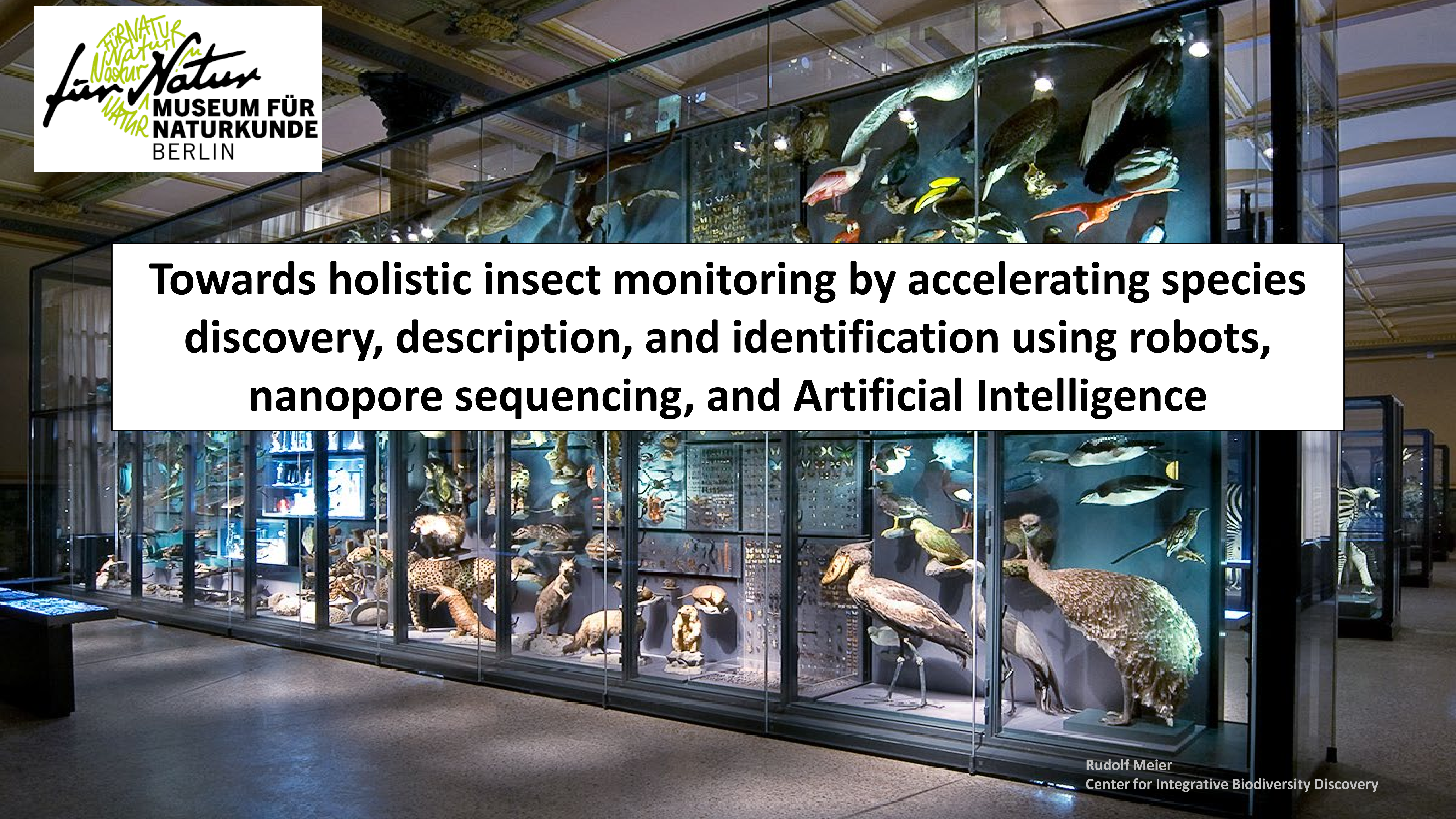
MUSEUM FÜR NATURKUNDE
BERLIN



Towards holistic insect monitoring by accelerating species discovery, description, and identification using robots, nanopore sequencing, and Artificial Intelligence

Rudolf Meier
Center for Integrative Biodiversity Discovery

Biodiversity research: 1990s versus 2020s

1990s: “Ivory Tower” discipline for biologists out of touch with modernity

2020s: Biodiversity loss threatens planetary health and society now cares

Who cares?

The Global Risks Report 2023

18th Edition

WORLD ECONOMIC FORUM

1	Failure to mitigate climate change
2	Failure of climate-change adaptation
3	Natural disasters and extreme weather events
4	Biodiversity loss and ecosystem collapse
5	Large-scale involuntary migration

Who else?

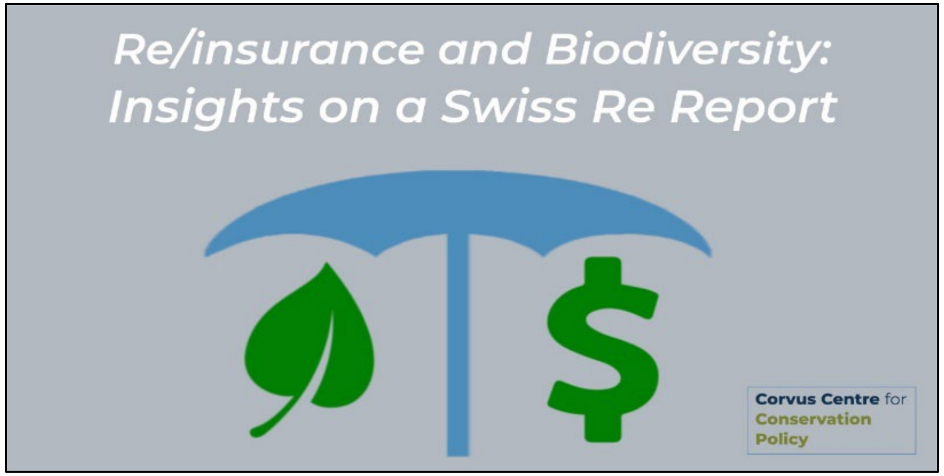
Goldman Sachs

GS SUSTAIN

Biodiversity

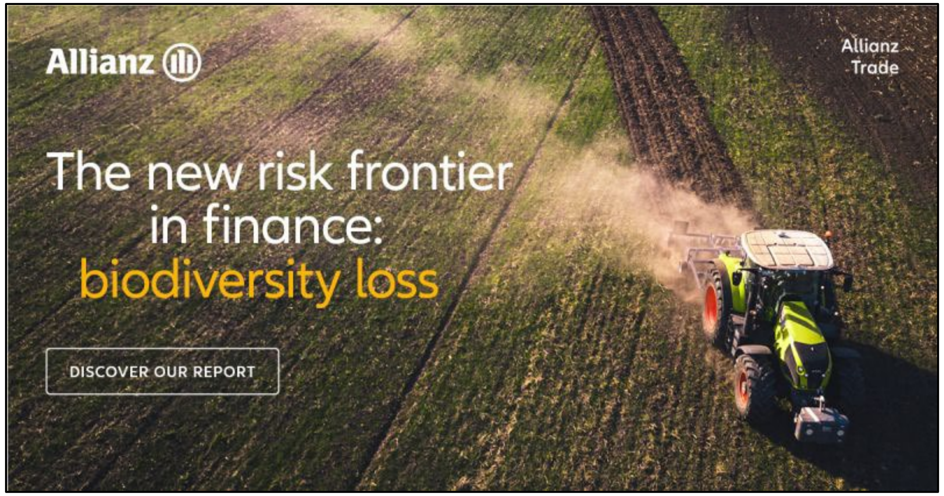
Assessing the Financial Links to Natural Capital

Biodiversity is the cornerstone around which most sustainability objectives sit and an area to which we think corporates and investors will likely pay increased attention in order to meet sustainability and economic



Re/insurance and Biodiversity: Insights on a Swiss Re Report

Corvus Centre for Conservation Policy



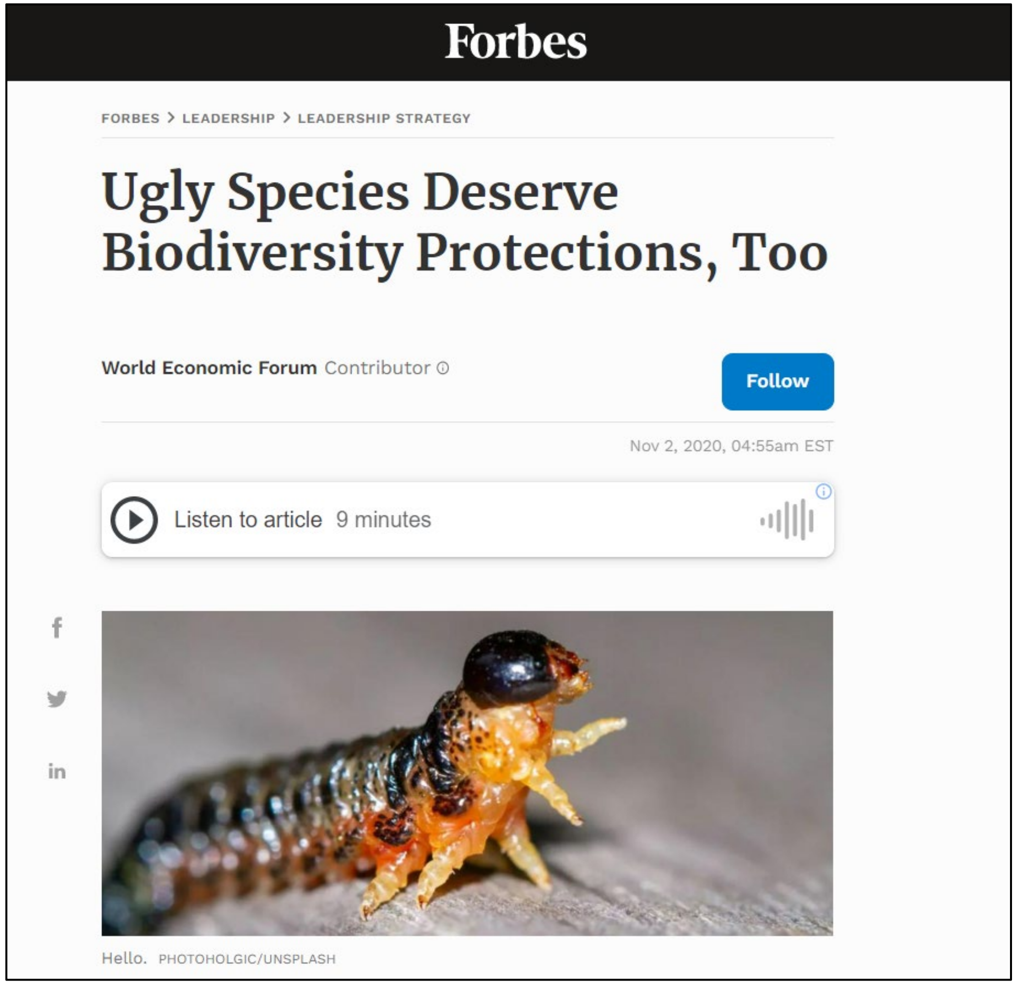
Allianz

Allianz Trade

The new risk frontier in finance: biodiversity loss

DISCOVER OUR REPORT

But is society interested in “Ivory Tower biodiversity”?



Forbes

FORBES > LEADERSHIP > LEADERSHIP STRATEGY

Ugly Species Deserve Biodiversity Protections, Too

World Economic Forum Contributor

Nov 2, 2020, 04:55am EST

Hello. PHOTOHOLGIC/UNSPLASH

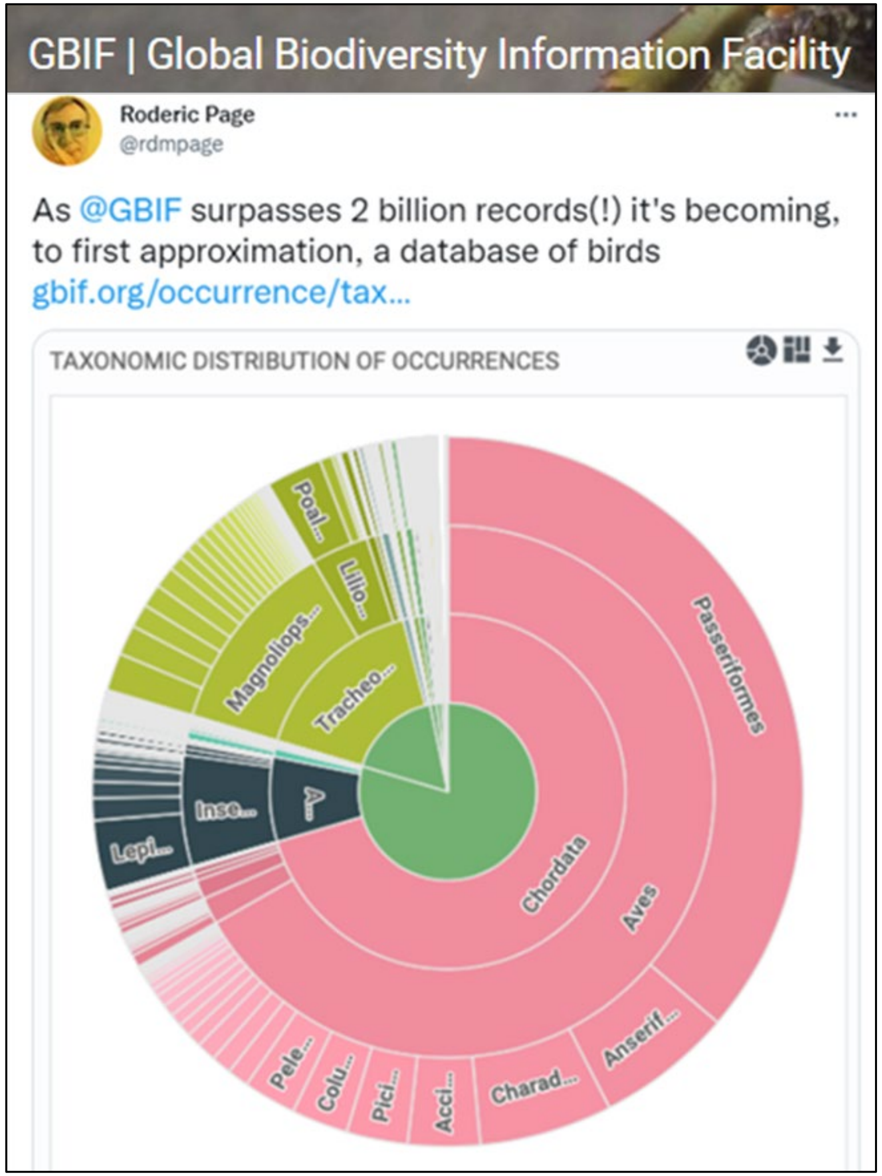
1990s: Ivory Tower taxon biases:

- pretty
- pretty large
- pretty diurnal
- pretty endangered
- pretty impressive on a tree
- pretty manageable taxonomically
- pretty unusual in many other ways…

What is missing is:

- pretty important but ugly…

How bad is the data gap for the uglies?



GBIF | Global Biodiversity Information Facility

Roderic Page
@rdmpage

As @GBIF surpasses 2 billion records(!) it's becoming, to first approximation, a database of birds gbif.org/occurrence/tax...

TAXONOMIC DISTRIBUTION OF OCCURRENCES

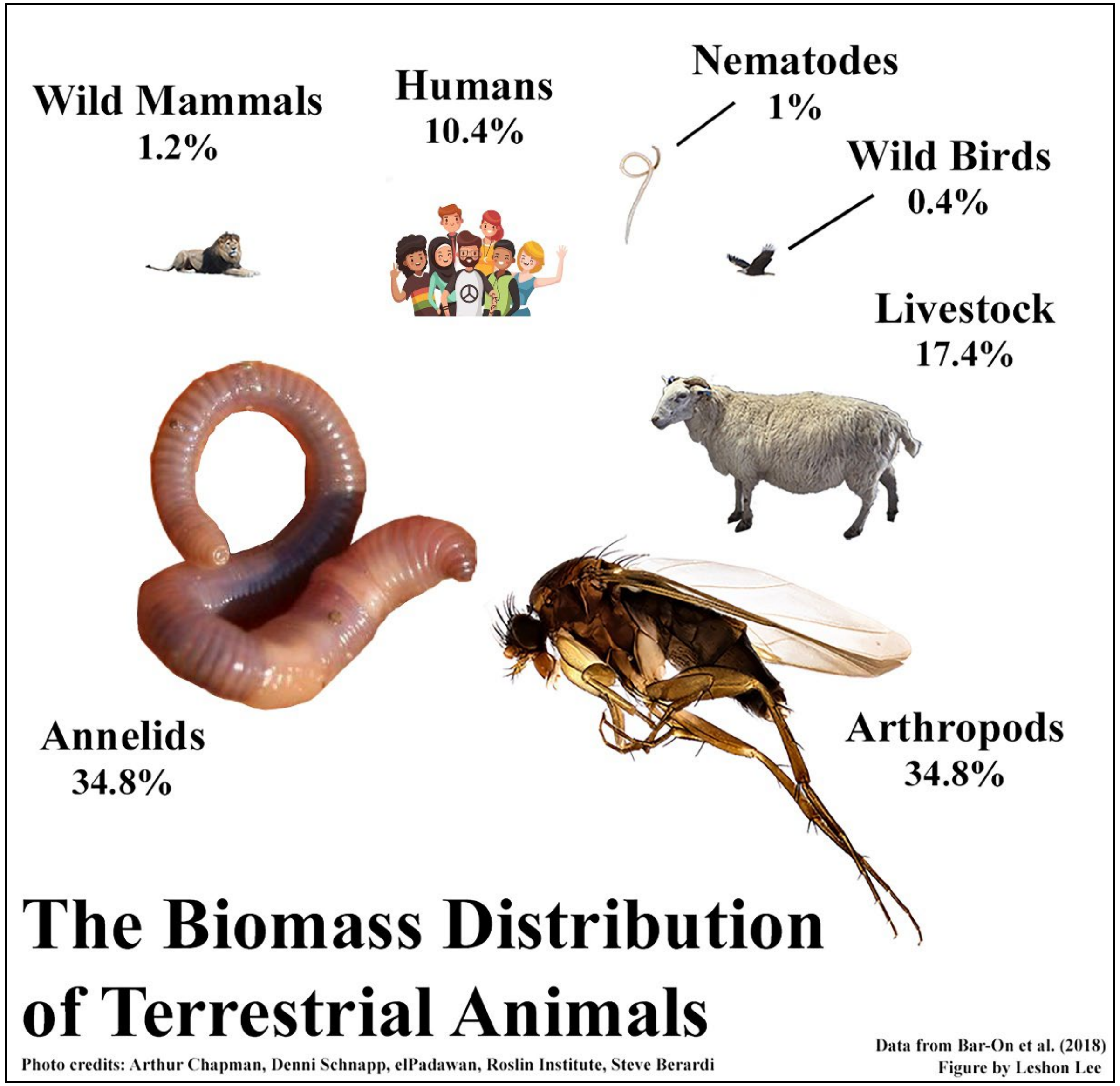
2024 update:

- 2.5 billion occurrence records
- ca. 2 billion for bird species
- Insecta: 238 million
    - 42 million butterfly records
    - top 10 bird species: 230 million

=> we need a new field:

- Ugly Zoology

Terrestrial animal biodiversity: biomass



The Biomass Distribution of Terrestrial Animals

Photo credits: Arthur Chapman, Denni Schnapp, elPadawan, Roslin Institute, Steve Berardi

Data from Bar-On et al. (2018)
Figure by Leshon Lee

Terrestrial animal biodiversity: species diversity

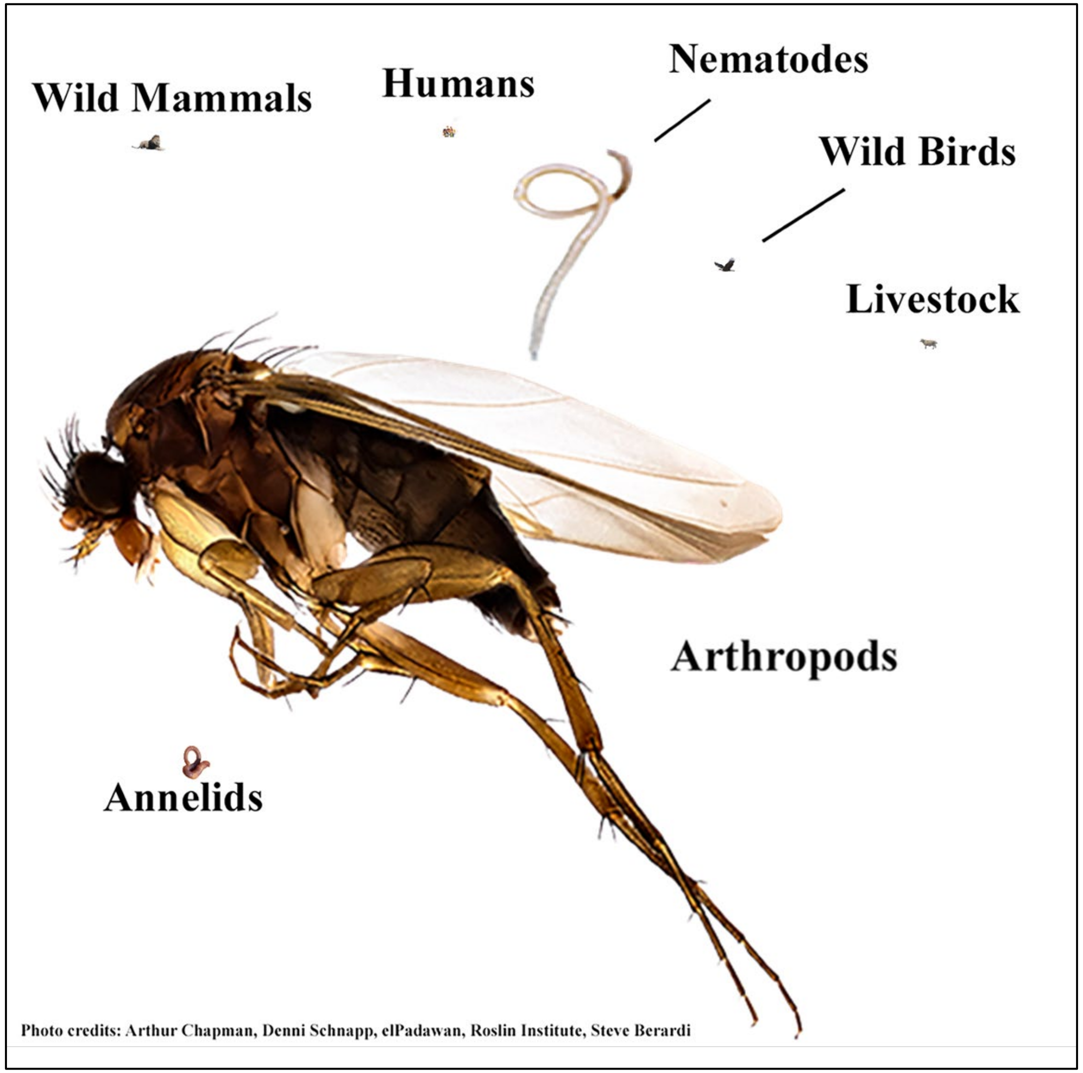


Photo credits: Arthur Chapman, Denni Schnapp, elPadawan, Roslin Institute, Steve Berardi

Shortcomings of the old ways…

- >80% of all species undescribed

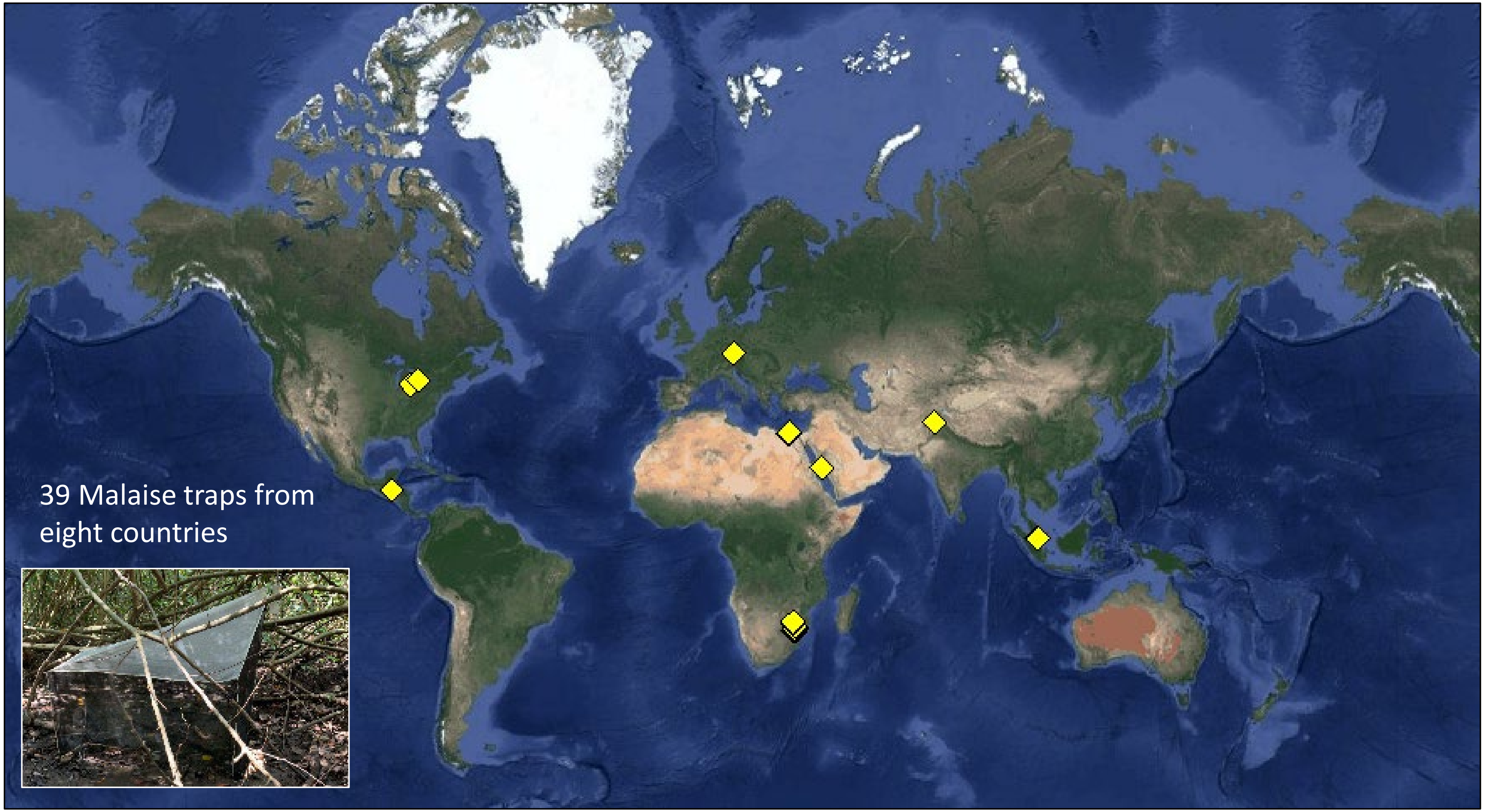
- 95% of all described species only identifiable for taxonomic experts

- very little baseline biodiversity data from standardized samples

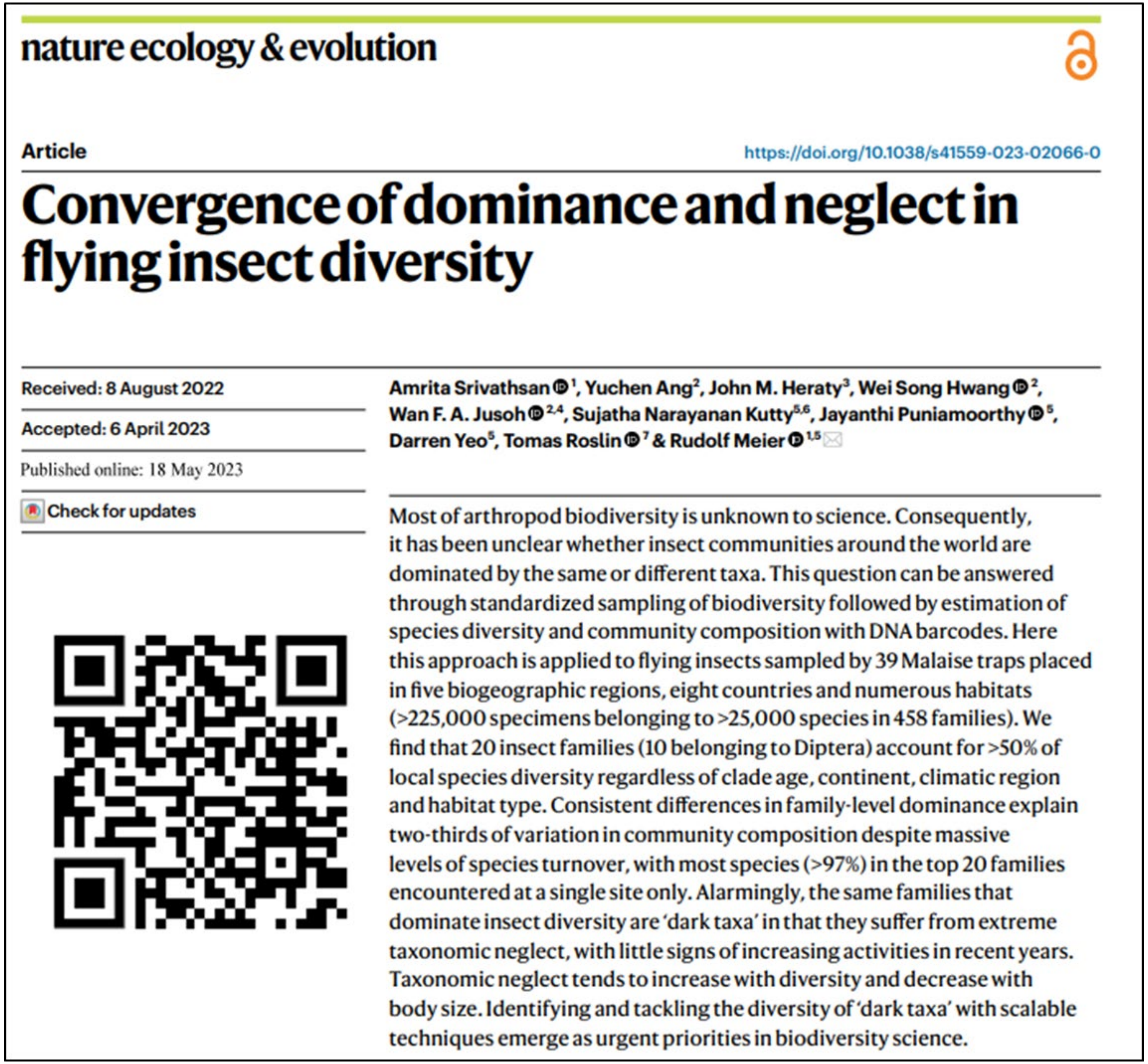
- natural history data only available for <5% of species

Triage: which taxa need our attention?

Dr. Amrita Srivathsan



39 Malaise traps from eight countries



nature ecology & evolution

Article https://doi.org/10.1038/s41559-023-02066-0

Convergence of dominance and neglect in flying insect diversity

Received: 8 August 2022

Accepted: 6 April 2023

Amrita Srivathsan[1], Yuchen Ang[2], John M. Heraty[3], Wei Song Hwang[2], Wan F. A. Jusoh[2,4], Sujatha Narayanan Kutty[5,6], Jayanthi Puniamoorthy[5], Darren Yeo[5], Tomas Roslin[7] & Rudolf Meier[1,5]

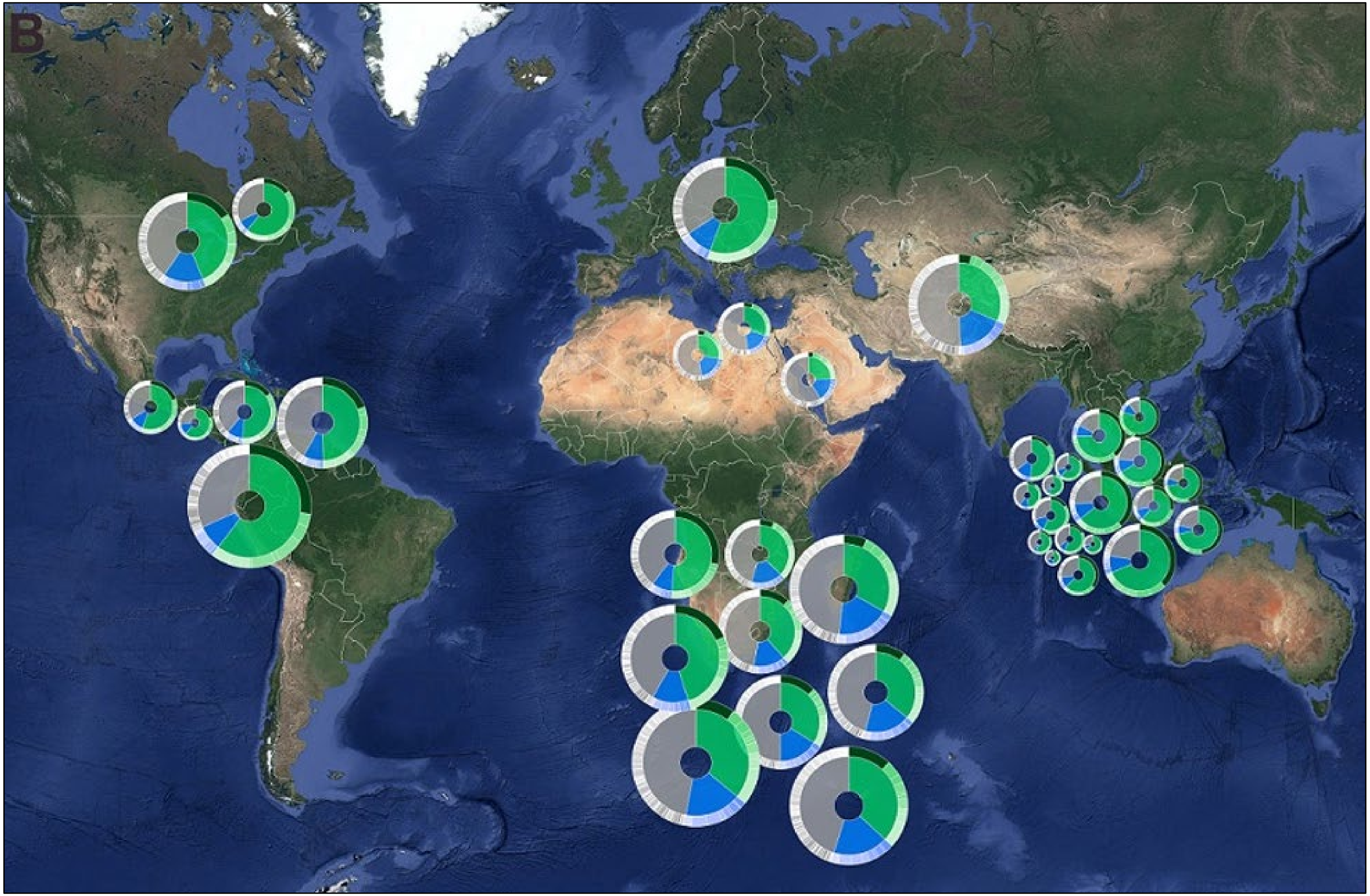
Most of arthropod biodiversity is unknown to science. Consequently, it has been unclear whether insect communities around the world are dominated by the same or different taxa. This question can be answered through standardized sampling of biodiversity followed by estimation of species diversity and community composition with DNA barcodes. Here this approach is applied to flying insects sampled by 39 Malaise traps placed in five biogeographic regions, eight countries and numerous habitats (>225,000 specimens belonging to >25,000 species in 458 families). We find that 20 insect families (10 belonging to Diptera) account for >50% of local species diversity regardless of clade age, continent, climatic region and habitat type. Consistent differences in family-level dominance explain two-thirds of variation in community composition despite massive levels of species turnover, with most species (>97%) in the top 20 families encountered at a single site only. Alarmingly, the same families that dominate insect diversity are 'dark taxa' in that they suffer from extreme taxonomic neglect, with little signs of increasing activities in recent years. Taxonomic neglect tends to increase with diversity and decrease with body size. Identifying and tackling the diversity of 'dark taxa' with scalable techniques emerge as urgent priorities in biodiversity science.

Dr. Amrita Srivathsan

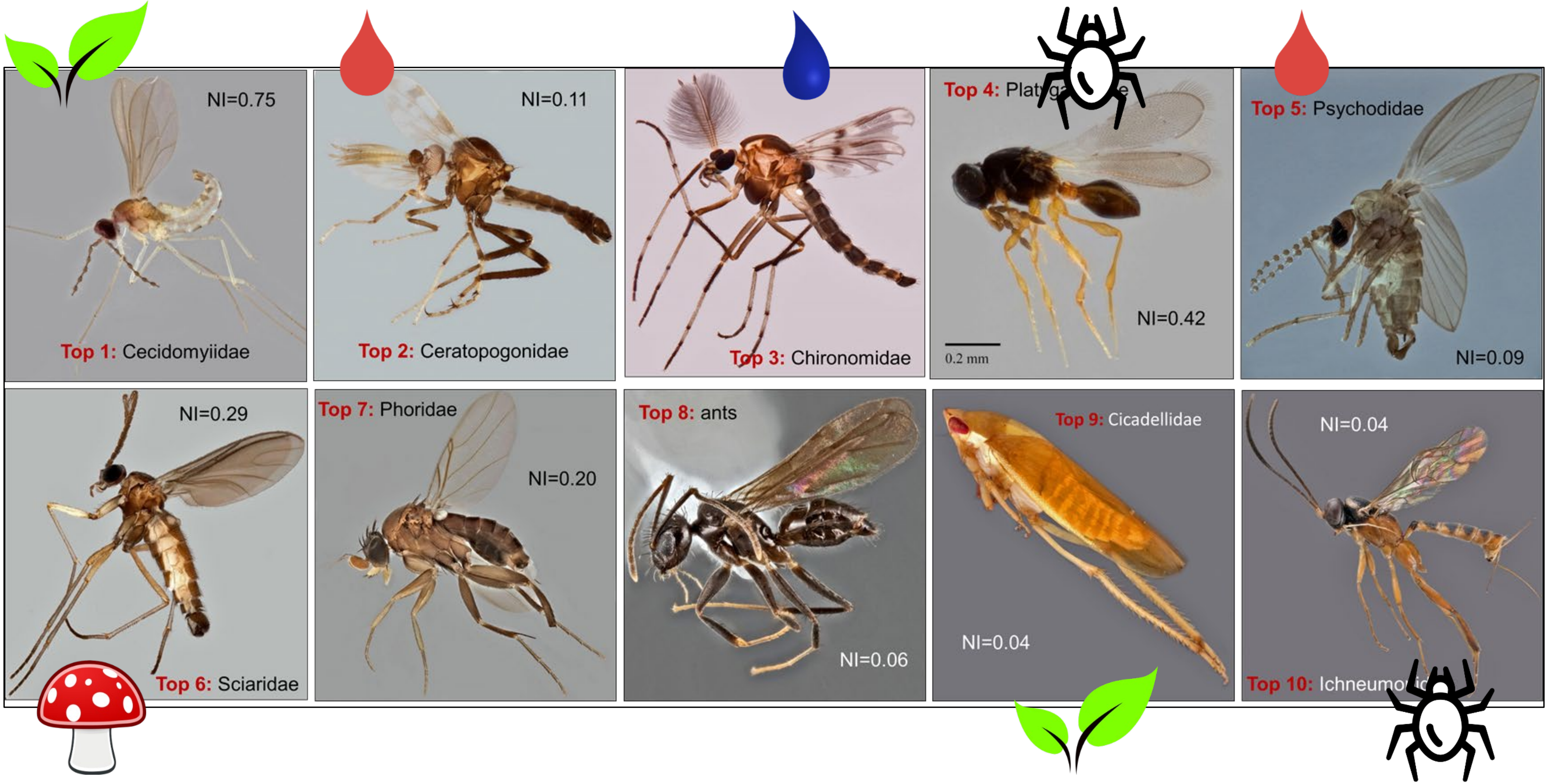
=> same 20 family-ranked insect clades contribute >50% of species diversity

=> same 20 clades also account for >50% of specimen abundance

Habitat, climate, etc. don't matter

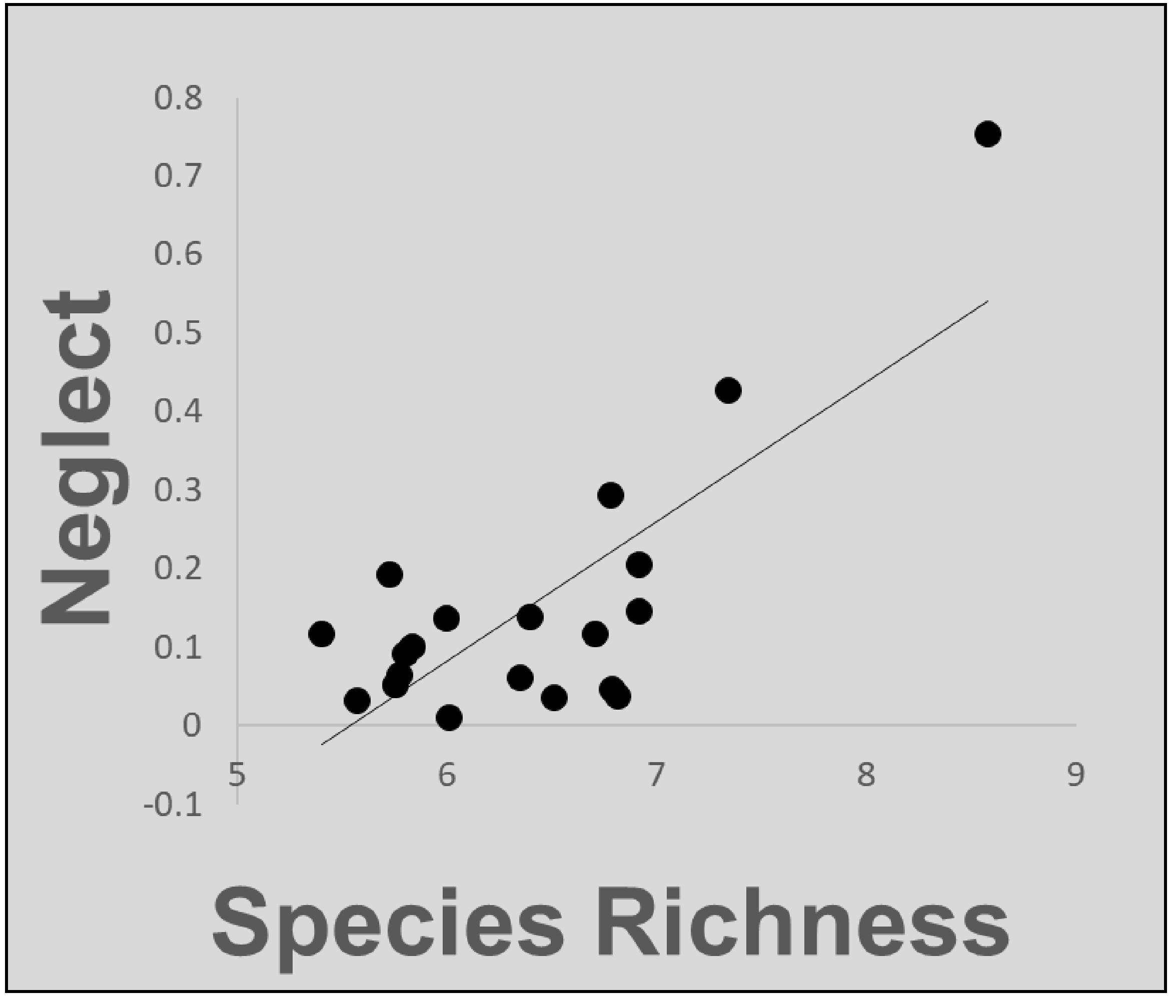

B

The top 10


NI=0.75
Top 1: Cecidomyiidae
NI=0.11
Top 2: Ceratopogonidae
Top 3: Chironomidae
Top 4: Platyg e
NI=0.42
0.2 mm
Top 5: Psychodidae
NI=0.09
NI=0.29
Top 6: Sciaridae
Top 7: Phoridae
NI=0.20
Top 8: ants
NI=0.06
Top 9: Cicadellidae
NI=0.04
NI=0.04
Top 10: Ichneumo

Most of these top taxa suffer from taxonomic neglect

Dr. Amrita Srivathsan



'Neglect Index' (NI): ratio between the number of mOTUs found across Malaise traps and number of species described

1. Neglect increases with species richness
2. Neglect is not improving over time
3. Small body size leads to neglect

Clearly, we need "Ugly Zoology"!

But we also need rebranding…

“Dark Taxon Zoology”: resolving biology’s dark matter

Roderik Page in 2011: “dark taxa”

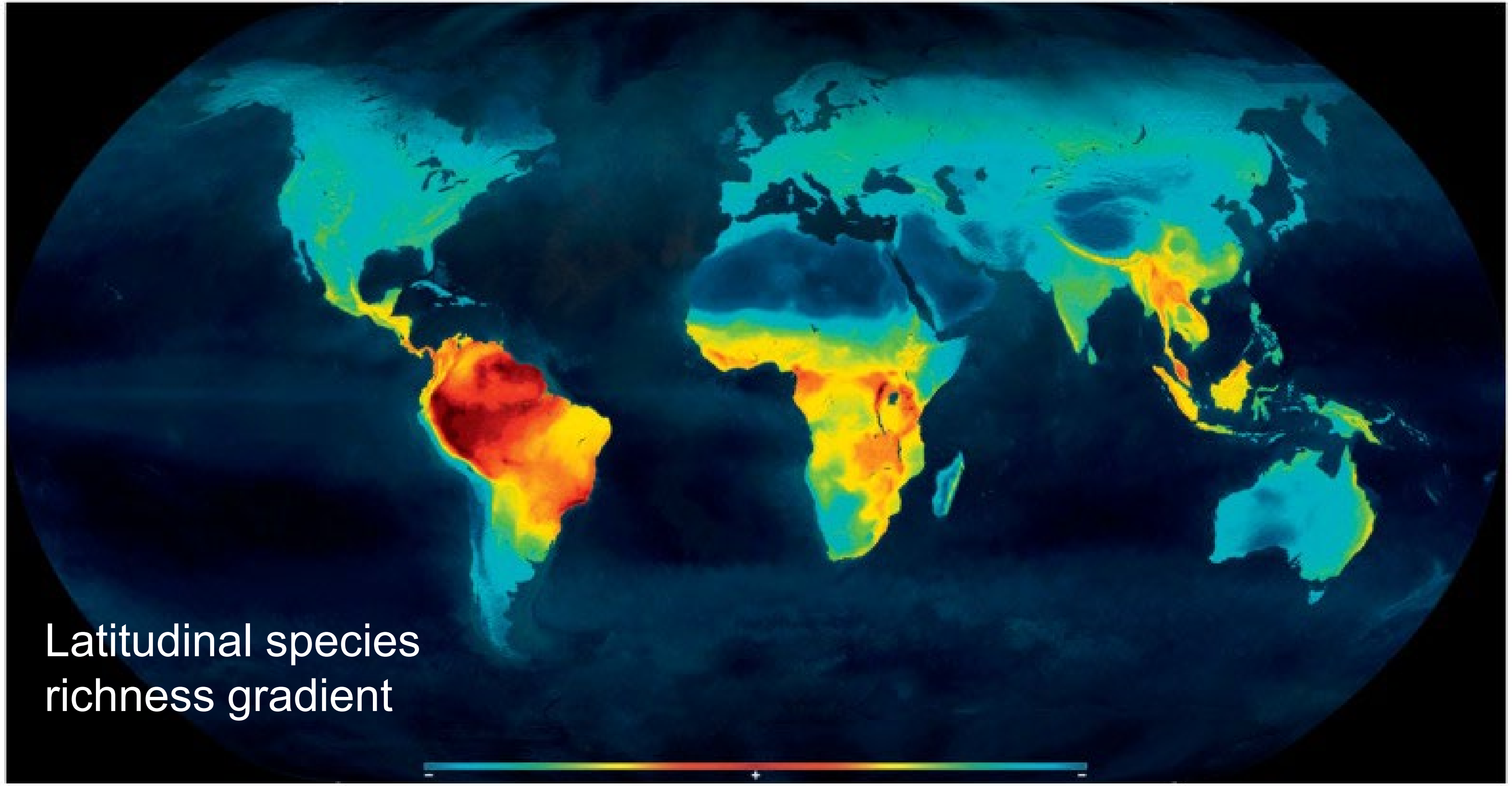
1. groups of organisms present in genetic databases
2. lacking formal scientific names

Hartop et al. 2022: “dark taxon”

1. clade whose diversity exceeds 1,000 species
2. of which fewer than 10% of species described

Biodiversity data crisis: more problems…

Geographic biases in biodiversity data

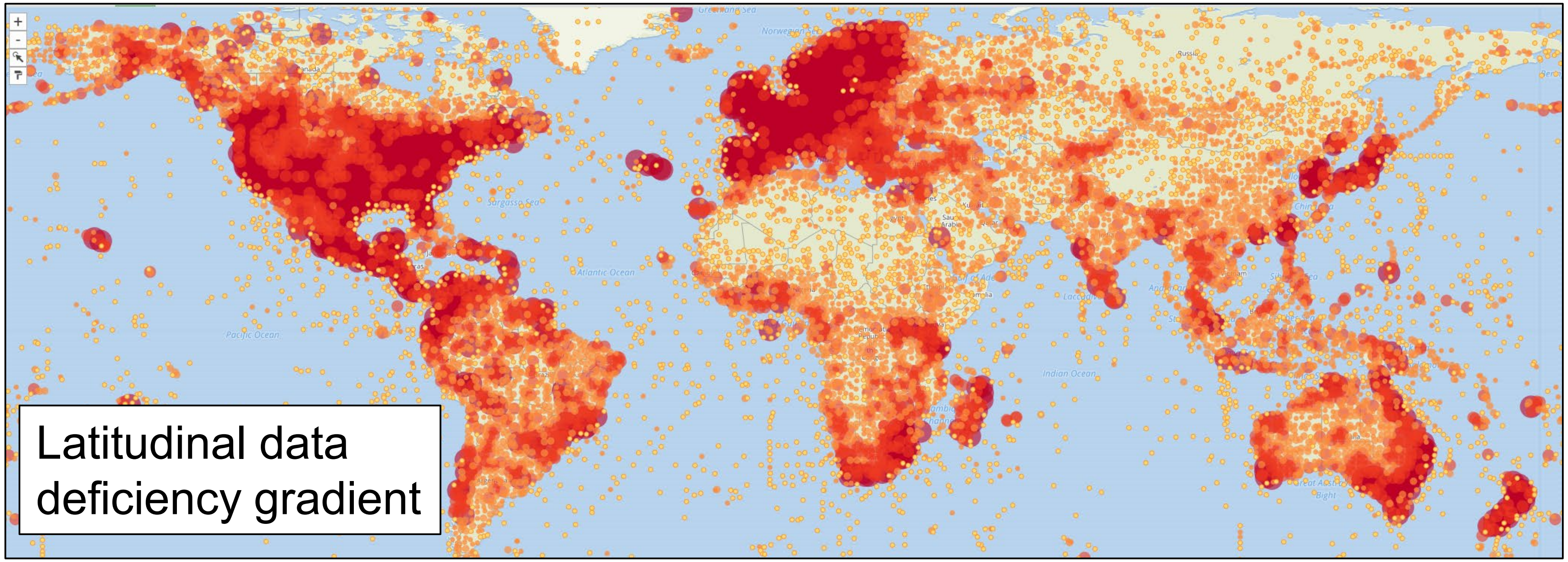


Latitudinal species richness gradient

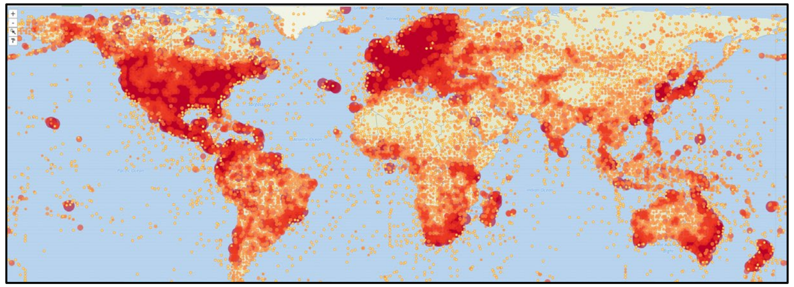
TRENDS in Ecology & Evolution

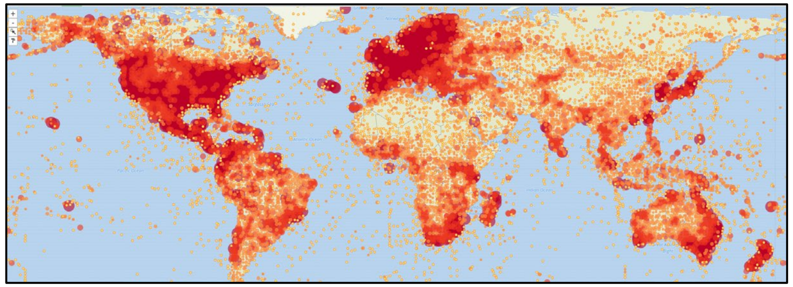
GBIF | Global Biodiversity Information Facility

Insect data in GBIF



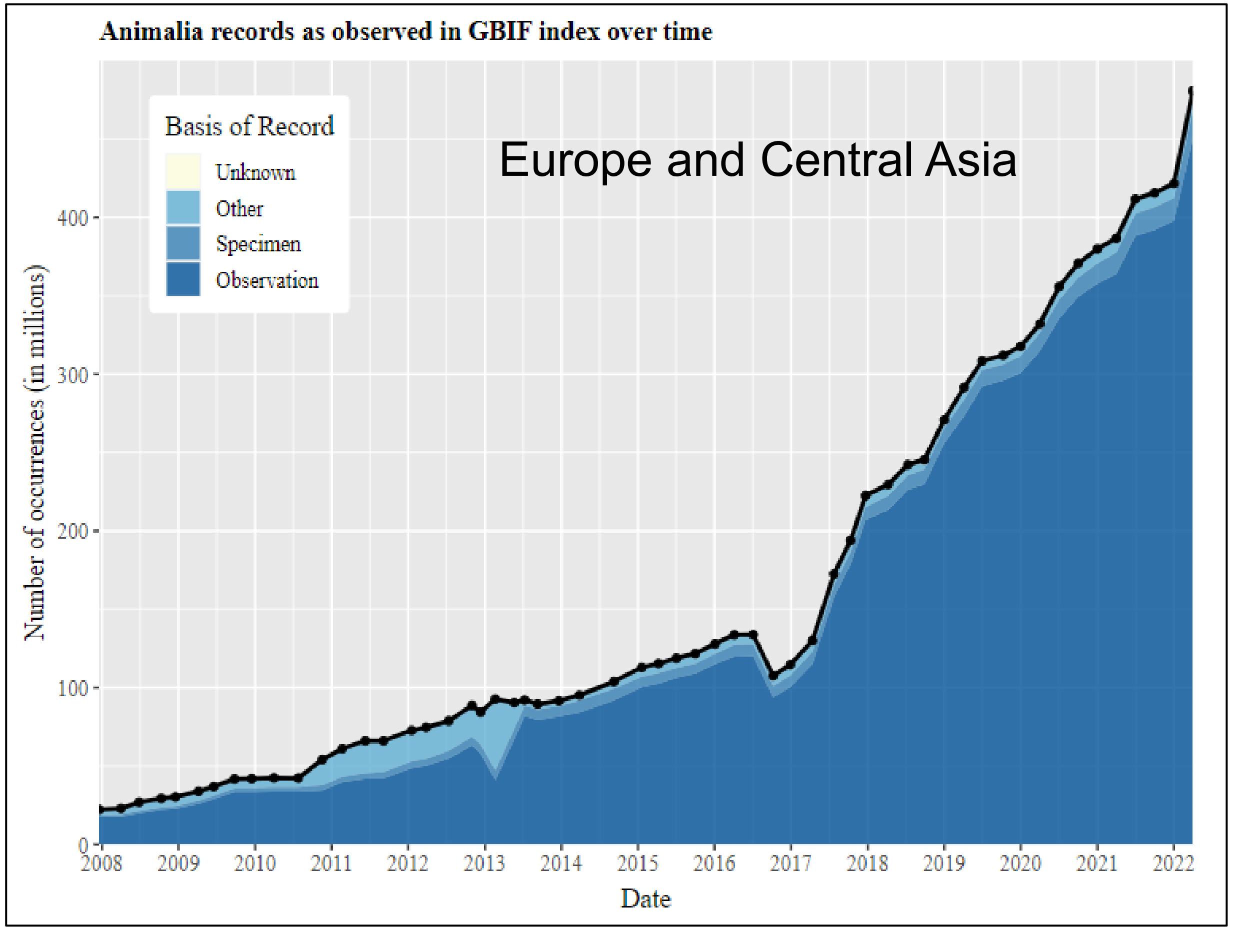
Latitudinal data deficiency gradient

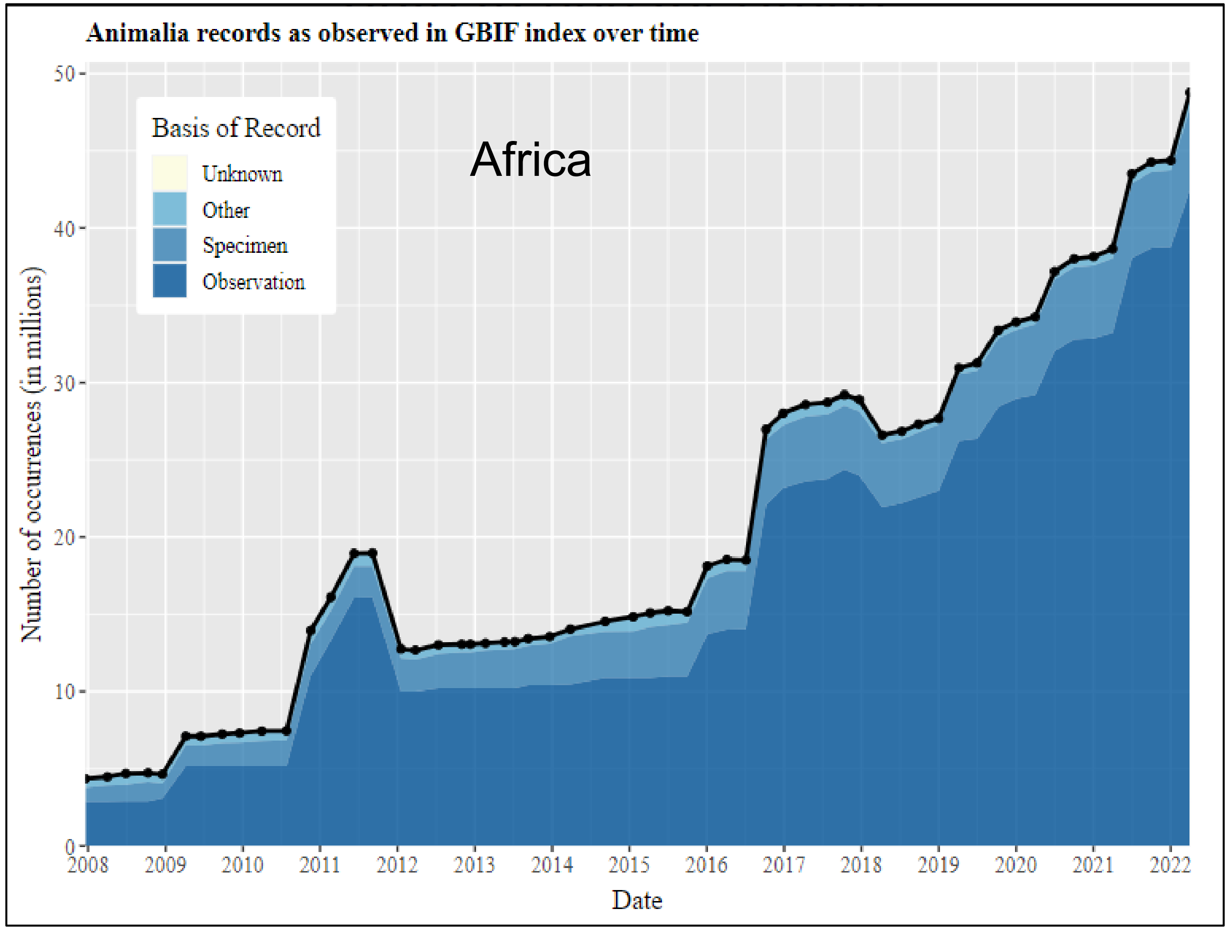




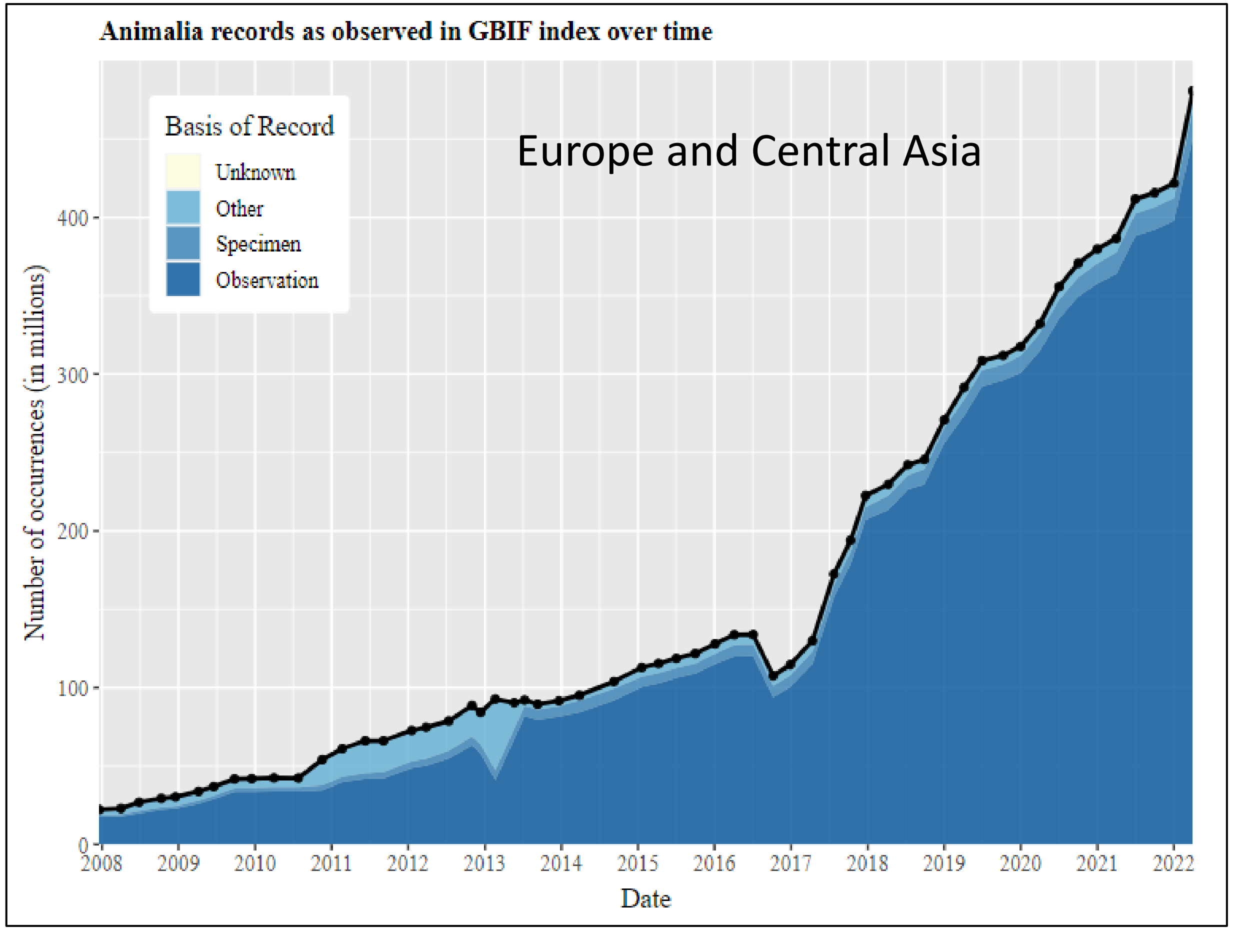
20% of insect data are for the UK

Is the situation improving?

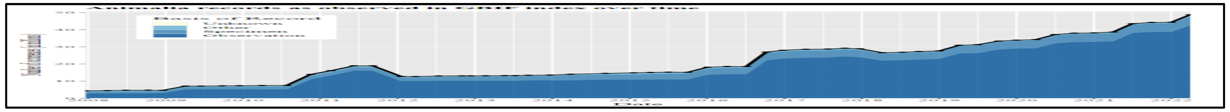




Is the situation improving?


Europe and Central Asia

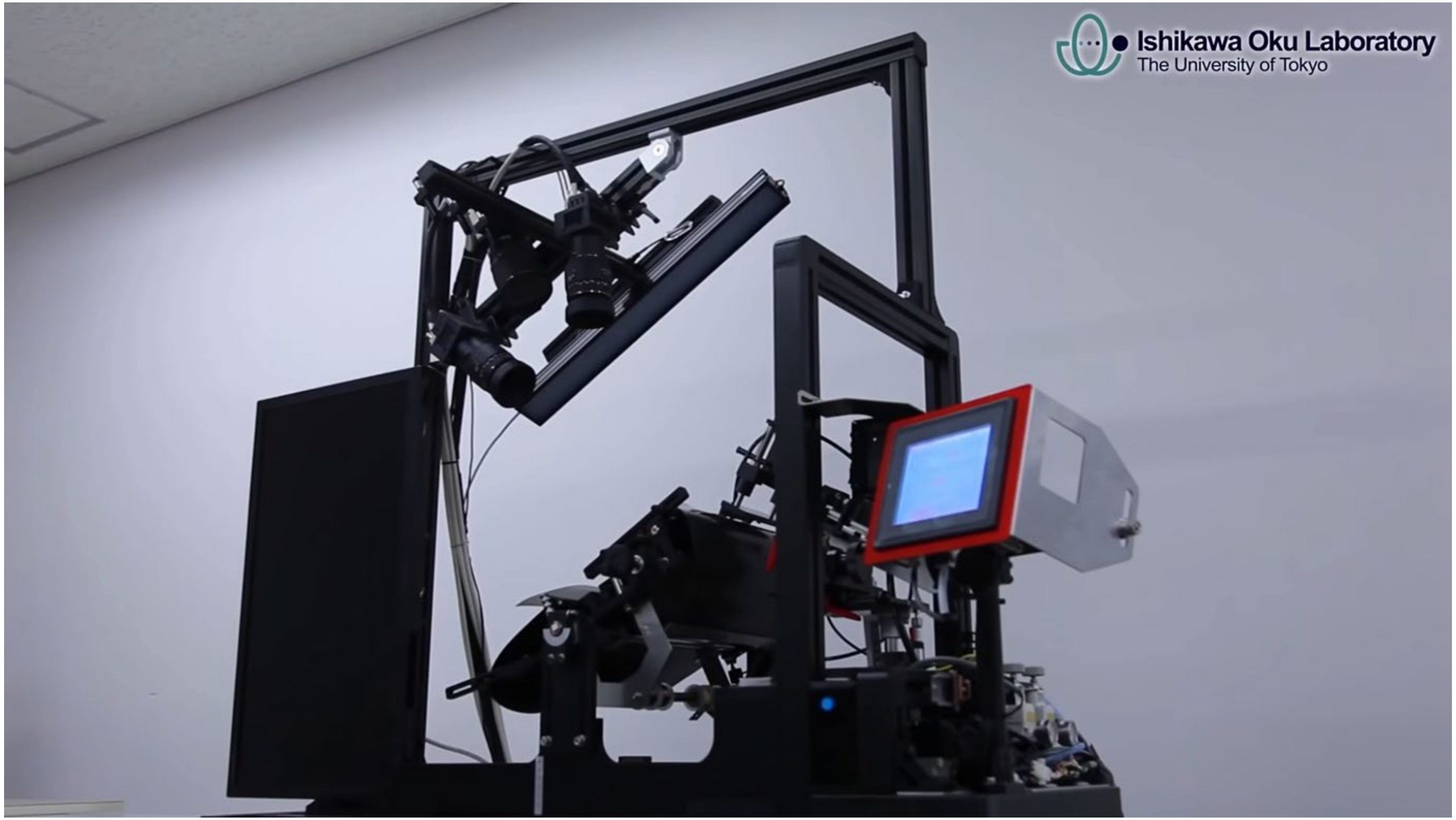
Africa

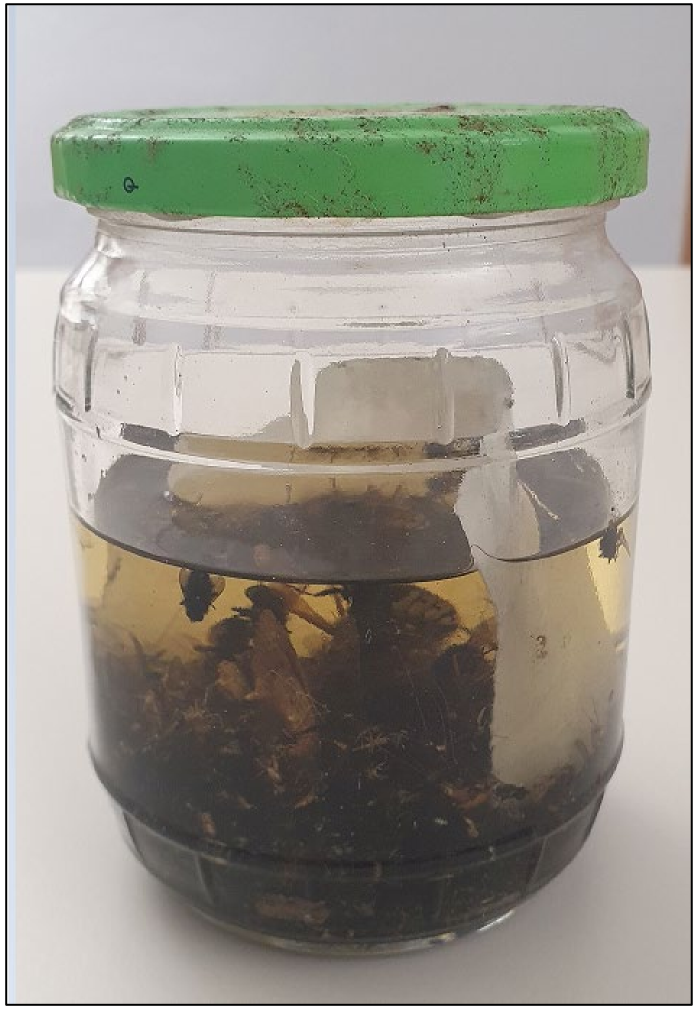


Biodiversity data crisis

- we need to generate more data fast
- data should be for standardized samples

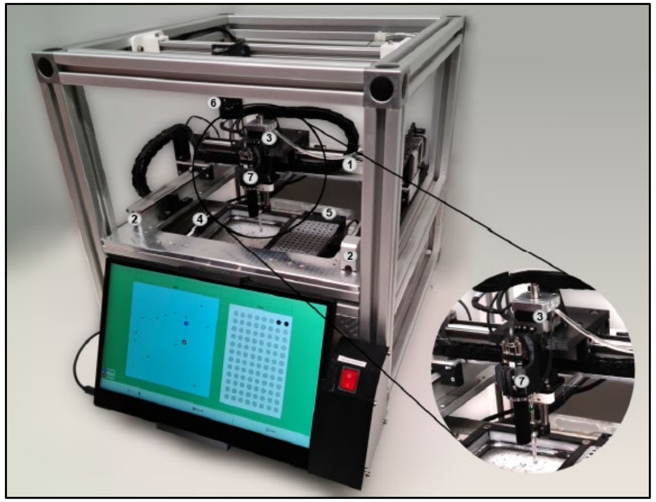
Let’s analyze biodiversity samples like libraries





Species Discovery Factory

1. DiversityScanner:
Digitization with robots



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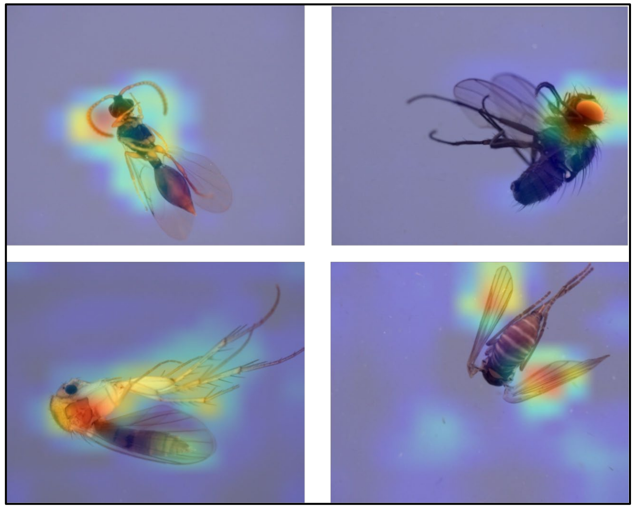
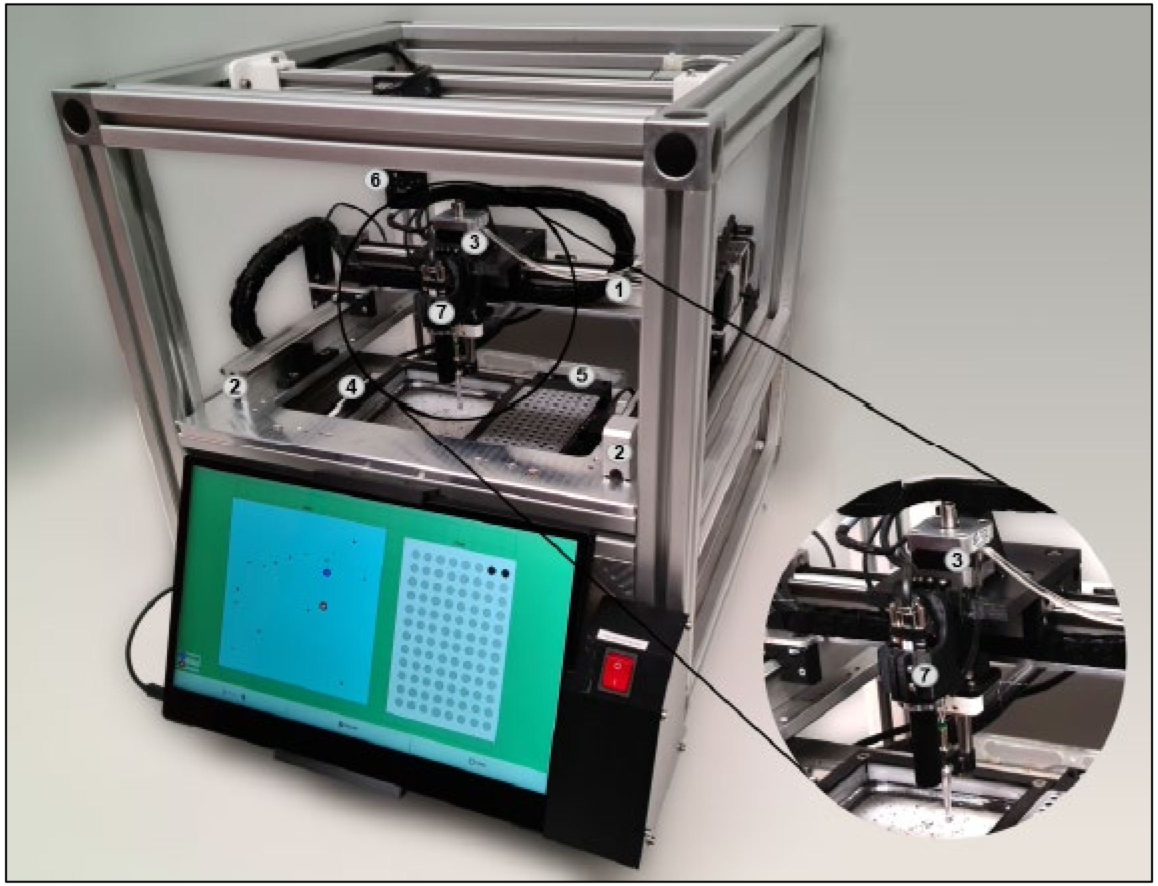
~~**2. Nanopore Sequencing:**~~
~~Sorting with DNA barcodes~~

=

3. Identified images:
biomonitoring with AI





- robot finds insects in a sample
- images each specimen
- classifies the specimen
- measures specimen
- prepares it for DNA sequencing

Lorenz Wührl, KIT

MOLECULAR ECOLOGY
RESOURCES

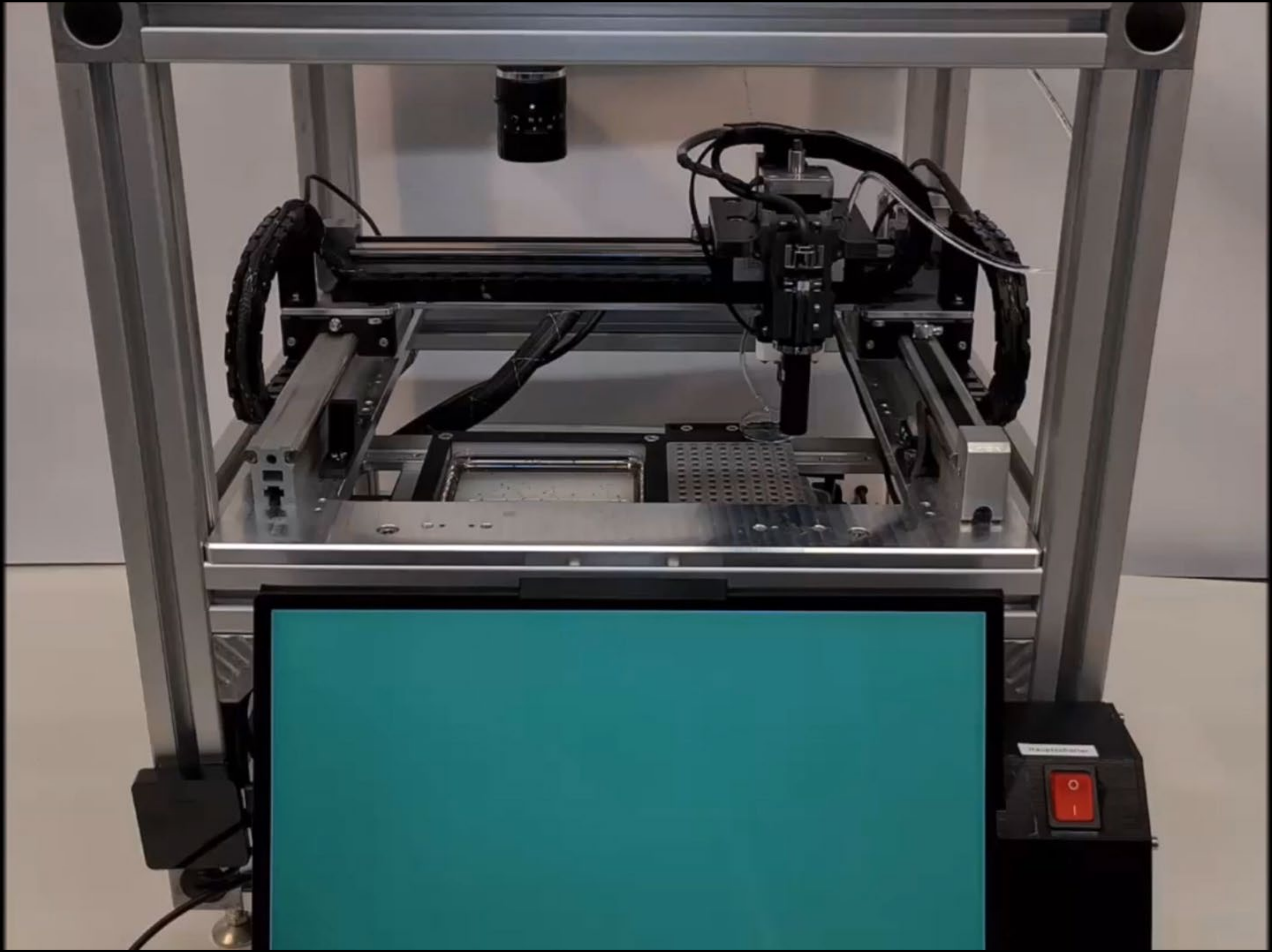
RESOURCE ARTICLE | Open Access

DiversityScanner: Robotic handling of small invertebrates with machine learning methods

Lorenz Wührl, Christian Pylatiuk, Matthias Giersch, Florian Lapp, Thomas von Rintelen, Michael Balke, Stefan Schmidt, Pierfilippo Cerretti, Rudolf Meier

First published: 04 December 2021 | https://doi.org/10.1111/1755-0998.13567

This article has been accepted for publication and undergone full peer review but has not been through the copyediting, typesetting, pagination and proofreading process, which may lead to differences between this version and the Version of Record. Please cite this article as doi:10.1111/1755-0998.13567



Karlsruher Institut für Technologie (KIT)

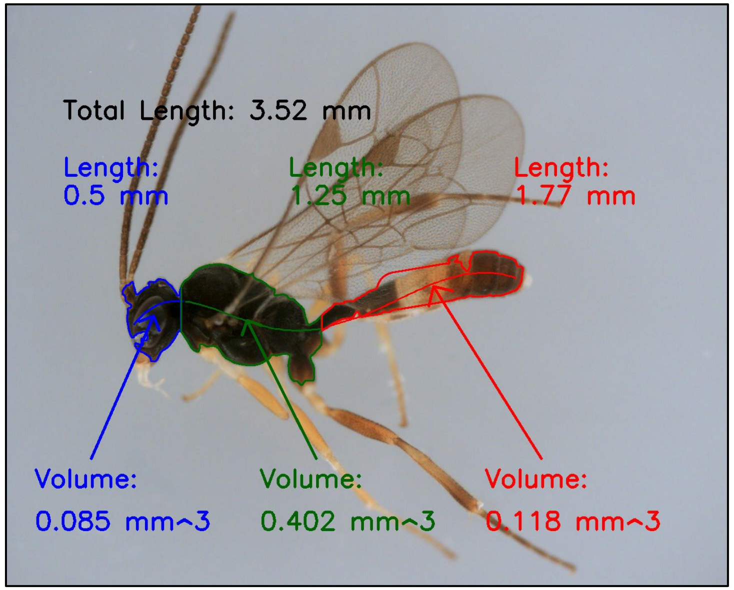
Prof. C. Pylatiuk

Lorenz Wührl

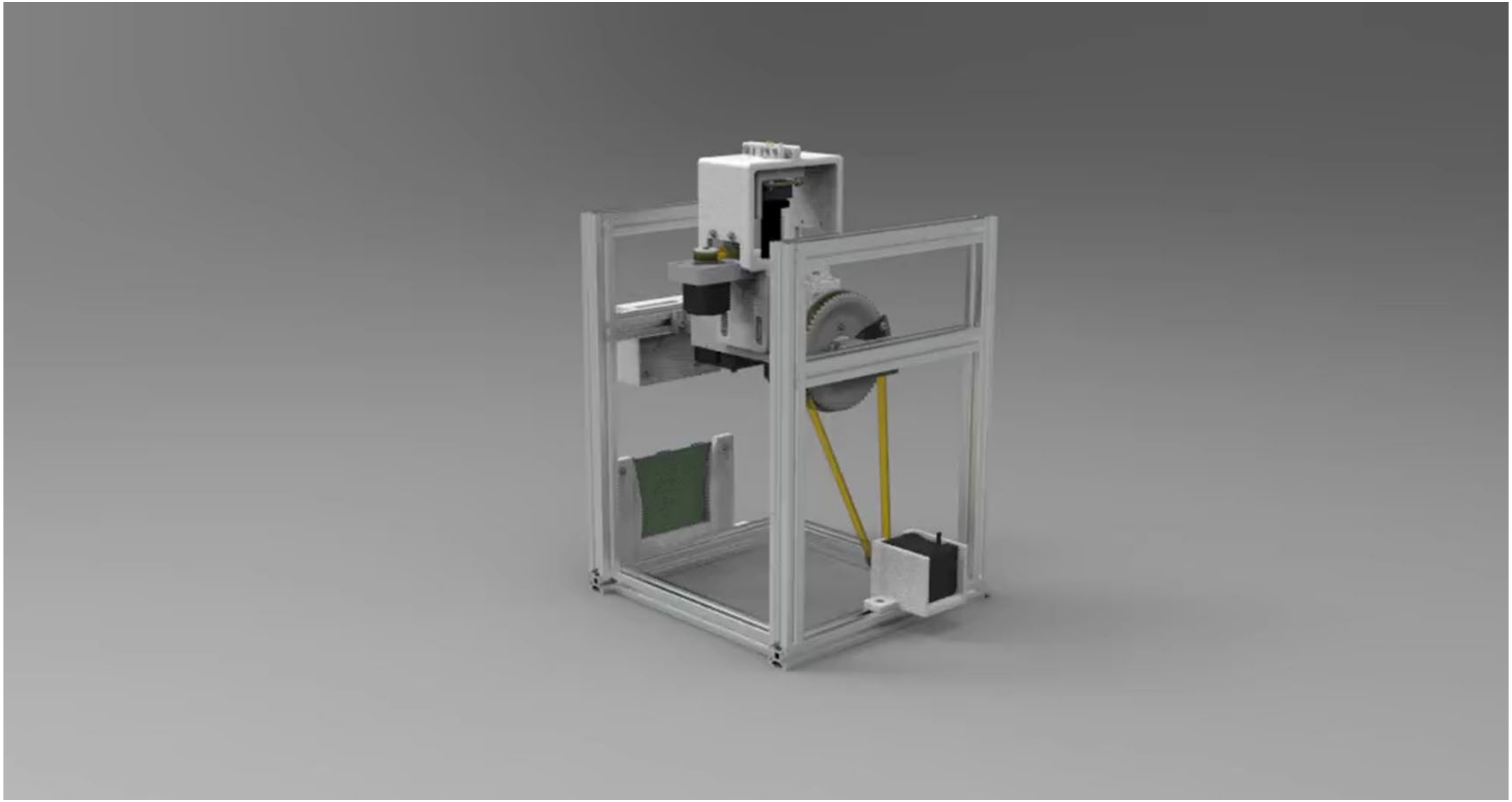
Detailed images for:
- taxonomic work
- training of AI models

=> image-based biomonitoring

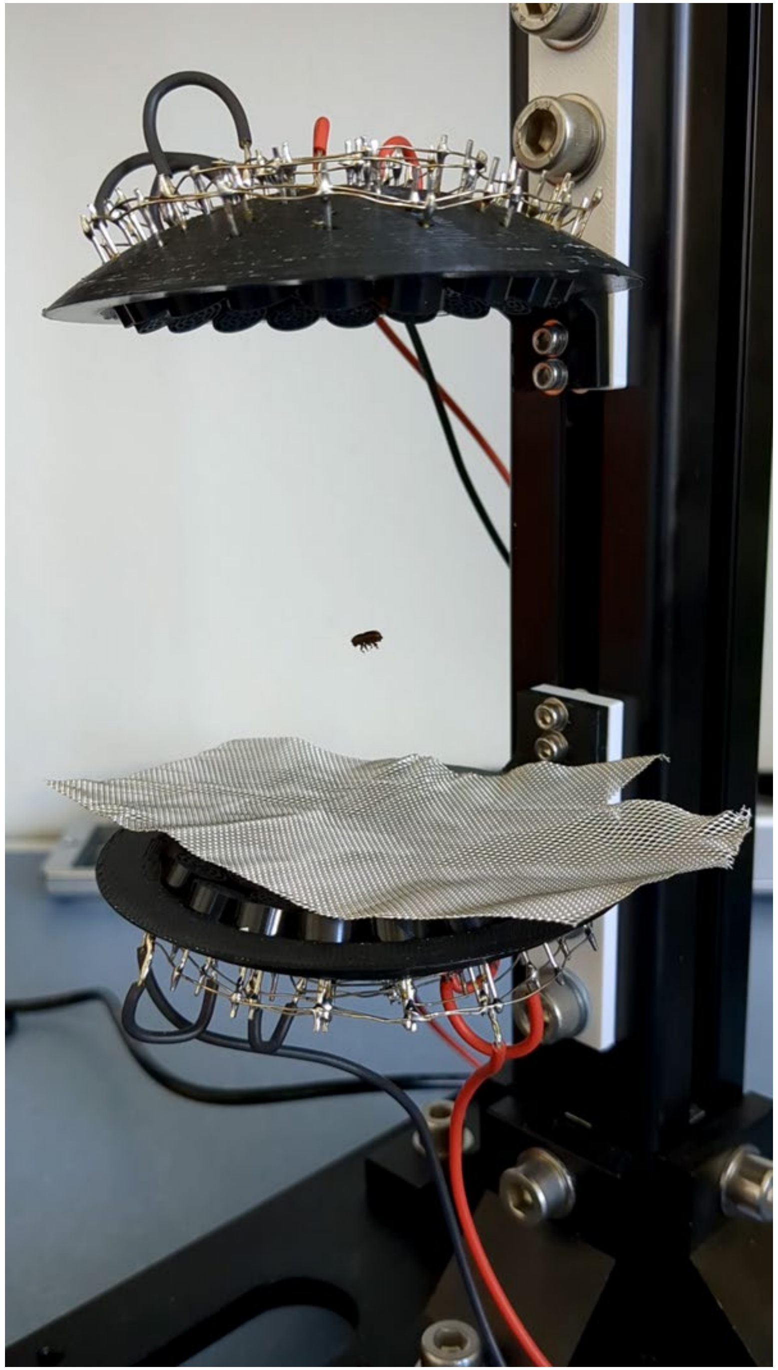
DiversityScanner
Biomass estimation



Lorenz Wührl, KIT



Lorenz Wührl, KIT



Nathalie Klug, KIT

Imaging from multiple perspectives also important for training AI identification models

Different “DiversityScanners” for different purposes

Lorenz Wührl, KIT

Entomoscope: An Open-Source Photomicroscope for Biodiversity Discovery

LORENZ WÜHRL[1], LUCA RETTENBERGER[1], RUDOLF MEIER[2], EMILY HARTOP[2], JULIEN GRAF[2], and CHRISTIAN PYLATIUK[1],

[1]Institute for Automation and Applied Informatics, Karlsruhe Institute of Technology, Karlsruhe, Germany {lorenz.wuehrl, luca.rettenberger, pylatiuk}@kit.edu
[2]Center for Integrative Biodiversity Discovery, Leibniz Institute for Evolution and Biodiversity Science, Museum für Naturkunde Berlin, Germany {rudolf.meier, emily.hartop, julien.graf}@mfn.berlin

Corresponding author: Lorenz Wührl (e-mail: lorenz.wuehrl@kit.edu).

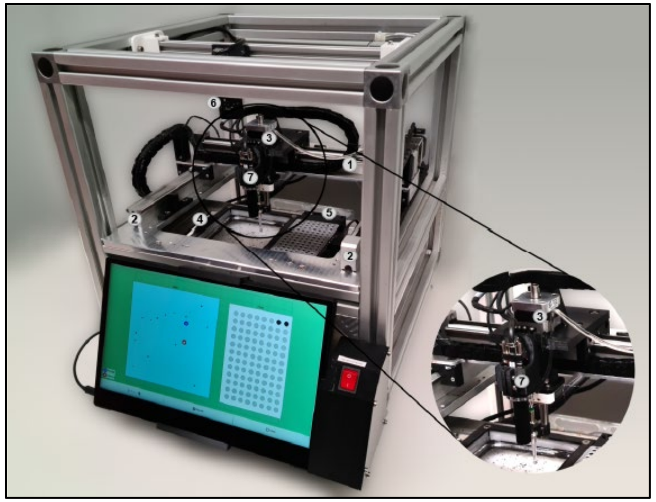
This research was supported by the Center for Integrative Biodiversity Discovery at the Museum für Naturkunde Berlin and the program Natural, Artificial and Cognitive Information Processing (NACIP) of the Helmholtz-Association.

Lorenz Wührl*, Leonard Keller, Nathalie Klug, Hossein Shirali, Rudolf Meier, and Christian Pylatiuk

Automated Handling of Biological Objects with a Flexible Gripper for Biodiversity Research

Species Discovery Factory

1. DiversityScanner:
Digitization with robots



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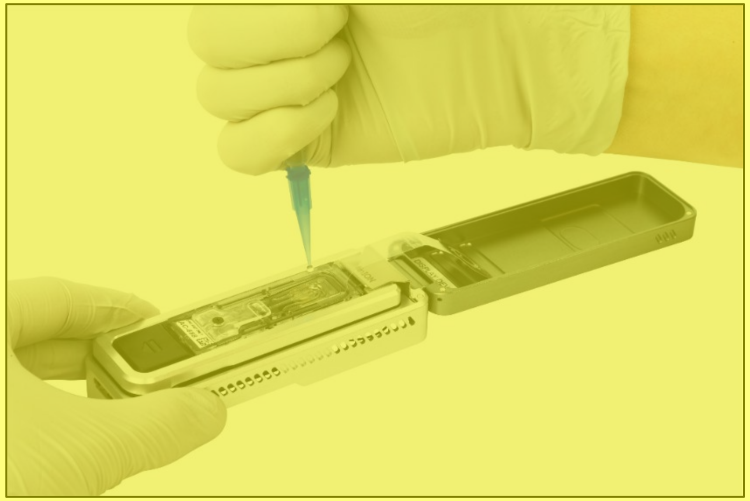
2. Nanopore Sequencing:
Sorting with DNA barcodes



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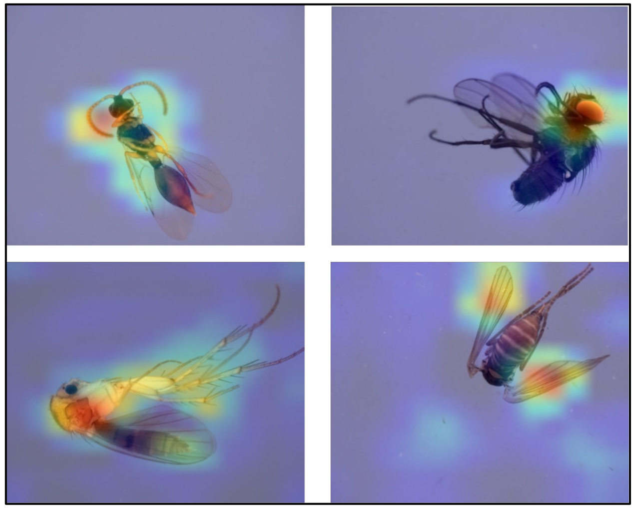


2. Nanopore Sequencing


Srivathsan *et al. BMC Biology* (2021) 19:217
https://doi.org/10.1186/s12915-021-01141-x

BMC Biology

METHODOLOGY ARTICLE **Open Access**

ONTbarcoder and MinION barcodes aid biodiversity discovery and identification by everyone, for everyone

Amrita Srivathsan[1], Leshon Lee[1], Kazutaka Katoh[2,3], Emily Hartop[4,5], Sujatha Narayanan Kutty[1,6], Johnathan Wong[1], Darren Yeo[1] and Rudolf Meier[1,7*]

“Megabarcoding”:

- simplified, fast, and robust techniques for specimen-based barcoding
- 10 cents/specimen

LOVE YOU

Matching real-time sequencing with real-time DNA barcoding

Dr. Amrita Srivathsan

Cladistics
The Willi Hennig Society

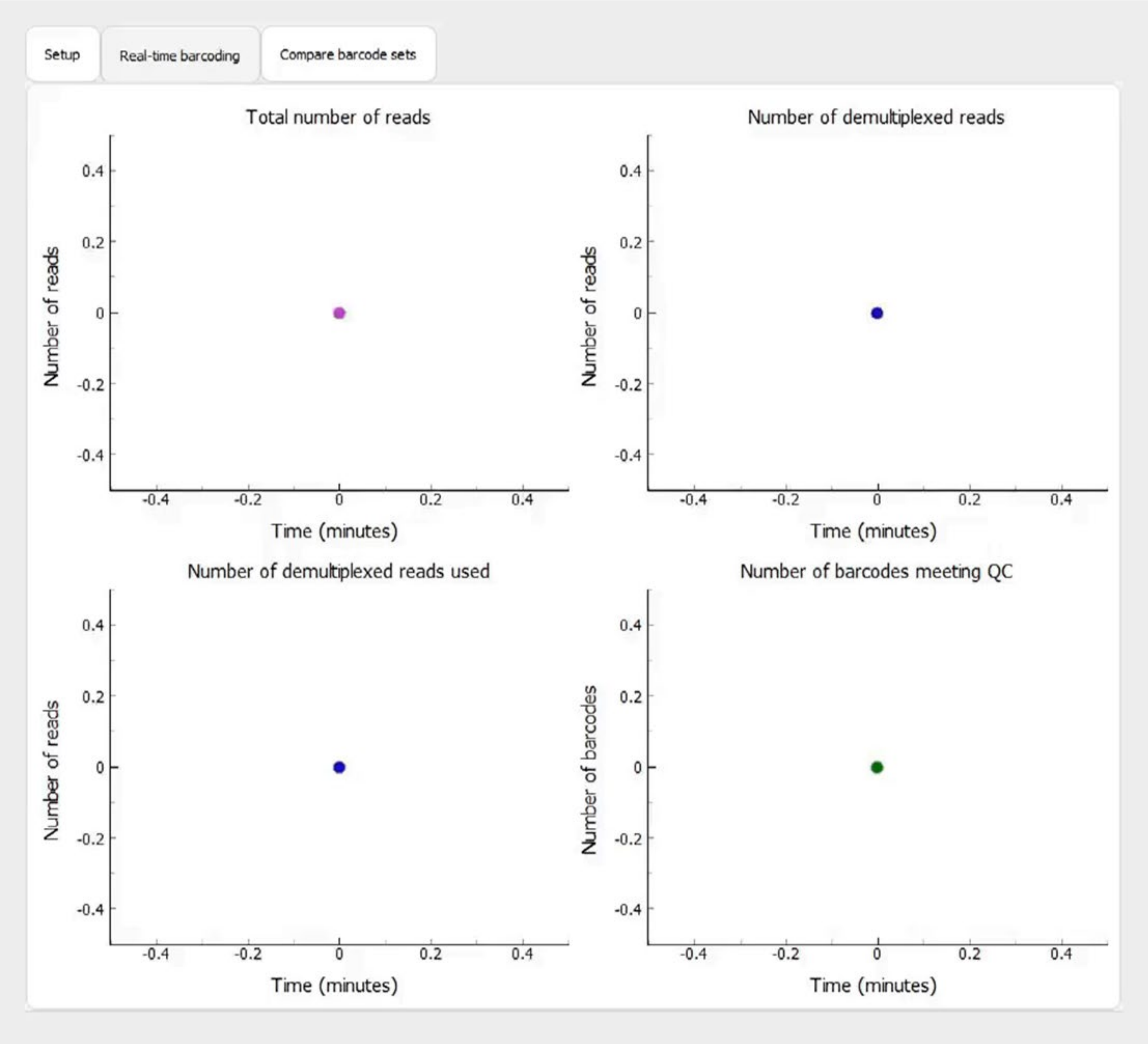
Research Article | Open Access

ONTbarcoder 2.0: rapid species discovery and identification with real-time barcoding facilitated by Oxford Nanopore R10.4

Amrita Srivathsan, Vivian Feng, Daniel Suárez, Brent Emerson, Rudolf Meier

First published: 02 December 2023 | https://doi.org/10.1111/cla.12566

Flongle R10.4



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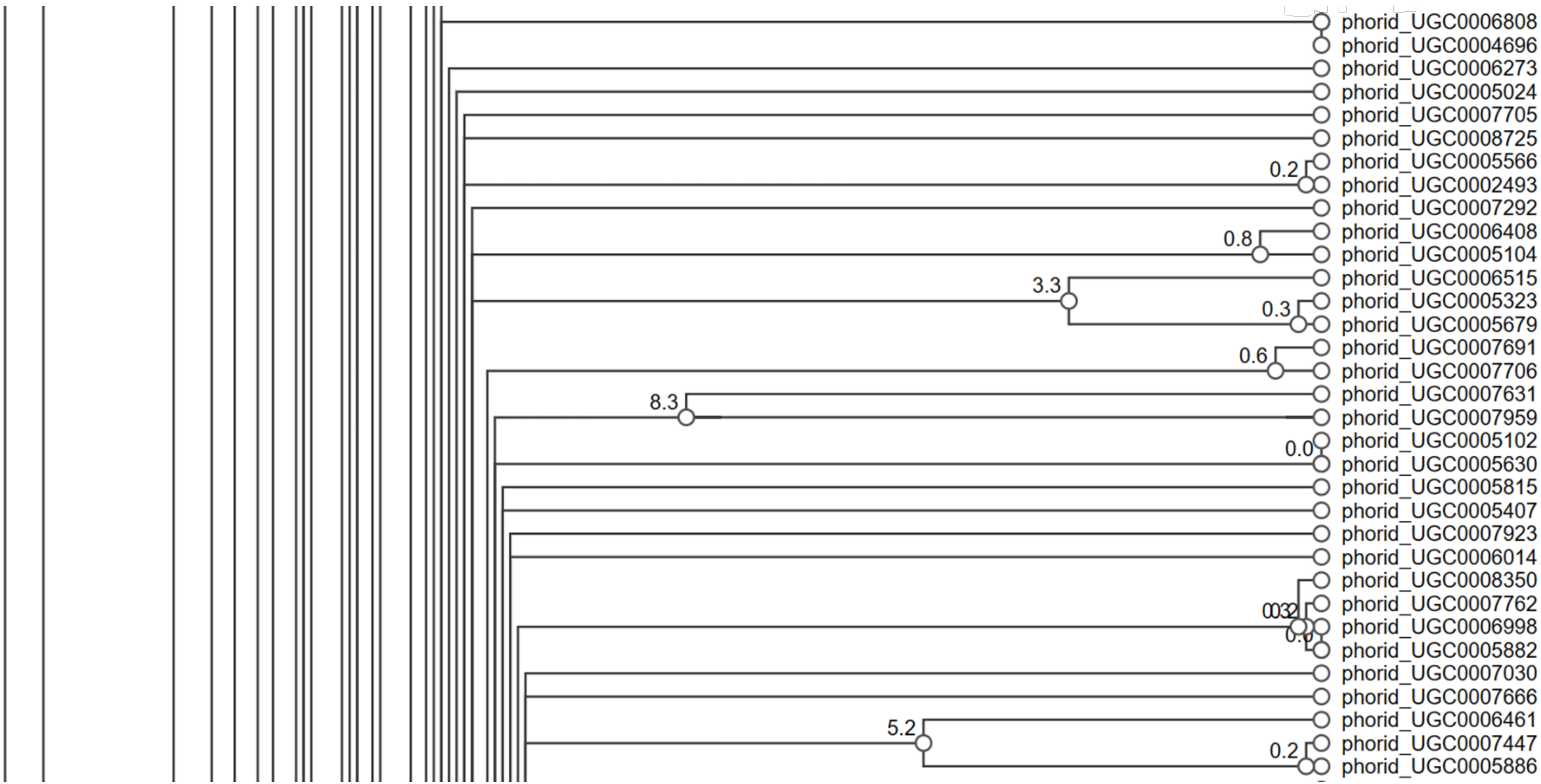
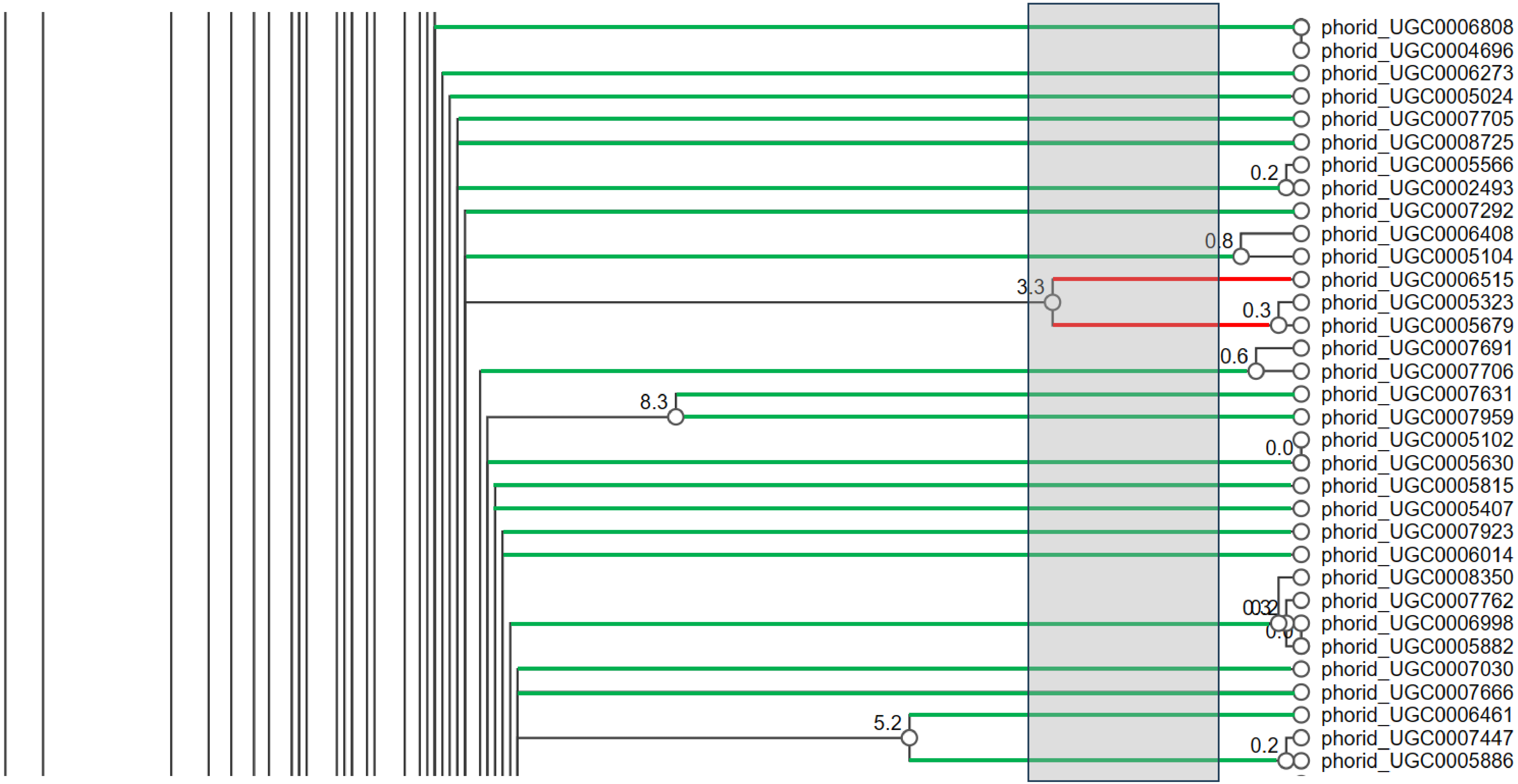
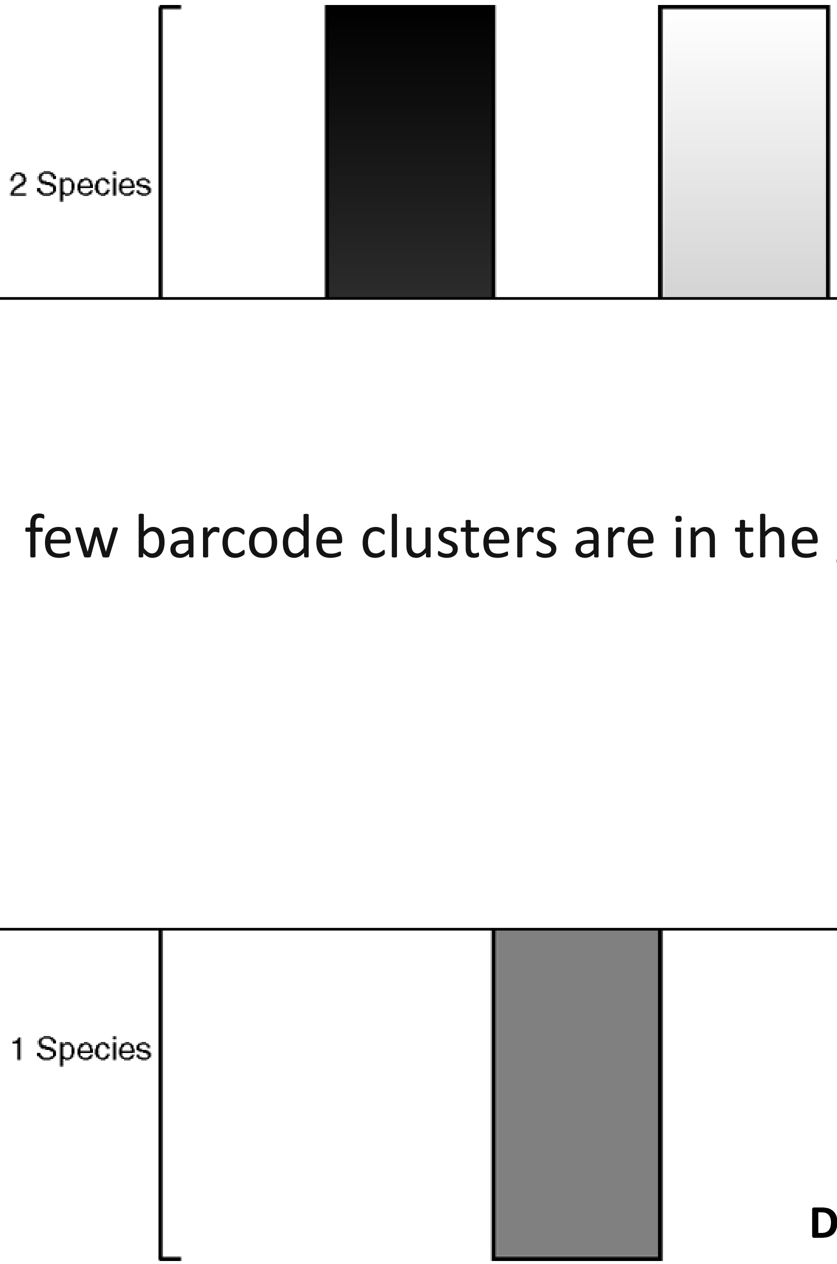
“Cluster Fusion Diagram”





1-3.5%

=> most haplotype distinct and rare => almost certainly species

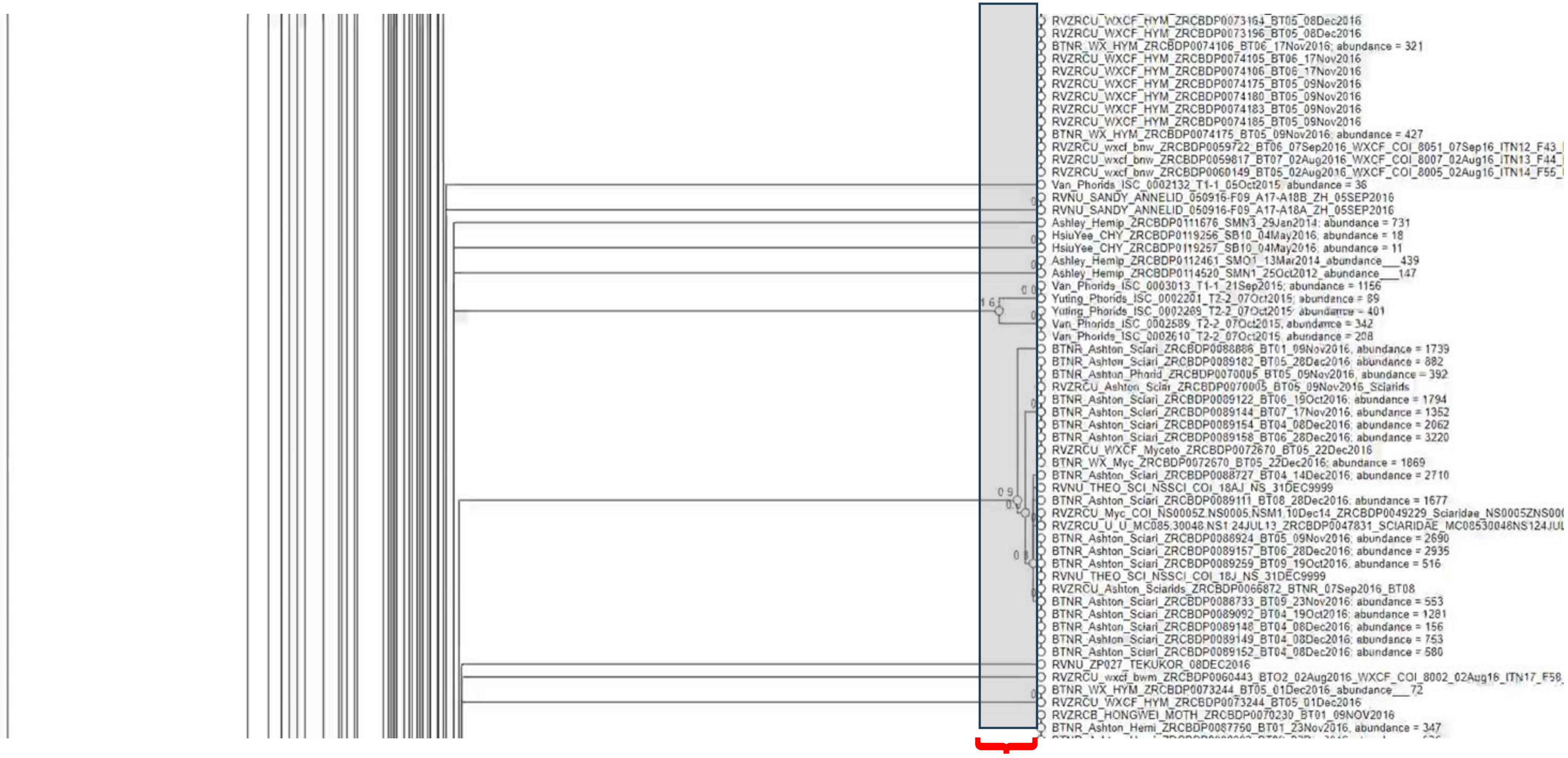


few barcode clusters are in the grey zone

De Queiroz, 2007

Singapore dataset: 350,000 specimens
=> ca. 9,000 species with distribution and abundance information
=> 45 minute-movie

Few species are in the "grey zone" where species limits are unclear



1-3.5%

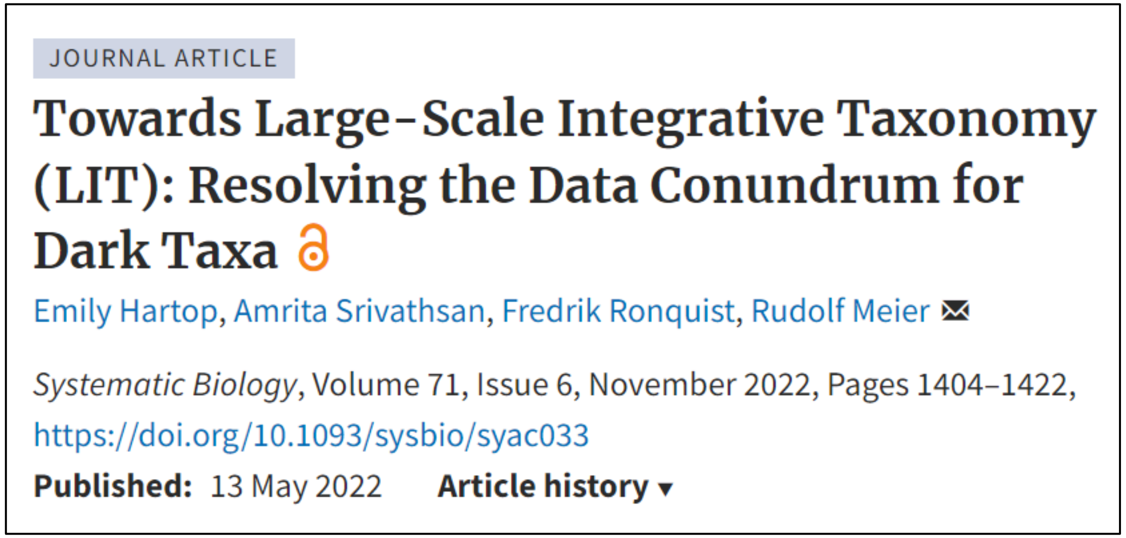
DNA barcoding or Integrative Taxonomy?


Dr. Emily Hartop

Step 1. DNA barcoding for presorting specimens to putative species

Step 2. Morphological checks using a subset of specimens representing
- main haplotypes for distinct barcode clusters
- main and divergent haplotypes for “grey-zone clusters”

Step 3: Resolving incongruence
Step 4: Identification or Description


JOURNAL ARTICLE

Towards Large-Scale Integrative Taxonomy (LIT): Resolving the Data Conundrum for Dark Taxa

Emily Hartop, Amrita Srivathsan, Fredrik Ronquist, Rudolf Meier

Systematic Biology, Volume 71, Issue 6, November 2022, Pages 1404–1422, https://doi.org/10.1093/sysbio/syac033

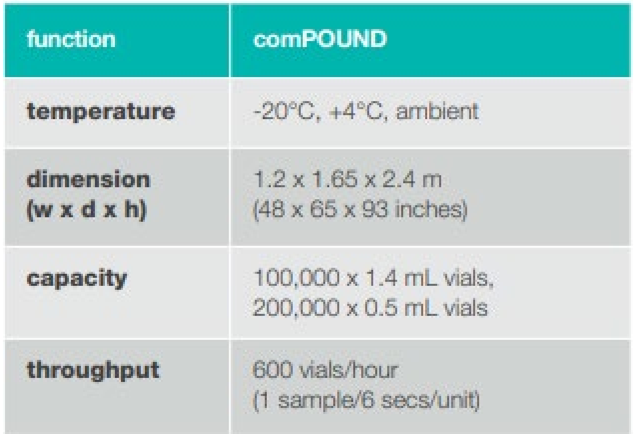
Published: 13 May 2022 **Article history** ▾

Managing specimens: voucher handling

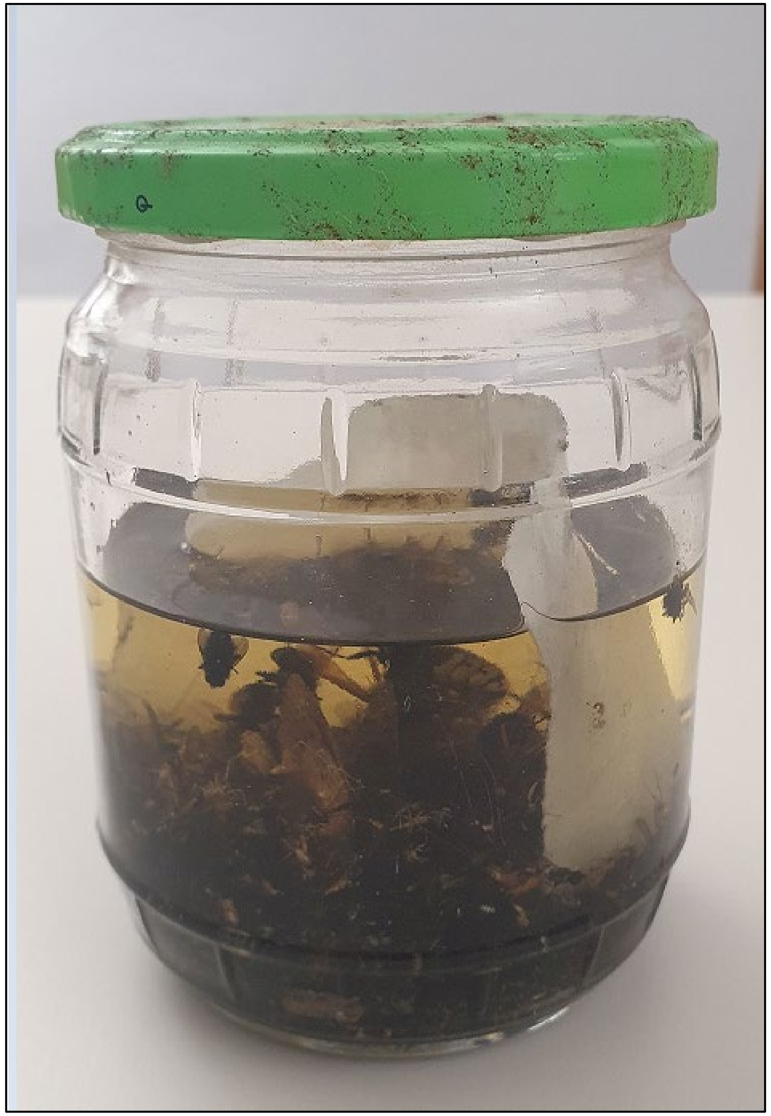
Vending machine for specimens

Carousel system for high density, low footprint storage

Pneumatic technology for vial storage and retrieval



function	comPOUND
temperature	-20°C, +4°C, ambient
dimension (w x d x h)	1.2 x 1.65 x 2.4 m (48 x 65 x 93 inches)
capacity	100,000 x 1.4 mL vials, 200,000 x 0.5 mL vials
throughput	600 vials/hour (1 sample/6 secs/unit)




comPOUND®

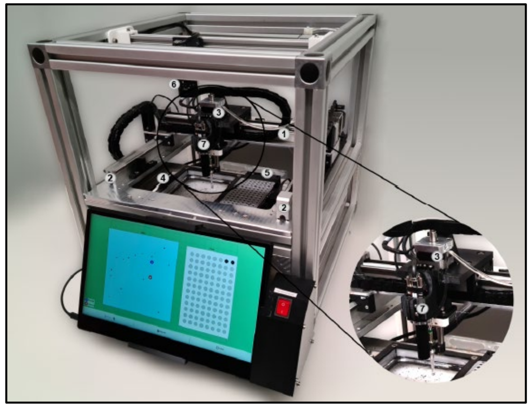
Managing data: from Entomoscope to Zenodo and GBIF

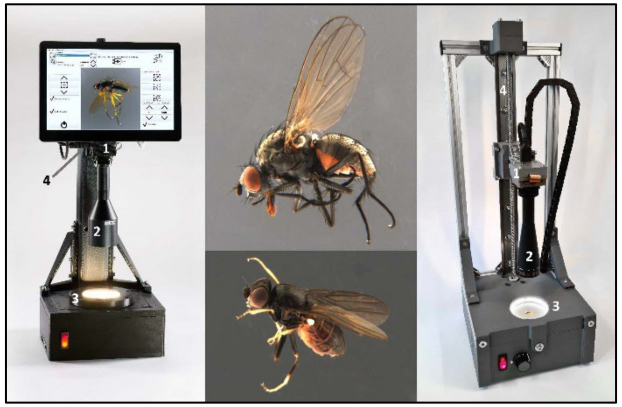
Automatic data upload to CERN’s Zenodo

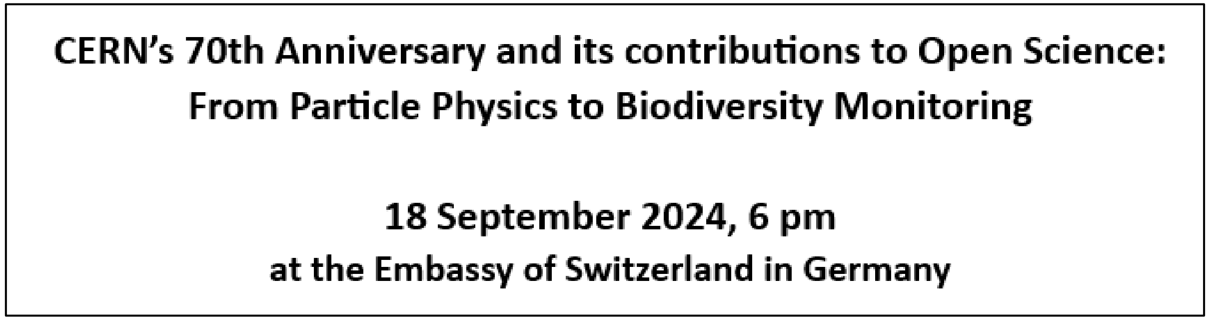
- Entomoscope and DiversityScanner software has Zenodo upload feature
- each specimen gets Zenodo entry with DOI (images, barcodes)

Dr. Amrita Srivathsan

Dr. Donat Agosti, Plazi



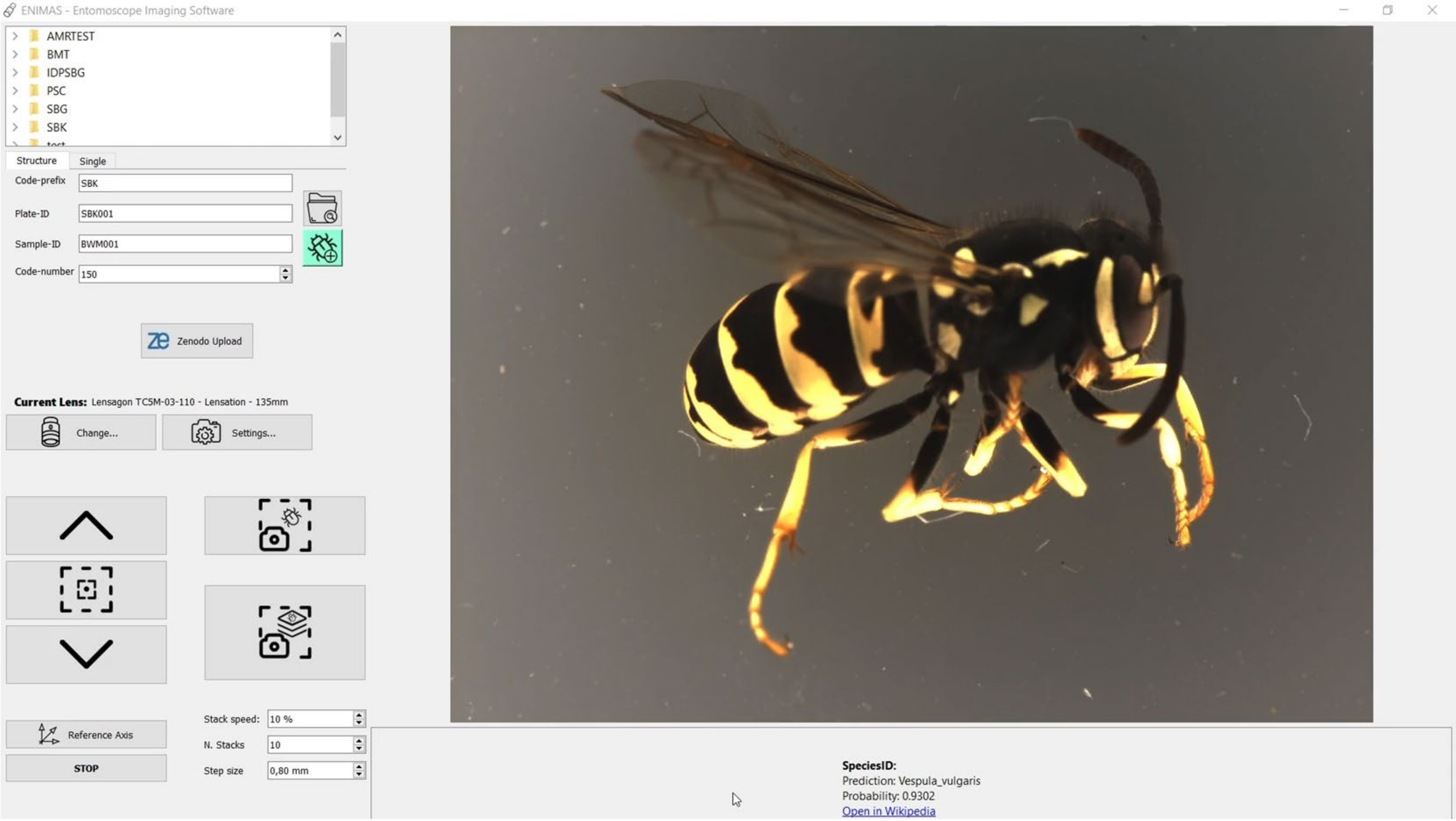




CERN’s 70th Anniversary and its contributions to Open Science:
From Particle Physics to Biodiversity Monitoring

18 September 2024, 6 pm
at the Embassy of Switzerland in Germany

Automatic data upload to Zenodo/GBIF



Code-prefix: SBK

Plate-ID: SBK001

Sample-ID: BWM001

Code-number: 150

Current Lens: Lensagon TC5M-03-110 - Lensation - 135mm

Stack speed: 10 %

N. Stacks: 10

Step size: 0,80 mm

SpeciesID:
Prediction: Vespula_vulgaris
Probability: 0.9302
Open in Wikipedia

Dr. Amrita Srivathsan

Dr. Donat Agosti, Plazi

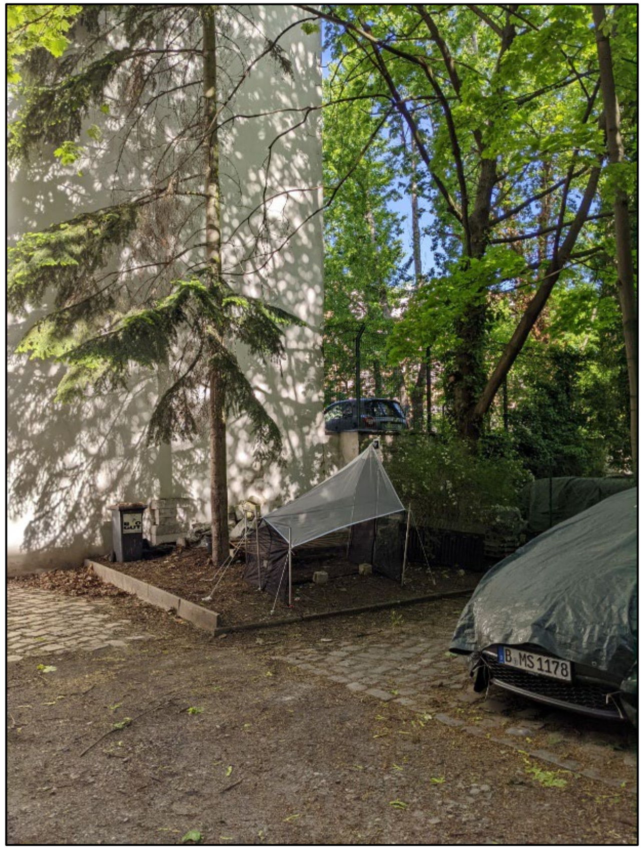
How quickly can we generate good biodiversity data?

How quickly can we generate good biodiversity data?

Cem Kir

5000 new data points for Berlin Kreuzberg

Bachelor's thesis
- 1 Malaise trap: 2-week July sample
- 5951 individuals
- 750 species



Let's imagine: 1000 BSc theses per year
- => 5 million new data points
- distribution of thousands of species known

imagine doing this for several years and in many places…

My challenge to you: be the first country that knows its arthropods

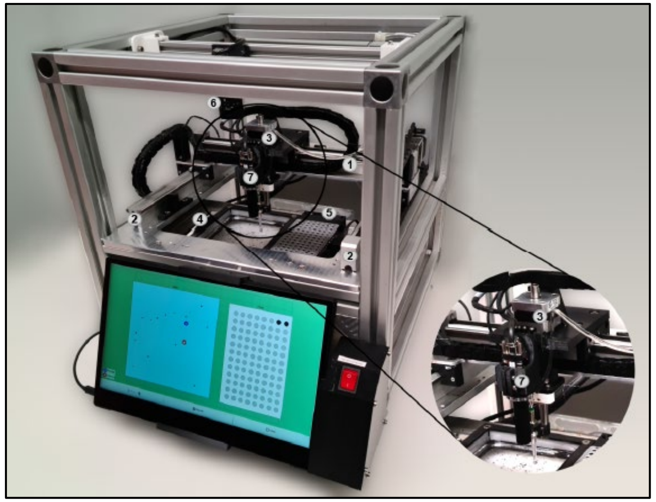
Step 1: barcode 1,000 specimens for 500 sites/sample (500,000 specimens)
=> species discovery rate can be 5-10% (i.e., 25,000 species)
Step 2: image numerically important species to train AI models
=> AI tools for common species

This would yield:

- unbiased abundance and diversity data for 500 sites
- barcodes for 25,000 species to interpret metabarcoding data
- vouchers, barcodes, and AI models for the most abundant species
- critical information on distributions and ecology of these species

Species Discovery Factory

1. DiversityScanner:
Digitization with robots

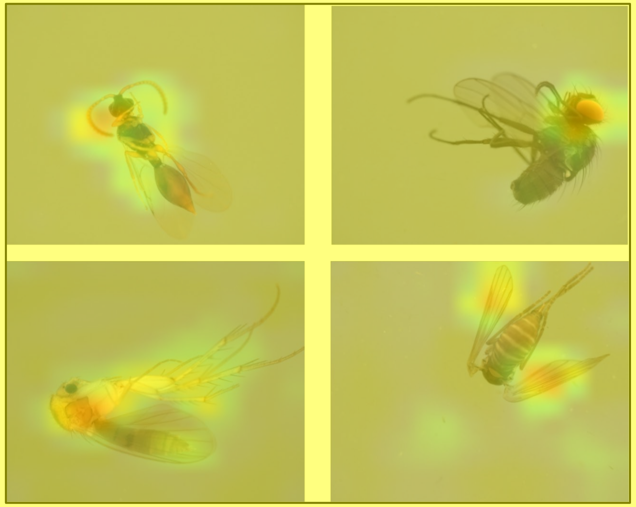


+

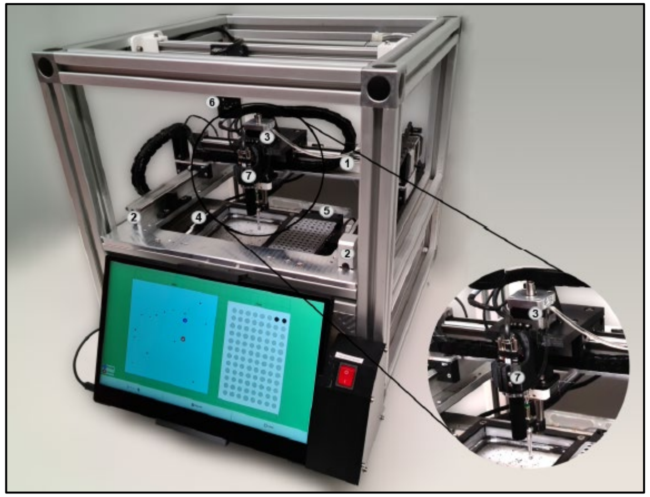
2. Nanopore Sequencing:
Sorting with DNA barcodes

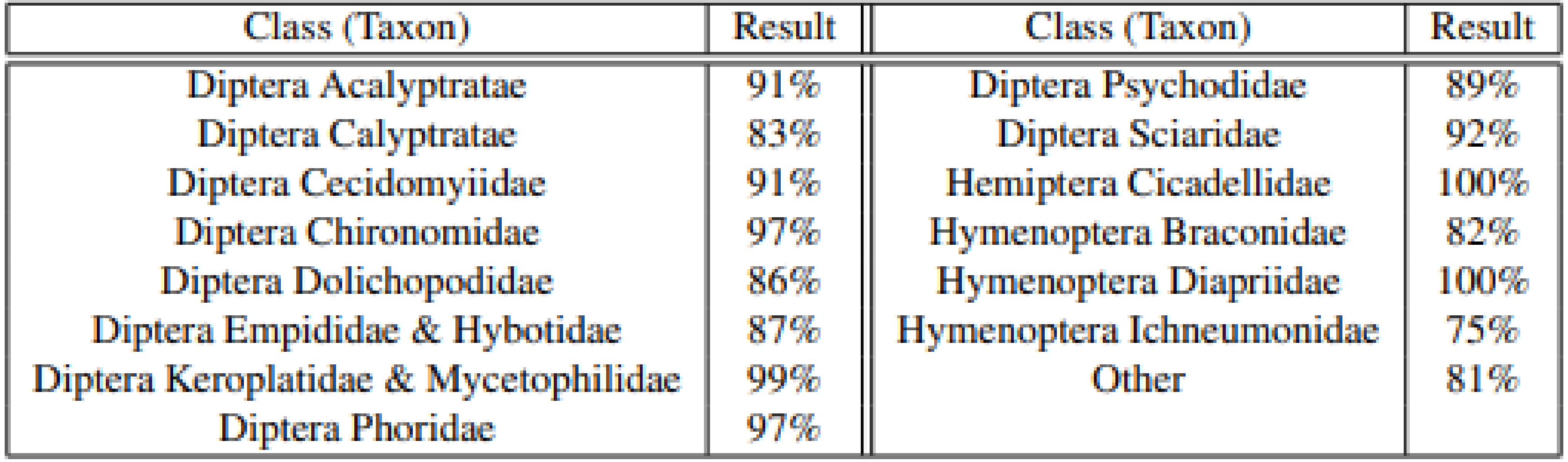
=

3. Identified images:
biomonitoring with AI



3. Identification with AI



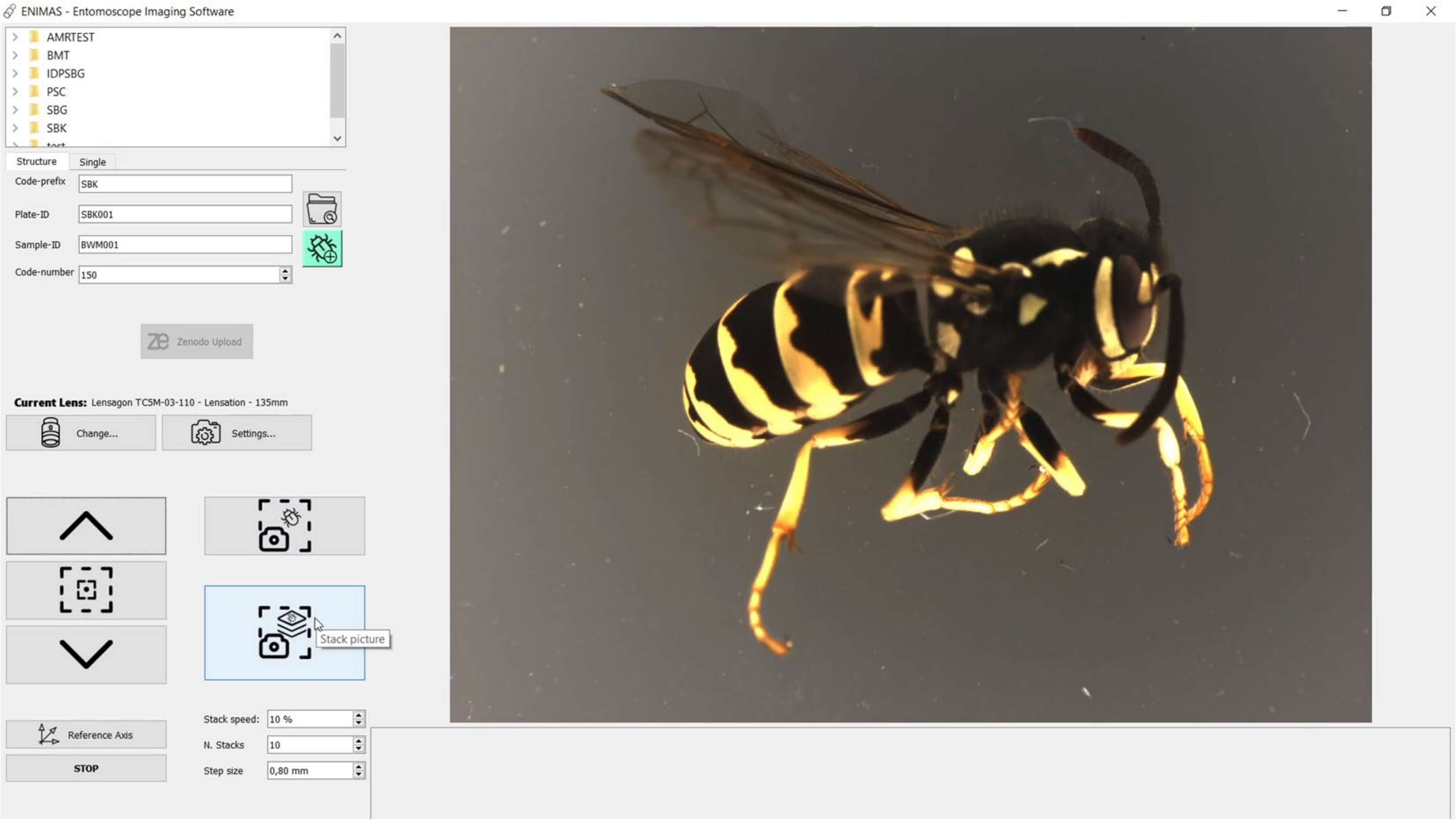


Class (Taxon)	Result	Class (Taxon)	Result
Diptera Acalyptratae	91%	Diptera Psychodidae	89%
Diptera Calyptratae	83%	Diptera Sciaridae	92%
Diptera Cecidomyiidae	91%	Hemiptera Cicadellidae	100%
Diptera Chironomidae	97%	Hymenoptera Braconidae	82%
Diptera Dolichopodidae	86%	Hymenoptera Diapriidae	100%
Diptera Empididae & Hybotidae	87%	Hymenoptera Ichneumonidae	75%
Diptera Keroplatidae & Mycetophilidae	99%	Other	81%
Diptera Phoridae	97%		

AI identification tools evolve

- common taxa benefit first
- tools evolve from family => genus => species
- latest version of DiversityScanner/Entomoscope has an AI model for common species in Berlin

Hossein Shirali, KIT



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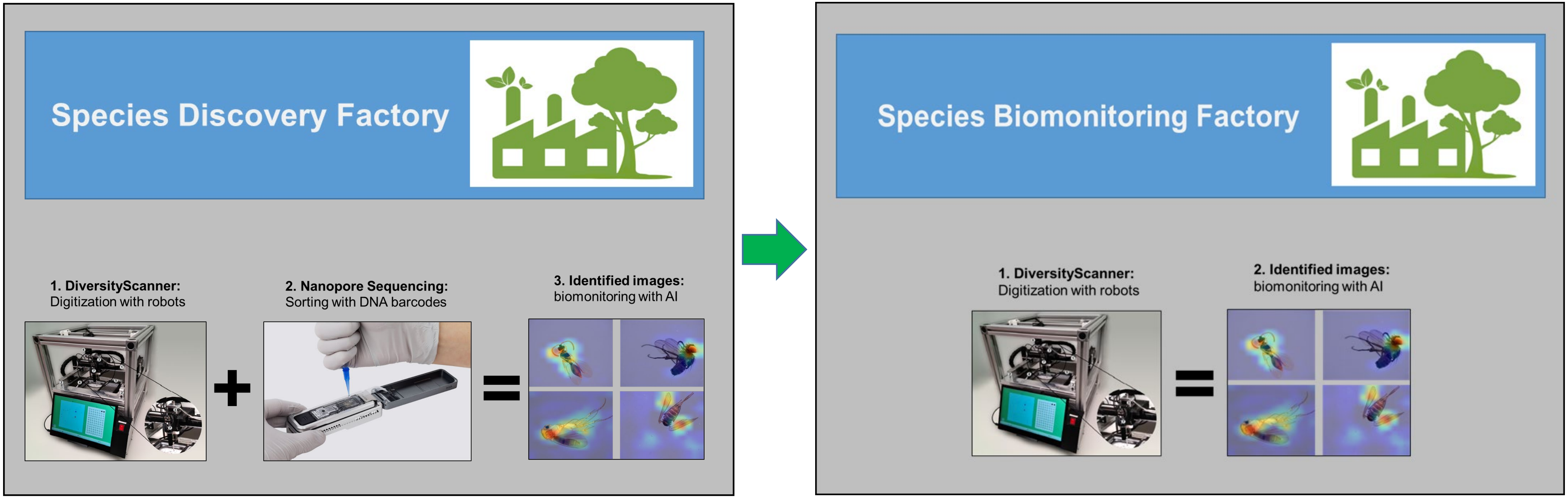
N. Stacks: 10
Step size: 0,80 mm

Hossein Shirali, KIT

Does the model really work? some species are easy some are hard



Species Discovery Factory

1. DiversityScanner:
Digitization with robots

2. Nanopore Sequencing:
Sorting with DNA barcodes

3. Identified images:
biomonitoring with AI

Species Biomonitoring Factory

1. DiversityScanner:
Digitization with robots

2. Identified images:
biomonitoring with AI

Could generate large amounts of data quickly

Shortcomings of the old ways…

- >80% of all species undescribed
- 95% of all described species only identifiable for taxonomic experts
- very little baseline biodiversity data from standardized samples
- natural history data only available for <5% of species

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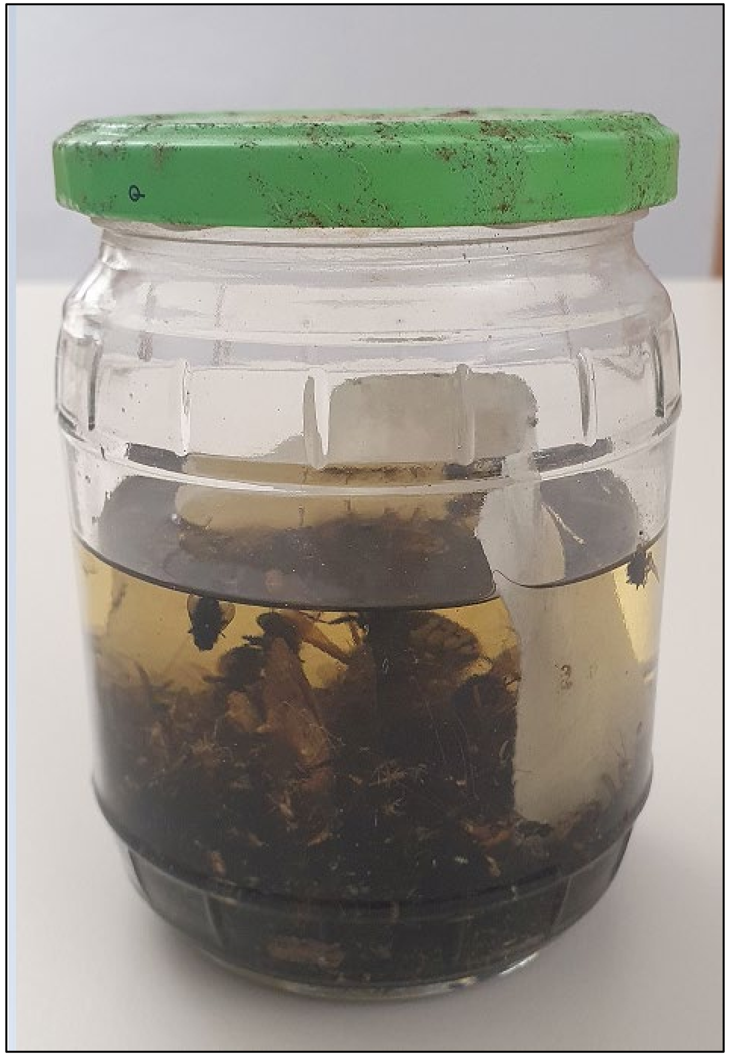
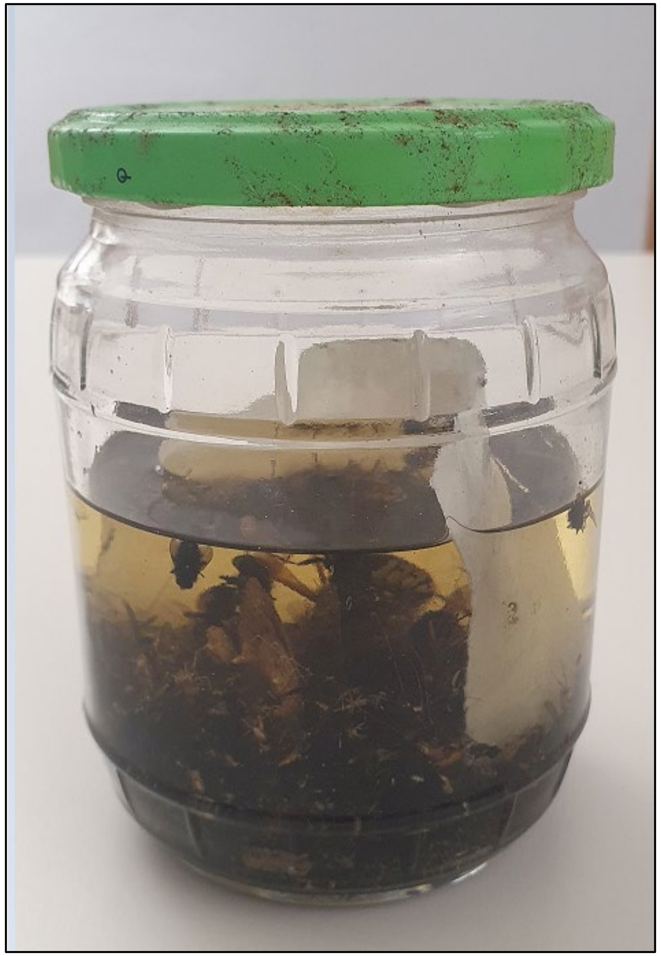
Too many species… isn’t it hopeless?



”Dark Taxonomy“: focus taxonomic work on species important for biomonitoring



”Dark Taxonomy“ with biomonitoring focus

Step 1: Get fresh material for a species-rich taxon using traps also used for biomonitoring

Taxonomic sample

”Dark Taxonomy“

Step 1: Get fresh material from many traps used in biomonitoring

Step 2: Pre-sort specimens with DNA barcodes

”Dark Taxonomy“: phase 1 of taxonomic exploration

Step 1: Get fresh material from many traps used in biomonitoring

Step 2: Pre-sort specimens with DNA barcodes

Step 3: Integrative taxonomy with two data sources

”Dark Taxonomy“

Step 1: Get fresh material from many traps used in biomonitoring

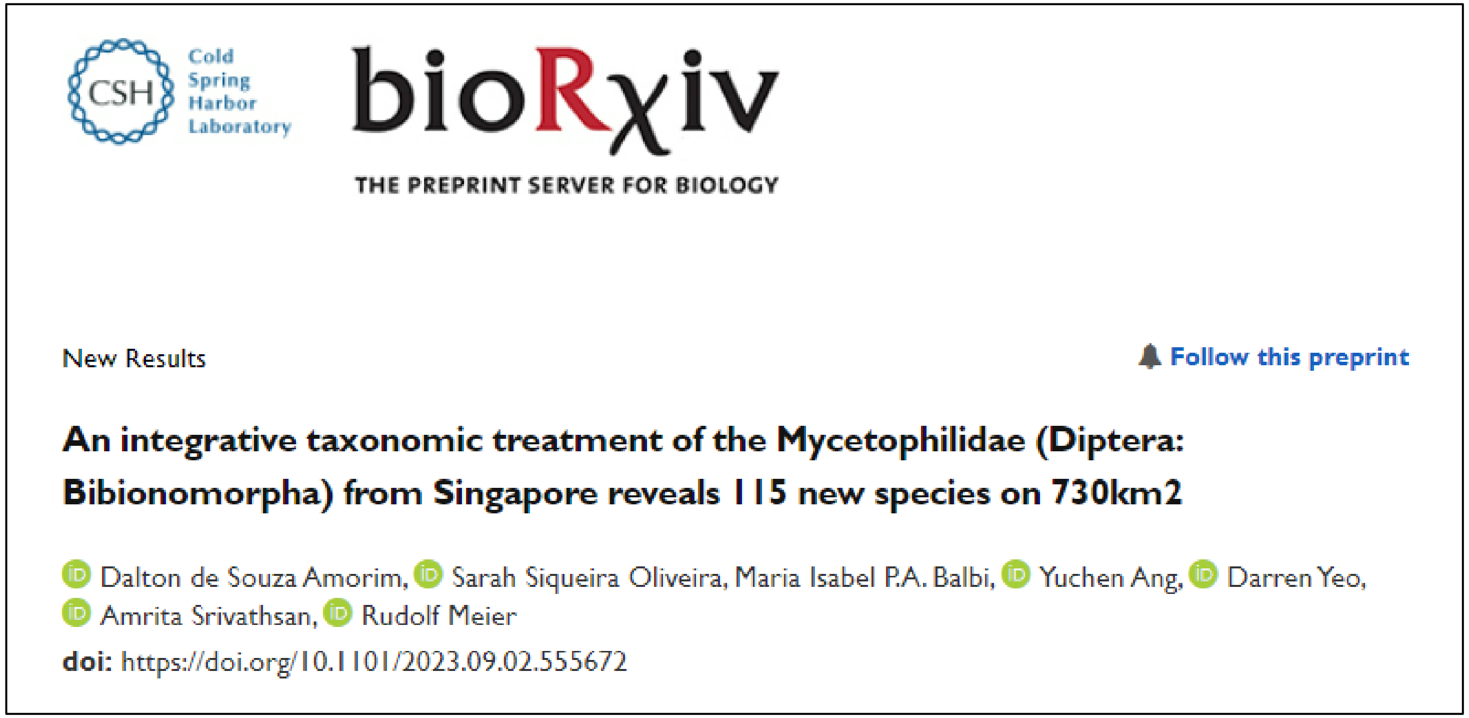
Step 2: Pre-sort specimens with DNA barcodes

Step 3: Integrative taxonomy with two data sources

Step 4: Identify/describe species

120 species of fungus gnats (Diptera: Mycetophilidae) discovered in Singapore:

- 5 described species
- 115 new species



New Results

An integrative taxonomic treatment of the Mycetophilidae (Diptera: Bibionomorpha) from Singapore reveals 115 new species on 730km2

Dalton de Souza Amorim, Sarah Siqueira Oliveira, Maria Isabel P.A. Balbi, Yuchen Ang, Darren Yeo, Amrita Srivathsan, Rudolf Meier

doi: https://doi.org/10.1101/2023.09.02.555672

bioRxiv
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”Dark Taxonomy“

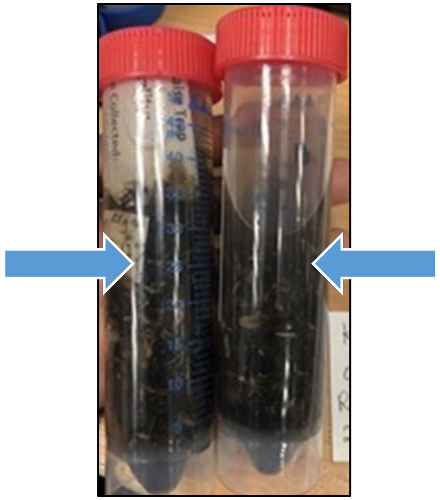
Step 1: Get fresh material from many traps used in biomonitoring

Step 2: Pre-sort specimens with DNA barcodes

Step 3: Integrative taxonomy with two data sources

Step 4: Identify or describe species

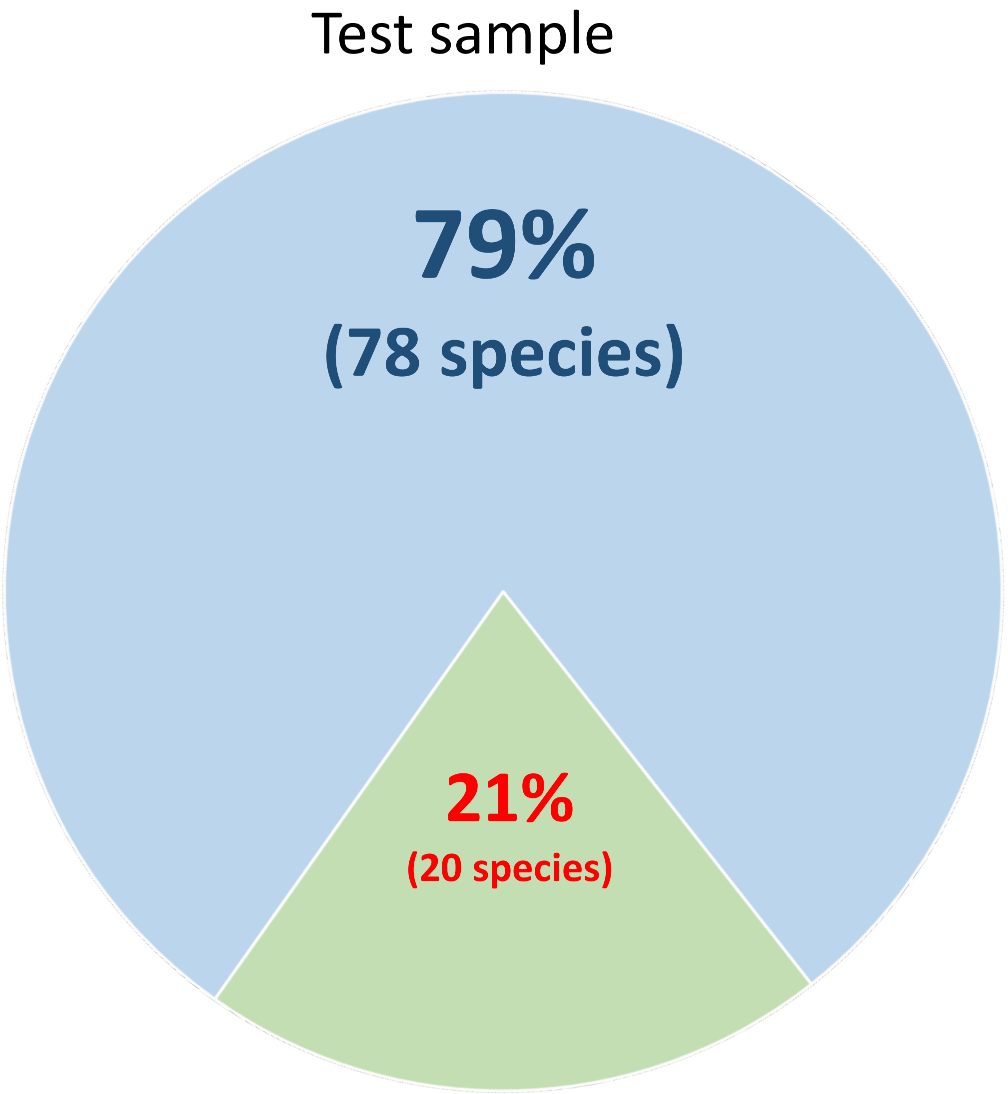
Step 5: Test whether all species important for biomonitoring are covered



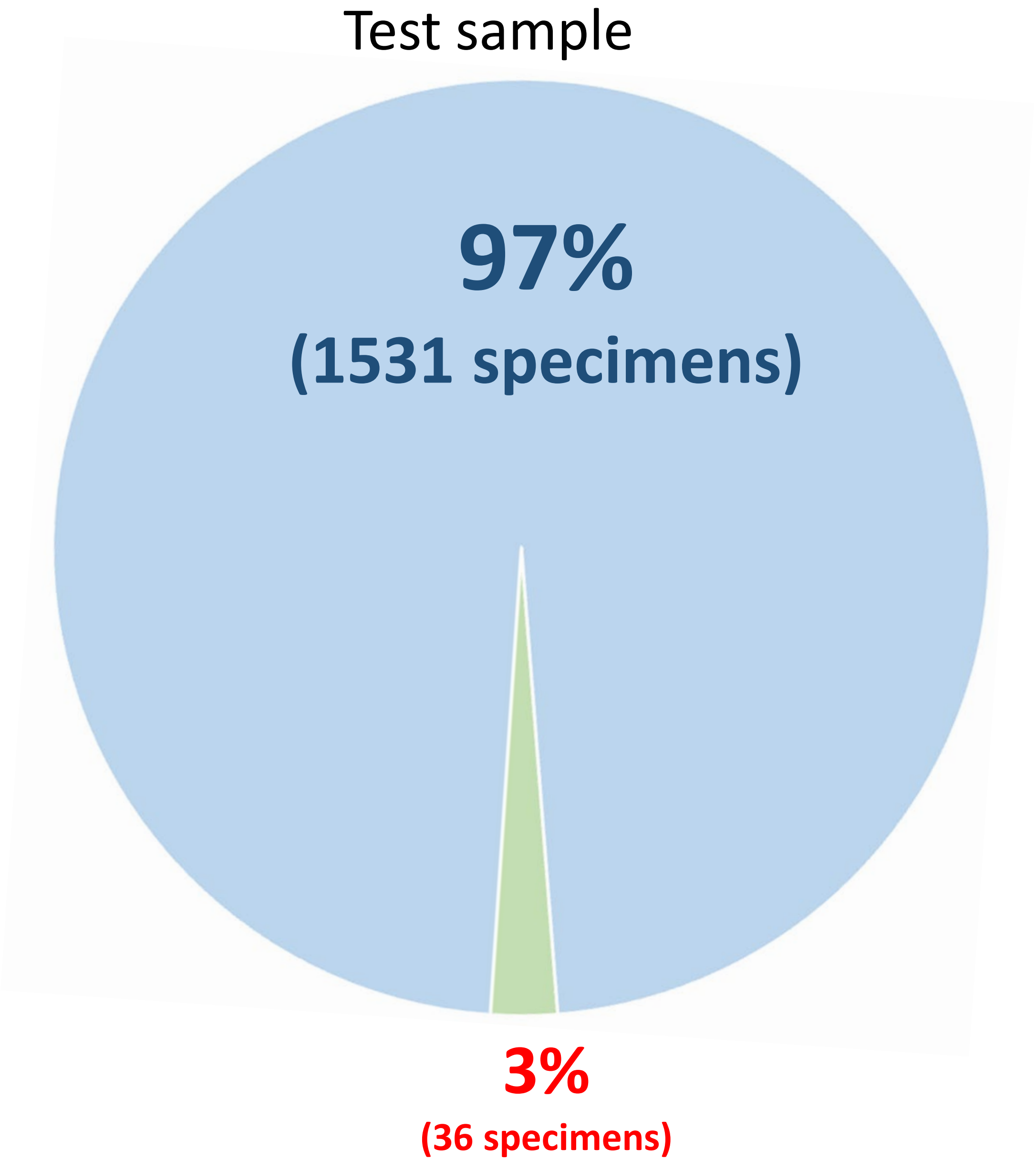
Taxonomic sample

Test sample

Mycetophilidae would be “biomonitoring-enabled“ if test sample consists mostly of described species also in taxonomic sample



What about specimens?



Dark Taxonomy:

from unknown to ready for biomonitoring in 5 steps

Cold Spring Harbor Laboratory

bioRxiv

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New Results

Follow this preprint

"Dark taxonomy": a new protocol for overcoming the taxonomic impediments for dark taxa and broadening the taxon base for biodiversity assessment

Rudolf Meier, Amrita Srivathsan, Sarah Siqueira Oliveira, Maria Isabel P.A. Balbi, Yuchen Ang, Darren Yeo, Dalton de Souza Amorim

doi: https://doi.org/10.1101/2023.08.31.555664

bioRxiv

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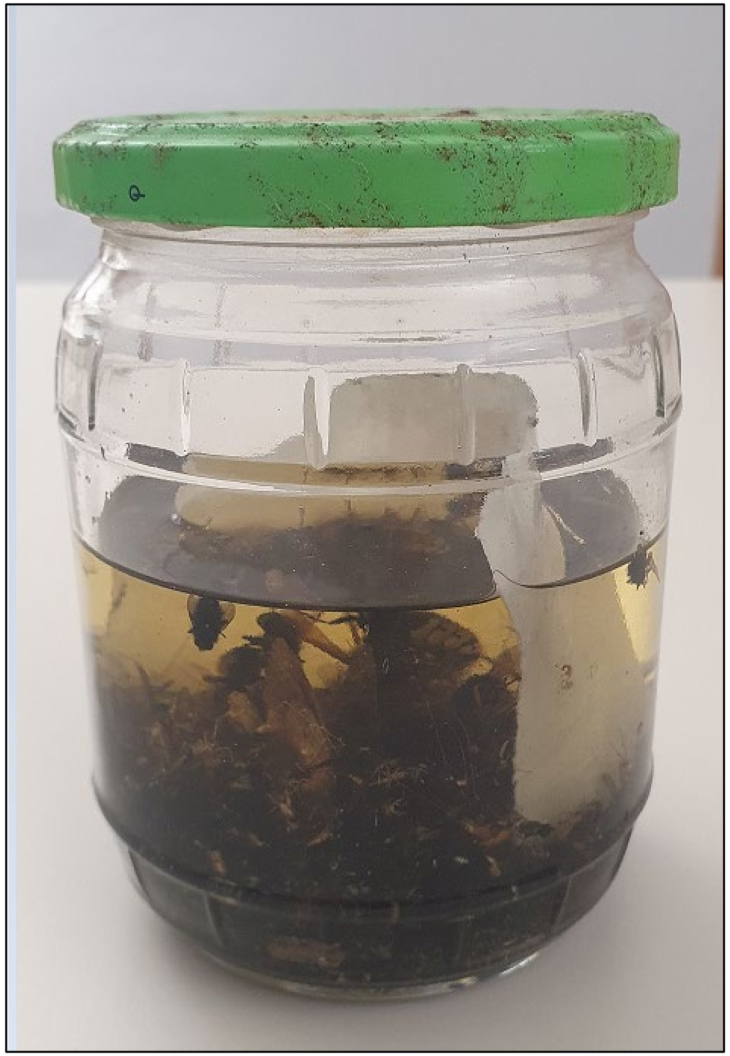
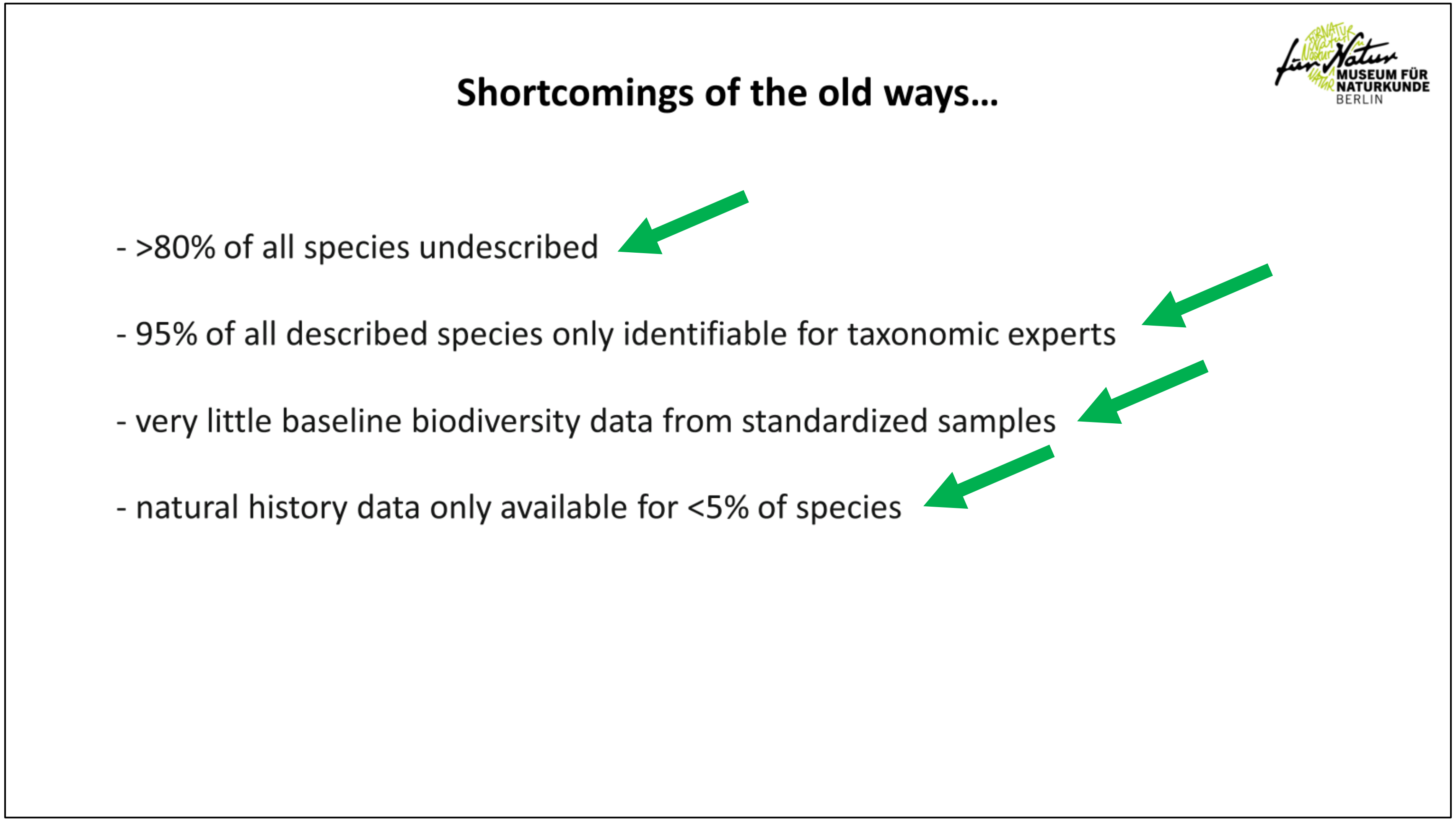
=> insect samples and faunas aren´t hopeless





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Shortcomings of the old ways…

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Our recommendation: resolve the natural history of common species first

Example 1: Mosquito diet

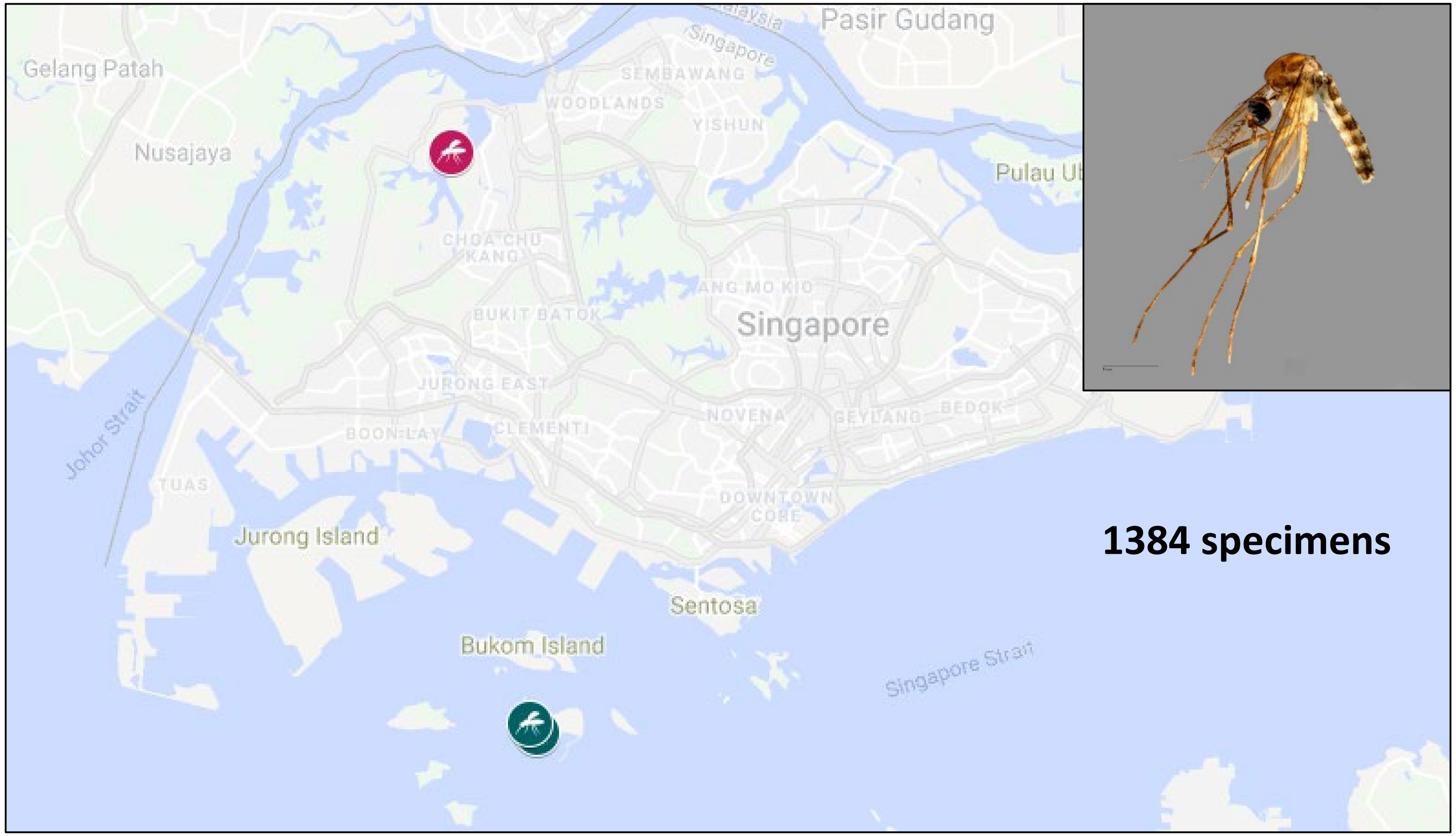
- 4000 mosquitoes barcoded

=> ca. 120 species detected (1/3rd identifiable via BLAST)

=> females for ca. 10 species had visible blood meals

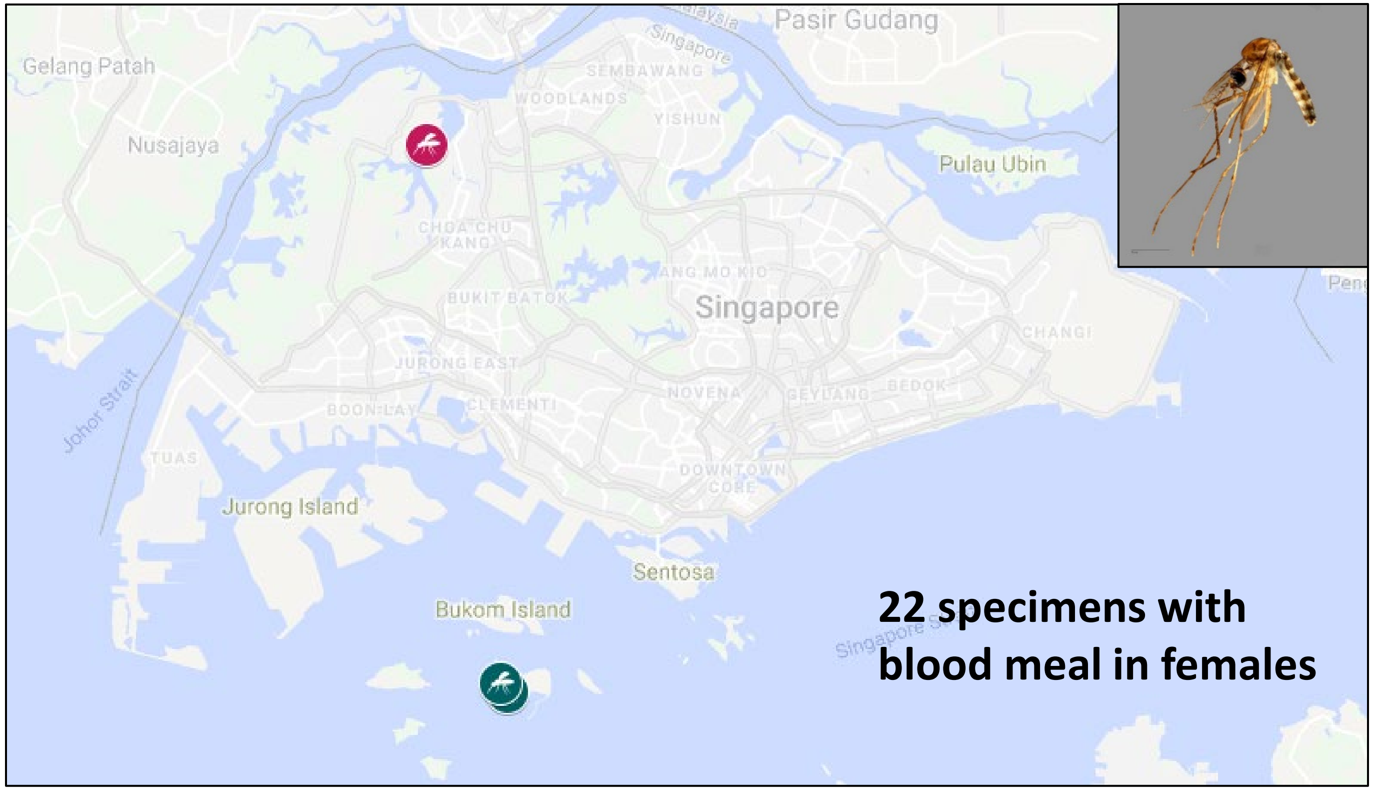
=> metabarcoding of gut content

Aedes sp. ZRCBDP00009685

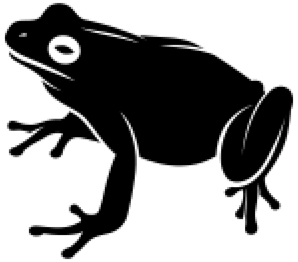


1384 specimens

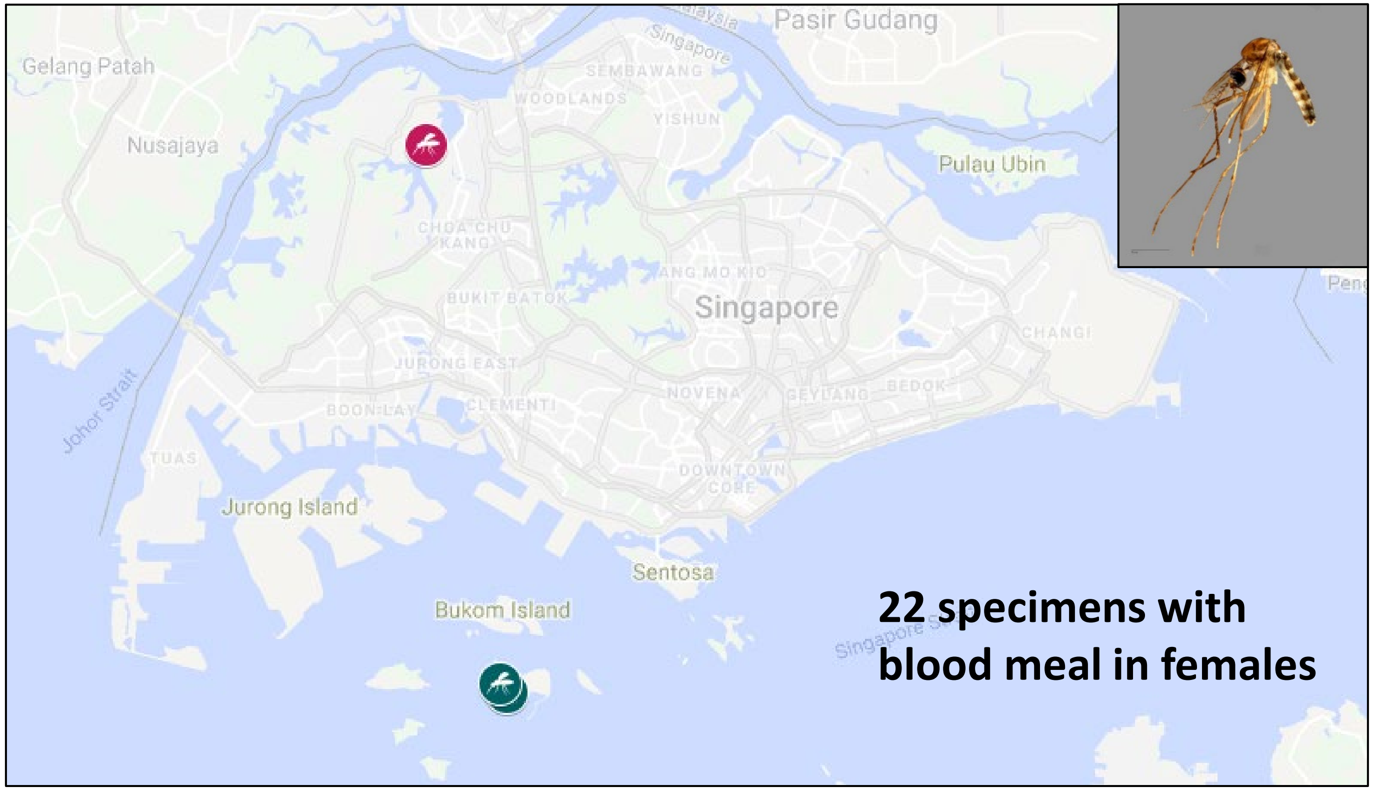
Aedes sp. ZRCBDP00009685



22 specimens with blood meal in females



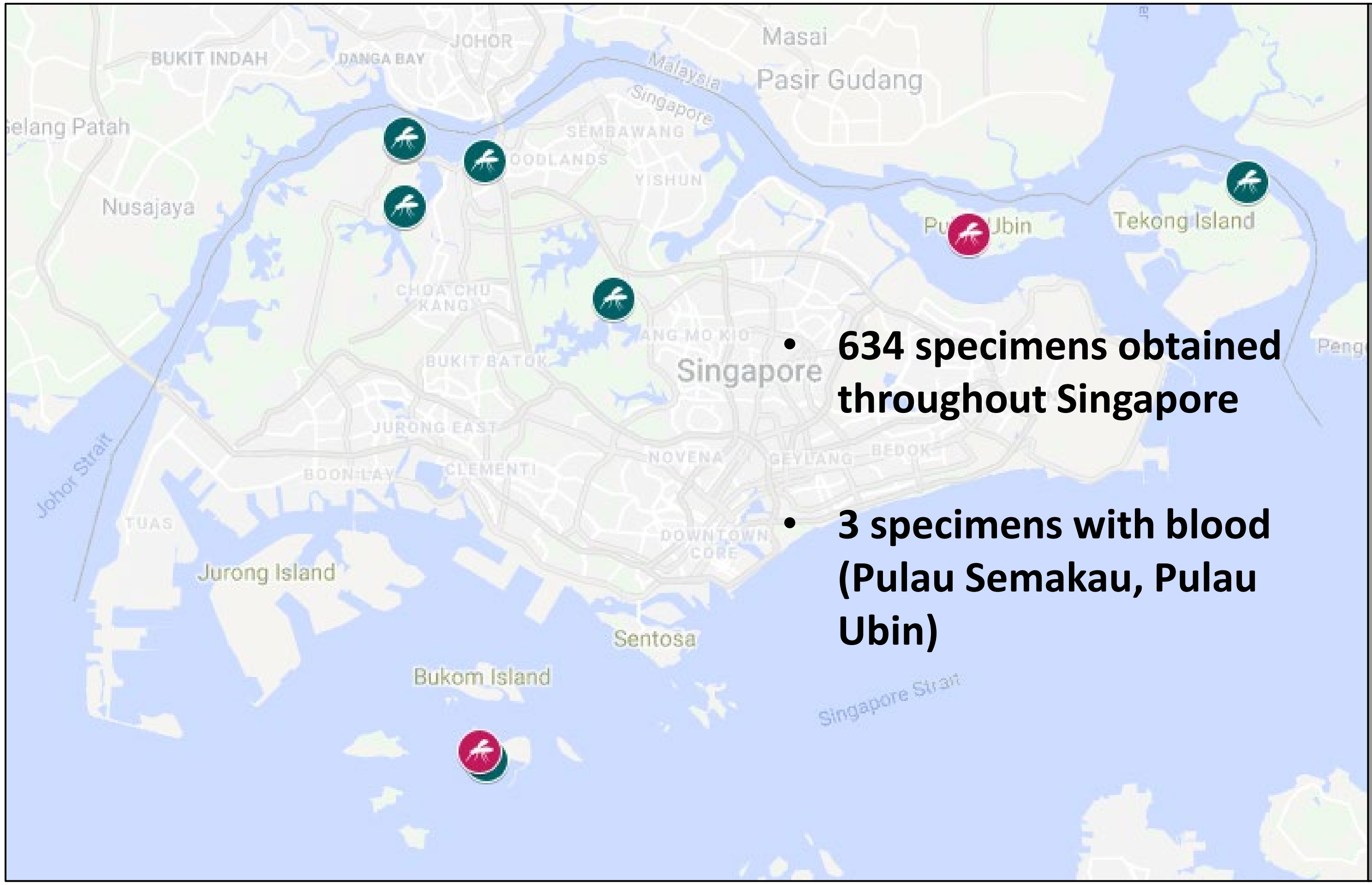
Aedes sp. ZRCBDP00009685



22 specimens with blood meal in females

=> high potential as an agent transmitting viruses between distantly related hosts, but fortunately with restricted distribution

Uranotaenia sp. ZRCBDP0009257



- **634 specimens obtained throughout Singapore**
- **3 specimens with blood (Pulau Semakau, Pulau Ubin)**

Leong
Tzi Ming

Example 2: Dragonfly nymph ecology

Singapore's odonates (122 extant spp)

- 1500 specimens sequenced

=> larval and adult matches for 59 species

=> molecular work: 1-week

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Conclusions

- Society expects us to monitor biodiversity holistically: we are failing
- We can catch up quickly as long as we use robotics, new DNA sequencers, and AI
- But we need “Dark Taxon Biology” to complement existing fields in biology
- Goal should be that biodiversity samples can be scanned like libraries
    - already realized for some samples and taxa
    - needs to be expanded to dark taxa

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Biodiversity Discovery

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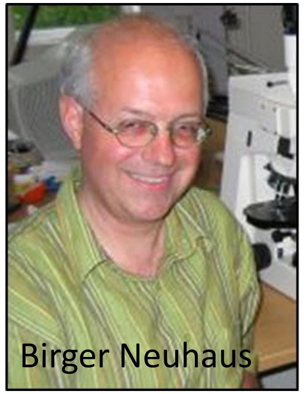
Amrita Srivathsan


Ambrosio Torres


Bernardo Santos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Bonnie Blaimer

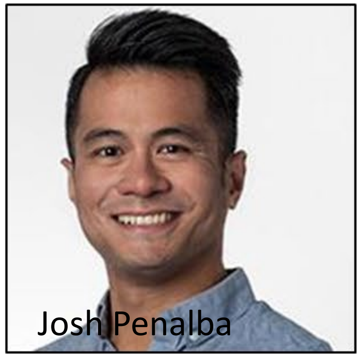

Dr. Liliia Serbina


Paco Hita-Garcia


Gregor Hagedorn


Johannes Frisch


Jörg Freyhof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