oraistes luridus Karsch, 1896	Bush laver	3

coastal stretch between the Kenyan border and Dar es Salaam that retains a substantial extent. In contrast, the other forests in this coastal corridor have been reduced to meager remnants of what was once a continuous forest belt, suffering from significant degradation. The Orthoptera composition, comprising Caelifera and Ensifera, within Zaraninge closely resembles that of the Kazimzumbwi and Pugu forests situated approximately 120 km further south. While the Kazimzumbwi and Pugu Hills forests have experienced substantial impact, with the latter now resembling more

of a bushland than a forest, Zaraninge's pristine forested patches persist due to its protective status within the Saadani National Park.

An intriguing discovery pertains to the presence of *Pseudophialosphera tectifera* (Fig. 7), initially described from forests around Tanga. In contrast, the Kazimzumbwi area to the south is inhabited by *P. severini*, described from Mkindani near Mtwara in southern Tanzania. Limited information is available about these two catantopine species that inhabit forested environments, primarily derived from their original descriptions based on a few specimens. Notably, both species have not been found cohabiting in the same habitat thus far. Zaraninge has proven to be abundant with *P. tectifera*, a species not hitherto observed in any other forested region in these numbers along the Tanzanian coastline.

As underscored by Hemp in 2016, Kazimzumbwi is under significant pressure from the surrounding human communities and the rapid expansion of cities, such as Dar es Salaam and Kisarawe. The depletion of precious timber species has been extensive, leading to various clearings of differ-



Figure 6. *Stenoschmidtia* sp. At the forest's edge, along the dirt road through Zaraninge, two *Stenoschmidtia* species were found, one of them *S. taurus*, widespread along the Tanzanian coast and on Zanzibar, and a new species, shown here.

ing sizes within the area. Coastal forests hold global significance, ranking among the top 25 regions worldwide for safeguarding endemic plant and animal species, as highlighted in studies by Burgess & Clarke (2000) and Olson & Dinerstein (1998). These coastal forests serve as a repository for comprehending both minor and large-scale mechanisms of speciation in Africa. However, there is a palpable concern that Tanzania's coastal forests are on the brink of disappearance, consequently erasing a treasure trove of rare species and numerous as-yet-

undiscovered organisms.

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Figure 7. Female of *Pseudophialosphera tectifera*. This species was plentiful on the forest ground.

Orthoptera from Kazimzumbwi Forest Reserve, Tanzania with the description of new species and discussion of the biogeographic patterns of threatened species. Zootaxa 4226 (2): 151–193. https://doi. org/10.11646/zootaxa.4226.2.1

Recap of the Cricket Course 2023

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mong all animals, insects show unparalleled diversity in hearing and sound production. Of all insects, the orthopteran suborder Ensifera includes more

than 15,000 described species of familiar singing insects, such as crickets and katydids, as well as interesting non-singing insects, such as cave crickets and wetas, and represents the most species-rich lineage using acoustic signals as a main mode of communication. Nevertheless, there has not been a platform in North America to provide structured hands-on training in these amazing insects' identification, ecology, behavior, and bioacoustics. Therefore, as part of the National Science Foundation grant titled "NSFDEB-NERC: Multidisciplinary approach to bioacoustics: Integrating phylogenomics, biophysics, and



Group photo of the 2023 Cricket Course participants.





Students learning to identify Ensifera.

functional genomics to unravel the evolution of hearing and singing in katvdids, crickets, and allies" (DEB-**1937815**), we assembled a team of currently active specialists to create and offer a unique workshop called "THE CRICKET COURSE." We offered this 5-day workshop for the first time from July 3-7, 2023, at the beautiful Archbold Biological Station (ABS) near Lake Placid, Florida, one of the most renowned biological stations in the world. ABS is home to numerous orthopterans, which makes it a perfect place to learn about crickets and katydids.

The instructors for the 2023 CRICKET COURSE were Dr. Hojun Song and Brandon Woo from Texas A&M University, who specialize in systematics, Dr. Fernando Montealegre-Z and Charlie Woodrow from the University of Lincoln (U.K.), who specialize in bioacoustics, and biophysics, and Dr. Nathan Bailey from the University of St. Andrews (U.K.) who specializes in behavioral genetics and mating biology. We had 16 participants, mostly from the U.S., but also from Taiwan and New Zealand. The participants included mostly graduate students, but also an undergraduate student, a postdoc, amateurs, and other professionals.

The first day (July 3) was dedicated to learning about the taxonomy, phylogeny, and identification of Ensifera. The course started with a brief introduction, followed by Hojun's lecture on the taxonomy and phylogeny of Ensifera and a primer on ensiferan morphology. These two lectures served as a crash course on the diversity of Ensifera of the world. Following these, students were given the key to subfamilies of Ensifera of the world, compiled by Hojun, and learned to identify various museum specimens using this key. Via assistance from the instructors, students recognized important diagnostic traits and worked their way through the key by spending time on their microscopes. In the afternoon, Brandon gave a lecture on the diversity of North American Ensifera. Brandon is an orthopterist extraordinaire and an excellent macro photographer, and his lecture featured many of his amazing photos. Following this lecture, students went back to identify more ensiferans, but this time using the key to genera of the North American and Florida Ensifera, compiled by Brandon. After dinner, we all headed to the Red Hill area to collect katydids and crickets. At night, ABS comes alive with numerous species of Ensifera singing loudly. Because we wanted to teach how to process the catches, the specimens were collected alive and kept in small cages. Fernando was very excited to find Phlugis chrysopa, a species in the Caribbean that somehow invaded Florida. This particular species sings at an ultrasound fre-

Learning how to analyze field recordings using the software Audacity.

quency and appears to be adapting to this new environment.

The second day (July 4) was dedicated to learning about bioacoustics, online resources, and sample processing. We started with a lecture from Fernando on sound production in Ensifera. Because Fernando is the leading authority on the biomechanics of cricket and katydid sound production, his lecture featured some of his own latest findings, which were very interesting. Using mathematical modeling, Fernando was able to reconstruct the songs from fossil specimens, which was quite amazing. Following this lecture on sound production, Charlie gave a lecture on hearing in Ensifera. Fernando and Charlie brought ultrasonic microphones and an oscilloscope, as well as 3D-printed models of katydid ears, which allowed us to visualize how different katydid species hear and how the shape of the ear covering (pinnae) affects hearing range of the katydids. Students also learned how to use the software Audacity to analyze the recorded songs. In the afternoon, Brandon lectured using online resources, such as the Orthoptera Species File (OSF), BugGuide, iNaturalist, and Singing Insects of North America (SINA). Because Brandon is a major contributor to some of these online resources, he gave an excellent overview of how these resources can be used for research. Following Brandon's lecture,



Learning how to use sound analysis equipment.

Hojun showed how to properly kill insects using kill jars and various preservation methods. At night, we went out with hand-held tablets connected to ultrasonic microphones (Echo Meter 2) and learned to field-record cricket and katydid songs.

The third day (July 5) was dedicated to learning about the ecology and natural history of Ensifera, how to set up behavioral experiments using Ensifera, sound analyses based on the field recordings from the previous night, and a field trip to Highlands Hammock State Park. In the morning, Nathan gave a lecture on the ecology of Ensifera. Nathan shared some of his own research on the evolutionary dynamics between the cricket Teleogryllus oceanicus and the parasitoid fly Ormia ochracea, and how these cricket males lost the ability to produce sound. Following his lecture, we brainstormed some ideas on conducting a behavioral experiment using some of the ensiferan insects found at ABS. We settled on the Florida true katydid Lea floridensis, which is abundantly present and calls loudly at night. We came up with an idea to experimentally test the effect of disturbance on the katydid's singing activity. In the afternoon, we all traveled to the Highlands Hammock State Park, well-known for cypress swamps and

an old-growth hammock. We chose this place to contrast with the scrub habitats at ABS. Unfortunately, the park did not have a lot of ensiferans, but the students enjoyed the boardwalks and very different habitats.

The fourth day (July 6) started with Charlie's lecture on interdisciplinary research using Ensifera. After a short lecture, the students were divided into groups to brainstorm ideas to develop bio-inspired products. Students came up with novel and exciting ideas, such as a katydid-inspired night vision, a mole cricket-inspired underground robot, ensiferan antennae-inspired sensory gloves, etc. It was a fun exercise to think outside the box and think about some real-life applications inspired by ensiferan insects. Following this lecture, Po-Wei Chen, a participant from Taiwan, gave a presentation regarding Orthoptera research in Taiwan. He talked about the history of orthopteran research in Taiwan, and shared his experience studying Orthoptera. It was very interesting to learn about a very different ensiferan fauna. After this lecture, Hojun and other instructors described various methods for rearing crickets and katydids for research. In the afternoon, we had a guest Zoom lecture by Dr. David Weissman, who has studied the taxonomy of crickets and Jerusalem crickets for decades. Dr. Weissman gave a brief history of how he became interested in these insects and shared his research and how he was able to collaborate with diverse researchers to achieve his goals. In the afternoon, students continued to work through their identifications and analyzed the field-recorded songs. We also visited the entomology lab, where Brandon explained the research that has been conducted by Dr. Mark Deyrup. After dinner, some folks went to Lake Annie and plunged into the water to cool off from the heatwave. At night, the students were divided into several groups and conducted behavioral experiments to study the effect of light disturbance on the calling behavior of Lea floridensis. For this experiment, 3-4 students formed a group, located a calling male, and tested whether shining a bright light (white or red) would affect its calling behavior. The students collected data, which were analyzed the following day.

The fifth day (July 6) was the last full day of THE CRICKET COURSE. We started with a group photo and went on a morning hike to find unique scrub habitats containing scrub rosemary (Ceratiola ericoides). We saw Florida scrub jays, as well as the Florida rosemary grasshopper, Schistocerca ceratiola. It was very hot and humid, but being out in the scrub during the morning was amazing. In the afternoon, we had another Zoom guest lecture by Dr. Ryan Ardoin, a food sensory scientist at USDA, who talked about developing cricket-based foods for the general public. It was a very informative talk, and we learned a lot about how people's perceptions of insects would affect the consumer market. After this invited lecture, we analyzed the previous night's behavioral experiment data, which collectively suggested that the light disturbance had a minimal effect on disrupting the calling behavior of the species. Afterward, we cleaned and packed our gear, and the students provided course evaluations. The first CRICKET COURSE was a transformative experience for many participants. In the evening, we had a nice social and dinner, followed by a salsa dancing lesson from Fernando. The course ended on a positive note and everyone was very satisfied.

Overall, the inaugural CRICKET COURSE was a resounding success and the students became much more knowledgeable about Ensifera. ABS was a fantastic place to host this course, with unlimited access to diverse natural areas where many crickets and katydids were abundantly present. We plan to offer THE CRICKET COURSE every other year, and hopefully, we will return to ABS sometime soon.

Editorial

By HOJUN SONG Editor, *Metaleptea* hsong@tamu.edu

any of us will gather in Merida for the 14th International Congress of Orthopterology in a few weeks. The ICO is my favorite scientific meeting, and I look forward to meeting old friends and new colleagues. Merida is near and dear to my heart because it is close to the permanent gregarious zone of the Central American locust (Schistocerca piceifrons), which I've visited several times. My dear friend Mario Poot-Pech has been tirelessly working to ensure this ICO runs successfully and I have no doubt that it will be a wonderful event. I am also so looking forward to the Yucatecan food, which is simply amazing.

This will be my 7th ICO. I remember my very first ICO meeting in Montpellier, France, in 2001. I was a first-year graduate student and presented a poster on the phylogenetic relationships of North American Schistocerca. I remember meeting Fernando Montealegre-Z, Axel Hochkirch, and Piotr Naskrecki at that meeting, who were all graduate students at that time. I remember being extremely nervous because it was the first time I met the giants in the field who I only knew from the papers. I was particularly nervous in front of the late Christiane Amedegnato, who had written extensively about Schistocerca. She turned out to be very kind and, later, she even allowed me access to the grasshopper collection at the Paris Museum when I visited her in 2009. I also remember how generous the late Ted Cohn was at that meeting. He knew I was a beginning graduate student and introduced me to many orthopterists I still interact with today. I also hung out with Greg Sword and Spence Behmer at that meeting, both of whom were in the early career stage. Now, the three of us are in the same department and working

together on the evolution of locusts. For me, ICO meetings have opened up many opportunities, enlarged my scientific network, and introduced me to many colleagues who have become good friends. So, I am excited about what new opportunities this upcoming meeting will bring!

This issue includes a number of Cohn Grant reports. It is very encouraging to see that we have a large community of young orthopterists who are doing excellent research. For many years, I had seriously worried about the decline of orthopterists around the world, but I do not worry anymore. The future of our field is bright, and this issue of *Metaleptea* is a good demonstration. I would like to thank our Associate Editor, Derek A. Woller, for his continued assistance in the editorial process. To publish in *Metaleptea*, please send your contribution to hsong@ tamu.edu with a subject line starting with [**Metaleptea**]. A MS Word document is preferred and images should be in JPEG or TIFF format with a resolution of at least 144 DPI. The next issue of *Metaleptea* will be published in January of 2024, so please send me content promptly. I look forward to hearing from you soon!

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