

10th International Congress of Dipterology

Reno, Nevada, USA
16-21 July 2023



55
**ABSTRACTS
VOLUME**

**10TH INTERNATIONAL
CONGRESS OF DIPTEROLOGY**

**RENO, USA
16-21
JULY, 2023**

10th International Congress of Dipterology

16–21 July 2023
Reno, Nevada, USA

Abstract Volume

Edited by
Stephen D. Gaimari



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Fly Times Supplement 5

Published by the North American Dipterists Society, © 2023

Printed by TheBookPatch, LLC, a service of Wilshire Press, Inc., Scottsdale
Freely available as PDF from https://dipterists.org/assets/PDF/flytimes_supplement05.pdf
Hard copy available for purchase at <https://app.thebookpatch.com/BookStore.aspx>

ISSN 2769-612X (online), 2769-6111 (print)

Suggested citation:

Dankowicz, Z.R. 2023. Methods of focus stacking for live insect photography. Page 39 in: Gaimari, S.D. (ed.), *Abstracts Volume, 10th International Congress of Dipterology, 16–21 July 2023, Reno, Nevada, USA*. Fly Times Supplement 5, 238 pp.

Front cover image: From the series “Entomologische Sammlung der Labors Diptera” by German artist Natalie Port. 55. an unknown fly from the Museum Koenig in Bonn (=Tachinidae, *Phasia subcoleoprata* (Linnaeus, 1767)).

Back cover image: From the series “Entomologische Sammlung der Labors Diptera” by German artist Natalie Port. 48. Syrphidae, *Palpada vinetorum* (Fabricius, 1799), painted from a specimen from the Museum Koenig in Bonn, collected in 2008 from Rancho Las Lajas, Sonora, Mexico.

Disclaimer: Following recommendations of the various nomenclatural codes, this volume is not issued for the purposes of the public and permanent scientific record, or for the purposes of taxonomic nomenclature, and as such, is not published in the meaning of the various codes. Thus, any nomenclatural acts contained herein (e.g., new combinations, new names, etc.) do not enter biological nomenclature or preempt publication in another work.

Preface

The International Congresses of Dipterology (ICD) have been consistently held every four years since the First one held in Budapest, Hungary in 1986. This Tenth congress has broken that mold, being postponed for a year due to the global Covid-19 pandemic, and the uncertainty that we would see dipterists wanting to travel. Although this extra year has seen a great improvement in this regard worldwide, certain factors have led to this being the smallest (I say coziest) ICD yet. Our delegation consists of 200 dipterists (yes, exactly 200, along with 30+ accompanying persons), 48 of which are students, from 35 countries from all continents except Antarctica (unfortunately no one came who could collect *Belgica antarctica* in their backyard). Their names, countries, and institutional affiliations (if any) are given in the Scientific Program.

This volume consists of 222 abstracts. The 190 oral presentations are arranged into 20 symposia, a General session, Plenary (5) and Banquet (1) addresses, and the Book launch event (3) for Volume 3 of the *Manual of Afrotropical Dipterology*. The Poster session consists of 32 posters. In total, students are giving 39 of the oral presentations and 10 of the posters.

This Abstract Volume is organized alphabetically by first author, and the presenting author's name is underlined except for cases of single authorship. The header on each abstract page gives its symposium or session. The Table of Contents is organized according to symposium or session in alphabetical order, and alphabetically by first author within each. An author index is given at the back, as well as a subject and taxonomic index.

We gratefully thank our numerous sponsors, without whom we could not have run such an excellent program. They are detailed on the back cover of this volume, with further acknowledgment in the Scientific Program.

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10th International Congress of Dipterology

16–21 July 2023
Reno, Nevada, USA

Abstracts

A molecular phylogeny of assassin flies (Diptera: Asilidae) with ancestral state reconstruction of arthropod prey preference

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Keywords: diet, phylogeny, predator, prey, sanger-sequencing

Assassin flies (Diptera: Asilidae) are a diverse family that plays an essential ecological role as top aerial and venomous predators. Little is known about the evolution of their predatory habits. This study provides a novel phylogenetic hypothesis of Asilidae along with prey preference and ancestral state reconstruction in a maximum likelihood framework. This study is based on 176 assassin fly species, 35 Asiloidea outgroup species, prey preference data accumulated from literature and museum collections, and approximately 7,913 bp of nuclear DNA from five genes (*18S* and *28S* rDNA, *AATS*, *CAD*, and *EF-1 α* protein-encoding DNA) and mitochondrial DNA from one gene (*COI*). Of the 12 asilid subfamilies included in the analysis the monophyly of six was supported. We used ancestral state reconstruction and stochastic character mapping to test whether a polyphagous arthropod predator is the ancestral state for Asilidae. Assassin flies are polyphagous arthropod predators, with specialized arthropod prey preferences evolving 20 independent times across the Asilidae phylogeny.

New species of Neotropical frog-biting midges (Corethrellidae)

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Keywords: biodiversity, Colombia, Cuba, Neotropical, new species, taxonomy

Frog-biting midges (family Corethrellidae) are a potentially species-rich group of dipterans that have most of its diversity still unknown. Despite the increase in the number of studies on this group over the past twenty years, the publications rate of new species, distributional records, and new taxonomic data does not show signs of plateauing. In this study we describe six new species of Corethrellidae, four of those from Cuba and two from Colombia. This is only the second report of frog-biting midges from Cuba, a highly biodiverse region with notable cases of endemism for insects. Even though Colombia has a larger number of frog-biting midge species recorded, the new species reported here come from small samples from two localities, suggesting high hidden diversity. With more extensive sampling from tropical and subtropical regions worldwide, we expect to find significantly more new species of frog-biting midges.

The conundrum of the biting “non-biting midges”

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Keywords: Chironomidae, evolution, hematophagy, fossil, morphology, mouthparts

Although usually described as aphagous with non-functional mouthparts, some adult representatives of the family Chironomidae, commonly known as “non-biting midges”, seem to defy such conceptions. Many adult chironomids are known to feed on nectar and honeydew, with some notable mouth apparatus adaptations, such as the very elongate labella in *Rhinocladius* Edwards and *Pseudorthocladius macrostomus* Sopenis. Besides that, there are a few known extant species with toothed mandibles in the genera *Archaeochlus* Brundin and *Austrochlus* Cranston, found in southern Africa and in Australia. Also, the fossil record reveals a much more complex and diverse evolution of adult feeding, with various adaptations, including the presence of short and long probosces, armored labra, toothed mandibles in both male and females, as well as toothed laciniae. Despite speculations by many authors, the feeding habits of species with well-developed mouthparts, even the extant ones, remain largely unknown. In this study we aim to infer potential feeding habits of chironomids by generating detailed morphological description of their mouthparts and comparisons to biting culicomorphans. For that, we perform and analyze light microscopy images of both fossil and extant species, as well as micro-CT scans of several fossil species of Chironomidae. Our results suggest multiple shifts of feeding habits during the evolution of the family, with entomophagous and blood-feeding species emerging among non-biting ones. We also report strong evidence of red-blood cells, likely from mammalian hosts, in the abdominal cavity, mouthparts and the vicinity of the body of biting “non-biting midges”.

Unusual as usual: conspicuous larvae from Cretaceous amber

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Keywords: Athericidae, Anisopodidae, Cretaceous, fossil, larvae, morphology, Tipulidae, Trichoceridae

The Cretaceous is marked by the presence of numerous fossils with distinct morphologies, so much it has been deemed as an “experimental phase” of insect evolution. One reason for that is a deep transformation of terrestrial ecosystems, largely related to the diversification of plants, notably the flowering plants. Not coincidentally, it was also in the Cretaceous that many large groups of insects arose and radiated. Therefore, it is not uncommon to find fossil specimens with conflicting traits, diagnostic of different extant taxa, or with morphologies not found in the modern fauna. In this work, based on images obtained via light microscopy and micro-CT scanning, we report four dipteran larvae from Kachin amber (Myanmar), with their identity still uncertain. Two of them resemble modern larvae of Athericidae, although their slender body, very elongate head capsules and shape of parapods differ in details from the extant forms. One of the larvae has anal papillae and a prominent furrow indicating a subdivision of the prothorax, the first character known in larvae of Trichoceridae the second in larvae of Anisopodidae. Finally, we report a specimen bearing a combination of characters only found in larvae of Tipulidae: a retractable head with longitudinal excisions, horizontally moving mandibles, and uncrocheted parapods. These specimens possibly represent lineages closely related to extant groups (“stem-lineage”) and their peculiar morphology may help us understand the evolution of modern morphologies.

Challenges and opportunities in Oriental acalyptrate taxonomy, with perspectives from a tiny island in Southeast Asia

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Keywords: cybertaxonomy, dark taxa, DNA barcoding, integrative taxonomy, Oriental, Singapore

Southeast Asia is a biodiversity hotspot with remarkable diversity and endemism. However, our understanding of acalyptrate flies in the region remains limited, with only a few researchers driven largely by personal interest working on this group. In this oral presentation, we delve into the history of acalyptrate taxonomy in Southeast Asia, highlighting the key challenges hindering its progress. One major obstacle is poor taxonomic understanding of the largely endemic fauna, making genus identification difficult. Much existing literature consists of poorly written and insufficiently diagnostic descriptions from the 1800s, often coupled with physically inaccessible or uninformative type specimens. Additionally, rampant inadequate taxonomic practices (e.g., erecting monotypic genera without comprehensive revisions or keys) have resulted in taxonomic confusion. These factors make it challenging for newcomers to enter the field. Moreover, the presence of numerous "dark taxa" within Acalyptratae further complicates conventional taxonomic approaches. To address these challenges, we present our collaborative efforts in studying acalyptrate diversity in Singapore, showcasing examples in Lauxaniidae, Chloropidae and Canacidae. We demonstrate how integrating affordable large-scale approaches in acquiring molecular (NGS-based DNA barcoding) and morphological (high-resolution specimen imaging) data can effectively address the scale and resolution required for delimiting species without expert-level taxonomic knowledge. We also test the optimal level of detail for taxonomic descriptions and discuss the use of accessible online tools, (e.g., Notion) for managing specimen and species information. Additionally, we highlight the collaborative potential of the 'biodiversity.online' platform in facilitating taxonomic research. Finally, we show how we aim to build capacity for regional dipterists via training programmes and online knowledge sharing, where the ultimate aim is to make acalyptrate taxonomy more accessible to newcomers.

Fly puke and poop: a simplified iDNA metabarcoding protocol for detecting vertebrate species using calyptrate flies

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Keywords: biomonitoring, iDNA, fly feces, nanopore sequencing, natural history

Invertebrate-derived DNA (iDNA) metabarcoding has proven effective for surveying vertebrate communities, but little is known about the interactions between invertebrate and vertebrate species. In this oral presentation, we investigate specialization in the calyptrate fly community in a swamp forest remnant in Singapore, along a disturbance gradient (10 sites: 80–310 m from a road). 407 flies were collected and individually sampled from baited traps. Here we employed a greatly-simplified iDNA protocol that eliminates DNA extraction by directly dissolving fly feces and regurgitates in water and then subject to nanopore sequencing, which yielded 294 identifications to 20 vertebrate species based on two COI fragments and 16S. We found that the calyptrate fly community sourced for iDNA was unexpectedly diverse, comprising 24 species from three families (Calliphoridae, Sarcophagidae and Muscidae), and carried DNA from both rare and common vertebrate species inhabiting different forest niches (ground-dwelling, arboreal, and flying). While we found no statistically significant specialization in the interactions between fly and vertebrate species, we discovered that uncommon fly species can carry the iDNA signal for vertebrate species that are challenging to detect using traditional methods. Our study highlights the need for further research using different bait types, developing traps that can automate/facilitate individual specimen sampling, and exploring experimental designs that can investigate the dispersal capabilities of flies carrying iDNA.

Integrating molecular and morphological approaches for taxonomic revisions of dark taxa: a case study on fungus gnats (Mycetophilidae) in Singapore

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Keywords: dark taxa, DNA barcoding, integrative taxonomy, next generation sequencing, Oriental, reverse taxonomic workflow, Singapore

Dark taxa, characterized by high species richness and abundance, pose significant challenges for traditional taxonomic efforts optimized for lower-diversity taxa with low abundance. To address this problem of scale, we propose compromise through comprehensive sampling at smaller geographical scales, advocating a 'reverse taxonomic workflow' combining fast, cost-efficient large-scale NGS-based DNA barcoding, followed by targeted morphology assessment for robust species delimitation. This allows integrating 1) information from traditionally excluded, morphologically 'uninformative' (e.g., damaged/female) specimens; 2) data-sources with independent evolutionary processes (e.g., sexual selection for morphology, genetic drift for DNA barcodes), providing perspectives that reciprocally illuminate speciation markers not captured by either data-source alone; 3) standardized subsequent faunistic surveys using DNA barcodes to allow a feasible, stepwise approach for comprehensively understanding biodiversity of a region. As a case study, we revised Mycetophilidae in Singapore, analyzing 1,347 sequenced specimens from diverse environments collected over eight years. Using these methods, we identified 120 species, 115 being new, increasing the described Oriental mycetophilid fauna by >25%. This exemplifies the taxonomic impediment for such taxa and the efficacy of our approach in enhancing species discovery. We emphasize several other critical aspects for successful large-scale species discovery: high-resolution specimen imaging, robust specimen data management, the importance of maintaining the connection between illustrations and specimens, and the continued relevance of completist approaches at higher taxonomic levels, aiding in generating identification keys and understanding morphology.

Succession and richness of acalyprate muscoid families with respect to vertebrate decomposition

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Keywords: decomposition, forensic entomology, Malaise trap, richness, succession

Acalyprate muscoid flies frequently participate in insect scavenging communities, though their role remains, with some exception, largely understudied due to their minute size. To better understand the role acalyprates play with respect to vertebrate decomposition, we set out a linear array of six malaise traps, three baited with neonatal pig carcasses alternating with three non-baited control traps in the Putah Creek Reserve near the University of California, Davis campus. By this we sought to (1) distinguish the acalyprate scavenger community from non-scavenging background, (2) determine the presence/absence and (3) the relative richness of acalyprate taxa at the family level with respect to time throughout the decomposition process. Sphaeroceridae, Lonchaeidae, Lauxaniidae, Heleomyzidae, Ephydriidae, Drosophilidae, Chloropidae, and Carnidae appeared more frequently in carrion baited traps in the early part of the study. Piophilidae appeared more frequently associated with the carrion baited traps later in the study. Additionally, the average number of unique taxa present in the carrion baited traps was significantly higher throughout the entire time frame of the study. Furthermore, the average number of taxa observed over time exhibited a slight increase in both baited and unbaited traps. These results indicate that the presence of vertebrate carrion attracts a majority of the acalyprate families observed in the environment, and qualitatively, that the presence/absence of families varies with respect to time, which could indicate a successional pattern of appearance at carrion and perhaps colonization.

Multilevel investigation of Diptera taxonomy: exploring coastal sand dunes in Italy

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Keywords: biodiversity, Brachycera, coastal sand dunes, community ecology, dark taxa, integrative taxonomy

Coastal sand dunes¹ serve as a crucial habitat for numerous insect species. However, challenging environmental conditions and human impact have a direct influence on the landscape of this extreme habitat. The impact on insect communities in Italy remains largely unexplored. This is the first extensive survey of sand coastal flies (Diptera Brachycera) in Italy. Three sampling rounds were conducted across ten study sites in 2022 to investigate the effects of land use (specifically urbanization), landscape structure, and latitude on brachyceran diversity and community composition. The study addresses the following questions: (I) How many brachyceran species inhabit Italian coastal sand dunes? (II) Does the taxonomic and functional diversity of brachyceran flies decrease or become disrupted with escalating impact of human activities? (III) To what extent do species vary between habitats and study sites? At each site, four distinct habitats were identified: shifting dunes, fixed dunes, retrodunal wetland, and retrodunal forest. Three sampling methods were employed, namely yellow pan-traps, net capturing, and sand sieving. Comprehensive data on plant communities, soil characteristics (temperature, humidity, grain size), climatic conditions (temperature, humidity), and land use were collected at each sampling site. DNA barcoding using Next-Generation Sequencing (NGS) techniques allowed sorting 11,000 specimens to putative species and identifying part of those via BLAST, covering various dark taxa like Chloropidae, Phoridae, Agromyzidae, as well as other acalyprate and calyprate groups. The Large-scale Integrative Taxonomy approach will be applied in the second phase to some groups, integrating morphological examination by expert taxonomists and validate preliminary species hypothesis. This study sheds light on community composition of several overlooked groups within highly endangered habitats like coastal sand dunes, also contributing to better understanding of the local fauna and relationship with the environment.

Amber dancers: phylogenetic affinities and ethology of enigmatic Mesozoic dance flies (Diptera Empidoidea)

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Keywords: Cretaceous, Dolichopodidae, fossils, palaeobiology, phylogeny

The fossil record of Empidoidea is extensive and dates back to the Jurassic period, though fragmentary and sometimes ambiguous. Recently, seven new fossil species of empidoid flies were discovered in late Cretaceous ambers from Kachin, Myanmar, which were assigned to a new genus, *Electrochoreutes*. *Electrochoreutes* exhibits a distinctive combination of ancestral and derived morphological features that challenge the phylogeny of Empidoidea. The use of high-resolution X-ray phase-contrast microtomography has been instrumental in studying the fossils' key morphological features, allowing to reconstruct their phylogenetic relationships within the empidoid clade. Phylogenetic analysis, which included representatives from all Empidoidea clades and extinct Mesozoic genera, recovered *Electrochoreutes* as a stem-group of Dolichopodidae. *Electrochoreutes* species are sexually dimorphic, with males being characterized by species-specific sexual traits on their forelegs, suggesting a crucial role in mating. These traits are reminiscent of those found in modern Dolichopodidae, whose males display exaggerated secondary sexual traits during elaborate pre-mating rituals. The diversity in the shape and structure of these features in *Electrochoreutes* species suggests that complex mating rituals evolved in this phylogenetic lineage during the Cretaceous epoch, shaping the evolution of the group since its emergence.

Using anchored hybrid enrichment to resolve the higher-level phylogeny of anthomyiid flies (Muscoidea: Anthomyiidae)

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Keywords: anchored phylogenetics, hybrid enrichment, phytophagy, systematics, trophic specialization

Anthomyiidae is a large family (~2,000 species) of ecologically diverse flies, exhibiting phytophagous, parasitic, and parasitoid lifestyles on a wide range of hosts. The majority of larvae in Anthomyiidae are plant-feeding, including several species of agricultural importance. Despite multiple attempts with both morphological and molecular characters, classification and relationships within Anthomyiidae have long been disputed. Monophyly has been identified among Neotropical taxa, but major relationships in the family have not yet reached consensus. Large-scale sampling efforts and increased sampling depth are necessary for successful resolution of systematics within anthomyiid flies. Here, we used anchored hybrid enrichment (AHE) and generated a phylogenomic dataset of 847 loci on 62 taxa to elucidate relationships among a broadly diverse sampling of anthomyiid genera. Based on our newly-supported phylogenetic hypothesis for the family, we also examine evidence for multiple origins of diverse modes of trophic specialization, new estimates of divergence times within the Anthomyiidae, and morphological support for major clades.

From taxonomy to conservation biology: lights and shadows of the citizen science applied to native flies in Chile

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Keywords: Linnean shortfall, Wallacean shortfall, Darwinian shortfall, Eltonian shortfall, Hutchinsonian shortfall, Prestonian shortfall, Raukiaeran shortfall

Flies are one of the most important and diverse insect orders on the planet, providing many key ecosystem services like pollination, recycling, and pest control. However, the knowledge of many families and fly species is still far to be sufficient, showing great shortfalls, mainly in South America. On the other hand, citizen science (CC) is recognized as an alternative to fill these knowledge gaps. The goal of this work is to determine whether CC can help to fill different scientific shortfalls only based on photographic records provided by volunteers. I worked on seven shortfalls during 8 years: Wallacean (gaps in geographic distribution), Linnean (gaps in species description), Darwinian (gaps in evolution), Eltonian (gaps in biological interactions), Hutchinsonian (gaps in abiotic tolerances), Prestonian and Raukiaeran (gaps in population dynamics and ecological function respectively). At least, 104 species of flies (some exotic) reported exhibited changes in known geographic distributions or are reported for the first time in Chile. Geographic distribution is responsible for the fact that 11 species of flies nationwide and 33% of the hover flies in Chile have a conservation category. The CC has detected new species present in the country like Stratiomyidae and volunteers have collected some rare species help to study the phylogeny of families such as Pelecorhynchidae. The CC is the main source of new biological interaction detected in Acroceridae and Asilidae. Some functional characteristics can be projected in cities to demonstrate the usefulness of CC in Anthropic ecosystems. However, CC has failed to fill major problems in population sizes or their dynamics, as well as abiotic tolerances. Challenges in the study of flies with CC are addressed toward bettering the IDs of flies with standardized protocols, strengthening public marketing and the flies' benefits as well as a high social media presence to maintain the number of records.

Over fifty years after the *Catalogue of the Diptera of the Americas South of the United States*: an update on the knowledge of Chile Diptera diversity

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Keywords: Acalypratae, Brachycera, Calypratae, citizen science, lower Diptera, Nematocera, Orthorrhapha, Schizophora

Chilean flies are key pieces to many evolutionary and phylogenetic aspects of Diptera because of their direct Gondwanan link. Through time, many dipterists considered the order among the most diverse in Chile, however, there are still many gaps to fill. Our goal is to update the number of genera and species known for each fly family in Chile and to understand the origin of all dipteran groups, including to which biogeographical layer each family belongs. We performed a thorough review of taxonomic literature since 1967 when the "Catalogue of the Diptera of the Americas South of the United States" was published. We found a total of 3,702 currently valid species and 922 genera grouped in 98 families, making Diptera one of the most species-rich taxa in Chile. Lower flies increase to 221 genera (89.83%) and 681 species (93.25%), whereas Brachycera increases to 239 genera (385.71%) and 739 species (48.26%). We present hypotheses of age of clades in Chile that belong to 60 families, of which 16 correspond to elements of Cretaceous layers of the fauna and 46 correspond to elements of Cenozoic layers, with a small number of truly Jurassic elements. Neriidae is cited for the first time in Chile. We discuss the following five major gaps: (1) lack of long-term systematic sampling and taxonomic, spatial, and temporal biases; (2) a low number of taxonomists residing in the country; (3) the presence of primary types in Chile; (4) difficulty to access undigitized primary literature and (5) poor understanding of biology and ecological processes related to Diptera in general and the implications of increasing anthropogenic impacts across the country. We propose citizen science and socio-political actions as part of the solutions to assess these gaps.

Morphological and molecular species delimitation of Mariobezziinae (Diptera: Bombyliidae) in South Africa

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Keywords: species delimitation, systematics, taxonomy

The genera *Corsomyza* (Wiedeman 1820) and *Callynthrophora* (Schiner 1867) (Diptera: Bombyliidae) are prominent pollinators of annual blooming Asteraceae and Aizoaceae in the Greater Cape Floristic Region (GCFR). *Callynthrophora* is a South African endemic genus found exclusively in the Western and Northern Cape. *Corsomyza* has a wider distribution from southern Namibia to the Eastern Cape, South Africa with a few historical records of this genus in Zimbabwe. Currently there are 27 described species of *Corsomyza* and three described species of *Callynthrophora* but the taxonomy of these two genera has not been investigated at a molecular level. This study aimed to assess the species boundaries of these two genera, using DNA barcodes and species delimitation models. The results of the molecular species delimitation of a large, spatially dense sample from the GCFR suggest there are three species of *Callynthrophora* and 13 species of *Corsomyza*. However, results of principle coordinate analyses of morphological characteristics show that there are likely four species of *Callynthrophora* and 16 species of *Corsomyza*. Determining the species boundaries of these genera will be beneficial in the assessment of the taxonomy of these genera and will have larger implications in revising the taxonomy of either genus.

On *Librella* (Cryptochetidae) and the limits of Drosophilidae

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Keywords: biodiversity, genomics, Ephydroidea, male terminalia, parasitoid, phylogeny

Phylogenomic advances in Ephydroidea demonstrate that the group exhibits remarkable ecological and morphological diversity. Modified, parasitic, and taxonomically challenging Mormotomyiidae, Cryptochetidae, and Braulidae are in fact ephydroid flies. This has major implications for the evolution of parasitoidism and for processes underlying the extraordinary diversification of Drosophilidae, including genetic model organisms and pests of health and agriculture. We aim to clarify the phylogenetic relationships of *Cryptochetum* scale parasitoids, and *Braula* bee lice by improving taxon sampling and alignment scope. Further, two additional controversial lineages are included in a phylogenomic study for the first time – *Librella* (Cryptochetidae) and *Diathoneura* (Cladochaetini). To construct this dataset, we processed dozens of published drosophilid genomes and sequenced critical taxa via de novo transcriptomes and genomes. Analyses of 160 taxa and 3000 markers yielded compelling hypotheses, clarifying the position of *Librella* and *Braula*. Key findings include the placement of *Cryptochetum* as sister group to Drosophilidae, the identification of *Librella* within Steganinae close to *Rhinoleucophenga* (Gitonini), the proximity of *Braula* to the *Stegana* group, and the placement of *Diathoneura* within Drosophilinae, consistent with traditional taxonomy. The placement of *Librella* is supported by examination of the first reported male, which has genitalic characters clearly linking it to Steganinae. Cryptochetidae and Drosophilidae (including Braulidae) are not monophyletic with respect to one another. Our study underscores the importance of comprehensive sampling in hyperdiverse clades, as taxa with newly clarified relationships significantly impact the reconstruction of life history evolution in Ephydroidea. This project provides the impetus and foundation to conduct a comprehensive revision of the Australian cryptochetid fauna. Cryptochetidae have never been revised, and Australian species of uncertain identities were deployed in biocontrol projects. Funding from the Australian Biological Resources Study will support the description of numerous new species in concert with museomics illuminating parasitoid host records.

The phylogenomic context of Tephritidae and Tephritoidea

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Keywords: Acalypratae, comparative methods, female terminalia, insect-plant interactions, phylogenomics, Neriodea, systematics

Tephritidae includes damaging pest species in which phytophagous larvae hatch from eggs laid by flies with aculeate terminalia. Other families in the Tephritoidea contain some plant feeding species, and other closely related flies possess modified female terminalia but are saprophagous or parasitoids. Tephritoidea was recently proposed to belong to a clade of flies with variously modified oviscapt, including Psilidae, Sepsidae, and Neriodea. The placement of this Modified Oviscapt Clade within the massive post-Cretaceous Schizophoran radiation is not resolved. Similarly, the monophyly of, and relationships between, the families of Tephritoidea and relatives needs further investigation. Deciphering the evolutionary context of fruit flies will illuminate patterns in their natural history and functional morphology. With a large international collaborative network, we have obtained genomic data for nearly all major lineages in an extensively sampled matrix. We present phylogenomic inferences for the evolutionary history of Tephritoidea and relatives based on 250 taxa and fragments of 3000 nuclear protein coding genes. This approach yields a robust and compelling hypothesis by integrating data from transcriptomes, long- and short-read genomes, and Anchored Hybrid Enrichment. Within Tephritoidea, we find evidence for non-monophyly in Pallopteridae, Tephritidae, and several subfamily lineages. The placement of Neriodea and Megamerinidae in context with Tephritoidea is not settled. Larval feeding on live plant tissue is present in at least eight clades, suggesting transitions from, and/or multiple origins of phytophagy. We are at the cusp of unraveling evolutionary patterns based on compelling and comprehensive phylogenies of tephritoid flies using extensive arrays of genes and taxa.

Plenary address

How scientific studies of the house fly in the 20th century influenced language and culture; the rise of filthy flies, flyswatters, and barflies

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Keywords: house fly, barfly, *Musca domestica*, historical entomology

Among the multitudes of free-living synanthropic insects, there has arguably been no more constant human companion than the aptly named house fly *Musca domestica* L. As humans have modified habitations over time, house flies have readily colonized them; in turn, as the nature of the relationship between humans and house flies has changed, cultures and languages have changed to reflect these altered relationships. At the turn of the 20th century, an unusual confluence of events exacerbated the longstanding bifurcation of attitudes toward *M. domestica*, historically spanning the continuum from reviling them for their association with filth to drawing inspiration from them for their remarkable biological abilities. The discovery of their role as disease vectors led to increased efforts to eradicate them at the same time scientific tools (including cameras) captured behaviors leading to (often strange) bio-inspired applications. What was likely the most rapid proliferation of new fly-associated words in the English language occurred during the first half of the 20th century.

Cryptic species diversity in North American leafmining *Eugaurax* Malloch (Chloropidae)

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Keywords: CO1, DNA barcoding, *Hydrocotyle*, *Sagittaria*, leafminer, phytophagous

At least sixteen families of flies are known to mine plant leaves as larvae. Chloropidae contains species that are phytophagous, but these are mostly stem borers of graminoids and leafmining is virtually unknown in this family. We reared chloropid flies from unknown larvae mining in hosts from two plant families (Araliaceae: *Hydrocotyle* and Alismataceae: *Sagittaria*) commonly found in wetland and aquatic habitats. Specimens were collected and reared from plants in seven states, including one location in Alabama where flies were reared from both hosts growing in sympatry. We further identified the adults as members of the uncommonly collected genus *Eugaurax*, which in the US contains one widespread species (*E. floridensis*; though a northern subspecies, *E. vittatus*, has been proposed as a valid species) and another restricted to Florida (*E. cf. quadrilineatus*). Because we reared specimens from distantly-related host plants, we hypothesized that the flies represented either one polyphagous species, or two cryptic, host-specific species. Based on CO1 barcode analysis, three clades of flies became apparent, with large molecular divergences between them. Flies also grouped by host plant rather than region, alluding to the presence of cryptic, host-specific species. Our study attempts to elucidate the taxonomy and host plant use of flies in this genus. Furthermore, since both host plant species are targets for biological control outside the US, proper species differentiation is critical to understanding host range during risk assessments.

Robust taxon sampling and targeted capture phylogenomics provide insights into the evolution of parasitism in blow flies (Oestroidea: Calliphoridae)

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Keywords: blow flies, phylogenomics, trophic specialization

Blow flies (Oestroidea: Calliphoridae) are a diverse family of flies consisting of approximately 1,800 species. Owing to their feeding biology as carrion feeders and parasites, they are of forensic and economic importance. Understanding the evolutionary history of parasitism within this group is important because it can aid in mitigating the effects of parasites that act as pests and vectors of diseases in humans and livestock. However, resolving the evolutionary relationships between members and related families in this group has been problematic. Recently, phylogenomic datasets have significantly advanced the circumscription of the family. Nonetheless, taxon sampling in these studies limits the full examination of feeding habits within this group. To address this limitation, we utilized Anchored Hybrid Enrichment (AHE) to reconstruct the phylogeny of Calliphoridae. Our dataset comprises approximately 1000 loci and nearly 200 blow fly species. We applied new analytical approaches, including specialized ortholog databases, alignment pruning, and potential paralog detection, and explored their effects on estimating tree topologies within Oestroidea. This new dataset provides valuable insights into the systematics of currently proposed subfamilies, their internal relationships, and the evolution of feeding habits within the group.

Balloon flies, the *Hilara* group of genera – complexity and uncertainty in a “dark taxon” (Family Empididae)

Daniel Bickel

Australian Museum, 1 William Street, Sydney, New South Wales, 2010 Australia

Keywords: *Atrichopleura*, *Hilara*, *Hilarempis*, polythetic classification, silk glands, uncertain characters

Hilara Meigen is a speciose and cosmopolitan genus (some 400 spp., many more undescribed), but we must consider the genus *sensu lato*, the “*Hilara* group of genera”, all with the strong *de novo* synapomorphy of a swollen basitarsus housing silk glands on male leg I. Silk is used to wrap prey or debris as nuptial gifts to females, with empty silken balloons from male imposters, and males form horizontal mating swarms over water. Characters are discussed, especially orientated body pruinosity. The European fauna comprises 174 *Hilara* species: taxonomy as a cultural activity, the Würm glaciation, and 90 species in Germany compared with some 40 spp. from entire Nearctic region (!?). But the greatest species richness and generic disparity are in the Southern Hemisphere, a so-called pear-shaped distribution. The related genus *Hilarempis* Bezzi (110 spp., many undescribed) is poorly defined by venation, but there is the burden of names. The extraordinarily rich Australian fauna is the least described for any biogeographic region. What happens when hilarines lose their silk-producing basitarsi - the strange case of the New Zealand Auckland Islands fauna. Also, the remarkable radiation of *Atrichopleura* Bezzi in Australia, including bizarre genitalia and transvestism in females. Finally, polythetic classifications are necessary when taxa have undergone frequent character reversal or parallelism during evolution, or why has Gauld & Mound (1982) been so ignored?

**Taxonomic disjunction in time and space:
Atlatlia (Dolichopodidae, Medeterinae) from
Australia, New Caledonia, and Baltic amber**

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Australian Museum, 1 William Street, Sydney, New South Wales, 2010 Australia

Keywords: Australia, Baltic amber, New Caledonia

While collecting Dolichopodidae off smooth-barked eucalypt trunks in Australia, I found a striking new genus with two strong apomorphies, the dm-m crossvein absent and male genitalia on a stalked abdominal segment 7, like a spear-thrower or “*atlatl*”, hence the generic name *Atlatlia* Bickel. I found more on tree trunks and some in collections, all from southern Australia. Three more species were collected in New Caledonia, and suggesting it was a Gondwanic genus (an Australia-New Caledonia connection occurs in a number of plants and terrestrial invertebrates, possibly an ancient connection with the “lost continent” of *Zelandia*). A German collector found *Atlatlia* in Baltic amber, and the genus was transformed from “Gondwanic” to “Pseudogondwanic”, with an enormous gap in time and space. Being associated with tree trunks, Medeterinae are abundant as inclusions in Baltic amber, and the genus *Atlatlia* now includes an additional ten amber species, a major radiation in Paleogene time.

Piecing together the Baltic amber fungus gnats: a comprehensive review and material assessment

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Keywords: Baltic amber, Eocene, fungus gnats, Sciaroidea

Fungus gnats (Diptera, Sciaroidea) represent the largest group in the fossil record and are among the most diverse groups of living flies. The Mycetophilidae family alone comprises approximately 400 described fossil species (surpassed only by Limoniidae) in 126 genera. The majority of these species were described from Baltic amber by Hermann Loew (in 1850 pamphlet) and Fernand Anatole Meunier (in a series of publications in 1899-1923). However, the study of Cenozoic fungus gnats has been significantly hindered by inadequate descriptions and, in many cases, the absence of type material. In this work, we review all species of fungus gnats (Diptera, Sciaroidea, excluding Sciaridae and Cecidomyiidae) known from Baltic amber and locate remaining type material. Fungus gnats in Baltic amber are represented by 258 species names, of which 44 are *nomina nuda* and 88 are *nomina dubia*. Our revision results in 53 new combinations, 6 new species synonyms, and 2 potential generic synonyms. Lectotypes are designated for 33 species, and neotypes for 8 species. Additionally, 23 species will be described as new. Unfortunately, for 88 species classified as *nomina dubia*, the type material has been lost, and the descriptions are inadequate for neotype selection. The type material of Loew and Meunier is located in the Institut und Museum für Geologie und Paläontologie, Georg-August-Universität, Göttingen (Königsberg collection), the Paläontologisches Museum, Humboldt-Universität, Berlin (Loew material from Berendt's collection), the Natural History Museum, London (part of Berendt's collection studied by Loew), and the National Zoological Museum of Franko Lviv National University.

Normalizing the genera of Phoridae

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Keywords: ranking, systematics, taxonomy

The genera of Phoridae are in great need of revision. There are 283 current genera for the approximately 4,500 species, some of which are not necessary. Many are known from one sex only, increasing the likelihood that species are described twice, and this situation is being exacerbated by authors who do not know the literature, are unable to make proper phylogenetic decisions, or who just do not care. I review this problem, its extent, and possible ways to correct the situation.

**Phylogenomics of the bee fly genus
Lordotus Loew, 1863 (Diptera: Bombyliidae)**

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Keywords: phylogenomics

Bombyliidae or bee flies are one of the most diverse groups of insects found in deserts across the world. Despite a diversity of over 5,000 species, most are poorly studied and in need of taxonomic and phylogenetic study. The genus *Lordotus* described in 1863 by Hermann Loew, exemplifies this problem. *Lordotus* is commonly found throughout the arid and semi-arid regions in the western United States and northern Mexico, these 29 species of *Lordotus* are classified into three species groups. However, due to the morphological variability in species, *Lordotus* species have been challenging to group or to establish distinct species delimitations. Beyond what might be implied by the species-group arrangement, the phylogenetic relationships within the genus have not been explored. Here, the first phylogenetic hypothesis of the bee fly genus *Lordotus*, generated using ultra-conserved elements (UCEs), is presented. A UCE dataset consisting of 936 loci from 76 individuals representing all species of the genus was generated using a combination of flies preserved in ethanol and historic, pinned museum specimens. Maximum likelihood analysis generated a well-resolved phylogeny and a monophyletic genus *Lordotus*. The three species groups suggested by previous authors were all found to be paraphyletic. These results provide a much-needed foundation for further detailed revision of *Lordotus*.

MesSciaridae

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Keywords: dark taxa, GBOL III, Germany, molecular, morphology, Sciaridae,
taxonomy

We are currently living through a biodiversity crisis; an ever-increasing loss of biodiversity. It has never been more important, therefore, to quickly and efficiently investigate, and make an inventory of, the planet's biodiversity. This is especially true regarding understudied yet megadiverse groups, often referred to as “dark” taxa. Dark taxa tend to be small sized and highly species rich, with their real species-level diversity still unknown. One project involved in exploring dark taxa is the German Barcode of Life III (GBOL III) project. The main purview of this project is attempting to improve the size and quality of Germany's DNA barcode library, whilst also utilising an integrative taxonomic approach to investigate megadiverse insect groups, focusing on select dark taxa. The Sciaridae are one such “problem group” under the purview of GBOL III. Despite having several taxonomists working on the group, there is still much more work to be done, especially considering that recent molecular work has uncovered many potentially unnamed species contained within named species complexes. This talk will provide an overview of some of the results obtained during this project. These include an update on the species-level diversity of German Sciaridae, and some results on hidden diversity in the *Bradysia polonica* (Lengersdorf), *Bradysia trivittata* (Staeger), and *Corynoptera tridentata* Hondru species-groups using the ASAP tool.

Starting point for investigation of German *Megaselia* (Phoridae)

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Keywords: biodiversity, dark taxa, DNA barcoding, MinION, species delimitation,
species estimates

Species extinctions increase at a global scale, rapid inventorying of our planet's biodiversity is becoming more and more important. It is a pressing need to investigate insect biodiversity and accelerate species discovery and description, especially for species belonging to megadiverse and understudied insect groups, also known as "dark taxa". Phoridae and are a great example of a "dark taxon", in particular the genus *Megaselia* Rondani, which contains more than 1,700 described species worldwide. The use of an integrative methodology, combining morphology with molecular methods, is probably the best approach to face up to the task of describing hyperdiverse and "dark" taxa. Species delimitation within these taxa should be based on multiple data sources (i.e., integrative taxonomy). Here, we use the Large-scale Integrative Taxonomy (LIT) approach to sort 10,000 *Megaselia* into 277 preliminary species hypotheses based on next-generation sequencing barcode (658 bp) clusters obtained using Objective Clustering with a 3% threshold. The specimens come from southern regions of Germany; sequences were obtained with MinION. Each cluster was passed through an evaluation of the predictors for incongruence indices between barcode clusters and morphology (maximum p-distance, stability index), and a subset of specimens were subsequently morphologically examined for each cluster. This study led to description of more than ten new species. Furthermore, we provided species estimates with Chao1, and our results suggest that 318 (± 14) putative species may have been present at the sampling sites, representing an increase in species richness by 15%, whereas 21–80 additional *Megaselia* species await discovery in the areas we sampled. As this estimate was mostly based on samples from southern Germany, the species count will likely increase with expanded geographic sampling. This work is part of the GBOL III: "Dark Taxa" project of the German Barcode of Life initiative.

1,467 papers and counting: the legacy of Dalcy de Oliveira Albuquerque in dipterology after 40 years

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Keywords: Anthomyiidae, Calliphoridae, Albuquerque's students, Fanniidae, Muscidae, Nycteribiidae, Streblidae, Tachinidae

Dalcy de Oliveira Albuquerque engaged at the Museu Nacional, Rio de Janeiro in the early 1940s, studied the Muscoidea families under the supervision of Professor Hugo de Souza Lopes, a taxonomist who mostly specialized in Sarcophagidae flies. Until his death in 1982, he described 126 new species in Anthomyiidae, Fanniidae, Muscidae, Piophilidae, Psilidae, Sapromyzidae (=Lauxaniidae), and Scathophagidae families. He supervised six students on Diptera, one in 1960s (TdeJPC) and five in late 1970s to early 1980s (DMP, CJBdeC, MSC, KM, SML). The students were considered as the Dalcy's F1 students as they were the beginning of the Dalcy' lineages. Four of the Dalcy' F1 students (DMP, CJBdeC, MSC, SML) supervised several students, which were termed Dalcy's F2 students, and students of two of them (CJBdeC, MSC) supervised the Dalcy's F3 and their students supervised the Dalcy' F4 students. The number of papers published was 1,467, half in Calyptratae. The number of families studied by Dalcy's students were over 100, and the five most published papers were in Muscidae, Cecidomyiidae, Tabanidae, Calliphoridae, and Tachinidae. The subjects covered were in biogeography, biodiversity, ecology, forensic, and other areas, but most were in taxonomy and systematics. To date, some Dalcy's students are still studying; most are working in the universities, colleges, schools, and scientific or technical institutions in Brazil; and a few are working in other countries. We believe that Albuquerque's students and their offspring students are the legitimate descendants of the Travassos' School, an informal school of scientific thought, obviously influenced by many other researchers and professors from Brazil and abroad. They seek to pass on to the next generation of students the principals of dedication to science, competence, honesty, and companionship, learned throughout their academic scientific life. The students are the major legacy of the Dalcy de Oliveira Albuquerque's scientific career.

Sampling with pan traps through time shows the importance of taking wide periods to catch the diversity of the fly community

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Keywords: Chile, diversity, pan traps, plant phenology

Flower insect visitors like flies are frequently sampled in the maximum blossom period of plants, usually with comparative technics such as pan traps. These studies focus on differences among sites and often assume that abundance of flower-visiting insect must be directly related with floral abundance. However, how phenological changes in floral resource affect insect communities within the same season is less studied. Here we study how the abundance and diversity of the fly community changed through wide sampling time and in parallel with flower richness. We sampled continuously eight months (from late winter to the end of Austral summer) in a unique remnant of native forest in central Chile. Once a month, 4 pan traps of three colors (white, yellow, and blue) were operated by one week, and an active search (net and hand capture) was conducted once per week. We capture 726 individuals from 29 families. NMDS analysis showed well-differentiated flower groups in the different microhabitats sampled but flies not presented this segregation. The families Acroceridae, Bibionidae, and Syrphidae were the most representative of active search, while Bibionidae and Tachinidae were the most abundant from pan traps. Diptera abundance did not change significantly with months and years, although it tends to be higher when floral richness is low (late winter and summer). On the contrary, diversity of flies was significantly different among months ($F = 4.15^{e30}$; $p < 0.0001$), presenting three peaks of diversity that differed in family composition. These peaks occurred in similar periods to the ones found to plant phenology. The changes in fly diversity through time evidenced that continuous sampling and not only single sampling by long periods, offer a better method to study the dynamic of native flies in remnants of native forests.

Using iNaturalist as a tool for asilidology

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Keywords: iNaturalist

iNaturalist is a website and mobile application whereby users can upload photographs, video, or sound clips of organisms they encounter. Any user can also add an identification to these observations. Robber flies (Asilidae) are one of the most highly observed dipteran families on iNaturalist, second only to hover flies (Syrphidae). I use this large dataset to evaluate the app's suitability for making inferences about asilid distribution and diversity. I also share records of various rare and unusual robber flies photographed for the first time on iNaturalist, and I conclude by discussing how the app can be integrated into asilidology and scientific outreach.

The first comprehensive molecular phylogeny of the crane fly family Tipulidae s.s. (Diptera: Tipuloidea)

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Keywords: crane flies, daddy longlegs, generic level, leatherjackets, molecular, Nematocera, phylogeny

Tipulidae s.s. is a hyperdiverse family of crane flies with cosmopolitan distribution. The family contains 4351 species with larval habitats from roots of desert plants to fast-flowing aquatic habitats, even decaying wood. Despite this diversity and near-global distribution, no comprehensive hypothesis of relationships has been proposed among the 37 genera and 86 subgenera. Here we provide the first comprehensive generic level molecular phylogeny for Tipulidae s.s. constructed from DNA nucleotide sequence data from the cytochrome oxidase I subunit. From 5197 publicly available barcode records, we excluded entries without specimen images and/or species identifications, with the exception of 5 taxa only represented by generic identifications. The resulting best Maximum Likelihood tree contained 1954 specimens representing 246 species (5%) and 48 subgenera (55%) among 21 genera (56%) with strong support at the tips but weak support at deep nodes. Tipulidae remained monophyletic and sister to *Cylindrotomidae*. We did not recover the three-subfamily topography based on accepted classification, instead finding a topography of eight main clades largely incongruent with current classifications. We recovered most of the *Lunatipula-Vestiplex* clade *sensu* Gelhaus at a deeper node and not grouped with the type subgenus of *Tipula* Linnaeus, but is sister to a clade including diverse subgeneric elements of *Tipula* such as *Platytipula* Matsumura and *Nippotipula* Matsumura. Ctenophorinae (excluding *Pselliophora* Osten Sacken) is sister to a clade of *T. (Tipula) + T. (Acutipula)* Alexander. *Pselliophora* forms a clade with *Leptotarsus* Guerin-Meneville, *Brachypremna* Osten Sacken, *Ptilogyna* Westwood, and *Ozodicera* Macquart. *Nephrotoma* Meigen was found to be sister to all other Tipulidae, and *Tipula (Yamatotipula)* Matsumura was found 6 times across the tree and is likely polyphyletic and paraphyletic. We suggest no taxonomic changes without similarly comprehensive analysis using robust, next-generation sequencing technique to confirm our results, which is underway.

Advances in the systematics of three unusual genera of Steganinae from the Western Hemisphere

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Keywords: Acletoxenina, comparative biology, phylogeny, vinegar flies

This study presents the advances on the systematics of the Steganinae (Drosophilidae) genera of the tribe Acletoxenina: *Pseudiasata* Coquillett, *Hyalistata* Wheeler, and *Mayagueza* Wheeler based on material from the Neotropical region. *Mayagueza* was a monotypic genus but in 2021 a second species was described: *Mayagueza lopesi* Costa et al., 2021. That was also the first record of this genus for South America. In the following year, *Hyalistata* was recorded in Brazil, and two new species were described: *Hyalistata cerradensis* Costa et al., 2022 and *H. emas* Costa et al., 2022. Furthermore, the Brazilian species of *Pseudiasata*—*P. brasiliensis* Costa Lima—originally described in 1937, was redescribed with designations of lectotypes, and paralectotypes. These three genera share similarities in regard to their geographic distribution (Nearctic and Neotropical regions), their biology (predatory flies), and shared morphologies (as setae on pregonite, and structure not divided of the phallus). Their similarities raised doubts about their taxonomy and circumscription of these three genera. We conducted a phylogenetic analysis for *Hyalistata*, *Mayagueza*, *Pseudiasata*, and close relatives (≈ 20 terminal species) based on 76 morphological characters. Our results indicate that they are reciprocally monophyletic and closely related. We provide characters that contribute to the delimitation of the groups, with a discussion of diagnostic characters.

A new illustrated identification key to Nearctic genera of Empidoidea (exclusive of Dolichopodidae s.str.)

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Keywords: Atelestidae, Brachystomatidae, Dolichopodidae *s.lat.*, Empididae, Hybotidae, Iteaphilidae, key, Nearctic

The Empidoidea (exclusive of Dolichopodidae *s.str.*), or dance flies, are a diverse group of six families and 72 genera in the Nearctic Region. An illustrated key to the dance fly genera is presented, updating the key from the Empididae chapter of the Manual of Nearctic Diptera and utilizing more than 400 colour photographs displaying many unique characters as well as some rarely seen or photographed genera. Since the 1981 key, 16 additional genera and two undescribed genera are now recognized in North America, north of Mexico. Currently, 953 species of Empidoidea (exclusive of Dolichopodidae *s.str.*) have been described. A brief synopsis of each genus is included with the key, providing information on number of species, taxonomic literature, biology, defining morphological characters and Nearctic distribution.

An attainable approach to specimen digitization with implications in data collection, research communication and end-user accessibility

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Keywords: character matrix, hypothesis testing, illustration, morphology, specimen database, taxonomy

Documentation of prepared specimens typically involves imaging, drawing or a combination of the two. Methods commonly used for both have advantages and disadvantages with respect to effectiveness and attainability. We present an approach to specimen imaging that involves capturing video of microscope observations in 4K resolution. Resulting MP4 video files include data captured from the X,Y and Z axes, similar to focus stacking or cross-sectional images comprising a CT scan. Video scrubbing speed and direction can be manipulated to create the effect of image continuity despite rapid variation in depth. This technique is especially useful for imaging of transparent specimens where pertinent information at points on the X and Y axes may be obscured at different points on the Z axis. Video files are readily transferrable, easily viewed using freeware and may be associated with specimens and uploaded to a database. They can also be annotated in real time to aid in communication or serve as the basis for production of drawings and still images.

**Untangling meniscus midge male terminalia:
development of descriptive language and visualization tools
to facilitate research communication**

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Keywords: 3D models, homology, species descriptions, taxon delimitation,
terminology

Pertinent aspects of the ground plan of dixid male terminalia have been poorly characterized or overlooked in treatments of Dixidae, with few exceptions. An assessment of homologies in the male terminalia of all major lineages of Dixidae, where specimens are available, was recently completed. Outgroup taxa include representatives from all remaining families of Culicoidea and few other families of Culicomorpha. Basic components of the ground plan for Dixidae and landmarks allowing for accurate delimitation of each have been identified. Focus stacked images, 2D color-coded illustrations and animated 3D models are employed to visualize complex structures and illustrate diagnostic characters. Challenges related to descriptive language range from a lack of terms to characterize specific details of complex structures to multiple terms being used to describe the same structure. Proposals for a revised terminology include novel terms, development of existing terms and elimination of redundancy.

The state of North American aphidophagous syrphid larvae (Syrphidae) descriptions and knowledge gaps

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Keywords: larval description, photo key, North America, taxonomy

North American aphidophagous syrphid (Syrphidae) larvae have been chronically understudied and infrequently described. Differences in description detail, identifying characters, incorrect species identification, and duplicate efforts make it difficult to navigate and apply existing literature. Adequate aphidophagous syrphid larvae descriptions, along with potential host range and larval habitats, are essential to future ecological studies and potential biological control efforts. This project will examine aphidophagous syrphid species of the subfamily Syrphinae found in Northeastern North America, as listed in Skevington et al (2019). I reviewed primary literature from authors situated in North America and Europe to determine the current status of North American syrphid larvae descriptions. Larval forms of at least 56 of 139 Northwestern North American Syrphinae species (40%) have been described to mixed degrees. Larvae of the genera *Allograpta* Osten-Sacken, *Eupeodes* Osten-Sacken, *Psuedodoros* Becker, and *Syrphus* Fabricius are relatively well circumscribed, whereas *Dasysyrphus* Enderlein, *Paragus* Latreille, *Parasyrphus* Matsumura, *Platycheirus* Lepeletier & Serville, and *Toxomerus* Macquart remain poorly known. This project involves extensive fieldwork to fill knowledge gaps identified throughout my literature review. Our 2022 collection season resulted in the successful rearing of about 15 species of aphidophagous syrphid larvae found in southern Ontario, Canada. The material will contribute to new descriptions or redescriptions where necessary. Future work will address knowledge gaps in select genera, generate high quality images and keys, and explore the biological control potential of common North American species.

**Scavenger to parasitoid, all in one species:
the amazing flexibility of many flesh fly life histories**

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Keywords: adult host discrimination, *Helicobia morionella*, *Helicobia rapax*, larval feeding, *Peckia lambens*, Sarcophagidae

Most parasitoids have developed obligate relationships with their hosts while most scavengers show no ability to develop as endoparasitoids. In the Sarcophagidae there are many examples of species that have been reared as scavengers, agents of vertebrate myiasis, and as endoparasitoids of other insects. Three recent rearings of flies from adult Japanese beetles and honey bees are discussed along with an overview of this unusual flexible life history among the families of calyprate Diptera.

Using 3rd instar larval morphology for identification of Sarcophagidae of forensic importance in Jamaica

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Keywords: forensic entomology, Jamaica, larval morphology, third instar

The field of forensic entomology is an important and established discipline proven throughout its development to be an asset to judicial systems and investigations. In most of the Caribbean, forensic entomology is currently more theory and research-based than practical in application, due to deficiencies in resources, training and/or baseline information needed. This is the current case for the island of Jamaica, which has long since battled with a backlog of unsolved crimes. For this discipline to be credibly used in practical applications a variety of baseline data are needed and this work is currently being done at the University of the West Indies Mona campus. The present research is a part of this initiative and investigates Sarcophagidae of forensic importance found in Jamaica and aims at the identification of these flies using 3rd instar larval morphology. Van Someren Rydon traps, baited with meat at various stages of decay were used to capture adult flies, which were subsequently allowed to larviposit. The resulting larvae were reared under laboratory conditions and morphological studies were done using light microscopy following microdissection and clearing techniques. The key morphological features studied include anterior and posterior spiracles, the cephalopharyngeal skeleton and the anal segment. Two species of international forensic importance were found during this study, these were *Peckia chrysostoma* (Wiedemann) and *Blaesoxipha plinthopyga* (Wiedemann) Illustrations of the third instar larval features were made for each species. This research aims to add the growing entries of baseline data for forensic entomology specific to Jamaica.

Methods of focus stacking for live insect photography

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Keywords: amateur, focus stacking, live insects, methods, photography, software, techniques

This poster describes the steps of focus stacking images of live insects for a wide depth of field, including how to avoid unintended alteration of color or morphology, as well as opportunities for image-based research and identification. Focus stacking is a powerful technique for creating images with a wide depth of field by combining multiple photographs from different focal planes into a single image. This is particularly useful for images that have been magnified to the extent that the natural depth of field is extremely narrow, as is the case in macro photography. Focus stacked images of insects enable significant characters such as setae and wing venation to appear entirely in focus. Such images facilitate research on the characteristics and behavior of live insects, as well as the discovery of new species. Focus stacking is widely used for imaging both specimens and live insects, but the mechanics of capturing the underlying data in the field differ from those in the lab. Variations in hardware and automation will be highlighted in this poster using data sets and images created by the author. Several software applications for focus stacking are readily available at various levels of functionality and price. Unintended effects of image processing, such as the accidental alteration of body parts, can impact the identification of insects, including by creating features that were not originally present or removing features that were. Examples of these effects will be shown in this poster.

Diptera pollination in Australian commercial cropping systems

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Keywords: *Calliphora*, Calliphoridae, cropping systems, *Eristalis*, *Eristalinus*,
Syrphidae

The growing recognition of Diptera as pollinators has led some fly species to be managed and deployed for crop pollination services in many regions of the world. However, their efficiency as pollinators of common global food crops is often overlooked and most management practices to support pollinators focus predominately on bee resource needs. Here we tested the pollination efficacy (single visit pollen deposition, fruit/seed set) of wild and/or managed flies in four Australian commercial cropping systems: blueberry, raspberry, blackberry, and hybrid seed carrot. We also investigated the life cycle needs of global crop pollinating flies to determine how to best support fly pollinators in agricultural landscapes. We found no significant differences in the pollination services provided by the Western honeybee (*Apis mellifera* Linnaeus, 1758) and one fly taxon, the cosmopolitan syrphid fly *Eristalis tenax* (Linnaeus, 1758), in all cropping systems tested. Two additional fly species endemic to Australia, the calliphorid fly, *Calliphora stygia* (Fabricius, 1752) and the syrphid fly *Eristalinus punctulatus* (Macquart, 1847), were also effective pollinators of *Rubus* (raspberry and blackberry) and hybrid seed carrot, respectively. In general, our results suggest that some flies can be effective supplementary pollinators to managed bees in the crops tested. Further, we highlight the resources needed to support the life cycles of wild flies known to visit global cropping systems and suggest ways to improve habitat within agroecosystems to support beneficial fly oviposition and larval development.

The unlikely siblings: an unexpected twist in Tanypodinae's subfamily reunion

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Keywords: mouthparts, morphology, evolution, Tanypodinae, fossil

Tanypodinae is the third most species-rich subfamily within the family Chironomidae, exhibiting a global distribution and occupying a diverse range of aquatic habitats, including lentic and lotic systems. The recent surge in enthusiasm for total evidence approaches through comprehensive analyses combining molecular and morphological data has reinvigorated the exploration of the evolutionary history of this subfamily. Despite the high quality of the data used in these analyses, it is important to note that they do not incorporate fossil information. Instead, they rely on indirect methods that, in the worst case, may lead to misrepresentations of the fossil record. In this investigation, we conducted a comprehensive analysis of molecular and morphological characters, examining all life stages and tribes, and incorporating evidence from both modern and fossil chironomids. Our main objectives were twofold: (i) to evaluate the impact of incorporating fossil evidence on the current hypothesis of phylogenetic relationships within Tanypodinae, and (ii) to investigate the evolutionary implications of mandibulate functional mouthparts and their role in shaping the relationships within this subfamily. The monophyly of the Tanypodinae is supported, with strong evidence for the internal relationships, confirming the existence of all proposed tribes. Moreover, despite the inclusion of fossil evidence, the overall internal relationships within Tanypodinae, previously established by other studies, remain largely unchanged. However, upon analyzing the fossils, an interesting finding emerges. Fossils displaying functional mandibles consistently formed a distinct cluster, positioned at the base and representing a sister group to the remaining tanypodine. This intriguing finding suggests that the presence of functional mouthparts, enabling efficient feeding strategies and adaptations to specific ecological niches, may have exerted a substantial influence on the early evolutionary processes and subsequent diversification of Tanypodinae. This provides valuable insights into the evolutionary significance of mandibulate functional mouthparts in shaping the evolutionary history and ecological success of this diverse group.

Diptera (Schizophora) from a mining area in the Brazilian Amazon

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Keywords: Amazon, biodiversity, Brazil, mining, restoration, sampling protocol

The lack of good ways for measuring biodiversity makes it hard to implement conservation plans, especially, in the Brazilian Amazon, where biodiversity is highly threatened by human changes, such as mining. To study the flies' biodiversity in a modified area by mining, samplings were carried out in the Hydro area of the Paragominas Mining Company S.A, localized in the "Arc of Deforestation" in the Brazilian Amazon. This sampling made part of the project "Entomological study and bioindicators selection to the biodiversity monitoring in the mining of Paragominas S.A. The specimens were collected with pitfall, Winkler, and malaise traps, in the years 2019 and 2020, in five expeditions. The samplings were realized in four different areas: forest (never mined), seedling planting, natural regeneration, and assisted natural regeneration (nucleation). These three last areas cited are restoration techniques used in the Brazilian Amazon. The material is still being processed, and, due to the pandemic, there are still some expeditions to make. Until now, were sampled 52 dipterans families, in a total of 16.465 specimens. The more abundant families are Cecidomyidae (n=6.447), Ceratopogonidae (n=1.856), Phoridae (n=1.264), Psychodidae (n=1.244), Dolichopodidae (n=943), and Sarcophagidae (n=722). The less representative families are Mesembrinellidae, Nemestrinidae, Neriidae, Ropalomeridae, Scatopisidae, and Tephrididae, with just one specimen caught. We also have several new species of different families, eg. Clusiidae (*Sobarocephala* Czerny), Agromyzidae (*Calycomyza* Hendel), Chloropidae (*Pseudogaurax* Malloch), and Sarcophagidae (*Boettcheria* Parker), and some to confirm. The area with the biggest abundance was nucleation (n=5.320), followed by seedling planting (n=4.751), natural regeneration (n=3.339), and forest (3.014). There is much more to study on the flies of this area, and, after careful analysis, we will be able to infer which restoration strategy is better to recover the biodiversity in the Amazon, besides discovering more new species and understanding the flies' distribution in that area.

Sarcophagidae (Diptera: Oestroidea) from two mangrove forests in the Brazilian Amazon

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Keywords: Amazon, anthropization, bait types, mangroves, species richness

The Sarcophagidae family is the second most diverse within the Oestroidea superfamily, with over 3100 known species. In the Brazilian Amazon, approximately 150 species are found. However, little is known about the community of Sarcophagidae in this region, especially in floodplain forests and estuaries. This study was conducted in two Amazonian mangroves forest in Pará, Brazil, one impacted by human interference and the other undisturbed. Four collections took place between December 2015 and April 2016, with traps exposed for 48 hours in 15 sampling units in each mangrove. The baits used included rotting beef lung, fish, crab, and shrimp. The integrity of the mangrove was assessed to determine differences in estimated and observed species richness and composition between the two treatments. A total of 805 specimens representing 26 species and 10 genera were collected, including three recently described species. The presence of the genus *Hallina* Lopes in Brazil, as well as the species *Peckia* (*Peckia*) *gulo* Fabricius, and *Peckiamyia minutipenis* Hall in the Brazilian Amazon, and *Helicobia morionella* Aldrich in Pará state, are new records. The preserved mangrove yielded 26 species, while the altered mangrove had 15 species, with 15 species shared between both environments. The Jack1 method estimated the richness of 36 species in the preserved mangrove and 15 species in the altered mangrove, which matched the observed richness in the latter. Significant differences in observed richness were found between the altered and preserved mangroves, and the composition of Sarcophagidae varied as well. While previous studies suggested higher richness in anthropized environments, this was not observed in these mangroves. The use of diverse bait types, including locally available resources, is crucial for attracting specific species. The results imply that human activities in the mangrove may account for the differences in Sarcophagidae richness and composition compared to the preserved mangrove.

Altitudinal homogenisation of mountain parasitoids across six decades

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Keywords: altitudinal range, diet specialization, Europe, Tachinidae

Elevation gradients, characterized by significant environmental changes over short geographical distances, offer valuable insights into how biodiversity responds to climate change. High-altitude species often occupy specialized niches that render them particularly vulnerable to rapid environmental transformations. Recent evidence indicates that elevation strongly influences both the composition and intensity of host-parasitoid interactions. Parasitoid insects, crucial in regulating host populations and preventing outbreaks of herbivorous insects, may see their regulatory role impacted by climate change, especially in the case of specialized feeders. In this study, an exceptional dataset comprising over 46,000 records of parasitoid bristle flies (Diptera: Tachinidae) from various regions in Europe, spanning a wide range of altitudes and six decades, was analyzed. The objective was to examine the temporal trend in the likelihood of encountering bristle flies with specialized diets (referred to as "oligophagous" species) at different altitudes. The aim was to determine whether the altitudinal gradient in specialization has undergone changes over time. The findings revealed a distinct reshuffling of bristle fly assemblages along altitudinal gradients, with specialized species becoming less abundant at higher elevations. Six decades ago, there was a clear altitudinal gradient in the proportion of specialized feeders, but over time, this proportion has become remarkably similar across different altitudinal bands. These results indicate an emerging homogenization in the dietary preferences of parasitoid communities, which could potentially reshape the ecological dynamics of mountain ecosystems by altering the mechanisms that regulate insect herbivory.

Mydidae of the central Namib Desert at the Gobabeb Namib Research Institute, Namibia

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Keywords: Afrotropical, desert

The Namib Desert is one of the oldest and driest deserts on Earth and harbors many unique taxa – the fly fauna is no exception. The central Namib Desert in and around the Gobabeb Namib Research Institute encompasses the ephemeral Kuiseb River, a linear oasis, the Namib Sand Sea, with towering shifting dunes, and the desert gravel plains. The plant and animal life is influenced by recurring fog from the Atlantic Ocean, which is approximately 60 km due West. Here, the Mydidae fauna at Gobabeb is summarized, with data obtained from taxonomic revisions, museum collections, and field-work. A year-long faunistic study by Wharton (1982) provided the groundwork for understanding the Mydidae diversity at Gobabeb. Because of the incomplete knowledge and difficult-to-use identification keys of the diverse Mydidae fauna in southern Africa at the time, Wharton's identifications were mostly incorrect. To date, 11 Mydidae species of nine genera, representing two subfamily taxa, have been recorded at Gobabeb. Of these, three species have only been collected in the Gobabeb area thus far and are considered endemic to this region. Three genera and five species remain undescribed. The uniqueness of this Namib desert fauna is its astounding diversity. There is no other place on earth where over a single year during different seasons 11 species of Mydidae can be observed and collected. While some species such as *Eremohaplomydas gobabebensis* Boschert & Dikow, 2022 have only been recorded in a single month (November), other species are active over several months such as *Parectyphus namibiensis* Hesse, 1972 (June–September) or *Namadytes vansoni* Hesse, 1969 (January–June). Photographs of the habitats and flies are provided and the seasonality and habitat preferences of species are summarized to illustrate the outstanding diversity in these unique desert habitats.

New discoveries and phylogenetic placement of assassin flies (Asilidae) in Cretaceous and Paleogene ambers

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Keywords: Baltic amber, Bitterfeld Amber, Burmese amber, Rovno Amber

The extinct assassin flies (Asilidae) preserved in Cretaceous and Paleogene ambers are reviewed and phylogenetically placed. The fauna in Cretaceous ambers from Myanmar and the USA (Burmese and Raritan) have recently been reviewed recognizing one species from each amber deposit based on three and one specimen, respectively. An additional female specimen preserved in Burmese Amber is described representing a new species, which does not belong in *†Burmapogon* Dikow & Grimaldi, 2014 as wing cell *cua* is closed, postsutural dorsocentral setae are present, and trowel-shaped metathoracic tibial spines are absent. Paleogene: Eocene amber from northern Europe (Baltic, Bitterfeld, and Rovno) is well-known for preserving insects. Three Asilidae species have been recorded from Baltic and Bitterfeld ambers to date. With the availability of 28 additional pieces of Baltic and Rovno amber with assassin-fly inclusions, the species diversity can be increased to six, representing two of the previously described species. Several inclusions are not well-preserved to allow formal description and placement. It is concluded that the Baltic, Bitterfeld, and Rovno deposits preserve the same insect fauna and that the most abundant Asilidae in Eocene amber, *Protolewinella keilbachi* Schumann, 1984 (Laphriinae, 16 specimens), is found in all three deposits. Five genera and four species are hypothesized to represent new, undescribed taxa. The phylogenetic placement is based on an expanded morphological matrix based on Dikow (2009) representing all subfamily taxa, additional extant species, and nine amber fossils. The Cretaceous amber species could not be coded for 32–78% and Paleogene amber species for 28–48% of characters. The backbone relationships within Asilidae remain largely unresolved but several subfamily taxa are recovered as monophyletic. Several Eocene fossil taxa enhance the relationships within these subfamily taxa and are grouped deeply within radiations. The unique character combinations found in some extinct taxa make phylogenetic placement a challenge.

**The Nearctic and Neotropical Asilidae fauna:
Eric M. Fisher and his contributions**

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Keywords: North America, Central America, South America

The assassin-fly fauna of the Americas is unique and diverse in many respects. The study of this region dates to the late 18th century when the first species were described by Linnaeus, Fabricius, Olivier, and later dipterists have added to the understanding. Since then, very few dipterists have known and understood this fauna as Eric Fisher does. Eric published peer-reviewed articles starting in 1977 and continues with taxonomic discoveries and reviews to this day. Eric has focused much of his work on the Laphriinae, with important publications on the Nearctic Laphystiini and his dissertation on Andrenosomatini. One of Eric's most impactful publications is the Asilidae chapter in the *Manual of Central American Diptera* published in 2009. However, the depth of his knowledge lies deeper than these publications. Throughout his career, he has assembled data, morphological observations, and specimens of Asilidae, which were instrumental in gaining this more profound understanding of the Asilidae fauna – knowledge and specimens that he has shared broadly with many students. Eric's field-work has been especially extensive in the south-western USA, Mexico, Costa Rica, Panama, and Peru. Additional trips have taken him to Brazil and Venezuela. Through this field-work and museum material, Eric established that there are 97 species in the western Amazon basin at Tambopata in Peru and 108 in the lowland rainforest at La Selva in Costa Rica, which make these localities the most diverse (known) areas for Asilidae. Specimens collected and pinned by Eric are exceptionally useful for research now and in the future because every specimen is perfectly prepared and ready for photography. It is a great pleasure to convene an Asilidae symposium at ICDX to honor Eric Fisher and his contributions to understanding the diverse Nearctic and Neotropical assassin-fly fauna.

TaxonWorks as a tool for managing large biodiversity projects

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Keywords: biodiversity, collaboration, database, nomenclature, taxonomy,
TaxonPages

Large systematic revisionary projects incorporating data for hundreds or thousands of taxa require an integrative approach, with strong biodiversity-informatics core for efficient data management to facilitate research on the group. TaxonWorks, a product of the SpecieFile Group, may offer such infrastructure. TaxonWorks is an open-source web application. It follows the best practices in the modern biodiversity database development (TDWG standards) to bring together multiple projects and users. The core of each project is taxonomy, which could be as simple as a list of name, represented by Operational Taxonomic Units (OTUs), with some biological properties and relationships; or it could be as complicated as hierarchical nomenclature with defined set of rules bases on the International Code of Zoological Nomenclature (ICZN), and accompanied with a set of nomenclatural and taxonomic statuses defined by NOMEN ontology. Hundreds of validations are implemented, to highlight the missing information and ensure the integrity of the data. The second largest component of TaxonWorks, is the set of interfaces for the efficient management of the specimen related data. Several large collections were migrated into TaxonWorks for their digitization needs. The tools are provided for specimen and loan management, semiautomatic data parsing, and georeferencing. Morphological and image matrices in TaxonWorks are used to build interactive keys or standard exports for phylogenetic analysis. TaxonWorks provides highly customizable annotations. Any object in the database could be annotated with tags, notes, illustrations, citations, confidence levels, translations, etc. Data from TaxonWorks could be exported in raw format, get accessed via Application Programming Interface (API), or be shared with external data aggregators, like Catalogue of Life, GBIF, iDigBio, using COLDP or Darwin Core exports. The public interfaces provided via separate application, TaxonPages and could be customized to meet the project needs.

Fleshy beauties – uncovering the megadiverse fauna of gall midges on succulent Aizoaceae in Southern Africa

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Keywords: Afrotropical, Cecidomyiidae, galls, host association, phylogeny, speciation

Succulent Aizoaceae (“ice plants”) constitute one of the most species-rich families in the Greater Cape Floristic Region (GCFR), with ca. 1200 species and over 70% endemism. Yet, insect diversity on these plants has hardly been studied, and knowledge on gall-inducing insects in the Afrotropics as a whole is scarce. In an ongoing study of the gall-midge fauna associated with succulent Aizoaceae in South Africa, we have so far discovered more than 50 species of cecidomyiids, representing an entirely unstudied and undescribed fauna, including at least 2 new genera. Diversification of the gall midges on these plants has occurred independently in two cecidomyiid tribes, showing similar evolutionary trends. The majority of species from both tribes are found on plants belonging to the “core” Ruschioideae, which is the most speciose and most rapidly diversified clade within the Aizoaceae, with most of the cecidomyiid species being highly host specific. Reflecting this host specificity, our molecular phylogenetic analyses suggest that diversification of the gall midges generally mirrors that of their host plants, albeit with certain exceptions and with strong geographic influences. This study highlights the great potential for the discovery of new taxa in the Afrotropical Region, particularly in little studied, mega-diverse groups such as gall midges, while also advancing our understanding of cecidomyiid evolution and their association with their host plants more generally.

Comparing population genomic strategies for developing diagnostic markers in *Anastrepha ludens* Loew (Tephritidae)

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Keywords: molecular diagnostics, phytosanitary management, population genomics, sterile insect technique, SIT

For hard-to-distinguish species and strains, genome-wide single nucleotide polymorphism (SNP) data sets have revolutionized diagnostic identification. The sheer size of these data sets, often ranging from hundreds to hundreds of thousands of markers, increases the genomic resolution for finding unique, population/species-specific alleles. Although numerous studies have used genomic-scale data to develop diagnostic panels (typically dozens to hundreds of SNPs), few have assessed 1) the efficacy of different population genomic strategies in identifying diagnostic markers, and 2) how to reduce large genomic datasets most effectively to a minimum set of highly informative markers. Here, using both wild and mass-reared individuals of the agricultural pest *Anastrepha ludens* Loew (Mexican fruit fly), we developed panels of diagnostic markers from two different types of genome-wide SNP data sets (double digest restriction-site associated DNA sequencing (ddRAD) and whole genome sequencing (WGS)). The ultimate goal of these panels is to facilitate determining geographic source of invasive flies and to delimit strains used for Sterile Insect Technique. For each type of SNP dataset, we identified highly informative markers and compared their individual and combined utility for identifying field-caught *A. ludens* samples. This information has the potential to reduce per-sample analysis costs, first, when creating the genome-wide SNP datasets, and second, by reducing the quantity of markers to genotype, and thus will be informative for research in many systems where diagnostic SNP markers are used.

Combining natural enemies and mass trapping for stable fly management

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Keywords: IPM, parasitoids, predators, *Stomoxys calcitrans*, Stomoxyinae, trapping

Stable flies (*Stomoxys calcitrans* (Linnaeus)) are serious livestock pests all over the world. Costs generated by these flies within the livestock sector has been estimated at US\$ 2.2 billion/year in United States. In France the losses in the meat sector are estimated at 145 M  per year, and 234 M  per year in the milk sector. These losses are due to the nuisance caused by painful bites, which prevent the animals from feeding properly, and potential transmission of pathogens. Using insecticides for control damages the environment and faces resistance problems. Resistance of flies to insecticides has been shown both phenotypically and genetically. An integrated pest management solution for the control of stable flies, using a combination of natural enemies to control immature stages and a specific trapping system for adults, is proposed. The Stomoxycc  trap (Alcochem company, <https://stomoxys.com>) is used for mass trapping of adult *Stomoxys*. As natural enemies of stable flies, Biomite and Biowasp (Bestico company) contain the predatory mite *Macrocheles robustulus* (Berlese) and 2 parasitoid species *Spalangia cameroni* Perkins and *Muscidifurax raptor* Girault & Sanders, respectively. Both predators and parasitoids are released monthly on animal bedding. This IPM has been implemented in a large donkeys/mules/horses sanctuary in the south of Spain (El Refugio del Burrito) in June 2022. Preliminary results indicate a good impact on the fly population. More results will be available in the 2023 fly season. Do not forget that good management of effluents is necessary in farms, but control of stable flies without the use of insecticides, associating trapping of adult flies and biocontrol of eggs and larval stages, would be a good solution.

Parasitoids, predators, and mass trapping for stable fly management, a new IPM approach

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Keywords: IPM, parasitoids, predators, *Stomoxys calcitrans*, Stomoxyinae, trapping

Stable flies, *Stomoxys calcitrans* (Linnaeus), are serious livestock pests all over the world. Larvae develop in various substrates such as rotten plant material or manure. Emerged adults bite animals and human beings for blood meals. Costs generated by these flies within the livestock sector has been estimated at US \$ 2.2 billion/year in United States. In France the losses in the meat sector are estimated at 145 M  per year, and in the milk sector it is rising a yearly average loss of 234 M . These losses are due to the nuisance caused by painful bites, which prevent the animals from feeding properly, and to the potential transmission of pathogens. Blood loss can be very significant during peaks of abundance. Chemical control by using insecticides is often used. This control method damages the environment and faces resistance problems. Resistance of flies to insecticides has been shown both phenotypically and genetically. We propose an integrated pest management solution for the control of stable flies: Using a combination of natural enemies to control immature stages and a specific trapping system for the adults. The Stomoxyc  trap from the company Alcochem is used for mass trapping of adult *Stomoxys* (<https://stomoxys.com>). As natural enemies of stable flies, we use Biomite and Bliowasp (Bestico) containing respectively the predatory mite *Macrocheles robustulus* (Berlese) and 2 parasitoid species *Spalangia cameroni* Perkins and *Muscidifurax raptor* Girault & Sanders. Both predators and parasitoids are released monthly on animal bedding. This IPM has been implemented in a large donkeys/mules/horses sanctuary in the south of Spain (El Refugio del Burrito) in June 2022.

Plenary address

Leaf-mining Diptera in North America

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Keywords: Agromyzidae, Anthomyiidae, Chironomidae, leafminer, natural history, Sciaridae, species discovery, Tephritidae

Leafminers are insects that spend at least part of their larval lives living and feeding between the epidermal layers of leaves. Although Agromyzidae is by far the most diverse group of leaf-mining Diptera, leafmining is also known in North American species of Anthomyiidae, Chironomidae, Chloropidae, Dolichopodidae, Drosophilidae, Ephydriidae, Lauxaniidae, Scathophagidae, Sciaridae, and Tephritidae (as well as ~50 families of Lepidoptera, Coleoptera, and Hymenoptera). Many Cecidomyiidae also feed within leaves, but none are true miners because the larva's living space is created by the plant's response to its presence rather than excavated by the larva. Some Cecidomyiidae, Chironomidae, Chloropidae, Drosophilidae, Lauxaniidae, and Phoridae also develop as secondary inhabitants in abandoned or occupied mines of other insects. This slideshow will give an overview of the groups of North American leaf-mining Diptera and their natural history, highlighting new discoveries made during the past 12 years as I prepared the first two editions of the *Leafminers of North America* e-book, traveled throughout the USA in search of leafmines, and perused tens of thousands of photos of mines on BugGuide.net and iNaturalist.org. These discoveries have included the description of 58 new leaf- and stem-mining species of Agromyzidae, Chironomidae, and Sciaridae; documentation of previously unreported introduced or Holarctic species of Agromyzidae, Ephydriidae, and Tephritidae; and new host and distribution records and natural history information for numerous other species in the eleven leaf-mining families.

Survey of Diptera species and habitats in seven states in Sudan

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Keywords: Asilidae, River Nile, *Scatella*, Sudan

Sudan is uniquely located, representing an interface region between the West-Palaeartic and Africa. West-Palaeartic insect species meet the northern limits of some African species here. Despite this interesting geo-zoological aspect, the region has been poorly investigated. Diptera research in Sudan has focused mainly on agricultural pests and flies with medical importance. Therefore, the proposed study aimed to survey Diptera and assess their preferred microhabitats in Sudan. The research was conducted in seven states (Khartoum, White Nile, North Kordofan, South Kordofan Red sea and River Nile state). Diptera collected from Semi desert, Savanna, Red Sea Coast and Nuba Mountains in June, July and August 2022. Of the 824 samples collected, the Diptera belonged to 24 families, with 139 morphospecies. Robber flies (Asilidae) showed the highest diversity, represented by nine species collected from all study areas. This was followed by the Bombyliidae. Samples were collected using three methods – sweep net, pitfall traps and coloured pan traps (yellow, blue, white; 10 cm*30 cm). Traps were set up in 50 m transects in all visited locations. The study confirmed open habitats with scattered trees and grass (North Kordofan and El Musawwarat) were preferred more than fully covered and shaded habitats (South Kordofan). River Nile state recorded most of species and showed high diversity in comparison to other states. Species of *Scatella* (Ephydriidae) and *Asyndetus* (Dolichopodidae) were collected from El Musawarrat and recorded for the first time from Sudan to Agricultural Research Corporation (ARC), Entomological Group Sudan.

The robber flies (Asilidae) of Mexico

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Keywords: diversity, Mexico, taxonomy

Asilidae is one of the most diverse subfamilies within the order Diptera with 7520 described species in 555 genera and 14 subfamilies worldwide. For Mexico the number of species remains unknown due to the group has been poorly studied and neglected. The aim of this contribution is to provide a checklist of Mexican species and an illustrated dichotomous key to the genera that are presented in the country. So far, 5000 specimens from 19 entomological collections have been examined. A database has been created which gathers information from both specimens and literature. Altogether some 490 species in 96 genera and 11 subfamilies of Asilidae have been determined for Mexico, including eight undescribed species, five new country records and 10 new state records (*Cystoprosopa sepi*a Hull, from Oaxaca; *Lastaurus lugubris* (Macquart), from Chiapas and Veracruz; *Lissoteles vanduzeei* Cole, from Baja California Sur; *Mallophora tsacasi* Artigas & Angulo, from Campeche, Chiapas, Michoacán, Oaxaca, Quintana Roo and Veracruz; *Orrhodops occidentalis* (Williston), from Nayarit; *Philonicus truquii* (Bellardi), from Estado de México, Guerrero, Jalisco, Michoacán, Morelos, Oaxaca, Puebla, Tamaulipas and Veracruz; *Ph. tuxpanganus* (Bellardi), from Guerrero, Morelos and Oaxaca; *Saropogon bryanti* Wilcox, from Oaxaca and Puebla; *Sintoria emeralda* Hull, from Morelos, Nuevo León and Zacatecas; and *Nevadasilus blantoni* (Bromley), from Durango). However, despite of these findings, the taxonomic knowledge for this group is still insufficient and further work is deeply needed in exploring both field and entomological collections.

Searching for synapomorphies: the case of Orthoclaadiinae Genus 5 (Chironomidae)

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Keywords: classification, generic concepts, life stage association, morphology,
synapomorphy

“Orthoclaadiinae Genus 5” is a member of the *Parakiefferiella* genus group which has been included as a provisional taxon in several keys to Nearctic Chironomidae pupae but has never been formally described. Newly associated larvae and pharate males confirm this placement but there are no unambiguous morphological synapomorphies defining the taxon, which blurs the distinction between the related genera *Parakiefferiella* Thienemann, *Rheosmittia* Brundin and *Krenosmittia* Thienemann & Krüger. This illustrates a larger problem with generic concepts in Orthoclaadiinae as numerous genera are based on typological character formulae rather than synapomorphies, demonstrating a need to reexamine generic diagnoses within the subfamily.

A brief review of *Diasemopsis* Rondani (Diopsidae) with discussion of its first species-group

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Keywords: Afrotropical, species-groups, systematics

Diasemopsis was erected by Rondani with as type species *Diopsis aethiopica* (Rondani), a common Afrotropical species. *Diasemopsis* is placed in the Afrotropical *Diasemopsis* genus-group together with *Chaetodiopsis* Séguy, *Trichodiopsis* Séguy and *Cobiopsis* Feijen. This genus-group forms one of the four clades in the stalk-eyed Diopsidae, together with Sphyracephalinae, the *Teleopsis* Rondani genus-group and *Diopsis* Linnaeus. The *Diasemopsis* genus-group forms a monophyletic clade characterised by autapomorphic reduction of spermathecae from 3 to 2. Other differential characters are 2-articled arista, abdominal syntergite with 3 or 4 tergites, fore femur with 2 rows of spinous setae and 2 rows of tubercles, apical seta and abdomen with pattern of pruinescence. Based on molecular analysis, *Chaetodiopsis* and *Trichodiopsis* are embedded in *Diasemopsis*. However, we do not (yet) follow the conclusion of placing these genera in synonymy with *Diasemopsis*. At present, *Diasemopsis* forms a rather inaccessible genus that requires subdivision in species-groups and maybe genera. Afterwards, the position of *Chaetodiopsis* and *Trichodiopsis* can be evaluated. A formal diagnosis of *Diasemopsis* does not yet exist. The shape of slender phallapodeme, ornamented spermathecae and non-articulated surstyli will form major elements for a diagnosis. The genus counts 46 valid species. Taxonomically, it is a difficult group. Séguy described it as a labyrinth. The last species was described 45 years ago while we know of around 100 undescribed species. Several dominant *Diasemopsis* species are distributed throughout the Afrotropical Region, but other species have an extremely restricted range. To make *Diasemopsis* accessible, our first aim is to split it up into species-groups. The foremost groups are listed and illustrated. The work on one of these groups, the *D. silvatica* Eggers group, will be highlighted. Criteria used will be discussed and illustrated: genitalia, photographs of dorsal abdomens and especially allometric aspects (sexual dimorphism of eye span and various morphological ratios).

A review of fossil Diopsidae with discussion of a new highly dimorphic amber species

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Keywords: amber, Eocene, *Prospyracephala*, Miocene, Oligocene, systematics

The first fossil diopsids were described from late Eocene Baltic amber as *Sphyracephala succini* Loew and *S. breviata* Meunier. Helm claimed in 1896 to own two long-stalked *Diopsis* in Baltic Amber. Hennig studied the holotype of *S. breviata* and provided arguments that these amber diopsids could not have been an ancestor of extant diopsids and therefore erected *Prospyracephala*. Hennig saw no reasons why more than one diopsid occurred in Baltic amber and proposed *P. breviata* as junior synonym of *P. succini*. The type of *P. succini* appears lost. It will now be argued that *P. succini* and *P. breviata* are not conspecific. The brief diagnosis of *Prospyracephala* needs to be revised. The characters on which Hennig assumed a sister group relationship between *Prospyracephala* and Diopsinae were correctly doubted by later authors. However, new data on *Prospyracephala* egg morphology supports Hennig's view. Oligocene compression diopsids were discovered in Montana and France. New specimens were also found in Miocene Saxon amber and Eocene Ukrainian amber. Kotrba made a morphometric comparison with 28 amber specimens of *Prospyracephala*. One specimen with large eye span distinctly fell outside allometric lines for eye span on body length. This fly was described as *Prospyracephala kerneggeri*. Kotrba suggested the presence of more cryptic species in the *P. succini* group. Now, an amber specimen is presented which is the only diopsid fossil with eye span longer than body length. Its remarkable characters will be discussed. Erection of a new genus is considered. Stalk-eyed Diopsidae count the subfamilies Sphyracephalinae and Diopsinae. Hennig did not place *Prospyracephala* as ancestor of a subfamily. Instead, he placed it in an Ancestral Group though noting that *Prospyracephala* has the general appearance of Sphyracephalinae. Likewise, the new genus can be placed in the Ancestral Group, but probably more towards the ancestral line of Diopsinae.

Hidden treasures: unraveling the diversity, natural history, and evolution of jackal flies (Diptera, Milichiidae)

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Keywords: Carnoidea, evolutionary adaptations, kleptoparasitism, milichiids, taxonomy

Among kleptoparasites, jackal flies (Milichiidae, Diptera) are a diverse group of insects that greatly illustrate the plethora of biological associations and evolutionary adaptations involved in kleptoparasitism. The family comprises approximately 400 extant and ten fossil species described in 20 genera, currently classified into three subfamilies: Madizinae, Milichiinae, and Phyllomyzinae. Recent studies demonstrate that we still have a long way to go in advancing the taxonomic, ecological, and evolutionary knowledge of these flies. It is hypothesized, for example, that the number of new species to be described may move the group diversity to over 1,000 in total. In this presentation, we will unravel the advances, the ongoing projects, and the knowledge gaps we still need to fill in the diversity, evolution, and natural history of jackal flies.

Towards a curated and optimized UCE probe set for investigating the evolutionary history of flies (Diptera, Insecta)

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Keywords: bioinformatics, genomics, phylogenomics, probe design, ultraconserved elements

Challenges remain regarding how best to design and curate UCEs probe sets for highly diverse organisms such as insects, and there is considerable debate on the performance of a universal probe set for the order versus a family-specific one. Although tailored probe sets might improve locus recovery, designing family-specific probe sets might also diminish the potential for combinability for future research. A good example is Diptera. In the last five years, at least three new family-specific probe sets for flies have been designed, which are not compatible with the universal probe set. Adopting the opposite trend, Hymenoptera researchers improved the original universal probe set for the group, including subsets for inclusive taxa and a set of legacy phylogenetic markers. We follow the aforementioned strategy to design an updated universal UCE probe set for flies. We aim to (i) improve the number of target loci; (ii) design subsets from the universal probe set; and (iii) include legacy phylogenetic markers and UCE loci from previous studies. We selected 19 fly genomes, representing lineages across the Diptera tree of life (e.g., Cecidomyiidae, Psychodidae, Tipulidae, Culicidae, Sciaridae, Stratiomyidae, Asilidae, Dolichopodidae, Phoridae, Syrphidae, Chloropidae, Sciomyzidae, Clusiidae, Tephritidae, Diopsidae, Ephydridae, Glossinidae, Muscidae, Calliphoridae). We used *Hermetia illucens* Linnaeus as the base genome, and the UCEs were identified by aligning the selected genomes to the base one. Currently, we are working in validating the identified conserved locus. Our next step is to perform in silico tests with this bait set to assess its efficiency across Diptera, and then sub setting the master probe list. This project not only improves an important tool to investigate the evolutionary history of flies, but also will address new analytical approaches and open new research avenues on the design and identification of UCE loci.

Variation in pollinating insect sampling associated with Malaise traps in marginal agricultural environments in Southern Ontario, Canada

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Keywords: Anthomyiidae, Apoidea, field margin, flight intercept trap, flower flies, hover flies, Hymenoptera, Syrphidae

Malaise traps have been gaining traction as an efficient tool to survey a variety of flying insects in various ecological studies. These traps have long been used in biodiversity surveys to study the presence of species in an area but are now becoming more commonly employed in agroecosystems to obtain estimates of pollinator abundance for pollination biology studies. While they are effective at capturing a large diversity of Hymenoptera and Diptera, the amount of variation associated with the proportions of insects caught in a single Malaise trap sample is unknown. The present study examines variance in individual Malaise trap capture of pollinators in crop fields in Southern Ontario. Five Malaise traps will be deployed 50 m apart along a single field margin at three sites with collection occurring weekly over a two-month period. Bees, flower flies, and Calyptrate Diptera will be sorted, identified, and counted for each sample. Statistical analyses will compare the total variance among the five traps at each of the three sites, the total variance at each site per week, and comparison of variance among different locations to assess for uniformity. Variance will be compared using the total biomass of each trap collection, the amount of each study group per collection event, and the diversity of the target groups between traps. The results will inform decisions regarding the number of traps required to capture a reliable estimate of the pollinating insect population present within a target sample area. By establishing an optimal methodology, future pollination studies across the agricultural sector that utilize Malaise traps will have increased confidence in observed results.

The North American Dipterists Society – where do we go from here? The Dipterists Society!

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Keywords: society, dipterists

During the 1987 annual ESA meeting, a group of dipterists agreed on the need to form a society. In connection with ICE 18, a dipterists field-day and informal meeting were held, during which an informal society for North American dipterists was born, including a newsletter, the Fly Times, to connect the community. The first field meeting of this group, now named the North American Dipterists Society, was held in Florida in 1989 and continues every two years since. Along with meetings, the Society developed a NA dipterists directory and a web presence early on. Fast forward to 2018 – a group of California dipterists bid to host ICDX. It was quickly apparent that organizational support is critical to run such a meeting in the US, so Gaimari (Fly Times editor since 2008) suggested to formalize the society, and did so in 2019, making it a US non-profit, an effort facilitated by its long history as an informal society. The international audience for Fly Times had grown steadily over the years, so our efforts focused on dipterists worldwide: developing a new internationally relevant website, directory of world dipterists (now 34 countries represented) and listserver (now with >1000 members from >80 countries). Membership has grown gradually, with a successful “Founding Member” program which will continue through 2024. The core mission is to support and further dipterology, with the keystones being publications, a grants program (nine students from six countries were supported to attend ICDX), and the constantly updated website <https://dipterists.org>. Donations and memberships fund all Society activities. We envision the society continuing to grow internationally, to soon be The Dipterists Society. As one of the major orders of animals on the planet, we want our robust and vibrant community to move forward cooperatively – a movement we want you to be part of!

Diversification of desert crane flies in western North America (*Tipula* Linnaeus, Tipulidae)

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Keywords: crane flies, desert, western North America

The long-palped crane flies (Tipulidae) are notable as one of the insect families to have successfully adapted to both aquatic and terrestrial environments. Most striking among these adaptations has been the broadscale evolution of many lineages of tipulids, particularly among the enormous genus *Tipula*, into desert and semi-desert landscapes of western North America, including (from south to north latitude) the Sonoran, Chihuahuan, Mohave, Colorado Plateau and Great Basin Deserts. Survival strategies of these desert groups have included diurnal inactivity of adults, larval development using shrub leaf litter, and an obligate egg diapause, which may extend several years during drought periods. The family Tipulidae, particularly the genus *Tipula*, is an ideal group for examining the historical biogeography of the desert regions and surrounding mountains of North America, for at least 10 separate lineages are found in arid and semi-arid areas and tend to have a high proportion of endemic species. This multiplicity of separate lineages inhabiting common regions and sharing common general attributes provides many independent “experiments” in which to examine the diversification of American deserts. Most of these groups fall within the *Lunatipula-Vestiplex* series of subgenera of *Tipula*, which the morphological analysis by Gelhaus (2005) supported as forming a natural evolutionary grouping. What is needed presently are species-level revisions of these desert groups. We are revising three species groups of *Tipula* (*Lunatipula* Edwards), the *splendens* Doane, *tergata* Doane and *unicincta* Doane species groups (encompassing 45 species), with the distributions overlapping those of *Tipula* (*Eremotipula*) (revised in 2005) and eventually providing important biogeographical comparisons. The species in these *T.* (*Lunatipula*) species-groups are poorly known and characterized, and our work examining the important male and female features for identification and subsequent phylogenetic analysis will be discussed.

Diptera of Haida Gwaii, British Columbia, Canada

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Keywords: biodiversity, biogeoclimatic zones, collections, collaboration, community comparison, First Nations, surveys

The archipelago of islands off the west coast of Canada, known as Haida Gwaii (“Islands of the People”), is a richly biodiverse region separate from the rest of the Nearctic. From sea level to over 1100m elevation, this group of hundreds of islands includes five distinct biogeoclimatic zones and dozens of microhabitat types. Surveys of insects have been conducted on the islands for the past century or more, including a 2022 collaborative trip. The results of some of these surveys, deposited in the Royal BC Museum collection, are presented here. From over 3600 specimens, 415 morphospecies in 60 Diptera families have been identified. Analysis indicates differences in community composition between biogeoclimatic zones. Future specialist attention will reveal complete species identifications and new species descriptions. More survey work also needs to be done as many habitat types remain undersurveyed. Future survey work must be conducted in collaboration with the Haida Nation. Also, much Haida Gwaii Diptera specimen data currently remains unknown and inaccessible in global entomology collections.

Systematics and natural history within the Sciomyzoidea – further questions for an uncontentious answer

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Keywords: classification, ecology, morphology, Nearctic, phylogeny, shoreline

In 1989, McAlpine described the Sciomyzoidea as “one of the least contentious of all the superfamilies of the Acalyptratae.” In the past seventy years, the only major change to the original grouping of six families (Helcomyzidae, Ropalomeridae, Sciomyzidae, Coelopidae, Dryomyzidae, Sepsidae) has been the addition of the Conopidae and the elevation of a few lineages to family status (Helosciomyzidae, Heterocheilidae, Huttoninidae, Phaeomyiidae, Natalimyzidae). Molecular methods have supported the monophyly of the superfamily. But what of the internal classification? Is the current family-level classification of the clade reflective of evolutionary history? None of the extant sciomyzoid families are globally speciose. Only Conopidae and Sciomyzidae have been classified into subfamilies. The presence of endoparasitism and marine shoreline adaptation within various genera suggest possible ancestral relationships. More questions than answers remain within this “uncontentious” taxon.

Rare Calypttratae flies (Muscidae and Sarcophagidae) preserved in fossil resin from *Hymenaea* spp.

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Keywords: amber, copal, Defaunation resin, Miocene, taphonomy

Fossil assemblages provide diverse information about ancient environments, but the record is incomplete and biased. Actuo-taphonomic studies are essential for understanding these gaps and interpreting what occurred in the past, as well as for reconstructing ancient ecosystems and relationships between organisms. The type and abundance of organisms preserved in resin depend on various taphonomic and ecological factors. In this communication, we present two rare flies preserved in amber and copal originating from the resin-producing tree *Hymenaea* and compare them with modern specimens collected in Defaunation resin and with sticky traps in *Hymenaea*. The first specimen, a Sarcophagidae, is preserved in a piece of Mexican amber from Simojovel, Chiapas (Mexico), dated to 23 million years old (Ma) and belonging to the Aquitanian age (Miocene). The second specimen, a Muscidae, is preserved in a piece of Holocene copal from Tanzania, with the resin produced between the years 772 and 885 AD, according to 14C analysis. Both families, Sarcophagidae and Muscidae, can be coprophagous, predators, parasitic, kleptoparasitic, necrophagous, or simply feed on nectar, fruit juice, and proteins from decomposing matter such as excrements and carrion. They are abundant in the forest in the proper habitats but do not frequently visit a tree, making their capture in resin extremely rare. Beside its poor presence on the trees, in our actuo-taphonomic studies, we collected ten specimens in the sticky traps and two specimens in the resin. This can be explained by the fact that Sarcophagidae and Muscidae are relatively young families, now abundant in the forest but with the most recent Calypttratae ancestor dating back to the Cretaceous period, around 64 Ma. The age of Sarcophagidae is estimated to be at least 23 Ma.

***Philopota* Wiedemann (Acroceridae) as an effective pollinator
and observation of Diptera visitors on two species of
Maranta L. (Marantaceae) in the atlantic forest of
Serra do Mendanha Natural Park, Rio de Janeiro**

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Keywords: Atlantic Forest, Marantaceae, *Stylogaster*

A less “relevant” aspect of the Diptera in the common sense is their anthophily, with many anthophilous taxa considered important pollinators, with some authors considering their importance only less than that of bees. Flowers pollinated by Diptera are usually small, white in color, pleasant or unpleasant odor, with a short floral tube, providing nectar or pollen resources. This study was made at Serra do Mendanha Natural Park, Rio de Janeiro, Brazil, from 2014 to 2023. Two species of Marantaceae were the focus of observation: *Maranta cristata* Ness & Mart. and *Maranta leuconeura* E. Morren. Observation hours were from 7 AM to 5 PM. *M. cristata* and *M. leuconeura* are rhizomatous herbs with erect inflorescences of which two to three flowers are in anthesis per day. Flowers of *M. cristata* and *M. leuconeura* can be visited by different groups of insects: bees, wasps, butterflies, ants, beetles, bugs and flies, but effective pollination is made by two genera of bees (*Paratetrapedia* Moure and *Plebeia* Schwarz) and by flies of the genus *Philopota* Wiedemann (Acroceridae). Visits happen in the morning, peaking at 10 AM. The flies land on the staminodes and insert their mouthparts into the floral tube to collect nectar, being able to fire the trigger, and may also visit flowers in which the stylet has already been triggered. Visiting time in each flower can reach 20 minutes. Visits can occur on the same inflorescence or on other inflorescences on the same individual and from close individuals. Up to three individuals of *Philopota* sp. were observed on the same plant. Two other species of Diptera were observed visiting the flowers, but they are not able to do effective pollination (*Stylogaster* Macquart [Conopidae] and an unidentified Stratiomyidae). This is the first observation of these taxa visiting flowers of Marantaceae.

**A comparison of the mycetophilid fauna of two aspen islands
in the Bloody Run Mountains of northern Nevada**

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Keywords: aspen islands, Bloody Run Mountains, fauna, Nevada

In 2022 two aspen forest islands in the Bloody Run Mountains of northern Nevada were surveyed for Mycetophilidae using Malaise traps and other methods. The forests were a few acres each, surrounded by miles of sagebrush desert, both centered around small springs, less than a mile apart, separated by a ridge. Despite similarities in the two forest habitats there were significant differences between the two in their respective mycetophilid faunas. There were also similarities in the mycetophilids collected. There were species reared out of mushrooms and leaf litter in both forests that were not seen in the Malaise traps.

Plenary address

How the fossil record of flies informs about distributions, extinctions, and diversity

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Keywords: amber, distribution, Eocene, fossil, geobiotic, Miocene, paleobiogeography, tropical forests

A brief summary of the chronology of major lineages of Diptera is presented, beginning with their earliest fossils in the Triassic, alongside geobiotic events in earth history. Among the unique evolutionary insights provided by fossils is their most common use for the calibration of divergence-time models. Pitfalls in methods and interpretation of these models are briefly discussed. Though used less for paleo(bio)geography, fossils have substantial implications for understanding earth history, modern distributions, and diversity. Examples are discussed of current research on Diptera and other insects in amber from the Early Eocene of western India and mid-Miocene of the Dominican Republic. Fossils from India address geophysical hypotheses of the tectonic collision of India with southern Asia in the Paleogene; these are also the earliest evidence from Asia of broad-leaved (e.g., dipterocarp) tropical forests. Flies and other insects in Miocene Dominican amber inform on how a fauna from a continental island evolves, and on Neotropical faunal stasis and extinctions. Overall, the gathering data indicates that tropical forests are significantly more ancient than north temperate forests. Late Pleistocene and Holocene fossils reveal the succession of ecosystems, sweeping changes in the distributions of living species, and modern extinctions.

Conflict and congruence between data from adult flies, immature stages, and molecular markers in the systematics of Calyptratae

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Keywords: systematics, taxonomy

Thanks to numerous past and recent studies, our understanding of phylogenetic relationships within Calyptratae is improving. However, several hypotheses still raise questions, mainly due to incongruence between molecular phylogenies and classifications based on adult morphology. To address this uncertainty, it may prove beneficial to utilize data from various sources, including different life-history stages. So, we present some recent studies where morphology of immature stages helped to resolve conflicting hypotheses, resulting in a more coherent understanding of the evolutionary history of calyptrate flies, rather than maintaining different classifications for different data sources. In muscids, both morphology of immature stages and molecular data reject the classification of *Aethiopomyia* Malloch, *Alluaudinella* Giglio-Tos, and *Ochromusca* Malloch within Phaoniinae, with new data supporting a transfer to Reinwardtiinae. Additionally, details of larval morphology confirmed the close relationship between *Achanthiptera* Rondani and *Potamia* Robineau-Desvoidy and their position within Azeliinae. The structure of the third instar cephaloskeleton tentatively supports the monophyly of a clade composed of Coenosiinae, Mydaeinae, and Phaoniinae. In flesh flies, both the morphology of first instar larvae and molecular data reject the classification of *Chauliooestrus* Villeneuve, *Chivamyia* Pape, *Eumacronychia* Townsend, *Goniophyto* Townsend, *Sarcotachina* Portschinsky, and *Xiphidiella* Zumpt within Eumacronychiinae proposed in 1998 by Yu.G. Verves. Instead, these genera should be classified partly within Miltogramminae, partly within Paramacronychiinae. Furthermore, the position of *Sphecapatoclea* Villeneuve, *Sphecapatodes* Villeneuve, and *Metopodia* Brauer & Bergenstamm within Phyllostelini, as proposed based on adult morphology, conflicts with results from first instar larval morphology and molecular data. These revealed that *Sphecapatodes* and *Metopodia* are closely related to *Taxigramma*, all within the "higher Miltogramminae", while *Sphecapatoclea* remain in "lower Miltogramminae".

Update of the Swiss Diptera checklist

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Keywords: checklist, Switzerland

The Swiss Diptera community met in 1998 to produce the first Diptera checklist in Switzerland. A couple of decades later, it was time to update this list by integrating all the new knowledge accumulated over the years. More than 50 experts, far beyond the borders of this small country, had to be contacted, collections were revisited, fieldwork data was closely examined. More than a thousand species could be added to the new checklist and countless changes and adaptations were made, which led to a better understanding of some groups and helped identify those that still require attention. This presentation will take you through the challenges of creating this new checklist and how we interpreted the results, including the knowledge gaps. Despite these gaps – which we hope will continue to narrow over time – this work is an important step towards a better understanding of the order Diptera, which, although numerous, is generally overlooked. It also allows the development of new tools, particularly for identification, ecology and ultimately to promote Diptera conservation at all levels, from the general public to the expert level. We hope that our approach can inspire others to embark in similar endeavors.

Forensic entomology case study: blow flies associated with dismembered and concealed remains

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Keywords: blow flies, Calliphoridae, forensic entomology, postmortem interval, time of colonization

To destroy or dispose of potential evidence, perpetrators use various methods to conceal remains, which may be problematic for forensic investigations. In forensic entomology, insect evidence can provide information on the minimum postmortem interval (mPMI) if insects arrive and colonize the remains quickly. Concealment of remains may delay insect arrival and alter the abundance of insect evidence available during the investigation. The dismemberment of remains, although rare, poses additional challenges in forensic investigations. In this case, the perpetrator confessed to the homicide and dismemberment, and willingly provided authorities with a timeline. The perpetrator provided a complicated account of the events that had transpired, which resulted in the dismemberment and concealment of the remains. The remains were dismembered with a chainsaw and concealed in large plastic garbage bags. Insect evidence was collected during the autopsy, and forensic entomology was able to provide a minimum time of colonization (TOC), that aided investigators in establishing a timeline. Insect evidence included third instar blow flies of a common species, *Lucilia sericata* Meigen (Diptera: Calliphoridae). Forensic pathology and forensic entomology were the two fields that yielded the most valuable information in this investigation. The information provided in this case study serves as an example of a complex investigation and further supports the application of insect evidence in forensic investigations.

**Managing *Megaselia* Rondani (Phoridae):
multifaceted solutions to one of the world's largest genera**

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Keywords: dark taxa, hyperdiverse

The taxonomic problems faced in hyperdiverse groups are not merely larger versions of the problems encountered in other taxa. Rather, the scale and complexity of these groups, combined with our ignorance of them, necessitates new and innovative solutions. How can we identify, organise, and classify groups with thousands of unknown species, countless numbers of individuals, challenging morphology coupled with very small size, scant life history data, troublesome trees, and species curves that refuse to plateau no matter how much sampling you do? This talk will focus on the arsenal of approaches that have been applied to mega-genus *Megaselia* in recent years, summarising an (ever evolving) approach to a dark taxon. Numerous efforts are contributing data to the comprehensive understand of this genus, ranging from systematic and integrative approaches to species discovery, delimitation and description, spatial and temporal analyses, ecological modeling, experimental taxon sampling, microbiome analysis, morphometrics, convolutional neural networks and robotics, and advanced morphological scans. All of these pieces contribute to a better understanding of one of the largest genera of animals. What is next and how do we bring all of this information together?

Mimicry in Stratiomyidae

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Keywords: Hymenoptera, Sarcophagidae, spider

Mimicry is widespread among insect groups, and well-known from other similar-sized Diptera like Syrphidae. Stratiomyidae display a remarkably wide range of size, shape, and coloration, for such a medium size family. A lot of striking colors and patterns are just imperfect mimics of Aculeata wasps and bees, and they just mimic the aposematic patterns of these defensive insects. But there is a number of species that display high fidelity mimicry. These soldier flies mimic Sarcophagidae, leafcutter bees (*Megachile chrysorrhoea*), mining bees (*Andrena*), Sphecidae, Formicidae, Pompilidae, Vespidae (*Agelaia*, *Brachygastra*, *Paralastor*, *Polybia*), and Braconidae. There are some indications that several genera are mimicking beetles, and *Boreoides*, with flightless females, might mimic spiders. Metallic blue species with contrasting yellow heads of the genus *Cyphomyia* are yet without explanation, especially because several unrelated Stratiomyidae genera have species imitating this color pattern. Even males (usually without yellow heads) show several independent forms which are mimicking the yellow heads of the females. There is no known model for this color pattern, although it can be found in bees (*Xylocopa*) and Scoliidae wasps, which occur outside the range of *Cyphomyia*. And it is found in fulgorid leafhoppers, Bombyliidae and Calliphoridae, but is in general rare.

Updates to the Afrotropical Stratiomyidae fauna

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Keywords: *Porpocera*, Prosopochrysiini, iNaturalist

In preparation of the Stratiomyidae chapter in the second volume of the “Manual of Afrotropical Diptera” (2017), we examined numerous types, checked the holdings of many museums, collected ourselves in Africa and received a lot of material from private and institutional collectors. Despite these extensive preparations, we found several new taxa in the years past the publication. This shows that we are still in the beginning of our understanding of the African Stratiomyidae fauna. Many undescribed species were already known to us for a long time, but because the Manual focused on the generic level, we are not mentioning new species. The rapidly increasing popularity of websites like iNaturalist provided another source of data. On this website we found an undescribed species of *Porpocera* in Ghana, while all known specimens are restricted to southern Africa. An unusual discovery on social media sites was the occurrence of an Afrotropical genus, *Sternobrithes*, outside Africa. This small species of Pachygastrinae spread, no doubt via human activity, into Europe, Asia, North America and the Pacific. While revising the generic limits of the Sarginae, it became clear that there is the need of several new genera in Africa. The taxonomic difficult subfamily Pachygastrinae is in need of a thorough revision, and while some genera (mainly monotypic) will likely be synonymized, there are some spectacular new genera to be described. An unsuspected new genus in the Nematelinae was found in the KwaZulu-Natal Museum collection. An undescribed genus of the little understood tribe Prosopochrysiini (subfamily Stratiomyinae) from Uganda was discovered, which is remarkable because there are only two monotypic genera known from mainland Africa, and all three genera are rather different in appearance.

Reassessment and discoveries of phorids from Cretaceous to present bring a fresh perspective to evolution of wing venation

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Keywords: amber, Cretaceous, evolution, homology, palaeodiversity, wing venation

The scuttle flies (Phoridae) have undergone an important radiation since the Cretaceous and have continued to evolve ever since. Currently, phorids are considered one of the most diverse families among the Diptera. The discovery of numerous specimens of phorids preserved in amber, copal and Defaunation resin ranging from the Cretaceous to the Holocene, has the potential to greatly enrich our knowledge of fossil biodiversity. However, only a small fraction of these specimens has been studied so far, accounting for less than a quarter of the known specimens. Fossil resins are particularly favourable for preserving morphological characters, allowing for exceptional details to be observed in specimens from different geological ages. Increasing the available data on phorid fossils is crucial for understanding the evolution of morphological characters, specifically how wing venation has changed through time. An initial re-evaluation of paleontological data, combined with the study of new specimens from various deposits, has revealed issues with morphological characters used in species descriptions. One such issue is the primary homology of the venation structures, which depends on the chosen wing nomenclature used by different authors. This issue poses challenges to conducting phylogenetic analyses of extant and extinct taxa. Therefore, our focus shifted toward investigating the evolution of wing veins in both extinct and extant specimens. Preliminary results indicate evolutionary patterns in the radial and medial veins. Specimens from the Cretaceous period exhibit wing venation with low variability and with abundant number of veins on the wings. In contrast, Cenozoic and modern taxa demonstrate a wider disparity in venation, with varying degrees of reduced wing venations. Furthermore, minimal changes are observed between the Cenozoic and modern taxa. Wing venation studies will contribute to the understanding of the evolution of the paleo-diversity through time, shed light on potential evolutionary pressures, and elucidate relationships with related outgroups.

Trapping techniques for *Stomoxys calcitrans* (L.) (Muscidae)

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Keywords: attract and kill, electric fence, Knight Stick trap, Vavoua trap

Stomoxys calcitrans (L.) has routinely been attracted to traps made of materials reflecting sun light in attractive wavelengths or combinations of attractive wavelengths. The first attractive material discovered in the US was Alsynite fiberglass. Flat panels were formed into a cross configuration and coated with an adhesive to trap the attracted flies. A cylindrical version was covered with adhesive-coated clear sticky sleeves and was more wind resistant. In 2002 a trap called the Knight Stick (KS) trap was introduced. This trap was a cylinder of plastic pipe covered with an adhesive-coated sleeve made of thin packing foam. The KS trap significantly outperformed the fiberglass traps. Placement of traps around animals was difficult because traps had to be where the animals could not contact them and destroy them. This problem was solved by placing traps inside the compounds with the animals and protecting the traps with electric fence. Traps inside of animal compounds captured significantly more stable flies than traps placed outside of the perimeter fence. The KS traps were usually serviced weekly as the sticky sleeves became covered with flies and were no longer attractive. The KS traps were placed inside of cylinders of pesticide-treated mesh that left a space of 2.5-cm between the mesh and the trap surface. An array of openings was made in the mesh to minimize any loss of attraction caused by the mesh and to allow an entryway for the attracted flies to try to reach the trap. Openings were large enough for the flies to pass through, but they could not fly through. In semi-field tests, the mesh killed more than 95 % of the flies before they could reach the trap surface. This greatly extended the life of the sticky wraps and the need to service the traps.

New data on *Basilia handleyi* Guimarães (Nycteribiidae) after 57 years

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Keywords: bat flies, Hippoboscoidea, new occurrence, taxonomy

Nycteribiidae is represented by hematophagous dipterans that are exclusively ectoparasitic on bats. *Basilia* is the only cosmopolitan genus of Nycteribiidae, but species that occur on the American continent are endemic. *Basilia* species are primarily defined by the morphological characteristics of females, particularly characteristics of tergites and sternites. The external morphology of males is quite homogeneous, and their identification is primarily based on the genitalia. The female of *B. handleyi* was described in 1966, from the bat *Lasiurus castaneus* Handley, collected in 1963 in San Blas, Panama. Since that time there have been no additional specimens collected. In 2012, in Puntarenas, Costa Rica, individuals *B. handleyi* were collected from *L. castaneus*. Among the collected individuals, two male individuals were found. These males have an external morphology similar to other *Basilia* species, however, the genitalia have three setae near the posterior margin of the phallobase, a feature unique to species of American *Basilia*. The other species of *Basilia* that presents a similar characteristic is *B. kerivoulae* Theodor, with distribution in South Africa and presents several morphological characteristics distinct from *B. handleyi*. The present summary indicates new distribution data for *B. handleyi*, about 660 kilometers (440 miles) from the type locality. Currently, *B. handleyi* represents the seventh species of the genus with distribution in Costa Rica.

Tephritoidea of economic importance

Taxonomic revision of subfamily Dacinae (Diptera: Tephritidae) from Taiwan based on morphological characters

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Keywords: genitalia, morphology, Taiwan, taxonomy

Family Tephritidae (the true fruit flies) is among Diptera's most prominent families. While larvae of most species developed in plant tissues, it is also one of the most economically important families. Subfamily Dacinae is an ecologically diverse group, with species predominantly native to South-East Asia. Most of the dacine species recorded from Taiwan belong to two major tribes, Dacini and Gastrozonini. Tribe Dacini is frugivorous, causing damage to the host plant through ovipositor punctures and larval growth. While non-pest species are associated with a narrow range of known hosts, particular pest species are polyphagous with a broad host range. Severe infestation on commercially produced fruits can lead to fruit rot or premature drop, leading to considerable crop loss. Thus taxonomy of Dacini may pave the way to improve pest management and quarantine. However, a current taxonomic system of Dacini relies on the color pattern as the primary diagnostic character for species delimitation, regardless of its high intraspecific variation. Tribe Gastrozonini is Poaceae-breeding. Taxonomic study of the group is relatively scarce and limited compared to other tephritids. This study aimed to revise the Dacinae species of Taiwan and search for a more stable set of traits for their taxonomic system. The checklist of Taiwanese Dacinae has been updated, with 32 Dacini species and 14 Gastrozonini species. The once-doubted record of *Zeugodacus yayeyamana* (Matsumura, 1916) from Taiwan has been verified. The male terminalia of 13 Dacini species and 4 Gastrozonini species has been studied, of which 14 species were described for the first time.

From a mosquito's (Culicidae) perspective – what happens when there are co-circulating viruses out there?

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Keywords: *Aedes aegypti*, arbovirus, co-infections, *Culex pipiens*, Mayaro virus, West Nile virus, Zika virus

Much of the entomological research into mosquito-borne viruses concentrates on surveillance to inform mosquito control efforts, to direct public health messaging and for building outbreak models. My lab has spent the past 20 years doing this type of research for West Nile virus in Ontario, Canada. Through our surveillance efforts we have been able to track when “new” species first appeared and then became established in Ontario. In 2016, *Aedes albopictus* (Skuse) and *Aedes aegypti* (Linnaeus) were first reared from larvae collected in Windsor, Ontario. The potential for exotic viruses to appear over time, influenced our decision to consider what would happen if mosquitoes were infected by two arboviruses simultaneously. We started by looking at co-infections and superinfections in cell culture (e.g., Vero cells and C6/36 cells, mammalian and mosquito cell lines, respectively) for West Nile virus and Zika virus (both *Flaviviridae*) and for Mayaro virus and Zika virus (*Alphaviridae* and *Flaviviridae*, respectively). We have since co-infected adult mosquitoes (*Culex pipiens* Linnaeus and *Ae. aegypti*) to determine infection rates, dissemination rates and transmission rates of co-infecting viruses. Data are often conflicting. For example, our cell culture assays have shown that Mayaro virus outcompetes Zika virus. However, in *Ae. aegypti* mosquitoes (fed on blood containing equal amounts of Mayaro and Zika and tested on day 10 post blood-meal), 7 of 9 mosquitoes were negative for both viruses, whereas 2 of 9 were positive for both. Data will be presented for our other co-infection studies, including preliminary work on Insect Specific Viruses (ISVs) and arboviruses. Discrepancies between cell culture assays and live mosquito assays will be discussed. The effects of co-infections on mosquitoes warrants further study.

Emergence patterns and community changes of aquatic dance flies (Diptera: Empididae: Clinocerinae, Hemerodromiinae) during 15 years

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Keywords: community changes, diversity, emergence, prey abundance, water discharge

Aquatic dance flies (Empididae: Clinocerinae and Hemerodromiinae) as an ecological group are important part of freshwater ecosystems, especially running waters. They are predators as larvae and adults and important for understanding aquatic food webs. The goals of this study were to analyze their preference for various microhabitats, the main ecological factors that affect the composition and abundance of specimens, and emergence of species at different freshwater karst habitats during a 15-year period. In addition, interactions between aquatic dance fly and prey abundances were explored. The study was performed at three sites in the National park Plitvice Lakes, which is situated in a Dinaric karst region of Croatia. Adult specimens were collected using a semi-quantitative method with six emergence traps placed at each site. Specimens were collected monthly from March 2007 until December 2021. In total 19 species were identified and in the year 2015 the highest number of species and diversity indexes was recorded. The dominant genus is *Chelifera*, while the most abundant species is *Hemerodromia unilineata* Zetterstedt. Except for the bivoltine *Chelifera precabunda* Collin, *Chelifera pyrenaica* Vaillant and *Chelifera stigmatica* (Schiner) all other species were univoltine. The composition of the species change during the years where some species disappeared from the community while others appeared. The highest emergence rates were recorded above microhabitats with highest prey densities (Chironomidae) and with highest current velocity. Water temperature and photoperiod are the main ecological factors determining the emergence. Water discharge seems to have the greatest effect on dance fly abundance patterns and community composition.

Taxonomic notes on tribe Gastrozonini (Diptera: Tephritidae: Dacinae) with a record of two new species from India

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Keywords: *Acroceratitis*, bamboo, *Taeniostola*

True fruit flies (Diptera:Tephritidae) are one of the most diverse groups of Acalyprtratae with a wide array of feeding habits. Fruit flies of tribe Gastrozonini, commonly known as bamboo-shoot fruit flies in subfamily Dacinae are specialized/ adapted to feed on shoots of grasses, bamboo (Fa. Poaceae). Gastrozonines are primarily tropical in distribution with 137 species in 28 genera in the world. Of these, 38 species belonging to 16 genera are recorded from India. 20 species in 11 genera, namely *Acroceratitis*, *Acrotaeniostola*, *Anoplomus*, *Chaetellipsis*, *Dietheria*, *Galbifascia*, *Gastrozona*, *Phaeospilodes*, *Taeniostola*, *Ichneumonopsis*, *Dacimita* and *Xanthorrhachis*, were collected and preserved as a part of explorative trips undertaken in India during the past decade. A new genus, *Dacimita* David & Hancock and four new species were described. Postabdominal structures of 21 species were studied and illustrated along with 3rd instar larval morphology of *Acroceratitis histrioinica* and *Gastrozona nigrifemur*. Two new species are recorded here, one resembling *A. striata* (Froggatt) in external morphology and wing pattern but can be differentiated from the former by the absence of scutellar spots, discal band of the wing not fused with costal band and an elongate oviscape longer than abdomen. The other is similar to *T. vittigera* Bezzi in scutal, abdominal colour and pattern but can be differentiated from the latter by the wing pattern with subapical band broadly joining apical spot, blunt aculeus tip with diverging sides, conical spicule with 5–7 spines and round/oval spermatheca. Examination of postabdominal structures of Gastrozonini revealed that the proctiger is larger than the epandrium, which can be considered a synapomorphy of the tribe Gastrozonini. All species of *Acroceratitis* examined have a tuft of fine hairs apically on the proctiger, which is a unique character of the genus.

Molecular phylogenetics of moth flies (Diptera: Psychodidae) using exon-capture sequencing

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Keywords: moth flies, Pericomaini, phylogeny, target DNA enrichment, tribal classification

The family Psychodidae, commonly known as moth flies, is a diverse group of small nematocerous flies with a worldwide distribution. Despite their importance in ecosystem functioning, and the medical importance of Phlebotominae, the phylogenetic relationships within this family remain poorly resolved. In recent years, several studies aiming to resolve the suprageneric classification of Psychodidae revealed that the current tribal classification comprises non-monophyletic groups; thus, the phylogenetic relationships between subfamilies and tribes remain unresolved. Using publicly available annotated genomes and newly generated transcriptomes, we created a bait-set with 18,651 baits targeting 1445 CDS regions. Our samples include 92 specimens of Psychodidae with members of four out of the six subfamilies currently recognized. We selected samples focusing on the tribe Pericomaini inside the subfamily Psychodinae. Moreover, we included representatives of the sister groups Culicomorpha and Tipulomorpha. We reconstructed a phylogenetic tree using different optimality criteria. The results provide a robust framework for understanding the evolutionary history of the family Psychodidae and highlight the need for further taxonomic and morphological studies to clarify the relationships within this group.

Review of the genus *Eugenys* Quate, 1996 (Diptera: Psychodidae) with the description of two new species from the Neotropical Region

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Keywords: moth flies, Pericomaini, Neotropical Region, tribal classification

We review the diagnosis of the genus *Eugenys* Quate, 1996 (Diptera: Psychodidae) which occurs in the Neotropical Region. Initially known from Costa Rica, Nicaragua, and Panama, we describe two species from Ecuador, bringing the total known species to five. This study provides detailed descriptions of the new species based on male and female specimens, along with the first DNA barcodes for the genus and the newly described species. We also provide an identification key for identifying male specimens of the genus worldwide. Finally, we discuss the morphological characteristics of *Eugenys* and compare the genus with several other taxa, tentatively suggesting a placement within Pericomaini *s.l.*

Automated mitogenome extraction from low-coverage genome skimming data facilitates integrative systematic revision of *Pollenia* Robineau-Desvoidy (Polleniidae) and a revised phylogenetic hypothesis for the Polleniidae

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Keywords: bioinformatics, Calypttratae, genome skimming, mitogenome, phylogenomics

The Polleniidae (Diptera) are a family of flies best known for species of the genus *Pollenia* Robineau-Desvoidy, which overwinter inside human dwellings. Previously divided across the Calliphoridae, Tachinidae and Rhinophoridae, the pollenid genera have only recently been united. Several studies have utilised molecular data to analyse polleniid phylogenetic relationships, although all suffered from low taxon sampling or insufficient phylogenetic signal in molecular markers. To alleviate these problems, we utilised two automated organellar genome extraction software, GetOrganelle and MitoFinder, to assemble mitogenomes from genome skimming data from 22 representatives of the pollenid genera: *Dexopollenia* Townsend, *Melanodexia* Williston, *Morinia* Robineau-Desvoidy, *Pollenia* Robineau-Desvoidy, and *Xanthotryxus* Aldrich. From these analyses, we provide 14 new mitogenomes for the Polleniidae and perform phylogenetic analyses of 13 protein-coding mitochondrial genes using both maximum likelihood and Bayesian inference. Subfamilial phylogenetic relationships within the Polleniidae are interrogated and *Pollenia* is found to form a monophyletic clade sister to *Melanodexia*, *Morinia* and *Dexopollenia*, providing no evidence for the synonymisation of any of these genera. Our topology conflicts with previous morphology-based cladistic interpretations, with the *amentaria*, *griseotomentosa*, *semicinerea* and *viatica* species-groups resolving as non-monophyletic. We provide support for our topology through analysis of adult morphology and male and female terminalia, while identifying new diagnostic characters for some of the clades of the *Pollenia*.

**When *Calliphora* are not *Calliphora*:
advances in Calliphoridae phylogenomics provide
unprecedented insights into Australian blow fly systematics**

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Keywords: anchored hybrid enrichment, Australasia, Calliphorinae, phylogenomics

The family Calliphoridae (blow flies) are a complex group of flies that include many well-defined subfamilies. However, as a result of very few strong unifying apomorphic character states, the family has been subject to several recent major changes in composition, such as the raising of the subfamily Polleniinae to family status as the Polleniidae. Furthermore, the position of and relationships between the Australian Calliphoridae and the blow fly fauna elsewhere on the globe have been widely debated over the past century, with species especially being interchanged between genera *Bellardia*, *Calliphora* and *Onesia*. Despite attempts at resolving the phylogeny of this group, to date no study has paired a broad representative taxon sample of Australian Calliphoridae with data from robust molecular markers capable of resolving complex relationships with strong support. Our application of an anchored hybrid enrichment approach, in combination with the most comprehensively sampled calliphorid phylogeny to date, has allowed us unprecedented insights into the systematics of the subfamilies Calliphorinae, Chrysomyinae and Luciliinae, and has revealed new information regarding the relationships between their constituent genera. Interestingly, there appears to be a strong divide within the Calliphorinae, between a mainly Australasian clade and a mainly Northern Hemisphere + Afrotropical clade, with the latter as the sister taxon to the Melanomyiinae. The Australasian clade has the genus *Bellardia* nested within a paraphyletic *Onesia*, and the *Bellardia*–*Onesia* clade is in turn nested within a paraphyletic *Calliphora*. This talk discusses the implications of our new phylogeny for the taxonomy and systematics of the Calliphorinae, with emphasis on the position of the Australian and New Zealand species. We also show how adult and larval morphology support our molecular results.

**More than meets the eye: a revision of the Nearctic species
of the genus *Amiota* Loew (Drosophilidae)**

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Keywords: asymmetry, chirality, fruit flies, lachryphagy, montane, saproxyly

Many gaps remain in our understanding of the diversity, distributions, and taxonomy of the North American biota. Well studied insects like the Drosophilidae (fruit flies) are no exception. Taxonomic investigations over 4 years revealed large species diversity in the obscure drosophilid genus *Amiota* Loew, which had previously been unknown. Montane areas represent the greatest areas of diversity including the Great Lakes, sky islands of Arizona and New Mexico, the Ozarks, and Appalachia. The interior highlands of Mexico, especially the Sierra Madre Occidental pine-oak forest, is found to contain very high species diversity. Morphologically diverse genitalia (asymmetry and chirality), feeding on tears and sweat, and requiring dead wood for their life cycle; there is still much to learn about this fascinating group of insects.

Overview and guide to Diptera trainings in sub-Saharan Africa

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Keywords: Afrotropical, agriculture, education, fruit flies, flower flies, hover flies, pest species, pollination

The Royal Museum for Central Africa, together with partner institutes in Africa and Europe, has organized and delivered nine training courses on Diptera taxonomy between 2009–2022 in Belgium, South Africa and Tanzania. This prompted us to compile a guide with all practical information and experience, and to present a best-way-forward in future organization of similar training courses. The practical guide has three sections: 1) a brief historical overview of the training courses, 2) a short evaluation of the profile (gender, age, educational and professional background) of the applicants and participants, and 3) comprehensive information on every aspect of the organization and delivery of the training courses. This will hopefully serve as a useful resource to stimulate and facilitate the organization of other entomological (or similar) training courses in the Afrotropical Region or elsewhere. Once published, all information will be freely available as Word or Excel files to facilitate future use by interested parties.

Overview of recent achievements in Afrotropical hover fly (Syrphidae) taxonomy and phylogeny

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Keywords: DNA barcoding, flower flies, morphology, Next Generation Sequencing

Over the last decade, there has been substantial progress on the taxonomy and systematics of Afrotropical Syrphidae, especially within the subfamily Eristalinae. Here, we present an overview of this progress and show how both morphological and molecular approaches have resulted in the detection of new species, improvements of identification keys, the phylogenetic placement of genera, and in resolving intergeneric phylogenetic relationships. We will illustrate how these data have stimulated ecological and biogeographical studies, and form the basis for country species checklist. We also present the first initiatives to include Afrotropical hover flies in conservation management plans in South Africa.

Taxonomic and ecological knowledge gaps in Afrotropical hover flies (Syrphidae)

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Keywords: DNA barcoding, flower flies, morphology

The network on taxonomists working on the Afrotropical Syrphidae (hover flies or flower flies) has expanded over the last decade. As a result, there has been substantial progress on the taxonomy and systematics of Afrotropical Syrphidae. Yet, many genera await taxonomic revision for different reasons. Here, we present an overview on the taxonomic and ecological knowledge gaps of the family Syrphidae in the Afrotropical Region and show how these gaps impede conservation efforts, plant-pollinator network studies, life-cycle history, biological control, and other applied aspects in which hover flies could be involved.

The diversity of pollinating Diptera in African Biodiversity Hotspots project

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Keywords: Afrotropical, citizen science, taxonomy, training

The Diversity of Pollinating Diptera in Afrotropical biodiversity hotspots (DIPoDIP) project studies the biodiversity of true fly families (Diptera) in Biodiversity Hotspots of the Afrotropical Region (AR). The project will improve the taxonomy and identification of these families and provide basic data on their distribution and (pollination) ecology. This is achieved through training of entomologists, conservationists and officials from the AR, including PhD and MSc students, as well as joint research endeavours. Workshops with local partners and stakeholders translate the results for policy making, Citizen Science (CS) and education. This will result in Red List assessments and improved conservation strategies for Diptera biodiversity in the AR, and CS and education activities raise awareness of the importance of Diptera in pollination, food security and nature conservation. An overview of current and future activities and outputs will be provided. The project is funded by the Directorate-general Development Cooperation and Humanitarian Aid through the Framework agreement with KMMA.

Do C₄ Chenopodiaceae constitute superior hosts for gall-inducing midges in harsh environments?

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Keywords: Chenopodiaceae, gall midges, oviposition, photosynthesis, plant vigor, preference-performance

The Plant Vigor Hypothesis (PVH) suggests that herbivorous insects prefer, and perform better on vigorously growing plants or plant parts, which is especially true for species that are intimately associated with plant tissues, such as gall-formers. Gall midges (Cecidomyiidae) constitute the largest group of gall-inducing insects, and one of the plant families on which they are particularly diverse is the Chenopodiaceae, supporting hundreds of cecidomyiid species in desert and salt marsh habitats worldwide. One of the adaptations enabling chenopods to thrive in such harsh habitats is the C₄ photosynthetic pathway. Interestingly, the overwhelming majority of gall midges associated with Chenopodiaceae are found on C₄ rather than C₃ species, but the reason for this trend has not been studied to date. To test whether this observation can be explained by the physiological superiority of C₄ over C₃ chenopods, we studied several species in the genus *Suaeda* that grow in sympatry along the Jordan Valley in Israel, some of which are associated with several species of gall midges. We subjected plants of C₄ and C₃ *Suaeda* species to shading and watering treatments in the field, and grew several *Suaeda* species of both types under controlled conditions in an attempt to offset the physiological disadvantage of the C₃ species. To test whether these manipulations will affect the preference and performance of the gall midges, we conducted choice and no-choice tests. In no-choice tests the midges completed their development on all C₄ *Suaeda* species, including a species which is not their natural host, and developed on a C₃ species to some extent. In choice tests on greenhouse plants, all cecidomyiid species completed their development only on the C₄ species. These results indicate a clear preference for C₄ plants, and thus support the Plant Vigor Hypothesis, although it is likely that other plant attributes are at play.

Cecidomyiidae (Diptera) diversity in Finland in the light of high-throughput DNA barcoding and nuclear sequencing

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Keywords: dark taxa, DNA barcoding, gall midges

Species inhabiting earth have been under taxonomic research for over 260 years, and less than 2 million species have been described thus far. It has been estimated that this number represents only a fraction of the world's true species number, and despite the admirable work of taxonomists, the vast majority of species are still yet to be found and described. Species lacking descriptions often belong to hyperdiverse groups and are commonly tiny in size, and sometimes lacking or having very few distinguishable external characteristics, making morphological examination impractical. These species are often referred to as dark taxa. Cecidomyiidae represent an excellent example of a species group of dark taxa with less than 7000 described species, but the true species number is regarded as unknown. While the most of the species are still to be found, we are simultaneously facing human induced biodiversity loss and high extinction rates. In order to study world's biodiversity more comprehensively in this current situation, we need efficient DNA-based tools to accelerate species discovery and species identification. Based on DNA barcodes, Cecidomyiidae is estimated to consist of 1.8 million species. In Finland, Cecidomyiidae are a poorly studied group and only 384 species are known from the country. With an objective to gain more information of Finland's Cecidomyiidae species diversity, over 9000 specimens were collected from 21 sites across the country. The DNA barcode region of the mitochondrial COI (Cytochrome c oxidase subunit 1) gene was analyzed non-destructively using high-throughput Sequel I platform. 7838 validated sequences were recovered and assigned to 1420 BINS. Additionally, 3 nuclear markers were sequenced from the same batch of specimens in order to gain support for BINS, that were used as species proxy. The further results will be elaborated in the presentation.

Taxonomic study of the genus *Dasysyrphus* (Enderlein) (Syrphidae) in Korea

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Keywords: cryptic species, DNA barcoding, Korea, Syrphinae, Syrphini

Dasysyrphus albostriatus (Fallén), *D. bilineatus* (Matsumura), and *D. tricinctus* (Fallén) were known in Korea. As a result of our ongoing study of Korean syrphid flies, we further recognized the following three species including one putative new species (code name is given): *D. hilaris* (Zetterstedt), *D. venustus* (Meigen), and *D. n.sp.-A*. These three species have been previously misidentified as *D. albostriatus* in Korea. Instead, *D. albostriatus* turned out to be a very rare species represented by a single male specimen in our collection. We conducted a DNA barcoding analysis of 20 species and 129 samples of *Dasysyrphus* (Enderlein) together with selected related taxa, and the resulting neighbor-joining tree supported the monophyly of this genus (50% bootstrap value). Our analysis also reasonably supported the four previous species groups (*albostriatus*, *pinastri*, *tricinctus*, and *venustus* groups). Interestingly, *D. bilineatus* which was previously placed in the *venustus* group, appears to be an independent lineage represented by three potential cryptic species. Similarly, all the Nearctic BOLD samples listed as *D. venustus* appear to be at least three closely related species.

Taxonomic study of the genus *Epistrophe* (Walker) (Syrphidae) in Korea

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Keywords: DNA barcoding, Korea, Syrphinae, Syrphini

As a result of our taxonomic study of the genus *Epistrophe* (Walker) in Korea, we recognized the following 16 species including seven putative new species (code names are given): *E. aeka* (Kimura), *E. aino* (Matsumura), *E. annulitarsis* (Stackelberg), *E. flava* (Doczkal & Schmid), *E. griseofasciata* (Matsumura), *E. grossulariae* (Meigen), *E. nitidicollis* (Meigen), *E. sasayamana* (Matsumura), *E. shibakawae* (Matsumura), *E. n.sp.-A*, *E. n.sp.-B*, *E. n.sp.-C*, *E. n.sp.-D*, *E. n.sp.-E*, *E. n.sp.-F*, and *E. n.sp.-G*. We also conducted a DNA barcoding analysis of 25 nominal species and 130 samples of *Epistrophe*. In our barcoding analysis, despite insufficient bootstrap support, *Epistrophe* formed a cluster with *Epistrophella* (Dušek and Láska) and *Leucozona* (Schiner), topologically supporting their close relationships. Our analysis also showed some interesting relationships worthy of attention: First, at least two distinct cryptic species were found within the *E. grossulariae* cluster. Second, the taxon identified as *E. nitidicollis* seems to represent at least three distinct species. Third, three BOLD-deposited European samples listed as *E. olgae* (Mutin) showed almost identical barcode sequences with the European *E. nitidicollis*.

**Diptera collection of the African Natural History Research Trust
– a developing global resource**

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Keywords: Afrotropical, collection development, Diptera collection, expedition

Situated in rural Herefordshire in the United Kingdom, the African Natural History Research Trust (ANHRT) is a charitable trust founded in 2010, which focuses exclusively on Afrotropical insects. The institution organizes and funds four or five expeditions to Africa each year focusing on remote, inaccessible region that have been poorly sampled. To date, 60 separate expeditions of 4–6 weeks duration have taken place in 14 countries in Central, East, Southern and West Africa. The main systematic focus of the institution has been Lepidoptera, with over 500,000 specimens, but the institution is now diversifying by developing collections of other insect orders, including the Diptera. Since my appointment in 2022, I have been developing the Afrotropical Diptera collection from scratch, firstly through the processing of samples from former expeditions and secondly through active fieldwork in the region. This presentation outlines the various processes in development of the rapidly expanding fly collection, its storage, accessibility and future development.

**Into the “*Heart of Darkness*” – Diptera survey of
Nouabalé-Ndoki National Park, Congo**

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Keywords: Afrotropical, Congo, rainforest, sampling protocol, surveys

The Nouabalé-Ndoki National Park in the remote far north of Republic of Congo (former Congo, Brazzaville) covers more than 4,000 Km² of contiguous lowland rainforest, that is arguably the best example of an intact forest ecosystem remaining in the Congo Basin. Unique to the park are open areas of wetlands known as “baïs” and research in the park has until recently focused on the study of large mammals, especially lowland gorillas and forest elephants and the park has never been sampled entomologically. The African Natural History Research Trust (ANHRT), in collaboration with the Wildlife Conservation Society (WCS), has organized four expeditions to the park over a two-year period, focusing on a survey of the Lepidoptera and Diptera. This presentation gives a brief outline of the park, its habitats, dipterological sampling methods, preparation techniques in the field and gives some broad preliminary results of the families of Diptera samples. A series of papers dealing with the Diptera fauna of the park are planned.

**Introduction and background to production
of Volume 3 and progress with Volume 4 of the
*Manual of Afrotropical Diptera***

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Keywords: Afrotropical, book launch, identification, systematics, taxonomy

The *Manual of Afrotropical Diptera* project was initiated in 2009, with the aim of providing a comprehensive guide to Diptera occurring in the Afrotropics. Volumes 1 and 2 were published in 2017, with Volume 1 included 11 general chapters covering diverse topics, such as forensics, natural history, biogeography, medical importance, *etc.* and also included a user-friendly “Key to Diptera families — adults” and a “Key to Diptera families — larvae”. Volume 2 included 56 chapters, covering the nematocerous Diptera and lower Brachycera and included a diagnosis for each family, information on biology and immature stages, classification, identification, a key to genera (if more than one is represented in the region) and a genus-by-genus synopsis of the fauna. Volume 3 was published in 2022 and includes 52 chapters dealing with Brachycera—Cyclorrhapha, excluding Calyptratae, with information and keys under the same headings as Volume 2. This Volume is officially launched at ICDX, in a special event fully sponsored by the African Natural History Research Trust. In this presentation a brief background is given of the production of Volume 3 and an update is provided of progress with the production of the fourth and final volume of the series, which deals with Brachycera—Calyptratae.

The automated taxonomist—prospects and perils of monitoring the Norwegian Sciaroidea fauna through metabarcoding

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Keywords: metabarcoding, Norway

Over the last decade a solid reference archive amounting to some 11000 Nordic DNA barcodes of Sciaroidea has gradually been built, largely by trained taxonomists, on the Barcode of Life online database BoldSystems (BOLD), resulting in relatively high taxonomic precision and coverage of all families except Cecidomyiidae which still is mostly represented with unidentified sequences. However, the work required to achieve high-quality reference libraries for species rich taxa like the Sciaroidea can be likened to family-level taxonomic revisions that will take decades to fully accomplish. Only when all the sequences and BINs are named and properly calibrated through a rigorously quality-checked reference library the great potentials of metabarcoding and eDNA ecology can be fully realized. In 2020 a national insect monitoring program was initiated by the Norwegian Environmental Agency. By utilizing any reference archive at hand, the metabarcoding program yearly associate Linnean names to millions of genetic reads obtained from Malaise trap samples collected throughout south-eastern Norway, gradually expanding to all of Norway. Those unvouchered genetic reads are dwarfing traditional taxonomic output from the museums like a tsunami in online databases, at the risk of undermining the core foundation of its base: the vouchered, collection-based reference archive. At the same time, the quantity and quality of the reference archive already at hand ensures that Sciaroidea is a dominating group among the identified Diptera reads in Norway, providing large amounts of potentially high-quality data on their distribution and population level trends over time. With examples pulled among Sciaroidea the prospects and perils of the current situation will be addressed, leading to questions like: Will traditional taxonomy survive this data tsunami? Why are the reference archive providers not acknowledged nor cited? How can we best assure the quality of metabarcoding data? How can we as taxonomist get in charge of the development?

Towards revising the entire Nordic-Baltic fauna of fungus gnats by building a complete image archive online

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Keywords: digital imaging, DNA barcoding, integrative taxonomy, Mycetophilidae, Nordic-Baltic Region, online cooperation, Sciaroidea, taxonomic revision

The fragmented taxonomic literature of Sciaroidea hinders both taxonomic progress and access to the group by non-specialists. To overcome this, we aim at revising the entire Nordic-Baltic fauna of the families Bolitophilidae, Diadocidiidae, Ditomyiidae, Keroplatidae and Mycetophilidae by publishing a monograph in five volumes and build a complete image archive for their identification online. Simultaneously we work towards completing a solidly curated reference archive of DNA barcodes of the same fauna on BOLD Systems. The Nordic-Baltic fauna of these five families approaches 1200 species recognized through our integrative studies, of which almost 200 are considered new to science. To accomplish this huge endeavor, we have since 2020 worked tightly together on Microsoft Teams where we daily share and discuss new images, gradually build the image archive and work to write the manual manuscripts. The team consist of five prime contributors from Norway, Finland, Russia, and Estonia with nine additional followers also from Sweden. We have refined our techniques for z-stacking and digital editing of images to ensure dissected and in situ terminalia viewed from different angles are rendered crisp and clear. Progress building the image archive is currently at 700 (60%) species fully covered and we are finalizing the first volume of the monograph treating 197 species, including 28 (14%) being newly described. Examples from our co-operative work and resulting image archive and monograph will be provided followed by a discussion of impact and funding for this kind of comprehensive taxonomic project.

Tabanidae of the Lower Suwannee Wildlife Refuge, Florida, USA

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Keywords: *Chrysops*, deer flies, horseflies, *Tabanus*, trapping, trolling, yellow flies

A multiyear study is in progress to determine the species composition and seasonal abundance patterns at the Lower Suwannee Wildlife Refuge (LSWR). This 54,000 acre refuge is located along the southern edge of the Big Bend Region of Florida's Gulf coast. The refuge was established to protect this riverine and estuarine ecosystem dominated by fresh and saltwater wetlands. The diverse mixture of uplands, freshwater, saltwater and their associated wetlands, creates a great variety of habitats. Various trapping techniques (primarily Nzi and H-traps and trolling) to collect adult specimens, which will be identified morphologically and molecularly. Thus far, the traps have mostly collected a diversity of *Tabanus* spp, *Chlorotabanus crepuscularis*, and *Diachlorus ferrugattus* while trolling collections have been mostly *Chrysops* spp.

The Bohart Museum Diptera collection need for systematist application

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Keywords: Bohart Museum, dipterists, museum collection, systematists

The Bohart Museum Entomology Collection was founded in 1946. Richard M. Bohart started with two Schmidt boxes of Calliphoridae and Apidae. Currently directed by Lynn Kimsey and managed by Brennen Dyer, the Bohart Collection contains over an estimated 8 million specimens. In more than 75 years the collection has grown through the donation of personal collections as well as global research projects and surveys. Notable collections from Culicidae are provided by Bohart, Barr, Nielsen, and Zavortink. Expansion of the Diptera Collection includes locations in Papua New Guinea, Scandinavia, Australia, Sulawesi, Indonesia, Somalia, Belize, Democratic Republic of the Congo, Africa, Brazil, Costa Rica, countless additional countries, plus expansive North American fauna. Aid is needed for the identification of unknown Diptera to family and species level taxonomy. Approximately 200 drawers of Diptera in the collection are entirely unidentified. Most significant unidentified Diptera by drawer are estimated as follows: Tachinidae (21), Culicidae (20), Syrphidae (14), Muscidae (12), Anthomyiidae (10). This extensive research collection provides the necessary information for continuing studies of various applications. Diptera systematists are needed at any capacity to further scientific research and utilization of the Bohart Museum Collection.

Importance of the Fur Formation outcrop (Denmark) for evolution of the Holarctic fauna of Diptera, Nematocera

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Keywords: Eocene, fossil, Fur Formation, imprint, Paleocene

The Eocene began with the episode of thermal maximum (PETM). Although the annual temperature gradually decreased, the climate in Europe in this epoch is considered subtropical. High volcanic activity in the North Atlantic area has produced very important paleontological sites. While the lava cooled and hardened, the volcanic ash particles were carried by winds towards Denmark. Ashes of approximately 200 separate volcanic eruptions sedimented in layers on the seafloor. The Fur Formation, restricted to the area around the western Limfjord (northwestern Denmark), is the oldest known Tertiary geological deposit (the earliest Eocene, ~55 Ma) known for its abundant fossils and the quality of perfectly preserved (3D) imprints covered with fine grained volcanic ashes. At this outcrop the nematoceran flies are abundant, and preserved not only with all morphological structures but also color patterns on the wings. The quality of these specimens can be compared with that of the amber inclusions. Numerous representatives of the families Limoniidae, Tipulidae, Cylindrotomidae, Culicidae, Anisopodidae, and Bibionidae were found. The preserved terrestrial fauna lived primarily in the land areas closed to the sea, and was brought out to sea by winds, and water currents of rivers and occasional floods. At the Paleocene-Eocene boundary there existed two continuous connections between North America and Europe: via Greenland (Thulean route) and via Iceland (de Geer route). Fossils also indicate an exchange of fauna between North America and Asia from the Late Cretaceous to the Eocene via the Beringia Land Bridge. The Fur Formation fauna shows the last moment of natural interchange of species of subtropical fauna in the Holarctic region.

**New species of the genus *Bactrocera* Macquart
(Diptera: Tephritidae: Dacinae) with red stripes on scutum**

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Keywords: *Bactrocera dorsalis* complex, cue lure, Dacini, identification key, pests

The tribe Dacini is the largest and most complicated taxon among tephritid fruit flies, with more than 1040 described species. The genus *Bactrocera* includes many species of economic importance as pests of fruits and fleshy vegetables. In 2019, our colleagues from the California Department of Food and Agriculture surveyed fruit flies in the Philippines (Luzon). While sorting the material, we discovered two previously undescribed species of *Bactrocera* with distinctive red stripes on their scutum. Another new species with similar features was collected from Borneo by Camiel Doorenweerd and colleagues that same year. Red stripes on the scutum are not a very common character among species in that genus, mostly among species in the *Bactrocera dorsalis* species complex, the most taxonomically difficult group in the genus. Newly discovered species *Bactrocera* sp.1, *B.* sp.2 and *B.* sp.3 are illustrated and discussed. The following fifteen other known species of *Bactrocera* sometimes or always also have red stripes on the scutum. To distinguish newly discovered species from similar species in the genus, an illustrated key is presented and discussed. COI DNA sequence data from the COI-5P and COI-3P mitochondrial DNA gene regions that can be used for molecular identification are also provided.

Revision of the New World species of the genus *Tephritis* (Diptera, Tephritidae, Tephritinae)

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Keywords: Asteraceae, distribution, fruit flies, key, Nearctic, new species, peacock flies, synonymy

Genus *Tephritis*, with more than 170 described species, is mostly found in the Palearctic Region, but about 10% of the known species occur in the Nearctic Region. R.Goeden from UC Riverside studied the biology, reared large series from the host plants, and studied morphology of the genus *Tephritis* in California. It allowed to discover and describe four previously unknown species from California between 1993 and 2001. During collection surveys for the California Insect Barcode Initiative and studies of the collections of the California State Collection of Arthropods, Bohart Museum of Entomology, and W.F.Barr Entomological Museum (Idaho), three previously unknown and unnamed species were discovered: *Tephritis* sp.1, *T.* sp.2, and *T.* sp.3. Illustrations and distribution maps are included in the presentation. In the collections, *Tephritis* sp.1 and *T.* sp.2 were previously misidentified as *T. araneosa* (Coquillett, 1894), *T.* sp.3 as *T. webbii* (Doane, 1899). Several other potential species of the *T. araneosa* complex are discussed, but not described, they require extensive host relationship studies and DNA sequence comparisons before any taxonomic changes can be made. Examination of the type specimens of *Urellia pacifica* Doane, 1899 and *U. aldrichii* Doane, 1899 showed that they were erroneously synonymized with *T. araneosa*. Both species undoubtedly belong to the genus *Tephritis*. *Urellia aldrichii* should be removed from the synonymy and become a valid name *Tephritis aldrichii*. Before removing *U. pacifica* from synonyms, additional research on the *T. araneosa* species complex is needed before taxonomic changes can be made. An illustrated identification key for North American *Tephritis* is prepared and presented.

Revisiting Tachiniscidae (Diptera: Tephritoidea): status, diagnosis, phylogeny, and diversification

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Keywords: new taxa, morphology, phylogeny, taxonomy

The concept, diagnosis and boundaries of the family Tachiniscidae were substantially changed by V. Korneyev (1999) by the inclusion of the genera *Ortalotrypeta* Hendel and *Cyaphorma* Wang. He also lowered its rank to a subfamily within Tephritidae, mainly for practical reasons of identification, as all the diagnostic characters of the latter family were present. Further studies (Korneyev & Norrbom 2006; Korneyev 2012) have increased the number and morphological diversity of included species, which mainly share a unique structure of the female ovipositor, apparently related to the parasitoid lifestyle of their larvae and oviposition in caterpillars. Recent discoveries of previously unknown taxa and the discovery of new morphological characters make it necessary to revisit the tachiniscines. Their diversity is found to be even greater, with the inclusion of several taxa that mimic wasps and have a fascinating superficial similarity to some Tephritidae Adramini, sharing only a few key features with the tachiniscines: the specific structure of the ovipositor (synapomorphy), the absence of fine erect hairs on the anatergite — plesiomorphy (their presence is an apomorphy of the Adramini), and the absence of the greater ampulla — plesiomorphy. The presence of the greater ampulla is a universal diagnostic character and apparently a synapomorphy present in all the Tephritidae including Blepharoneurinae, but excluding tachiniscines. The latter fact makes the inclusion of tachiniscines in Tephritidae controversial. However, they may appear as a sister group to other Tephritidae (Korneyev, 1999), a sister group to Pyrgotidae (McAlpine, 1989), or even to Pyrgotidae + other Tephritidae. Phylogenetic relationships among known tachiniscine taxa are analysed and new tribes are proposed for several monophyletic lineages.

**Discovery and revision of the bamboo picture-winged flies
(Diptera: Tephritoidea: Ulidiidae: Lipsanini):
unknown diversity and taxonomy**

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Keywords: bamboo, bionomics, phylogeny, Poaceae, taxonomy

In 2008 and 2011, unknown species of Lipsanini picture-winged flies were discovered for the first time in association with bamboo in Peru and Bolivia. Detailed analysis of morphological characters of adults and larvae showed that they are related to the genera *Aspistomella* Hendel and *Ulivellia* Speiser, both very rare in collections and known from 3 species originally included. Further study of the collection material added 15 previously undescribed new species and 4 species described in the genera *Euxesta*, *Paraphiola* and *Polyteloptera* from the Neotropical Region, which together form a group of species that share some characters of head morphology, but are very diverse in wing venation and pattern, and male genital structure. These species are described and illustrated in detail, including SEM and high-resolution macro- and microphotographs. Phylogenetic analysis based on morphology shows that they form a monophyletic lineage, either sister to or nested within a larger complex of genera, some of which are associated with maize and other tall grasses (Poaceae). Known larvae inhabit water-filled bamboo internode cavities with small holes created by Crambidae (Lepidoptera) caterpillars that attack young bamboo culms that sprouted during the previous bamboo shooting season. The semiaquatic larvae are saprophagous on wet bamboo walls or in the water, often floating with their posterior spiracles attached to the water surface.

Phylogenetic relationships within the Diptera Nematocera in light of new paleontological data

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Keywords: evolution, fossils, Mesozoic, phylogeny

The relationships between different groups within the order Diptera have fascinated dipterologists since long time. The first attempts to construct a general phylogenetic tree of flies were made by Rohdendorf and Hennig in the 1960s and 1970s. Initially, phylogenetic trees were constructed based solely on morphological features. Recent years have provided us with abundant new data from molecular studies. Unfortunately, molecular studies that relatively well explain relationships within the Brachycera, have failed to meet expectations within Nematocera. Although the oldest Brachycera appeared as early as the beginning of Middle Triassic, they evolved very slowly and only a few families existed in the Jurassic. The greatest radiation in Diptera Brachycera occurred relatively late in the Cretaceous period, and as a result, the vast majority of families have modern representatives. Evolution of Diptera Nematocera began in the Lower Triassic (~ 245-248 Ma), and in contrast to the Brachycera they radiated rapidly during the Upper Triassic and Jurassic. Unfortunately, almost 100 Ma of their evolution is documented only in sedimentary rocks. The imprints are difficult to study and interpretation, and provide incomplete information on morphology. Moreover, there are large gaps in this record, especially among flies with small and delicate body structure. Fortunately, we have an increasing number of specimens from Cretaceous fossiliferous resins with remnants of Triassic and Jurassic faunas. They enable us not only studying all external morphological structures of extinct evolutionary lineages but also facilitate a correct interpretation of those traces which are preserved in imprints. We should be aware that elucidation of phylogenetic relationships, especially within the Diptera Nematocera, is not reliable without the data on fossil taxa.

A glimpse into the Afrotropical Mycomyinae, with emphasis on the genus *Mycomyiella* Matile (Mycetophilidae)

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Keywords: Afrotropical, biodiversity, fungus gnats, new species, systematics

The subfamily Mycomyinae, a uniform group of fungus gnats, comprises 11 extant genera containing more than 600 species worldwide. Members of the subfamily are characterized by legs with tibial setae arranged in regular rows, head with two ocelli situated far from eye margins (except for *Syndocosia* Matile that has ocelli missing), wings without macrotrichiae and empodia absent. Mycomyinae has shown to be monophyletic by morphological as well as by molecular characters. In the Afrotropical Region, the subfamily is represented by 86 species in eight genera, viz. *Dinempheria* Matile (7), *Moriniola* Matile (1), *Mycomya* Rondani (9), *Mycomyiella* Matile (9), *Neoempheria* Osten Sacken (11), *Parempheriella* Matile (38), *Syndocosia* Matile (9) and *Viridivora* Matile (2). Except for globally distributed *Mycomya* and *Neoempheria*, the other six genera are endemic to the Afrotropical Region. However, the diversity of Afrotropical Mycomyinae is still poorly understood and the majority of the species are known from their type localities only. Study of extensive material from Tanzania, Zambia, Uganda, Congo DR, and São Tomé and Príncipe revealed remarkable undescribed diversity, especially in the monophyletic genus *Mycomyiella*, with more than 30 new species recognised. The newly discovered species are well delimited based on the structure of the male terminalia allowing division into species groups. Unexpectedly, the current concept of other endemic genera of Afrotropical Mycomyinae based mostly on wing venation and thoracic vestiture does not completely hold up and needs to be revised.

Revising the Australian Sphaeroceridae

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Keywords: Australia, *Howickia*, taxonomy, wing loss

The Australian sphaerocerid fauna is reviewed based largely on a study of almost 7,000 specimens collected by S.A. Marshall. We recognize 19 native and three endemic genera and a total of 114 recognized species, of which 13 remain to be described and another 11 are newly recorded from Australia. Introduced sphaerocerid species, including some newly recognized, make up 22% of the known fauna. Many of these native Australian species can also be found in the neighbouring countries of Southeast Asia and Oceania. *Howickia* Richards, now redefined and revised to hold 43 species, including all wingless and short-winged Australian sphaerocerids, is the largest, most heterogenous, and most commonly collected genus of Sphaeroceridae in Australia. *Howickia* is also found throughout southern Asia and New Zealand, the latter fauna including both wingless and winged *Howickia* species. In Australia, most species of *Howickia* are known from the rainforests along the eastern coast, specifically the Gondwanan rainforests of New South Wales, Queensland, and Tasmania, though the genus is known from almost every state.

**Advances and perspectives in the systematics
of South American bee flies and micro bee flies
(Diptera: Bombyliidae and Mythicomyiidae)**

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Keywords: Brazil, Exoprosopini, Lomatiinae, Neotropical, systematics

Ongoing projects concerning the systematics of Bombyliidae (Exoprosopini and Lomatiinae) and Mythicomyiidae, based on comparative morphology, reveal important advances in knowledge. Such projects assist in training needed human resources in helping reveal Brazil's Diptera diversity. Recent publications on Exoprosopini corroborate its monophyly and show New World species of "*Ligyra*" Newman as monophyletic and separate from its Australasian species, forming the new genus *Nyia* Márquez-Acero, Lambkin & Lamas. When more Neotropical Exoprosopini species are added to the matrix, the same pattern is observed for related genera. Preliminary results reveal that New World species of the genus *Exoprosopa* Macquart *s.l.* form a separate monophyletic group (a result similar to previous studies focusing Afrotropical and Australasian exoprosopines), and a new genus will be necessary for this clade. Regarding Lomatiinae, preliminary results corroborate the monophyly of the subfamily and of its known tribes. Some New World genera will be synonymized, and seven new South American (SA) species are being described. However, the most outstanding results concern the diversity of Mythicomyiidae in Brazil. Previous to 2014, just one species of *Pieza* Evenhuis was known from Brazil and three genera sharing 14 species were known for SA. Recent work shows a much more diverse and widely distributed family. Three new genera were erected, 15 new species were described, the first records of *Glabellula* Bezzi in Brazil and Colombia were published, and our ongoing studies reveal that the Brazilian fauna is much more diverse, not only from a taxonomic perspective [currently, in the Neotropics, there are 37 species in seven genera including species not yet described [new species of *Amydrostylus* Lamas, Falaschi & Evenhuis, *Heterhybos* Brèthes, *Mythicomyia* Coquillett – until now restricted to the Nearctic – and *Pieza*] and new genus records for Argentina, Peru and Brazil], but also a biological one (a cave-dwelling species was found).

A fruitful decade of tephritid fly surveys in Bangladesh (Diptera: Tephritidae)

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Keywords: *Bactrocera*, *Dacus*, *Zeugodacus*

Since June 2013, snap-shot surveys of dacine fruit flies have been carried out in rural environments and protected forest areas of Bangladesh, using traps baited with male lures (cue-lure, methyl eugenol, zingerone). These surveys have increased the number of species known in the country from 7 to 34, including a new species, based on 195,483 specimens collected and identified to species level. We also present data from two years of population monitoring for pest species that provided baseline data for a planned area wide suppression exercise. Starting in 2021, further sampling using traps baited with synthetic food attractants (torula yeast, BioLure) has yielded an additional 27 other species of non-dacine tephritids, based on 1,180 collected specimens. Through host fruit surveys, immatures of a number of species were made available for study, including the first rearing of *Dacus longicornis* Wiedemann, being described by Gary Steck.

Tephritoidea of economic importance

A photographic atlas of the Dacini of the world (Diptera: Tephritidae)

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Keywords: *Bactrocera*, *Dacus*, *Monacrostichus*, *Zeugodacus*

The dacine fruit flies (Diptera: Tephritidae: Dacini) is a large and diverse group, currently comprised of 1,016 described species, among which 84 are pests that cause considerable damage to cultivated fruits and cucurbits. Taxonomic revisions and keys available to identify the Afrotropical, South-East Asian and Australasian species mostly rely on line drawings representing average-looking specimens of each species. To complement current identification tools, we compiled an open access photographic atlas with color photographs (face, scutum, abdomen, wing, lateral view, intraspecific variants whenever available) for 759 of the species, including all the pest species, along with current information on synonyms, distribution, lure response, pest status, and references to published line drawings. An up-to-date checklist of all the species is also included.

Experimental taxonomy: addressing the taxonomic impediments through localizing and prudent disregard

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Keywords: dark taxa, DNA barcoding, experimental taxonomy, integrative biodiversity discovery, large-scale species discovery, next generation sequencing

Most biodiversity remains undescribed and is concentrated in “dark taxa” characterized by few described species in an ocean of undescribed diversity. This interferes with biodiversity discovery, inventories, monitoring, and filing new information in the biological literature. Recent advances in sorting specimens to putative species with molecular markers have greatly sped up the discovery process. However, two challenges remain: discovering efficient methods for integrative species description and distinguishing between new and described species. We address both by testing feasibility of an integrative “outward moving” regional approach to tackling dark taxa, demonstrating that species boundaries are stable even with increased spatio-temporal sampling. 69 of 71 morphospecies of scuttle flies from a single trap on Sentosa Island, Singapore, remained distinct when additional samples from other parts of Singapore and Indonesia from 2013 to 2018 were added to the single trap data. Our findings indicate that increased spatiotemporal sampling generally adds new morphospecies. However, if morphospecies have borderline morphological and molecular differences in the single trap dataset, increased spatiotemporal sampling might show those morphospecies as single species with large intraspecific morphological and molecular variation. Subsequently, we propose to what extent existing literature and types can/should be used. Lastly, we describe 69 new species from a single trap placed on Sentosa Island, with short, but integrative diagnoses. We demonstrate how an efficient workflow consisting of targeted literature analysis and type consultation based on region can help taxonomists working on dark taxa to navigate taxonomic impediments to yield integrative species descriptions for large numbers of species. These findings have far-reaching implications for understanding the vast diversity of life, opening new avenues for scientific exploration and discovery.

Bot flies on flatworms: hyperparasitism or just a fluke?

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Keywords: Congo, Gasterophilinae, Oestridae, parasites, Platyhelminthes, rhinoceros

Bot flies have specialized on a variety of mammal hosts, and larvae of *Gyrostigma* Brauer are exclusively known as stomach endoparasites of rhinoceroses. An unusual association of rhino bot fly larvae and parasitic platyhelminths, briefly mentioned by Bequaert (1916), is here resurrected from oblivion and explored using photography and non-destructive x-ray microcomputed tomography. Third instars of *Gyrostigma rhinocerontis* from a white rhino were found firmly anchored by their mouthhooks to gastrodiscid trematodes and an anoplocephalid tapeworm. Possible biological explanations are discussed.

Revisiting the New World bot flies: towards the delimitation of the genera of the subfamily Cuterebrinae (Oestridae)

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Keywords: Cuterebrinae, morphology, Nearctic, Neotropics, phylogeny

The subfamily Cuterebrinae is a clade of parasitic dermal bot flies that are restricted to the Americas, with a gamut of mammal host – marsupials, rodents, lagomorphs, and primates. Currently, this group is composed of approximately 70 species from two grossly different genera: the monotypic *Dermatobia* Brauer with the human bot fly (*D. hominis*) as the sole species, and all other species in *Cuterebra* Clark. Recent rediscovery of very distinct undescribed species of *Andinocuterebra* Guimarães spurred reexamination of its taxonomic treatment and phylogenetic placement. Along with other previously proposed Neotropical generic concepts such as, *Metacuterebra* Bau, *Pseudogametes* Bischoff, and *Rogenhoferia* Brauer are preliminary evaluated using morphological data. Insights on generic boundaries and diagnoses are presented and discussed.

Discovering new Acalyptratae species through DNA barcoding

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Keywords: Carnidae, diversity, DNA barcoding, Heleomyzidae, Milichiidae, Nearctic, new species

The Centre for Biodiversity Genomics is a known leader in DNA barcoding. A lesser-known fact, we have a very extensive voucher collection, acquired through the deployment of Malaise traps around the world, and all that data is accessible through BOLD (<https://boldsystems.org/>). The data includes many rarely encountered families with highly divergent sequences, which gain family level identification through our curatorial work on new Barcode Index Numbers (BINs aka species proxy). One of those specific groups is the Acalyptratae, a highly understudied, species rich, and ecologically diverse group, hence with many undescribed species. Can DNA barcoding help find those species and estimate their numbers? This will be explored using three case studies from recent publications on Nearctic Acalyptratae. Case study 1: *Paramyia* Williston (Milichiidae), your ideal scenario, where the BIN/species ratio is 1:1. With the described species tally, it is easy to estimate the number of undescribed species on BOLD based solely on the BIN count. Case study 2: *Meoneura* Rondani (Carnidae), where your species are split across multiple BINs. This makes estimating the number of undescribed species more difficult, but still possible by looking at the barcode gap, or visually by using a neighbor-joining tree. Case study 3: *Neossos* Malloch (Heleomyzidae), the opposite scenario, where multiple species are lumped into one BIN. This makes the estimation of new species practically impossible without morphology and can point out to possible synonymy. This is a cautionary tale about using DNA barcoding blindly. It is a great tool, but morphology remains an important first step!

Migratory birds and louse flies – a Canadian review

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Keywords: Aves, Acari, Canada, DNA barcoding, diversity, ectoparasites, Hippoboscidae, louse flies

Louse flies (Hippoboscidae) are large, attractive ectoparasites that can be found on birds and mammals. Due to their specific life cycle, museum specimens are often rare and old, and their high morphological variation within species also makes their identification difficult. Most of the knowledge on North American species comes mainly from two authors, Bequaert (1950s) and Maa (1960s), with very little addition since then. Recent works were often small-scale and limited to one location. My recent work at a raptor banding station in Ontario (fall 2020) has showed that raptors have a high prevalence of louse flies, even more on sharp-shinned hawk (*Accipiter striatus* Vieillot) and red-tailed hawk (*Buteo jamaicensis* (Gmelin)). COI has also proven to be quite reliable to separate species, track misidentification on online databases (e.g, GenBank), and helped confirm the first record of *Ornithomya avicularia* (Linnaeus) in North America and the first record of *Ornithomya bequaerti* Maa in Europe. With discoveries being made with only one year of data, the project was expanded to 20 stations/locations across Canada, collecting primarily during fall migration (2021-2022). Data processing is still under way. This project focuses on the species distribution across Canada, but also interactions with their host and their own parasites, by looking at the identity of their hyperparasitic mites via morphology and DNA barcoding. So far, year 1 count louse flies collected off 841 birds (1 vial per bird), representing 84 distinct species from 6 different orders, dominated by the Passeriformes. 176 vials had flies with mites and/or egg clusters. Occasionally, phoretic lice were also found on the flies (27 instances). This will be the most extensive study looking at louse flies in Canada and will likely generate many new provincial records, while expanding our knowledge on the group.

Phylogenomics reveals accelerated late Cretaceous diversification of bee flies (Bombyliidae)

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Keywords: lower Brachycera, maximum likelihood, phylogenomics

Bombyliidae is a very species-rich and widespread family of parasitoids with more than 250 genera in 17 extant subfamilies. However, little is known about the evolutionary history shaping their present day diversity. Phylogeny was reconstructed using 94 bee fly species in 14 subfamilies using transcriptomes of 15 species and anchored hybrid enrichment (AHE) sequences of 86. We integrated data from transcriptomes across each main lineage in our AHE tree to build a data set with more genes (550 loci versus 216 loci) and higher support levels showing strong congruence with current classification, with 11 of 17 subfamilies recovered as monophyletic. Heterotropinae and Mythicomysiinae are successive sister-groups to the remainder of the family. We examined the evolution of key morphological characters through our phylogenetic hypotheses and show that neither “sand chamber subfamilies” nor “Tomophthalmae” are monophyletic in our phylogenomic analyses. Based on our results, we reinstate two tribes at the subfamily level (Phthiriinae stat. rev. and Ecliminae stat. rev.) and we include the genus *Sericosoma* Macquart (previously *incertae sedis*) in the subfamily Oniromysiinae. Our dating analyses indicate a Jurassic origin of the family, with the sand chamber evolving early on, in late Jurassic or mid Cretaceous. We hypothesize that the angiosperm radiation and the hothouse climate established during the late Cretaceous accelerated diversification of bee flies by providing an expanded range of resources for parasitoid larvae and nectarivorous adults.

Taxonomic revision of the genus *Megistopoda* (Macquart) (Streblidae) parasites of new world leaf-nosed bats (Chiroptera: Phyllostomidae) and preliminary results of species delimitation

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Keywords: bat flies, ectoparasites, morphology, species delimitation, taxonomy

Bat flies (Diptera: Streblidae and Nycteribiidae) are obligate, blood-feeding ectoparasites of bats, parasitizing only bats. Bat fly species belonging to the genus *Megistopoda* (Macquart) are characterized by presenting the femur III longer, thorax shield-like, and stenopterous wings. *Megistopoda* species are therefore unable to fly and their ability to move among host individuals is restricted and this lack of flight can lead to strong host associations. The genus *Megistopoda* is composed of three described species *M. aranea* (Coquillett), *M. proxima* (Séguy) and *M. theodori* (Wenzel), each of which has been found to be associated with host species belonging to the New World leaf-nosed bats (Chiroptera: Phyllostomidae) genera *Artibeus* (Leach) or *Sturnira* (Gray). With only three widely distributed species, *Megistopoda* is an understudied genus and species delimitation has not been rigorously assessed. Most of the used characters to identify *Megistopoda* species are ambiguous and intraspecific variation is usually not considered. In this talk, I will present the results obtained during my master's studies, where we analyzed morphological characters and a total of 12 body measures of 852 *Megistopoda* individuals (obtained from museum collections and recent fieldwork) from across their geographic range. We identified nine putative species, six new to science, but describing diagnostic characters. Two analyses (ANOVA and PCA) were performed to recognize significant differences among the quantitative characters. Thorax chaetotaxy, the femur III length, and the wing measures were the most informative to recognize morphospecies. Future research will include phylogenetic analyses using morphological and molecular data to determine relationships, host associations, and geographic distributions among *Megistopoda* species.

Flies on small islands: zooming in on Pacific sphaerocerid diversity (Acalyptratae) from Malaise trap samples in Fiji

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Keywords: Fiji, Limosininae, Malaise, Oceania, Pacific, Sphaeroceridae, surveys

The Sphaeroceridae of Fiji are reviewed based on over 5,000 specimens taken by Malaise sampling from 2002–2006 as part of the NSF-Fiji Terrestrial Arthropod Survey. No fewer than two dozen species in thirteen genera are found, many for the first time in the region. The genera *Leptocera* Olivier, *Rachispoda* Lioy, *Pachytarsella* Richards, *Parapterogramma* Papp, and *Pseudopterogramma* Papp are newly recorded from Fiji. One putative undescribed genus and at least seven undescribed species are recognized, including five species of *Parapterogramma*. This study of the Fijian Sphaeroceridae is summarized in the context of the poorly known sphaerocerid fauna of Pacific islands and more broadly the Oceanian region.

Evaluating the suitability of a transgenic blow fly for insect age estimation empirical validation

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Keywords: Calliphoridae, development rate, forensic entomology, *Phormia*,
postmortem interval, prediction, recombinant DNA, statistical confidence

The most common forensic entomological (FE) analysis is estimating the age of an insect thought to have been associated with the corpse all of its life. We favor age prediction as a statistical confidence set. Forensic insect age estimation relies on laboratory development rate data to predict insect age under the very different environmental conditions of a human corpse. Age prediction performance has been assessed in an artificial setting but not under realistic conditions. A technical barrier to realistic validation is the difficulty of knowing the true age of a maggot in a corpse exposed to wild insect colonization processes. This problem could be solved by mark-release-recapture in the field. Recover a marked individual added to the corpse as an egg and 1) its age will be known, and 2) it developed under natural conditions. We “marked” *Phormia regina* (Diptera: Calliphoridae) by engineering them to express a fluorescent protein. However, to be suitable for a validation experiment the transformation must not have altered development rate. This presentation will describe how we assessed the effect of the transgene on *P. regina* development rate in the context of future FE method validation. Wild-type, heterozygous transgenic, and homozygous transgenic strains were reared under identical laboratory conditions. Life stage was recorded at each of 18 ages at 27°C. The wild-type data were then used to predict the age of the transgenic individuals based on life stage, as would happen in a field validation study, in the form of a 95% confidence set. Greater than 98% of age predictions included the true age. The transformation did not alter development rate to a practical extent.

An integrated overview of *Paralucilia* species (Diptera, Oestroidea, Calliphoridae) in the Amazonian biome

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Keywords: Amazon, blow flies, Neotropical, integrative taxonomy

Paralucilia Brauer & Bergenstamm is a Neotropical blow flies genus that inhabit exclusively the wild environment. This behavioral aspect makes their species potential bioindicators of environmental quality. Two out five valid species, *P. fulvinota* (Bigot) and *P. paraensis* (Mello), have been recorded in the Amazon biome, but with some imprecision about their respective distribution limits due to the difficulty in diagnosing them since they present great morphological similarity. Thus, this study aimed to delimit the Amazonian *Paralucilia* species using an integrative taxonomy approach. Adult specimens were collected in the Amazon biome using baited traps. After identification, the specimens had their DNA extracted, amplified for the COI, 16S, ITS2 and 28S genes, and sequenced. Morphological analyzes were conducted with both collected and preserved in collections specimens. Bayesian Inference analyzes were performed in the BEAST™ v. 2.4.4. ABGD, mPTP and GMYC analyzes were also carried out, based on the COI gene data set, to propose hypotheses for *Paralucilia* species delimitation of the Amazon biome. In addition to the sequences obtained in this study, other available on Genbank and BOLD System were included in the analyses. *Paralucilia fulvinota* and *P. paraensis* were recovered as monophyletic lineages, and as distinct species in all analyzes for species delimitation. Furthermore, the Amazonian lineage of *P. fulvinota* is monophyletic in relation to other populations of the same species. Amazonian *Paralucilia* species can be distinguished from each other by the number of posterodorsal seta on the posterior tibia and longitudinal stripes on the mesonotum, and by male and female terminalia characters. We conclude that the use of integrative taxonomy successfully assisted in the delimitation of Amazonian *Paralucilia* species. In addition, we emphasize that this knowledge associated with the evolutionary history of species can provide essential information for the preservation of the environment in which they live.

An updated phylogeny of Neotropical Chrysomyinae (Diptera, Calliphoridae), with notes about biogeography

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Keywords: biogeography, blow flies, integrative taxonomy, Neotropical, phylogeny

Chrysomyinae (Diptera, Oestroidea, Calliphoridae) is a subfamily of blow flies of recognized medical, veterinary and forensic importance, but its evolutionary position is still unclear. In the Neotropics, five endemic genera are found (*Chloroprocta*, *Cochliomyia*, *Compsomyiops*, *Hemilucilia* and *Paralucilia*), recovered as a monophyletic group in previous phylogenetic studies. This study aimed to present an updated molecular-based phylogeny of Neotropical Chrysomyinae with a historical description of the biogeography of the group. A Bayesian Inference phylogenetic analysis calibrated with divergence times was performed based on 78 terminal taxa (57 species of Chrysomyinae + outgroup), using eight molecular markers: COI, COII, 16S, CytB (mitochondrial), CAD, ITS2, 28S, EF1-a (nuclear). For the biogeographic analyses, the resulting tree and species distribution information were evaluated using the DEC model, with or without the inclusion of speciation by founder event. For the Neotropical regionalization, the proposal of Morrone (2014) was adopted. Neotropical Chrysomyinae was recovered as a monophyletic group, sister to *Chrysomya* + Nearctic Chrysomyinae. *Chloroprocta* was retrieved as a sister group to other Neotropical Chrysomyinae, while *Hemilucilia* is as a sister group to the clade [*Compsomyiops* + (*Paralucilia* + *Cochliomyia*)]. Regarding the biogeographical analyses, the Neotropical Chrysomyinae probably emerged during the Miocene, between Panama and South America. Furthermore, the southern Brazilian domain was recovered as the probable area of origin and diversification of all genera of this subfamily. So far, this is the hypothesis of phylogenetic relationships with the largest number of Chrysomyinae taxa included, as well as the first study of historical biogeography for Calliphoridae. We hope that the hypotheses discussed in this study can contribute to a better understanding of the evolutionary history of Chrysomyinae.

**Communicating morphology to a broad audience:
a primitive crane fly story**

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Keywords: natural history, nature documentary, outreach, rearing, science
communication, storytelling, Tanyderidae

Field and laboratory observations of live organisms often provide insights valuable to investigations of form and structure typically based on preserved material. Despite this, information such as natural history data for many taxa remain limited or unavailable. An ongoing study of morphological characters in primitive crane flies and related families highlights the benefit of observing species throughout their life cycle. Tools and techniques found useful for recording observations of live insects are discussed. Common approaches used for communication of research results among scientists may not be effective for making information available to non-experts. Nature documentaries and storytelling are explored as avenues for presenting morphology in light of pertinent natural history observations.

**Review of Neotropical *Nothodixa* (Edwards) (Dixidae)
with observations on natural history**

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Keywords: aquatic, meniscus midges, microhabitat, morphology, taxonomy

The Neotropical *Nothodixa* are revised. Specimens collected during expeditions in Patagonia include mostly undescribed taxa, while redescriptions of known species are based primarily on museum material. Morphological characters previously unexplored in the *Nothodixa* of South America aid in reliable distinction of species and evidence multiple supraspecific groups. Association of life stages of recently collected species was based on rearing of individual larvae to adults or sequence data where rearing was unsuccessful. Field observations indicate microhabitat preference in some species; lab observations include feeding in larvae and adults and progression of the life cycle. Eggs were obtained from gravid females that oviposited in captivity. Morphological and natural history information recorded during this study are organized as an identification guide for larvae, pupae, males and females.

“Dark taxa” in Diptera collected for the California Insect Biodiversity Initiative reveal a need for taxonomic expertise

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Keywords: biodiversity, California, CO1, DNA barcoding, dark taxa, Malaise trap, winter

California is the state with the greatest diversity of habitats and wildlife in the USA, yet it is also one of the least explored scientifically. Hence, multiple institutions across California, along with the Canadian Centre for DNA Barcoding at the University of Guelph, are collaborating on the California Insect Biodiversity Initiative (CIBI). CIBI is an ongoing project to collect and sequence Californian insects and upload their CO1 genes to the BOLD database for further research and identification purposes, using newly-collected specimens as well as curated and identified museum specimens. Since October 2022, our group has been collecting insects across Northern California as part of CIBI, primarily using malaise traps. An abundance of insects comes to the traps even in snow-covered mountains during winter, largely comprising “dark taxa” in Diptera such as Cecidomyiidae, Sciaridae, Chironomidae, and Phoridae. Such families are expected to contain numerous undescribed species even in well-studied regions, including cryptic species that are morphologically indistinguishable. Given that most collectors tend to avoid such time and conditions in favor of ones with higher productivity, we assume that there is some under-collected and undescribed diversity among these groups in our sampling, which may be detected via DNA barcoding. For malaise trap samples from select sites in Northern California, we counted and estimated the number of flies from each family caught therein. We compared how their relative abundances changed from month to month between October 2022 and June 2023. In this presentation we discuss CIBI as a project and Diptera families collected during the winter months that may need increased attention from taxonomists.

Colour, characters and fly photography for MAD

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Keywords: Afrotropical, book launch, key, manual, photography

Color and visible structures have always been critical to how we identify and appreciate flies, but it is only relatively recently that it has been practical to take full advantage of photography to convey concepts of habitus and characters in printed manuals. The first of the Diptera Manuals, the *Manual of Nearctic Diptera*, 1981 set new standards for illustrated keys, but it contained no photographs of flies. The *Manual of Palaearctic Diptera* followed suit in 1998. The *Manual of Central American Diptera*, published in 2009, included 132 fly photographs in plates made up of single images illustrating almost all Central American families, but color photos were not used in the keys. In the meantime, the *Journal of Arthropod Identification* pioneered the publication of photo-based keys, including keys to flies on a PowerPoint template since 2007. The *Manual of Afrotropical Diptera* was in its initial stages during the same period and the editors planned from the start to make extensive use of color photographs not only in the keys but also in every chapter of the Manual. This short talk will touch on how those plans came to fruition thanks in part to some unprecedented opportunities for field photography in Africa.

Survey sweet spots ... looking for flies in all the right places

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Keywords: inventories, Micropezidae, Pseudopomyzidae, Sphaeroceridae, surveys

Diptera surveys and inventories, like the flies themselves, are almost infinitely variable and apparently adapt themselves to every available niche. Despite the resultant disparity in survey types, the things we call “surveys” all have the general properties of improving baseline knowledge of an area or habitat and providing an essential foundation for biodiversity and conservation work. Some kinds of surveys can also expedite collaboration, provide unparalleled educational opportunities, offer critical synergies with revisionary systematics and generate or test interesting hypotheses in ecology. In this brief talk I will contrast some of the different kinds of surveys from the perspective of a collaborator primarily interested in revisionary systematics of large and patchily documented Diptera families, emphasizing the difficulties in matching the objectives of survey organizers with collaborating specialists.

**Distributional range limits of undescribed long-legged flies
(Dolichopodidae: Hydrophorinae) inhabiting the intertidal zone
of remote islands in Japan**

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Keywords: *Acymatopus*, *Cymatopus*, new species, taxonomy

The morphology and taxonomic positions of three undescribed species of Hydrophorinae (Dolichopodidae) that inhabit the intertidal zone of remote islands in Japan were investigated. Undescribed species 1, 1.8 mm long, was found on sandy beaches dotted with coral rocks in the Ryukyu Islands, over a range that coincides with the distribution of coral reefs in Japan. It is assigned to the genus *Cymatopus* Kertész, 1901 based on the following setal features: antennal postpedicel lacking setae; thorax with 5 dorsocentrals and no acrostichals; fore femora with strong bristles; surstylus with hook-shaped bristle. *Cymatopus* is distributed mainly in the tropics, and this new species, which occurs north of the Tropic of Cancer, is its northernmost representative. Undescribed species 2, 2.0 mm long, was collected from the southern tip of the Satsuma Peninsula in Kyushu and from the Ryukyu Islands. It is assigned to the genus *Acymatopus* Takagi, 1965 based on the following setal features: antennal postpedicel bearing setae; thorax with 6 dorsocentrals and minute, uniserial acrostichals; fore femora lacking strong bristles; fore basal tarsomere with long bristle and membranous parts; distal margin of epandrium with some long bristles. *Acymatopus* occurs in Japan and China, and the present new species is the southernmost among the member species. Undescribed species 3, the smallest among the three species with a body length of 1.5 mm, was collected in the Ogasawara Islands. On account of having 5 dorsocentrals, no especially strong setae or other ornamentation on the legs, and a long, not apically bifid hypandrium, it cannot be assigned to any known genus of Hydrophorinae but instead seems to represent an undescribed genus closely related to *Cymatopus*. This new genus and species may represent an endemic, locally-evolved lineage in the Ogasawaras, an oceanic island chain that has never been connected to other lands.

Banquet address

Charismatic Diptera – who are we kidding?

Erica McAlister

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Keywords: dipterists, morphology, nomenclature, outreach, taxonomy

Flies. Not much liked by the average person. Not even liked by many entomologists. And frankly, you can understand why – terrible names, terrible behavior and terrible taxonomists associated with them. In this talk I will highlight some of the issues that beset Dipterists and their species of fancy, and maybe, just maybe, tell of some stories to help change the minds of the most ardent of disbelievers.

**Investigating factors associated with larval
Culicoides Latreille (Ceratopogonidae) emergence
from sylvatic and agricultural habitats**

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Keywords: ecology, emergence, larval habitat, soil analysis, water analysis

Culicoides Latreille biting midges are small, hematophagous Diptera in the Ceratopogonidae family. The larval stage of these flies typically occurs in semi-aquatic habitats at the periphery of ponds, streams, springs, and puddles, with some exceptions including manure or treehole developing species. In North America, over 150 *Culicoides* species exist, each with a unique ecology. Currently, the majority of available data focus on a single species, *Culicoides sonorensis* Wirth & Jones, which is a common North American vector of several viruses of veterinary importance. The larval habitat used by this species is predominantly dairy wastewater ponds, which are extremely organically enriched environments. However, aspects of larval habitats for most of the other North American *Culicoides* species are still poorly resolved. Data on larval habitat associations of *Culicoides* species collected in Northeastern Kansas will be presented as well as preliminary data investigating the impact of soil and water chemistry on larval presence. Understanding the larval habitat associations of diverse midge species can help in the identification of productive sites for putative vector species that may be contributing to the spread of midge-borne viruses and may aid in the development of novel control strategies.

Reassessing the rare robber fly genus *Dasylechia* Williston (Asilidae) with new morphological details, DNA barcoding, an African synonymy, and information gleaned from digital media

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Keywords: assassin fly, Bugguide, citizen science, CO1, iNaturalist, Laphriinae, taxonomy

The asilid genus *Dasylechia* Williston is redescribed and detailed figures are given for the first time of the terminalia, mouthparts, and eggs of the lone North American species, *D. atrox* (Williston). Several new museum specimens are reported for *D. atrox*. While specimens remain relatively scarce, online insect forums like BugGuide and iNaturalist provide a venue to connect amateurs serendipitously photographing this species with experts capable of recognizing it. We have compiled 88 such photo-based records, which complement the published record with added information on microhabitat, prey selection, phenology, and distribution. This species is newly reported from four US states: Indiana, Maryland, West Virginia, and Wisconsin. The mitochondrial barcode (COI) was sequenced from a recently collected specimen. Phylogenetic analyses of this gene and of 220 morphological characters support classifying *Dasylechia* in the asilid subfamily Laphriinae, but these analyses both failed to reliably resolve finer scale phylogeny.

Plenary address

Accelerating biodiversity discovery in hyperdiverse arthropod clades with robots and Nanopore sequencing

Rudolf Meier

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Keywords: biodiversity loss, species delimitation, species discovery

The biodiversity of Diptera and arthropods remains poorly understood although it comprises much of the terrestrial animal biomass, most species, and supplies many ecosystem services. One obstacle is specimen-rich samples obtained with quantitative sampling techniques (e.g., Malaise trapping). Traditional “morpho-species” sorting requires too much time. At the Center of Integrative Biodiversity discovery at the Museum für Naturkunde in Berlin, we work on specimen-based approaches that pick individual specimen from bulk samples for barcoding. We developed a robot (“DiversityScanner”) that detects, images, and measures individual specimens and moves them into the wells of a 96-well microplate. The images are used to train convolutional neural networks (CNNs) that are capable of assigning the specimens to the 14 most common insect taxa in Malaise trap samples. Many of these taxa are Diptera. To obtain biomass information, the images are also measure specimen length and estimate body volume. In order to obtain DNA barcodes, we have developed robust and cost-effective barcoding techniques involving ONT sequencers and bioinformatics tools that allow for approximate species-level sorting. The next step will be developing high-throughput species description techniques for “dark taxa”.

German Barcode of Life III: Dark Taxa – Diptera

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Keywords: dark taxa, DNA barcoding, GBOL III, Hymenoptera, integrative taxonomy,
Limoniidae, Psychodidae, Tipulidae

The German Barcode of Life (GBOL) initiative, funded by the German Federal Ministry of Education and Science (BMBF), has built a useful DNA barcode reference library for the animals, plants and fungi of Germany. While some species-rich taxa of animals are well represented in our DNA database (started in 2011), there are important gaps in the reference library in the insect orders Diptera and Hymenoptera. The megadiverse parasitoid wasps and “lower Diptera” contain many of the so-called Dark Taxa, for which only little or no taxonomic information is available. Quite often these groups also lack taxonomic experts. The present phase of GBOL, named “GBOL III: Dark Taxa”, aims at tackling both: on the one hand, the study and discovery of the (so far) unknown insect biodiversity of Germany and Central Europe in an unprecedented way; and on the other hand, the training of a new generation of taxonomists (PhD candidates) in the latest methods using integrative taxonomy. Three additional subprojects/tasks complement the research of dark taxa: "New and optimised approaches for poor quality samples and taxonomics (including DNA target enrichment)", where we research and develop new protocols to sequence poor quality samples; "Extension of the data infrastructure and communication channels (web portal and database for OTU-based data) - ASV-Table-Registry", where an extension of the database concepts is being developed so that ASV data from metabarcoding studies can be databased and assigned to species names at later stages; and "knowledge transfer and citizen science", that we use to attract attention to the diversity and importance of dark taxa in science and society. Results of the “GBOL III: Dark Taxa” will not only increase the completeness of the DNA barcode reference library, but they will heavily improve the significance of future biodiversity studies, including biodiversity monitoring programs and insect conservation management.

Molecular phylogenetics of predatory flower flies (Diptera: Syrphidae) using exon-capture sequencing

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Claudia Etzbauer¹, Sander Bot⁶, Martin Hauser⁷, Kurt Jordaens⁸,
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Keywords: flower flies, hover flies, phylogeny, Syrphinae, target DNA enrichment,
tribal classification

We developed a new bait kit (SYRPHIDAE1.0) to capture and enrich phylogenetically and evolutionary informative exonic regions. Using high-throughput sequencing we targeted 1945 CDS regions belonging to 1312 orthologous genes in 121 flower fly species. Our analyses based on amino acid and nucleotide data sets (1302 loci and 154 loci) were performed using Maximum Likelihood and Multispecies Coalescent models. Our analyses resulted in highly supported similar topologies, although the degree of the SRH (global Stationarity, Reversibility and Homogeneity) conditions varied greatly between amino acid and nucleotide data sets. Pipizinae and Syrphinae were recovered sister taxa in all our analyses, corroborating a common origin of taxa with predatory larvae feeding on soft-bodied arthropods. Based on our results, we define Syrphini stat.rev. to include the genera *Toxomerus* Macquart and *Paragus* Latreille. Our divergence estimate analyses using BEAST inferred the origin of the Syrphidae in the Lower Cretaceous (125.5–98.5 Ma) and the diversification of predatory flower flies around the K-Pg boundary (70.61–54.4 Ma), concurring with the diversification of their prey.

The integrated management of the Asian tiger mosquito (*Aedes albopictus*) in Greece with emphasis on the Sterile Insect Technique

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Keywords: IPM, population suppression, SIT, sterile insect techniques

Aedes albopictus (Culicidae) is of high concern in public health due to its severe nuisance and its vectorial capacity for many pathogens such as dengue, chikungunya, yellow fever and Zika. Its management plan is a complex system that includes coordinated actions. In this context a pilot integrated management of *Ae. albopictus*, is implemented in Greece including the synergistic action of the Sterile Insect Technique (SIT) with the simultaneous implementation of citizens' educational visits ("door-to-door"/D-t-D). During the implementation of D-t-D method a questionnaire distributed, based on KAP methodology (Knowledge-Attitudes-Practices). Results revealed that a visit-inspection of the potential breeding sites in citizens backyards can influence the residents' behavior and raise their awareness towards the reduction of mosquito breeding sites. Systematic releases of sterilized males of *Ae. albopictus* resulted in significant reduction of egg hatchability and population suppression. The outcomes from the pilot trials (both D-t-D and SIT) and the systematic entomological surveillance plan will be further evaluated to develop new management plans against *Ae. albopictus* to reduce the use of biocides in urban areas, as foreseen by EU Regulation 528/2012. Future steps also involve boosted SIT aiming to increase our SIT knowhow to invest on the production of sterilized mosquito males in Greece in all stages (mass rearing, sterilization, and releasing) for the first time. The paper supported by the projects: «moSquITo» (TAEΔK06173- National Recovery and Resilience Plan, "Greece 2.0" & EU Funding – Next Generation EU); "Research project for the entomological surveillance of mosquitoes in Attica Region (Athens, Greece)"; International Atomic Energy Agency (IAEA) – TC Project RER/5/022 "Establishing Genetic Control Programmes for *Aedes* Invasive Mosquitoes"; IAEA- Programme of Coordinated Research Activities "Quality control bioassays for irradiated *Aedes albopictus* males"(IAEA-CRP Contract No: 23915).

**A new species of *Nyassamyia* Lindner
(Diptera: Stratiomyidae) from South Africa**

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Keywords: Afrotropical, new species, taxonomy

Nyassamyia Lindner, 1980 is a small genus of Stratiomyidae endemic to the Afrotropical Region. Specimens are rarely collected, but occur from South Africa to the Democratic republic of Congo. The two described species are densely pilose, likely mimicking bees. Here we present a new species in this genus from KwaZulu-Natal, South Africa that closely resembles *N. deceptor* Curran, 1928. A brief diagnosis of the adults will be given, along with pictures of all species. Geographical distribution will be discussed, and biological observations of juvenile stages presented.

Biological observations of the larvae of *Meromacroides meromacriformis* (Bezzi)

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Keywords: DNA barcoding, flower flies, species descriptions, Syrphidae

Meromacroides meromacriformis (Bezzi) is an enigmatic hover fly, first collected in South Africa in the 1850s, but not again until 2020. A further collection in 2022 was also made in the country. While widespread in Africa, the species is rarely encountered and information about its biology is scarce. The recent collections in South Africa were associated with colonies of *Schedorhinotermes lamanianus* (Sjöstedt) (Isoptera: Rhinotermitidae) found in rot-holes within the trunks of standing living trees and in 2022 larvae were collected from a frass midden. The link between termites and *M. meromacriformis* requires further investigation, but this habitat specialization may explain why the species is encountered infrequently. Larvae collected in 2022 have been reared to adulthood in 2023. Efforts are underway to enhance conservation of both sites through education panels in collaboration with local wildlife authorities.

***Spheginobaccha pamela* Thompson & Hauser, additional locality records and description of the female**

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Keywords: DNA barcoding, flower flies, species descriptions, Syrphidae

Spheginobaccha de Meijere is a rare genus of hover flies, restricted to the Afrotropical and Oriental regions. Only eight species are known from the Afrotropics, of which only three are known from both sexes. The recent discovery of a female *Spheginobaccha pamela* Thompson & Hauser, specimen as well as recent collections of the species in South Africa brings this number to four. We present photographs and taxonomic notes on the female of *Spheginobaccha pamela*.

The Diptera of Lesotho: a special issue in the journal *African Invertebrates*

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Keywords: Afrotropical, biodiversity, species checklists, surveys

The Kingdom of Lesotho is a landlocked country, surrounded by the Republic of South Africa. While many vegetation types are shared between the countries, the extent of alpine vegetation is much greater in Lesotho, potentially holding a greater diversity of specialized species. Historic collecting in Lesotho has been limited, but some insect collections exist at various institutions. We undertook three field trips to Lesotho in December 2021, November 2022 and January 2023, visited 30 sites and collected over 6000 Diptera specimens. An overview of the recent collecting sites with habitat photographs is given. One new species has already been described from the 2021 field trip. We invite contributions to a special collection of papers in *African Invertebrates* and provide an overview of specimens and families collected for this purpose.

**The female of *Atherimorpha latipennis* Stuckenberg,
the first record of brachyptery in the Rhagionidae**

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Keywords: brachyptery, lower Brachycera, taxonomy

The genus *Atherimorpha* White is a Gondwana relic with 51 species recognized as valid. Globally, the genus has received sporadic taxonomic attention, with most revisions only addressing regional faunas, though acknowledging the link between Australia, Africa and South America. Of the twelve Afrotropical species, both sexes are known in only four species, and the female is unknown for seven species. Many species are known from mountainous habitats and emerge as adults for a limited time. Brachypterous Diptera present a collecting challenge, as the usual techniques employed often rely on passive movement by the target individuals (e.g. Malaise traps) or active searching for flying adults. We describe the female of *Atherimorpha latipennis* Stuckenberg, reporting brachyptery in Rhagionidae for the first time.

The genus *Amphoterus* Bezzi: taxonomic notes on a rare Afrotropical hover fly genus

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Keywords: Afrotropical, DNA barcoding, flower flies, species descriptions, Syrphidae, taxonomic revision

The genus *Amphoterus* Bezzi is a rare group of hover flies restricted to the Afrotropical Region. Three species are recognised, *Amphoterus cribratus* Bezzi, known from only one male and one female, *Amphoterus braunsi* van Doesburg, described from a single male and *Amphoterus londti* Midgley et al, described from a single female. While not often collected, a large series of *A. braunsi* has been accumulated over time at the KwaZulu-Natal Museum. Apart from the usual sexual dimorphism, patterns of pilosity on the face can be used to differentiate males from females in the genus. Photographs, DNA barcodes and notes on useful taxonomic features will be presented.

Diptera and vector borne diseases under a One Health perspective in Spain

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Keywords: Ceratopogonidae, Culicidae, culicoides borne diseases, mosquito borne diseases, Phlebotominae

Europe and particularly Spain have experienced an increase of vector borne diseases during the last decade. During the pandemic by SARS-CoV-2 virus, the concept One Health has been vastly promoted as a cornerstone for the understanding of diseases related to humans, animals and the environment, such as vector borne diseases. A detailed description of the major vector borne diseases outbreaks related to Diptera in Spain is provided, as well as comments about the One Health implications. In 2012, sandflies, such as *Phlebotomus perniciosus* (Phlebotominae), were the major vectors in the unprecedented outbreak of human leishmaniasis in the area of Madrid (central Spain). Hares were identified as the main reservoir of the parasites. The outbreak is considered to be the biggest in Europe still active after 11 years. West Nile virus circulation has been detected in Spain since the 80's, in birds, horses and sporadically in humans, however, in 2020 and 2021 an important outbreak of West Nile fever in humans took place in Andalusia (south Spain).. Two mosquito species, *Culex pipiens* and *Cx. perexiguus* (Culicidae) were considered the major vectors in the area. Bluetongue virus has circulated in Spain since year 2000, including the Balearic Islands in year 2000, 2004 and 2021. The virus is transmitted by species of the genus *Culicoides* (Ceratopogonidae). Wild ruminants and vector bridge species play a role in the persistence and circulation of the pathogen. More recently in 2022, another biting midge transmitted disease, epizootic haemorrhagic disease, was detected in the south of Spain affecting deer populations. Finally, several outbreaks of local transmission of Dengue virus were detected in south of Spain and Ibiza (Balearic Islands). In all cases, the implicated vector was *Aedes albopictus* (Culicidae). Diptera are the most important group related to the transmission of vector borne diseases in Spain.

Diptera sampling in remote islets: combining direct sampling and non-destructive automatic electronic traps

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Keywords: Balearic Islands, insect monitoring, internet of things, Tephritidae

General insect decline is a major concern nowadays. Obtaining long-term data on the species diversity and population abundance is of major importance to build proper managing and conservation policies. Sampling in remote places (i.e., islets) represents an important challenge for researchers due to logistic and need for resources. Relatively lower cost methods based on electronic trap types implemented in a long-term sampling may provide an optimal solution for sampling insects in remote places. In this study a multidisciplinary team of researchers from the University of Balearic Islands developed a sampling system based on non-destructive automatic monitoring traps with sensors integrated into a GIS network (BioView System). Each trap is equipped with two high-resolution cameras (18 megapixels) at two different heights (50 cm and 120 cm from the ground). Three sites with high ecological value were selected for the study in the Balearic Islands: the islets of Sa Dragonera (Mallorca), Illa de'n Colom (Menorca) and Sa Conillera (Ibiza). One electronic trap was placed in each islet in June 2022 and they are still operative. A conventional sampling was conducted in each islet for comparison with electronic traps. The conventional sampling was carried out using sweeping nets in transects over two consecutive days in May 2021 and on one day in June 2022 in the three islets. Analysis of pictures taken by the electronic traps showed that Diptera was the most frequent order detected (24% detections), followed by Hymenoptera. Diptera was also one of the most represented orders found in the conventional sampling. Interestingly, new citations of Tephritidae were reported for the islets: *Acanthiophilis helianthi*, *Aciura coryli*, *Capites ramulosa*, *Tephritis nigriacuda*, *Trupanea stellata* and one species of *Tephritis* still pending of identification. We discuss about the use of electronic automatic systems as a complementary tool for conventional systems, in particular for the long-term study of Diptera.

Role of Chloropidae in the cross pollination of the endemic *Aristolochia bianorii* (Aristolochiaceae) and other species in the Balearic Islands (Spain)

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Keywords: deceptive pollination, pollination, pollinator, pollinator entrapment

Plants of the genus *Aristolochia* use modified trap flowers to select and attract potential pollinators. Micromyioiphily and sapromyioiphily are the two main strategies recognized in *Aristolochia* for attracting pollinators. Four species of the genus *Aristolochia* are found in the Balearic Islands (Spain): *A. bianorii*, *A. clematitis*, *A. rotunda* and *A. paucinervis*. All the species show small tubular flowers that are designed for entrapping pollinators. *Aristolochia bianorii* is the only endemic species found in Mallorca and Menorca. In this study, the anthesis of *A. bianorii* was monitored in the field as well as the volatile organic compounds (VOCs) emitted by flowers. Potential pollinators were collected from a total of 344 flowers from May to June 2021. Anthesis showed four stages, from which at stage 1 (female phase), the flower was receptive for pollinators. 109 entrapped insects were counted from 344 flowers. All insects were from the family Chloropidae. The most abundant species was *Oscinomorpha longirostris* followed by *Aphanotrigonum femorellum*. In general, all insects were entrapped for 48h. Analysis of VOCs showed volatiles possibly related to brood site deception and kleptomyophilous pollination. Based on these results, we discuss about the role of Chloropidae in the pollination of the different species of *Aristolochia*.

A Tale of Two Synonymizations: progress in Neotropical Chloropidae taxonomy

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Keywords: Acalypratae, diversity, grass flies, Neotropical, new species, synonymy

The Neotropical Chloropidae is a speciose assemblage of insects with several poorly-known genera that share striking similarities with other genera. With new specimens being regularly investigated, the boundaries of the genera are not yet clearly established. Such are the cases with *Agrophaspidium* Wheeler & Mlynarek 2008 resemblance with *Acanthopeltastes* Enderlein 1911 and the likeness of *Strandimyia* Duda 1930 to *Enderleiniella* Becker 1912. Based on the examination of type specimens and reliable morphological features such as the male terminalia, we propose the synonymization of *Agrophaspidium* with *Acanthopeltastes*. Similarly, we propose *Strandimyia* as a junior synonym of *Enderleiniella*. We will discuss the limits of the genera and these new synonymies in the light of eight new *Acanthopeltastes* and two new *Enderleiniella* species. Our findings further the knowledge of Neotropical chloropids and provide a more comprehensive view of their diversity.

Resolving the Diptera tree of life through advances in bioinformatics

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Keywords: bioinformatics, phylogenetics, phylogenomics

The order Diptera is the result of one of four insectoid super-radiations that give rise to most of the animal diversity on Earth. Since the development of molecular phylogenetics, studies have attempted to reconstruct the phylogenetic relationships within this order. However, a comprehensive next generation phylogeny spanning the entire breadth of this group is yet to be published. Questions remain regarding Dipteran relationships including, but not limited to, resolving the earliest diverging lineages, the placement of enigmatic parasitic families and the higher-level relationships within the acalyprates. Advancing our understanding of the relationships within this highly diverse order would provide a critical foundation for further investigations into Dipteran evolution, development and genome. We harvest publicly available whole genome, transcriptome, ultra-conserved element, and targeted enrichment data to assemble a dataset comprised of over 130 families and 250+ subfamilies. This data was processed using SAPPHYRE, a newly developed bioinformatics pipeline. Using SAPPHYRE, we resolve relationships previously obscured by multiple episodes of rapid diversification. SAPPHYRE eschews traditional assemblers in favor of mapping reads directly against references. Using data from Dietz et al. 2023 we compare the output of SAPPHYRE against PHYLUCe, HybPiper, Orthograph, BWA mapping, IBA, and the combined approaches of Orthograph + Phyluce and Orthograph + Phyluce + Trinity. We show SAPPHYRE averages between 1.5x to 14x more data recovery and 1.7x to 20x the number of parsimoniously informative characters than the next most performant and least performant solutions.

Resolving the relationships of Eristalinae (Diptera: Syrphidae) using exon-capture sequencing

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Keywords: biogeography, flower flies, phylogeny, target DNA enrichment

Exon-capture sequencing, in combination with the dataset preparation pipeline SAPPHYRE, cuts through the stubborn knot represented by the paraphyletic subfamily Eristalinae (Diptera: Syrphidae) to reveal a new hypothesis of Syrphidae relationships. The study utilizes probes developed specifically for use with Syrphidae and samples 1291 orthologous genes. 136 of 145 described genera within Eristalinae are sampled, with the use of pinned museum specimens allowing for inclusion of rarer genera, for a total sampling of more than 300 taxa. Eristalinae is revealed as a paraphyletic assemblage of four monophyletic clades. Furthermore, multiple independent Australian-Chilean relationships are revealed, raising the question of whether Syrphidae underwent Trans-Antarctic dispersal or Gondwanan vicariance.

Phylogenomic reconstructions of meniscus midges (Dixidae) and black flies (Simuliidae) inferred from orthologs recovered from Illumina libraries using the aTRAM2 pipeline

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Keywords: aquatic, Culicomorpha, Nematocera, phylogeny

Supraspecific relationships within two culicomorphan families, Dixidae and Simuliidae, are largely unknown and under-resolved, respectively, and sorely in need of study using a phylogenomic approach. Toward this goal relationships within both families were reconstructed from data sets comprised of more than two thousand orthologous nuclear genes extracted from Illumina draft genomes using aTRAM2 and its associated pipeline and analyzed phylogenetically using maximum likelihood criteria with IQ Tree2. Included taxa in these works-in-progress include multiple relevant outgroup and ingroup taxa, the latter representing a significant proportion of suspected or known generic diversity. Inferred trees for both families are strongly supported throughout and form the underpinning used to inform both revisionary and homology studies aimed at establishing evolutionarily informed supraspecific classifications and enabling parallel studies of relative utility of genes to biogeographical and evolutionary time-scale analyses. Finally, an interesting although extremely concerning pitfall of using this pipeline as originally conceived and how the issue was resolved are discussed.

**Systematic revision of the *philadelphicus* group
in the genus *Proctacanthus* (Macquart) (Asilidae)**

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Keywords: morphology, Nearctic, phylogenetics, revision, taxonomy

We will present a systematic revision of the robber flies in the Asilinae genus *Proctacanthus* Macquart, with a special focus on the *philadelphicus* group. *Proctacanthus* are large-bodied and highly noticeable flies, often collected and photographed by the lay-person as well as the expert. Many of these flies lie unidentified to species in museum collections, making their use in behavioral and ecological studies difficult. In this talk we outline the parts of our revision of the widespread *philadelphicus* group, including the descriptions of several new species, and highlight notes on the geography, morphology, and phylogeny of these flies. We will particularly use a variety of illustrations and photographs to aid in the identification of members of the group based on mouthparts, overall coloration, and details of both female and male terminalia. All these will be included in the most comprehensive keys and phylogenies of the genus ever presented.

**Niche-modelling for Afrotropical hover flies
(Diptera, Syrphidae): a case study using
the invasive species *Toxomerus floralis***

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Keywords: Afrotropical, invasive species, pollinator, species distribution modeling

Toxomerus floralis (Fabricius, 1798) is indigenous to the Nearctic and Neotropical Regions. The larvae are pollen-feeding and closely associated with the invasive plants *Mitracarpus hirtus* (L.) (Rubiaceae), a geographic contemporary, and the much more widely spread *Cyperus rotundus* (L.) (Cyperaceae). *Toxomerus floralis* was detected in 2013 for the first time in the Afrotropical Region, and has since spread swiftly through large parts of the Afrotropical Region. The potential species distribution of *T. floralis* was modelled using MaxEnt and Worldclim Bioclimatic variables. The Bioclimatic variables were tested for collinearity using Variance Inflation Factor in the R package “usdm”. This identified 9 correlated variables which were removed to improve upon estimations and to more accurately determine potential environmental variables that could have an influence on the species’ distribution. The resulting models suggests that the distribution of *T. floralis* is potentially associated with the distribution of its host plant, *M. hirtus*, and also with *C. rotundus*, which is not native to its home range but does occur in almost all invaded areas.

Genus *Ornithoica* Rondani in Hippoboscidae evolution

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Keywords: attachment apparatus, evolution, Ornithomyiinae, paraphyletic

Hippoboscidae – a specialized group of blood-sucking ectoparasites – is divided into three subfamilies: Ornithomyiinae, Lipopteninae and Hippoboscinae. The representatives of Hippoboscidae spend all or most of their lives in the fur or among the feathers of their hosts. These flies have a large number of unique morphological and physiological adaptations, that are closely related to their ectoparasitic life. Of particular importance are those that are associated with reliable attachment and efficient movement on the host. The morphology of their attachment apparatus is associated with the evolution of this group. Our analyze of the phylogenetic relationships in Hippoboscidae shown that the genus *Ornithoica* Rondani, which is part of the subfamily Ornithomyiinae, emerged earlier than other large groups of louse flies and formed a separate cluster. And the rest of the genera from Ornithomyiinae are the latest. According to the literature, the genus *Ornithoica* species have many plesiomorphies, such as the presence of seven developed tergites on the abdomen of females and six on the abdomen of males, while in the rest of the louse flies their reduction occurs, weakly developed humeral calli, the presence of a well-developed notopleural suture, a short and simple frons, completely developed ocelli, a concealed interantennal area etc.. This genus has even been proposed to be classified as a separate taxon Ornithoicinae. We noted the following features of the attachment apparatus in representatives of this genus: the claws are simple, like those of mammalian parasites, while the empodium is covered with long identical setae and transverse folds, which are not found in representatives of other genera. Thus, we believe that the tribe Ornithomyini is a paraphyletic group. The genus *Ornithoica* can be considered as a separate line of evolution.

**Louse fly *Ornithoica momiyamai* Kishida
(Diptera: Hippoboscidae): morphological and molecular data**

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Keywords: evolution, Ornithomyiinae, paraphyletic, species status

Species status of *O. momiyamai* Kishida described from Japan has been controversial for many years. Female specimens of the species were collected from two birds named only common Japanese names and the type locality not given. The type probably lost. Bequaert synonymized *O. momiyamai* with *O. turdi* Latreille and Maa – with *O. exilis* Walker and in 1963 – with *O. stipituri* Schiner. Maa did not included *O. momiyamai* in the list and in the key of species in his revision of the genus *Ornithoica*. Maa wrote his new species *O. tridens* from Taiwan might be identical with *O. momiyamai*. Maa treated *O. momiyamai* as a valid species in his review of the Japanese species. Soós listed *O. momiyamai* as a synonym of *O. stipituri*. Maa treated *O. tridens* as a synonym of *O. stipituri* in the Oriental catalog. Iwasa synonymized *O. momiyamai* and *O. tridens*. Mogi with colleagues published a special paper on identity of *O. momiyamai* and illustrated the three-lobed structure of infra- anal plate of its gynandromorph. We investigated the morphology of, supposedly, *O. momiyamai* males and females from Primorsk Territory, Russia and southern Vietnam and found only insignificant differences. The genetic distance between the sequences of Vietnamese samples was 0.2%, between the samples from Primorsk Territory, Russia – 0.3%, between the samples from two geographic populations from 4.4 to 4.7%, which may correspond to the subspecies. Than *O. bistativa* Maa and *O. momiyamai* Kishida may be treated as subspecies. Maa considered *O. bistativa* as a subspecies of *O. stipituri*. To solve the problem with the identity of all these species it is necessary to receive molecular data of *O. stipituri* from Australia or New Guinea.

Arbovirus modification of midge behavior and considerations for vector surveillance and management

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Keywords: behavior, biting midge, virus, vision

Culicoides biting midges (Diptera: Ceratopogonidae) are important biological vectors of arboviruses such as bluetongue virus (BTV), epizootic hemorrhagic disease virus (EHDV) and vesicular stomatitis virus (VSV), resulting in significant economic losses worldwide. Previous studies have shown that during the course of dissemination to the salivary glands, these viruses highly infect ommatidia, cerebral ganglia (brain), and other sensory tissues in the midge. Our group has shown that many genes associated with vision, memory, and other behaviors were downregulated in midges infected with EHDV. We surmised that infection of the brain and ommatidia might lead to a phenotype of visual impairment and changes in light-seeking behavior. In subsequent studies we tested the visual performances and light-seeking preferences of midges that were infected arboviruses. Female *Culicoides sonorensis* were infected with either BTV, VSV, or control (PBS) via microinjection, and at 6 days post-injection were placed in the center of a circular aerial bioassay arena with LEDs (ultraviolet (350-400 nm), blue (400-500 nm), and green light (500-570 nm)) mounted to collection cups along the periphery. Midges were exposed for 12 h and spectral preferences were determined by counting midges in the cups and elsewhere in the arena. Taken together, these studies will give insight into the effects of arbovirus infection on midge visual performance which will improve our ability to trap and survey infected midge populations in the field.

A new phylogeny of true fruit flies (Diptera: Tephritidae) based on nuclear loci obtained by phylogenomic methods

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Keywords: fruit flies, phylogeny

True fruit flies (Tephritidae) are one of the largest and most economically important families of Diptera, with approximately 5000 recognized species, including dozens of species that are significant pests of commercial and subsistence fruit crops or beneficial biological control agents of weeds. Tephritid higher classification has changed drastically over the last 30-40 years, largely based on increased study of genitalic characters and adoption of phylogenetic methods. Despite these improvements, major questions remain regarding the monophyly of and relationships among many higher taxa and even the extent of the family (e.g., status of subfamily Tachiniscinae). Molecular phylogenetic analysis across the family has been very limited. To address this problem our international team of collaborators is investigating tephritid phylogeny by analyzing sequences of nuclear loci obtained by anchored hybrid enrichment (AHE) and transcriptome sequencing. We are attempting to sample as many genera as possible across the Tephritidae and related families of Tephritoidea. Samples of nearly 200 of the 500 genera of the family, representing all seven subfamilies and 25 of the 31 tribes are included, contributed by 14 scientists in nine countries. Progress is discussed and preliminary results are presented.

Catalogue of the Tachinidae (Insecta: Diptera) of the world

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Keywords: catalogue, classification, distribution, nomenclature, Tachinidae, world

The Tachinidae are one of the largest families of true flies (Diptera) with a species diversity that cannot be accurately estimated even to the nearest 10,000. The number of valid species names is approximately 8900, but undescribed species might exceed this number by two, three or more times based on preliminary data from surveys in biologically diverse parts of the world and caterpillar rearing programs such as those in Area de Conservación Guanacaste, Costa Rica and Yanayacu Biological Station, Ecuador. The present authors and the late D.M. Wood initiated a project about 15 years ago to prepare a comprehensive catalogue of the world Tachinidae to replace aging regional catalogues and to introduce a unified classification for the family. We are getting close to finishing this goal and have completed data entry for all generic and specific names (ca. 17,400), species distributions, and most of the name-bearing types (including sex(es), type locality(-ities) and depository(-ies)). The database can be queried to produce outputs of various sorts including catalogues/checklists by country or region, species lists by author, and name-bearing type lists by author, depository, country, and/or type locality. Two checklists of world Tachinidae have been prepared to date (2019, 2020) and a new one is planned for late 2023. The new checklist will include a hierarchical classification of the family with all valid generic and specific names, synonyms, species distributions, and references. Name-bearing type data, misidentifications, misspellings, and nomenclatural and other notes will be included in the world catalogue.

Evaluating aversion behaviors to develop a standardized method of evaluating fly worry and fly control product efficacy in horses

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Keywords: horse, machine learning, pyrethroid, stable fly

Stable flies (*Stomoxys calcitrans*) are obligate blood feeding muscid flies which feed at least once a day. Horses are a preferred host and are frequently parasitized with bites causing significant pain and irritation. Aversion responses, also called fly worry, include leg stamping, head and body shaking, tail swishing and skin twitching. Under high fly pressure, horses attempting to escape parasitism can injure themselves or others. On-horse fly control is primarily achieved through the use of chemical fly sprays and/or physical barriers like fly sheets. Horse owners often select products without the ability to evaluate the efficacy of the product. This has resulted in an overuse of pyrethroid based fly sprays due to the low cost and perceived efficacy. This study aimed to evaluate the efficacy of physical barriers (fly sheet, mask, and leg coverings), pyrethroid-based spray, and bio-based spray composed of plant essential oils in horses. Horses were evaluated for fly worry behavior (skin twitch, leg stamp, tail swish, and head movement) before and after intervention by counting the number of fly worry events. Physical barriers were more effective than sprays in reducing the number and intensity of fly worry events with bio-based spray outperforming pyrethroid based spray. Using this data and a machine-learning approach, a standardized method and platform to evaluate biting fly protection will be developed enabling horse owners to select the most effective intervention strategy for their horse and area.

Diptera surveys in the Cerrado, the Brazilian savannah: updates and first results

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Keywords: conservation, diversity, inventories, occurrence, surveys, taxonomy

The Brazilian savannah, known as Cerrado, is the most threatened landscape in South America. Every year, natural areas are converted into open areas for livestock rearing and agriculture. Diptera surveys in the core area of the Cerrado, in the Goiás state, were not realized so far. Some expeditions occurred in the 1950 decade, but there is no continuity in the subsequent years, only punctual records. Since 2015, Diptera surveys have been performed in the conservation reserves of Cerrado, in the state of Goiás. The Emas National Park (ENP), the biggest Cerrado reserve, was sampled with four malaise traps, in three different landscapes, thought three years, with monthly samples. Almost 180 thousand insects were collected, of which 158.834 are dipterans, which means 88% of the collected specimens. A total of 57 Diptera families were sampled, with five new families' records from the state of Goiás. The diversity of species is expressive. In Dolichopodidae, for instance, only one species were known from the Cerrado in the state of Goiás so far. In ENP were collected 4.147 specimens in 28 genera and 55 species. Besides the study done in ENP, four additional conservation areas have been sampled, also with malaise traps: the Legados Verdes do Cerrado reserve, the Sylvania National Forest (FLONA), the Parque Estadual dos Pirineus (PEP), and the Altamiro de Moura Pacheco (PEAMP). The data provided by the inventories are being used in taxonomic studies, including systematic and biogeography, as well as by the SEMAD (Secretaria Estadual de Meio-ambiente e Desenvolvimento) to organize the conservation list from the state of Goias. This is the first conservation list in Brazil with Diptera data. The list is being organized, with almost 500 Diptera species included so far, all of them categorized as low, medium, and high endangered.

Mycetophilidae from the Cerrado, the Brazilian savannah

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Keywords: diversity, fungus gnats, inventories, occurrence, Sciaroidea, taxonomy

The Brazilian savannah, known as Cerrado, is the most threatened landscape in South America, being converted to livestock rearing and agriculture quickly. The Mycetophilidae data from the state of Goiás, a core area of the Cerrado, is scarce. Some expeditions occurred in the 1950 decade, performed by Lane and Coher, and some punctual records were add posteriorly. From this period, a total of 13 genera and 25 species of Mycetophilidae, mainly into the genera *Neoempheria* Osten Sacken and *Leia* Meigen, are known from the region. Since 2015, Diptera surveys have been performed in the conservation reserves of Cerrado, in the state of Goiás. The Emas National Park (ENP), the biggest Cerrado reserve, was sampled monthly with four malaise traps, in three different landscapes, thought three years. One of these malaises, installed in a semideciduous forest, collected 21.535 specimens of Mycetophilidae. Regarding the previous genera known from the state of Goiás, were collected 28 species. Additionally, where collected species from 12 genera are still not recorded from the state, with the identification of 10 species of Sciophilinae. Hence, the fauna of the core area of Cerrado in Brazil encompasses 25 genera and 38 species, plus the new species not yet described. The diversity unknown so far is impressive. We highlight the genus *Monoclona* Mik as an example. This is the first record of the genus from the Cerrado and was find one species known, *M. maculata* Edwards, plus five new species. Besides, the study done in ENP, we are performing Diptera surveys in four additional conservation areas. The data provided by the inventories are being used in taxonomic studies, including systematic and biogeography, and we hope to have a wide view of the Mycetophilidae distribution in the Cerrado for future studies and public policies.

Insecticidal activity of *Dysphania ambrosioides* (Amaranthaceae) essential oil against house fly (*Musca domestica*) (Muscidae)

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Keywords: essential oils, insecticidal activity, house fly

The house fly (*Musca domestica* L., Diptera: Muscidae) is worldwide known as a key pest in urban and agricultural environments. Its main control method are synthetic insecticides sprays, even so, resistance in this species is a growing problem with more than 80 cases worldwide. In Chile, reports of resistance in general are scarce and given the need to seek control alternatives, the objective of this research was to assess under laboratory conditions the insecticidal activity of the essential oil from paico (*Dysphania ambrosioides*) (Amaranthaceae) and to compare its effectiveness with the synthetic insecticides pirimiphos methyl and cypermethrin, and natural ones such as pyrethrins, D-limonene and azadirachtin. Toxicity bioassays were carried out on larvae and adults, obtaining that the commercial doses of synthetic insecticides and pyrethrins eliminated at least 70% of the adults with the commercial dose, while the insecticides derived from D-limonene, azadirachtin and *D. ambrosioides* essential oil at 1.0% had a mortality lower than 6%. Conversely, all commercial insecticides exhibited a mortality lower than 40% in larvae, except for pyrethrins and paico essential oil diluted to 2.0%, where a mortality of 61 and 100% was registered respectively. In general, it was observed that the commercial alternatives had powerful insecticidal activity against adults from the assessed population, however, a high tolerance in larvae is reported. The results with the essential oil suggest that this compound has the potential to be used for control in larvae. However, it is necessary to study further its activity as a formulated product, identify its main active chemical components and carry out trials under field conditions.

Latitudinal variation in survival and immature development of *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae) populations reared in two key overwintering hosts

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Keywords: latitudinal variation, life history, medfly, overwintering hosts, survival rates

The Mediterranean fruit fly, *Ceratitis capitata* (medfly), is a major agricultural pest, expanding its geographic distribution to cooler, more temperate areas of Europe. In coastal Mediterranean regions, high population densities during summer and autumn and overwintering are accomplished mainly in bitter oranges as larvae. In cooler, more temperate areas, peak population densities occur in autumn with apples as the overwintering host of larvae. Following a common garden experimental approach, we assessed the overwintering capacity of different medfly populations obtained from a wide latitudinal range of the northern hemisphere. Survival and development of immature stages were assessed in two overwintering hosts (apples, bitter oranges) under laboratory conditions. Eggs from six populations, spanning a latitudinal range, from Southern Europe (Crete, Greece) to Central Europe (Vienna, Austria), were artificially inserted in the flesh of the two hosts. Infested fruits were kept at 15, 20, and 25°C, and relative humidity 60±5%, until pupation and adult emergence. Analysis revealed combined effects of origin-latitude, host, temperature, population and microclimatic conditions on survival rates and developmental duration of immatures. Immature stages exhibited higher survival rates and prolonged developmental duration when reared in apples compared to bitter oranges. In medflies originating from southern latitudes (warmer areas), developmental duration was longer than those originating from northern regions, demonstrating differential plastic responses to constant temperatures. Lower temperatures decreased adult emergence rate and delayed developmental duration regardless of the host and population. Our results contribute to understanding invasion dynamics of medfly, deciphering biological traits that influence invasion success, with emphasis on range expansion to northern, more temperate areas of Europe. Funding was provided by projects FF-IPM (HORIZON 2020, GA818184) and the MEDFLY (HFRI-FM 17-4289).

Adults of geographically distant populations of *Ceratitis capitata* (Wiedemann) (Tephritidae) respond differently to thermal stress depending on age, acclimation and mating status

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Keywords: age, CTmax, CTmin, mating, thermal acclimation, thermal biology

The Mediterranean fruit fly (medfly), *Ceratitis capitata* (Wiedemann) (Diptera: Tephritidae), is a highly invasive pest of fresh fruits and vegetables that is currently expanding its geographic ranges to cooler temperate areas of the Northern Hemisphere. The invasion potential of medfly depends on its intrinsic ability to tolerate the thermal stress induced by environmental temperature shifts in the invaded regions. Insects use physiological adjustments (i.e., acclimation) to achieve thermal tolerance that is influenced by several factors such as age, mating status and thermal history. The effect of acclimation and thermal history has been studied extensively, unlike age and mating status, or their combination. Here, we assessed the Critical Thermal Limits (CTmin, CTmax) of three geographically distant medfly populations from Greece (Crete, Volos) and Croatia (Dubrovnik). We tested adults (a) at the ages of 1, 6, 15, and 35 days old, without acclimation, (b) 6 and 35 days old (acclimated at 15, 25, and 30°C), and (c) mated and virgin. Age significantly affects CTmin and CTmax of non-acclimated flies. CTmin differs among populations, and older individuals are more heat tolerant. Furthermore, acclimation at lower and higher temperatures increased both cold and heat tolerance, respectively, at younger ages for all populations, but its effect is not apparent at older ages. Finally, the mating status of females is related to both cold and heat tolerance and it seems it is related to population as well. Our results contribute towards understanding the invasion dynamics of medfly and the biological traits that are related to success of establishment in northern, more temperate regions of Europe. The study was funded by the project MEDFLY (HFRI-FM 17-4289).

Calibrated calyptrate classification

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Keywords: Calyptratae, classification, Hippoboscoidea, muscoid grade, Oestroidea

Calyptrate phylogeny has seen major progress during the last two decades, and with an increasingly robust family-level topology driven by large amounts of molecular data, it is time to calibrate the classification. The position of Hippoboscoidea as a basal calyptrate lineage is solid, and the division into two families, Glossinidae and Hippoboscidae, has much to recommend it. The concept of a non-monophyletic ‘muscoid grade’ is growing in acceptance, and apart from uncertainty relating to the first few splits at the base of the Anthomyiidae–Scathophagidae clade, families are well-supported, and their emerging internal phylogenetic resolution provides the first hints at a realistic subfamily classification. Oestroidea are undoubtedly monophyletic, and although the exact position of each of the mono-specific families Mystacinobiidae and Ulurumiidae is not yet fully settled, family-level phylogeny within the superfamily has finally achieved sufficient support to allow blow fly classification to rest on robust monophyly. A subfamily classification for blow flies is now up for scrutiny. Botflies were long suspected as derived blow flies, but this is rejected by recent analyses, while molecular and morphological data disagree on botfly monophyly. Polleniidae classification has shifted from being a blow fly subfamily to being sister to Tachinidae. Rhinophorinae, now in Calliphoridae with a surprising position as sister to the macrolarviparous Ameniinae, are now ready for a tribal classification. Rhiniinae have also returned to the Calliphoridae, with both molecular and morphological evidence corroborating Bengaliinae + Rhiniinae as a clade dominated by termite-associated species. The traditional classification of Tachinidae into four subfamilies has largely stood the test of molecular data, with only minor modification needed if Macquartini + Myiophasiini is corroborated as a basal tachinid branch.

Biogeographical history of genus of *Robsonomyia* Matile (Keroplastidae)

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Keywords: amber, biogeography, first record, fossil

During examination of the Baltic amber inclusions belonging to the Natural History Museum in Copenhagen, flies of the genus previously unrecorded from fossil materials have been found. This newly discovered species belongs to the genus *Robsonomyia* Matile belonged to the family Keroplastidae. This family of nematoceros flies is one of the largest in the infraorder Bibionomorpha and gathered nearly 1,000 living species and 53 fossil ones. The oldest representative of the family was found in the Lower Cretaceous. They appear in various types of fossil materials; however, the largest number of species come from Baltic amber, which is dated on the middle Eocene. So far, we have known 19 species from 10 genera from this fossil resin. Right now, we can add not only the 20th species, but also the 11th genera to this list. Moreover, this is the first species of *Robsonomyia* recorded on the European continent. Until now, this genus was represented by only two extant living species: *R. reducta* Matile & Vockeroth known from North America (USA) and *R. sciaraeiformis* (Okada) found only in Asia (Japan). Therefore, this discovery gives us new information about the biogeographical history of that genus and suggests that it was widely distributed throughout the Holarctic region one day.

Radiations within radiations: phylogenomics and morphological evolution of the tachinid tribe Polideini (Diptera: Tachinidae)

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Keywords: Andes, biodiversity, bristle fly, convergent evolution, homoplasy, Neotropical, UCE

Polideini comprise a relatively small, eclectic, and rapidly diversifying tribe of Tachinidae (Bristle flies) in the subfamily Tachininae. This mostly New World tribe was assembled by O'Hara in 2002, in which he drew diverse taxa from nine tribes and multiple subfamilies together into one well-supported tribe. This important work also revised the North American genera of Polideini, however, the diverse Neotropical fauna was left largely unstudied. Here, we report on a phylogenomic analysis of the tribe using UCEs with focus on Neotropical taxa, including 59 taxa belonging to approximately 36 genera. Trees inferred via ML analysis of 676 (average) protein-coding UCE loci distribute the included taxa into 11 major clades, with most nodes being robustly supported. However, backbone nodes are very short, indicating an extremely rapid radiation of lineages, possibly associated with Andean orogeny. The tribe likely originated and diversified in South America, with multiple colonizations and one major radiation in the Nearctic. Each of the 11 major clades is morphologically diverse in itself, with limited external similarity of many contained taxa, although male terminalic traits support some relationships. Rampant intra-clade morphological disparity and convergent evolution plagues the phylogeny, with “hedge-hog body forms”, bright coloration, setal patterns and “sarcophagiform” body habitus evolving repeatedly in different lineages. This morphological confusion makes it difficult to determine the affinities of unsequenced taxa. Hosts are unknown for most taxa, but the ancestral host was likely lepidopteran caterpillars and several lineages have colonized unusual host groups. This analysis provides a first step towards understanding the diversity and evolution of the Polideini. However, the majority of species and genera of Neotropical Polideini remain undescribed and unknown, and those included here likely represent just the “tip of the iceberg” of Neotropical Polideini diversity.

Diversity of *Eloeophila* crane flies (Diptera: Limoniidae)

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Keywords: dimorphism, diversity, fossils, Limnophilinae, morphology, new species, phylogeny, taxonomy

Eloeophila Rondani, 1856 (Diptera, Limoniidae) is a medium-sized genus of crane flies with a total of 85 described extant species worldwide. Within the genus the most diverse is the Palearctic fauna with 33 species. Three fossil species are described from the Eocene period. Four undescribed species were discovered during our studies from South Korea, India, Malaysia and Western United States. The most obvious feature of *Eloeophila*, making it easily recognizable among other crane flies, is the supernumerary cross-vein in cell bm. This character occurs also in other Limnophilinae crane flies, it was considered as a synapomorphy shared between the genera *Eloeophila* and *Idioptera* by Ribeiro (2008), but this additional cross-vein is also known in families Tipulidae and Pediciidae. Preimaginal stages are unknown for most species. Larvae are reported to live in mud beside streams or in moist soil, some are more often found at seepages. This is the first attempt to gather all possible information on this genus and to document type specimens located in different museums across the world.

How a deep look into the morphology of crane fly larvae helps to solve taxonomic problems

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Keywords: crane flies, integrative systematics, larvae, morphology

Superfamily Tipuloidea, or crane flies, is one of the largest groups of Diptera. It includes about 10 percent of all species of the order. The immature stages of crane flies are still very poorly known. Less than half genera have described larvae, and only few percent of the species have described larvae or pupae. Crane flies larvae have successfully exploited nearly every kind of habitat so it is not surprising that they exhibit a great diversity of morphological characters - from huge variety of head capsules to a multitude of different types of spiracular discs - like no other group of diptera. The taxonomy of crane flies is mainly based on adult characters, but an integrated taxonomic approach, including not only adult but also immature characters and molecular markers, may change the current taxonomy of this large group. Recent comprehensive studies of the genus *Hexatoma* have revealed that *Eriocera*, the largest subgenus of *Hexatoma*, is not monophyletic, a hypothesis supported by both morphological and genetic characters of the larvae. The set of unusual larval characters of the newly described genus from subfamily Chioneinae was the first hint that science might have a new genus. The genera *Geranomyia* and *Dicranomyia* have good distinguishing adult characters, but their larval characters overlap. Phylogenetic analysis of the mitochondrial COI fragment (DNA barcode) shows that *Geranomyia* and *Dicranomyia* species are intermixed in the phylogenetic tree, which reveals unclear phylogenetic relationships between the two genera.

Review of the Dolichopodidae (Diptera) of the Netherlands: diversity, status and the significance of citizen scientist data

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Keywords: checklist, citizen science, first record, GBIF, Netherlands, new introductions, opportunistic species

In 2021 a NLBIF project was started to assemble all data on Dolichopodidae of the Netherlands in order to generate a dataset for publication to GBIF (Global Biodiversity Information Facility). Both nearly 17,000 historical data and about 8,500 new identification records as well as over 22,000 observations by citizen scientists were processed. Ultimately, about 97% and 93% of the historical and new identifications resp. were considered useable, while this applied for only 51% of the citizen science data. The final dataset consists of nearly 35,500 species records. As compared to the most recent checklist with 246 certain and 2 doubtful occurrences (Meuffels, 2002) and Fauna Europaea with 250 certain and 1 doubtful occurrences (Pollet, 2011), our research confirmed the presence of 268 species. While 12 species have been removed from the previous checklist, first records of 23 species could be added. The new checklist also includes three species of uncertain identity. As part of an atlas of Dolichopodidae of the Netherlands, for each species a brief data sheet will be generated with information on distribution, phenology, ecology and current rarity. Our research revealed that, surprisingly, a few species like *Sciapus pallens* (Wiedemann) and *Hercostomus rusticus* (Loew) seem to favour urban environments. Some of them were even discovered indoors during winter which suggests man-induced introduction. On the other hand, the southern European *Campsicnemus simplicissimus* Strobl appears to enter the country from the southwest (Belgium), where it has recently also been discovered for the first time. Its introduction might have been facilitated by global warming. The usefulness of citizen science data for the establishment of distribution ranges and current rarity is discussed.

The Our Planet Reviewed Corsica 2019-2021 expedition: building and sharing knowledge on Diptera diversity

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Keywords: Corsica, Dolichopodidae, ecology, endemics, expedition, expert network,
pan traps, sampling protocol

The Our Planet Reviewed Program currently run by the Muséum national d'Histoire naturelle (Paris, France) aims at discovering unknown biodiversity in remote sites around the globe and sharing this information publicly. The most recent edition was conducted in Corsica focusing on undersampled regions in the southern mountains (2019) and coastal areas in the north (2020) and southeast (2021). Thirty-four experts were involved in the field campaign and over 80 in the identification of specimens. As in previous editions, the terrestrial mission employed a vast array of sampling methods to collect invertebrates. In 2019 and 2021, 16 sampling sites in 4 different locations were selected for pan trap sampling. In each site, 15 pan traps (5 blue, 5 yellow and 5 white) were in operation on soil surface level for mostly four days. The survey produced 876 Diptera samples that were ultimately split into more than 4,300 workable taxonomic subfractions. Thirty-seven dipteran families were pulled from the samples and disseminated to a network of taxonomic experts. A number of specimens from various families was used for barcoding. During the expedition special attention was drawn to Dolichopodidae. With 62 species recorded prior to the expedition, the dolichopodid fauna of the island was considered rather well known. Nearly 26,500 specimens of 115 different species were retrieved from the 477 samples, including 10 new species to science, 2 possible subspecies and 6 species of doubtful identity. Only 31 of the previously recorded species was rediscovered, while 84 species were recorded from Corsica for the first time. Similar assessments were made for the other fly families as well. The current status of the Corsican Diptera fauna and the impact of this kind of surveys on the knowledge of its invertebrate fauna is discussed.

Oak and chestnut forest in Canton of Ticino (southern Switzerland): biodiversity and sampling protocol

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Keywords: biodiversity, forest, sampling protocol, surveys, Switzerland

In the outcome of the first biodiversity research campaign of *Querco-castagnetum* forest carried out in Canton of Ticino, southern Switzerland, a new sampling plan was set up, with the aim of establishing a protocol to be applied to a broader research project. Three stations were still sampled, but this time not in close proximity but in three different valleys; one of the stations already investigated in the first year was nevertheless retained. In terms of methodology, two of the three attractant traps were removed, as the results obtained were comparable, while two different interception traps were added. Each sampling station, in operation from July 2017 to July 2018, was composed as follows: Malaise trap, aerial Malaise trap (SLAM trap), window trap (Polytrap), “Kombifalle”, a trap which combines window trap and coloured pan, and an attractant trap with wine. Eighteen taxonomic groups were sorted, and Diptera, representing 60% of the individuals, were identified to the family level. Of the 66 families found, 27 (40% of the captured Diptera) were identified to the species level, resulting in a total of 155 species. Whilst both methods, attractant and interception, show a peculiarity in the community of species caught, the difference between the three stations tested by multivariate analysis was not significant. The difference tested between the two years, evaluated for the one station surveyed during both sampling campaigns, also was not significant. In conclusion, the methodology resulted to be the most important factor affecting Diptera species community, showing that to maximize diversity it is advisable to invest in a variety of trapping methods at one station, representative of the forest typology, and to sample for an entire year.

Book launch event

Official launch of Volume 3 of the *Manual of Afrotropical Diptera*

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Oxford University Museum of Natural History, Parks Road, Oxford OX1 3PW,
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Keywords: Afrotropical, book launch, exploration history, publication history

A few reminiscences are given about dipterological publishing and African dipterology as a prelude to welcoming volume 3 of the *Manual of Afrotropical Diptera*, which continues and confirms the tradition of excellence established by the first two volumes.

Computer-based identification of immature forms of Neotropical blow flies (Diptera, Calliphoridae)

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Keywords: forensic entomology, identification, interactive key, larvae, morphology, Neotropical, taxonomy

Approximately 70 species of Calliphoridae are recorded in the Neotropics. Although various feeding habits have been reported among blow flies, the necrophagy is dominant. For this reason, biological, behavioral, and distributional data are often used to provide useful information about the postmortem interval. Identifying forensic importance species is not always easy, particularly the immature forms whose inter and intraspecific variability can be low and high, respectively, in addition to other aspects such as scarcity of descriptions and taxonomic keys. With the aim of reducing the impediment and making the Neotropical Calliphoridae immature taxonomy more accessible to experts, researchers and students, we chose to build computer-based interactive keys for first- and third-instar larvae of forensic importance. To obtain larvae, adult individuals attracted by putrefied baits were actively collected in the field. In the laboratory, identified females were kept in cages, under controlled conditions ($27\pm 1^{\circ}\text{C}$, $70\pm 10\%$ RH, 12 h of photoperiod), to stimulate oviposition. Immatures were reared on ground beef until they reached the first and third instars. Examination of internal and external anatomical characters and acquisition of images were performed under stereomicroscope. An array of characters and their states has been included in the LUCID v. 3.6 for preparing the key. The following were listed as diagnostic characters: (i) internal: presence, shape, size and pigmentation of cephaloskeleton structures; and (ii) external: oral ridges, distribution of spines on the body, distribution and size of papillae in the anal division, presence of buttons and peritreme shape. Our interactive key offers an intuitive approach and numerous features, as well as an extensive image bank, glossary, and instructions and tips for using the key. Keys with this interactive approach to Neotropical blow fly immatures are unpublished. We hope that the keys can provide support for more secure and reliable identifications of immatures.

Exon-capture sequencing reveals a new hypothesis on Microdontinae relationships

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Keywords: ant flies, phylogeny, Syrphidae, target DNA enrichment

The subfamily Microdontinae is a cosmopolitan group of ant-associated hover flies. Phylogenetic relationships of the more than 500 described species are unclear. Until recently, more than 300 species were classified in *Microdon*. Reemer & Ståhls (2013) published the first phylogenetic study dealing specifically with Microdontinae. Yet many relationships remained uncertain or unresolved, in part because many taxa were unavailable for molecular study. In the present work, exon-capture sequencing was used to obtain DNA from pinned museum specimens, which allowed for the inclusion of rarer taxa. Our phylogenetic study is based on baits developed specifically for Syrphidae, which target 1302 orthologous genes. A total of 91 taxa, including specimens from all six biotic regions, were successfully sequenced. A new hypothesis on Microdontinae relationships is presented with the resulting tree compared to those that came before. Paraphyly in the tree is discussed. The potential implications of our results for the classification of microdontines, interesting biogeographic patterns, and evolutionary history are also discussed.

Local ecosystem and abiotic variable impact on carrion fly communities

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Keywords: abiotic variables, Calliphoridae, Fanniidae, forensic entomology, local ecosystems, Muscidae, Sarcophagidae

For forensic entomologists to use carrion flies as an accurate indicator of how long a decedent has been dead, they must have a comprehensive understanding of the local carrion fly community and how local ecosystem conditions affect fly behavior and distribution. Given plants are significant indicators of the conditions and resources available in an ecosystem, I identified five different local ecosystems, as defined by their plant group classification, and examined the carrion flies present in each ecosystem by setting out baited traps at the beginning, middle, and end of summer. I ran a Multiple Analysis of Variance and found no significant relationship between the plant group ecosystem and the flies captured in that ecosystem. Then, I used Poisson regressions to model the effects of abiotic variables I measured at each trap site on different families of carrion flies. Calliphoridae, Muscidae, and Fanniidae showed a statistically significant relationship with humidity and the distance from water. Calliphoridae also showed a relationship with the ambient temperature, and Fanniidae had a significant relationship with wind speed. Future studies should focus on the impact of local abiotic variables on carrion fly communities so entomologists can make accurate contributions to forensic investigations.

Syrph-ing habitats: investigating the impact of agricultural landscape composition on native flower fly populations in Southern Ontario

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Keywords: agricultural landscape, flower fly, habitat, hover flies, pollinator conservation, Syrphidae

Native pollinator diversity provides significant pollination services in many agroecosystems in Ontario and globally, with estimated economic valuations running to hundreds of billions of dollars. Habitat studies are lacking in Canada but are critical to understanding species-specific niches, as well as pollinator abundance and diversity patterns in various landscape compositions. As such, there is a critical need for more research investigating the nesting and foraging requirements of these native species. Our objectives are to assess differences in pollinator diversity (restricted to wild bees (Apoidea) and flower flies (Syrphidae)) and abundance among three habitat types (forest, field margin and restored prairie grass) on agricultural land and to understand how biodiversity is affected by habitat type, patch size and connectivity. We conducted two field seasons, deploying three Malaise traps per farm (10 farms in 2018/20 farms in 2021) with one trap within each habitat of interest. Samples were collected every three days, over an eight-week sampling period. Targeted transects were also performed to check whether traps were consistently missing any species. Our research provides a novel dataset with an inclusive focus on both wild bees and flower flies that can be used to benefit farmers, landowners, and NGOs to inform government policy to improve food security of essential crops while simultaneously protecting our threatened native pollinators. A substantial amount of data were collected across both field seasons, and preliminary results show an extremely high diversity of flower flies in the region as well as pockets of unusually high aphidophagous flower fly abundance. These results, alongside an analysis of the abundance and diversity presence of Syrphidae in each habitat will be presented.

Novel approaches on the taxonomy and systematics of grass flies (Chloropidae)

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Keywords: biodiversity, DNA barcoding, frit flies, integrative taxonomy

With over 3,000 species classified into 200 genera and thousands yet to be described, Chloropidae is a hyperdiverse family of flies. The importance of this group is evidenced by the high abundance and richness, global distribution, and diversified life-history strategies, with special reference to grasslands. The challenging taxonomy and poorly understood ecological role make this lineage a great fit for implementing large-scale integrative taxonomy pipelines, cutting-edge imaging techniques and exploring the genomic diversity. The latest advancements in the taxonomy of the group have generated DNA barcodes of over 1,300 specimens from Sweden resulting in the identification of 63 species which corresponds to 15% of the European grass flies diversity. The study included the description of new species and permitted to address boundaries of long-standing species complexes. More developments in the taxonomy, systematics and functional morphology of grass flies on a global scale are under development. These novel approaches not only expand our knowledge on this globally important lineage of flies which are fundamental for exploring further evolutionary questions but also serve as a model for the implementation of cutting-edge technologies on other neglected groups of flies.

Pesticides and blow flies: how do clothianidin and bifenthrin affect Calliphoridae (Diptera) oviposition?

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Keywords: blow flies, forensic entomology, oviposition, pesticides, toxicology

Blow flies (Diptera: Calliphoridae) arrive to remains within minutes and insect colonization can occur soon after in the form of oviposition (egg laying). This is known as the time of colonization (TOC), which forensic entomologists use to estimate a minimum post-mortem interval (minPMI), or time between insect colonization and discovery of the remains. There are many factors, however, that can influence blow fly colonization and oviposition. The presence of chemicals including insecticides can, accidentally or intentionally, mask the odors of decomposition resulting in detection and oviposition changes. In this study, we aimed to determine how oviposition substrates treated with different concentrations of clothianidin (neonicotinoid) or bifenthrin (pyrethroid) influenced blow fly oviposition under laboratory. We hypothesized the following: if the presence of insecticides has a direct effect on the time it takes blow flies to detect a substrate, then as insecticide concentration increases, oviposition events will take longer. To test this, we treated beef liver with one of three concentrations of each insecticide or distilled water (control). We then placed the liver into small cages containing 40 females and 20 males of either *Phormia regina* (Meigen) or *Lucilia sericata* (Meigen)—two common blow fly species in the United States. Observations for first oviposition event were recorded every 15 minutes for the first 2hr and every hour until the 8hr mark. Using ANOVA analyses, we determined differences between treatments and species. We found that both bifenthrin and clothianidin have a significant effect on *L. sericata* oviposition, $p=0.0376$ and $p=0.0199$, respectively. However, bifenthrin and clothianidin appear to have no significant effects on *P. regina* oviposition ($p=0.742$ and $p=0.566$, respectively). Interestingly, clothianidin resulted in high rates of paralysis and mortality for both species. These results can help to improve entomological evidence analysis in legal cases where insecticide presence is suspected.

State of art of *Stylogaster* Macquart (Conopidae) after the Museu Nacional/UFRJ fire

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Keywords: biodiversity, biogeography, Museu Nacional, phylogeny, taxonomy

Stylogaster Macquart (Conopidae, Stylogastrinae) contains 116 described species, 115 of them extant. *Stylogaster* is recorded for all biogeographic regions, except for the Palaeartic, and is very different from the other conopid subfamilies, both in morphology and biology. Non-stylogastrine conopids prefer hymenopteran hosts. In contrast, *Stylogaster* species apparently prefer Orthoptera and Blattaria as hosts, but little work has been done on host associations. There are few researchers working on conopids (and *Stylogaster*) across the world, based mainly in Canada, Brazil, Germany and Australia. The last identification keys are from Camras & Parrillo (1985) for the New World, and Stuke (2012) for Africa. The tragic fire at Museu Nacional on 2 September 2018 destroyed 90% of the entomological collection, including all mounted specimens of *Stylogaster*. The types of 16 species described by Lopes, Monteiro, Lopes & Monteiro and Rocha & Mello-Patiu were all lost, as well as many other specimens identified or yet to be studied. The following species are now lacking their name fixation: *Stylogaster alvarengai* Lopes, *S. amapaensis* Monteiro, *S. australis* Lopes, *S. fidelis* Monteiro, *S. fluminensis* Rocha & Mello-Patiu, *S. hirtinervis* Lopes & Monteiro, *S. hugoi* Rocha & Mello-Patiu, *S. jactata* Lopes & Monteiro, *S. lepida* Lopes & Monteiro, *S. longicornis* Lopes, *S. macrura* Lopes, *S. nigrifrons* Lopes, *S. pilosa* Lopes & Monteiro, *S. planitarsis* Rocha & Mello-Patiu, *S. shannoni* Lopes & Monteiro, *S. souzai* Monteiro, 1960 and *S. tarsata* Lopes. On the other way, some unmounted material from a survey project survived the fire and now there are 231 specimens of *Stylogaster* under study in a major revision of the Neotropical species that is in course. By now, we know that at least to *S. macrura* a neotype is to be assigned and there is great chance of more species have their name fixation recovered.

Interactive identification key to third instars of *Anastrepha* Schiner (Diptera: Tephritidae)

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Keywords: agriculture, fruit flies, key, larvae, Lucid, morphology, taxonomy, fruit flies

Anastrepha is the most diverse and economically important genus of Tephritidae in the American tropics. Of the 328 species, multiple species are serious agricultural pests, attacking crops such as mango, citrus, and other commercial fruits. *Anastrepha* taxonomy is based largely on the adult females, and the immatures stages remain understudied with most species lacking a description of the larva. Third instars of only 38 species have been described. We conducted comparative studies on larvae collected from naturally-infested fruits and compiled data from those and previously described larvae. The scope of this study was to create an interactive identification key to described larva using morphology. Qualitative and quantitative character data were generated from the facial mask (oral ridges, accessory plates, stomal organ), cephalopharyngeal skeleton (mouthhooks), anterior and posterior spiracles, dorsal spinules, and anal lobes. The interactive larval key, developed in Lucid, includes 41 *Anastrepha* species, including all that are recognized as minor to major pests or that are potential emergent pests. This presentation describes the development and structure of the key. The interactive key is free to access at <https://idtools.org/tools/1062/index.cfm?pageID=3353>.

Nematode-induced demasculinization of Nearctic Dolichopodidae (Diptera) and the taxonomic implications

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Keywords: Nematoda, Mermithidae, parasitism, Parasitylenchidae, *Dolichopus*,
Rhaphium, *Tachytrechus*, synonymy

Demasculinization, the reduction of male sexual characters due to parasitic nematode infection, is described in Nearctic Dolichopodidae. Five species are confirmed to be demasculinized (3 species of *Dolichopus* and 1 species each of *Rhaphium* and *Tachytrechus*) by nematodes in two families (apparent Mermithidae and Parasitylenchidae). On at least five occasions, nematode-infected males were inadvertently described as new species. A conspectus is presented of the scope of taxonomic problems caused by nematodes in Nearctic *Dolichopus* and *Tachytrechus*, which suggests that ten additional names in *Dolichopus* could be based on infected specimens. Guidelines are provided for recognizing nematode infection to avoid erroneous species descriptions.

Evaluation of different larval diets for the laboratory rearing of *Aedes albopictus* (Skuse) (Culicidae)

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Keywords: laboratory rearing, larval diets

The Sterile Insect Technique (SIT) is an innovative method to suppress populations of important mosquito vectors, like *Aedes albopictus*. Prerequisite for successful SIT applications is the efficient mass rearing of mosquitoes and the high performance of released sterilized males. Larval diet affects survival and developmental rates of immatures and the size, survival and performance of adults. In the current study we compared five different larvae diets: (a) cat-food (CF), (b) fish-food (FF), (c) milled larvae from *Tenebrio molitor* (YM) and (d) *Hermetia illucens* (BSF), and (e) a standard diet used in Centro Agricoltura Ambiente, Italy (CAA) (mixing of CF, FF and brewers' yeast). We placed 300, 1st instar larvae in small trays containing 1.5lt of tap water and 0.8g larvae food provided either at once or sequentially over several days. Immatures survival and developmental duration, and adult survival up to day 20 of age were recorded in constant laboratory conditions (25°C). The mode of food supply affected immature survival rates but not developmental duration and sex ratio of emerging adults. High mortality of immatures and decreased adult emergence was recorded when food was provided at once compared to sequential provision of the same amount of food. Higher larval survival and adult emergence was recorded in CAA and CF in both modes of food provision compared to other diets tested. Larvae reared in CF and FF developed faster but a higher number of males produced from the CAA and YM diets. Adults emerged from the CAA diet showed higher survival to adult day 20 compared with all other diets. It seems that although CAA performs better, its cost is quite high and can be reduced considering as ingredients milled insect larvae. This study was supported by the MoSquITo project (Greece 2.0; EU; T2EΔK-02020).

Earthworm-feeding Diptera: a review and evolutionary insights

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Keywords: Annelida, Calliphoridae, endophagy, Lumbricidae, parasitoid, Polleniidae, Sarcophagidae

Earthworms (Annelida: Oligochaeta) play a crucial role in terrestrial ecosystems, representing approximately 35% of the total animal biomass. They serve as a significant trophic resource for various animal groups, including arthropods such as Formicidae, Staphylinidae, Carabidae, and Diptera. Notably, among these, dipterans are the sole group of parasitoids that have exploited earthworms as hosts. Specifically, the Oestroidea superfamily has independently evolved this developmental strategy at least four times. The objective of this review is to provide a comprehensive overview of earthworm parasitoids, with a specific focus on Oestroidea species that undergo endophagous development. The review consolidates existing knowledge on the feeding characteristics of these flies. Our findings indicate that endoparasitoids in this group of families primarily target Lumbricidae (Annelida: Oligochaeta) as their preferred hosts and display a considerable degree of polyphagy, utilizing multiple host species from different genera. Various taxonomic groups have employed diverse developmental strategies, ranging from rapid development spanning 2–14 days, as observed in *Sarcophaga*, to prolonged and intricate cycles characterized by diapause and potential host-parasite interactions, as seen in *Pollenia*. In all cases, the larval feeding activity of parasitoids induces noticeable behavioral abnormalities and discernible body lesions in earthworms at advanced stages of infection. Furthermore, we underscore the importance of conducting further studies to unravel the complete parasitoid-host specificity of each species, including their feeding mechanisms within earthworms. Finally, the review delves into the evolutionary origins of earthworm feeding, shedding light on this intriguing phenomenon.

Diptera surveys in Inner Mongolia, China

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Keywords: Daxinganling Mountains, diversity, Hanma National Nature Reserve, new record, new species, sunflower, Xinlinguole grassland

Located in the north of China, Inner Mongolia forms forests, forest steppe, typical steppe, desert steppe and desert from east to west, with diverse habitats and rich resources of insects in Diptera, which play an important role in maintaining ecological balance, ecological restoration and carbon neutrality. From 2013 to 2023, 285,482 insects of Diptera were collected by Malaise traps and ground pitfall traps. A comparative analysis of the species-level diversity of Diptera in Xilinuole grassland, sunflower field and Hanma National Nature Reserve from Daxinganling Mountains was carried out, and the species diversity of flies in the forests of Hanma National Nature Reserve from Daxinganling Mountains was classified and identified. The result showed that 292 species, 168 genera and 33 families of Diptera were identified. Among them, the flies belong to 242 species, 140 genera, 18 families, and the Acalyptratae includes 147 species, 77 genera, 20 families with one family, 6 genera and 18 species newly recorded for China, and 8 species new to science. All the data will be input into the National Specimen Platform database with the DNA sequence information data, contributing to the conservation of Diptera resources in China.

Empidoidea

**New World species of the aquatic dance fly genus
Roederiodes (Diptera: Empididae)**

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Keywords: Clinocerinae, distribution, Nearctic, Neotropical

The distinctive aquatic dance fly genus *Roederiodes* Coquillett is found in fast-flowing rocky streams and at the base of waterfalls. The genus is recorded from all geographical regions except Australasia and includes 25 described species. In the New World, 13 species are newly recognized, extending from northern Canada to Costa Rica. The distribution of the New World species is discussed and COI mitochondrial DNA barcodes are known for seven Nearctic species.

The global classification and description of mosquito larval habitat communities

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Keywords: community structure, Culicidae, diversity, ecoinformatics, larval habitat

Mosquito larval habitats are an integral part in a mosquito life cycle with species thought to be on a gradient from generalists to specialists. However, a species-level global analysis of mosquito larval habitats and community structures has never been undertaken. To address this gap in knowledge, we obtained larval habitat descriptions for approximately 1,200 mosquito species from the scientific literature. There were approximately 1,900 unique ways in which larval habitats are described throughout the literature. To categorize these habitats, we developed a classification schema for mosquito larval habitats based on the EUNIS freshwater habitat classification and Greeneley (2001) phytotelmata classification systems. Our classification system reduced the 1,900 unique larval habitat descriptions in the scientific literature into 79 habitat categories. This allowed us to investigate larval habitat community structure for 1,200 mosquito species worldwide. Additionally, we investigated network connectivity, habitat uniqueness, and other metrics describing how mosquito species cluster in respect to their proposed larval habitats. To demonstrate the utility of this approach, we used our data to examine how mosquito species diversity is affected by specific larval habitats in different countries.

A review and reorganization of the Natural History Museum's Diptera collection

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Keywords: museum collection, NHM London

The Diptera collection at the Natural History Museum in London, which contains approximately two and a half million specimens, is being reorganized to reflect our current understanding of Diptera phylogeny and to manage collection expansion. Here we present an overview of the NHM collection, the changes that are being made, and highlight parts of the collection that have the greatest potential for taxonomic research.

The first reported case of accidental intestinal myiasis in a domestic dog by the flesh fly, *Sarcophaga africa* (Wiedeman) (Sarcophagidae)

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Keywords: DNA barcoding, identification, myiasis

Myiasis occurs when fly larvae, or maggots, feed on the tissue, secretions or digestive content of a live vertebrate. Here, a rare case of accidental intestinal or enteric myiasis is reported in a domestic dog. The species of fly was identified as *Sarcophaga africa* (Wiedeman) using the barcoding region of cytochrome oxidase I (COI). A brief discussion on the usage of the term “pseudomyiasis” is provided and the complex taxonomy of *S. africa* is briefly summarised in order to shed light on the erroneous use of *S. cruenata* Meigen and *S. haemorrhoidalis* Fallen with obvious downstream effects. Finally, a comparative assessment to the limited cases in the literature is provided. These few cases are however highly fragmented and our understanding of accidental intestinal myiasis and the clinical manifestations thereof remain incomplete.

An enduring enemy: evolutionary history of host association in the Culicidae

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Keywords: host association, macroevolution, phylogenomics

Mosquitoes have profoundly affected human history and continue to threaten human health through the transmission of a diverse array of pathogens. The phylogeny of mosquitoes has remained poorly characterized due to difficulty in taxonomic sampling and limited availability of genomic data beyond the most important vector species. Here, we used phylogenomic analysis of 709 single copy ortholog groups from 256 mosquito species to produce a strongly supported phylogeny that resolves the position of the major disease vector species and the major mosquito lineages. Our analyses support an origin of mosquitoes in the early Triassic (217 MYA [HPD: 188–250 MYA]), considerably older than previous estimates. Moreover, we utilize an extensive database of host associations for mosquitoes to show that mosquitoes have shifted to feeding upon the blood of mammals numerous times, and that mosquito diversification and host-use patterns within major lineages appear to coincide in earth history both with major continental drift events and with the diversification of vertebrate classes.

Examining Diptera entrapment bias in two different taxa of resin-producing trees

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Keywords: amber, Cretaceous, fossil, Miocene, Phoridae, taphonomy

Amber is well known for its finely preserved biological inclusions, providing a unique glimpse into ancient ecosystems. Different amber deposits from various geological periods and locations have different plant origins. Resin-producing trees such as Araucariaceae, Hamamelidaceae, Cupressaceae, and the extinct Cheirolepidaceae contributed to Cretaceous amber deposits. Paleogene deposits contain both gymnosperms and angiosperms, e.g., Baltic amber produced by Pinaceae and/or Sciadopityaceae and Cambay amber produced by Dipterocarpaceae. Deposits of Neogene amber and Quaternary copal, and extant Defaunation resin, predominantly originated from angiosperms such as Fabaceae and Dipterocarpaceae. Not all amber deposits provide bioinclusions, and abundance of trapped organisms varies widely depending on ecological and taphonomical variables. Resin-producing trees, e.g., *Agathis* (Araucariaceae) and *Hymenaea* (Fabaceae), are used as models for investigating different taphonomic aspects related to formation of amber. We compared the abundance of arthropod inclusions in resins from *Agathis* and *Hymenaea* trees, and in sticky traps located on these genera, to evaluate the primary agents for promoting or preventing entrapment in resin. Diptera is studied in detail because of its abundance in the forests, and also in the resins and sticky traps. We observed that resin collected from *Agathis* spp. trees contained few bioinclusions. The number of arthropods was unexpectedly low compared to large numbers found trapped in *Hymenaea verrucosa* resin. Surprisingly, the sticky traps from *Agathis lanceolata* yielded “atypical” results with respect to insect groups trapped. For example, within Diptera, Phoridae dominated the *A. lanceolata* samples, making up almost 90% of total Diptera. Our results indicate significant differences in entrapment of organisms among different types of resin and emphasize the importance of considering entrapment bias and the faunal composition in an environment for paleontological reconstructions.

A surveillance program of invasive mosquitoes based on citizen science in Hungary

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Keywords: *Aedes albopictus*, *Aedes japonicus*, *Aedes koreicus*, citizen science, distribution map, Hungary, invasive mosquitoes, validation

Climate change, intensified tourism and trade activity result in several exotic mosquito species to invade the temperate zone, which has considerable ecological and economic consequences and threatens human health due to the pathogen-transmitting role of these organisms. In the last decade three invasive mosquito species (*Aedes albopictus*, *Ae. japonicus*, and *Ae. koreicus*) have been detected in Hungary. We have established a citizen science program, in which we asked the public to submit reports on their observations of invasive mosquitoes. Between 2019 and 2021, we have collected and taxonomically validated about 3,000 reports that can be arranged along both the temporal and spatial scales. We aggregated these observations into 35 km² quadrats and examined if these can be reliably used for scientific inferences. We first found that the number of validated reports in a quadrat depends on the underlying sampling effort (i.e., number of total reports), but this relationship varies among species and study years. Second, after controlling for study effort, we showed that the prevalence and presence/absence of invasive mosquitoes within quadrats are significantly repeatable among years, but this consistency varies in a species-specific way. Third, we demonstrated that conclusions about the local presence/absence of focal species based on citizen reports corroborate well the results of direct field sampling with conventional trapping protocols. If the reporting intensity is appropriate (i.e., the number of reports reaches a species-specific threshold), citizen science results can be used to derive biologically meaningful conclusions about the distribution of invasive mosquitoes in a country. Distribution maps of the three invasive species in Hungary can be used to identify ecological predictors that determine such spatial patterns and also to develop a mosquito control program and assess epidemiological risk.

The association between urbanization and the presence of invasive mosquito species in Hungary based on community science

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Keywords: *Aedes albopictus*, *Aedes japonicus*, *Aedes koreicus*, citizen science, distribution map, Hungary, invasive mosquitoes, validation

Urbanization can be a significant contributor to the spread of invasive mosquito vector species, and the diseases they carry, as urbanized habitats provide access to a great density of food resources (humans and domestic animals) and offer abundant breeding sites for these vectors. Although anthropogenic landscapes are often associated with the presence of invasive mosquito species, we still have little understanding about the relationships between some of these and the built environment. This study explores the association between urbanization level and the occurrence of invasive *Aedes* species, specifically *Aedes albopictus*, *Aedes japonicus*, and *Aedes koreicus*, in Hungary, using data from a community (or citizen) science program undertaken between 2019 and 2022. The association between each of these species and urbanized landscapes within an extensive geographic area was found to differ. Using the same standardized approach, *Ae. albopictus* showed a statistically significant relationship with urbanization, whereas *Ae. japonicus* and *Ae. koreicus* did not. The findings highlight the importance of community science to mosquito research, as the data gathered using this approach can be used to make qualitative comparisons between species to explore their ecological requirements.

**Two-hours life trapped in amber –
first fossil representatives of the Deuterophlebiidae
and their evolutionary and ecological implications**

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Keywords: amber, fossil, Mesozoic, mountain midges, Nematocera, palaeoecology

Deuterophlebiidae (mountain midges) currently includes one genus, *Deuterophlebia*, with 19 species, most of which have circumpacific Holarctic distribution; several occur deep in the Asiatic continent. Recent preimaginal stages occur on rocky substrates in cold, running water with velocity sometimes exceeding 3-4m/s. Adult males live up to two hours and are renowned for their peculiar morphology. Their head capsule is reduced with vestigial mouthparts while wing venation is composed mostly of numerous pseudo-veins in a fan-like arrangement. Their phylogenetic position has remained unclear, partly due to lack of fossils which could provide information on early states of unique characters and unambiguous evidence supporting a relationship with other families of Diptera. This stalemate situation can be overcome now with the first amber fossil deuterophlebiid specimens confirming their presence in the Cretaceous, already at this time with some of their bizarre characters. The evolutionary history of the family can now be reconstructed to a greater extent, with the possibility to evaluate earlier lineages. Until now, Deuterophlebiidae was the only recent dipteran family without known fossil representatives. One inclusion of a deuterophlebiid male is accompanied by a syninclusion of an ephemeropteran wing. Presence of these two short-lived insects in the same piece of amber suggests that the flight period for both species was short. Moreover, their time of flight was apparently synchronous with flow of fluid resin. The chance that all these events occur at the same time seems low. Preference of recent species to cold, running waters may indicate presence of such habitats in amber-producing forests of the Cretaceous.

**Comparative morphology of epandrium in
Tipula (Vestiplex) Bezzi, 1924 crane flies (Diptera, Tipulidae):
from ventral lobes to the tergal saucer**

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Keywords: diversity, genitalia, homology, tergite 9

The subgenus *Tipula (Vestiplex)* is large group of crane flies (Diptera, Tipulidae) with 189 recognized species distributed in Holarctic and Oriental Realms. The male terminalia is extremely polymorphic, among genital structures epandrium is surprisingly diverse and of great taxonomic and phylogenetic importance. Typically, the epandrium is forming so called “tergal saucer”, a sclerotized plate while other species of *T. (Vestiplex)* have epandrium divided middorsally by pale membrane with ventral portion bearing a pair of membranous plates or heavily sclerotized, lobes; other species have ventral portion hypertrophied, occupying nearly entire area of epandrium. Though Alexander briefly provided some insight on the origin of ventral lobes from tergal saucer (1965), however no further comparative analysis was provided to establish homology of different parts of epandrium in *T. (Vestiplex)*. Comparative morphology of epandrium of crane flies in subgenus *T. (Vestiplex)* is presented. Homology of tergal saucer with ventral portion and the main evolutionary ways of epandrium are discussed.

Genital bridge and interbase – same structure, different names

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Keywords: diversity, genitalia, homology, tergite 9

As was noted by Wood (1991), there are specialized terminology for structures in different family groups developed by taxonomist on their own with no regard for homology with other groups. This is also true for crane flies (Diptera, Tipuloidea) where different terms are used for the same structure. The homology of interbase (=dorsolateral portion of parameres) in Limoniidae with genital bridge (= sclerites sp1+sp2; = gonocoxal fragment) in Tipulidae is discussed based on criterion of connection to other parts. The possible origin of appendages of sternite 9 (A9s) in genus *Tipula* is also presented.

**Taxonomy and systematics of the American bristle fly genus
Xanthophyto Townsend (Diptera: Tachinidae)**

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Keywords: CO1, Nemoraeni, parasitoid

Xanthophyto is a small genus of New World Tachinidae consisting of four described species, two Nearctic and two Neotropical. Members resemble flesh flies (Sarcophagidae) in overall appearance, often with the tip of the abdomen reddish in color. They are parasitoids of various families of caterpillars, often concealed feeders associated with conifers. Over the past several years I have been in the process of taxonomically revising *Xanthophyto*, evaluating species relationships, and delimiting and describing new species using external morphology, male terminalia, CO1 mtDNA sequence data, distributions, and host associations. Despite their relatively large size and the abundance of several (undescribed) species, at least 10 undescribed species exist in North America and at least 20 undescribed species occur in the Neotropics. The genus appears to have originated in the Neotropics but there have been several recent radiations of species in North America, making species boundaries difficult to recognize. Male terminalia exhibit relatively little variation among species except at the broadest scales and there is considerable convergence in external traits. I highlight evolutionary relationships among taxa, patterns of morphological evolution, and ecological/biogeographical patterns in the genus.

**Estimating *Dirofilaria* (Railliet & Henry)
(Nematoda: Onchocercidae) occurrence and distribution
in Hungary using different approaches**

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Keywords: Culicidae, *Dirofilaria*, emerging disease, invasive mosquitoes

Dirofilariosis is an emerging mosquito-borne disease that is caused by the nematode species *Dirofilaria immitis* (Leidy) and threatens domestic dogs worldwide. *Dirofilaria immitis* has been established in Hungary since 2007, and two approaches were used to understand the occurrence, diversity, and distribution of these pathogens within the country. In one approach, data were collected from dog owners in a community (or citizen) science campaign aiming to describe the spatiotemporal patterns of heartworm disease in Hungary, in which the practices of dog owners were also considered. In the second approach, we collected mosquitoes nationwide, to molecularly test the occurrence of the parasite in several potential vectors, including a native, *Aedes vexans* (Meigen), and three invasive species, *Ae. albopictus* Skuse, *Ae. japonicus* (Theobald), and *Ae. koreicus* (Edwards). The results of the questionnaire based on ~1400 respondents show that the disease is present across the country but with varying prevalence (of which the average is 19.7%). Overall, the occurrence of heartworm disease in Hungary shows an increasing trend compared to the results of previous years, and the central and eastern regions of the country show the highest prevalence. Dog ownership habits, such as the daytime/nighttime housing of dogs, also contribute to the prevalence of the disease in Hungary. The molecular survey showed the highest parasite prevalence in the native *Ae. vexans* (14.5%), while *Ae. albopictus* (4.3%), *Ae. japonicus* (0.0%), and *Ae. koreicus* (13.2%) were infected less frequently. Accordingly, the presence of the parasite can be confirmed in invasive mosquitoes that have been introduced in the last decade to the country. Developing new and improved surveillance and control strategies to better manage this emerging disease is increasingly important.

First data on early instar Afrotropical Miltogramminae (Diptera: Sarcophagidae) reveal unexpected morphological diversity

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Keywords: Afrotropical, larva, morphology, SEM, CLSM

First instar larvae of the genera *Hoplcephala* Macquart, *Khowaba* Pape and *Lamprometopia* Macquart are described for the first time, along with the first instar larva of '*Chauliooestrus*' *leza* Pape and *Phylloteles picifrons* Pape. The larval morphology is analysed using a combination of light microscopy, confocal laser scanning microscopy and scanning electron microscopy. Pseudocephalon, antennal complex, maxillary palpus, facial mask, thoracic and abdominal segments, anal region, spiracular field, posterior spiracles, and details of the cephaloskeleton are documented. Substantial morphological differences are observed between the three genera, as well as in comparison to other known larvae of Miltogramminae. Notable unique character states are found in the shape of the labrum and mouthhooks of the cephaloskeleton, the sculpture of the integument, and the sensory organs of the thoracic and abdominal segments. The first instar larval morphology corroborates the close phylogenetic affinity of '*Chauliooestrus*', *Hoplcephala*, *Khowaba*, *Lamprometopia*, and *Phylloteles* with other members of the grade of 'lower' Miltogramminae. The larvae of *Khowaba atrox* Pape and *Hoplcephala* sp. are the most peculiar by massive cephaloskeleton, integument covered by cuticular hair-like spines/scales or body papillae modified into fleshy processes. The first instar larva of *Phylloteles picifrons* possesses a large, bladder-like structure with an unknown function dorsally on the first thoracic segment. All documented larvae are characterized by a maxillary palpus forming a distinct protuberance with an elongated second sensillum basiconicum. The dorsal bridge of the cephaloskeleton is present only in '*Chauliooestrus*' *leza* Pape. The possible breeding habits are discussed and an association with termites is suggested.

Towards the most resolved phylogeny of Polleniidae – implications for their systematics

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Keywords: *Pollenia*, phylogeny, systematics

The family status of Polleniidae has gained wide acceptance among dipterists, yet research evaluating subfamilial phylogenetic relationships is limited. Phylogenetic hypotheses for the family have suffered from limited taxon sampling, and in most cases, phylogenetic trees have contained only a small representation of species from a few genera. Two recent molecular studies have delivered a comprehensive intraspecific polleniid phylogenetic hypothesis based on broad taxon sampling, including 22 species belonging to five genera, representing almost all species-groups defined by Rognes for the West Palearctic species of *Pollenia* Robineau-Desvoidy. Analysis of mitogenomic data showed strong support for a monophyletic *Pollenia* and rejected the synonymisation of the genera *Dexopollenia* Townsend and *Xanthotryxus* Aldrich with the Palearctic *Pollenia*. Analysis of the three molecular markers (COI, Ef1 α , CAD), subsequently supported by mitogenomic data, allowed for critical re-evaluation of the species-group system within the genus *Pollenia*. Our results are incongruent with previous morphology-based cladistic interpretations, with the *amentaria*, *griseotomentosa*, *semicinerea* and *viatica* species-groups resolved as non-monophyletic. Morphological analysis enabled identification of key structures supporting the topology of our phylogenetic trees. Details of male terminalia, particularly appears to provide the best support for certain nodes. However, only a subset of the received clades may be defined by morphological characters, and a morphological definition of the family Polleniidae cannot be established.

Study on the metathoracic segments in the Nematocera

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Keywords: homology, metapleural suture, metepimeron, metepisternum, morphology

Flies have atrophied metathoracic segments due to the reduction of wings into halteres. The pleura of the wing-bearing (pterothoracic) segments are important for mechanics and contain significant phylogenetic information. The metathoracic segments are also involved in the movement of the abdomen. This study focuses on the characteristics of the metathoracic segments in nematocerans. Based on the metapleural suture, the homologies of metepisternal and metepimeral pleura have been redefined. This suture is an internal apophysis that usually extends from the coxa to the wing process. Findings indicate that in Tipuloidea, both the metepisternum and metepimeron are usually broad, while in Culicomorpha, the metepimeron is generally small and strongly reduced. In Psychodomorpha, the pleural suture is indistinct or incomplete. Most families of Bibionomorpha have a pair of short apodemes or invaginations in the metathoracic postnotum, except for Scatopsidae. Additionally, in Anisopodidae, the genus *Sylvicola* Harris has a forked metathoracic postnotum at the dorsal portion, a unique joint structure to the first abdominal tergite.

Unveiling the black hole of 'non-EU Tephritidae': identification of fruit fly larvae intercepted in the EU

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Keywords: COI, DNA barcoding, European Union, import pathways, interception,
larval morphology, quarantine fruit flies

Recent Regulation (EU) 2021/2285 has considerably expanded the list of quarantine fruit flies (Diptera: Tephritidae) for the European Union. It now includes 75 taxa (species and genera) whose introduction and spread over the EU territory must be promptly detected and prevented. Whereas under the previous legislation National Reference Laboratories (NRLs) could identify all exotic flies as 'non-EU Tephritidae', they must now identify them at the species or genus level. This is not an easy task as diagnostic protocols, reference material and sequence records in the databases are lacking for a large part of the listed taxa, especially at the larval stage, which is the one most commonly encountered during plant product inspection at import. To try to unveil what is hidden beyond the black hole of 'non-EU Tephritidae' intercepted over time, the staff of the EURL has collected real interception data over the last twenty years and re-analysed larval specimens using both morphological and molecular methods. The aim is to build a clear and comprehensive picture of Tephritidae taxa actually intercepted in the EU and their respective import pathways (origin-host plant association). The expected output is the development of a simplified and up-to-date tool for the rapid morphological identification of third instar larvae of Tephritidae intercepted at the EU border. This identification tool will help NRLs dealing with fruit flies larvae identification from import control. Preliminary data show that few taxa are regularly intercepted over time and that most of them can be identified morphologically, provided that pathways are known with certainty.

Processing of entomological traces to supply the morphological and molecular diagnosis: a proposal for a standard protocol for the Neotropical region

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Keywords: blow flies, forensic entomology, identification, laboratory techniques, molecular, morphology, Neotropical, protocol

Blow flies (Calliphoridae) can provide evidence to estimate the postmortem interval (PMI), determine the cause of death, and indicate cases of abuse and negligence. In order to continue or even complete a forensic investigation, in addition to correctly identifying the insects associated with a given crime, access to biological and ecological information on one or more species is essential. It is known that abiotic or biotic factors can affect insect development and, consequently, alter the morphometric data that are used PMI estimation. Other studies have also shown that certain techniques for preserving and preparing specimens can also modify the length, weight and anatomical characters of larvae. This study aimed to evaluate the effects of sample preservation and preparation on the morphometric and morphological aspects of first and third instar larvae of *Lucilia cuprina* (Wiedemann), a necrophagous species, widely distributed and of great forensic relevance in the Neotropics. Larvae of approximately 12h and 72h of *L. cuprina* (N=640) were obtained from flies kept in the laboratory at 27±1°C, 70±10% RH and 12:12 photoperiod. After removing the diet consisting of fresh ground beef, larvae were deposited in Falcon tubes containing 30mL of distilled water and vortexed at 2500rpm/30sec to remove food residues from the integument. They were then divided into two groups: (I) (N=360) those preserved directly in 70% ethanol, 96% ethanol, isopropanol and Kahle's solution; (II) (N=360) those that had body segments distended in water at 80°C/30sec and transferred to the same preservative media. Some larvae (N=80) from each experimental group had DNA extracted and amplified with a barcode primer set. From 50–100% of DNA of samples from all experimental groups were amplified, except those in Kahle's solution. In all aspects, 70% ethanol was the best preservative.

Discovery of paedogenetic gall midges (Diptera: Cecidomyiidae) from decayed wood and branches in natural environments in Japan and their laboratory rearing techniques

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Keywords: gall midges, mushroom, paedogenesis

Paedogenesis is a unique reproductive mode shown only by several fungivorous gall midges (Diptera: Cecidomyiidae) and a coleopteran species in insects. Some paedogenetic gall midges are regarded as serious pests of mushroom cultivation. In Japan, *Mycophila speyeri* and *Heteropeza pygmaea* were previously found to infest the mycelia of mushrooms in indoor cultivation factories, but the occurrence of paedogenetic species in natural habitats was not previously reported. During our recent field surveys, we first found paedogenetic gall midges in natural habitats, such as decayed trunks and branches on the forest floor in Japan. Based on morphological examinations of larvae as well as adults of some species, we confirmed the occurrence of paedogenetic gall midges belonging to at least five genera as follows: *Miastor*, *Mycophila*, *Heteropeza*, *Heteropezula*, and *Leptosyna*. These gall midges were found on white-rotten woods but not on brown-rotten ones in the field. These results indicate that wood-decaying fungi responsible for white rotting trunks and branches harbor diverse paedogenetic gall midges on the forest floor in Japan. We established a continuous rearing technique for paedogenetic gall midges in the laboratory using *Flammulina velutipes* mycelia growing on potato dextrose agar (PDA) media. In addition, the effects of temperature on the paedogenetic larval development were examined for *Mycophila* sp. A. The generation period of one paedogenetic cycle was 5.9 days at 24°C and a mother larva produced 15-16 young larvae on average. The developmental zero and thermal constant in the paedogenetic development were estimated to be 8.3°C and 92.7 degree-days, respectively, with the traditional linear method. The laboratory rearing method established in this study is useful for studying the developmental traits of paedogenetic gall midges as well as for examining the mechanism of reproduction mode shift from a paedogenetic to holometabolic life cycle.

Curation of the El Salvador gall midge types (Diptera: Cecidomyiidae) at the State Museum of Natural History Stuttgart

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Keywords: Asphondyliini, Alycaulini, Camptoneuromyiini, digitized collection, SMNS

The family Cecidomyiidae includes 6,651 species and 832 genera organized into six subfamilies, five of them with fungivorous habit. The sixth subfamily, Cecidomyiinae, includes mainly the gall-forming species. The State Museum of Natural History Stuttgart (SMNS) houses one of the most important collections of galls and gall midges (Diptera: Cecidomyiidae) of the World, with 25,776 registers. This collection was built and studied by several specialists, including Edwin Möhn, the curator of the Diptera collection at SMNS between 1959 and 1975. The holotypes of 21 genera and 150 species of gall midges described by Möhn from El Salvador were deposited in the SMNS collection, but many were considered indistinguishable from other type or non-type specimens they were preserved with. After an exhaustive search for types stored in ethanol and on semi-permanent slides, it was established that the current number of primary types of Neotropical Cecidomyiinae from El Salvador at SMNS is representative of 124 nominal taxa in 19 genera, all valid. All types were mounted on permanent slides, labeled, and digitized during a 3-month curatorial project to be made available in the Diversity Workbench (DWB). The tribes Asphondyliini, Alycaulini, and Camptoneuromyiini contain the largest number of recovered types, with 54, 52, and 14 holotypes, respectively. Lasiopterini and Trotteriini are also represented with one and three holotypes, respectively. Considering these updates and the recommendation of the International Code of Zoological Nomenclature, an annotated catalog of the Neotropical primary types of Cecidomyiinae from El Salvador deposited at the SMNS was prepared.

Molecular phylogeny and biogeography of the subfamily Clinocerinae (Empididae)

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Keywords: biogeography, molecular phylogeny, polyphyletic, *Proclinopyga*

The phylogenetic relationships and biogeography within subfamily Clinocerinae (Empididae) have been previously investigated morphologically, but now a molecular aspect is added. The subfamily consists of 17 genera: *Afroclinocera* Sinclair, *Asioclinocera* Saigusa & Sinclair, *Asymphyloptera* Collin, *Bergenstammia* Mik, *Clinocera* Meigen, *Clinocerella* Engel, *Dolichocephala* Macquart, *Hypenella* Collin Kowarzia Mik, *Oreothalia* Melander, *Phaeobalia* Mik, *Proagomyia* Collin, *Proclinopyga* Melander, *Roederiodes* Coquillett, *Rhyacodromia* Saigusa, *Trichoclinocera* Collin and *Wiedemannia* Zetterstedt. There are more than 465 described species in the subfamily, with genera *Clinocera* (122) and *Wiedemannia* (121) being the richest in species. In this study, we sequenced two mitochondrial (COI and Cyt β) and two nuclear (CAD and EF-1 α) genes to reconstruct the phylogenetic relationships within the subfamily, using both Bayesian inference and maximum-likelihood approaches. From 17 genera, 15 were represented, as *Asioclinocera* was only recently described in 2022, and we could not obtain successful amplification of marker genes for the genus *Rhyacodromia*. The subfamily is found to be polyphyletic, but most of the genera are found to be natural groups. Genera *Asymphyloptera*, *Proagomyia*, *Hypenella* and *Afroclinocera* are placed outside of the remaining genera, within Empidinae. We should take this with caution as *Hypenella* and *Afroclinocera* were only represented by one gene, COI, so their status is not certain. *Proclinopyga* is placed as sister to the remaining Clinocerinae. *Clinocera* appears to be polyphyletic. Subfamily Clinocerinae has a worldwide distribution, but some genera, such as *Bergenstammia*, *Phaeobalia* and *Clinocerella* are strictly confined to Europe. *Oreothalia* is restricted to the Nearctic Region, *Afroclinocera* restricted to the Afrotropical Region and *Asioclinocera* restricted to the Oriental Region. *Clinocera* is the only genus that is recorded from all biogeographical regions.

To see the unseen: on confocal microscopy in Diptera morphology studies

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Keywords: cephaloskeleton, confocal microscopy, genitalia, morphology

Insect morphology has traditionally been studied using light microscopy (LM). However, LM has certain optical limitations in regards to resolution, illumination and depth of field, which can hinder the recognition of fine structures, their shape, and their interactions. Confocal laser scanning microscopy (CLSM) has revolutionized the field, providing high-resolution, high-quality images and three-dimensional visualizations of structures under examination. CLSM has a significant impact on the quality of information when compared to more traditional methods of imaging, yet its use in Diptera morphology studies is not yet widespread. Using both larval instars and adult flies of cyclorrhaphan Diptera, we show the usefulness of CLSM for studying the morphological characters by taking advantage of the autofluorescent properties of cephaloskeleton and adult genital structures. Application of CLSM allowed for detailed examination not only the fresh but also more than 100-year-old museum material. In the latter case, LM did not provide satisfactory results because of sample bleaching resulting from storage conditions, yet CLSM allowed for successful examination. CLSM application revealed the presence of structures previously unknown in examined taxa, e.g., the rami, or considered absent, i.e., the parastomal bar in Muscidae. We urge dipterists to implement CLSM in morphological studies for a better understanding of the complex morphological structures of Diptera, particularly the fine sclerites of the cephaloskeleton, which are often challenging to observe using traditional microscopic techniques. We believe that the use of CLSM will improve the understanding of dipteran morphology but also facilitate the recognition of previously unknown morphological structures and shed more light on the evolution and phylogenetic relationships of Diptera.

Forensic entomology and the Sonoran Desert: progress and challenges

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Keywords: blow flies, Calliphoridae, colonization, decomposition

The utility of blow flies in legal investigations has increased due to the plethora of information they can provide to law enforcement including if a body has been relocated, an amount of time the remains have been present along with detecting the presence of various toxins within the body. Much of the work conducted in the United States pertains to areas that consist of temperate climates. Although this information is useful to a wide geographical area, it is limited in its applicability to areas like the American Southwest. This presentation will review the historical work conducted related to forensic entomology within the Sonoran Desert and discuss current research focusing on blow fly populations and initial colonization of decomposing remains, specifically in Arizona, USA.

**Inducing and observing the *Chrysomya megacephala*
(Diptera: Calliphoridae) morning twilight male mating swarm:
a preliminary study**

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Keywords: behavior, eye morphology, low light, sexual dimorphism

Chrysomya megacephala has sexually dimorphic compound eyes in which the male has distinctly enlarged upper ommatidia. Such an arrangement should permit greater ability to detect motion in low light. The biological purpose for this was observed by Alan Olsen and Thomas Sidebottom during July of 1986 at one site in the Palau Archipelago. Males formed “dawn” swarms, hovering in place and apparently chasing females passing through. The setting was an open area with no adjacent vegetation. To our knowledge this behavior was not observed since. In blow flies, such sexually dimorphic eyes occur only in *C. megacephala* and close relatives, and nothing like this behavior is known for any other blow fly species, which are generally inactive at night. We found that the swarm can be induced in south Florida by attracting *C. megacephala* to a tree using carrion bait. Flies that spend the night on the tree swarm the next morning between dawn and sunrise. The behavior never occurred at sunset. In contrast to Olsen and Sidebottom’s experience, *C. megacephala* males appeared to be using the tree as a landmark to maintain hovering position, i.e., the height of the tree determines the height of the swarm. This presentation will include a variety of casual observations of this behavior as well as images and high-speed video of swarming, the chase, and copulation on the ground.

Afrotropical flesh flies (Diptera: Sarcophagidae) – a synopsis

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Keywords: Afrotropical, biogeography, Miltogramminae, Paramacronychiinae, Sarcophaginae, *Sarcophaga*, taxonomy

Flesh flies (Sarcophagidae) are a diverse family of calyptrate Diptera in the superfamily Oestroidea, with an estimated age of ~35 million years and a possible sister-group relationship to bot flies (or to bot flies + Mystacinobiidae). They have diverse ecologies, gravitating towards necrophagy, parasitoidism and kleptoparasitism, primarily on other invertebrates. Although most described Afrotropical flesh fly species are classified in the large, cosmopolitan genus *Sarcophaga*, the bulk of their diversity in the region, at the generic level, is found in the mostly kleptoparasitic subfamily Miltogramminae. The subfamily Paramacronychiinae is remarkably sparsely represented, with only two species so far known to occur south of the Equator. At the present state of knowledge, the Afrotropics host around 350 sarcophagid species in about 28 genera, although recent collecting activities and the study of museum material provide hints to a much greater diversity, not only of species but also of genera. Here, we present an updated synopsis of the fauna based on recent work in preparation for the Manual of Afrotropical Diptera project.

Assessing phylogenomic support for a renewed fly tree of life

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Keywords: divergence times, genomics, phylogeny, phylogenomics

Phylogenomic data sets are revolutionizing our ability to evaluate competing hypotheses in fly classification and evolution, but as this data source is more widely used, we also uncover new challenges in analysis, interpretation, and conflict among data types. Disagreement among estimates of relationship from alternative data sets is commonplace, but methods for evaluating and reducing misleading signal are just beginning to be applied to the largest datasets ever assembled for Diptera. Here, we present the most recent results of analyses of data from published fly genomes, transcriptomes and anchored hybrid enrichment that resulted from a decade of collaboration across the insect (1Kite) and dipterological communities. These data establish well-supported clades that repeatedly emerge in analyses of genomic data, establish divergence times for the origin and diversification of the earliest fly lineages, the lower Brachycera, and Cyclorrhapha, and bring increased resolution to some of the most difficult phylogenetic radiations in the fly tree. Exploring information content, homoplasy, rates of evolution, and sampling bias remain major elements in the role of the Diptera systematist – even as we come to a greater consensus on evolutionary relationships throughout the fly tree.

Genomic data supports a new, phylogeny-enabled, identification system for *Anastrepha* (Tephritidae)

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Keywords: anchored hybrid enrichment, identification, species diagnostics

With over 300 named species, *Anastrepha* is the largest group of fruit flies within Latin America and the Caribbean. They include the most economically important pest species in the region, some that have invaded the US and others that remain serious threats to U.S. agriculture. As part of a large collaborative effort to address the need for new integrated identification systems, we developed a new phylogenetic tree of *Anastrepha* species and species groups from anchored hybrid enrichment phylogenomics to serve as reference system and diagnostic data resource. These genomic data are also used to evaluate individual nuclear loci as markers in interactive phylogenetic placement systems, Bayesian-classifier analyses, and PCR-based rapid identification assays. To date, we have screened over 400 loci that yield valuable evidence on the phylogeny and diversity of the genus. AHE generated gene loci are also used to characterize, compare and screen for genetic diagnostic variation through an analytical workflow useful for characterizing diagnostic features at different taxonomic levels.

The evolutionary diversification of the Culicomorpha and the earliest origins of blood feeding in Diptera

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Keywords: anchored hybrid enrichment, blood feeding, phylogeny

The phylogenetic placement of the lower dipteran infraorder Culicomorpha has changed according to recent phylogenomic analyses of the fly tree. This higher-level clade is well-supported as monophyletic, but is now well-supported as one of the earliest diverging lineages in the entire order, originating approximately 225–245 MY. Relationships among the constituent families have also recently been addressed using transcriptomes, but these data have left some remaining uncertainty as to relationships among culicomorphan families. Here, we present a new analysis of family-level relationships from combined transcriptome and genomes sequences to assess support for interrelationships among families, the estimated age of divergences in the clade, and for placing the origins of blood feeding in multiple culicomorphan clades.

Challenges in identifying Afrotropical horse flies of the genus *Atylotus* (Osten-Sacken) (Tabanidae)

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Keywords: Afrotropical, DNA barcoding, identification, vectors

Species of the horse fly genus *Atylotus* are known vectors of pathogens. Previous studies of this genus in the Afrotropics has shown little to no differentiation into species based on the barcoding region of cytochrome oxidase I (COI), largely due to morphological misidentifications. This study analysed all available *Atylotus* COI sequences from GenBank and BOLD to determine if COI is suitable for delimiting species of this genus. Morphological assessments of the different Afrotropical species were done to determine if these species have been accurately identified in recent publications. Using field caught specimens and a museum reference collection together with type comparisons, COI and 16S ribosomal RNA sequences were generated from specimens of *Atylotus agrestis* (Wiedemann), *A. albipalpus* (Walker), *A. fuscipes* (Ricardo) and *A. nigromaculatus* Ricardo. Phylogenetic analysis of these sequences produced four separate species clades with strong support. The results showed that COI does delineate the species of Afrotropical *Atylotus* and that misidentifications of specimens is a common problem. This is of concern as misidentifications have serious implications for management practices.

Numerous new species of nectar feeding *Geranomyia* crane flies (Haliday) (Limoniidae) from a small, forested fragment of Zurqui de Moravia, Costa Rica

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Keywords: Costa Rica, Neotropical, pollination, taxonomic impediment, Tipuloidea, ZADBI

A Diptera inventory project, ZADBI (Zurqui All-Diptera Biodiversity Inventory) took place at Zurqui de Moravia, Costa Rica with the goal of inventorying a megadiverse group of organisms (Diptera) in a small, forested fragment of land. While there were 224 species of crane flies collected primarily at Zurqui, our current research focuses on the genus *Geranomyia* (Haliday) (Diptera: Limoniidae), whose adults visit flowers for feeding on nectar, and larvae develop in wet environments where they feed on algae. We have identified 24 species occurring at Zurqui, with 19 of these species new to science and presumably endemic to this region. Before these sampling efforts, there were only 8 species of *Geranomyia* described from the entire country of Costa Rica. The previously known species at Zurqui all have broad distributions through tropical America. Our work to document and describe these species has been achieved through comparisons to regional species (type specimens and/or original descriptions), photographic imaging of key body parts (particularly wing and leg patterns, and male genitalia), assembling species description pages, and creating a key to the species of *Geranomyia* at Zurqui and for Central America. We have also documented a range of mouthpart lengths across these sympatric species, which might be one factor allowing these congenetics to coexist at Zurqui. This research has been faced with many taxonomic impediments, including scattered species descriptions, few or no illustrations, and no comprehensive taxonomic identification guides, demonstrating the importance and need for more taxonomists and scientific researchers in Diptera studies.

Ultrastructure of antennal sensory organs in Calyptratae

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Keywords: sensilla, sensory pits

Calyptratae flies comprise the largest and ecologically most diverse clade with the schizophoran super-radiation. The antennae are the main olfactory organ of Calyptratae flies, playing crucial roles in their various sensory functions. Compared with other insects, the aristate antennae of Calyptratae flies have been an idea subject for the study of the morphological evolution of insect antennae for their relatively small size, complex structure and high efficiency. Ultrastructure of antennae of Calyptratae from different families were examined with the scanning electron microscope, paraffin section and Laser Scanning Confocal Microscopy. On the antennae of Calyptratae, seven major types of surface sensilla are found and are classified as (1) setiferous plaques, (2) pedicellar buttons, (3) chaetic sensilla, (4) trichoid sensilla, (5) basiconic sensilla, (6) coeloconic sensilla and (7) clavate sensilla. Sensory pits and sacculi with few or clustered sensilla were detected on the postpedicel. These sensory pits and sacculi are vary in size, number and distribution pattern on the postpedicel. Combined, the obligate parasitic species and saprophagous species tend to have more sensory pits than those of other ecotypes. The existence of clustered bottle-shaped sensilla in the sensory pits in all three subfamilies of the sarcophagid species suggests a potential synapomorphy of sarcophagids and a new morphological diagnosis character of the flesh flies. *Gyrostigma rhinocerontis* (Hope), as the largest species of Oestridae, has significantly larger antennae with more sensilla and sensory pits than any other Oestridae species, which could be an adaptation to locate their rare and endangered hosts. A large number of branched or unbranched trichoid and basiconic sensilla are identified on some oestrid flies (*Portschinskia magnifica* Pleske, *Oestromyia leporine* (Pallas)). With the ever-deepening ultrastructure study of this diverse group, more detailed sensilla structures are being found, and their evolutionary adaption and functions need further study.

Population increase and its management of container-inhabiting mosquitoes (Diptera: Culicidae) in northeastern Florida

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Keywords: control, ecology, disease vector, habitat, invasive mosquitoes

Due to fast growth of population and development in St. John's County, Northeastern Florida in the past decades, populations of the container-inhabiting *Aedes* mosquitoes have been growing and increasing, especially *Aedes aegypti* and *Aedes albopictus*, major vectors of dengue fever, Zika, yellow fever, and other viruses. Also, West Nile virus vector, *Culex quinquefasciatus*, another contain-breeding mosquito population has been increased. Due to the increases of the disease vector mosquito populations and their speed development of resistance to insecticides, the control and management of the vector mosquitoes have met and faced many new challenges and difficulties. Anastasia Mosquito Control District has redirected the budget emphasis and increased and enhanced the surveillance of mosquito populations and adopted several new technologies to control and manage the situation and prevent the potential public health threats caused by the increased populations of container-inhabited vector mosquitoes, such as, increasing numbers of BG sentinel traps, mass deployment of autocidal gravid traps for control, release of SIT radiated male mosquitoes and *Wolbachia*-infected male mosquitoes for control of natural population. Due to many efforts and promotion of the best management practices, the District has been successfully preventing from further spreading and possible outbreak of mosquito-borne diseases after several imported cases presented in our area.

**Phylogeny of Oestridae (Calyptratae: Oestroidea):
evidence from transcriptomes**

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Keywords: phylogeny, transcriptomes

Bot flies (Calyptratae: Oestridae) are obligate endoparasites of mammals, living (sub)dermally (Cuterebrinae and Hypodermatinae), in the nasopharyngeal cavities (Oestrinae) or in the intestinal tract (Gasterophilinae) of their hosts. Their extraordinary diversification is therefore of great importance in understanding the evolution of parasitism. However, phylogeny of Oestridae has been in debate depending on the characters or data used for phylogeny reconstruction. With the emerging of phylotranscriptomic data and advances in genomic data processing, it is possible to target thousands of genes for phylogenetic analysis. In this study, we used transcriptomic data to reconstruct the phylogeny of bot flies and hopefully reach a better phylogenetic resolution of the oestrids.

Settling a half-a-century-old debate: a revision of the family Pseudopomyzidae (Diptera, Acalyptratae) with a reclassification of the family as part of the Cypselosomatidae

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Keywords: comparative morphology, family revision, new species, phylogeny, reclassification, taxonomy

Pseudopomyzidae (Nerioidea, Diptera) are relatively uncommon but charismatic flies traditionally treated as a family of Nerioidea although the monophyly, family status and superfamily placement of the group have been debated for decades. A new revision of the family and a reconsideration of the relationships among the families of Nerioidea on the basis of morphological characters and multiple genes indicate that the family Pseudopomyzidae is paraphyletic with respect to Cypselosomatidae. These families should therefore be treated together as the Cypselosomatidae.

Diversity, distribution and DNA-barcoding of forensically important blow flies (Diptera: Calliphoridae) in the Caribbean Region

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Keywords: Caribbean, diversity, distribution, DNA barcoding, forensic entomology

Blow flies (Diptera: Calliphoridae) are among the most dominant and conspicuous insects in the decomposition process. They are important in forensic entomology to determine time of death and, in certain situations, cause of death or relocation of a body. Insects are now included as standard operating procedures in crime scene investigations in many countries, however, this is not standard procedure in the Caribbean area due to lack of knowledge of insects involved in cadaveric decomposition. Successful application of forensic entomology depends on solid underlying data. Our main goal is to advance the knowledge of Calliphoridae in the Caribbean to enable forensic entomology studies. We performed a mega-transect across the Caribbean and extensively collected flies attracted to rotten meat baits during five years from 2011 to 2015. Overall, we collected 61,332 flies of which 34,650 were Calliphoridae. We morphologically identified freshly collected specimens, did phylogenetic analyses and employed several species delimitation methods for a total of 468 individuals representing 19 described species. Our results show that endemic species are the most abundant and dominant species in each island. Molecular data shows that combination of COI + ITS2 genes yields more accurate identification and diagnoses, and better agreement with morphological data, than the mitochondrial barcodes alone. All of our results from independent and concatenated trees and most of the species delimitation methods yield considerably higher diversity estimates than the distance-based approach and morphology. Molecular data support at least 24 distinct clades within Calliphoridae recovering substantial geographic variation for *Lucilia eximia* (Wiedemann), *Lucilia retroversa* (James), *Lucilia rica* (Shannon) and *Chloroprocta idiodea* (Robineau-Desvoidy) probably indicating several cryptic species.

Evolution and biogeography of blow flies (Diptera: Calliphoridae) from the Caribbean Region

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Keywords: biogeography, Caribbean, evolution, forensic entomology

Calliphoridae is a species-rich family of Diptera important to the field of forensic entomology. Calliphorids are abundant throughout the Neotropics, however their phylogenetic relationships and geographic distributions throughout the Caribbean are still being resolved. The Caribbean Island archipelago comprises a complex geologic history, with both continental, volcanic, and sedimentary islands forming. Historically calliphorids have been categorized as good dispersers throughout the Caribbean, suggesting possible intermediate dispersal and/or reflecting species-specific habitat suitability. Intermediate dispersers tend to have evolutionary histories that mirror geologic histories, making them useful for better understanding biogeographic histories as their dispersal does not obfuscate ancient geologic signatures. We tested dispersal three hypotheses into the Caribbean Islands including (1) a single over-water dispersal into the islands and subsequent dispersals among islands, (2) multiple dispersals to the islands from the mainland across geologic time scales, and (3) Calliphoridae dispersed into the Greater Antilles via GAARlandia, a land-bridge hypothesized to have existed 33-35 million years ago. Bayesian and Maximum likelihood methods were implemented on a Standard Sanger Sequencing dataset with four molecular markers (COI, Ef-1a, ITS2 and 28s) to infer phylogenetic relationships across mainland and islands Calliphoridae. We estimated divergence times and ancestral ranges across while including parameters to account for the historical geologic changes across the Greater and Lesser Antilles. We found evidence for both endemism and widespread species within Caribbean Calliphoridae as well as multiple over-water dispersals into the Caribbean. This study is the first biogeographic analysis of Calliphoridae in the Caribbean and is important for better understanding the evolution of this group in in this region.

An unusual myiasis by *Lucilia eximia* (Wiedemann) (Calliphoridae) in a dog and evidence of mistreatment: case report and identification key for Neotropical third-instar larvae

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Keywords: blow flies, domestic animal, forensic entomology, identification, key, mistreatment, Neotropical, parasitism

Due to the economic losses associated with parasitism and also because it can affect humans, myiasis receives more attention in the areas of livestock and public health. In the forensic field, infestations have been reported to help identify cases of negligence, in general, involving people with some fragility. In the same way, we report an unusual case of infestation by *Lucilia eximia* (Wiedemann) larvae in a dog in Southeast Brazil, where it was possible to identify a situation of mistreatment. A female adult mixed breed dog, weighing approximately three kilograms, was found inside a raffia pack abandoned in a vacant lot in an urban area of a city of Minas Gerais, Brazil. The examining veterinarian reported paresis, mucous membrane paleness, ascites, feces accumulated in the perianal region and cachexia. It was also reported superficial lumbar, sacral and perineal wounds, and deep rectum and posterior limbs wounds, with tendons and bones exposure, especially in the tarsal region. Dipteran larvae were removed from wounds. A portion of the collected larvae were deposited in 50 mL plastic vials with 70% ethanol, and another portion were separated for rearing on fresh ground beef under controlled conditions for confirmation of identification. Larvae and emerged adults were examined in a stereomicroscope and identified. Taking into account morphological characteristics such as weight and larval length, it was possible to estimate an infestation interval between four and five days. To help forensic experts and other animal health practitioners with the diagnosis of larvae and the identification of cases of mistreatment with domestic animals we present an identification key for Neotropical third-instar larvae of myiasis-causing flies and, for the first time, guidelines and standard operating procedures for the collection of entomological samples that may be used as criminal evidence.

A taxonomic review of Canadian Oestridae (Calypttratae: Oestroidea)

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Keywords: bot fly, identification, key, parasite, taxonomy, warble flies

Oestridae, known as bot flies and warble flies, are a small group of flies whose larvae are obligate mammal parasites that cause myiasis in their host. Oestrid parasitism of domesticated livestock has been well-documented by agricultural and veterinary entomologists, however, research on oestrids that primarily parasitize wild mammals remains understudied. In Canada, Oestridae taxonomy has not been studied for the past 40 years, and the absence of an identification key makes species determination insufficient and questionable. Our research aims at filling in this knowledge gap and aims at diagnosing the species by adult morphology. An extensive examination of Canadian Oestridae includes more than 1900 specimens and 150 photos. We propose new diagnostic characters to aid in the identification of Canada's 17 recognized species of Oestridae and provide an extensively illustrated dichotomous key based on external morphology. This project also establishes a full database of all examined specimens, contributing crucial information on the biogeography and incidence of Oestridae in Canada. The updated identification resources of the Canadian Oestridae will facilitate further research of this fauna and support research in parasitology, ecology, veterinarian, zoology and among other fields.

Evolution and diversification of snail-killing flies (Diptera, Sciomyzidae)

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Keywords: feeding behavior, macroevolution, parasitoid, phylogenomics, Sciomyzoidea, snail-killing flies

Parasitoids feed inside or on another organism to complete their development, resulting in the host's death. Parasitoidy is present in different taxonomic groups, ranging from insects, fungi, to nematomorphs. The unique ecological and evolutionary aspects of the parasitoid habit, as well as their specialized host associations, warrant a detailed investigation of the evolution of their feeding behaviors. Insects are the most diverse parasitoids, of which the majority are wasps (Hymenoptera) and flies (Diptera). Diptera is an excellent biological model for the study of parasitoids, mostly due to the multiple independent origins of parasitoidy within the group and the diverse behaviors and morphologies they exhibit. Snail-killing flies (family Sciomyzidae) are parasitoids of snails (Gastropoda) and may play an important role in the biological control of schistosomiasis and fascioliasis. This study aims to understand the evolution of parasitoidy and host associations in sciomyzids, in order to advance our knowledge of their evolutionary biology and natural history. This study is the first to use phylogenomics and macroevolutionary analysis in Sciomyzidae.

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We gratefully acknowledge the support of the numerous sponsors below, as well as the individual sponsorships of several people, including and Mike & Bonnie Irwin and Terry & Faye Whitworth.



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https://dipterists.org/assets/PDF/flytimes_supplement05.pdf
Edited by Stephen D. Gaimari
ISSN 2769-6111 (print)
ISSN 2769-612X (online)