

ESOVE 2024

The XXIIIrd European
Society for Vector Ecology
Conference



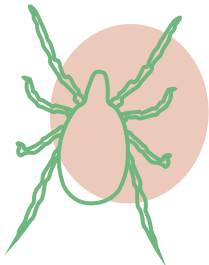
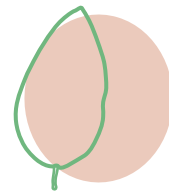
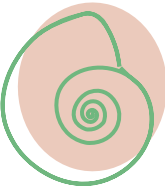
ONE HEALTH IN ACTION:
supporting and accelerating
the bridging of the vertebrate
and plant health communities

14-17th Oct. 2024
Montpellier, France

BOOK OF ABSTRACTS

Program - Abstracts - Posters
Lists of participants, partners & exhibitors

[https://www.alphavisa.com/
e-sove/2024/index.php](https://www.alphavisa.com/e-sove/2024/index.php)





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Tel: +33 4 86 94 97 30

WELCOME TO THE 23rd EUROPEAN SOCIETY FOR VECTOR ECOLOGY CONFERENCE

Dear Conference Participants, Dear Colleagues, Dear Friends,

Twelve years after ESOVE 2012 in Montpellier, we're excited to welcome you again to the **ESOVE 2024** congress, and we hope you have a fantastic time both at the conference and exploring the local area.

Your presence is highly appreciated, and we are thrilled to have you join us for what promises to be an inspiring and informative event in the light of One Health! We believe that a friendly atmosphere fosters collaboration and innovation. We have a fantastic lineup of speakers and keynote speakers, as well as engaging posters targeting arthropod-borne pathogens and vector species, not only in vertebrates but also in plants. This conference aims at highlighting the numerous bridges between scientific communities, and emphasizes the need for collaborative, multisectoral and transdisciplinary research for surveillance, warning, and innovative vector and pest control.

We believe **ESOVE 2024** is the opportunity to network and connect with fellow attendees. We believe that these interactions can lead to collaboration, inspiration, and professional growth. We strongly encourage the junior community to engage into networking opportunities: we trust that you will gain insights and connections that will benefit your professional journey. **Indeed, ESOVE and the local committee are happy to award three prizes to Best oral communication (PhD students and postdoctoral researchers), Best poster (Master students and PhD students) and Best poster (postdoctoral researchers).**

At last, we would like to deeply thank all the partners and sponsors for their ongoing support of our organization's event. It plays a key role in the success of our event, and we deeply appreciate your participation.

Thank you for being part of this exciting journey. Together, we'll make this conference a memorable and rewarding experience.

Warm regards,
Vectopole Sud and the local organizing committee



Group picture from ESOVE 2012 in Montpellier

WELCOME ADDRESS

Dear Friends and Colleagues,

Welcome to the 23rd European Society for Vector Ecology Conference, and welcome back to Montpellier!

This year's conference is particularly exciting as we broaden our focus beyond human and veterinary health to include plant health, underscoring the importance of a multisectoral and transdisciplinary approach in vector ecology. By linking diverse perspectives, we hope to inspire fresh ideas and new approaches for those working with insects as they engage with different scientific disciplines. Thank you for bringing your brilliant minds to the meeting! Although this is our first attempt to make this link, since SOVE has had many parts of the One Health concept in its logo for over 50 years, I believe you are at the right place at the right time. It is my absolute pleasure to be a part of it.

Throughout the conference, we will meet to explore critical topics such as advances in vector control, the role of genomics in disease surveillance, and the impacts of climate change on vector-borne diseases. These discussions will highlight the essential bridges between our scientific communities and the need for collaborative research to tackle the complex challenges we face.

This event is not just about sharing knowledge; it's about building and strengthening the connections that make our community resilient and forward-thinking. As always, I encourage you to engage fully, challenge ideas, and forge new partnerships that will shape the future of our field.

My heartfelt gratitude goes to the local organizing committee, the members of the Vectopole Sud, and the ESOVE Scientific Committee. Their dedication and hard work have made this gathering possible and have shaped such an outstanding scientific program!

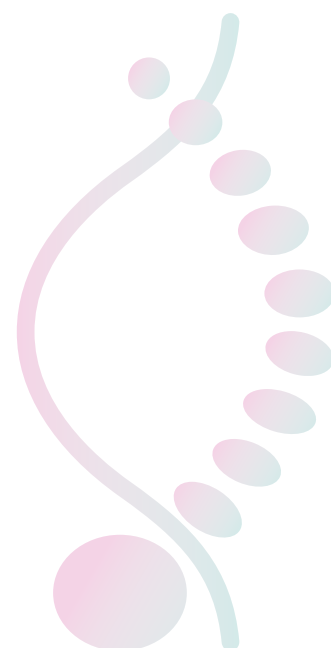
I wish you all a productive and enjoyable conference enriched by diverse viewpoints and approaches.

Stay curious, and stay healthy.

Warm regards,

Filiz Günay

European Region of Society for Vector Ecology Director



ORGANIZING AND SCIENTIFIC COMMITTEES

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- **Michelle BROWN** - SOVE Executive director, USA
- **Alex CHASKOPOULOU** - USDA-EBCL, Thessaloniki, Greece
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- **Filiz GÜNAY** - European SOVE director - Hacettepe University, Turkey

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- Emilie BOUHSIRA - ENVT, Toulouse
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Hacettepe University, Turkey
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- Andrei MIHALCA - USAMV Cluj-Napoca, Romania
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- Nicolas SAUVION - INRAE, France

Administrative secretariat

Registration, accommodation - Sponsorship

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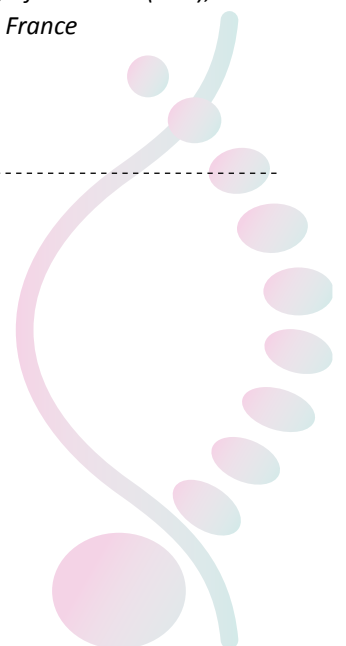
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ESOVE 2024

ORGANIZERS



<https://www.vectopole-sud.fr/>



<https://www.sove.org/>

PARTNERS

Vectopole Sud, SOVE and ESOVE acknowledge our partners and sponsors



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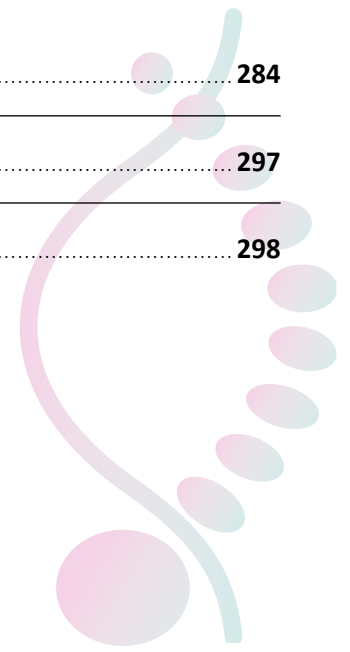
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One Health in action: supporting and accelerating the bridging of the vertebrate and plant health communities

MONDAY 14 OCTOBER

08:00	Conference registration
09:00	Welcome Introduction
09:30	Keynote 1 - Cindy Morris (Plant Pathology Research Unit, INRAE, France)
10:05	SESSION 1 Integrative taxonomy and evolution
11:05	Coffee break - Posters
11:30	SESSION 1 (continued)
13:10	Lunch & Coffee Posters
14:10	SESSION 2 Vector ecology and biology
15:50	Coffee break - Posters
16:10	SESSION 2 (continued) Vector ecology and biology
17:30	Poster Session 1
18:30	Welcome Cocktail

TUESDAY 15 OCTOBER

08:25	Keynote 2 - Cyril Caminade (Earth System Physics Department, The Abdus Salam International Center for Theoretical Physics, Italy)
09:00	SESSION 3 Eco-epidemiology of VBDs under anthropogenic and climatic changes
10:40	Coffee break - Posters
11:10	SESSION 3 (continued) Eco-epidemiology of VBDs under anthropogenic and climatic changes
12:30	Conference Group photo
12:40	Lunch & Coffee
13:40	Poster Session 2
15:00	SESSION 4 Interactions between host(s)-vector(s)-pathogen(s)-microbiota-environment
16:20	Coffee break - Posters
16:50	SESSION 4 (continued) Interactions between host(s)-vector(s)-pathogen(s)-microbiota-environment
18:30	

WEDNESDAY 16 OCTOBER

08:25	Keynote 3 - Tine Huyse (Royal Museum for Central Africa, Belgium)
09:00	SESSION 5 Novel research avenues for innovative vector control strategies
10:00	Coffee break - Posters
10:30	SESSION 5 (continued)
11:30	SESSION 6 Networks and projects: What's new?
12:45	Lunch box
14:00	14:00 - Bus departure <i>From Le Corum - Level 0</i>
	15:00 - Cultural trip Abbey of Valmagne
19:00	Gala Dinner at the Abbey of Valmagne

THURSDAY 17 OCTOBER

09:00	Keynote 4 - Fredros Okumu (University of Glasgow, School of Biodiversity, One Health and Veterinary Medicine, Scotland)
09:35	SESSION 7 Vector surveillance systems, community-based surveillance and management of VBDs
10:35	Coffee break - Posters
11:00	SESSION 7 (continued)
12:40	Closing Session Student awards presentation
13:00	Lunch & Coffee
14:00	

Area Antigone - Level 2
 Auditorium Einstein - Level 0

Colored stars are for oral communications in competition for the awards

★ Best Oral communication - PhD and Post doctoral researchers

PROGRAM - MONDAY 14 OCTOBER

CET (Central European Time)

08:00-09:00 **Conference registration - Welcome coffee**

Area Antigone - Level 2

09:00-09:30 **Welcome Introduction**

Auditorium Einstein - Level 0

Chairs: Didier FONTENILLE (*IRD, Vectopole Sud, Montpellier, France*) & Claire GARROS (*Cirad, Vectopole Sud, Montpellier, France*)

- Prof Jacques MERCIER (*Vice-Président en charge de la Recherche, Université de Montpellier*)
- Mme Clare HART (*Vice-Présidente de Montpellier Méditerranée Métropole, Déléguée au Rayonnement international et Coopération européenne*)
- Mme Claire GATECEL (*Conseillère Régionale, Région Occitanie, Présidente de la Commission Enseignement Supérieur et Recherche*)

09:30-10:05 **Keynote 1**

- **KN-1- What is the interest of Plant Health for One Health?**
Cindy MORRIS (*Plant Pathology Research Unit, INRAE, Montfavet, France*)

10:05-11:05 **Session 1 - Integrative taxonomy and evolution**

Chairs: Thierry LEFEVRE (*CNRS, Montpellier, France*) & Alessandra DELLA TORRE (*Universita Sapienza, Rome, Italy*)

Oral presentations

- ★ **S1-1 - Pangenomic variations and differentiation between the two African malaria vector sister species *Anopheles gambiae* and *Anopheles coluzzii***
Clothilde CHENAL (*MIVEGEC, Univ. Montpellier, CNRS, IRD, Montpellier, France*)
- **S1-2 - Speciation within the *Anopheles gambiae* complex: high-throughput whole genome sequencing reveals evidence of a putative new cryptic taxon in 'far-west' Africa**
Alessandra DELLA TORRE (*Dpt of Public Health and Infectious Diseases, Sapienza University, Rome, Italy*)
- **S1-3 - Genomic diversity, evolution, and adaptation of *Anopheles bwambae* to the extreme thermal spring waters in Uganda** - Michael C. FONTAINE (*MIVEGEC, CNRS - U Montpellier - IRD, Montpellier, France*)

11:05-11:30 **Coffee break - Posters**

Area Antigone - Level 2

11:30-13:10 **Session 1 (continued)**

Auditorium Einstein - Level 0

Chairs: Thierry LEFEVRE (*CNRS, Montpellier, France*) & Alessandra DELLA TORRE (*Universita Sapienza, Rome, Italy*)

Oral presentations

- ★ **S1-4 - Characterization of *Aedes aegypti* from three West African countries using morphological, morphometric and genetic analysis**
Eric AGBOLI (*Vector control, Bernhard Nocht Institute for Tropical Medicine, Hamburg, Hamburg, Germany*)
 - **S1-5 - A worldwide study of structural variants of insecticide resistance genes in *Culex pipiens s.l.***
Pierrick LABBÉ (*ISEM, Université de Montpellier, Montpellier, France*)
 - **S1-6 - A decade of mass spectrometry applications on phlebotomine sand flies: what we know and what we want to know more** - Vit DVORAK (*Dpt of Parasitology, Faculty of Science, Charles University, Prague 2, Czechia*)
 - **S1-7 - An integrative approach to combat cryptic speciation in a complex of biting midges**
Phillip SHULTS (*ARS, USDA, Manhattan, Kansas, United States*)
- ... /...

- **S1-8 - Integrative taxonomy of nearly 200 years old type specimens of *Culicoides vexans* Staeger leading to unexpected taxonomic implications**
Bruno MATHIEU (*UR3073 PHAVI - Medical School, University of Strasbourg, Strasbourg, France*)

13:10-14:10 **Lunch & Coffee - Posters**Area *Antigone* - Level 214:10-15:50 **Session 2 - Vector ecology and biology**Auditorium *Einstein* - Level 0Chairs: Filiz GÜNAY (*University of Florida, Vero Beach, USA*) & Chris SANDERS (*The Pirbright Institute, Woking, UK*)*Oral presentations*

- ★ **S2-1 - Temperature and humidity limits for flight activity and survival of field-collected *Culicoides* in the UK**
Laura JONES (*The Pirbright Institute, Woking, United Kingdom*)
- ★ **S2-2 - Tick-borne encephalitis virus in the Netherlands: Exploring the interplay between ticks, viruses and rodents**
Julian BAKKER (*Laboratory of Entomology, Wageningen University & Research, Wageningen, Netherlands*)
- **S2-3 - The cellular and molecular components of the proboscis in the tiger mosquito *Aedes albopictus***
Paolo GABRIELI (*Dept. Biosciences, University of Milan, Milano, Italy*)
- ★ **S2-4 - The genetic bases of deltamethrin resistance in *Aedes albopictus* - A genomic travelling around the world**
Tiphaine BACOT (*Laboratoire d'écologie alpine, Université Grenoble Alpes, Gières, France*)
- **S2-5 - The seasonal thermal ecophysiology and egg metabolome point to different overwintering strategies in two invasive *Aedes* mosquito species**
Ruth MÜLLER (*Unit Entomology, Biomedical Sciences, Institute of Tropical Medicine, Antwerp, Belgium*)

15:50-16:10 **Coffee break - Posters**Area *Antigone* - Level 216:10-17:30 **Session 2 (continued)**Auditorium *Einstein* - Level 0Chairs: Filiz GÜNAY (*University of Florida, Vero Beach, USA*) & Chris SANDERS (*The Pirbright Institute, Woking, UK*)*Oral presentations*

- **S2-6 - Unlocking *Aedes albopictus* Expansion: Integrating Microhabitat Climate in Species Distribution Models**
Katie Milligan SUSONG (*Pathobiological Sciences, University of Wisconsin - Madison, Madison, WI, United States*)
- ★ **S2-7 - Resting site selection and microclimate of adult mosquitoes along a land use gradient**
Carmen VILLACAÑAS DE CASTRO (*Carl von Ossietzky Universität Oldenburg, Oldenburg, Germany*)
- ★ **S2-8 - Toc-toc: who is inside? Uncovering indoor host-seeking mosquito dynamics by Citizen Science**
Eleonora LONGO (*Center Agriculture Food Environment, University of Trento, San Michele all'Adige, Italy*)
- **S2-9 - Diversity of the *Aedes aegypti* group in the Southwest Indian Ocean**
Andrea GLORIA-SORIA (*Department of Entomology, The Connecticut Agricultural Experiment Station, New Haven, CT, United States*)

17:30-18:30 **Poster Session 1**Area *Antigone* - Level 218:30 **Welcome Cocktail**Area *Antigone* - Level 2

PROGRAM - TUESDAY 15 OCTOBER

CET (Central European Time)

08:25-09:00

Keynote 2

Auditorium Einstein - Level 0

- **KN-2 - Impact of recent and future climate change on vector-borne diseases**
Cyril CAMINADE (*Earth System Physics Department, The Abdus Salam International Center for Theoretical Physics, Trieste, Italy*)

09:00-10:40

Session 3 - Eco-epidemiology of VBDs under anthropogenic and climatic changes

Chairs: Didier FONTENILLE (*IRD RIVOC, Montpellier, France*) & Antoine BARREAUX (*Cirad, Nairobi, Kenya*)

Oral presentations

- ★ **S3-1 - Predicting the current and future geographic distribution of *Lutzomyia* spp., vectors of *Leishmania* spp., under climate change, in North America**
Sydney DEWINTER (*AEROH Lab, Department of Population Medicine, Ontario Veterinary College, University of Guelph, Guelph, Ontario, Canada*)
- **S3-2 - The rising global economic costs of invasive *Aedes* mosquitoes and *Aedes*-borne diseases**
David ROIZ (*MIVEGEC, Institut de recherche pour le Développement (IRD), Montpellier, France*)
- **S3-3 - One Health innovative sampling to understand the ecology and spread of Usutu virus and West Nile virus in the Netherlands, 2016-2022**
Reina SIKKEMA (*Virology, ErasmusMC, Rotterdam, The Netherlands*)
- ★ **S3-4 - Modeling mosquito thermal performance and West Nile virus transmission suitability with Bayesian hierarchical models** - Julian HEIDECHE (*Heidelberg University, Heidelberg, Germany*)
- **S3-5 - Molecular Xenomonitoring (MX) allows real-time surveillance of West Nile and Usutu virus in mosquito populations**
Nazli AYHAN (*Unité des Virus Émergents, Aix-Marseille University, Marseille, France*)

10:40-11:10

Coffee break - Posters

Area Antigone - Level 2

11:10-12:30

Session 3 (continued)

Auditorium Einstein - Level 0

Chairs: Didier FONTENILLE (*IRD RIVOC, Montpellier, France*) & Antoine BARREAUX (*Cirad, Nairobi, Kenya*)

Oral presentations

- ★ **S3-6 - Alien lymnaeid snails in the Western Mediterranean basin: new invaders and trematode-transmitters in the region?** - Annia ALBA MENÉNDEZ (*Université de Montpellier, Perpignan, France*)
- ★ **S3-7 - Lyme Borreliosis in Bonn, Germany under an integrated approach of One Health and Socio-Ecology**
Maria Luisa ESPINEL RAMOS (*Center for Development Research (ZEF), University of Bonn, Bonn, Germany*)
- ★ **S3-8 - Green cities and the risk for vector-borne diseases transmission** - Mathilde MERCAT (*UMR MIVEGEC, University of Montpellier, IRD, CNRS, Montpellier, France*), presented by Colombine BARTHOLOMÉE
- **S3-9 - Phenotypic plasticity in vector traits drives trends in global disease incidence: *Aedes albopictus* and dengue** - Steven WHITE (*UK Centre for Ecology and Hydrology, Wallingford, United Kingdom*)

12:30-12:40

Conference Group Photo

12:40-13:40

Lunch & Coffee - Posters

Area Antigone - Level 2

13:40-15:00

Poster Session 2

Area Antigone - Level 2

15:00-16:20

Session 4 - Interactions between host(s)-vector(s)-pathogen(s)-microbiota-environment

Chairs: Thomas POLLET (INRAE, Montpellier, France) & Maryline UZEST (INRAE, Montpellier, France)

Auditorium *Einstein* - Level 0*Oral presentations*

- ★ **S4-1 - West Nile virus-related activity alterations and neuronal infection in *Culex pipiens***
Jaume GARDELA (IRTA, Centre de Recerca en Sanitat Animal (CRESA, IRTA-UAB), Bellaterra, Barcelona, Spain)
- **S4-2 - Understanding factors shaping mosquito microbiome assembly using gnotobiotic approach**
Shivanand HEGDE (School of Life Sciences, Keele University, Newcastle, United Kingdom)
- **S4-3 - Analyzing the Complexity: interactions of arboviruses and insect-specific viruses in vector competence studies with two invasive *Aedes Species***
Anna HEITMANN (Department of Arbovirology and Entomology, Bernhard Nocht Institute for Tropical Medicine, Hamburg, Germany)
- ★ **S4-4 - Impacts of insecticide resistance on Usutu virus transmission and associated life-history parameters in *Culex quinquefasciatus* mosquitoes**
Maxime PRAT (ISEM, Université de Montpellier, Montpellier, France)

16:20-16:50

Coffee break - PostersArea *Antigone* - Level 2

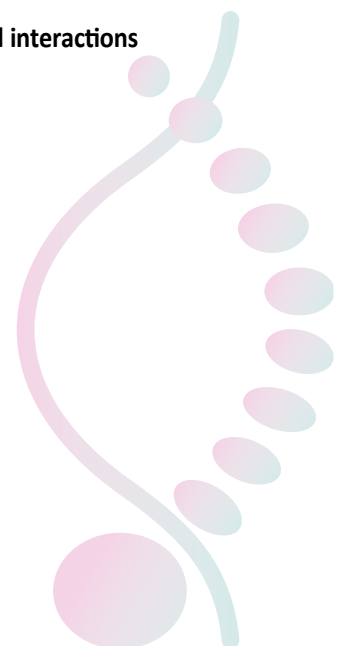
16:50-18:30

Session 4 (continued)Auditorium *Einstein* - Level 0

Chairs: Thomas POLLET (INRAE, Montpellier, France) & Maryline UZEST (INRAE, Montpellier, France)

Oral presentations

- **S4-5 - Infection dynamics and impact of blood feeding on the susceptibility of *Culicoides biting* midges to bluetongue virus (BTV)**
Christopher SANDERS (Entomology, The Pirbright Institute, Woking, Surrey, United Kingdom)
- ★ **S4-6 - Identification of receptors of the plant virus Turnip mosaic virus (*Potyviridae*) in their aphid vectors**
Yu FU (Plant Health Institute Montpellier (PHIM), Montpellier, France)
- **S4-7 - The aphid factor: how aphid genetic diversity and the endosymbionts they carry may impact virus epidemiology**
Trisna TUNGADI (Keele University, Keele, United Kingdom)
- ★ **S4-8 - A flower pathway for arbovirus transmission? Unravelling sand fly - Phlebovirus interactions**
Lison LAROCHE (Life Sciences, Imperial College London, London, United Kingdom)
- ★ **S4-9 - The *Hyalomma marginatum* tick holobiont: composition, dynamics and interactions**
Charlotte JOLY KUKLA (ASTRE, Cirad-INRAE, Montpellier, France)



PROGRAM - WEDNESDAY 16 OCTOBER

CET (Central European Time)

08:25-09:00

Keynote 3

Auditorium *Einstein* - Level 0

- **KN-3 - sNailed it: Unlocking the potential of Citizen Science to control and prevent snail-borne diseases**
Tine HUYSE (*Royal Museum for Central Africa, Tervuren, Belgium*)

09:00-10:00

Session 5 - Novel research avenues for innovative vector control strategies

Chairs: Fabrice CHANDRE (*IRD, Montpellier, France*) & Eleonora FLACIO (*SUPSI, Mendrisio, Switzerland*)

Oral presentations

- **S5-1 - Monitoring individual rice field flooding dynamics over large scales to inform mosquito surveillance and control** - Mauricianot RANDRIAMIHAJA (*ONG PIVOT, Ranomafana, Madagascar*)
- ★ **S5-2 - Reducing densities of zoophagous Anopheles by treating cattle with an innovative formulation of Ivermectin while preserving the environment: a One-Health approach**
Angélique PORCIANI (*MIVEGEC, Université de Montpellier-CNRS-IRD, Montpellier, France*)
- **S5-3 - The use of Wolbachia for dengue control - New Caledonia as a case study**
Edwige RANCÈS (*World Mosquito Program, Pau, France*)

10:00-10:30

Coffee break - Posters

Area *Antigone* - Level 2

10:30-11:30

Session 5 (continued)

Auditorium *Einstein* - Level 0Chairs: Fabrice CHANDRE (*IRD, Montpellier, France*) & Eleonora FLACIO (*SUPSI, Mendrisio, Switzerland*)

Oral presentations

- **S5-4 - Development of N-arylamide insecticides to control hemipteran pests of agricultural systems**
Daniel SWALE (*Emerging Pathogens Institute, Department of Entomology, University of Florida, Gainesville, FL, United States*)
- **S5-5 - Vectrap: a 3 years-field evaluation of mass-trapping strategy against *Aedes albopictus* and *Aedes aegypti* implemented in residential areas**
Nicolas LE DOEUFF LE ROY (*EID Méditerranée, Montpellier, France*)
- ★ **S5-6 - Exploiting sugar-feeding behaviour for the control of *Culicoides* biting midges**
Alec HOCHSTRASSER (*Vector Entomology Group, Institute of Parasitology, Zürich, Switzerland*)

11:30-12:45

Session 6 - Networks and projects: What's new?

Auditorium *Einstein* - Level 0Chairs: Florence FOURNET (*IRD, Montpellier, France*) & Vit DVORACK (*Faculty of Science - Charles University, Prague, Czech Republic*)

Oral presentations

- **S6-1 - VectorNet3: The third iteration of the European network for medical and veterinary entomology**
Cedric MARSBOOM (*Avia-GIS, Zoersel, Belgium*)
- **S6-2 - MEDILABSECURE A One Health Network to improve preparedness and response capacities to emerging vector-borne diseases increased by environmental and climate change**
Vanessa LAGAL (*Department of International Affairs, Institut Pasteur, Paris, France*)
- **S6-3 - VECLim Advances: Climate Sensitive Models of Vector Dynamics and Disease Transmission**
Kamil ERGULER (*CARE-C, The Cyprus Institute, Nicosia, Cyprus*)

.../...

- **S6-4 - Shortening the gap between vector surveillance and first response in arboviruses outbreaks: from REVIVE network program to MOBVEC project**
 Hugo Costa OSÓRIO (*Centre for Vectors and Infectious Diseases Research (CEVDI), National Institute of Health (INSA), Águas de Moura, Palmela, Portugal*)
- **S6-5 - A federative scientist Network under One Health approach to enhance people well-being and sustainable development in Côte d'Ivoire**
 Mathurin KOFFI (*Research Unit of Genetic and Molecular Epidemiology, UJLoG, Daloa, Côte d'Ivoire*)

12:45-14:00 **Lunch box**

Area *Antigone* - Level 2

14:00-19:00 **Cultural trip: Abbey of Valmagne**

14:00 Bus departure from Le Corum

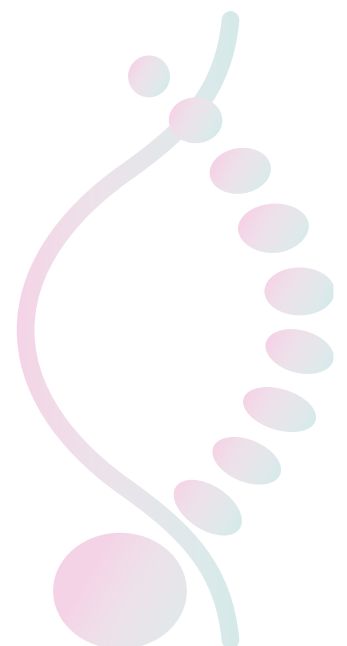
Level 0 - Allé de la Citadelle

15:00 Arrival at the Abbey of Valmagne for guided activities

19:00 **Gala Dinner at the Abbey of Valmagne**

22:00 Return first bus to Montpellier Triangle

00:00 Last departure to Montpellier Triangle



PROGRAM - THURSDAY 17 OCTOBER

CET (Central European Time)

09:00-09:35

Keynote 4

Auditorium *Einstein* - Level 0

- **KN-4 - Exploiting the Biology of Dominant Vector Species to Improve Malaria Control in Africa**
Fredros OKUMU (*Ifakara Health Institute & University of Glasgow, Glasgow, United Kingdom*)

09:35-10:35

Session 7 - Vector surveillance: surveillance systems, community-based surveillance and management of VBDs

Chairs: Alex CHASKOPOULOU (*USDA ARS EBCL, Thessaloniki, Greece*) & Jerome BARON (*Heidelberg University Hospital, Heidelberg, Germany*)

Oral presentations

- **S7-1 - Integrating Citizen and Authoritative Surveillance Data in Mosquito Distribution Models**
Catuxa CERECEDO-IGLESIAS (*Centre d'Estudis Avançats de Blanes (CEAB-CSIC), Blanes, Catalunya, Spain*)
- **S7-2 - Preventing Another Useless Modelling Task**
Heidi BROWN (*University of Arizona, Tucson, AZ, U.S.A.*)
- **S7-3 - Success in Mosquito Control: An Integrated Approach in Istanbul, Turkey**
Bulent ALTEN (*Ecology/vErg Lab./Biology, Hacettepe University, Ankara, Turkey*)

10:35-11:00

Coffee break - Posters

Area *Antigone* - Level 2

11:00-12:40

Session 7 (continued)

Auditorium *Einstein* - Level 0

Chairs: Alex CHASKOPOULOU (*USDA ARS EBCL, Thessaloniki, Greece*) & Jerome BARON (*Heidelberg University Hospital, Heidelberg, Germany*)

Oral presentations

- **S7-4 - Circulation of West Nile virus in active *Culex pipiens* mosquito populations in the Attica region 2021-2023** - Georgios BALATSOS (*Laboratory of Insects & Parasites of Medical Importance, Benaki Phytopathological Institute, Kifisia, Attica, Greece*)
- **S7-5 - Unraveling Dissemination Trajectories of Insect Vectors : Towards Enhanced Prophylaxis in Plant Health** - Margaux DARNIS (*Pathologie Végétale, INRAE, Avignon, France*)
- **S7-6 - Novel eDNA-based surveillance strategy targeting quiescent ticks in soil**
Androniki CHRISTAKI1 (*European Biological Control Laboratory, USDA-ARS, Thessaloniki, Greece*)
- **S7-7 - Testing a novel Optical Sensor for *Aedes* and *Culex* adult automatic count and identification and BG-trap capture rate determination**
Martina MICOCCI (*Dpt of Public Health and Infectious Diseases, Sapienza University of Rome, Rome, Italy*)
- **S7-8 - Estimating the probability that observed absence of tsetse (Diptera: Glossinidae) indicates elimination**
John HARGROVE (*South African Centre for Epidemiological Modelling and Analysis (SACEMA), Stellenbosch University, Stellenbosch, South Africa*)

12:40-13:00

Closing Session

- Filiz GÜNAY (*European SOVE director, Hacettepe University, Turkey*)
- Alexandra CHASKOPOULOU (*USDA-EBCL, Thessaloniki, Greece*)
- Fabrice CHANDRE (*IRD, Montpellier, France*)

- Student awards presentation

13:00-14:00

Lunch & Coffee

Area *Antigone* - Level 2

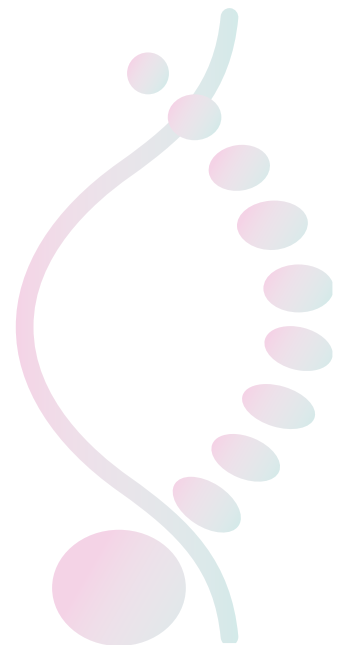


ONE HEALTH IN ACTION

14-17th October 2024
MONTPELLIER, FRANCE



ABSTRACTS MONDAY 14 OCTOBER



What is the interest of Plant Health for One Health?

Cindy MORRIS (cindy.morris@inrae.fr)

Plant Pathology Research Unit, INRAE, Montfavet, France

One Health professes that the health of organisms is interconnected through exploitation of planetary resources, trade and transportation, in particular. The impetus for the emergence of this concept in the early 2000's was knowledge of the epidemiology of zoonotic diseases that put humans at risk to diseases carried by animals. In spite of the intended comprehensiveness of One Health, the place of plant health in this concept is vague and few issues about plant health are debated in the scientific literature related to One Health. The history of concepts on which the modern One Health concept is founded suggests why there is this schism between the plant sciences and the medical and veterinary sciences concerning the adoption of the modern One Health concept. There is a rich history in the plant sciences of concepts concerning the oneness of plants, animals and humans and the debates about the definition and scope of sustainability that are precursors to One Health. These concepts continue to be foundations for research and development, particularly for food security and food safety, under the umbrellas of agroecology and sustainable development. The emergence of these concepts from plant sciences was based on fundamental understanding of the food web – where plants are food for humans and animals whose digestive processes create important resources for plant growth and health. Yet, this latter part of the food web – recycling of manures in particular - was ruptured during modernization of agriculture. Attaining sustainable One Health will depend on restoring this part of the food web via soil stewardship, whose principal guarantors are the ensemble of actors in plant production.

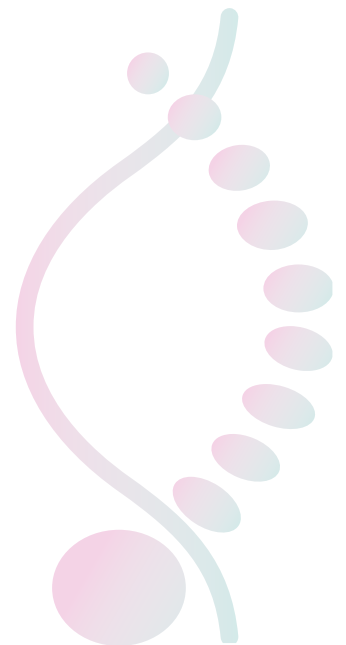


ONE HEALTH IN ACTION

14-17th October 2024
MONTPELLIER, FRANCE



Integrative taxonomy and evolution



Pangenomic variations and differentiation between the two African malaria vector sister species *Anopheles gambiae* and *Anopheles coluzzii*

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Structural variations (SVs) play pivotal roles genome architecture, potentially involved in adaptation and speciation, yet they are often overlooked. Here, we delved into the genomic landscape of SVs within *Anopheles gambiae* and *Anopheles coluzzii*, principal African malaria vectors. We used a «map-then-assemble» pangenomic strategy with short-read sequencing data, validated with samples for which short- and long-read data were available. Then we assessed pangenomic diversity and divergence in the two sister species by screening 200 samples across Africa from the *Ag1000g* consortium. Our study revealed strong disparities in read mapping efficiency across five reference genomes available in the literature. Using the *Ngouso* reference as a backbone for building the panreference, we identified substantial SVs within the reference, with ~10% (23Mb) as dispensable. *De novo* assembled SVs further augmented the reference genome with 1.5M new variants, totalling 521.4Mb (223% larger than *Ngouso*) across the 200 genomes. However, only 15% (234k SVs; 82.1Mb) of these SVs uniquely anchored into the reference, and 3% (46k SVs; 15.4Mb) with high confidence. Notably, our analysis delineated distinct pangenomic dynamics between autosomes and the X chromosome, particularly within heterochromatic regions. In particular, low-recombining regions previously identified as «*genomic islands of differentiation*» between *An. gambiae* and *An. coluzzii* were enriched in diagnostic pangenomic variations, dominated by young transposable elements. Moreover, SVs captured the key axes of ancestry variation along the genome previously identified with single nucleotide polymorphisms (SNPs), including well-known chromosomal inversions and islands of species differentiation, implying that SVs are impacted by the same evolutionary forces. This study offers critical insights into the genetic diversity and evolutionary trajectories of *An. gambiae* and *An. coluzzii*, highlighting SVs, particularly transposable elements, as pivotal drivers in their adaptive and speciation processes.

Speciation within the *Anopheles gambiae* complex: high-throughput whole genome sequencing reveals evidence of a putative new cryptic taxon in 'far-west' Africa

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¹ Department of Public Health and Infectious Diseases, Sapienza University, Rome, Italy ; ² Wellcome Sanger Genomic Surveillance Unit, Wellcome Sanger Institute, Hinxton, Cambridge, United Kingdom ; ³ Harvard T.H. Chan School of Public Health, Boston, MA, United States ; ⁴ Broad Institute, Cambridge, MA, United States ; ⁵ Liverpool School of Tropical Medicine, Liverpool, United Kingdom

Introduction and Objectives

The two main Afrotropical malaria vectors - *Anopheles coluzzii* and *An. gambiae* – are genetically distinct and reproductively isolated across West Africa. However, populations at the western extreme of their range are assigned either as “intermediate” between the two species by Whole Genome Sequencing (WGS) data, or as hybrids by conventional molecular diagnostics. We carried out population genomic, divergence and demographic analyses on populations across west Africa to understand the actual nature of these populations, based on WGS data from the *Anopheles gambiae* 1000 Genomes network.

Materials and Methods

Bioinformatic genomic analyses were conducted on 1,190 fully whole genome genotyped adult females from 5 *An. coluzzii*, 5 *An. gambiae*, and 3 “intermediate” far-west populations: i) PCA and ADMIXTURE analyses were employed to elucidate patterns of genetic variation; ii) nucleotide diversity and divergence measures were utilized to quantify levels of genetic differentiation and divergence; iii) TreeMix, Patterson’s D statistics, and F3 statistics were employed to infer gene flow and introgression events; iv) $\delta a \delta i$ was applied to explore the demographic history in the far-west; v) CNV analyses and haplotype diversity selection scans were conducted to identify genomic regions under positive selection associated with metabolic traits and target site resistance.

Results, Discussion and Conclusion

Results suggest that “intermediate” populations belong to a novel demographically stable cryptic taxon in the far-west region, strongly differentiated from *An. coluzzii* and *An. gambiae* and originated at the same time as the split between the two species. Intriguingly, this taxon lacks insecticide resistance mechanisms commonly observed in the two main species. These findings lead to a change of perspective on malaria vector species in the far-west region with potential for epidemiological implications, and a new challenge for genetic-based mosquito control approaches.

Genomic diversity, evolution, and adaptation of *Anopheles bwambae* to the extreme thermal spring waters in Uganda

Michael C. FONTAINE^{1,2} (michael.fontaine@cncrs.fr), Frédéric LABBÉ^{3,4}, R. Rebecca LOVE³, Loïc TALIGNANI¹, Adeyemi T. AKINADE¹, Scott T. SMALL³, Christina BERGEY⁵, Robert W WATERHOUSE⁶, Jorge E. AMAYA-ROMERO^{7,8}, Jonathan K. KAYONDO⁹, Krystal N. BIRUNGI⁹, Victor BALYESIMA⁹, Harold TOWNSON¹⁰, Nora J. BESANSKY³

¹ MIVEGEC, CNRS - U Montpellier - IRD, Montpellier, France ; ² GELIFES, University of Groningen, Groningen, Netherlands ; ³ Department of Biological Sciences, University of Notre Dame, South Bend, IN, United States ; ⁴ UMR PVMT, CIRAD, Saint Pierre, La Reunion ; ⁵ Department of Genetics, Rutgers University, Piscataway, NJ, United States ; ⁶ Swiss Institute of Bioinformatics, Lausanne, Switzerland ; ⁷ GELIFES, University of Groningen, Groningen, Netherlands ; ⁸ T.H. Chan School of Public Health, Harvard University, Boston, MA, United States ; ⁹ Department of Entomology, Uganda Virus Research Institute, Entebbe, Uganda ; ¹⁰ Liverpool School of Tropical Medicine, Liverpool, United Kingdom

The closely related and partially interfertile mosquito species of the *Anopheles gambiae* complex (AGC) include the major vectors of human malaria in Africa. While these have attracted much attention, little is known about minor vectors like *An. bwambae*. This species is highly specialized to the thermal springs in Bwambae County (Uganda), in close vicinity with other major vectors (e.g., *An. gambiae*). Previously reported potential introgression between them raised questions about how such species remain adapted despite the homogenizing effects of gene flow.

Here we investigated the genomic diversity and evolutionary history of *An. bwambae*, together with its extreme adaptations to thermal spring waters. We first *de novo* assembled the reference genome for this species and compared it to those from other species of the AGC. Then, using whole genome sequencing, bioinformatics, population genomics, and phylogenomics approaches, we investigated the genetic diversity, evolutionary history, and adaptation of *An. bwambae* (n=42) compared to *An. gambiae* (n=25). Analyses of 13.5 million SNPs across the genomes of the two species revealed major genomic heterogeneity including a new inversion system on the X chromosome of *An. bwambae*. Genetic structure confirmed that the two species are well distinct, with no evidence of recent gene flow. Nevertheless, historical introgression was detected not only with *An. gambiae*, but also with other species of the AGC. In contrast to *An. gambiae*, *An. bwambae* displayed low diversity, strong inbreeding, and autozygosity, indicative of a very small population. Meanwhile, while genome scans revealed strong positive selection signals on known insecticide resistance loci in *An. gambiae*, no such signal was identified in *An. bwambae*. Genome scans revealed a few genomic regions potentially undergoing introgression, while the majority did not. How *An. bwambae* persisted and adapted despite its small population size and strong inbreeding remains a paradox warranting further investigation.



Characterization of *Aedes aegypti* from three West African countries using morphological, morphometric and genetic analysis

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¹ Vector control, Bernhard Nocht Institute for Tropical Medicine, Hamburg, Hamburg, Germany ; ² Arbovirus and Entomology Department, Bernhard Nocht Institute for Tropical Medicine, Hamburg, Hamburg, Germany ; ³ Laboratório de Transmissores de Hematozoários, Instituto Oswaldo Cruz, FIOCRUZ, Rio de Janeiro, Brazil ; ⁴ Centre d'Entomologie Médicale et Vétérinaire, Université Alassane Ouattara, Bouake, Côte d'Ivoire ; ⁵ Département de Recherche et Développement, Centre Suisse de Recherches Scientifiques en Côte d'Ivoire, Abidjan, Côte d'Ivoire ; ⁶ Department of Epidemiology and Public Health, Swiss Tropical and Public Health Institute, Basel, Switzerland ; ⁷ Laboratory of Fundamental and Applied Entomology, Université Joseph Ki-Zerbo, Ouagadougou, Burkina Faso ; ⁸ Faculty of Mathematics, Informatics and Natural Sciences, Universität Hamburg, Hamburg, Hamburg, Germany

Aedes aegypti is the most important vector for arboviruses worldwide. The evolutionary origin of *Ae. aegypti* lies in Sub-Saharan Africa, from where it has spread to tropical and subtropical regions throughout the world. In West Africa data on disease and vector ranges are sparse and issues such as genetic and ecological diversity are poorly understood. In this study, we investigated *Ae. aegypti* from Burkina Faso, Côte d'Ivoire, and Ghana collected at urban, peri-urban, rural and sylvatic sites. The characterization included the sequencing of the mitochondrial DNA, the analysis of wing shape and size, and the classification of the percentage of white scales on the first abdominal tergite. Our findings revealed the presence of 4 subclades that can be broadly categorized into two major mitochondrial lineages. The majority of samples from West Africa formed one subclade showing no clustering by country or landscape type. Specimens from the sylvatic site in Burkina Faso were clustering in the other major lineage together with samples from Brazil. Wing shape analysis showed low differences between the country and land use classes. In our study, we identified a population of *Ae. aegypti* that genetically differs from the other populations found in West Africa. It is closely related to new world populations, leading to the assumption that it could be a reintroduction from outside of Africa. The interbreeding with local populations will lead to an increase in genetic variation which could also influence the ability to transmit viruses. Monitoring populations in West Africa will help to anticipate the spread of the diseases that this mosquito is so efficient at transmitting.

A worldwide study of structural variants of insecticide resistance genes in *Culex pipiens s.l.*

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While small-scale variations (substitutions, indels, etc...) and their impact on fitness are well-understood, the impact of large-scale structural variants (duplication, inversion, ...) is now becoming a major trend in evolutionary biology. Their impact on mosquitoes biology and adaptation has however long been recognized, both in terms of speciation and for their impact on insecticide resistance. For example, it is well known that inversions play a key role in the diversification of the species complex *Anopheles gambiae*. In the same complex, the role of duplicated resistance alleles at the *ace-1* locus has also been the focus of several studies: several alleles have been found, either multiple copies of the resistance alleles (homogeneous duplications) or alleles resulting from tandem duplications of resistance and susceptible alleles (heterogeneous duplications).

In this talk, I will present an overview of the diversity of resistance alleles duplications found in the species complex *Culex pipiens s.l.*, at different locus (the major locus implicated in insecticide resistance), taking advantage of our recent analysis of a worldwide sample of mosquitoes whose full genome was sequenced. I will also discuss more generally the role of structural variants in adaptation to environmental variation and their implications in terms of mosquito control.

A decade of mass spectrometry applications on phlebotomine sand flies: what we know and what we want to know more

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¹ Department of Parasitology, Faculty of Science, Charles University, Prague 2, Czechia ; ² Institute of Microbiology of the Czech Academy of Sciences, BioCeV, Vestec, Czechia

Phlebotomine sand flies (Diptera: Psychodidae) are important vectors of diverse pathogens like *Leishmania* protozoans, bacteria and viruses. While many regions suffer from emerging outbreaks of human leishmaniasis, often exacerbated by natural disasters and political unrest, and evidence grows that sand fly-borne viruses, many just newly discovered, are of previously underestimated medical significance, sand fly-borne diseases generally still remain neglected.

When studying the epidemiological role of sand flies, both conclusive species identification and spectrum of hosts help to understand the dynamics of local transmission cycles and to assess potential emergence of sand flies in new regions. MALDI-TOF protein profiling of sand flies from Old World endemic regions like the Mediterranean, East Africa or south-east Asia with standardized trapping, storage and sample preparation allowed conclusive, time-, labour- and cost-effective species identification of field-collected specimens including those yielded by large-scale multinational entomological surveys in the last decade. Species-specific protein profiles proved as useful tool in integrative taxonomy, supporting formal description of new species, suggesting existence of yet undescribed cryptic species, discrimination among sibling species with compiled morphological resolution and challenging validity of established taxons while allowing parallel application of other complementary approaches (DNA barcoding, morphological analysis) and screening for sand fly-borne pathogens. MALDI-TOF peptide mass mapping of host-specific haemoglobin peptides in engorged sand fly females successfully identified the host blood, including mixed bloodmeals, up to 48 hours post feeding, a period longer than analyzable by conventional assays. The method successfully identified also bloodspots on filter paper, enabling easy sample handling, storage and shipment for analysis in field surveys.

Future challenges of mass spectrometry deployment in vector biology include rigorous application of standard protocols, dissemination of necessary machinery and expertise, assessment of intraspecific variability within studied species, modifications to allow parallel pathogen detection and creation of publicly available open-access database of reference protein profiles.

An integrative approach to combat cryptic speciation in a complex of biting midges

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ARS, USDA, Manhattan, Kansas, United States

Introduction and Objectives

Proper species identification is paramount to the surveillance of vector insects, however, this can prove particularly difficult when dealing with cryptic species. In North America, *Culicoides sonorensis* is one of the primary vectors of bluetongue virus. This biting midge species belongs to the *C. variipennis* complex, a group of closely related species whose morphology and genetics overlap substantially. As identification within this group can be challenging, we sought to investigate species delimitation within the complex and develop a tool for identifying the vector species *C. sonorensis*.

Materials and Methods

We used morphological, microsatellite, and genomic data to examine the *C. variipennis* complex. Museum specimens were examined from populations throughout the known ranges of each member of the species complex. Additionally, 206 individuals from 17 populations were sequenced using RADSeq to obtain SNP data. Finally, we utilized several bioinformatic tools to develop and optimize a set of microsatellites to differentiate the members of the *C. variipennis* complex.

Results

In analyzing the SNP data, we found evidence for several cryptic species within this group. Upon examination of the museum species given this new information, slight but novel morphological differences were identified. These results led to a taxonomic revision of the complex by raising two subspecies to full species status as well as describing a new species. A set of seven microsatellites were developed capable of separating the species within this group in addition to a PCR-based assay to identify *C. sonorensis*.

Discussion and Conclusion

The complexity of species identification within the *C. variipennis* complex appears to have been due to cryptic species and incomplete reproductive isolation. Fortunately, this work has offered some clarity in this regard and has produced several effective ways to differentiate the vector species from the non-vector members of this group.

Integrative taxonomy of nearly 200 years old type specimens of *Culicoides vexans* Staeger leading to unexpected taxonomic implications

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¹ UR3073 PHAVI - Medical School, University of Strasbourg, Strasbourg, France ; ² Science Department, Natural History Museum, London, United Kingdom ; ³ MGI Tech UK, Ltd, Scale Space Building C, White City Campus, London, United Kingdom ; ⁴ NIHR Imperial BRC Genomics Facility, Hammersmith Hospital, Imperial College London, London, United Kingdom ; ⁵ Tromsø University Museum, UiT, The Arctic University of Norway, Tromsø, Norway

Introduction and Objectives

The genus *Culicoides* of biting midges (Diptera: Ceratopogonidae) is highly diverse. Most species have spotted wings but in the Palearctic approximately a third of them show plain wings. The latter group includes *C. vexans* Staeger, 1839. The type materials of the species are deposited the zoological museum ZMUC in Copenhagen, and we were authorised to examine and slidemount them and to attempt molecular work. Our study aims to redescribe *C. vexans* and clarify its position within the genus using an integrative taxonomy approach.

Materials and Methods

Nine pinned dry syntypes of *C. vexans* were processed one at a time with particular care. After using a semi-destructive method of DNA extraction, the specimens were mounted on microscope slides. First SANGER sequencing attempt involved amplification of COI fragments. With second approach, the libraries were prepared using Santa Cruz method and sequenced through MGI platform.

Results

Unexpectedly, morphological observation of the slide mounted type specimens didn't fit with the current interpretation of *C. vexans* but match another species of *Culicoides*. Two out of nine specimens were successfully sequenced for a partial COI fragment of 95 bp. Notwithstanding the limited length, those sequences confirm the morphological observations.

Discussion and Conclusion

While SANGER sequencing had given a first insight of the COI of such old type specimens, the application of Santa Cruz method combined with sequencing through MGI platform enables new possibilities. Integrative taxonomy approach on nearly 200 years old syntypes of *C. vexans* implies a deep taxonomic revision and opens the possibility of using such small, historical specimens for phylogenetic and taxonomic analyses.

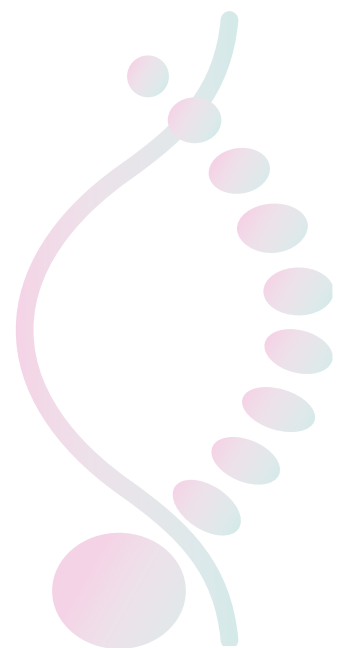


ONE HEALTH IN ACTION

14-17th October 2024
MONTPELLIER, FRANCE



Vector ecology and biology



Temperature and humidity limits for flight activity and survival of field-collected *Culicoides* in the UK

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Introduction and Objectives

Culicoides biting midges (Diptera: Ceratopogonidae) are biological vectors of internationally important arboviruses, including bluetongue virus (BTV) and Schmallenberg virus (SBV), which significantly impact livestock production in Europe. In temperate regions, temperature fundamentally influences the transmission and persistence of *Culicoides*-borne arboviruses by regulating virus replication within the insect as well as impacting vector activity and survival. Studies were conducted to determine the low and high temperature and humidity thresholds of flight activity for UK *Culicoides* vector species.

Materials and Methods

More than 60,000 adult *Culicoides* were collected from field sites in the south east and north east of England using CDC light traps during spring, summer, and autumn. Bioassays conducted under laboratory conditions were used to determine temperature and humidity thresholds of *Culicoides* flight activity by measuring the proportion of *Culicoides* flying towards an ultraviolet light source over a 24-hour period.

Results

Optimal temperatures for flight were recorded between 20-25°C with activity of *Culicoides* declining above and below this. Moreover, seasonal variation in low temperature thresholds for flight were recorded within the subgenus *Avaritia*, with a lower threshold for flight recorded in the autumn (4°C) compared with the summer (10°C) populations. Species differences were also detected with *Culicoides scoticus* significantly more active across all low temperatures tested than *Culicoides obsoletus* ($P < 0.05$). Finally, humidity also influenced *Culicoides* flight activity and survival, significantly reducing these under low humidity conditions (<50% RH).

Discussion and Conclusion

Combined these studies have provided insights into the low and high temperature and humidity thresholds of flight activity of UK *Culicoides* vector species, as well as highlighting seasonal and species differences between thresholds. Data presented here have facilitated the refinement of existing models used to predict incursion and spread of *Culicoides*-borne viruses.



Tick-borne encephalitis virus in the Netherlands: Exploring the interplay between ticks, viruses and rodents

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¹ Laboratory of Entomology, Wageningen University & Research, Wageningen, Netherlands ; ² Dutch National Health Institute (RIVM), De Bilt, Netherlands ; ³ Laboratory of Virology, Wageningen University & Research, Wageningen, Netherlands ; ⁴ Wildlife Ecology and Conservation group, Wageningen University & Research, Wageningen, Netherlands ; ⁵ Wageningen Bioveterinary Research, Lelystad, Netherlands

Tick-borne encephalitis (TBE) is an important arboviral disease in Europe, with 2,000 to 3,500 human cases annually. The transmission of the TBE virus (TBEV) relies on a complex interaction between the virus, ticks, and small rodents. While TBEV is endemic in central and eastern Europe, it is expanding towards western European regions including the Netherlands. The emergence of TBEV in these regions was unforeseen according to previous TBEV distribution models, with factors promoting its geographical spread remaining poorly understood.

In the current study, we sought to understand whether vector competence of ticks is affected by different strains of TBEV. An artificial blood-feeding system was used to infect ticks with different viruses to study the infection of TBEV strains in *Ixodes ricinus* ticks. Furthermore, we studied the role of wood mice (*Apodemus sylvaticus*) and yellow-necked mice (*A. flavicollis*) as reservoir hosts for TBEV. For this, wood mice and yellow-necked mice were trapped using life traps and inoculated with either a classical European subtype of TBEV or a more distinct related virus, TBEV-NL in a BSL3 laboratory.

Our findings showed that ticks from two distinct geographical populations differed in their susceptibility to TBEV strains, which could be the result of differences in the genetic background of the tick populations. Furthermore, experiments in rodents showed that all animals survived the challenge with TBEV and without clinical signs, yet, TBEV was detected up to 21 days in whole blood of both rodent species and TBEV did infect the brain of the mice from 3 to 21 days after infection. Our research provides a foundation for future studies to understand which factors affect the spread and distribution of TBEV in Europe.

The cellular and molecular components of the proboscis in the tiger mosquito *Aedes albopictus*

Irene ARNOLDI (irene.arnoldi@unimi.it), Marta VILLA, Simone PITTON, Claudio BANDI, Sara EPIS, Paolo GABRIELI

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Introduction and Objectives

Female mosquitoes are annoying ectoparasites of humans and animals that can transmit numerous pathogens. Their vectorial capacity and their extreme reproductive success reside in the ability of females to acquire blood from vertebrates, which is mostly needed to complete the oogenesis. The efficiency of this process is guaranteed by the mouth apparatus of the mosquitoes, the proboscis, which is perfectly adapted to accomplish its function. Many studies have been performed on this organ to unravel its anatomy and to study its function. Despite these efforts, the exact 3D anatomy of the proboscis and its cellular and molecular components are largely elusive. To gain a better understanding of these features, we are studying the proboscis of females of the tiger mosquito *Aedes albopictus*. This mosquito is an invasive species in Europe that can sustain local and autochthonous transmission of arboviruses, such as dengue and chikungunya. A better knowledge of how the mosquito proboscis works would help identifying which clues the mosquitoes use to find blood in the animal skin.

Materials and Methods

We are integrating morphological and molecular studies to obtain a clear picture of the anatomy of the mosquito proboscis. We used classical electron microscopic studies (SEM and TEM), array tomography, X-ray nanotomography and FIB-SEM to study the anatomy of the proboscis. Spatial transcriptomic has been attempted to study the gene expression of the cells in the proboscis.

Results, Discussion and Conclusion

The integration of multiple techniques allowed us to have a clearer picture of the 3D anatomy of the mosquito proboscis. The combination of this data with the molecular data allowed us to provide new insights into the function of this organ, which is an efficient microneedle that plays a central role in disease transmission.



S2-4

The genetic bases of deltamethrin resistance in *Aedes albopictus* - A genomic travelling around the world

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Pyrethroid insecticides remain largely used against *Aedes* mosquitoes to limit arboviral transmission. Such massive use of insecticides led to the emergence and the current spread of resistance in *Ae. albopictus*. Until alternative vector control tools or arboviral vaccines are widely implemented, understanding the mechanisms of resistance, identifying the involved genes and developing robust molecular markers is critical for improving surveillance and resistance management. We thus aimed at identifying the top resistance genes involved in deltamethrin resistance in *Ae. albopictus* in each studied region.

This study focuses on 5 regions: southern Europe, the Indian Ocean, middle-Western Africa and South-East Asia. To approximate the genetic diversity of each studied region, one composite population per region was constituted by assembling several field populations. Those composite populations were selected or not by deltamethrin for several generations. Genomics and transcriptomics approaches were then combined to identify resistance alleles.

Distinct deltamethrin resistance mechanisms were identified in each region, including target site mutations but also detoxification and cuticular resistance. The contrasted mechanisms observed across regions supports a dynamic landscape of resistance in *Ae. albopictus* worldwide and the need to expand the panel of molecular markers available for surveillance. From an applied perspective, such study paves the way for defining a set of molecular markers to track deltamethrin resistance in the field.

The seasonal thermal ecophysiology and egg metabolome point to different overwintering strategies in two invasive *Aedes* mosquito species

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The Asian tiger mosquito *Aedes albopictus* being carrier of dengue and chikungunya viruses and the Asian bush mosquito *Ae. japonicus* known as vector of Japanese Encephalitis and West Nile viruses are currently invading the (cold-)temperate areas of Europe. The fact, that *Ae. japonicus* has its origin in more temperate areas and *Ae. albopictus* originates in sub(tropical) regions suggests that their thermal ecophysiology and specifically their thermal stress tolerance and metabolic strategies to overwinter are different.

Here, we test the hypothesis that both species contrast in their seasonal ecophysiological strategies: We monthly collected *Aedes* eggs (*Ae. albopictus*: Italy; *Ae. japonicus*: Germany) and measured their seasonal metabolism using liquid chromatography and mass spectrometry (LC/MS-MS). We experimentally tested the cold tolerance (-2°C, -6°C) of the most cold-resistant life stage of *Aedes* species, the egg stage, for potential differences between species and during the mosquito season.

Through this comparative and high-resolution characterization of the metabolic and low-temperature phenotypes of the two *Aedes* species, different seasonal adaptation mechanisms could be identified. Metabolite content of *Ae. albopictus* appears to deplete during the onset of diapause while *Ae. japonicus* eggs generally retain metabolite levels during the mosquito season. Stable isotope-resolved metabolic profiling further reveals species-specific signatures in hatching-implicated pathways, such as fatty acid β -oxidation. Both, *Ae. albopictus* and *Ae. japonicus* show a decrease in egg hatching success in August indicating the onset of diapause. *Aedes japonicus* eggs show increased stress tolerance upon cold exposure in July, whereas *Ae. albopictus* show increased stress tolerance upon cold exposure in October.

The analysis of the metabolic and low-temperature features of the eggs of the two *Aedes* species allowed biochemical insights into an ontogenetically early development phase, but also into a stress-resistant mosquito life stage. We can conclude that the two *Aedes* species adopt different survival strategies under challenging environmental conditions.

Unlocking *Aedes albopictus* Expansion: Integrating Microhabitat Climate in Species Distribution Models

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Global climate change is significantly altering the landscape of mosquito-borne diseases, posing unprecedented risks to human populations worldwide. The expansion of *Aedes albopictus* into temperate regions has intensified the threat of pathogens such as Dengue and Chikungunya, necessitating a deeper understanding of the mechanisms driving its proliferation. Insulative oviposition sites, such as microhabitats, are crucial overwintering sites for *Ae. albopictus* in temperate regions. Microhabitats such as tires exhibit significantly warmer and more stable climates than the surrounding environment, impacting mosquito survival and distribution. Our previous work shows that temperate *Ae. albopictus* survive harsh winters in microhabitats specifically, tires and in particular snow-covered tires. Despite the known importance of these microhabitats, it has proven difficult to apply the presence and effects of microhabitats in regional, national, or global efforts to predict the distribution of *Ae. albopictus*. Consequently, many prior species distribution model (SDM) efforts have yet to consider microhabitats across a large geographic region, such as the Midwest, U.S.A. Here, we integrated microhabitat climate regression models derived from our previous work in the Midwest with traditional environmental climate data to develop SDM for *Ae. albopictus* that consider the environmental conditions within common microhabitats. Using Maximum entropy species distribution modeling, we evaluate the performance and predictive capabilities of SDMs when considering both environmental and microhabitat conditions. Incorporating microhabitat variables leads to notable shifts in predicted habitat suitability. Models incorporating microhabitats predict increased suitability at higher latitudes, reflecting the expanding range of *Ae. albopictus* observed in recent years. By comparing models' predictions with and without microhabitat data, this study highlights the importance of considering fine-scale ecological factors in SDMs for a complex understanding of distribution. Our findings underscore the need for further research to improve the representation of microhabitats in SDMs, particularly regarding the impact of climate change on mosquito-borne disease dynamics.



Resting site selection and microclimate of adult mosquitoes along a land use gradient

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Most mosquitoes spend a huge part of the day inactive in resting sites, which are used e.g. as refuge from unfavourable weather conditions or predators. Therefore, at least part of the blood meal digestion and development of pathogens also takes place in these sites. Understanding the microclimatic conditions therefore is highly relevant, as there is a direct impact on the extrinsic incubation period.

We selected 45 sites along three land use classes: forest, grassland, and urban. Data loggers were installed to hourly measure temperature and light incidence data. Samples were taken bi-weekly using a manual aspirator from June to September 2023, and thermal images were taken of the surrounding environment. The abundance of mosquitoes found in each sampling event was analysed as a function of land use together with mean temperature, the average of the 10th percentile temperature values, and the average of the highest 10% values in each interval between sampling events. The same was applied to light incidence.

In total 1,039 mosquitoes were collected from 43 resting sites, with a significantly higher abundance in forest areas (714) compared to grassland (168) and urban (157) areas. The abundance was analysed as a function of the microclimatic variables, the best predictors was the average of the highest 10% temperature values as well as the mean light incidence. Our results confirm that adult mosquitoes show a preference for resting sites with lower temperatures and lower light incidence compared to their surrounding areas.

With a focus on epidemiologically relevant *Culex* species, we provide high-resolution data on the resting behaviour and microclimate conditions, and a basis for improving predictive models of the spatio-temporal distribution of mosquitoes, e.g. to plan targeted interventions

Toc-toc: who is inside? Uncovering indoor host-seeking mosquito dynamics by Citizen Science

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Introduction and Objectives

Mosquito sampling in Europe is primarily implemented outdoors, thus providing limited knowledge on the exploitation of indoor habitats, which is assumed to be strongly different in the two main arbovirus vectors *Culex pipiens* (known as endophagic) and *Aedes albopictus* (known as exophagic in its native range). Here, we analysed indoor and outdoor mosquito photographic records sent by citizen scientists involved in Mosquito Alert (MA) project in Italy, based on the assumption that mosquito photos were taken during the host-seeking activity.

Materials and Methods

Georeferenced records of mosquitoes from Italy (identified by MA experts) were analysed in relation to information provided by the citizens on whether the photos were taken indoors or outdoors. The dynamics of *Ae. albopictus* relative frequencies were compared to those of *Culex* spp. (herein considered as *Cx. pipiens*, due to the negligible presence of other species) by binomial regression models as a function of indoor/outdoor location.

Results, Discussion and Conclusion

From October 2020 (when MA was launched in Italy) to the end of 2023, a total of 1,791 and 5,076 mosquito photos were recorded outdoors and indoors, respectively; 88% of outdoor records and 51% of indoor records were of *Ae. albopictus*. Models show that the probability of receiving *Ae. albopictus* records outdoors is always higher than the probability of receiving *Cx. pipiens* records, consistently with known host-seeking behaviours of the species. Notably, the trend is the same when considering indoor records received from June to October, while in colder months *Cx. pipiens* records prevail. While the seasonal difference is in line with the species known seasonality, *Ae. albopictus* indoor records suggest an unexpectedly high indoor host-seeking behaviour. The implications of these results and the potential of citizen science in enhancing mosquito monitoring and public health preparedness will be discussed.

Diversity of the *Aedes aegypti* group in the Southwest Indian Ocean

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Aedes aegypti is the primary vector of the World's most harmful arboviruses causing human diseases. This mosquito has a worldwide distribution in the tropics and subtropics and has been expanding its distribution due to human transportation, as well as climate change. Populations from the Southwest Indian Ocean region are phylogenetically basal to all other *Ae. aegypti* and this region harbors more genetic/taxonomic diversity than any other part of the range of *Ae. aegypti* previously characterized. This study characterizes genetic population structure across the ancestral range of *Ae. aegypti*, placing these populations into the global context of the species by using an extensive worldwide genetic database generated throughout the last decade. We have generated and analyzed whole-genome-sequencing data from *Aedes aegypti* and sister species from Madagascar (17 populations), Mauritius (4 populations), La Reunión (2 populations) and Mayotte (2 populations); as well representatives of the two *Ae. aegypti* subspecies: *aegypti* (Aaa) and *formosus* (Aaf). Genetic structure in the Southwest Indian Ocean reflects the islands where samples were collected. Our analysis shows low differentiation within Madagascar, *Aedes mascarensis* from Mauritius, and an *Aedes aegypti*-like group from Mayotte, while significant genetic structure was detected within La Reunion. Analysis of the mitochondrial genome uncovered signatures of putative admixture between some of these populations, likely the result of re-introduction of populations across the region. We will discuss these findings and implications in the context of *Aedes aegypti* evolution and consequences for disease transmission.



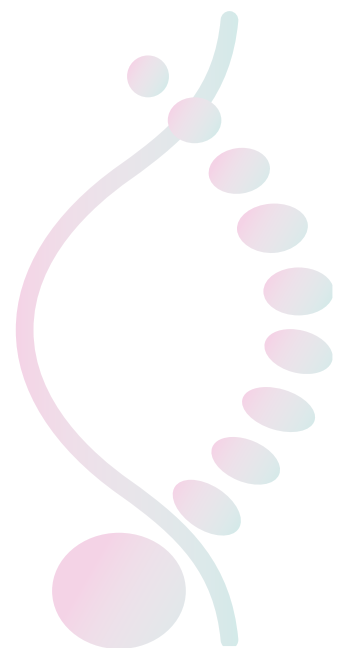
ONE HEALTH IN ACTION

14-17th October 2024
MONTPELLIER, FRANCE



ABSTRACTS TUESDAY 15 OCTOBER

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Impact of recent and future climate change on vector-borne diseases

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Climate change has direct impacts on human, animal and plant health through weather extremes such as droughts, heatwaves and floods. It has also indirect impacts on water-borne and vector-borne diseases and food security. Vector-borne diseases are primarily transmitted by hematophagous ectotherm arthropods that do not regulate their internal temperature. Temperature affects development stages of the insect vector, its biting rate, and the so-called extrinsic incubation period, which is the time, required for an arthropod to become infectious following a viraemic blood meal. Increase in temperatures can affect the distribution and seasonal activity of arthropods and the disease they transmit. As an example, arboviruses transmitted by *Aedes* mosquitoes have had significant impacts worldwide, with recently observed disease transmission in temperate regions. This presentation will review recent advances in our understanding of the impact of climate change on vector-borne diseases affecting humans and animals. Recent and future directions for this inter-disciplinary field of research will also be discussed.



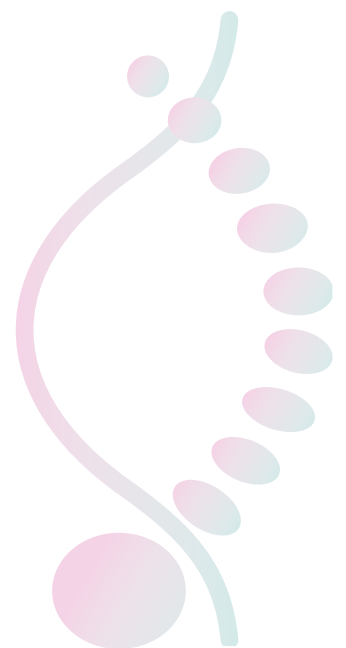
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Eco-epidemiology of VBDs under anthropogenic and climatic changes

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Predicting the current and future geographic distribution of *Lutzomyia* spp., vectors of *Leishmania* spp., under climate change, in North America

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Leishmania infantum and *L. braziliensis* are vector-borne zoonotic parasites, and the etiologic agents of leishmaniases in both dogs and humans. In South and Central America, species within the *Lutzomyia* genus drive *Leishmania* spp. transmission. While currently absent in Canada, *Lutzomyia longipalpis*, and *Lu. shannoni*, are of interest. *Lutzomyia longipalpis* is the primary vector of *L. infantum* across South, Central, and North America (i.e., Mexico), and *Lu. shannoni*, widely distributed across the United States, is a suspected vector, but its competency remains unproven. While dogs infected with *L. infantum* or *L. braziliensis* are imported into Canada on a regular basis, it is unknown if climatic changes are facilitating *Lu. longipalpis* and *Lu. shannoni* range expansion.

Objectives: (1) Determine current suitable area across North America for *Lu. longipalpis* and *Lu. shannoni*, (2) the potential for range expansion of *Lu. longipalpis* and *Lu. shannoni*, into Canada, and (3) identify climatic and land use factors impacting habitat suitability and range expansion.

Sandfly occurrence records were obtained from the Global Biodiversity Information Facility and the Disease Vectors Database. Environmental variables were obtained from ClimateNA (<https://climatena.ca>). Historic climate data (1981 – 2010) and projection data (2041 – 2070) were obtained to predict current and future distribution of sandflies. Ecological niche modelling (ENM), using MaxEnt algorithms, were used to predict the current distribution of northernmost *Lutzomyia* spp. A projection layer was applied to identify areas in North America that may become environmentally suitable for these sandflies. Results provide information regarding current potential distribution of *Lu. longipalpis* and *Lu. shannoni*, along with the potential distribution of these sandflies in the future. Climatic and land use variables that are associated with current distribution will be determined. These findings identify current suitable areas for *Lu. longipalpis* and *Lu. shannoni*, along with future, non-endemic regions where these sandflies may become established.

The rising global economic costs of invasive *Aedes* mosquitoes and *Aedes*-borne diseases

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Invasive *Aedes aegypti* and *Aedes albopictus* mosquitoes transmit viruses such as dengue, chikungunya and Zika, posing a huge public health burden as well as having a less well understood economic impact. We present a comprehensive, global-scale synthesis of studies reporting these economic costs, spanning 166 countries and territories over 45 years. The minimum cumulative reported cost estimate expressed in 2022 US\$ was 94.7 billion, although this figure reflects considerable underreporting and underestimation. The analysis suggests a 14-fold increase in costs, with an average annual expenditure of US\$ 3.1 billion, and a maximum of US\$ 20.3 billion in 2013. Damage and losses were an order of magnitude higher than investment in management, with only a modest portion allocated to prevention. Effective control measures are urgently needed to safeguard global health and well-being, and to reduce the economic burden on human societies. This study fills a critical gap by addressing the increasing economic costs of *Aedes* and *Aedes*-borne diseases and offers insights to inform evidence-based policy.

One Health innovative sampling to understand the ecology and spread of Usutu virus and West Nile virus in the Netherlands, 2016-2022

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Outbreaks of emerging arbovirus infections are increasing worldwide. In Europe, several arboviruses show a growing geographical spread and an increase in cases in recent decades. Extensive surveillance of mosquitoes, live wild birds, dead birds and chickens was therefore set up in 2016 to monitor the introduction and spread of a selection of high-risk arboviruses in the Netherlands. In addition, the risk of arbovirus infections for humans and predators in close contact with wild birds, was investigated. Mortality in captive and wild birds and wild carnivores is reported through a citizen science-based alerting system. Live wild birds are sampled (throat swab, cloaca swab and serum) in collaboration with bird ringers. A network of mosquito trapping sites was set up in July 2020, linked to bird ringing locations. Chickens from petting zoos around WNV outbreak locations were sampled in 2021, as well as bird ringers in the whole country. Samples were tested for USUV and WNV RNA using RT-PCR and full genome sequencing. Flavivirus antibodies were detected using protein microarray and FRNT. Endemic Usutu virus circulation was first detected in 2016, with continued enzootic presence of USUV lineage Africa 3 in subsequent years. Indeed, USUV lineage Africa 3 was found during winter, in live wild birds and hibernating mosquitoes. Multiple USUV infections in blood donors, bird ringers and wild carnivores, show that USUV also infects mammals in the Netherlands. Local West Nile virus (WNV) was first detected in 2020 in passerine birds, mosquitoes and humans. In 2021, seroconversions in petting zoo chickens showed continued WNV circulation. This was confirmed in 2022, when WNV was detected in a Grey Heron.

We show that a One Health framework for research and surveillance of WNV and USUV can serve as early warning for human infections, and provides deeper insights in flavivirus ecology and spread.

Modeling mosquito thermal performance and West Nile virus transmission suitability with Bayesian hierarchical models

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Introduction

The expansion of West Nile virus (WNV) in Europe, exacerbated by climate change, represents a major concern for human and animal health. WNV circulates between birds and *Culex* mosquitoes and can spillover into humans potentially resulting in severe disease. We provide an extensive analysis of experiments investigating the response of mosquito-pathogen traits to temperature. Focusing on *Culex* mosquitoes and their ability to transmit WNV, we investigate the generality of their temperature response and apply mathematical models to track temperature-driven changes in suitability for mosquito populations and disease transmission.

Methods

Through a literature review we update a dataset of experiments on mosquitoes acting as important disease vectors in temperate regions. Bayesian hierarchical models, accounting for differences between species and experiments, serve to fit thermal response functions for each mosquito-pathogen trait. The derived thermal response functions are incorporated into models of mosquito population dynamics and transmission suitability. The resulting models are applied to monitor changes in WNV transmission suitability across Europe.

Results

We find that temperature response estimates can vary widely between experiments on the same species but find striking similarities between the expected temperature responses of each species. Based on our models we show that WNV transmission suitability peaks around 25°C and aligns with current disease patterns in Europe. We find that the annual average temperature suitability of WNV transmission in Europe increased by 36.6% from 1951–60 to 2013–22.

Discussion

Our analysis enhances the knowledge in mosquito thermal biology. Models parameterized by our trait thermal performance estimates offer vital tools to monitor climate-driven disease risk changes. The increasing trend of WNV suitability in Europe underlines the importance of climate change mitigation and adaptation. Future experiments should focus on traits currently lacking extensive data and should investigate potential discrepancies between laboratory and field estimates to enhance the robustness of our approach.

Molecular Xenomonitoring (MX) allows real-time surveillance of West Nile and Usutu virus in mosquito populations

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West Nile (WNV) and Usutu (USUV) virus are vector-borne flaviviruses causing neuroinvasive infections in both humans and animals. Entomological surveillance is a method of choice for identifying virus circulation ahead of the first human and animal cases, but performing molecular screening of vectors is expensive, and time-consuming.

We implemented the MX (Molecular Xenomonitoring) strategy for the detection of WNV and USUV circulation in mosquito populations in rural and urban areas in Nouvelle-Aquitaine region (France) between July and August 2023, using modified BG Sentinel traps. We first performed molecular screening and sequencing on excreta from trapped mosquitoes before confirming the results by detecting, sequencing and isolating viruses from individual mosquitoes.

We identified WNV and USUV-infected mosquitoes in 3 different areas, concurrently with the first human cases reported in the region. Trapped mosquito excreta revealed substantial virus co-circulation (75% of traps had PCR+ excreta for at least one of both viruses). *Cx. pipiens* was the most common species infected by both WNV and USUV. Genomic data from excreta and mosquitoes showed the circulation of WNV lineage 2 and USUV lineage Africa 3, both phylogenetically close to strains that circulated in Europe in recent years. Four WNV and 3 USUV strains were isolated from trapped mosquitoes.

Interpretation

MX strategy is easy and rapid to implement on the field, and has proven its effectiveness in detecting WNV and USUV circulation in local mosquito populations.

Alien lymnaeid snails in the Western Mediterranean basin: new invaders and trematode-transmitters in the region?

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Biological invasions have the potential to foster the emergence of diseases, either through geographic spread or adaptive evolution. In the Western Mediterranean basin, two non-native snails have been introduced: the American *Pseudosuccinea columella*, to Aquitania and Corsica (France), and the Asian *Orientogalba viridis*, to Spanish Catalunya. Both play significant roles in transmitting the liver fluke *Fasciola hepatica* (Trematoda) elsewhere. This zoonotic emerging parasite circulates endemically in Europe within livestock, facilitated by a single vector species, the local snail *Galba truncatula*. Here, we aim to address four crucial questions: (i) Have the introduced exotic snails established and spread throughout the region? (ii) What are their ecological patterns in comparison to the local vector snail? (iii) What is their population genetic structure, and what is their history of invasion? (iv) Are they compatible with and capable of transmitting local parasites? Spatial surveys conducted within the Catalunya/Occitania region, alongside field ecology, were complemented with COI sequencing, microsatellite genotyping, and trematode metabarcoding analyses. Furthermore, laboratory populations were established to evaluate experimental compatibility with local parasites. The successful establishment of a COI haplotype of *O. viridis*, closely related to a Malaysian haplotype, is patented within the Ebro delta. Of particular concern is the spread of the globally invasive genotype of *P. columella*, known for its high compatibility with *F. hepatica*, in Occitania. This spread is primarily associated with hydrographic systems connected to the Canal Latéral de la Garonne/Canal du Midi. However, multiple introduction events may have occurred, as populations of these species are also documented in distant and non-connected artificial habitats, suggesting the existence of high propagule pressures. Both species exhibited high compatibility with local isolates of *F. hepatica* (70-100%). This comprehensive study provides a thorough overview of the invasion history and the associated risks of parasite transmission in the region driven by these exotic snails.

Lyme Borreliosis in Bonn, Germany under an integrated approach of One Health and Socio-Ecology

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Lyme Borreliosis is a common vector-borne disease in Europe transmitted by *Ixodes ricinus* ticks. While the disease is generally associated to rural forested environments, urban green spaces are potential tick habitats, posing a risk of disease transmission among urban residents. The present study assesses disease transmission dynamics using an integrated approach of One Health and Socio-Ecology. Therefore, a systematic tick sampling was conducted over a one-year period to determine tick abundance and *Borrelia s.l.c.* prevalence in ticks in urban and peri-urban green spaces in Bonn, Germany. The species and stage of the ticks sampled were determined by microscopy, and pathogen prevalence was assessed by PCR analysis. Additionally, an in-field survey among green space users was conducted to evaluate their tick-borne disease knowledge, preventive behavior and risk perception. The highest abundance was observed in the peri-urban forest and the lowest in the urban park. However, *Borrelia s.l.c.* prevalence ranged from 21,79% to 24,42% in the urban forests compared to an average *Borrelia s.l.c.* prevalence of 18,15% in the peri-urban forest. Despite lower tick abundance in urban green areas, findings of inadequate preventive measures and awareness among green space users may result in human health risks. Among the study participants, 26,2% reported being bitten by a tick within the past 12 months and 10,98% reported being diagnosed with a tick-borne disease in the past. By embracing a holistic and an integrated approach, this study enhances our understanding of urban local risk factors associated with Lyme Borreliosis transmission and underscores the importance of integrated surveillance and prevention strategies.

Green cities and the risk for vector-borne diseases transmission

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Introduction and Objectives

Greening cities represents a nature-based strategy aimed at fostering sustainable urban development by mitigating climate change effects and improving human health and well-being. However, integrating nature to urban environments can influence the distribution and the abundance of disease vectors, potentially impacting the emergence and transmission of vector-borne diseases (VBDs). Urban green infrastructures may expand the overall habitat capacity by providing suitable breeding and resting sites for urban vectors. These green areas could also enhance opportunities for managing VBD risks within cities, by facilitating predation, competition, and dilution effects in the urban ecosystem.

To begin unravelling the complexities of how urban greening influences VBD transmission in urban settings, we conducted a scoping review.

Materials and Methods

We implemented a systematic search following the PRISMA guidelines, identifying 110 articles investigating the interplay between urban green infrastructure and the risk of VBDs affecting human, animal, and plant health. We summarized identified impact by vector, and describe methodologies used to characterize urban green infrastructure and VBD risk.

Results

The majority of the articles addressed mosquito-borne diseases (75) and tick-borne diseases (29). Overall, urban vegetation appears to heighten the risk of tick-borne diseases. The influence of urban green infrastructure on mosquito-borne diseases varied depending on the specific vector system and environmental context. Additionally, we observed a wide diversity of indicators used to assess both VBD risk and urban green infrastructure. Depending on the selected indicator, the impact on the VBD risk can differ.

Discussion and Conclusion

Considering the growing interest in urban greening, it is crucial to investigate its potential implications for public health. However, the absence of standardized protocols hinders accurate interpretation of the effects of urban green infrastructure on the risk for VBD emergence and transmission. Our analysis highlights gaps in knowledge and identifies opportunities and priorities for research.

Phenotypic plasticity in vector traits drives trends in global disease incidence: *Aedes albopictus* and dengue

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The incidence of vector-borne disease is on the rise globally, with burdens increasing in endemic countries and outbreaks occurring in new locations. Effective mitigation and intervention strategies require models that accurately predict both spatial and temporal changes in disease dynamics, but this remains challenging due to the complex and interactive relationships between environmental variation and the vector traits that govern the transmission of vector-borne diseases. Predictions of disease risk in the literature typically assume that vector traits vary instantaneously and independently of population density, and therefore fail to capture the delayed response of these same traits to past biotic and abiotic environments. We argue here that to produce accurate predictions of disease risk it is necessary to account for environmentally driven and delayed instances of phenotypic plasticity. To show this, we develop a novel stage and phenotypically structured model for the invasive mosquito vector, *Aedes albopictus*, and dengue, the second most prevalent human vector-borne disease worldwide. We find that environmental variation drives a dynamic phenotypic structure in the mosquito population, which accurately predicts global patterns of mosquito trait-abundance dynamics. In turn, this interacts with disease transmission to capture historic dengue outbreaks. By comparing the model to a suite of simpler models, we reveal that it is the delayed phenotypic structure that is critical for accurate prediction. Consequently, the incorporation of vector trait relationships into transmission models is critical to improvement of early warning systems that inform mitigation and control strategies.

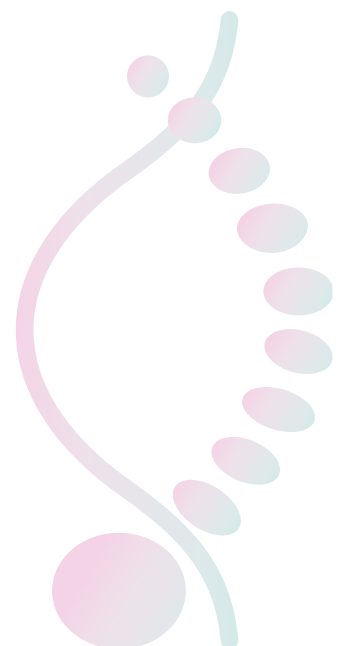


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Interactions between
host(s)-vector(s)-pathogen(s)-microbiota-environment



West Nile virus-related activity alterations and neuronal infection in *Culex pipiens*

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Mosquito-borne pathogens transmission is dependent on mosquitoes' behavior. Arthropod-borne viruses (arboviruses) may target mosquito neuronal tissues, potentially impacting a range of behavioral traits along the blood-feeding trajectory and subsequently influencing their transmission. Little research investigated the influence of arbovirus infection on mosquito behavior. Our study aimed to determine how West Nile virus (WNV) targets *Culex pipiens*' neuronal tissues and induces changes in its activity. Female mosquito *Culex pipiens* ecotype *pipiens* were intrathoracically inoculated with WNV lineage 1 and 2 ($7 \log_{10}$ TCID₅₀/mL) or with cell culture media as a control group. Infected females from both lineages were tested for WNV presence via immunohistochemistry at 1, 3, 5, 7, 10, and 14 days post-infection (dpi). Non-infected females were analyzed at 14 dpi. Mosquito bodies and saliva were also collected at each time point to analyze viral replication and transmission, respectively. To monitor the mosquito activity, female mosquitoes (WNV-1, WNV-2, controls; n=30/group) were kept in three individual plexiglass cages and tracked using a camera with infrared capture capacity for 14 days. Images were processed using a custom-developed code to monitor mosquitos' position variations throughout the captured frames. WNV neurotropism was confirmed by positive staining in the central nervous system (head, thoracic, and abdominal ganglia) and peripheral sensory organs (ommatidia and Johnston's organ). The presence of WNV in the mosquito nervous system indicates potential alterations in neuronal communication, also associated to the changes in activity observed. Salivary glands were also positive for the stain and viral titration. Our preliminary results reveal activity modifications caused by WNV infection in *Culex pipiens* f. *pipiens*. These behavioral changes may have meaningful consequences for virus transmission. However, further research is needed to understand the impact of mosquito behavior on WNV transmission dynamics.

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Understanding factors shaping mosquito microbiome assembly using gnotobiotic approach

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The holometabolous life cycle of mosquitoes contributes to the variation in the microbiome across different life stages. However, it remains unclear how different factors influence the colonization of the microbiome in mosquitoes. Our limited knowledge in this regard was mainly due to the inability to manipulate the mosquito microbiome. The gnotobiotic rearing of mosquitoes allows us to manipulate the microbiome and create known communities. This approach is useful to understand factors such as host and microbial genetic factors, larval environment, and host-microbe and inter-microbial interactions. Utilizing this we have shown that surface-associated protein *ompA* of symbiotic bacteria facilitates the colonization and biofilm formation in the mosquito gut. Inter-species microbiome transplantation between diverse mosquito species revealed interactions between microbiome and host factors. Our results show that the variation in mosquito microbiome communities is associated with variability in host-microbiome interactions. Mono-association (MA) infection of *Aedes aegypti* mosquitoes has shown that the inter-microbial interactions shape the gut microbiome. Here, we have shown that bacteria-bacteria and interkingdom interactions influence the microbiome composition. Using MA infection, we were able to investigate interactions of gut bacteria with arboviruses at the strain level. Further, we have used the gnotobiotic approach to assess the role of microbial density in larval in shaping microbiome stochasticity in larval and adult gut. The higher density of bacteria in the larval water contributes to the lower randomness and vice versa. Hence, our results show that microbial density in the larval water contributes to the microbiome variability. Altogether, these results show that we can use a gnotobiotic approach to investigate the microbiome assembly and host-microbes interactions in mosquitoes. Further, we can expand these observations to investigate mechanisms at the molecular level to gain deeper insights into microbiome and mosquito interactions.

Analyzing the Complexity: interactions of arboviruses and insect-specific viruses in vector competence studies with two invasive *Aedes* Species

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The global spread of invasive mosquito species increases the risk of arbovirus infections in humans and livestock. Besides *Aedes albopictus*; *Aedes koreicus* and *Aedes japonicus* have recently spread within Central Europe. Extensive information on the vector competence of the latter two species is missing. Therefore, we evaluated the vector competence of field caught *Ae. koreicus* and *Ae. japonicus* mosquitoes from Germany for different arboviruses. Additionally, the virome of the examined mosquitoes was investigated, and the impact of co-infection with arbovirus and insect-specific virus (ISV) on vector competence was evaluated.

Mosquitoes were orally infected and incubated under different temperature conditions ($27 \pm 5^\circ\text{C}$; $24 \pm 5^\circ\text{C}$, $21 \pm 5^\circ\text{C}$), to evaluate the influence of temperature on transmission dynamics. A salivation assay was performed to investigate the vector competence. Using a whole virome analysis via NGS, novel insect-specific viruses (ISVs) were identified.

Ae. koreicus had the potential to transmit Chikungunya virus (CHIKV) and Zika virus (ZIKV) but not West Nile virus (WNV). Transmission was only observed at the higher temperature with low transmission efficiency. *Ae. japonicus* was able to transmit CHIKV, Sindbis virus (SINV) and western equine encephalitis virus (WEEV). The risk of CHIKV transmission by *Ae. japonicus* was also rather low and exclusively observed at the higher temperature. Transmission of SINV and WEEV was observed at all investigated temperature profiles with high transmission efficiency.

While we detected only one ISV in *Ae. koreicus*, we could identify seven ISVs in *Ae. japonicus*. Correlating the ISV infection status of individual specimens with their transmission capability for specific arboviruses revealed a potential interference of ISVs on arbovirus infection, characterized by highly complex dynamics.

Impacts of insecticide resistance on Usutu virus transmission and associated life-history parameters in *Culex quinquefasciatus* mosquitoes

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Mosquitoes of the *Culex pipiens* complex are known for their role as vectors for many arboviruses. Among these, Usutu virus (USUV) is an emerging pathogen in Europe that impacts bird species as well as other animal such as humans. To control the spread of arboviruses, vector control is the only affordable way in absence of treatments. But the extensive use of insecticides led to select several resistant alleles that have invaded natural populations. The objective of our study is to determine the potential impact of insecticide resistance on arbovirus transmission and on vector-arbovirus interaction. We investigated the influence of several insecticide resistance alleles on vector competence for USUV and the impact of both resistance and infection on survival and fecundity of *Cx. quinquefasciatus*. We performed experimental infections of four *Cx. quinquefasciatus* colonies (one susceptible and three resistant) with USUV EU3 lineage. These colonies differ by the genomic architecture around *ace-1* locus conferring resistance to organophosphate but share a common genetic background after introgression with the genome of the susceptible colony. We then used an RNAseq approach to analyze the key genomic components involved in vector competence.

The results showed specific effects of genotype on vector competence and life history traits of infected mosquitoes. The susceptible colony was significantly more competent than the three resistant colonies. Survival and oviposition of females varied between the 4 strains. Interaction was found between resistant alleles and USUV infection in these parameters for some of our colonies.

Based on these phenotypic results, our experimental design allowed us to conduct a transcriptomic analysis to explore the differential gene expression between mosquitoes colonies and competence status, i.e. competent individuals, non-competent and controls groups. This study helped characterizing the genetic factors involved in transmission and virulence and improves our understanding of vector-pathogens interactions.

Infection dynamics and impact of blood feeding on the susceptibility of *Culicoides* biting midges to bluetongue virus (BTV)

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Culicoides biting midges are biological vectors of several arboviruses of veterinary importance, including bluetongue virus (BTV). Bluetongue virus causes bluetongue disease in domestic and wild ruminants leading to significant morbidity and mortality. We investigated the infection and dissemination dynamics of BTV in its insect host and identified key behavioural factors that may affect the vector competence of *Culicoides* biting midges. We examined the temporal-spatial dynamics of BTV infection in different *Culicoides* tissues in unprecedented detail. Novel insights into the distribution of viral infection in the midgut were revealed through the use of RT-qPCR in parallel with 3D immunofluorescence microscopy. BTV infection occurred in discrete foci of midgut cells that could be observed as early as 2 days post-infection (dpi), a finding that has previously been obscured by the presence of BTV in the remains of the blood meal at this time point. Additionally, midgut infection was not spatially restricted by region and replication continues in the midgut after dissemination. Furthermore, experimental infections demonstrated the enhancement of BTV replication, infection and dissemination following acquisition of a subsequent non-infectious bloodmeal. The statistical significance of this effect was dependent on both *Culicoides* species and BTV serotype used. Investigation of the mechanism underlying the increased infection following blood feeding was impeded by the susceptibility of *Culicoides* midgut cells to infection from the abluminal side, even in the absence of blood feeding.

Identification of receptors of the plant virus Turnip mosaic virus (*Potyviridae*) in their aphid vectors

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Hundreds of plant viruses are transmitted by aphid vectors, among which non-circulative ones are acquired and inoculated from one host to another within seconds. These viruses are retained on receptors located at the surface of the cuticle of insect mouthparts. Members of the family *Potyviridae* are the most abundant RNA viruses infecting plants, and they cause significant economic losses. Because of their importance, they are widely studied. However, their receptors in aphid stylets are still poorly characterized. Identification of these molecules remains a major challenge for understanding the transmission mechanisms and would pave the way for alternative control strategies as potential targets to prevent vector-borne transmission. Using the *Turnip mosaic virus* (TuMV, *potyvirus*) as a model, we developed several approaches to characterize potyvirus-vector interactions. Using immunolocalization and transmission electron microscopy studies, we mapped the retention sites of TuMV at the apical part of aphid stylets in the vicinity of the acrostyle, a cuticular organ known to host and expose receptors on the stylet surface. In addition, peptide array assay revealed an interaction between the viral ligand of TuMV, the helper component proteinase (HC-pro), and cuticular peptides present at the stylet surface. We further characterize the role of cuticular proteins of the acrostyle in TuMV transmission, using RNA interference (RNAi) to reduce gene expression or aphid mutants generated via CRISPR-Cas9 technology. Altogether, our results lead to the identification of receptor candidates for TuMV.

The aphid factor: how aphid genetic diversity and the endosymbionts they carry may impact virus epidemiology

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Majority of plants viruses are vectored by insects such as aphids. Controlling aphid population is challenging due to rising insecticide resistance and legislation that limits use. Virus infection can alter the production of plant natural chemicals which can attract or repel aphids to feed on the infected plants and pick up viruses. Previously we found that infection of Cucumber mosaic virus (CMV) in the model plant, *Arabidopsis thaliana* deterred peach potato aphids (*Myzus persicae*) from prolonged feeding. Contrastingly, aphid fitness was enhanced on CMV-infected tobacco. Both outcomes support virus transmission by promoting vector dispersal and population growth respectively. However, it is not yet well understood how natural genetic variation of each 'partner' in this three-way plant-virus-vector interaction influences virus spread. Within an aphid species the asexual progeny from a single female aphid are typically defined as clones. Aphids also harbour symbiotic bacteria (endosymbionts), which can influence their fitness, pathogen resistance and virus vectoring efficiency amongst others. Here we explore if aphid genetic diversity and the bacterial endosymbionts they carry can influence the dynamics of plant-virus-vector interactions. Barley yellow dwarf virus (BYDV) is an aphid-vectored cereal pathogen causing significant economic loss worldwide. The two main BYDV vectors are the bird cherry oat aphid (*Rhopalosiphum padi*) and the grain aphid (*Sitobion avenae*). We collected *R. padi* and *S. avenae* aphids from different regions in the UK. Their genetic diversity was characterised using microsatellite genotyping. The presence of bacterial endosymbionts identified using PCR. Virus transmission experiments using BYDV-PAV (*R. padi*) and BYDV-MAV (*S. avenae*) strains on these aphid clones will be performed. We aim to determine if there is a link between aphid clones, endosymbiont profile, and efficiency to vector BYDV. Ultimately, the results will enable better prediction of BYDV outbreaks through improved monitoring and identification of aphid clones with varying vectoring capacities.

A flower pathway for arbovirus transmission? Unravelling sand fly - Phlebovirus interactions

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Vector-borne diseases represent one of the world's major public health challenges. Among these pathogens, arthropod-borne viruses (arboviruses) have become the most important cause of (re) emerging epidemic diseases worldwide. For many arboviruses, there are major unknowns regarding their transmission route and maintenance in nature. This knowledge gap is exemplified by phleboviruses, such as the Toscana virus (TOSV), which are mainly transmitted by sand flies and can cause meningitis or encephalitis. Despite decades of research, no reservoir host has been identified for TOSV, and based on the idea that the main transmission route must involve sand fly blood feeding, few alternative pathways have been explored. By investigating sand fly - Phlebovirus interactions experimentally, we have shown that infected sand flies spit TOSV into sugar while feeding. Furthermore, our study reveals that TOSV remains persistent and infectious for seven days in the sand fly sugar meal. Because most arbovirus vectors require a daily sugar meal, these results highlight a potentially novel horizontal transmission route operating through sand fly sugar feeding. These findings prompt further exploration of Phlebovirus transmission dynamics and emphasise the importance of environmental factors in their eco-epidemiology, particularly the hitherto unexplored role of plants.

The *Hyalomma marginatum* tick holobiont: composition, dynamics and interactions

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Ticks transmit pathogens of medical and veterinary importance and are an increasing threat to human and animal health. The invasive tick *Hyalomma marginatum* newly settled in France is the vector of several human pathogens including the deadly Crimean-Congo hemorrhagic fever virus. While few information about the *H. marginatum* holobiont are currently available, we characterized the composition, the spatio-temporal dynamics and the interactions of *H. marginatum* microbial communities from 792 ticks collected in 28 sites in Southern France. Different high-throughput sequencing approaches were used to identify the composition and the dynamics of both the *H. marginatum*-borne pathogens and microbiota (qPCR microfluidic and Illumina MiSeq). Interactions between all identified tick microbes were finally assessed by network analysis.

The *H. marginatum* microbiota was dominated by two endosymbionts, *Francisella*-like endosymbiont and *Candidatus Midichloria*. The spatio-temporal dynamics were mainly influenced by environmental bacteria. While the pathogen screening revealed that 11% of ticks were infected by two animal pathogen genera *Theileria* and *Anaplasma*, a very high infection rate was observed for the human pathogen *Rickettsia aeschlimannii* (87%) regardless the detection method. Both infection rate and bacterial loads were influenced by the spatial and temporal scales. Considered as a human pathogen, this maternally-transmitted bacterium is likely to be a tick symbiont with potential important functions. Beyond composition and dynamics, several microbial interactions have been highlighted, paving the way for the development of potential new strategies for the control of ticks and tick-borne diseases.



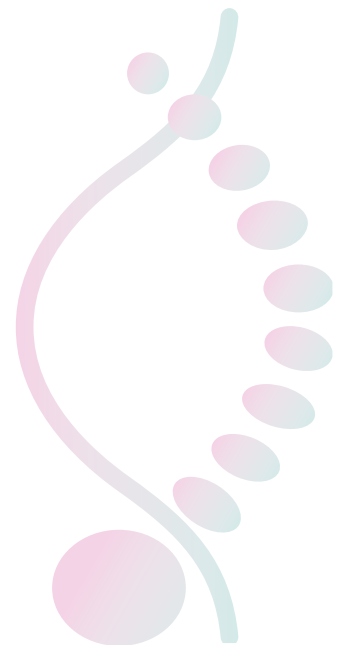
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ABSTRACTS WEDNESDAY 16 OCTOBER

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Snailed it: Unlocking the potential of Citizen Science to control and prevent snail-borne diseases

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Background

Citizen science is gaining popularity in many research domains, but it has never been applied in the field of snail-borne diseases. In this talk I will outline the challenges and opportunities of implementing this approach to monitor and prevent schistosomiasis in Africa. This disease affects over 200 million people, mainly in Africa. Its transmission is influenced by a complex interplay of behavioral practices and ecological factors, including the presence of snail hosts. Since drug treatment alone does not suffice to control the disease, the WHO recommends including snail control and community involvement. However, the shortage of malacologists and hence snail data, presents a major challenge design and implement targeted control measures.

Methods & Results

In the ATRAP project we established a network of 25 citizen scientists in each country dedicated to monitoring snail populations and risky water practices. We demonstrate that citizens can monitor snail presence and relative abundance in space and time with congruence of up to 80% with an expert scientist, but eight times cheaper. The resulting dataset is unprecedented in terms of resolution and coverage, and is used in modelling snail population dynamics at fine spatial-temporal scales. We also constructed risk maps maps incorporating both ecological and behavioural data, offering valuable insights for public health officials to design local control measures.

Discussion

Apart from data generation, citizen participation also empowers communities and increases knowledge of snail-borne diseases and their control and prevention. This way of doing research enables a two-way dialogue between citizens and scientists, exchanging local knowledge and scientific findings. We therefore argue that this community-based approach presents a very powerful and sustainable addition to classical surveillance programs. It also has a huge potential for upscaling to other other vector species and regions, thereby generating the much-anticipated data and community support for targeted control.



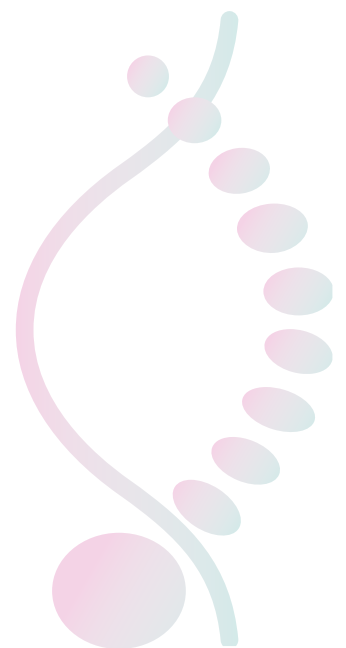
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Novel research avenues for innovative vector control strategies

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Monitoring individual rice field flooding dynamics over large scales to inform mosquito surveillance and control

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Progress in malaria elimination has been hindered by recent changes in mosquito behaviour and increases in insecticide resistance in response to traditional vector control measures, such as indoor residual spraying and long-lasting insecticide-treated nets. There is increasing interest in the use of larval source management (LSM) to supplement current insecticide-based interventions. However, LSM implementation requires the characterization of larval habitats at fine spatial and temporal resolutions to ensure interventions are well-placed and well-timed. The most compelling approach to achieve this high-resolution information is the application of remote sensing via the use of drones, which are limited in their spatial reach and temporal frequency, preventing their routine use at scale. Here, we propose a method to monitor flooding dynamics in individual rice fields, a primary larval habitat, over very large geographic areas relevant to national malaria control programs aiming to implement LSM at scale. We demonstrate this for a 3,971 km² malaria-endemic district in Madagascar with over 17,000 rice fields. We trained a classification model of surface reflectance on over 200 field observations from over 50 rice fields to produce time-series of bi-weekly flooding dynamics for thousands of rice fields, by combining rice field mapping on *OpenStreetMap* with Sentinel-1 satellite imagery (radar, 10m) from 2016 - 2022. From these time-series, we obtained key indicators of each rice field useful for LSM implementation such as the timing and frequency of flooding seasons. These monitoring tools were integrated into an interactive GIS dashboard for operational use by vector control programs, with results available at multiple scales (district, sub-district, rice field) relevant for different phases of LSM intervention (e.g. prioritization of sites, implementation, follow-up). Scale-up of these methods could enable wider implementation of evidence-based LSM interventions and reduce malaria burdens in contexts where irrigated agriculture is a major transmission driver.

Reducing densities of zoophagous *Anopheles* by treating cattle with an innovative formulation of Ivermectin while preserving the environment : a One-Health approach

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In rural Sudano-Sahelian zones, livestock live close to humans and are alternative blood sources for *Anopheles* vectors of Plasmodium, even the most anthropophilic. Thus, vectors remain at densities sustaining transmission of the parasite, despite massive coverage using conventional control tools. In Burkina Faso, we work on a holistic approach: treating livestock with ivermectin to render animal blood toxic to *Anopheles*, hence promoting both human and animal health. Yet, approved ivermectin formulations are of too short remanence to impact malaria incidence. In this context, Long-Acting Ivermectin Formulations (LAIFs) that deliver, upon a single injection, efficient ivermectin concentrations spanning the transmission season would represent substantial advantage, but would raise ecotoxicological concerns for already fragile agro-ecosystems.

In the Anivermate project, a LAIF candidate targeting more than 6 months efficacy was designed using the BEPO® technology (IVM-BEPO®) and administered at 2.4 mg/kg to local cattle. Efficacy against wild-derived *Anopheles coluzzii* was determined through survival experiments and compared with that of an already approved formulation injected monthly at 0.4 mg/kg. Data were used in a population model to predict effectiveness, according to *Anopheles* trophic preference, infectious status, and human-animal ratio in the villages. Plasma and fecal ivermectin concentrations were measured and risks for the environment explored (thorough bibliography, dissipation in feces and soil sorption experiments). A single injection of the IVM-BEPO® is mosquitocidal for over 6 months, with median *Anopheles* lifespan being 3-5 days. A single injection would reduce infectious *Anopheles* populations by 50-90% in all tested scenarios. By comparison, efficacy of a single injection of approved formulation lasts 14 days. Risks for agro-ecosystems are likely, notably through toxicity to coprophagic insects. However, local dissipation tools (fosses fumières) and agro-pastoral practices would be efficient mitigation tools. Hence, our integrative and holistic approach could reach sustainability, while providing efficient tool to combat malaria.

The use of *Wolbachia* for dengue control - New Caledonia as a case study

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This year, 2024, has been unprecedented for the size, intensity and geographic range of dengue epidemics occurring globally. Urgent solutions are needed beyond conventional control, which has shown to be largely ineffective, to reverse the current disease trend. One novel solution that is having dramatic impacts on reducing dengue transmission is the use of *Wolbachia*.

Wolbachia is one of the most common endosymbiont, partly due to efficient vertical transmission and manipulation of its host's reproduction. The stable introduction of *Wolbachia* into *Aedes aegypti*, showed that it can also limit the replication of human pathogenic viruses such as dengue in the mosquito. The past 13 years, the introduction of *Wolbachia* into *Aedes aegypti* population around the world showed that in areas where *Wolbachia* is self-sustaining at a high level, dengue transmission has been significantly reduced. *Wolbachia* is also safe, no adverse events have been reported so far.

A case study on its use is run in the French Territory of New Caledonia in collaboration with the government of New Caledonia, the cities, the South Province, Pasteur Institute New Caledonia, the French government, and the World Mosquito Program. I will describe the implementation of this innovative and sustainable approach to fight mosquito-borne diseases, the community acceptance and its current impact on dengue.

Development of *N*-arylamide insecticides to control hemipteran pests of agricultural systems

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Aphid-mediated transmission of plant viruses occurs during distinct feeding events, such as probing for non-persistent viruses or phloem feeding for persistent viruses. We have shown that initiation of aphid feeding, and thus transmission of non-persistent viruses, occurs between 4-6 minutes after landing on the plant. Thus, we aimed to develop novel chemicals capable of inducing rapid (<6 min) mortality or altering feeding behavior to reduce horizontal transmission of plant viruses. We screened a newly synthesized *N*-arylamide series for acute toxicity against two aphid pests, *Myzus persicae* and *Aphis gossypii*, through direct spray exposure and leafdip bioassays. Twelve molecules resulted in >90% mortality within 6 minutes of exposure at 200 ng/cm² and two molecules, *N*-(2,6-dichlorophenyl)-2,2,3,3,3-pentafluoropropanamide (GMR031) and *N*-(2,6-dichloro-4-(trifluoromethyl)phenyl)-2,2,3,3,3-pentafluoropropanamide (GMR138), resulted in >90% mortality at 3-4 minutes at 50 ng/cm². Electrical penetration graph (EPG) technique was used to quantify changes to feeding behavior and an 8-fold increase in time-to-first probe, 5-fold reduction of time spent in probes, and significantly ($P < 0.05$) reduced ingestion of xylem and phloem were observed after foliar application of GMR031 and GMR138. Based on the changes to feeding behavior, assays to quantify changes to horizontal transmission of potato leaf roll virus (PLRV; persistent) and sweet potato feathery mottle virus (SPFMV; non-persistent) after foliar treatment with *N*-arylamides were performed. Data indicate significant reduction or inhibition of aphid-mediated transmission of both, PLRV and SPFMV, after foliar application of *N*-arylamides. Structure-activity relationships, preliminary mode of toxicity, and implications of these findings for aphicide development efforts will be discussed.

Vectrap: a 3 years-field evaluation of mass-trapping strategy against *Aedes albopictus* and *Aedes aegypti* implemented in residential areas

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Within mosquito species of public health importance, *Aedes aegypti* and *Ae. albopictus* are among the most important due to their extensive areas of presence throughout the world and their ability to transmit major human arboviruses, as well as being a major source of nuisance. Apart from using insecticides against these vectors (and its disadvantages: resistance phenomenon, hard to reach breeding sites), the only method used to reduce their abundance is communication and social mobilisation. The VECTRAP project aimed to demonstrate the effectiveness, feasibility and sustainability of mass trapping against both species in 3 geographical areas (Mediterranean France, Alpine France, and the tropical island of Martinique) by using gravid *Aedes* trap (BG-GAT[®] trap from Biogents). During 3 years from 2021 to 2023, the effectiveness of 3 phases were assessed: Trapping only in year 1; Trapping and door-to-door breeding sites suppression in year 2; autonomy of the strategy (traps managed by inhabitants) in year 3. The entomological monitoring consisted in fortnightly captures using BG-Sentinel[®] traps in intervention and control areas, and a sociological survey was carried out focusing on the behaviour and perception of the residents from intervention and control areas. Our results tend to show that the use of gravid *Aedes* traps has an effect when this strategy is combined with door-to-door and control of breeding sites. In addition, a regular monitoring of the passive gravid traps seems essential to guarantee the trapping network efficiency. The ultimate goal of the project, implemented for 3 years, is to demonstrate the effectiveness of this mass trapping method and identify key factors that could maximise the adult mosquito population reduction, in order to provide recommendations to municipalities. Potential factors that could explain the observed efficiency variability among sites will be discussed.



S5-6

Exploiting sugar-feeding behaviour for the control of *Culicoides* biting midges

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Biting midges of the genus *Culicoides* are vectors of veterinary relevance, as they can transmit a plethora of pathogens such as bluetongue and African horse sickness disease virus. Novel control options may include push-pull systems to alleviate the impact of insect vectors upon their hosts. While spatial repellents have proven to be valid “push” candidates, we still lack an effective and selective attractant. A previous study revealed that the vast majority of field-collected biting midges are positive for sugars, an essential energy source for both female and male individuals. We collected biting midges from three field sites, and analyzed their sugar source by sequencing with two different primer pairs, to unravel recurring plant sugar sources. Over 100 clear DNA sequences were retrieved, and the most recurring plant species were *Solanum tuberosum* (potato), *Citrus spp.* (orange, lemon or mandarin), *Glycine max* (soya bean), and *Ficus carica* (fig). The attractiveness of these plants is being verified with behavioural bioassays in Y-tubes and large-cage setups and plant volatiles will be analysed with GC-MS. The attractants to be developed can be employed to bait traps, and alleviate the biting pressure exerted by *Culicoides*.

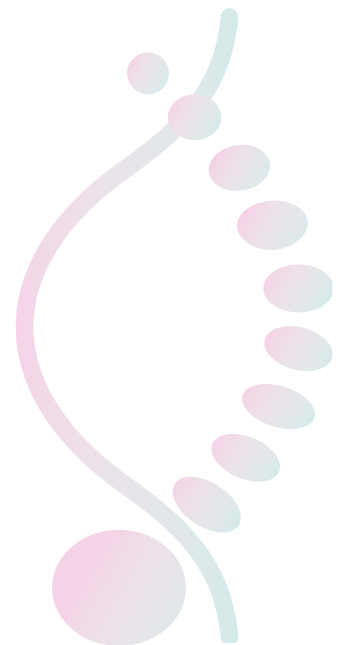


ONE HEALTH IN ACTION

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Networks and projects: What's new?



VectorNet3: The third iteration of the European network for medical and veterinary entomology

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The VectorNet network is co-funded by the European Food Safety authority (EFSA) and European Centre for Disease Control (ECDC). The network got its start in 2014 with the first iteration of VectorNet build upon the network started in the VBORNET project. The network has now entered its third iteration. In the project we cover four different vector groups: mosquitoes, ticks, biting midges and sandflies. The network comprises of the consortium, in charge of executing the project and the One-Health VectorNet Entomological Network (OHVEN), comprised of medical entomologist, public health professionals and animal health professionals. The aim of this OHVEN is to create efficient exchange of information and strengthen the collaboration between the stakeholders to support the prevention of vector-borne diseases (VBDs) in the EU and its surrounding countries.

In this third iteration of VectorNet we will continue the previous work such as providing scientific advise to ECDC and EFSA, maintaining and updating of the database for the different priority species and expand on the work conducted in the previous iteration of VectorNet. In order to make the database more accessible we will make extracts of the VectorNet database easy obtainable from public databases such as the Global Biodiversity Information Facility (GBIF). To further improve the usability of the VectorNet output maps we will add interactivity to the maps. The gaps in the current database will be analysed and the results of this analysis will be used to design specific field sampling campaigns. In this new iteration of the project we also aim to investigate the cost-effectiveness of vector control methods in a one-health setting via clustered randomized controlled trials.

MEDILABSECURE A One Health Network to improve preparedness and response capacities to emerging vector-borne diseases increased by environmental and climate change

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The geographical distribution and incidence of vector-borne diseases is increasing favoured by environmental degradations and climate change. The implementation of an integrated surveillance is of paramount importance to efficiently prevent vector-borne diseases. MediLabSecure is a capacity-building project funded by the European Commission that aims at reinforcing a OH network of national reference laboratories and public health institutions to improve preparedness and response capacities to vector-borne diseases, in 22 EU neighbouring countries of the Mediterranean, Balkans, Black Sea and Sahel regions.

The main lines of action are capacity building through training activities and multisectoral networking, both centred on the promotion of the added value of a One Health approach.

The key successes of the project are: i) the setting up of rapid detection capacities in case of emergence/re-emergence and epidemics ii) the strengthening of identification, mapping, and control capacities for vectors of interest. iii) the production of 19 operating procedures and 5 tools for vector identification iv) the formulation of recommendations to improve the integration of data between sectors for risk assessment and in national surveillance plans v) the organization of 5 global and regional meetings to foster regional and multisectoral networking and promote OH added value.

MediLabSecure is now a well-established OH network in the peri Mediterranean region bringing together 111 national reference laboratories and public health institutions from the human and animal health sector; and gathering more than 200 multidisciplinary experts. The main achievement in terms of detection capacities, risk mapping and intersectoral networking is the demonstration of how concrete actions can lead to a successful OH implementation approach. The network is now ready for further development to empower members by promoting in-country initiatives and by supporting the OH advocacy to national stakeholders to leverage the political engagement.

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VEClm Advances: Climate Sensitive Models of Vector Dynamics and Disease Transmission

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The Climate-Driven Vector-Borne Disease Risk Assessment platform (VEClm) is an early warning decision support system to improve the prediction and management of climate-sensitive vector-borne diseases. The project is supported by the Wellcome Trust as part of Digital Technology Development Awards in Climate-Sensitive Infectious Disease Modelling. The platform is operationally maintained at The Cyprus Institute and is permanently available via veclim.com.

The VEClm platform employs data-driven mechanistic modelling to represent vector populations and pathogen circulation as a result of environmental and meteorological changes. The platform displays maps of vector presence, predicted activity, and outbreak risk and impact. In addition, seasonal profiles and medium-range predictions are presented through the GIS interface. The platform hosts a repository of customisable models that incorporate physiological processes driven by relevant climate and environmental factors. A comprehensive application programming interface extends the capacity of advanced data analysis tools, such as Python and R, to accommodate climate-sensitive mechanistic modelling.

The model repository supports a range of models and frameworks, as well as the third-generation implementation of the dynamically structured matrix population framework. Accordingly, a population can be assigned multiple processes, such as survival, latency, and reproduction, that apply sequentially to each individual. Both age-dependent (10.12688/f1000research.15824.3) and accumulative (10.1038/s41598-022-15806-2) processes are integrated into this coherent framework (<https://github.com/keruler/Population>).

To facilitate the standardisation, communication, and storage of models, we propose a JavaScript Object Notation representation (PopJSON). Decorated with custom tags and operations, PopJSON describes the essentials of a dynamically structured multi-process matrix population model and will soon extend to cover frequently used vector dynamics and disease transmission models.

VEClm aims to contribute to reducing the environmental impact of insecticides due to better timing and higher efficiency of vector control and to prevent disease outbreaks due to the incorporation of climate-sensitive risk assessments into early warning systems.

Shortening the gap between vector surveillance and first response in arboviruses outbreaks: from REVIVE network program to MOBVEC project

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Introduction

Aedes mosquito species can transmit dengue, Zika or chikungunya and have been introduced and established in Europe. Prevention and control of vector-borne diseases largely depends on effective and sustainable vector surveillance and timely response to outbreaks. In Portugal, vector surveillance has been under the National Vector Surveillance Network—REVIVE. However, in the event of an outbreak, a rapid entomological and epidemiological response may not be immediately guaranteed. MOBVEC project proposes to be the first VBD Mobile Bio-Lab to be rapidly operational in the heart of outbreaks to support first-responders.

Methods

REVIVE has been ongoing since 2008. CDC light traps and BG Sentinel traps baited with CO₂ or lure have been used to collect adult mosquitoes. Automated field and laboratory classifications were performed in different scenarios and analysed under MOBVEC to evaluate smart-trap technology as a viable tool for first-responders in outbreaks.

Results

In 2023, mosquito monitoring was carried out in 231 municipalities in Portugal and 40,565 mosquitoes were identified by REVIVE. *Aedes albopictus* was identified for the first time in Lisbon. More than 15,000 mosquitoes of different species of *Aedes* and *Culex* genera were automatically classified in tests with smart-traps. These demonstrated the ability to identify and track mosquito dynamics in real time (>85% accuracy) and to perform age grading by differentiating young (<5 days) from older (>6 days) mosquitoes (>70% accuracy).

Discussion

The implementation of networks of sustainable and real-time vector surveillance strategies is essential and is the precursor to interoperable new capacities for first-responders to outbreaks. MOBVEC project will introduce a new technology capable of providing real-time information on vector populations, GEOSS compliant vector risk maps, disease VBD transmission models, and epidemiological maps. This technology will help to prevent and control disease outbreaks.

Acknowledgements

REVIVE Network and MOBVEC project (Ref. 101099283 —MOBVEC— HORIZON-EIC-2022-PATHFINDEROPEN-01) funded by European Commission.

A federative scientist Network under One Health approach to enhance people well-being and sustainable development in Côte d'Ivoire

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In Côte d'Ivoire, the current economic transformations result in significant territorial and environmental changes and are probably linked to an epidemiological transition, leading us to consider jointly issues of health and sustainable development

As a contribution to this agenda, scientists and partners of various disciplines from human, animal, plant and environmental health, decided to set up a federative, inclusive and operational network to encourage an integrative and bottom-up One Health approach.

Our objectives are (i) bringing together scientists to share and resume their knowledge, experience in their field of health science; (ii) giving them the opportunity to co-construct a common understanding and vision of the "One Health" concept (iii) promoting the One-Health concept and collaborating for its implementation to reach sustainable development in Côte d'Ivoire.

Our first action was to organise the "One Health meeting in Côte d'Ivoire" with a diversity of actors sharing their own knowledge about health according to the local practices. This bottom-up approach was resolutely initiated to root our reflexion with the field reality, and to initiate a constructive dialogue between stakeholders and scientists.

Further, a workshop made possible to take advantage of these interactions fed with multi-actors, and multidisciplinary experiences. We therefore initiated, in a participatory way, the actual co-construction of future work through a concept note for an integrative project «One Health in Côte d'Ivoire».

Moreover, as scientists, we were pushed to question our own practices, and rethink the way of producing knowledge by addressing complex health challenges in an integrated and collaborative way for networking. Our network aims at becoming a proposal force for the national One Health platform, initially created to organize the response against pandemics such as COVID 19 with focus on providing expertise and recommendations for health policy development in Côte d'Ivoire.

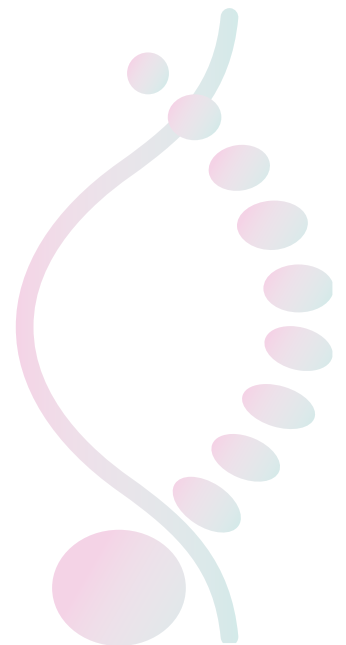


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ABSTRACTS THURSDAY 17 OCTOBER



Exploiting the Biology of Dominant Vector Species to Improve Malaria Control in Africa

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This talk will focus on the biology of *Anopheles funestus*, the leading malaria vector in east and southern Africa, with references to other vectors in the region. Despite challenges in studying this species, due to difficulties in establishing laboratory colonies, emerging evidence points to unique ecological vulnerabilities that present opportunities for targeted control measures. Drawing on data from southeastern Tanzania, where *An. funestus* is responsible for over 85% of malaria cases and exhibits strong resistance to key insecticides. We will discuss its life-history traits, aquatic and adult ecologies, and how these factors influence the efficacy of various control interventions. Ultimately, the presentation will outline strategies for exploiting vector ecology to reduce malaria transmission in different settings.

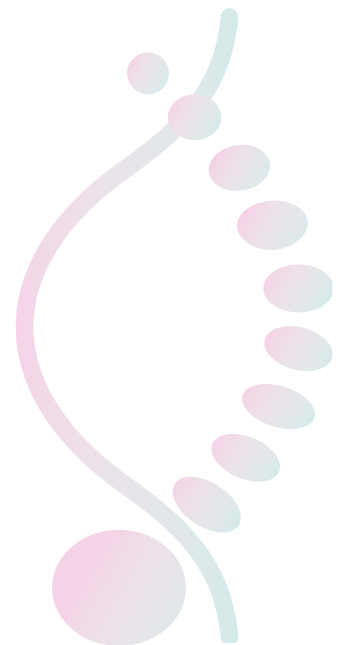


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**Vector surveillance: surveillance systems
community-based surveillance and management of VBDs**



Integrating Citizen and Authoritative Surveillance Data in Mosquito Distribution Models

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In recent years, citizen scientists have become vital in monitoring the spread of mosquitoes that can transmit diseases in Europe. Novel technologies, like mobile phones and the Internet, can be used to track the presence and activity of mosquito species of public health concern. To incorporate these novel data streams into mosquito distribution modelling frameworks, we first need to understand how they are related to authoritative surveillance data and whether they can augment our predictive capacity. Here, we compare and integrate distribution models created from authoritative surveillance and citizen science data for different species, scales, and resolutions. We specifically focused on *Aedes albopictus* at the municipality level in Spain and *Culex pipiens* at a 9x9 km grid level in Europe. We developed three hierarchical Bayesian models for each species: a suitability model (using presence-absence data from authoritative surveillance; binomial distribution), a count model (using count data from authoritative surveillance; negative binomial distribution), and a mixed model (combining mosquito counts from authoritative surveillance with citizen scientists' observations; zero-inflated binomial distribution). The latter model allows differentiation between two processes: one explaining presence/detectability processes and the other explaining mosquito counts. Our results showed that citizen-based estimates are a better proxy for mosquito counts than for presence/absence. This is so because: (i) mixed model predictions are more correlated to count than suitability models, and (ii) model fit improves when citizen information is incorporated into the count part, not into the detectability/zero part of the model. In addition, the large spatial coverage of citizen science data improves the spatial predictions of models. Overall, we show that citizen science data improves mosquito distribution predictions and, therefore, the nowcasting and forecasting of mosquito-borne disease surveillance and epidemiological modelling.

Preventing Another Useless Modelling Task

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Introduction

Vector control agencies across the globe are collecting data on the species and distribution of disease vectors within their jurisdictions. Often, these data are shared with disease modelers to predict vector abundance, disease dynamics, early warning and to forecast future disease risk. However, the uptake of these models into daily vector control activities is limited.

Objective

To identify challenges vector control managers are facing – in particular in the use of statistical and epidemiological models – we sought representation from EU countries and US states along the US/Mexico border.

Methods

We identified 66 vector control or public health agency representatives, of which 31 replied to our email. Interviews were conducted with 26 individuals between July and December 2023. Among the EU interviews, 64% worked at public health entities and 46% were academic partners. In the US, 58% were vector control and 42% public health entities. We performed qualitative data analysis to identify themes related to challenges, successes, capacity, and collaboration.

Results

Almost all of those interviewed visualize their data, either on internal or public facing dashboards. Models were seen as useful, but “not on a day-to-day operational scale at this point in time” where easy-to-calculate vector indices or pathogens rate in vector/host/human are preferred. Models have been successfully integrated when modelers are part of the organization, or where there are resources and culture for collaboration between academic modelers and vector control managers.

Conclusion

Vector-borne disease control intervention development needs the epidemiology of the disease to be fully understood and the intervention effectiveness needs to be measured. These two elements are slowing down preparedness and elimination of endemic and emerging vector-borne diseases.

Success in Mosquito Control: An Integrated Approach in Istanbul, Turkey

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Invasive mosquito species and their emerging/reemerging pathogens are rapidly moving towards previously unoccupied geographical areas, spreading, and establishing themselves in these regions. Despite the genetic and physiological characteristics of mosquitoes, the expansion of mosquito-borne diseases has significantly increased over the past four centuries due to human activities. It is evident that a methodology beyond classical methods is needed to combat these organisms. The One Health concept, which employs a multidisciplinary approach to address such problems, is now recognized as the most important tool we can access and implement.

In this study, the results obtained between 2020 and 2024 of a «State-of-art Integrated Control Program and Action Plan» developed by the Istanbul Municipality and Hacettepe University VERG laboratory under the One Health Concept to combat the invasive *Aedes albopictus* (Asian tiger mosquito) species, which has become a significant public health issue in Istanbul, Turkey, since 2014, was examined and evaluated. In comparisons made with the results from the year 2019, our program's outcomes revealed a 36.3% reduction in the *Aedes albopictus* population size and a 62% decrease in the usage of insecticides in Istanbul. In contrast, approximately 64 million Euros less financial resources were utilized. Since 2022, using a large-scale stochastic time/space model, the likelihood of the occurrence of vector species and the status of *Aedes albopictus* populations were modeled. Models like these predict the situation based on assumptions due to the lack of observation-based high-resolution biological data but reveal crucial findings with high resolution. In this presentation, on the other hand, we evaluated overview of current field studies conducted in Istanbul and how the data obtained from these studies are used in functional modeling studies.

Circulation of West Nile virus in active *Culex pipiens* mosquito populations in the Attica region 2021-2023

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Since 2010, when Greece experienced a major outbreak of West Nile virus (WNV), annual cases have been consistently recorded across various geographical regions. In 2021, the Hellenic National Public Health Organization (NPHO) reported three cases of WNV among humans in the Attica Region (East Regional Unit) and no human cases were recorded in the following years. Since 2021, 24 mosquito monitoring and sampling sites have operated year-round in selected areas in the region, ensuring comprehensive geographical coverage, including sites historically associated with the detection of WNV in humans or mosquito vectors. From May 2021 until June 2023, two distinct trapping systems were established: a BG Sentinel trap with BG lure and a BG Sentinel trap with BG lure and carbon dioxide (CO₂) as attractants, while from July 2023 until December 2023, only the trapping system of the BG Sentinel trap with BG lure operated. Each trap functioned for 24 hours biweekly, and the *Culex pipiens* mosquitoes collected after morphological identification to the species level were pooled according to date and site. A specific TaqMan real-time PCR protocol was used for the detection of WNV. From May 2021 until December 2023, 2,471 samples were collected in the Attica Region, and a total of 20,022 female *Cx. pipiens* were divided into 1,450 tested pools. WNV-positive pools were detected as follows: 19 in 2021, 22 in 2022 and 4 in 2023. WNV-positive samples of overwintering adult mosquitoes were detected in Piraeus and West Attica, regardless of the presence of CO₂ in the traps. The comprehensive mosquito sampling program implemented in the Attica Region revealed, for the first time, the presence of WNV in overwintering vectors captured in adult mosquito traps. The epidemiological importance of our findings is reflected in the necessity of implementing a vector surveillance program annually, aiming to serve as an indispensable component in monitoring both vectorial and WNV dynamics in the area.

Unraveling Dissemination Trajectories of Insect Vectors : Towards Enhanced Prophylaxis in Plant Health

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Introduction and Objectives

Given the tremendous crop losses caused every year by vector-borne pathogens, it is crucial to better understand epidemic processes. This study aims to contribute to this knowledge base by identifying likely dissemination trajectories of insect vectors of plant pathogens. The overarching goal is to inform the development of new surveillance and prophylactic strategies, aligning with the global imperative to reduce pesticide usage.

Materials and Methods

Two types of vectors are studied: psyllids and aphids. The approach consists in investigating probable vector flight trajectories, considering the relative importance of short and long-distance dispersal, local and distant climate, air mass movements, and historical vector abundance data. Initial analyses focus on exploring the statistical relationship between vector population dynamics and local climate using R and/or assessing air mass trajectories and connectivities with the Tropolink tool, leveraging historical vector abundance. This leads to the formulation of probable vector arrival patterns in fields, encompassing their origin, trajectory, and timing. Targeted sampling follows, allowing scenario validation through complementary analyses (e.g., population genetic and dynamics).

Results

Here we will focus on one of our models, the pathosystem psyllids-phytoplasma-apricot tree. Expected outcomes include the precise determination of the spatio-temporal scale of vectors dissemination, unveiling their origins, trajectories, and arrival timings in the fields. Factors influencing these parameters as well as their recurrence or fluctuation over time will also be determined. Results for the aphids models will also be presented and methodology compared with the psyllids model.

Discussion and Conclusion

The main prospects for this work is informing the development of new prophylactic methods in plant health. Moreover, the diversity of models studied lays the groundwork for broader generalizations of the approach, fostering discussion on the genericity of the approach and possible adaptation to other biological models of huge interest to farm animal or human health (e.g., mosquitoes, culicoids).

Novel eDNA-based surveillance strategy targeting quiescent ticks in soil

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Introduction and Objectives

Ticks are obligate hematophagous ectoparasites with worldwide distribution and the ability to transmit tick-borne diseases to both humans and animals. Their rapid geographic expansion and abundance of multiple vector species emphasize the need for systematic tick surveillance. Current tick surveillance efforts focus on the host-seeking phase even though ticks spend the majority their life in the soil digesting their blood-meals, molting, and egg laying. In an effort to expand the existing tick surveillance strategies, a novel molecular approach was developed for the detection, identification, and quantification of tick DNA in soil samples.

Materials and Methods

Tick species of medical and veterinary health importance belonging to 3 genera of ixodids (*Amblyomma*, *Ixodes* and *Rhipicephalus*) were employed for protocol optimization. Ticks (individual or pooled) and from all life-stages (eggs, larva, nymph, and adult) were spiked in soil, and DNA was extracted using a verified arthropod-specific extraction protocol. Tick DNA was detected with PCR using new species-specific primers that were designed based on sequence dissimilarities at the ribosomal RNA gene cluster (5.8S-ITS2-28S rRNA genes) in such a way that the PCR products for each species would be distinguishable by size in gel electrophoresis. PCR amplicons were verified by sequencing.

Results, Discussion and Conclusion

All tick life stages, and targeted species were successfully detected and identified from soil while positive controls (extracted tick DNA without the addition of soil) and negative controls (soil only; blank) were used to validate the results. Specificity tests revealed absence of off-target amplicons showing that these species-specific primers do not cross-react with DNAs of other tick species. This approach could provide a robust, standardized, and economical technique for tick detection and identification in locations of putative tick abundance, complementing existing surveillance methods and facilitating control interventions.

Testing a novel Optical Sensor for *Aedes* and *Culex* adult automatic count and identification and BG-trap capture rate determination

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Introduction and Objectives

Effective mosquito vector monitoring is increasingly needed to optimize control interventions and predicting risk of pathogen transmission. However, conventional methods are time- and labor-intensive and difficult to implement on a large spatial and temporal scale. Recently, an optical sensor combined with a supervised machine learning algorithm (Vectrack), was developed with the goal to allow automatic count and identification of adult mosquitoes in conventional BG-traps. Here we summarize the results of field experiments carried out in summer 2023 in Italy to evaluate: (i) the accuracy of Vectrack in counting and identifying *Aedes* and *Culex* adults, and (ii) whether BG-trap catching capacity is affected by the sensor presence. The experiments also provided the opportunity to estimate the capture rate of BG-traps based on the known capture rate of Sticky Traps.

Materials and Methods

The experimental design implied the rotation every 48h of three types of traps – one BG-Mosquitaire (BG-M), one BG-M equipped with the sensor, and 4 Sticky Traps (considered as a single trap due to their lower catching capacity) – in three sites within an area of about 1,000m². The same experimental design was carried out in 4 Italian provinces (Bergamo, Padua, Rome, Naples) and replicated 3 times/area. Collected mosquitoes were counted and identified both automatically by the Vectrack and by visual inspections.

Results, Discussion and Conclusion

The capture performance of the BG-M with and without Vectrack was comparable both overall and when assessed against *Aedes* and *Culex*, or sexes (females, males) (Tukey's Honest Significant Difference method, p-value NS). High correlation was observed in *Aedes* and *Culex* counts between Vectrack and operator (Pearson cor 0.985, pval <0.0001). Overall, results support Vectrack high potential for continuous monitoring with minimal human effort and opened the possibility of unprecedented studies on mosquito seasonal and circadian rhythms in 2024.

Estimating the probability that observed absence of tsetse (Diptera: Glossinidae) indicates elimination

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Introduction

There have been numerous reports since the 1910s of tsetse control exercises resulting in claims of elimination. However, the associated evidence was often insufficient and elimination only became evident with time and the continued absence of tsetse, and diseases. We develop methodologies providing greater confidence that vector elimination has been achieved. These are particularly important for both sleeping sickness and African animal trypanosomosis elimination, where vector control has contributed to disease elimination

Methods

We combine processes that: (i) relate the probability that failure to capture tsetse over a given period, using sampling techniques of known efficiency, indicates population elimination; (ii) predict extinction probabilities of small tsetse populations with given vital parameters; (iii) estimate the growth of small populations of tsetse; (iv) model the dispersal of tsetse by diffusion. We apply it to real field data and case studies in Tchad and Kenya.

Results

We show that the difficulty of being sure of elimination increases with the area targeted. Zero catches from stationary traps with low capture efficiency often provide – by themselves - insufficient evidence of tsetse elimination. Mobile baits could cover greater proportions of infested areas and capture tsetse with higher probability. Increased confidence in elimination resulted from estimating the growth of surviving populations, and the resulting increased probability of capture, or estimates that population densities were so low that autonomous extinction would occur with high probability.

Discussion

To declare that elimination has been achieved it is necessary to stop all control and then monitor vector and disease levels for prolonged periods. This comes with the danger that, if tsetse have not been eliminated, the disease may return. Accordingly, control measures should always be maintained until it can be concluded that the tsetse population is sufficiently low that it can be controlled even if has not already been eliminated.



ONE HEALTH IN ACTION

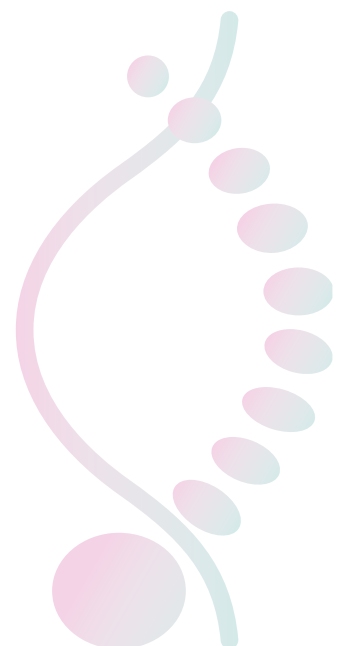
14-17th October 2024
MONTPELLIER, FRANCE



LIST OF POSTERS

Colored stars are for posters in competition for the awards

- ★ Best Poster award - Master students and PhD
- ★ Best Poster award - Post doctoral researchers



Integrative taxonomy and evolution

Last name	First name	Title of poster	# poster
BALCAZAR	Dario	Genetic Structure and Evidence of Introgression in Saudi Arabian <i>Aedes aegypti</i> Populations	S1-P01
BARRÉ-CARDI	Hélène	What species do these <i>Culex</i> (Culicidae) larvae belong to? Description of a surprising set of criteria	S1-P02
BON	Marie-Claude	A new record of <i>Haemaphysalis bispinosa</i> Neumann and <i>Haemaphysalis cornigera</i> Neumann on cattle in the Thai Nguyen Province in Vietnam	S1-P03
★ CEVIDANES	Aitor	Development of a duplex real-time PCR assay for rapid Identification of <i>Aedes albopictus</i> and <i>Aedes japonicus</i> eggs	S1-P04
★ CURRAN	Thomas	Genetic identification of mosquitoes in Ireland using a multi-sampling approach of direct and indirect samples	S1-P05
★ DIONE	Mamadou Lamine	Eco-geographical characterization of an <i>Anopheles gambiae</i> hybrid zone in 'Far-West' Africa	S1-P06
DUJARDIN	Jean-Pierre	Reliability of AI versus manual digitization of anatomical landmarks	S1-P07
FONTAINE	Michael C.	Mitochondrial variation in <i>Anopheles gambiae</i> and <i>An. coluzzii</i> : phylogeographic legacy and associations with metabolic resistance to pathogens and insecticides	S1-P08
★ HELLEMAN	Pepijn	How ecology and population history shape mosquito population genetics: a case study from the Caribbean	S1-P09
KAMPEN	Helge	Incorrect DNA sequence entries in data bases and their consequences, exemplified for <i>Culicoides</i> biting midges (Diptera: Ceratopogonidae)	S1-P10
		Comparative analysis of the mitochondrial full genome of Central European <i>Culicoides</i> species (Diptera: Ceratopogonidae)	S1-P11
KRATZ	Fanny	Unlocking sand fly biodiversity in Nepal: Enhancing disease control through DNA-barcoding	S1-P12
LA CORTE	Roseli	Analysis of the genetic structure of <i>Haemagogus leucocelaenus</i> (Diptera: Culicidae), yellow fever vector, using the Cytochrome Oxidase I gene	S1-P13
★ LORANG	Camille	Non-destructive extraction of DNA in ticks: a method to keep the shape!	S1-P14
★ PEREIRA NEVES	Aline	Faithful tenants: the subgenus <i>Microculex</i> (Diptera: Culicidae) and its predilection for bromeliad phytotelms	S1-P15
★ SAUER	Felix	Mosquito species identification with geometric morphometrics based on wing images captured with different devices	S1-P16
SERENO	Denis	Wing Interferential Patterns (WIP) in Medically Important Dipteran Insects for Automatic Classification Using Deep Learning	S1-P17
TIZON	Charles	A simple key to identify <i>Aedes albopictus</i> larvae to the naked eye in the field in Western Europe	S1-P18
★ VAN DER BEEK	Jordy	What the mitogenomes of 27 European mosquito species about the discriminative power of the different mitochondrial genes	S1-P19

Vector ecology and biology

Last name	First name	Title of poster	# poster
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ANDRADE	Pâmela	Understanding the behavior of <i>Aedes albopictus</i> : Zika Infection and Locomotor Activity in Laboratory Conditions	S2-P02
★ BALVERS	Carlijn	Blood-fed mosquitoes as 'flying syringes' in a field setting	S2-P03

BARCELO	Carlos	Effect of ABO blood group on the bionomics of <i>Aedes albopictus</i> under laboratory conditions	S2-P04
★ BILLOT	Maelys	How do host contaminants affect vector fitness ? An experimental study of <i>Ornithodoros</i> ticks and mercury-contaminated host blood	S2-P05
★ BLANCO-SIERRA	Laura	On the Move: Tracking Passive Dispersal and Survival of <i>Aedes albopictus</i> in North-Eastern Spain	S2-P06
BOURQUIA	Maria	Diversity of biting midges, mosquitoes and sand flies at four dog shelters in rural and peri-urban areas of Central Morocco	S2-P07
COOPER	Rodney	Molecular gut content analysis to identify non-crop sources of insect vectors	CANCELLED
DELLA TORRE	Alessandra	How old are you? Assessing <i>Aedes albopictus</i> age by mid-infrared spectroscopy	S2-P09
★ GANDY	Sara	Tick-borne disease hazard in urban green spaces and their surrounding rural environment	S2-P11
★ GIUPPONI	Carla	Expansion and adaptation of the <i>Hyalomma marginatum</i> tick in the Occitanie region	S2-P12
GUIS	Helene	What information can a longitudinal survey carried out in a single multi-host site provide on mosquito dynamics and their drivers in peri-urban Antananarivo, Madagascar?	S2-P13
HERREN	Jeremy	Harnessing mosquito symbionts for malaria transmission blocking	S2-P14
IPPOLITI	Carla	Deep Learning techniques to predict <i>Culex pipiens</i> suitable habitat in central Italy using Sentinel-2 data	S2-P15
★ ISIYE	Elsie	A DNA-based approach to surveillance of <i>Culicoides</i> vectors and their vertebrate hosts in Ireland	S2-P16
★ JAQUET	Mathilde	Unraveling the Impact of Plant-Based Diets on <i>Aedes albopictus</i> Mosquitoes and Arboviral Transmission in Occitanie	S2-P17
★ LOPEZ-PEÑA	David	Altitudinal distribution pattern of <i>Aedes (Stegomyia) albopictus</i> (Skuse, 1894) in Valencia province, Eastern Spain	S2-P18
★ LOUNNACI	Zohra	Biodiversity and bioecology of <i>Culicidae</i> (Diptera, Nematocera) in rural, suburban and urban areas of Tizi-Ouzou (Algeria)	S2-P19
★ LUCATI	Federica	Tiger mosquito blood-feeding behaviour and possible implications for disease transmission	S2-P20
LÜHKEN	Renke	Broad host preference and host-feeding patterns of <i>Culex pipiens</i> s.s./ <i>Cx. torrentium</i>	S2-P21
★ MAIA	Pollyana	Pyrethroid resistance profile of <i>Aedes aegypti</i> (Diptera: <i>Culicidae</i>) populations from Sergipe, Brazil	S2-P22
★ MARTINS AFONSO	Sara	Distribution of <i>Culex pipiens</i> s.s. and <i>Cx. torrentium</i> in Europe	S2-P23
★ MERCIER-LARRIBE	Chiara	The role of biodiversity on the dynamics of trematode communities and their circulation	S2-P24
MIGNOTTE	Antoine	Track the Tiger: Unraveling Resting Site Preferences of <i>Aedes albopictus</i> in Occitania, France	S2-P25
NABET	Cécile	Estimating the age of field <i>Anopheles</i> mosquitoes using mass spectrometry and deep learning	S2-P26
★ O'DONNELL	Aidan	Dancing out of step: The adaptability of <i>Anopheles stephensi</i> rhythms and their impact on malaria development	S2-P27
★ OKE	Catherine	Poorly-resourced mosquitoes exhibit increased host seeking and early evening biting	S2-P28
★ OUEDRAOGO	Cheick Oumar Wendpagnandé	Comparing Ivermectin susceptibility in wild-derived and laboratory-reared <i>Aedes</i> and <i>Anopheles</i> larvae: Preliminary study on selecting for resistance mechanisms	S2-P29
★ PARDO ARAUJO	Marta	The spread of <i>Aedes albopictus</i> in Spain: linking human mobility and habitat suitability	S2-P30

★ PARVY	Jean-Philippe	Assessing the risk of mosquito vector-borne diseases in Scotland and their response to environmental change	S2-P31
DELLA TORRE	Alessandra	Longitudinal survey of insecticide resistance in a Burkina Faso village reveals co-occurrence of 1014F/S and 402L mutations in <i>Anopheles coluzzii</i> and <i>Anopheles arabiensis</i>	S2-P32
RANAIVOARISOA	judith	Breeding of Tsetse Flies and Animal Welfare at CIRAD Baillarguet	S2-P33
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★ RODRIGUEZ VALENCIA	Victor Manuel	Host Preferences of <i>Culex pipiens</i> and its Significance for West Nile and Usutu Flaviviruses transmission in Camargue, France	S2-P35
★ RUIVO	Margarida	Tick-borne microorganisms detected in small mammals collected in the surroundings of two villages in Styria, Austria	S2-P36
★ SHAHIN	Keana	Investigating the buzz: What environmental and weather factors are associated with <i>Aedes albopictus</i> in Southern Ontario, Canada?	S2-P37
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TSIKOLIA	Maia	Exploring Positive Chemotaxis in <i>Phlebotomus papatasi</i> Larvae	S2-P40
★ VAZQUEZ	Antonio	Unveiling a snail host of a major trematode in Europe: <i>Galba schirazensis</i> (Lymnaeidae) facilitate the transmission of Paramphistomidae while disengaging <i>Fasciola hepatica</i>	S2-P41
VILLARI	Sara	Role of microhabitat diversity and interspecies competition in the study of Culicidae larvae interactions	S2-P10
★ WIJNVELD	Michiel	Updated distribution of <i>Dermacentor</i> ticks in Austria	S2-P42
★ ZELA	Lamidi	Does Ivermectin impair <i>Anopheles</i> mosquitoes' attractiveness toward treated cattle under field and laboratory conditions?	S2-P43

Eco-epidemiology of VBDs under anthropogenic and climatic changes

Last name	First name	Title of poster	# poster
★ BARTHOLOMEE	Colombine	How urban green infrastructures influence <i>Aedes albopictus</i> presence and abundance? A study in the city of Montpellier (South of France) in 2023	S3-P01
★ CHEVALIER	Laure J.G.	Modelling RVF vector population dynamics in a tropical setting in South-Eastern Madagascar	S3-P02
COSTANTINI	Carlo	Diurnal and outdoor biting by <i>Anopheles gambiae</i> complex malaria vectors reveals residual transmission along an urbanisation gradient in Burkina Faso	S3-P03
DEBLAUWE	Isra	Monitoring the public health impact of climate change through environmental indicators: a case study on the Asian tiger mosquito (<i>Aedes albopictus</i>) in Flanders	S3-P04
★ FUSTEC	Benedicte	The Remote Emerging Disease Intelligence-Network: From Concept to Active Surveillance	S3-P05
★ GARCIA VAN SMEVOORDE	Margot	Multifactorial characterization of dengue transmission dynamics in the French Caribbean islands to better prepare for future epidemics	S3-P06
GILBERT	Lucy	Environmental risk factors for <i>Anaplasma phagocytophilum</i> in Scotland	S3-P07
★ HARTUNG	Tijmen	Wetland development and its effects on avian arbovirus hosts in nearby urban areas	S3-P08
IBANEZ-JUSTICIA	Adolfo	Unraveling the effect of urban green on mosquito species in the Netherlands	S3-P09

KASHEFI	Javid	Interface between invasive hard ticks and weeds in the context of climate change	S3-P10
★ KRUPA	Eva	Ticks in and around Paris: maps of tick's presence and associated tick-borne pathogens	S3-P11
MARTIN	Estelle	Impact of socio-economic factors on mosquito abundance and diversity	S3-P12
MOHAN	Iswaryalakshmi	Tick- host dynamics underpinning transmission of Kyasanur Forest Disease in degraded forest landscapes in India: insights from MonkeyFeverRisk	S3-P13
★ MUCCI	Giuliano	Assessing the potential impact of anthropization in the forest ecosystem in Southern Cameroon on vector communities and the risk of zoonotic virus emergence	S3-P14
RADZIJEVSKAJA	Jana	The distribution of Ixodes ricinus ticks and their infection with Borrelia spp., Anaplasma phagocytophilum and Neorhlichia mikurensis in urban green spaces in Lithuania	S3-P15
★ RAHARINIRINA	Manou	Variation in the spatial distribution of Aedes aegypti and Aedes albopictus in Madagascar	S3-P16
SCHÄFER	Stefanie M.	Identifying socio-ecological drivers of tick-borne zoonosis impacts for disease management: insights from a OneHealth systems approach to Kyasanur Forest Disease in India	S3-P17
SOHIER	Charlotte	West Nile virus monitoring in Flanders (Belgium) during 2022-2023 reveals endemic Usutu virus circulation in the wild bird population	S3-P18
SZENTIVANYI	Tamara	Urban adaptation, host behavior, and distribution affects the presence of Dirofilaria parasites: a phylogenetic comparative study of wild carnivores	S3-P19
★ TACONET	Paul	modis2fast : an R package for efficient access to satellite-based MODIS (and similar) data used in landscape eco-epidemiology	S3-P20
★ VARGA	Zsaklin	Studying the effects of Aedes invasive and native mosquito species on the circulation of Filarial nematodes in a hungarian southwestern city	S3-P21
VERHULST	Niels	Influence of climate change and infection on thermal preferences of mosquitoes	S3-P22

Interactions between host(s)-vector(s)-pathogen(s)-microbiota-environment

Last name	First name	Title of poster	# poster
★ ALBA	Annia	Microbiome and host resistance to a trematode parasite: the case of a vector snail	S4-P01
ALOUT	Haoues	Virulence and transmission vary between Usutu virus lineages in Culex pipiens	S4-P02
ALTINLI	Mine	Unraveling Mechanisms of ISV-Arbovirus Interactions in Mosquito Cells	S4-P03
★ BAUDON	Daphné	Mosquito infection dynamics by Rift valley fever virus	S4-P04
★ BERNARD	Célia	First detection of Crimean–Congo haemorrhagic fever virus in Hyalomma marginatum ticks, southern France.	S4-P05
★ BOURNE	Mitchel	Deciphering the role of skin bacterial odours in the attraction of mosquitoes using in vivo and in vitro studies	S4-P06
★ BRAMS	Evi	Vector competence of Belgian Culex pipiens and Anopheles plumbeus mosquitoes for West Nile Virus	S4-P07
★ BURGAS	Albert	The West Nile virus lineages circulating in Spain can be transmitted by Culex pipiens reared in two different types of water	S4-P08
★ BUYSSE	Marie	Worldwide distribution of Candidatus Borrelia capensis in ticks associated with seabirds	S4-P09
★ FATTAR	Noor	Tissue tropism convergence among tick major nutritional symbionts: insights from preliminary results	S4-P10
★ FERRARI	Giulia	Spatio-temporal re-colonization of top-predators modulates zoonotic infections in ticks	S4-P11
GONZALEZ FERNANDEZ	Estela	Blood Source Impact on WNV lineage1 transmission dynamics in Culex pipiens	S4-P12

★ GOUSI	Fani	A virus-induced gene silencing (VIGS) approach to study plant-geminivirus-insect vector interactions and virus transmission	S4-P13
GRADONI	Francesco	Occurrence of tick-borne pathogens in ticks collected from wildlife and domestic animals in northeastern Italy	S4-P14
★ HARTKE	Juliane	Insecticide resistant Anopheles from Ethiopia but not Burkina Faso show a microbiome composition shift upon insecticide exposure	S4-P15
★ HÖLLER	Patrick	Abstract Title: Unveiling Tahyna Virus Transmission: Vector Competence of Mosquitoes from Europe	S4-P16
★ JAGTAP	Swati	Interactions of insect-specific viruses (Bunyavirales) with the mosquito host and Rift Valley Fever Virus	S4-P17
JEANNEAU	Melanie	Arbovirus infection modulates deltamethrin susceptibility of Culex pipiens	S4-P18
KORYTAR	Ľuboš	First evidence of flavivirus circulation in Greater Mouse-eared bats Myotis myotis (Borkhausen, 1797) in Slovakia	S4-P19
KURUCZ	Kornélia	Characterizing the bacterial microbiome of the invasive vector Aedes albopictus in Hungary: a pilot study using Oxford Nanopore sequencing	S4-P20
LOISEAU	Claire	Understanding interactions between humans, mosquitoes, waters and plastics: an interdisciplinary approach in Montpellier, France	S4-P21
★ MANNUCCI	Mélissa	The social construction of Aedes albopictus as a public problem: views cross between daily nuisances and public health issues in Occitania	S4-P22
★ MICHELS	Emile	Non-native game birds released for recreational shooting amplify Lyme disease risk via spillback	S4-P23
PAULAUŠKAS	Algimantas	Ticks and flies as vectors of Bartonella spp., Mycoplasma spp. and Rickettsia spp. pathogens in cats	S4-P24
PONDEVILLE	Emilie	Microbiota-nutrition-physiology interactions in Aedes mosquitoes: treat and trick?	S4-P25
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★ RAKOTOBÉ HARIMANANA	Ravo Niaina	The vector competence of Synopsyllus fonquerniei and Xenopsylla brasiliensis fleas from Madagascar to transmit the plague bacillus	S4-P27
SCROGGS	Stacey	Impact of timing on co-infection dynamics of bluetongue virus serotypes 13 and 17 in Culicoides biting midge cells	S4-P28
★ SY	Victoria	Aedes aegypti vector competence for two genetically divergent Kenyan Rift Valley fever virus isolates	S4-P29
★ TROZZI	Gabrielle	Exploring the interaction between Tick-Borne Encephalitis Virus (TBEV) and Ixodes ricinus ticks after infection via an Artificial Blood Feeding system	S4-P30
★ ZAHRI	Abderrahmane	First detection and molecular characterization of Hepatozoon canis, in Rhipicephalus sanguineus sensu lato ticks collected from dog shelters in central Morocco	S4-P31

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★ BELLVER ARNAU	Jesús	Optimisation of tiger mosquito control in Barcelona	S5-P02
★ BERGER	Audric	Development of dPCR and NGS methods for improved surveillance and detection of new mosquito densovirus	S5-P03
BRANDOLINI	Teresa	Production and supply of Aedes albopictus sterile males	S5-P04
CHASKOPOULOU	Alexadra	Biting Back! Wild harvested mosquitoes as an alternative protein ingredient in broiler feed: Insights from the first pilot study	S5-P05

COHUET	Anna	Partial clearance of pre-established <i>Plasmodium falciparum</i> infection in mosquitoes by feeding on patients treated with Artemether+Lumefantrine + Atovaquone-Proguanil	S5-P06
★ CHANDRE	Fabrice	Insecticidal activity of essential oils of <i>L. alba</i> harvested in Burkina Faso and Côte d'Ivoire against mosquito vectors	S5-P07
★ CSIBA	Rebeka	Impacts of prolonged use of unilateral mosquito control methods in Hungary: Evaluating pyrethroid resistance in disease vectors <i>Culex pipiens</i> and <i>Aedes albopictus</i>	S5-P08
★ DUCHAUDE	Yolène	Study of the chemotype of Caribbean plants in the vector control of the <i>Aedes Aegypti</i> mosquito	S5-P09
FERRE	Jean-Baptiste	Mosquifen: are mosquito traps an effective tool in the battle against mosquitoes?	S5-P10
FLACIO	Eleonora	Application of sterile male of <i>Aedes albopictus</i> as an integrated control measure in Switzerland: Results of two consecutive release seasons	S5-P11
JEAN	Frédéric	Assessing the impact of a low-cost door-to-door strategy on <i>Aedes albopictus</i> populations	S5-P12
★ LE MAUFF	Anais	Defining repellent properties and electrophysiological responses of essential oils against the Lone Star tick, <i>Amblyomma americanum</i>	S5-P13
MOULINE	Karine	Using the ectocide Fluralaner administered per os to <i>Rattus rattus</i> to control the flea <i>Xenopsylla cheopis</i> , vector of <i>Yersinia pestis</i> in Madagascar: the Ectopeste project	S5-P14
MÜLLER	Pie	Citizen action for sustainable control of dengue vectors in Abidjan, Côte d'Ivoire: a cluster-randomised control trial	S5-P15
★ NDOYE	Baye Bado	Ecological alternatives to insecticides for controlling vector-borne diseases: effects of caffeine on mosquito ageing	S5-P16
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★ ROONEY	Sara	Exploring <i>Wolbachia</i> and Usutu virus interactions- a potential control strategy?	S5-P18
SHIAO	Shin-Hong	Development of a Broad-Spectrum Antiviral System Using Flavivirus-Inducible Gene Expression to Control Mosquito-Borne Diseases	S5-P19
TANNIERES	Mélanie	An innovative method of using <i>Agrobacterium tumefaciens</i> to screen for AMPs against insect-transmitted plant vascular diseases: demonstrated with <i>Xylella fastidiosa</i>	S5-P20
★ TSAFRAKIDOU	Panagiota	Repellent activity evaluation of Cumin Essential Oil and its components against <i>Phlebotomus papatasi</i> (Diptera: Psychodidae) via a high-throughput screening bioassay	S5-P21

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POINSIGNON	Anne	The INOVEC Project: a pan-European partnership for enhancing the surveillance and control of mosquito vectors of emerging arboviruses	S6-P04
RADICI	Andrea	VECTOCLIM PROJECT. Assessing the impact of climate change on two vector-pathogen pairs: <i>Aedes albopictus</i> & <i>Culicoides</i> midges in Occitania, France and Europe	S6-P05
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Vector surveillance: surveillance systems, community-based surveillance and management of VBDs

Last name	First name	Title of poster	# poster
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★ ANDRIAMAMORISOA	Lalaina Mirado	Historical review of insecticide resistances of Anopheles in Madagascar: Origin and spatio-temporal dynamics	S7-P03
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BAKRAN-LEBL	Karin	The Asian tiger mosquito - Current situation and surveillance programs in Austria	S7-P05
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★ BARON	Jerome	First findings from "Rapportera Fästing" – a citizen science tool for reporting ticks in Sweden	S7-P07
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CHAKHUNASHVILI	David	Investigation of tularemia in Shida Kartli (Georgia) with One Health approach	S7-P10
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D'ALESSIO	Silvio Gerardo	West Nile and Usutu surveillance in Abruzzo and Molise regions (Central Italy): entomological approach and data management	S7-P12
DEFILIPPO	Francesco	Tick infestation on migratory birds and spread of different Borrelia and Rickettsia species: a seasonal study in Lombardy region (Northern of Italy)	S7-P13
FOURNET	Florence	Use of mobile applications and Geographic Information Systems for enhanced characterization of Aedes albopictus risk areas in urban environments	S7-P14
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KNAPIČ	Tea	Comprehensive Surveillance of Mosquitoes and Sandflies in Slovenia: Implications for Public Health and Emerging Pathogens	CANCELLED
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ONE HEALTH IN ACTION

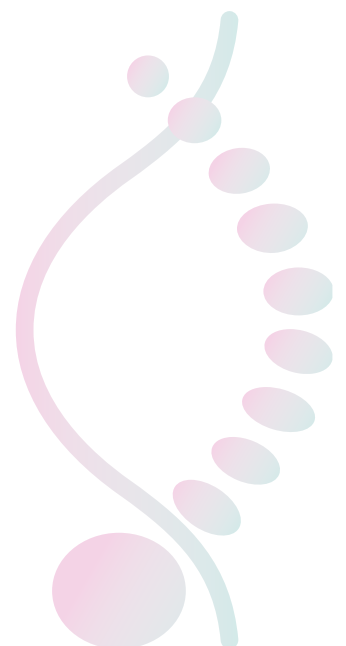
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ABSTRACTS POSTERS

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- ★ Best Poster award - Master students and PhD
- ★ Best Poster award - Post doctoral researchers



S1-P01

Genetic Structure and Evidence of Introgression in Saudi Arabian *Aedes aegypti* Populations

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The mosquito *Aedes aegypti* is an important vector of viruses that cause human diseases, such as dengue, chikungunya, Zika, and yellow fever. There are two genetically distinct subspecies of *Aedes aegypti*: *Aedes aegypti formosus* (Aaf) and *Aedes aegypti aegypti* (Aaa). Aaf is exclusively found in sub-Saharan Africa, breeds in nonhuman-disturbed habitats, and prefers animal blood meals; while Aaa is widely distributed outside Africa, breeds in human habitats, and prefers human blood meals. Saudi Arabia presents an opportunity to study the interaction between these two subspecies due to its proximity to African Aaf and a native Aaa population.

We collected mosquito populations from various locations in Saudi Arabia: Jeddah, Mecca, Najran, Sahil, Madinah, and two regions of Jazan (low and high altitude). Mosquitoes were genotyped using the *aegypti* SNP-Chip and genotypes analyzed to determine genetic structure and relationships with African Aaf and Asian Aaa populations.

Our analysis revealed the presence of 4 distinct genetic groups within Saudi Arabian mosquitoes, with Jazan and Sahil populations forming separate clusters away from the rest of the mosquitoes. The Jazan low and high populations appeared to be isolated from most Saudi Arabian mosquitoes, except for Sahil. Additionally, some Aaf ancestry was detected in the Jazan and Sahil populations, likely a consequence of introgression of neighboring Aaf into Saudi Arabia. Our results unveil the spread of Aaf outside Africa, affecting the genetic structure of Saudi *Aedes aegypti* populations and introducing novel genetic elements that could have implications for viral disease transmission.

What species do these *Culex* (Culicidae) larvae belong to? Description of a surprising set of criteria

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The corsican mosquito fauna, studied for many years by the Corsican Agency of Environment (OEC) and its partners, was considered relatively well known. However, some rarer species would need to be studied again, or even redescribed.

On November 2022, we have found four larvae presenting surprising morphological criteria: they are all already described, but in different species. Our first hypothesis was that the larvae belonged to *Culex brumpti*, a species never been found again in Corsica since its first description more than 90 years ago. In fact, the majority of criteria could correspond to this species. But after more in-depth observations, some divergences concern us. *Culex theileri* is well known in Corsica and a few criteria of our larvae are more similar to this species.

After years of field observations, the presence and distribution of these species in Corsica must be clarified.

Until the question can be answered with the help of genetics, we therefore discuss here the criteria observed in our specimens and compare them to the four neighboring species, *Cx. brumpti*, *Cx. theileri*, *Cx. univittatus* and *Cx. perexiguus*.

S1-P03

A new record of two *Haemaphysalis* tick species on cattle in the Thai Nguyen Province in Vietnam

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The invasive Asian longhorned tick (ALT), *Haemaphysalis longicornis* Neumann, is rapidly becoming an agricultural and epidemiologic concern in the United States. Native to eastern Asia, this tick is a major pest of domestic livestock throughout its invasive range in Australia, New Zealand, and surrounding islands and parasitizes wildlife and humans. Classical biological control is being explored as a strategy to control ALT in the United States, and special attention is given to the search of parasitoid wasps belonging to the genus *Ixodiphagus* (Hymenoptera: Encyrtidae) in Southeast Asia, and particularly in Northern Vietnam. In recent years, intensive efforts have been made to survey the diversity of ticks in Vietnam, though available data on ticks in Vietnam remain limited. The reason is that the lack of reference data and standard taxonomic keys specific to Vietnamese tick species makes the morphological identification of Vietnamese ticks difficult. Nevertheless, 48 species of nine tick genera have been reported so far and recently two new *Dermacentor* species have been described. Our recent exploration efforts in the Thai Nguyen Province in Northern Vietnam revealed the presence on cattle of several *Haemaphysalis* specimens which did not match the morphological descriptions of ALT. By using an integrative taxonomic approach based on morphological and two-gene phylogeny analyses we assigned these specimens to *Haemaphysalis shimoga* Trapido and Hoogstraal and *Haemaphysalis bispinosa* Neumann, which both originated in India. This is the first record confirmed by a molecular analysis of the presence of these two species on cattle, and moreover in the Thai Nguyen Province. Both species are of particular interest as both have been found in association with cattle *Babesia* though their role as vectors remains unclear.



Development of a duplex real-time PCR assay for rapid Identification of *Aedes albopictus* and *Aedes japonicus* eggs

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Aedes Invasive Mosquitoes (AIM) are rapidly colonizing regions across Europe, posing significant challenges to public health. In the Basque Country (northern Spain) established populations of *Aedes albopictus* (*Aea*) and *Aedes japonicus* (*Aej*) are spreading. Previous studies have revealed distinct spatial distribution patterns of these species: *Aea* tends to dominate in densely populated cities, particularly urban and suburban areas, whereas *Aej* shows a preference for periurban areas and cities with lower population density. Despite these general trends, both species are now being identified co-occurring in an expanding number of municipalities. Traditionally, species identification has relied on labor-intensive methods, such as hatching eggs from ovitraps and morphological identification larval or adults. In cases where hatching fails DNA sequencing becomes necessary, albeit time-consuming and impractical for rapid response scenarios. Therefore, this study aims to address this challenge by developing a duplex real-time PCR assay specifically designed for the rapid identification of both *Aea* and *Aej* in the region. The qPCR was designed targeting the COI gene and selecting sequences to differentiate between both species. Primers and probes have been meticulously designed for specificity considering all relevant haplotypes and other non-target *Aedes* species. The assay utilizes the SensiFast Probe Lo-ROX kit, and FAM and CY5 probes for species-specific detection.

During qPCR optimization, two life stages (the egg and the adult) were used. Various quantities of eggs were tested to assess sensitivity, demonstrating the assay's ability to detect even a single egg. Furthermore, the qPCR assay was evaluated with mixed-species samples, yielding positive results for species identification even in mixed populations.

By streamlining the identification process, this assay will significantly enhance the efficiency and effectiveness of AIM surveillance programs. This improvement will facilitate prompt interventions to prevent the rapid dispersion of these vectors into new municipalities.



S1-P05

Genetic identification of mosquitoes in Ireland using a multi-sampling approach of direct and indirect samples

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Ecosystem services provided by insects include soil aeration, plant pollination and pest control. Insects also pose concern, particularly vectors for disease, such as some of the 3500 mosquito species that occur worldwide. Across Europe, cases of mosquito borne diseases have been recorded and are associated with anthropogenic changes. Mosquito monitoring is vital to create informed mitigation measures in advance of potential disease outbreaks. Morphological identification of mosquito larvae and adults requires a high level of expertise and is susceptible to error. Here, we demonstrate the application of DNA technologies to identify mosquitoes from larvae, adults and indirectly from the diet of bats and from water (i.e. environmental DNA (eDNA)) using a combination of DNA-based techniques including Sanger sequencing, DNA metabarcoding and real-time PCR (qPCR). Sanger sequencing facilitated the identification of previously unrecorded mosquito species in Ireland (i.e. *Culex torrentium*). Using DNA metabarcoding on bat faecal pellets a highly diverse diet was revealed and included the identification of four mosquito species, one of which was also previously unrecorded in Ireland, (*Culex quinquefasciatus*). Finally, species-specific qPCR assays were designed and optimised and allowed for the rapid detection of mosquitoes and have been applied to DNA extracted from bat faecal pellets and water samples and allowed for Irish mosquito distribution maps to be updated and identified the spread of various species throughout Ireland resulting from the lack of Irish mosquito surveillance efforts. The combination of these molecular techniques have facilitated the identification of mosquito species occurring in Ireland via direct and indirect approaches.



S1-P06

Eco-geographical characterization of an *Anopheles gambiae* hybrid zone in 'Far-West' Africa

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Afrotropical malaria vectors *Anopheles gambiae* and *An. coluzzii* are sister species in the Gambiae complex diverging ecologically and genetically despite occasional hybridization (<1%). An exception is a region in 'Far-West' Africa, ranging from Guinea-Bissau to Senegal, where unusually high hybridization rates (3%-24%) have been recorded. Recent whole-genome analyses of populations from Guinea Bissau and The Gambia highlighted the occurrence of a distinct hybrid taxon having a mosaic genome inherited from the two species. This hybrid zone constitutes a suitable region where to study the genetic architecture of speciation, and identify mechanisms allowing these taxa to maintain their integrity in the face of gene flow.

To characterize the genetic and eco-geographical structure of 24 populations along the Casamance river drainage basin in southern Senegal—a region lying inside the hybrid zone and in-between the localities where the hybrid taxon was reported—we carried out two surveys separated by a 5-yr interval, across a 300-km transect spanning a gradient of water salinity. More than 3,000 *An. gambiae* s.s. from 175 breeding sites characterized for their physico-chemical profile, were genotyped. The hybrid status of each larva was assessed by the the Divergent Islands SNP assay aimed at distinguishing parentals, hybrids, and backcross individuals.

Larvae that could be considered as F1 hybrids were rare, indicating that hybridization between parentals is occasional in this region too. Hybrid genotypes on the X chromosome were markedly less than on the 2L and 3L divergence islands. A positive correlation between hybridization and salinity emerged from the distinct distribution of genotypes on the opposite sides of the salinity ecotone generated by an anti-salt dam: less hybridized genotypes prevailed in the freshwater section, while more hybridized genotypes prevailed on the saltwater side. Larvae with mostly *An. coluzzii* ancestry-informative markers on chromosome X clustered in populations at and just upstream of the ecotone.

Reliability of AI versus manual digitization of anatomical landmarks

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Digitization of anatomical landmarks is a critical process in geometric morphometrics, challenged by the labor-intensive, and error-prone nature of manual methods. Application of machine learning or AI-automation is accordingly highly anticipated, but only a few publications have so far reported success in this area and offered suitable software.

Such an application would be highly beneficial, particularly when dealing with abundant material. It is recommended that users (human) repeat their manual digitizing at least once to verify their own precision. User error is generally less than the inter-user error, i.e., the discrepancies observed when different operators digitize the same objects. This inter-user error is generally high, preventing data digitized by different operators from being used together.

The use of automated digitization by AI is still in its infancy and requires a clear answer to a few questions. Two of them are:

- Do different AI-based software produce differences when digitizing the same objects?
- Is AI-based software built for species A effective for digitizing species B?

We present a first analysis which provides elements of answer to these two questions. Our methodology involves the repeated digitization of a diverse set of landmarks on multiple species by both AI algorithms and human experts, measuring precision and inter-user variability.

Mitochondrial variation in *Anopheles gambiae* and *An. coluzzii*: phylogeographic legacy and associations with metabolic resistance to pathogens and insecticides

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Mitochondrial DNA (mtDNA) has been a popular marker in phylogeography, phylogeny, and molecular ecology, but its complex evolution is increasingly recognized. Here, we investigated mtDNA variation in *An. gambiae* and *An. coluzzii*, in perspective with other species in the *Anopheles gambiae* complex (AGC), by assembling the mitogenomes of 1219 mosquitoes across Africa. The mtDNA phylogeny of the AGC was consistent with a previously reported highly reticulated evolutionary history, revealing important discordances with the species tree. The three most widespread species (*An. gambiae*, *An. coluzzii*, *An. arabiensis*), known for extensive historical introgression, could not be discriminated based on mitogenomes. Furthermore, a monophyletic clustering of the three salt-water tolerant species (*An. merus*, *An. melas*, *An. bwambae*) in the AGC also suggested that introgression and possibly selection shaped mtDNA evolution. MtDNA variation in *An. gambiae* and *An. coluzzii* across Africa revealed significant partitioning among populations and species. A peculiar mtDNA lineage found predominantly in *An. coluzzii* and in the hybrid taxon of the African “far-west” exhibited divergence comparable to the inter-species divergence in the AGC, with a geographic distribution matching closely *An. coluzzii*'s geographic range. This phylogeographic relict of the *An. coluzzii* and *An. gambiae* split was associated with population and species structuration, but not with *Wolbachia* occurrence. The lineage was significantly associated with SNPs in the nuclear genome, particularly in genes associated with pathogen and insecticide resistance. These findings underline the mito-nuclear coevolution history and the role played by mitochondria in shaping metabolic responses to pathogens and insecticide in *Anopheles*.



S1-P09

How ecology and population history shape mosquito population genetics: a case study from the Caribbean

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Despite their medical and veterinary importance, little is known about population dynamics, structure and population history of mosquitoes, and how these relate to their ecology. The scarce information that is available comes from cosmopolitan (often disease-transmitting) species, while precious little information is available on most other species. Therefore, this study aimed to unravel how population genetics of different mosquito species relate to the ecology and population history, using a representative subset of the mosquito biodiversity of the Caribbean islands of Aruba, Curaçao, and Bonaire as a case study.

Mitochondrial COII sequences were obtained from 258 mosquitoes belonging to six species, which were collected during a recent a biodiversity survey. Sequences were used in phylogenetic analysis and haplotype network analysis to assess the variation in mosquito population genetics of six ecologically diverse species, varying in both population history and ecology. In addition we employed high-resolution (10x10m) Maxent ecological niche models to assess mosquito distribution patterns and link these to underlying environmental preferences.

Both the genetic diversity and population genetic structure were found to differ strongly between sets of species, leading to a subdivision into three species groups: i) introduced species with low genetic diversity; ii) native species with high genetic diversity and closely related haplotypes; iii) native species with high genetic diversity and locally restricted haplotypes. This subdivision is highly linked to the respective ecologies and population histories of the groups of species.

Overall these results clearly show that population genetics differ profoundly between introduced and native species. Furthermore, the results suggest that native populations may display distinct population genetic structures depending on their respective ecologies, likely related to variation in dispersal capacity and availability of breeding habitat. We hypothesize that similar contrasts in mosquito population genetics along historical and ecological axes may be present worldwide.

Incorrect DNA sequence entries in data bases and their consequences, exemplified for *Culicoides* biting midges (Diptera: Ceratopogonidae)

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Concomitantly with the loss of taxonomic expertise, most scientists relying on the identification of species, particularly small ones that can morphologically only be examined with considerable effort, make use of DNA sequence alignment with databases such as GenBank. These, however, have to be fed with information before data can be extracted. Since sequence data are not critically checked by an authority when being entered, the databases abound with erroneous data and data incorrectly assigned to their taxa.

While phylogenetically analysing DNA sequence data of *Culicoides* biting midges (Diptera: Ceratopogonidae), we came across numerous cytochrome c oxidase subunit I (COI) sequences in GenBank incorrectly assigned to a species or assigned to a taxon name (or synonym) not valid anymore. When referenced to consensus sequences generated from all available GenBank COI sequences of *Culicoides* species, such sequences matched with the consensus sequence of the species they had been assigned to to a much lesser extent than to other taxa. The incorrect assignment of the original specimens to a taxon is probably due to preceding incorrect morphological identification, sequence alignment with an incorrect dataset or the renaming of taxa.

In some cases, re-analysis of COI sequences from GenBank and correction of species assignment resulted in geographic distribution areas of a taxon other than described. Additionally, these data questioned the phylogenetic status of some closely related species and genetic variants whose role in disease agent transmission is not yet clarified.

Incorrect data and species identification cannot only spoil scientific analyses but have practical consequences. In the case of vectors, such as *Culicoides* species, control approaches must be designed according to spatiotemporal distribution, habitat and ecological requirements of the target species. It is therefore a challenge and a responsibility of researchers both to enter data into databases and to critically use them for scientific analysis.

S1-P11

Comparative analysis of the mitochondrial full genome of Central European *Culicoides* species (Diptera: Ceratopogonidae)

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Biting midges of the genus *Culicoides* (Diptera: Ceratopogonidae) play an important economic role in Europe as vectors of animal pathogens. In particular, species of the *Obsoletus* and *Pulicaris* Groups have been identified as significant vectors of bluetongue and Schmallenberg viruses.

For the reliable species identification of biting midge females within a group, morphological characters are not existent or distinct enough. Molecular biological methods, such as PCR and/or DNA sequence analysis of genetic markers, facilitate the differentiation between many, but not all, described species and their genetic variants (haplotypes). However, genetic differences within and between those vary considerably, making species statuses questionable or suggesting genetic variants to be true species. In general, phylogenetic relationships and systematic assignments of *Culicoides* species and haplotypes are often unclear. In addition, sequences in databases that could serve as references are often incorrectly assigned to taxa. To identify markers better suited for genetic species identification and clarify phylogenetic relatedness, comparative analyses of the mitochondrial full genome of important Central European *Culicoides* species have been initiated. So far, 11 full genomes of species of the *Obsoletus* Group, one full genome of a *Pulicaris* Group species and six full genomes of *Culicoides* species not belonging to one of these two groups could be generated. For annotation and analysis, only a single *Culicoides* mtDNA sequence (*C. arakawae*) is available in GenBank, but mtDNA sequences of other nematocerans may be helpful. While all *Culicoides* species analyzed so far possess an AT-rich region, which is characterized by inverted repeats in the *Obsoletus* and *Pulicaris* Groups, a second AT-rich region with inverted repeats was registered in the mitochondrial genomes of all *Obsoletus* Group taxa. The second AT-rich region does not occur in *C. dewulfi*, confirming its systematic demarcation from the group. Analysis is ongoing and promises to solve some systematic puzzles.

Unlocking sand fly biodiversity in Nepal: Enhancing disease control through DNA-barcoding

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Sand fly-borne diseases have a significant impact on global public health, although they are typically overlooked. In the Indian subcontinent, particularly in Nepal, India, and Bangladesh, over 46 species within the genera *Phlebotomus* and *Sergentomyia* are reported, of which only few *Phlebotomus* species play a key role in transmitting the protozoan parasite *Leishmania* spp. causing diseases such as visceral leishmaniasis (VL) and cutaneous leishmaniasis (CL). The role of *Phlebotomus* (*Euphlebotomus*) *argentipes* as a primary vector for *L. donovani* and the presence of other potential vector species in Nepal highlight the need for accurately identifying and monitoring sand fly species in order to create targeted disease control methods. This study used both morphological and molecular DNA barcoding techniques to examine the genetic diversity of Phlebotomine sand flies across Nepal, in order to support the national VL elimination program—a critical public health initiative given the disease's fatality rate, which is over 95% in untreated cases. A total of 315 sand fly specimens from six districts in Nepal were analyzed, targeting the mtDNA-COI gene, together with available data from BOLD and GenBank. Inter and intraspecies genetic divergence was calculated to check for diversity. The identification success rate using DNA barcoding was 97%, with interspecific genetic divergence ranging from 12.2% (*S. babu* and *S. punjabensis*) to 23.5% (*P. argentipes* and *Sergentomyia sp1*). For the genus *Phlebotomus*, including the vector species, the overall divergence averaged 14.9%. The lowest intraspecific genetic divergence was observed in *P. transcaucasicus* (0.1%), and the highest in *P. major* (5.9%). The results confirm that the COI barcoding method is an effective tool for identifying sand fly species in Nepal, providing critical data for the VL disease elimination initiative and enhancing global understanding and surveillance of sand fly-borne diseases.

S1-P13

Analysis of the genetic structure of *Haemagogus leucocelaenus* (Diptera: Culicidae), yellow fever vector, using the *Cytochrome Oxidase I* gene

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Haemagogus leucocelaenus is considered primary vector of yellow fever in forest areas of Brazil, and its wide geographical distribution may lead to genetic variations between populations. This study aimed to characterize *Hg. leucocelaenus* populations (Diptera: Culicidae) using molecular techniques. Samples were collected in fragments of the Atlantic rainforest and Cerrado (Tropical savanna), as well as in a transition zone between these two Brazilian biomes. Samples were processed to obtain sequences of the mitochondrial gene *Cytochrome Oxidase I* (DNA barcoding). DnaSP v. 4.0, NETWORK v. 4.6 and Structure v. 2.3 were used to analyze genetic diversity and population structure. The phylogenetic analysis revealed the existence of two distinct monophyletic clades: one composed of samples from the Cerrado and the transition Area, and the other composed of samples from the Atlantic rainforest. High levels of interpopulation divergence were observed between the Atlantic rainforest population with the other biomes, *Fst* values of 0.7830 (Cerrado) and 0.7513 (transition area). On the other hand, Cerrado and transition area populations showed an *Fst* value of 0.0011, indicating that geographical distance and environmental differences did not significantly modify the ancestral relationship between them. Nine haplotypes were identified in the populations analyzed, five in the Atlantic rainforest population and the remaining four shared between the two other areas. The genetic distance between the populations of *Hg. leucocelaenus* was calculated using the Kimura-2-parameter method, revealing values that ranged from 0.08% to 2.0%. The genetic similarities suggest that, from an evolutionary point of view, the Cerrado and transition area populations are more closely related to each other than to the Atlantic rainforest population. This genetic connection indicates a common ancestry and implies results that point to a geographical isolation of the Atlantic rainforest population when compared to the others.



S1-P14

Non-destructive extraction of DNA in ticks: a method to keep the shape!

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“My DNA? At the price of my good shape? No way” it is what are thinking all of the ticks when we ask them for more information about their genetic and morphology. Even old specimens from Museum collection (the alcohol-preserved ticks), want to keep their integrity. That is why, to help researchers in their study and to preserve morphology (and mood) of these specimens, non-destructive DNA extraction protocol was applied on *Ixodes ricinus* (larvae, nymphs and adults -female and male) conserved since 2007 in alcohol 70%. The method can extract on average of 69ng of DNA (in a final volume of 30 µl). 16S fragments were successfully amplified and sequenced for all the stages. Photo shoot under stereomicroscope for all of the stages, reassures ticks and researchers that ticks keep there chitinous exoskeleton intact but clarified. Using the outline-based geometric morphometry approach, the contour of the first coxa of larvae, nymphs and adults, a diagnostic character frequently used to discriminate different species, was investigated. Our result shows that the first coxa is not altered by the DNA extraction. The method is fast, cheap. No body shaming for ticks: the non-destructive DNA extraction preserves the ticks and their morphological characteristics for future reference. It is a valuable aid for integrative taxonomy of ticks.



S1-P15

Faithful tenants: the subgenus *Microculex* (Diptera: Culicidae) and its predilection for bromeliad phytotelms

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The subgenus *Microculex* of the genus *Culex* (Diptera: Culicidae) has an almost exclusive preference for bromeliad tanks, where the larvae develop amidst the organic matter present in the water. This study aims to survey the fauna of mosquitoes of the subgenus *Microculex* in bromeliads in the Atlantic Forest Protection Area on the urban perimeter of the city of Aracaju-SE, Brazil. Collections were carried out bi-monthly from June to September 2022, covering 11 bromeliad clusters, four of which were on the ground and seven epiphytes. The samples collected were taken to the laboratory at Federal University of Sergipe for rearing and associated mounting of the larval and pupal exuviae, adults and male genitalia. The subgenus *Microculex* had a relative abundance of 40% compared to the other species. To date, 296 immatures belonging to this subgenus have been identified, covering a richness of six species. Preliminary results indicate the highest abundance of *Cx. (Mcx) pleuristriatus* (76,0%), followed by *Cx. (Mcx) aphyllactus* (17,9%), *Cx. (Mcx) albipes* (2,0%), *Cx. (Mcx) inimitabilis* (1,7%), *Cx. (Mcx) davisii* (1,4%) e *Cx. (Mcx) microphyllus* (0,7%). *Culex (Mcx) pleuristriatus* e *Cx. (Mcx) aphyllactus* showed greater constancy among the species, occurring in 100% and 66.7% of the collections, respectively. In Atlantic Forest regions, *Cx. (Mcx)* can make up to 80% of the Culicidae fauna in bromeliad tanks, being commonly associated with preserved environments, although some species show adaptive potential to anthropized regions. Currently, none *Microculex* has been incriminated as a vector of pathogens for humans. However, due to the scarcity of information resulting from its taxonomic complexity and the ecological aspects involved, it is important to conduct studies to investigate the species present in bromeliads, as well as monitoring artificial containers, especially in impacted areas, since studies indicate a reduction in richness and strong dominance of one species, which demonstrates synanthropic potential.



Mosquito species identification with geometric morphometrics based on wing images captured with different devices

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Accurate species identification is crucial to assess the medical and veterinary relevance of a mosquito specimen, but it requires high experience of the observers and well-equipped laboratories. Herein, we would like to present the use of geometric morphometrics to distinguish invasive, morphologically similar *Aedes* species based on wing images. Thereby, we compared the use of images captured with two different devices: a professional stereomicroscope (Olympus SZ61, Olympus, Tokyo, Japan) and a macro lens (Apexel-25MXH, Apexel, Shenzhen, China) attached to a smartphone.

The right wings of 670 female specimens covering 184 *Ae. aegypti*, 156 *Ae. albopictus*, 166 *Ae. japonicus japonicus* and 164 *Ae. koreicus*, were removed, mounted and photographed with the two devices. Subsequently, the coordinates of 18 landmarks on the vein crosses were digitalised by a single observer for each image. The resulting two-dimensional landmark coordinates were superimposed to produce size-free wing shape variables, which provide the basis to statistically compare the shape variation between the mosquito species.

Species classification conducted using linear discriminant analysis was evaluated through a leave-one-out cross validation approach, demonstrating an overall reclassification accuracy of 94% based on microscope images and 93% based on smartphone images. *Aedes aegypti* and *Ae. albopictus* could be identified with an accuracy greater 96%, while the species identification accuracy was slightly lower for *Ae. japonicus japonicus* and *Ae. koreicus* at approximately 90%.

Our results show, that geometric wing morphometrics provides a reliable method to distinguish the most common exotic *Aedes* species in Europe. Thereby, the image quality obtained by smartphones equipped with a macro lens is sufficient for the use of geometric morphometrics and can provide a cost-effective alternative to professional microscopes. Further research is planned to contrast the performance of different classification methods (e.g. support vector machine or XGboost) and to assess the degree of observer error during landmark collection.

S1-P17

Wing Interferential Patterns (WIP) in Medically Important Dipteran Insects for Automatic Classification Using Deep Learning

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Blood-sucking insects such as mosquitoes, ticks, and sandflies are vectors for various pathogens causing diseases like arboviroses, malaria, and Lyme disease. Climate change, global economic growth, migration, and increased trade significantly affect the distribution of these insects. For instance, the expansion of *Aedes albopictus*, a vector for Zika, chikungunya, and dengue, highlights the need for accurate species identification during surveys to mitigate health risks. Traditional morphological methods are often inadequate for damaged samples or extensive surveys, while biological protocols are costly and unsuitable for field analysis. Wing Interferential Patterns (WIPs) have emerged as a promising method for species identification, as these colored patterns on insect wings vary significantly among species.

This study utilized the ARIM collection from the Institut de Recherche pour le Développement (IRD), consisting of over 100,000 insect specimens. Insect wings were dissected, placed on glass slides, and imaged using a Keyence™ VHX 1000 microscope. The resulting database contains 5,516 images of seven families (Culicidae, Calliphoridae, Muscidae, Glossinidae, Tabanidae, Ceratopogonidae, Psychodidae) and 21 genera. A Convolutional Neural Network (CNN) approach was developed for Diptera species classification. The CNN extracts hierarchical features through convolutional layers and classifies them using fully-connected and softmax layers. The confusion matrix showed minimal misclassification, with an overall accuracy of 95.5%, demonstrating the high taxonomic value of WIP imaging. Specific accuracies were: Anopheles 97.0%, Glossina 97.5%, Culex 97.0%, Aedes 97.1%, and Phlebotomus 96.2%. This study confirms the efficiency and semantic power of CNNs in identifying insect species using WIP imaging, providing a robust and rapid method for species identification essential for addressing the threats posed by emerging vector-borne diseases.

A simple key to identify *Aedes albopictus* larvae to the naked eye in the field in Western Europe

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Introduction and Objectives:

Aedes albopictus is a major vector of arboviruses in urban environments. The larvae develop in a wide variety of artificial containers. Mosquito control operators (MCO) have to assess vector population presence and abundance in these artificial breeding sites. A rapid method, initially developed for marsh mosquitoes, was adapted to study abundance of container mosquito in the field: The abacus method, based on a visual comparison between the abundance of larvae placed in a standardized container and reference images of abundance classes. This method is based on the ability of an agent to rapidly identify larvae of the target species with the naked eye. We developed an identification key of *Aedes albopictus* larvae a) without need for stereomicroscope and b) country-dependent and therefore evaluated it with various audiences.

Materials and Methods:

The identification key was developed using color differences, at individual scale, between parts of the larvae, especially respiratory siphon and head, among mosquito species with a wide cylindrical siphon (genus *Aedes* and *Coquillettidia*). Living larvae from artificial breeding sites, belonging to species found in France (*Aedes albopictus*, *Ae. geniculatus*, *Ae. japonicus*, *Culiseta longiareolata*...) were collected and color differences were analyzed. The key was tested with a varied audience who were challenged to identify live or photographed larvae.

Results, Discussion and Conclusion:

Aedes albopictus is the only species in France to present fourth instar larvae (L4) with a darker respiratory siphon than the head (in >98% of specimens), allowing a rapid differentiation with all instar larvae of *Culiseta longiareolata* (<2%). Coloration of abdominal segment permits to differentiate L4 of *Ae. albopictus* and *Ae. geniculatus*. This key permit easy identifications of larvae by non-specialist audience with few errors. It can be used in the field by MCO or in participatory science systems to identify *Ae. albopictus* larvae.



S1-P19

What the mitogenomes of 27 European mosquito species tell about the discriminative power of the different mitochondrial genes

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In recent decades, entomologists have significantly expanded their genomic toolkits, deepening our understanding of insect evolution, population dynamics, species identification, and monitoring practices. Mitochondrial genes have emerged as ideal markers for showcasing inter- and intra-specific variation. Particularly, the cytochrome oxidase subunit I (COI) barcoding region has been embraced by the mosquito research community for species identification. However, this gene often lacks discriminative power, particularly between certain cryptic and sibling species, does not allow for research on intraspecific variation, and poses challenges for primer design. The strong focus on COI has resulted in a lack of references for other genes, limiting researchers' potential to select a gene better suited to their application.

We sequenced and assembled 83 mitochondrial genomes of 27 European mosquito species to better understand the variability of different genes across taxonomic levels. Among the 13 protein-coding genes (PCGs), two ribosomal RNA (rRNA) genes, and 22 transfer RNA (tRNA) genes, we identified several genes with potential use for species identification. Particularly, ND6 stood out, exhibiting within-species similarity comparable to COI but showing greater variability between species. The variability is particularly high in *Anopheles* and *Culiseta*. This gene therefore presents a promising candidate for identifying closely related species. This detailed and comparative understanding of the evolutionary dynamics of different mitochondrial genes aids researchers in selecting a gene that better fits their purpose. Additionally, 21 of the sequenced species were sequenced for the first time, forming a unique library for various genomic studies, including primer development.

Interspecific larval competition between invasive *Aedes albopictus* and established *Culex pipiens* mosquitoes from the Black Sea Region of Turkey

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Mosquitoes often experience intraspecific and interspecific competition among larvae attributable to nutrient limitation and crowded larval conditions, especially container mosquitoes including *Aedes albopictus* and *Culex pipiens*. Density-dependent effects on larvae influence recruitment of adults and their life history traits, including those traits relevant to their ability to transmit arboviruses. The application of insecticides may alleviate density-dependent competition and differential species response to insecticides may modify interspecific competition. We tested the hypothesis that exposure to insecticide pyriproxyfen alters interspecific resource competition between *Ae. albopictus* and *Cx. pipiens* derived from the Black Sea Region in Turkey. The progeny of field-collected *Ae. albopictus* and *Cx. pipiens* from Turkey were used in studies to assess competitive interactions between the two species. Pyriproxyfen and control treatments were crossed with six different density combinations of *Ae. albopictus* and *Cx. pipiens*, respectively (0:30, 0:60, 15:15, 30:30, 0:60, 0:30). The experiments were conducted in plastic containers with 0.1 g fish food, 1.0 liter of tap water, and 1 ml of microbial inoculum. The containers were maintained in an insectary at 25°C and a 14:10 h light:dark photoperiod. Pyriproxyfen treatment containers received a low dose of pyriproxyfen, whereas control containers received only tap water. Mosquito performance measurements showed negative density-dependent effects with similar effects of intra- and interspecific interactions. *Culex pipiens* were more negatively affected by competitive treatments than *Ae. albopictus*. Development times and sizes of adult females were similar or negatively affected by high density and control conditions. *Culex pipiens* survivorship to adulthood was negatively affected by increasing densities of *Ae. albopictus*. High density conditions experienced during the larval stage yielded adult females with 30-50% reduction in lifespan for both mosquito species. We found little support to suggest that pyriproxyfen alters competitive interactions between *Ae. albopictus* and *Cx. pipiens*.

Understanding the behavior of *Aedes albopictus*: Zika Infection and Locomotor Activity in Laboratory Conditions

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Aedes (Stegomyia) albopictus is a globally significant mosquito vector of arboviruses, particularly in temperate countries such as Italy, France and Spain and potentially Brazil. This species is considered a diurnal/crepuscular mosquito, with daily activities concentrated in the early and late hours of the day. However, it is possible that factors like viral infection can alter this vector's activity pattern. This study aimed to investigate the effect of Zika virus (ZIKV) infection on the locomotor activity of *Ae. albopictus* under laboratory conditions, using two infection methods. Eight-to twelve-day-old females of *Ae. albopictus* were infected via intrathoracic injection or blood feeding with ZIKV. Both methods were triplicated. Post-infection, females were individually placed in glass tubes with sugar solution-soaked cotton at the ends. Locomotor activity was continuously monitored for 8 days in activity monitors within an incubator. Data were analyzed considering total, diurnal, and nocturnal activity means. Both the control and infected groups exhibited a diurnal and bimodal activity pattern, with peaks at the beginning and more pronounced at the end of the light period. Statistical analyses showed no significant differences in locomotor activity means between control and infected groups for both infection methods. This pioneering study on the locomotor activity of ZIKV-infected *Ae. albopictus* using different infection methods found no evidence that Zika virus infection alters the locomotor activity of this mosquito under laboratory conditions. These results suggest the reason why *Ae. albopictus* is a vector of arboviruses in some European countries, but considered a potential vector in American countries, may be complex, involving biological and ecological factors.



S2-P03

Blood-fed mosquitoes as 'flying syringes' in a field setting

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Introduction and Objectives

Mosquitoes are well known for their role in vector-borne disease transmission, but blood-fed mosquitoes could represent an effective sampling tool to monitor non-vector-borne pathogens. We aimed to evaluate whether blood-fed mosquitoes can be used as a tool for monitoring pathogens in wild and domesticated animals in different habitats in the Netherlands. Previous studies showed the potential of fermenting molasses in capturing blood-fed mosquitoes. Hence we tested fermenting molasses and derivatives to catch blood-fed mosquitoes in the field. To determine the availability of potential hosts, we examined the diversity of vertebrate species present at the field sites, using airborne eDNA sampling.

Materials and Methods

We tested the importance of CO₂ and the bioactive compounds of fermenting molasses to catch blood-fed mosquitoes at farm, wetland and peri-urban habitats in the Netherlands. DNA and RNA were extracted from blood-fed mosquitoes. Host and bacteria species were identified using DNA metabarcoding and sequencing. Virus species were identified using SISPA on DNA and RNA, followed by probe capturing and sequencing. Animal species that were present in the mosquitoes' environment were identified by metabarcoding and sequencing of airborne eDNA captured by air filters.

Results, Discussion and Conclusion

Non-vector-borne pathogens were identified in blood-fed mosquitoes, but the number of collected blood-fed mosquitoes was limited. From the different mosquito species, different host species were identified. Targeting different mosquito species may thus result in targeting different animal populations. Based on the eDNA samples, the sampling locations were similar in animal composition, although, the number of animals per species and the sampling range of these air filters remain unknown. Acknowledging these limitations, airborne eDNA sampling represent a promising tool to screen the host availability at a location. We conclude that blood-fed mosquitoes can be used to screen for non-vector-borne pathogens, but optimisation of sample collection is required.

Effect of ABO blood group on the bionomics of *Aedes albopictus* under laboratory conditions

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Aedes albopictus is an invasive mosquito species that has spread to many regions worldwide. The public health concern of this species relies on its capability to transmit several viruses, including dengue, chikungunya and Zika. In Spain, this species is well established in many cities, especially in the Mediterranean Basin. Several studies conducted in field or laboratory conditions demonstrated that *Ae. albopictus* is anthropophilic but also bite other mammal hosts. In this study, the effect of the blood group on the bionomy of *Ae. albopictus* was tested. For this, eggs from *Ae. albopictus* obtained from wild populations and a laboratory colony were used to rear the species under laboratory conditions. Obtained females were fed with human blood from groups A+, B+, AB+ and O+ using the Hemotek. Four variables were measured: percentage of *Ae. albopictus* blood-fed females, oviposition, adult lifespan, and eggs viability. Results showed that the percentage of blood-feeding was higher in *Ae. albopictus* obtained from the colony than the wild ones, however, none of the colonies showed any preference for a specific blood type. Females fed with B+ and O+ groups laid a significantly higher number of eggs. In addition, the viability of eggs was significantly higher from colony females that fed with B+ and O+ groups. Therefore, females that bite humans from those groups may lay higher number of viable eggs compared to other blood types. Further studies using negative Rh blood must be performed to better clarify the blood preferences of *Ae. albopictus*. Finally, our study contributes to the knowledge and learning of mosquito rearing techniques in the laboratory to improve efficiency and productivity.



S2-P05

How do host contaminants affect vector fitness ? An experimental study of *Ornithodoros* ticks and mercury-contaminated host blood

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Mercury pollution is ubiquitous and known for its high toxicity. This contaminant bioaccumulates through the trophic chain, and can pose a significant threat for organisms once levels reach specific thresholds. This pollution is especially problematic in aquatic environments, where mercury is methylated by aquatic microorganisms into methylmercury MeHg, its most dangerous form. Although much work has focused on the impacts of MeHg on vertebrate organisms, little is known about its effects on the organisms that live in association with these species, and in particular, their parasites. Our study experimentally investigated the effects of methylmercury (MeHg) contamination of host blood on the development and reproduction of soft ticks (Family Argasidae ; genus *Ornithodoros*). We considered two species, *Ornithodoros (Alectorobius) maritimus* collected from natural populations of its seabird host – *Larus michahellis*, and a laboratory population of *O. moubata*. Ticks were artificially fed with blood containing five different methylmercury concentrations, defined from field based levels present in *L. michahellis* (0, 0.76 µl/ml, 3.8 µl/ml, 7.6 µl/ml, 38 µl/ml). At the highest concentration, juvenile ticks were unable to moult and died within a few days. No other effects at the other concentrations were found after a single bloodmeal, suggesting that ticks may either have high tolerance to this contaminant or are able to excrete it during blood digestion. More detailed and longer term studies are now required to evaluate more subtle impacts of MeHg contamination on tick life history traits, and the potential consequences this may have for the transmission dynamics of tick-borne disease.



S2-P06

On the Move: Tracking Passive Dispersal and Survival of *Aedes albopictus* in North-Eastern Spain

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Global trade and travel have accelerated the spread of invasive mosquito vectors carrying disease-causing pathogens. In Europe, invasive *Aedes* species, including *Ae. albopictus*, have arrived through various pathways such as international trade and natural dispersal. *Ae. albopictus*, capable of adapting to diverse environments and overwintering as diapause eggs, has rapidly spread across continents, facilitated by its ability to survive transportation over long distances. While active dispersal of *Ae. albopictus* is limited, its spread correlates with global shipping routes and road networks. However, research on passive dispersal, particularly via vehicles, remains limited. Our study aims to quantify *Ae. albopictus* survival within private vehicles and identify influencing factors. To perform the study, we introduce three *Ae. albopictus* females in individual containers with different feeding regimes (i.e., fasting, sugar-fed and blood-fed) in vehicles from volunteers, from March to September 2024. We also introduce in each car a datalogger with an ID connected to each volunteer's car, to monitor humidity and temperature. Volunteers provide us the information of their movements with the car through the Space Mapper app, a generic research app for human mobility projects which brings the possibility to share their positioning data (at GPS resolution). This allows us to measure the kilometres travelled in the car during the experiment and associate them to specific features of the car and driving usual conditions through an anonymized ID that the same app provides for each participant. Our findings allow us to quantify the distance travelled and in-vehicle survival of laboratory-reared females introduced into volunteer vehicles for the first time. These results can enrich future invasion and transmission models, helping to predict establishment in neighbouring areas.

Diversity of biting midges, mosquitoes and sand flies at four dog shelters in rural and peri-urban areas of Central Morocco

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Introduction and Objectives

Blood feeding arthropods are involved in the transmission of several pathogens that have a major impact on public health. This study focused on entomological investigations highlighting the composition, abundance and diversity of flying hematophagous arthropods at four dog shelters (DS) located in central Morocco with the aim of discussing their vectorial roles and assessing the risk of these shelters as foci for zoonotic diseases.

Material and Methods

Culicoides, mosquito and sandflies species composition was investigated in four DS (Sidi Yahya Zaer, Bouskoura, Fes and Khouribga) from 1 April to 30 November 2022. *Culicoides* were collected using OVI (Onderstepoort Veterinary Institute) traps. Mosquitoes were collected using BG-Pro mosquito traps combined with a CO₂ source as an attractant. Sandflies were sampled using castor oil on sticky papers (20 × 20 cm) disposed on walls inside the different DS. The traps and sticky papers were functional from 6 pm to 10 am on two consecutive days every two weeks.

Results

2,321 biting midges, 575 mosquitoes and 475 sand flies were collected. Fourteen *Culicoides* species were recorded, with *Culicoides imicola* (55.96%), *Culicoides paolae* (16.07%), *Culicoides circumscriptus* (10.29%), and *Culicoides newsteadi* (5.77%) as the most abundant species, whereas three mosquito species were collected including *Culex pipiens* (96.84%), *Culiseta longiareolata* (2.80%), and *Culex perexiguus* (0.36%). Ten sand flies species were collected, i.e. seven *Phlebotomus* species (62.70%) and three *Sergentomyia* species (37.30%). *Sergentomyia minuta* was the most dominant species (34.31%), followed by *Phlebotomus sergenti* (32.42%), *Phlebotomus perniciosus* (8.63%), *Phlebotomus alexandri* (6.94%), and *Phlebotomus riouxi* (6.52%).

Discussion and Conclusion

within these findings, the coexistence of several vectors in these study areas indicates the potential circulation of a wide range of pathogens. Indeed, a study is underway on carried pathogens within the collected vectors (stored at -80°) and in dogs from the four DS (n=234).

How old are you? Assessing *Aedes albopictus* age by mid-infrared spectroscopy

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Introduction and Objectives

The age structure and dynamics of mosquito populations are crucial for understanding their ability to spread diseases and assessing the effectiveness of anti-mosquito control measures. Available methods for estimating mosquito age are labour-intensive and imprecise. We developed a promising Supervised Machine Learning (SML) algorithm based on Mid-Infrared Spectrometry (MIRS) of semi-field reared *Aedes albopictus* specimens to predict the age of single adult females and males. We here present data on the validation of the accuracy of this approach on field samples and preliminary results on its application to assess the effectiveness of conventional and innovative control interventions.

Materials and Methods

Mid-Infrared spectra from 1,881 semi-field reared adults of known age were used to train the SLM algorithm and assess its accuracy when predicting mosquito age, either as a multilevel variable (with a resolution of 3, 6, and 9 day-long classes) or as a binary variable of epidemiological relevance (e.g. young vs older). The accuracy of the approach will be validated on field samples aged by morphological approaches. Samples collected in summer 2024 before and after pyrethroid treatments in Rome and during a SIT trial in Procida Island (Naples) will be processed by MIRS.

Results, Discussion and Conclusion

Results from semi-field females showed that the mean accuracy of SML-algorithm for 3-day age is 85% (78%-90%) for specimens <15-day old and 71% (52%-83%) for older ages. Mean accuracy increased to 85% (77-100%) when grouping females into 6-day long classes and to >96% when young (>3-day old) and older (>6-day old) females were compared. This high accuracy will be validated in the field by analysing the spectra of morphologically distinguished nulliparous and parous females. Results of collections carried out before and after anti-mosquito interventions will provide preliminary indications of the potential of MIRS approach in assessing their effectiveness.

Role of microhabitat diversity and interspecies competition in the study of Culicidae larvae interactions

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Introduction and Objectives

Vector Borne Diseases (VBDs) represent a health scourge; their spread is influenced by vector's presence and circulation, but also by its interactions with host and pathogen in the environment; therefore, investigating on direct and indirect effects on biodiversity in ecosystems helps to develop more effective preventive measures. The aim of the work is to study the inter- and intraspecific relationships between the larval stages of native species (*Culex pipiens*) and invasive ones (*Aedes albopictus*) settled by colonizing the same environments.

Materials and Methods

Breeding sites were sampled for collecting eggs, larvae and adult mosquitoes to set up colonies of both studied species, creating artificial habitats to have specimens at any stage of biological cycle. All tests were conducted in our Entomology Laboratory under controlled conditions of humidity, temperature and photoperiod. Adult mosquitoes were reared and fed sugary substances or animal blood, artificially supplied through Haemotek. Feeding plans were created for larvae, changing the matrix and doses, including fasting.

Results, Discussion and Conclusions

Interspecific dynamics of larval stages were observed by testing their adaptability as completion of the biological cycle and evaluating responses to microhabitat sharing by changing available space, food quantity and composition. Inter- and intraspecific competitiveness were assessed as percentage survival of final adults' number compared to the initial larvae' number. The collected data show that acting indirectly on the biological cycle of culicids is interesting for observing their adaptive abilities and it provides clearer indications of the most favorable conditions for the establishment and proliferation of exotic culicids, increasing the gradual loss of biodiversity as well as the risk of circulation of VBD pathogens. The obtained results add information on the inter- and intraspecific dynamics of the studied species, thus enhancing entomological surveillance by providing fundamental notions for monitoring exotic vectors' introduction sites and increasing the predictive value of Arthropod Surveillance.



Tick-borne disease hazard in urban green spaces and their surrounding rural environment

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Introduction and Objectives

Urban greenspaces provide ecosystem services such as climate change mitigation and recreation. However, ticks and tick-borne diseases have been reported in urban greenspace across Europe, which could present the disservice of a public health hazard. Most studies investigate only urban centres where ticks are present, so the extent of the issue across large regions is unknown. Furthermore, few studies test the influence of the surrounding landscape on tick or tick-borne pathogen hazard inside towns. The objective of this study was to take a nationwide approach to characterize tick and Lyme disease hazard in 16 towns and three cities across the United Kingdom.

Methods

For each town, we surveyed questing *Ixodes ricinus* ticks and the *Borrelia burgdorferi* s.l. bacteria (causing Lyme disease) in five urban greenspaces and five sites in the surrounding rural environment. For each of the three cities, we also surveyed five suburban sites. Surveys were conducted between 2021 and 2023.

Results

We found ticks in 37.5% (6/16) towns and 75% (12/16) hinterlands, with fewest ticks across the central parts of England. The probability of ticks and Lyme disease hazard in urban greenspaces were significantly lower than in the surrounding countryside. Tick presence and Lyme disease hazard were positively correlated with the density of ticks and the percentage of woodland cover in the rural environment (between 5 and 10 km of the town centre) and negatively associated with the percentage of urban cover whilst we did not find any correlation with land cover metrics within 5 km of the town centre.

Discussion

These results highlight that recreating in urban greenspaces poses a lower hazard of ticks and Lyme disease than in rural woodlands and demonstrates tick presence and Lyme disease hazard in urban greenspaces are influenced by the hinterland context within which the town lies.



S2-P12

Expansion and adaptation of the *Hyalomma marginatum* tick in the Occitanie region

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The *Hyalomma marginatum* tick is endemic in several Mediterranean countries, from the Maghreb to the Iberian Peninsula and the eastern Mediterranean basin. Its range is gradually expanding towards northern Europe. Described since the 1950s in Corsica, this tick was reported in mainland France in 2016. It therefore seems that we are witnessing a biological invasion of this species in France. Such change in species distribution can be due to three main eco-evolutionary drivers: migration change, phenotypic change and environmental change. First, we studied the migration pattern of this ticks and the origin of this invasion as revealed by its current genetic structuring. A campaign to collect samples in France between 2016 and 2023 and collaborations with foreign partners provided us with more than 600 individuals of *H. marginatum*. Our phylogeography study using mitochondrial markers enabled us to define three differentiated genetic clusters in mainland France, indicating potentially different origins and therefore several discrete colonization events by this tick. These results will be confirmed using nuclear markers developed by sequence capture targeting ultraconserved elements designed in Arachnida. Second, phenotypic changes allowing successful establishment in the novel environment can be due to either to phenotypic plasticity or to evolutionary changes. These two hypotheses were tested with two different experiments: one with field ticks brought back to the laboratory, where life-history traits were compared between different French populations to see if there had been any local adaptation; the other with laboratory-bred ticks, which were placed into the field to see whether or not their life cycle was successful and how long this cycle lasted as a function of environmental conditions. The latter showed that *H. marginatum* can complete its cycle in a climate that differs other than that of the Mediterranean, and that the species is likely to extend its range still further.

What information can a longitudinal survey carried out in a single multi-host site provide on mosquito dynamics and their drivers in peri-urban Antananarivo, Madagascar?

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Madagascar is endemic to several diseases of public health importance such as malaria, dengue, chikungunya, Rift Valley fever (RVF), West Nile fever (WNV) or lymphatic filariasis. This study aims to assess, in a resource limited setting, what information a longitudinal entomological study carried out in a multi-host single-site can provide on mosquito population dynamics and transmission risk in the vicinity of Antananarivo, the capital city.

A fortnightly collection of mosquitoes using six CDC-light traps was set up between January 2017 to April 2018 in a multi-host farm in peri-urban area of Antananarivo. Multivariable generalized linear models were developed using indoor and outdoor densities of the predominant mosquito species as response variables and moon illumination, environmental and climatic data as the explanatory variables.

Overall, 46,737 mosquitoes belonging to at least 20 species were collected. *Culex antennatus* (68.9%), *Culex quinquefasciatus* (19.8%), *Culex poicilipes* (3.7%) and *Anopheles gambiae* s.l. (2.3%) were the most abundant species. Mosquito densities were observed to be driven by moon illumination and climatic factors interacting at different lag periods. The outdoor models demonstrated biweekly and seasonal patterns of mosquito densities, while the indoor models demonstrated only a seasonal pattern.

Results show that (1) an important diversity of mosquitoes exists in the peri-urban area of Antananarivo, (2) the four main species (>94% individuals) are major or potential vectors of viruses (WN and/or RVF viruses) and/or parasites (*Plasmodium*, *Wuchereria*) (3) despite a decline in winter of three of these species, they are present all year round. The main drivers of their abundance were temperature, relative humidity and precipitation, as well as, for outdoor abundance only, moon illumination. Identifying these drivers is a first step towards the development of pathogen transmission models (R0 models) which are key to inform public health stakeholders on the periods at risk of vector-borne diseases.

S2-P14

Harnessing mosquito symbionts for malaria transmission blocking

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A possible malaria control approach involves the dissemination in mosquitoes of inherited symbiotic microbes to block *Plasmodium* transmission. However, in the *Anopheles gambiae* complex, the primary African vectors of malaria, there are limited reports of inherited symbionts that impair transmission. We have established the SYMBIOVECTOR project to investigate the prospect of deploying a recently discovered *Anopheles* symbiont, *Microsporidia MB*, as a *Plasmodium* transmission blocking tool. The ability of *Microsporidia MB* to block *Plasmodium* transmission together with vertical transmission and avirulence makes it an excellent candidate for symbiont-based transmission blocking. We show that a vertically transmitted microsporidian symbiont (*Microsporidia MB*) in the *An. gambiae* complex can impair *Plasmodium* transmission. *Microsporidia MB* is present at moderate prevalence in geographically dispersed populations of *An. arabiensis* in Kenya, localized to the mosquito midgut and ovaries, and is not associated with significant reductions in adult host fecundity or survival. We investigated the mechanistic basis and efficiencies of *Microsporidia MB* transmission between *Anopheles arabiensis* mosquitoes. We show that *Microsporidia* can be transmitted both vertically (mother to offspring) and sexually between adult mosquitoes. The dynamics of spread and optimal dissemination strategies have been investigated under semi-field conditions and used to determine the likely outcomes of releasing *Microsporidia MB* in the field.

Deep Learning techniques to predict *Culex pipiens* suitable habitat in central Italy using Sentinel-2 data

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Climate and environmental conditions strongly influence the presence, distribution and dynamics of the vectors responsible for the spread of animal and human diseases. Understanding the distribution of adult mosquitoes can be an indication of disease risk.

Culex pipiens is a species capable to feeding on a wide variety of hosts and adapting to different environments, responsible for the transmission of vector-borne disease as West Nile, Usutu, Rift Valley fever and Japanese encephalitis.

In this research, Sentinel-2 Satellite Earth Observation data from the European Copernicus program were used to study the vector habitat at unprecedented spatial (20 meters), spectral (11 bands, corresponding to different wavelengths of the electromagnetic spectrum) and temporal (5-days) resolutions.

A dataset consisting of four years of mosquito field collection data (2019-2022) was coupled with satellite imagery (patches of 4480x4480 meters in the surroundings of each trap location, obtained through a Python script) around each site and for each satellite revisit time. Data were subject to a Deep Learning (DL) analysis through a multitemporal model: the DL model is trained with a sequence of local conditions occurring approximately in the two months (comprising a series of ten Sentinel-2 acquisitions) preceding the mosquito collection. We also evaluated 15 days of temporal lags between the multi-band EO datacube date of acquisition and the mosquito collection: this "empty time" is valuable when the model is used as a base for operational predictive tools in support of surveillance activities.

The model achieved an F1 score of 81.4% in correctly classifying the presence or absence of *Cx. pipiens* in central Italy, 15-days in advance. These findings will advance our ability to identify suitable times and areas for *Cx. pipiens* presence, optimising entomological surveillance activities and reducing high-risk exposure to VBDs within Italian landscapes.



A DNA-based approach to surveillance of *Culicoides* vectors and their vertebrate hosts in Ireland

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Introduction

Culicoides biting midges serve as vectors for Bluetongue (BTV) and Schmallenberg (SBV) viruses, which impact the well-being and productivity of livestock. Outbreaks of these viruses highlight the urgent need for precise and efficient vector surveillance. Determining the vertebrate hosts for these vectors is also crucial in understanding the potential transmission of BTV/SBV to susceptible hosts.

Objectives

To conduct DNA barcoding for species identification.

To develop species-specific qPCR assays for the rapid detection of *Culicoides* vectors in pooled samples.

To determine vertebrate hosts for *Culicoides* through blood meal analysis.

Materials and Methods

Adult *Culicoides* were captured using OVI traps at 13 livestock farm locations in Ireland from May 2022 to December 2023. Individual specimens were barcoded using PCR amplification and sequencing of cytochrome oxidase subunit 1 (CO1) and internal transcribed spacer (ITS) gene regions.

Real-time qPCR tests were designed to discern specific *Culicoides* species within pooled samples, focusing on vectors of BTV and SBV.

Sources of *Culicoides* blood meal were determined by PCR and sequencing of vertebrate CO1 gene region and by species-specific qPCR, providing a comparative approach to identify vertebrate hosts.

Results and Discussion

A total of 354 biting midges belonging to 12 species were barcoded from six sites (Co. Dublin, Co. Waterford, Co. Galway, Co. Laois, Co. Kilkenny, and Co. Cork).

qPCR assays were designed to detect vectors including *C. obsoletus*, *C. scoticus*, *C. chiopterus*, *C. dewulfi*, *C. pulicaris*, *C. lupicaris*, *C. punctatus*, and a specific assay for detecting *C. impunctatus* in pools of 100 midges.

Blood meal analysis of 21 *Culicoides* showed that 10 of these vectors obtained their blood meal from cattle.

Conclusion

The molecular techniques established in this research provide a valuable tool for the efficient surveillance of *Culicoides* vectors and their vertebrate hosts, essential for preventing and managing the transmission of viruses to livestock.



Unraveling the Impact of Plant-Based Diets on *Aedes albopictus* Mosquitoes and Arboviral Transmission in Occitanie

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Accumulating evidence suggests that the diet of mosquitoes, particularly those derived from plants, may significantly impact their biology. These meals can influence various aspects of mosquito fitness, such as survival and fecundity, as well as their vector competence for pathogens. However, the precise mechanisms underlying the plant-mediated effects on vector capacity to transmit pathogens remain unknown.

In light of global changes and the shifting spread of arboviruses in Europe, including the south of France, where autochthonous cases of chikungunya and dengue are on the rise, we hypothesize that specific plants growing in Occitanie (South of France) could influence the vector competence of *Aedes albopictus*.

To test this hypothesis, we exposed newly emerged *Ae. albopictus* females for 5 days to different plant species (*Lavandula angustifolia*, *Perovskia atriplicifolia*, *Scabiosa atropurpurea*), and to water and sucrose solution as controls. Females were then subsequently challenged with chikungunya (CHIKV) and dengue (DENV) viruses using oral experimental infection procedures. Females that ingested a complete infectious blood meal (composed of viruses and blood erythrocytes) were then incubated for 7 and 14 days post infection (dpi). At each time point post-infection, 20-30 per condition were analysed using qPCR to detect and quantify viral genomes in their bodies (abdomen+thorax) and heads, respectively, to approximate the infection (IR) and the dissemination (DR) rates as proxies of the vector competence. Our initial results showed significant effects of mosquito diet on the vector competence, depending on the time points (7 vs. 14 dpi), the virus (DENV vs. CHIKV) and the tissue (body vs. head), but will need to be repeated to confirm them. These preliminary results suggest that plant species in Occitanie might have an impact on the vector competence of *Ae. albopictus*, which could warrant further investigation before considering implications for managing local arboviral transmission.



S2-P18

Altitudinal distribution pattern of *Aedes (Stegomyia) albopictus* (Skuse, 1894) in Valencia province, Eastern Spain

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Introduction and Objectives

Aedes (Stegomyia) albopictus (Skuse, 1894) was detected for first time in the Valencian Autonomous Region (Eastern Spain) in 2005 at the province of Alicante. From that moment on, the Laboratory of Entomology and Pest Control of the University of Valencia began to study this species in the Valencian territory. Established tiger mosquito populations were recorded in 2013 from the municipality of Cullera, being the first record in the province of Valencia. Detection and surveillance tasks have subsequently continued both autonomously and through contracts with regional public institutions.

The objectives of this communication showcase the work carried out over 16 years and point out how the altitude and other associated factors negatively affect the establishment of this species.

Materials and Methods

A sampling of the preimaginal stages employing the dipping technique was conducted throughout Valencia province in all those potentially optimal places for the establishment and development of the life cycle of this mosquito, such as tree cavities, scuppers, ornamental fountains, graveyard vases, flowerpot holder plates and others ones.

Results, Discussion and Conclusion

The advance and colonization along the province of Valencia has been recorded year after year, and it is currently present in 244 of the total of 266 municipalities. It has been proven that the tiger mosquito is capable of reproducing in breeding sites located between 3 and 898 m a.s.l., but its capacity is reduced or limited at higher altitudes. In fact, its dispersion in high mountain municipalities located above 1100 m has not yet been recorded.

It is concluded that the elevation together with factors such as local climate, the small resident population, and the reduced trips and transportation carried out in mountain towns limit the capacity of dispersion and establishment of the tiger mosquito in this area of Spain.



Biodiversity and bioecology of Culicidae (Diptera, Nematocera) in rural, suburban and urban areas of Tizi-Ouzou (Algeria)

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Context/Purpose

The study of the biodiversity and bioecology of Culicidae in the region of Tizi-Ouzou is carried out during the period from December 2018 to May 2019.

Methods

The sampling of the pre-imaginal stages of Culicidae was carried out using the dipping technique, with ten dips in different parts of the breeding sites. fifteen stations belonging to three different rural, suburban and urban environments.

Results

Ten species were recorded: *Aedes caspius*, *Anopheles labranchiae*, *Anopheles claviger*, *Culex pipiens*, *Culex hortensis*, *Culex impudicus*, *Culex theileri*, *Culex perexiguus*, *Culex mimeticus* and *Culiseta longiareolata*. This study makes it possible to specify the distinctive morphological characteristics of the species collected. The use of ecological indices allowed us to determine that *Culex pipiens* is the most dominant species in the urban area of Tizi-Ouzou with 420 individuals and a frequency varying between 57% and 39% in the peri-urban stations. These environments represent the most favourable breeding sites for larva and their presence in the vicinity of urban agglomerations can compromise the quality of life of citizens. On the other hand, in the rural stations, *A. labranchiae*, a potential vector of the causal agent of malaria (*plasmodium phalciparom*), is the most dominant species with 147 individuals (55%). The monitoring of spatio-temporal variations in the larval effects of *C. pipiens* and *A. labranchiae* made it possible to show positive correlations between some physico-chemical parameters such as temperature, conductivity and organic matter

Conclusion

This study contributes to the understanding of the bioecology of the aquatic stages of mosquitoes, an essential prerequisite for achieving effective control.

Keywords: Biodiversity - Tizi-Ouzou - Culicidae - Ecological indices - Population dynamics.



Tiger mosquito blood-feeding behaviour and possible implications for disease transmission

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Introduction and Objectives

Determination of human-mosquito contact patterns and how bites are distributed among different people is of great importance for better understanding the transmission dynamics of mosquito-borne arboviruses and developing effective strategies to mitigate risk. Here, we used human DNA blood meal profiling of the Asian tiger mosquito (*Aedes albopictus*) to quantify its contact with human hosts and infer epidemiologic implications of its blood feeding behaviour.

Materials and Methods

During the course of two years (2021 and 2022) we collected 361 bloodfed tiger mosquitoes throughout Spain. In order to determine the number of different people bitten, bloodmeals were screened for a set of 16 human microsatellites commonly used for human fingerprinting.

Results, Discussion and Conclusion

We will report on the proportion of collected mosquitoes in which human DNA signal was found, as well as on the frequency of mosquitoes that contained single, double and triple bloodmeals, and use this information to make inferences on potential disease transmission. Preliminary results indicate that ~40% of mosquitoes in which human DNA signal was found had ingested a single blood meal, while ~60% contained double meals. No triple meals were detected so far. In Europe, *Ae. albopictus* is regarded as an epidemiologically important vector for the transmission of many human pathogens. Our results provide new details on tiger mosquito biting frequency distribution, which is crucial to achieve a correct understanding of the networks of interaction between hematophagous mosquitoes and their human hosts.

Broad host preference and host-feeding patterns of *Culex pipiens s.s./Cx. torrentium*

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Mosquito host-feeding patterns are an important factor of the species-specific vector capacity determining pathogen transmission routes. *Culex pipiens s.s./Cx. torrentium* are competent vectors of several arboviruses, such as West Nile virus and Usutu virus. However, studies on host-feeding patterns rarely differentiate the morphologically indistinguishable females. We analyzed the host-feeding preference of *Cx. pipiens* and *Cx. torrentium* in host-choice studies for bird, mouse, and a human lure. In addition, we summarized published and unpublished host-feeding patterns of field-collected specimens from Germany, Iran and Moldova from 2012-2022 genetically identified as *Cx. pipiens* biotype *pipiens*, *Cx. pipiens* biotype *molestus*, *Cx. pipiens* hybrid biotype *pipiens x molestus* and *Cx. torrentium* and finally put the data in context with similar data found in a systematic literature search. In the host-choice experiments, we did not find a significant preference for bird, mouse and human lure for *Cx. pipiens pipiens* and *Cx. torrentium*. Hosts of 992 field-collected specimens were identified for Germany, Iran and Moldova with the majority determined as *Cx. pipiens pipiens*, doubling the data available from studies known from the literature. All four *Culex pipiens s.s./Cx. torrentium* taxa had fed with significant proportions on birds, human, and non-human mammals. Merged with the data from the literature from 12 different studies showing a domination of blood meals from birds, more than 50% of the blood meals of all four taxa were identified as birds, while up to 41% were human and non-human mammalian hosts. However, there were considerable geographical differences in the host-feeding patterns. In the light of these results, the clear characterization of the *Culex pipiens s.s./Cx. torrentium* taxa as ornithophilic/-phagic or mammalophilic/-phagic needs to be reconsidered. Given their broad host ranges, all four *Culex* taxa could potentially serve as enzootic and bridge vectors.



S2-P22

Pyrethroid resistance profile of *Aedes aegypti* (Diptera: Culicidae) populations from Sergipe, Brazil

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Pyrethroid insecticides are widely marketed in Brazil to *Aedes aegypti* control. This group of insecticides acts directly on the sodium channels (Nav) of the vector's nervous system, producing a knockdown effect. However, some *Ae. aegypti* populations have mutations in Nav, which affect the effect of pyrethroids on the nervous system, leading to resistance (Kdr). The aim of this study is to analyze the pyrethroid insecticide resistance profile in *Ae. aegypti* populations from five municipalities in the state of Sergipe, northeast Brazil. Eggs were collected using ovitraps in five populations in the state: Capela, Laranjeiras, Neópolis, Pinhão and Umbaúba. The bioassays using deltamethrin were carried out in the laboratory using the impregnated bottle test, with the diagnostic dose and exposure time following the recommendations of the Centers for Disease Control and Prevention: 10µg/vial of deltamethrin for 30 minutes. The presence of the Nav mutation (Val1016Ile) was investigated on samples of 30 males of *Ae. aegypti* of each municipality. DNA was extracted individually and allele-specific primers were used for the PCR reaction. Three populations showed altered sensitivity in the bioassays: Capela (92.7%), Umbaúba (91.2%) and Pinhão (91.2%), while Laranjeiras (99.4%) and Neópolis (100%) were considered susceptible to the insecticide. Of the 148 *Ae. aegypti* males analyzed for the kdr mutation, 32.4% were homozygous dominant, 42.6% were heterozygous and 25% were homozygous recessive. The municipality of Neópolis showed the highest frequency of recessive homozygotes (42.9%), followed by Pinhão and Umbaúba (26.7%), Laranjeiras (23.3%) and Capela (6.7%). The altered sensitivity (phenotype) and mutation frequency (genotype) presented in the populations tested may be a reflection of the use of this group of insecticides by the population over time, or agriculture since this insecticide is no longer used in public health to *Ae. aegypti* control.



S2-P23

Distribution of *Culex pipiens* s.s. and *Cx. torrentium* in Europe

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Zoonotic viruses transmitted by native *Culex* mosquito species are (re-)emerging widely across Europe, as evidenced by the detection of human West Nile virus infections in 126 NUTS-3 regions in 2023, a new maximum since the largest outbreak in 2018. Other circulating *Culex*-associated pathogens in Europe include Sindbis virus and Usutu virus, all of which may lead to febrile illness and severe neurological symptoms, with no specific treatment or vaccine for humans currently available. *Culex pipiens* s.s. is considered the most important vector species in Europe. However, *Cx. torrentium* has been shown to also have a very high vector capacity. Although the species is considered more prevalent North of the Alps, knowledge on the exact difference in the distribution of *Cx. pipiens* s.s. and *Cx. torrentium* taxa remains limited as the females are morphologically indistinguishable. Therefore, in the present work, species occurrence records of *Culex pipiens* s.s. and *Cx. torrentium* were extracted from 260 European studies with molecular tools used for taxa identification. The data were compiled in standardized datasets and used in an ensemble of 18 species distribution models. It was found that the areas suitable for *Cx. pipiens* s.s. and *Cx. torrentium* largely overlap in Central Europe, but *Cx. torrentium* extends further to the North and *Cx. pipiens* s.s. to the South of Europe. Interestingly, there were several areas in Eastern Europe (e.g., Poland, Hungary, Romania, Croatia) and regions of lower latitude such as Northern Spain and Northern Italy, which were highly suitable for *Cx. torrentium*. The suitability maps generated by this study represent an important tool to understand the risk of *Culex*-associated pathogen transmission or identify areas for potential vector control in Europe.



S2-P24

The role of biodiversity on the dynamics of trematode communities and their circulation

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Predicting and acting against infectious diseases that are increasingly emerging and spreading, as well as halting biodiversity loss, are intertwined challenges that are crucial to ensure global health. Several links were already established between biodiversity levels and the circulation of pathogens at global scales. On one hand, a high diversity of potential hosts can lead to a high diversity of pathogens present in ecosystems, with a greater risk of host hopping and possibly the emergence of infectious diseases. On the other hand, biodiversity can also drastically influence (either positively or negatively) the circulation of a given pathogen species through several biotic interactions and ecological processes. This is particularly true for trematodes that display complex life cycles hence involving several hosts and several, sometimes free-living, life stages. We here investigate how biodiversity at the ecosystem scale influences the composition of trematodes communities and their circulation in Mediterranean coastal wetlands (Bagnas Nature Reserve). We first assess the theoretical parasitic (trematode) pressure exerted by the fluctuating diversity of definitive hosts (particularly birds) on the Bagnas aquatic ecosystems and the potential risks of local circulation of trematodes based on the reconstructed communities of aquatic snails, the first obligatory intermediate host for all trematodes species. We next investigate the seasonal dynamics of local snail diversity and abundance, to study their effects on the circulation of the most prevalent trematodes species identified locally. This empirical study shed lights on the ecological mechanisms linking biodiversity and the circulation of trematodes.

Track the Tiger: Unraveling Resting Site Preferences of *Aedes albopictus* in Occitania, France

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The Asian tiger mosquito, *Aedes albopictus*, thrives in urban environments by exploiting artificial containers for larval breeding. Despite the importance of adult mosquitoes' resting sites for public health preventative and curative measures, limited attention has been given to their exploration in temperate and European contexts. Correlations have been reported between vegetation and vector density at different developmental stages, but the role of urban vegetation elements, as resting sites for mosquito populations, remains uncertain. This study offers a thorough examination of adult *Aedes albopictus* resting sites at a detailed level. Our objectives encompass: (i) identifying and characterizing resting sites, (ii) phenotypic profiling of mosquito populations occupying these sites, and (iii) establishing standardized indicators to enhance targeted preventive and reactive control strategies.

We studied 72 resting sites across a 100-hectare village, including natural and artificial habitats. During almost 1300 resting site sampling replicates local and global climatic conditions (temperature, wind, humidity, etc.) and vegetation characteristics were recorded. Our capture method combines 5-minute human aspiration technique, followed by 1 minute 30 seconds of aspiration per resting site. Dissections of adult mosquitoes allowed us to observe key anatomical markers indicating vector age structure, including ovarian follicle development, parity, and spermatheca fillings in females, and genitalia rotation in males.

We demonstrate significant effects of resting site type on mosquito capture probability and abundance. Additionally, the presence of males was found to impact female capture probability. Habitat type exhibited a significant influence on mosquito sex ratio, with certain habitats favoring young or mature female mosquitoes.

These results are complemented by ground releases of batches of coloured sterile male mosquitoes to better document resting site preferences and dispersal patterns. Identifying preferred resting sites will help vector control operators, modelers, and decision-makers adjust strategies for more effective public health management of *Aedes albopictus* in the area.

Estimating the age of field *Anopheles* mosquitoes using mass spectrometry and deep learning

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Introduction

Mosquito survival is critical for epidemiological monitoring of pathogen transmission and evaluation of vector control strategies, as the risk of pathogen transmission increases with mosquito age. An accurate determination of mosquito age would also provide a better understanding of vector biology. However, available tools for estimating the age of adult field mosquitoes are often approximate and time-consuming. In this context, matrix-assisted laser desorption and ionization time-of-flight mass spectrometry (MALDI-TOF MS) is emerging as a promising tool for mosquito age estimation.

Objective

This study aimed to evaluate a rapid method combining MALDI-TOF MS with deep learning for mosquito age estimation.

Methods

To establish a genetically variable dataset, field larvae of *Anopheles arabiensis* were collected from two ecologically distinct sites in Senegal. Mosquitoes were laboratory-reared under uncontrolled temperature and humidity while controlling for age. We developed deep learning frameworks such as convolutional neural networks, conventional classification, rank-consistent classification, and regression, to detect protein variations linked to mosquito aging and to improve the granularity of mosquito age estimation.

Results

Using 2,763 mass spectra from head, legs, and thorax of 251 field *An. arabiensis* mosquitoes, we achieved a best mean absolute error of 1.74 days for mosquito age estimation. We showed protein biomarkers of age that were conserved between the two populations of mosquitoes collected. Modeling studies confirmed that the accuracy of our approach was independent of the age distribution of the samples, which is promising for the monitoring of the age structure of wild *Anopheles* mosquito populations.

Conclusions

We have developed a rapid method for estimating the age of *Anopheles* mosquitoes, using deep learning analysis of MALDI-TOF mass spectra. Accurate and expandable, we anticipate that our approach will have strong implications for malaria control and the field of vector biology, benefiting other disease vectors such as *Aedes* mosquitoes.



S2-P27

Dancing out of step: The adaptability of *Anopheles stephensi* rhythms and their impact on malaria development

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Biological rhythms mediate interactions between organisms; from predators and prey, to mating behaviors between males and females, to hosts and parasites. The role of daily rhythms in infections is gaining traction because explaining the regulatory mechanisms and fitness consequences of biological rhythms exhibited by parasites, vectors and hosts offers new avenues for infection control.

Our lab has shown that malaria parasites exhibit asexual development that is synchronized with host feeding time and that the development of transmission stages are also synchronized with the biting time of the mosquito vector. Disruption of parasites synchrony in the mammalian host can lead to a reduction in growth stages and in transmission stages, greatly reducing potential for mosquito transmission. Yet, while the rhythms of mammalian hosts significantly impact parasite fitness, the role of mosquito rhythms in parasite development during transmission remains uncertain. Moreover, field observations reveal the adaptability of mosquitoes' daily rhythms, such as adjusting their feeding times in response to interventions like bed nets and thriving amidst urban environments flooded with artificial light at night. However, the potential fitness repercussions for mosquitoes with altered rhythms remain unexplored.

Here we disrupted the rhythms of *An. stephensi* by subjecting mosquitoes to photoschedule durations (i.e the total length of day plus night) that are longer or shorter than the standard 24 hours and investigate the consequences for both mosquito life history traits such as egg lay, survival, nutrition and the timing of flight activity. Second, we explore the impacts of this altered environment for malaria parasite development in the mosquitoes including the likelihood of infection, the duration of development, and overall parasite density. By unraveling the intricate interplay between biological rhythms, parasite development, and vector behavior, our study offers insights into the complex landscape of infectious disease transmission.



S2-P28

Poorly-resourced mosquitoes exhibit increased host seeking and early evening biting

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Introduction and Objectives

Vector-borne disease transmission is reliant on vector biting behaviour, where parasites/pathogens are acquired from an infected host and transmitted to a new host during blood-feeding events. Vector control tools exploit vector behaviour to reduce transmission, and in the case of malaria, tools such as insecticide treated bednets, which target the nocturnal biting behaviour of *Anopheles* mosquitoes, have been effective for curbing transmission. Despite this success, residual transmission persists, and could be partly explained by growing reports of mosquitoes shifting their biting time to when people are unprotected by bednets. However, the extent to which mosquito behaviour can shift, and its underlying causes, remain unclear. Given that mosquito feeding ecology revolves around their need for both blood and sugar meals, we investigated how differences in mosquito nutritional status impacted biting time and host seeking behaviour.

Materials and Methods

We perturbed the sugar and blood availability of adult mosquitoes, which were F2 progeny of wild-caught *Anopheles gambiae* s.l. larvae collected across Accra, Ghana, and released them into a semi-field system with traps mimicking human hosts. We programmed the traps to turn on in the evening, night-time and morning to capture mosquitoes biting at different times across the night, and collected mosquitoes from each trap after the final trap had closed.

Results, Discussion and Conclusion

We show that mosquito nutritional status affects propensity to host seek and the time of day that host seeking occurs. Specifically, poorly-resourced mosquitoes were more likely to host seek, with the majority of these mosquitoes trapped during the evening. Our results reveal that mosquito biting time is a plastic trait which can be modulated by resource availability, and highlights the importance of considering the role of environmental conditions on vector behaviour, especially in cases where behavioural shifts can reduce the efficacy of control interventions.



Comparing Ivermectin susceptibility in wild-derived and laboratory-reared *Aedes* and *Anopheles* larvae: Preliminary study on selecting for resistance mechanisms

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Introduction and Objectives

Administering ivermectin to humans and livestock renders their blood toxic for mosquitoes like *Anopheles* and *Aedes*, offering a promising approach for controlling these vectors. However, the impact of such treatment on larval stages exposed to the drug through contaminated breeding sites is not fully understood. This study looked at how ivermectin affects the development of *Aedes* and *Anopheles* larvae.

Materials and Methods

We exposed laboratory-reared (*Anopheles gambiae* Kisumu and *Aedes aegypti* Bora Bora) and wild-derived (*Anopheles coluzzii* VK5 and *Aedes aegypti* Bobo) larvae to ivermectin concentrations ranging from 1 to 100 ng/ml for 24h, and transferred surviving larvae into free-ivermectin medium to monitor development until adult stage. Parameters measured were : survival, pupation dynamics, emergence rates, and fecundity of the adult females. Ivermectin effects were characterized by comparison with larvae raised in control medium.

Results

Data indicated that highest ivermectin concentrations (100, 75, and 50 ng/ml) reduced larval survival by over 50% within 24 to 48 hours post-exposure, with varying effects across different strains. Wild-derived larvae showed lower susceptibility to ivermectin compared to laboratory larvae for both *Anopheles* and *Aedes* species. The concentrations leading to 50% larval mortality (4-day-LC50) were 3.65 and 1.86 ng/ml for *Anopheles* VK5 and Kisumu strains, and 15.60 and 2.56 ng/ml for *Aedes* Bobo and Bora Bora strains, respectively. The transition from larval to adult stage was significantly affected, particularly in the Kisumu strain ($p = 0.001$). No significant effects on the number of laid eggs were observed across different strains.

Discussion and Conclusion

These data showed how lab-raised and wild-derived *Anopheles* and *Aedes* larvae are affected differently by ivermectin, highlighting potential implications for vector control strategies. Further investigations are planned to understand potential existing mechanisms allowing wild-derived larvae to better survive to ivermectin exposition than laboratory ones.



S2-P30

The spread of *Aedes albopictus* in Spain: linking human mobility and habitat suitability

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The tiger mosquito, *Aedes albopictus*, is an invasive urban vector responsible for transmitting diseases like Dengue and Chikungunya. Its introduction to Spain was first documented in 2004 in Sant Cugat del Vallés, and since then, it has progressively spread along the Spanish Mediterranean coast and is moving inland. Experimental research indicates that these mosquitoes have two dispersal modes: natural dispersal, flying, and human-mediated dispersal, they are capable of hitchhiking in vehicles, facilitating their ability to cover vast distances.

We study the colonization patterns of *Aedes albopictus* over the past two decades, utilizing data collected from the citizen science platform, Mosquito Alert. Our analysis focuses on two primary factors: human mobility and habitat suitability. To comprehensively understand the effect of different dispersal modes on the colonization process, we integrate human mobility, habitat suitability, and distance to previously invaded regions in a metapopulation model. This model represents a variation of the well-known Hanski metapopulation model. Within this metapopulation model, the Mosquito basic reproduction number, R_M determines the habitat suitability of individual patches. This variable incorporates parameters associated with the mosquito life cycle: temperature, rainfall, and human population density. Moreover, we enrich our analysis by leveraging fine-scale mobility data sourced from the MITMA dataset for Spain at hourly scale.

Our ultimate goal is to gain insights into the invasion process and identify key factors that promote colonization. To achieve this, we aim to infer key parameters, including colonization rates, extinction rates and a key parameter measuring the impact of each dispersal mode. After that, we intend to project our findings into the future using CMIP6 climate change predictions to anticipate potential future colonization hotspots.



S2-P31

Assessing the risk of mosquito vector-borne diseases in Scotland and their response to environmental change

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In recent decades, invasive mosquito species have spread across Europe, leading to an increase in mosquito-borne diseases. This spread is linked to increased travel, international trade, and climate changes, which create favourable conditions for new mosquito species and their diseases. While the United Kingdom (UK) has been less affected due to its geography and climate, extensive travel and climate variations still pose a risk for vector-borne diseases (VBDs) to emerge. Birds are key reservoirs for many emerging zoonotic VBDs in Europe, like West Nile Virus (WNV) and Usutu Virus (USUV), potentially carrying these diseases into the UK along migratory routes. Since 2009, mosquito surveillance in England and Wales has identified various vector species. However, there is limited data for Scotland, despite the presence of favourable wetland habitats for both potential vector species and avian reservoirs.

The primary objective of the Mosquito Scotland project is to evaluate the risk of mosquito-borne disease emergence in Scotland in the light of climatic change by addressing surveillance gaps. We will present initial findings regarding the establishment and characterization of a *Culex pipiens pipiens* colony from field collected mosquitoes. We also carried out some RNAseq for pathogen detection in mosquito vectors and avian reservoir populations in Scotland. Additionally, we tested the vector competence of a Scottish *Cx. pipiens* population in transmitting Usutu and Sindbis arboviruses.

Longitudinal survey of insecticide resistance in a Burkina Faso village reveals co-occurrence of 1014F/S and 402L mutations in *Anopheles coluzzii* and *Anopheles arabiensis*

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Introduction and Objectives

Pyrethroid resistance (PR) of *Anopheles* mosquitoes is threatening the effectiveness of pyrethroid-treated bednets (ITNs), a crucial tool for malaria vector control. Pyrethroids target the voltage-gated sodium channel (VGSC) and genotyping of mutations in the VGSC gene is widely used to assess the spread of target-site resistance among malaria vectors. L1014F and L1014S substitutions are the commonest and best characterized VGSC-mutations in African malaria vectors of the *Anopheles gambiae* complex. Recently, a substitution involved in PR, V402L, has been detected in west-African *Anopheles coluzzii*. We here monitored the temporal dynamic of target-site resistance mutations L1014F/S and V402L in *A. coluzzii* and *A. arabiensis* specimens from a Burkina Faso village over 10 years after the massive ITN scale-up started in 2010.

Material and Methods

Anopheles coluzzii (N= 300) and *A. arabiensis* (N=362) specimens, collected in 2011, 2015 and 2020 in Goden village were genotyped for the three target-site resistance mutations by TaqMan-assays and sequencing.

Results, Discussion and Conclusion

Over the study period, 1014F-frequency decreased in *A. coluzzii* (from 0.76 to 0.52) but increased in *A. arabiensis* (from 0.18 to 0.70); 1014S was detected only in *A. arabiensis* (from 0.33 to 0.23); 402L increased in *A. coluzzii* (from 0.15 to 0.48) and was found for the first time in *A. arabiensis*. In 2020 resistance alleles co-occurred in 43% of *A. coluzzii* (410L and 1014F) and 32% of *A. arabiensis* (1014F and 1014S). After 10 years from first ITN implementation in Goden, a significantly increasing level of target-site resistance was observed in the vector population, with only 1% of specimens being wild-type at both loci. This, together with the evidence of co-occurrence of different mutations in the same specimens, calls for future investigations on the possible synergism between resistance alleles and their phenotypic effects to implement locally tailored vector-control strategies.

Breeding of Tsetse Flies and Animal Welfare at CIRAD Baillarguet

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Tsetse flies, also known as Glossines, are vectors of trypanosomes, the causative agents of sleeping sickness in humans and Nagana in animals. Their habitat is located in sub-Saharan Africa, mainly in savannahs (gallery forests) and humid forests, under specific physical and climatic conditions. Therefore, breeding them in insectariums poses a real challenge.

At the CIRAD insectarium platform in Baillarguet (Montpellier, France; <https://doi.org/10.18167/infrastructure/00001>) member of the Vectopole Sud network), three colonies of *Glossina* from the Palpalis group are raised: *Glossina palpalis gambiensis*, *Glossina palpalis palpalis*, and *Glossina fuscipes fuscipes*, under controlled temperature and humidity conditions. Moreover, tsetse flies are strictly hematophagous for both sexes, which represents an additional difficulty for their breeding. Until May 2018, the colonies were fed on rabbits. In line with the 3Rs (“**R**eplace, **R**educe, **R**efine”) rule in animal experimentation, we have implemented a new artificial feeding protocol using a membrane with sheep blood. The artificial feeding device consists of a heating device (maintaining a temperature of 37°C for the blood meal), an aluminum plate of 49x49 cm², and a 46x46 cm silicone membrane 1 mm thick that mimics the host’s skin. For flies feeding, 200 ml of blood is spread on the plate and covered by the membrane. The cages are placed on the membrane, and the device allows feeding around 600 flies simultaneously.

The entire breeding protocol has also been revised to improve and simplify the work, particularly the adult sorting system (separation of sexes). Tsetse flies are now anesthetized with cold, under strictly controlled conditions to facilitate and secure handling.

The insectarium houses a minimum of 800 to 1000 female individuals of each strain and can increase production according to demand of partners for research and teaching.



Detection of eDNA of *Aedes albopictus* in water samples from tree bags in the Viennese Prater

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Introduction and Objectives

In the last decades, the Asian tiger mosquito, *Aedes albopictus*, has been introduced to Europe and was able to establish new populations. Due to its potential to transmit pathogens, such as dengue virus or *Dirofilaria* spp., *Ae. albopictus* poses a threat to human and animal health. Therefore, monitoring is of great importance. Currently, surveillance strategies most commonly include the collection of mosquito eggs by ovitraps and/or the use of CO₂-baited traps for detection of adult mosquitoes. In this project, we aim to develop a new approach for mosquito monitoring by applying molecular analyses to environmental samples, such as water.

Materials and Methods

In July and August 2024, tree bags located in the Viennese Prater area were monitored and checked for the presence of *Ae. albopictus*. Therefore, the presence of adults and larvae of *Ae. albopictus* within or around the tree bags was documented weekly. Adults and larvae were identified either morphologically and/or using molecular techniques (PCR, DNA barcoding). Additionally, water samples were collected from the tree bags and prepared for eDNA detection. After filtration and DNA extraction, the subsequent qPCR assay was performed, which targets the ITS2 gene of *Ae. albopictus*. The results of the water analyses are then evaluated by comparing them with the monitoring data of adult and larval *Ae. albopictus*.

Preliminary Results

Water samples from 5-10 sampling sites will be collected and analyzed for the presence of *Ae. albopictus* using molecular methods/techniques.

Discussion and Conclusion

The results of this project will allow to explore a complementary strategy to traditional mosquito surveillance and to evaluate its potential as a stand-alone approach. Moreover, the outcome may be helpful to assess the efficacy of vector control measures in the future.

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Host Preferences of *Culex pipiens* and its Significance for West Nile and Usutu *Flaviviruses* transmission in Camargue, France

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Introduction and Objectives

The spread of the *Flaviviruses* West Nile (WNV) and Usutu (USUV) in Europe has highlighted the critical need to comprehend the enzootic cycles of these pathogens for effective decision-making. These viruses are part of a complex disease cycle that involves birds as principal hosts and humans as dead-end hosts. Our study aims to visualize the intricate relationships between the main vector of this viruses, *Culex pipiens*, and its host preference for several host species, primarily birds, and the potential changes into a land use gradient that could pose a risk factor in WNV and USUV transmission.

Materials and Methods

We conducted mosquito sampling for host-seeking and engorged females, coupled with bird censuses in the Camargue region in southern France, where these two viruses' co-circulation had been well reported. Several localities were sampled monthly along a land-use gradient of peri-urban, agricultural, and natural areas from May to November 2021. Engorged *Cx pipiens* were analyzed by PCR amplification and sequencing of mitochondrial 12S and 16S DNA

Results

Culex pipiens was found to feed primarily on birds and mammals. We identified 55 vertebrate species in 110 engorged *Cx pipiens*, 51 bird species, two mammals, one amphibian, and one reptile. The Forage ratio was estimated for several bird species. Based on these results, we described a preference of *Cx. pipiens* in Camargue to selectively bite *Columbia livia domestica* in rural areas and *Passer montanus* in agricultural and peri-urban areas. Passeriformes is the order preferred to be bitten by *Culex pipiens*.

Discussion and Conclusion

In our analysis, *Columbia livia* and *Passer montanus* have been identified as potential super-spreader. Results demonstrate species selection and discuss interactions concerning West Nile host competence and potential Flaviviral transmission risk. These results will be helpful in disentangling host-vector interactions and the relationship between bird diversity and emerging mosquito-borne disease.



S2-P36

Tick-borne microorganisms detected in small mammals collected in the surroundings of two villages in Styria, Austria

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Tick-borne microbes rely on a complex infection model involving ticks and reservoir animals to sustain in nature. In this study, we aimed to get further insight into the role of small mammals as reservoirs of tick-borne pathogens.

To this end, 87 small mammals (i.e., *Mus musculus*, *Sorex araneus*, *Apodemus flavicollis* and *Myodes glareolus*) were collected near two Austrian villages: Unterpurkla and Hummersdorf. Both villages are in the federal state of Styria.

Samples of the bladder, ear, heart, kidney, liver, lungs, paw and spleen were collected for DNA isolation and subsequent reverse line blot (RLB) analysis. This method allows to screen multiple samples for the presence of DNA of numerous pathogens at once. Briefly, the RLB consists of a membrane that has up to 43 (geno) species-specific oligonucleotides bound to it. To screen for pathogens, tissue DNA extractions were subjected to several genus-specific PCRs targeting *Anaplasma*, *Babesia*, *Borrelia*, *Ehrlichia*, *Rickettsia* and *Theileria* species. The resulting PCR products were subsequently hybridised to the RLB probes using a mini-blotter. Bound PCR products were visualized using chemiluminescence. Of all screened animals, 56% (49/87) tested positive for at least one pathogen. The identified pathogens were *A. phagocytophilum*, *Ba. canis*, *T. (Ba.) microti*, *B. afzelii*, *B. garinii/bavariensis*, *Cand. Neoehrlichia mikurensis*, *R. helvetica*, *R. raoultii* and *R. slovaca*. Some RLB signals required further sequencing to give a definitive answer. This revealed the presence of a Hepatozoon species with an 18S rRNA sequence 99.3% similar to *Hepatozoon ophisauri*. A pathogen previously not found in Austria and normally associated with legless lizards (*Pseudopus apodus*) found from Southern Europe to Central Asia.

This study revealed a surprising variety and high prevalence of tick-borne microbes in various species of collected animals, emphasizing the important role of small mammals as reservoirs of pathogenic agents in nature.



Investigating the buzz: What environmental and weather factors are associated with *Aedes albopictus* in Southern Ontario, Canada?

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Background

Since 2016, *Aedes albopictus*, responsible for transmitting Zika, chikungunya, yellow fever, and dengue viruses, has been identified in Ontario, Canada. It was previously thought that this species could not survive Canada's cold climate; however, *Ae. albopictus* is now established in some parts of southern Ontario. The factors facilitating introduction, survival and establishment are currently unknown. With the growing impacts associated with climate change, it is important to understand the contributing factors associated with the presence and expansion of *Ae. albopictus* so that preventative measures can be implemented.

Objective

To explore the habitat and weather factors associated with the presence or absence of *Ae. albopictus* in the region of Windsor, Ontario.

Methods

Our 8-year dataset includes mosquito trap type, the number of *Ae. albopictus* found in each trap, the date of collection, as well as the location of the trap from month-year to month-year. We will use a mixed-effects regression model to examine the association with adult *Ae. albopictus* counts and explanatory variables including precipitation, temperature, relative humidity, wind speed, land use, and population density.

Results

The number of trapped adult *Ae. albopictus* increased year by year. In 2016 only 17 adults were found in 4 traps, this number has grown to 822 adults in 54 traps in 2022. We predict there will be associations between environmental and weather factors and the entomological data collected in Windsor, Ontario.

Conclusions

Weather and habitat factors are known to be influential in terms of mosquito development and survival. These factors have not been investigated in Southern Ontario to determine drivers of absence or presence of *Ae. albopictus*. The results from this study identify which of these factors are most influential to *Ae. albopictus* survival in the first region of Canada where this species has established and can inform public health and mosquito control programs.

Host identification, environmental and climatic variables associated to host-feeding patterns of mosquito vectors of West Nile and Rift Valley fever viruses in Madagascar

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The burdens of West Nile and Rift Valley Fever viruses vary across Madagascar, probably resulting from heterogenous abundance of mosquito vector species and variations in the level of vector-host contacts. Here we explore the association of host, climatic and environmental factors with mosquito feeding behaviour. Bimonthly mosquito collections were performed in 24 districts of Madagascar from February to October 2019. Engorged mosquitoes were collected indoors after indoor pyrethrum spraying (IPSCs) and outdoors using Muirhead-Thomson pit traps (MTPTs). Blood meal identification was performed using PCRs. For the two predominant mosquito species, beta regression models were developed using four blood indices (BI) corresponding to human (HBI), cattle (CaBI), bird (BiBI) and small ruminants (SRBI) as response variables. Twelve variables including seasonality, average number of vertebrate hosts per house, climatic and environmental factors were included as explanatory variables. In MTPTs, origin of the blood meals were identified in 770 females of 843 females tested. In IPSCs, they were identified in 297 females of 334 females tested. Human was the dominant vertebrate host identified (44.86%), followed by cattle (44.52%) and Galliformes (25.53%). Blood meals from Passeriformes and pigs were rarely detected (<1%). Blood indices of *Culex antennatus* and *Culex quinquefasciatus* (two predominant species) varied in time and space ($p < 0.05$), and were correlated to the collection method used ($p < 0.05$). Significant associations between the mosquito blood indices and explanatory variables were observed only for *Cx. antennatus* : (i) HBI and the average number of humans per house [Odds Ratio = 2.36], (ii) CaBI and the outdoor resting location [OR = 0.28], the precipitation [OR = 1.03] and relative humidity [OR = 0.93]. (iii) BiBI and the type of house [OR = 3.89] and the number of humans per house [OR = 0.41]. The different results obtained from *Cx. antennatus* and *Cx. quinquefasciatus* demonstrate differences in biological and ecological traits between mosquito species.

Effortless Counting of Sorted *Culicoides* (Diptera: Ceratopogonidae) from Light-Traps

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Introduction and Objectives

Culicoides biting midges are among the most abundant of haematophagous insects. They transmit a great number of pathogens, but it is as vectors of arboviruses, and particularly arboviruses of domestic livestock, that they achieve their prime importance. Counting sorted *Culicoides* from light traps can be time-consuming, especially with hundreds or thousands of specimens. Despite advancements in artificial intelligence (AI) and AI-based object counting applications, implementing these techniques can still be costly or challenging.

The study aimed to develop a simple, cost-effective, and easy-to-implement method for counting sorted *Culicoides* from light traps.

Materials and Methods

Culicoides, ranging from 10 to 1000, were accurately counted using a stereomicroscope and then photographed in alcohol on a white enameled plate with an Android cell phone. Subsequently, these samples were mixed to create various densities, up to several thousands, and photographed again.

The IMAGEJ (FIJI) software was utilized to automatically count the specimens using a combination of simple commands based on the final "FIND MAXIMA" process. Optimal parameters for the command chain were estimated by comparing the results from several photographs of various insect densities with known exact counts.

After defining the process, its agreement with exact manual counting using the stereomicroscope was evaluated through Bland-Altman analysis and Passing-Bablok regression.

Results

The statistical analysis showed that the method is highly repeatable, and as precise as the standard manual counting. It can thus be used in routine analysis.

Conclusion

The presented method is rapid and simple, and can even be implemented in a cell phone with the IMAGEJ. JS software. To simplify the process, particularly for the cell phone app where screen manipulation is challenging, an ImageJ MACRO containing all the necessary commands and parameters has been developed to simplify the process and is available from the authors.

Exploring Positive Chemotaxis in *Phlebotomus papatasi* Larvae

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Introduction and Objectives

Phlebotomine sand flies are important vectors of medical and veterinary importance, transmitting pathogens, such as the *Leishmania* parasites, responsible for 700,000 – 1 million new cases of leishmaniasis every year. The vast majority of the current sand fly surveillance and control tools are tailored against the adult stages, due to the limited knowledge on the ecology of the larval stages. It is well known that chemical cues play an important role in insect behavior. While there are numerous studies investigating the behavior of adult sand flies in response to chemical sources, there is currently no information available on the response of their larval stages. To address this gap, we developed novel bioassays for investigating the effect of chemical cues (gustatory and olfactory) on the behavior of sand fly larval stages.

Materials and Methods

Phlebotomus papatasi sand fly larvae were used for all bioassays (three bioassays were constructed in total). Responsiveness to chemical cues was assessed using the colony standard larval feeding diet, and an alternative stimuli source.

Results

The larvae exhibited a clear food preference within a few hours of exposure in a two-choice assay, while, also, demonstrated positive chemotaxis in response to volatile stimuli emitted from their preferred food source.

Discussion and Conclusion

The present study introduced innovative feeding and olfactory attractancy bioassays for sand fly larvae, unveiling their capacity to perceive and respond to gustatory and olfactory cues. Identification of the specific chemical compounds eliciting attractance response to sand fly immature stages could lead to the development of innovative, and targeted (larval-specific) tools for the surveillance, and management of these important public health pests.



S2-P41

Unveiling a snail host of a major trematode in Europe: *Galba schirazensis* (Lymnaeidae) facilitate the transmission of Paramphistomidae while disengaging *Fasciola hepatica*

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Introduction and Objectives

Trematodes parasites –major players of zoonoses of worldwide relevance- have tightly evolved with snails, being the latter ultimately mandatory hosts for their existence. Species of Lymnaeidae (Mollusca: Gastropoda) are hosts of a wide spectrum of trematodes. Particularly, *Galba* spp. play a key role in the transmission of *Fasciola hepatica* (liver fluke, affecting over 30 million people worldwide) and *Calicophoron daubneyi* (rumen fluke, infecting ruminants). Both parasites are in re-emergence in Europe. Genus *Galba* comprises small, amphibious species of high crypticity that usually differ in their ecology, ultimately affecting the rate of snail-parasite encounter. Molecular identification of *Galba* spp. is then capital to the understanding of the epidemiological patterns. Here we provide novel results of their experimental susceptibility and overall compatibility to the circulating isolates of the above-mentioned trematodes.

Materials and Methods

We used a combined field ecology and experimental parasitology approach to unveil the distribution and susceptibility of cryptic *Galba* spp. to circulating flukes.

Results, Discussion and Conclusion

While *G. truncatula* (a long widespread species in Europe) has been considered the main intermediate host for both parasites, the exotic *G. schirazensis* (native to the Americas) showed 100% prevalence when experimentally exposed to *C. daubneyi* but failed to develop infection of *F. hepatica* with significant reduced survival. As expected, *G. truncatula* showed 100% prevalence after exposure to both trematodes but significant less parasite intensity (total rediae and metecercariae at day 30 post-exposure) in the case of *C. daubneyi* when compared to *G. schirazensis*. Hence, we provide the first evidence of the ability of *G. schirazensis* to transmit the rumen fluke. Given that both *Galba* spp. differ in their ecology, we suggest that the spread of *G. schirazensis* in Europe will inflect dramatic shifts in the transmission patterns of both trematodes in the frame of the ongoing climatic change.



S2-P42

Updated distribution of *Dermacentor* ticks in Austria

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Introduction and Objectives

Recent studies have shown that *Dermacentor* ticks are expanding to new areas in Europe. These ticks feed on ground-dwelling mammals, including humans, and are not known to feed on birds. As a result, their spread is limited to the range of these mammals and the obstacles they encounter, such as rivers, mountains, and other landmarks. In Austria, only limited studies have been conducted to investigate the distribution of *Dermacentor* ticks. Thus, we aimed to investigate the current occurrence, distribution, and associated pathogens of *Dermacentor* ticks.

Materials and Methods

Ticks were collected from the vegetation by hand in multiple locations in Vienna, Lower Austria, and Burgenland. The specimens were morphologically identified using established identification keys. A subset of the specimens underwent molecular identification to confirm their taxonomic classification. Subsequently, DNA was extracted from all collected ticks individually. PCR-reverse line blot analysis is used to identify the presence of pathogens (*Ehrlichia*, *Anaplasma*, *Babesia*, *Theileria*, *Rickettsia* and *Borrelia* spp.) in the obtained DNA extracts.

Results

We have identified several novel locations where *Dermacentor* ticks are present. From these locations, we have so far collected more than 150 specimens and are currently in the process of screening these questing ticks for tick-borne pathogens.

Discussion and Conclusion

This study will help to better understand the ecology and epidemiology of *Dermacentor* ticks and their associated pathogens.

The expansion of *Dermacentor* ticks to new geographical areas has led to an increased incidence of tick-borne pathogens in regions previously believed to be free of these ticks. *Dermacentor* ticks are known for their prolonged activity during winter months. This means that ticks are always active in Austria, not only during the spring, summer and autumn seasons.



Does Ivermectin impair *Anopheles* mosquitoes' attractiveness toward treated cattle under field and laboratory conditions?

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Introduction

The administration of ivermectin (IVM) to humans or cattle is viewed as a promising complementary tool to control malaria vectors. However, potential modification of treated hosts' attractiveness to mosquitoes owing to IVM metabolization has not been studied. This study aimed to investigate whether the malaria vector *An. coluzzii* was more attracted to IVM-treated cattle compared to control groups.

Methods

A two-arm study was designed using 8 calves: 4 controls and 4 treated with a long-acting ivermectin formulation at a dose of 1mg/kg (3-4 months efficacy, IMPACT project). The calves were exposed to wild mosquitoes at t=2 days, 1-, 2-, 3-, and 4-month post-treatments, under nets in the field (Bama, Burkina Faso) and to *An. coluzzii* (lab colony) using a dual-choice olfactometer in the laboratory. In field experiment, trapped wild mosquitoes were counted and identified into species. In laboratory experiment, female *An. coluzzii* were released into an olfactometer, and activated females were counted according to their preference for treated or control calves.

Results

A total of 181,696 mosquitoes was collected in field experiments, of which 70% were *Culex*, 27.3% *Anopheles*, and 2.7% *Mansonia*. IVM treatments did not influence mosquito attractiveness ($X^2=0.8791$; $P=0.3484$) when all mosquitoes were taken together. However, IVM-treated calves were more attracted by *Anopheles* mosquitoes than controls, but this was only significant at month 4 post-treatment ($Z=0.584$; $P=0.001$). *An. coluzzii* tended to be more trapped around treated than control calves for all timepoints, but this difference was not significant. At the laboratory, dual choice-tests on *An. coluzzii* colony showed similar attractiveness regardless of cattle treatments ($Z=0.215$; $P=0.83$).

Conclusions

IVM seemed to increase *Anopheles* mosquitoes, including *An. coluzzii* attractiveness toward treated calves in the field, but not in the laboratory. Further studies are needed to decipher the main odor molecules driving this attractiveness toward IVM-treated calves.

How urban green infrastructures influence *Aedes albopictus* presence and abundance? A study in the city of Montpellier (South of France) in 2023

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Urban greening is regarded as a solution to mitigate global urbanization challenges due to its positive impact on human health. However, research on potential adverse effects, especially concerning mosquitoes and mosquito-borne diseases, remains limited. This study aimed to investigate the relationship between urban vegetation and the presence and abundance of the invasive *Aedes albopictus* in Montpellier, the second greenest city in France, considering weather conditions, microclimate, and landscape as potential influencing factors.

Adult mosquitoes and microclimate were monthly sampled from May to October 2023, using Odor+CO₂ traps across vegetated parks and residential areas in the city center. Environmental (meteorological and landscape) variables were extracted from open-access databases. Entomological data underwent two-stage analysis. First, bivariate analysis employing generalized linear mixed models was performed between the presence/abundance of *Aedes albopictus* and environmental variables, considering various time lags and spatial proximities relative to trap location. Secondly, multivariate random forest models were used to highlight nonlinear relationships between environmental factors and the presence/abundance.

The most influential variables for *Aedes albopictus* presence were meteorological: weekly cumulative growing degree days (1 week prior), mean daily wind speed (5 weeks prior), and weekly cumulative rainfall (5-6 weeks prior). Landscape factors (building percentage in a 100m buffer), microclimate (hourly temperature during sampling), and meteorological (weekly cumulative growing degree days (1 week prior)) were the key variables for abundance. Urban vegetation minimally impacted presence but moderately influenced abundance within a 50-meter radius.

To the best of the authors' knowledge, this study is the first to explicitly examine urban vegetation's impact on *Aedes albopictus* in France. Further research is necessary to solidify the findings presented and to explore the influence of vegetation on other determinants of *Aedes*-borne diseases risk (including vector and parasite biology). These findings can inform urban greening strategies while monitoring and managing mosquito populations.



Modelling RVF vector population dynamics in a tropical setting in South-Eastern Madagascar

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Introduction and Objectives

Rift valley fever (RVF) is an emerging zoonotic arbovirolosis circulating in Africa, the Middle East and the Indian Ocean. It has been identified by the WHO as a priority disease for research, surveillance and prevention, due to the high human, animal and economic toll it has on affected communities. Three large epizootic waves have hit Madagascar since RVF virus (RVFV) was introduced there, including in 2021 in South-Eastern Madagascar. Interepizootic circulation has been demonstrated.

We studied the population dynamics of two suspected RVFV vector species in South-Eastern Madagascar: *Anopheles coustani* and *Culex antennatus*. The outputs will be used to model RVFV transmission, and ultimately allow for risk assessment of RVF flares according to climate variables.

Materials and Methods

A generic mechanistic compartmental model, representing mosquito life cycle and driven by temperature and rainfall, was adapted to each of the two vector species. We used the model to estimate the relative abundances of host-seeking mosquitoes over time, which we then compared to observed mosquito captures.

Results, Discussion and Conclusion

Our model was able to predict vector population dynamics according to climate variables, with predictions in good agreement with field observations, especially for *Anopheles coustani*. Peaks in predicted mosquito abundance occurred at the end of the wet season, which is concurrent to a high RVFV circulation. These first results are promising when it comes to incorporating the population dynamic model into a transmission model for RVFV in Madagascar.

Diurnal and outdoor biting by *Anopheles gambiae* complex malaria vectors reveals residual transmission along an urbanisation gradient in Burkina Faso

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Introduction

Despite considerable progress in malaria control, challenges remain because of spreading resistance to insecticides and antimalarials. Moreover, idiosyncrasies in vectors biting behaviour can lead to persistent transmission despite effective implementation of control interventions based on insecticidal nets. Malaria is more prevalent in rural as opposed to urban areas. However, urban malaria may increase in the future due to adaptation of malaria vectors to urban conditions. Here, we investigated biting of malaria vectors along an urbanisation gradient to gain insights into the emerging issues of urban malaria and vector behaviour in residual transmission.

Methods

We assessed biting rates in three localities next to and within Ouagadougou, Burkina Faso, each locality representing urban, peri-urban, and rural environments. Human volunteers conducted landing collections during 3 months of the 2023 rainy season. In order to determine the relative contribution to transmission of indoor *versus* outdoor biting, as well as daytime *versus* nighttime biting, each sampling occasion consisted of parallel indoor and outdoor collections during 48 hours round-the-clock.

Results

The distribution of 4,428 *Anopheles gambiae s.l.* across indoor/outdoor sites and daytime/nighttime periods varied between urban and non-urban settings. In the urban area outdoor landings accounted for >2/3 of the total, whereas in non-urban settings this proportion decreased to ~1/2. Diurnal samples taken indoors contributed 13-19% of biting in non-urban sites. Assuming a transmission season of 100 days and average mosquito infectiousness of 1% (assessment underway), we expect ~90 and ~1 infectious bites/person/year outdoors in the urban locality, and during daytime in non-urban settings, respectively.

Conclusions

Growing evidence indicates limitations in malaria control interventions relying on nocturnal indoor biting by vectors. Besides insecticide resistance, the strength of diurnal and/or outdoor biting can set a ceiling to the effectiveness of insecticide-treated nets, suggesting considering vector control strategies against residual transmission in malaria elimination efforts.

Monitoring the public health impact of climate change through environmental indicators: a case study on the Asian tiger mosquito (*Aedes albopictus*) in Flanders

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The Asian tiger mosquito (*Aedes albopictus*) is a highly invasive species, originally endemic to Southeast Asian rainforests but now present on all continents except Antarctica. Its presence is essential for the local transmission of dengue, Zika, and chikungunya viruses, posing significant public health concerns. *Ae. albopictus* thrives in warm climates, where its life cycle is accelerated, and female longevity is prolonged. Furthermore, as a result from climate change, the increase in global temperatures has led to an increase in the environmental suitability and geographic expansion of the tiger mosquito, especially towards higher latitudes.

Recently, an environmental indicator set was proposed for the Flemish Department of Environment and Spatial Development to monitor the public health impact of climate change. The occurrence of *Ae. albopictus* was selected due to its direct relation with climate change and its significance in disease transmission dynamics. This indicator will remain policy-relevant as the risk for public health increases with an increased occurrence of *Ae. albopictus*.

We developed three sub-indicators with data from the *Ae. albopictus* surveillance and meteorological datasets. Firstly, the distribution of *Ae. albopictus* across Flanders was mapped to depict its presence (establishment or introduction) at each location annually, based on passive monitoring data. Secondly, the overwintering probability in Flanders was calculated using winter temperatures and active monitoring data, indicating the likelihood of *Ae. albopictus* egg survival during the winter at each location per year. Lastly, the seasonal activity duration of *Ae. albopictus* in Flanders was estimated, representing the duration of activity at each location annually.

Visual representation of these sub-indicators through maps and graphs facilitates public accessibility and comprehension. Structural passive and active monitoring programs are essential for monitoring the yearly dynamics of *Ae. albopictus* in Flanders, thereby enhancing preparedness and response to public health risks associated with climate change-induced vector expansion.



S3-P05

The Remote Emerging Disease Intelligence-Network: From Concept to Active Surveillance

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The past decades have seen a dramatic increase of emerging and/or re-emerging infectious diseases worldwide. More outbreaks are foreseen for the future, yet proactive surveillance remains limited due to many challenges including lack of technical expertise and characterization of reliable sentinel sample types for accurate detection of circulating pathogens reflecting potential zoonotic spillover threats. The Remote Emerging Disease Intelligence-NETwork (REDI-NET) is a phased initiative project which aims to enhance current surveillance efforts to detect, predict and contain potential emerging infectious diseases in an efficient and timely manner using real-time long read shotgun metagenomic sequencing surveillance outputs in a flexible, scalable data repository and computing platform to support detection of pathogens, estimate risk of exposure and guide policy-decisions on animal, environmental and public health.

Our core objectives focus on leveraging partner expertise to fill gaps in existing surveillance efforts to include 1) strengthening infrastructure where capacity may need to be built and/or enhanced (remote research stations); 2) standardizing sample collection, storage, and testing processes for assurances in data rigor and big data management; 3) leveraging existing networks and health data to widen the global surveillance footprint; and 4) transferring knowledge on surveillance system activities and providing actionable data. Here we will present program successes to date from early phases and describe next steps in expanding the operational framework with broad applicability across international regions.



S3-P06

Multifactorial characterization of dengue transmission dynamics in the French Caribbean islands to better prepare for future epidemics

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Dengue is now the most widespread arboviral disease with incidence increasing more than 10-fold over the past 20 years, reaching a peak of 5.2 million cases in 2019, according to World Health Organization (WHO). While it is endemic in all tropical and subtropical areas of the world, the Caribbean is one of the most affected regions, especially since the reintroduction of the *Aedes aegypti* vector in the 1970s. Since 2006, all four serotypes of dengue virus (DENV) have been detected during different epidemics, with heterogeneous circulation in the French Caribbean islands (Guadeloupe and Martinique). During the recent outbreak in 2019-2021, three DENV serotypes were detected (DENV-1, DENV-2 and DENV-3), but curiously, their circulation was not homogeneous, with DENV-2 predominating in Guadeloupe, while DENV-3 was the main serotype circulating in Martinique. To investigate if genomic features of serotypes circulating in both islands were different, full genome sequencing was carried out on strains collected during the recent 2019-2021 outbreak in Guadeloupe and Martinique. Phylogenetic data revealed the homogeneous presence of genotype V for DENV-1, cosmopolitan genotype for DENV-2 and genotype III for DENV-3 in both French departments. To determine whether these differences in circulation of these viral strains are linked to a difference in vector transmission capacity, recent evaluations of vector competence of the 3 serotypes circulated during the 2019-2021 outbreak, as well as the current epidemic (DENV-2 only), were carried out on 6 populations (3 in Guadeloupe and 3 in Martinique) of *Aedes aegypti*, the local mosquito vector. Overall, these results confirm the active circulation of DENV in these regions, will help identify the factors inducing the epidemiological differences observed between the two islands and contribute to a better preparedness to cope with DENV emergences in the French Caribbean islands.

Environmental risk factors for *Anaplasma phagocytophilum* in Scotland

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Introduction and Objectives

Anaplasma phagocytophilum (AP) is a complex group of tick-borne pathogens with several ecotypes that vary in their pathogenicity and transmission hosts. The ecological drivers of AP across landscapes often receives less research attention than some other tick-borne pathogens such as *Borrelia*. We aimed to test the hypothesis that higher densities of transmission hosts (sheep and deer) are associated with higher prevalence and hazard of AP ecotypes I and II, while hosts that do not transmit AP I and II (rodents and birds) will dilute prevalence in the tick population.

Materials and Methods

We conducted surveys of questing *Ixodes ricinus* ticks at ~150 sites of contrasting habitats (coniferous, deciduous and mixed woodlands, open habitats and urban green spaces) through much of the UK, especially Scotland, and analysed the ticks for AP using PCR. We estimated deer, sheep, rodent and bird abundance indices at sites in NW Scotland. We conducted a series of general linear mixed models (GLMMs) to identify the environmental factors associated with AP I and II prevalence.

Results, Discussion and Conclusion

The highest prevalence AP sites were in the north of Scotland. Of the AP positive nymphs 86% were ecotype I and 14% were ecotype II. As predicted, higher AP prevalence was associated with higher deer and lower bird abundance. We found no effect of sheep presence, reflecting a need for more sites with sheep abundance data. We found no effect of rodent abundance, perhaps reflect issues with the live trapping method. We found a negative relationship between AP (I+II) prevalence and *Borrelia* prevalence in questing *I. ricinus* nymphs. This may be because red and roe deer transmit AP I and II but not *Borrelia*, while birds and rodents transmit *Borrelia* but not AP I and II.



S3-P08

Wetland development and its effects on avian arbovirus hosts in nearby urban areas

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Introduction and Objective

Wetland abundance is predicted to increase in the Netherlands, to mitigate the effects of climate change in regards to increased precipitation and flood risks. Wetland development may have an effect on local mosquito and bird richness and densities. The development of wetlands might influence the risk of mosquito-borne disease outbreaks, by driving changes in the community structure of wild birds, which serve as hosts for these pathogens. This study aims to understand the effects of wetland development on the community structure of bird hosts in wetlands and nearby cities. Specifically, we will research changes in wild birds that are known hosts for the mosquito-borne viruses Usutu and West Nile.

Material and Methods

We conducted a systematic literature review to understand which European wild birds are most frequently infected with Usutu and West Nile virus. Wetland development was retrospectively mapped throughout the Netherlands for a 20 year period using high resolution Land Use Netherlands maps (LGN). Species occurrence and abundance data was obtained from the Dutch ornithological management institute SOVON. The species occurrence data will be used to analyze changes in host birds in cities near recently developed wetlands.

Results

We expect increased numbers of foraging of known wetland host birds in nearby cities as a result of wetland development.

Discussion

This study described the effect of wetland development on the risk of arboviral host occurrence in nearby urban areas. While this study focused on arboviruses in the Netherlands, this framework can be applied for other bird-borne pathogens and on a global scale as well.

Conclusion

We provided insight into the risks of wetland development for nearby urban areas in regards to the arbovirus host densities and occurrence, which is vital in understanding the interactions between climate change, human health, animal ecology and (emerging) arboviruses.

Unraveling the effect of urban green on mosquito species in the Netherlands

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Introduction

Cities offer diverse landscapes with sometimes unexpected niches for arthropods such as mosquitoes. Greening of urban areas is considered important for human well-being in both living and working environments but could also have implications for the presence of potential resting, feeding- and breeding sites for mosquito species, including vectors of infectious diseases.

Objective

To investigate the effect of urban green on mosquito species. We aim 1) to obtain densities and species composition data of mosquitoes in areas in cities with different percentages of green space, and 2) analyze the densities of *Culex pipiens*, a known vector of Usutu- and West Nile virus.

Methods

To investigate the effect of urban green on mosquitoes, a longitudinal monitoring was implemented between 2020-2022 using adult mosquito traps. Traps were placed in four Dutch cities and sited in three different green categories: 10-20% green, 30-40% green and >50% green. For the city of Utrecht, mosquito monitoring was done for three years in row, for the other cities (Leiden, Zwolle and Arnhem) a single year.

Results

A total of 18 mosquito species were captured during the study. *Culex pipiens/torrentium* and *Culiseta annulata* were the most dominant species in all cities and present in all three green categories. Except for Arnhem, *Cx. pipiens/torrentium* was always more abundant in the greener areas. Species composition and abundance did not follow a clear pattern linking urban green to mosquito presence but instead differed between the cities. In Utrecht, consecutive years also gave varying results between the green categories.

Discussion

Our equivocal results demonstrate the challenges entomologists face when aiming to measure the effect of green space on vectors in the city. We aim to open up a discussion to standardize and streamline these methods for broader use in European (greening) cities and beyond. Better green than sorry.

Interface between invasive hard ticks and weeds in the context of climate change

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Invasive species contribute to the spread of various tick-borne diseases (TBD). These include the invasive weeds which afford tick vectors with an unnaturally beneficial habitat, invasive hard tick species (including those from outside the United States, such as the Asian Longhorned tick and cattle fever ticks), and species of tick native to one region of the U.S. which are spread to another region, and invasive disease hosts (including wildlife species that serve as reservoirs for the diseases of concern). Although climate change combined with an increase in the international trade of domestic and wild animals amplifies the adverse effect of these TBD vectors on food safety and human and animal health, little attention has been paid to the characteristics enabling hard tick species to establish and invade. Commonalities among invasive hard ticks are that they thrive in anthropogenically modified habitats, feed on either domestic animals or wildlife occurring in high density and can survive across a broad range of climatic conditions. In the context of climate change, rise of temperature creates new zones of establishment for ticks by giving them the chance of surviving during winter in high numbers. The cattle fever ticks i.e. *Rhipicephalus annulatus* and *Rhipicephalus microplus*, both species of veterinary importance due to their potential to transmit babesiosis to cattle are good examples of this situation. Along the U.S Mexico border, the invasive *Arundo donax* L. (the giant reed) has been shown to be associated with abiotic and biotic conditions that are favorable to cattle fever tick survival, and persistence especially when compared with other nearby habitats. Based on the above information regarding invasive ticks and weeds, climate change and tick resistance to many acaricides, TBDs can be a serious threat to human and animal health and food security.

Keywords: Ticks - Climate change - Invasive - Resistance.



Ticks in and around Paris: maps of tick's presence and associated tick-borne pathogens

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Introduction and Objectives

Ticks are strict hematophagous acarids that can transmit a high diversity of tick-borne pathogens (TBPs) such as *Borrelia* spp., *Babesia* spp., or the tick-borne encephalitis virus. Environmental, socio-economical and climate changes impact the geographical distribution of these vectors, contributing to the emergence of associated tick-borne diseases. Additionally, urban greening, providing suitable environments for ticks and their hosts, and promotion of outdoor activities increase the risk of encountering ticks and their pathogens.

In that context, our aim is to provide evidence-based recommendations and tick risk maps for the general public on the green spaces in and around Paris, France.

Materials and Methods

Several urban parks, peri-urban woods, green infrastructures and forests, selected on the basis of significant human attendance, were surveyed for questing ticks by flagging the vegetation. Tick collections were performed in 166 sites of 32 different green areas of Ile de France region, in both spring and autumn of 2022 and 2023. TBPs were detected by dedicated PCR and Next Generation Sequencing.

Results

In total, 3456 ticks were collected, most of them being *Ixodes ricinus*. Forest but also peri-urban woods hosted the majority of ticks collected. Urban parks and green infrastructures were not exempt of risk, as ticks were also collected in these areas, even if they seemed to be less suitable for the installation of tick populations. *Borrelia burgdorferi* sensu lato, *Anaplasma phagocytophilum*, *Bartonella* spp., *Rickettsia* spp. and *Babesia* spp. were detected.

Discussion and Conclusion

Whilst the environment is a key parameter in the distribution of ticks and hence tick bite risk and pathogen transmission, exposure is ultimately crucial. Tick abundances and TBP prevalence rates stratified by type of green space will be mapped across the region and contextualised with respect to human frequentation through social media information to generate a population level estimation of risk.

Impact of socio-economic factors on mosquito abundance and diversity

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Arboviral diseases are increasingly prevalent both globally and within the United States. With the absence of effective vaccines and therapeutic treatments, integrated vector management serves as the primary strategy to control mosquito populations and mitigate the spread of vector-borne diseases. Sociodemographic and socioeconomic factors, such as income, education, and housing structure, play pivotal roles in influencing mosquito abundance and transmission risk in endemic regions.

Despite the United States' developed status, approximately 10.5% of its population lives below the poverty line. Taking Gainesville, Florida, as a case study, where 30.6% of residents fall below the poverty threshold with an average per capita income of \$23,018, our project aims to identify sociodemographic and socioeconomic factors that impact mosquito diversity and abundance. Additionally, we seek to gauge residents' perceptions of mosquito burden, mosquito management practices, and vector-borne diseases through a Knowledge, Attitude, and Practices (KAP) survey.

Over a 13-month surveillance period, we collected 5,680 mosquitoes, representing 31 species across 8 genera, including *Aedes aegypti* (n = 1,350), *Mansonia dyari* (n = 1,111), *Culex quinquefasciatus* (n = 1,048), *Aedes albopictus* (n = 330), *Aedes infirmatus* (n = 279), and *Coquillettidia perturbans* (n = 177). Our preliminary findings indicate that income level did not impact total mosquito abundance. However, we observed higher mosquito diversity, particularly during the wet season in higher socio-economic status neighborhoods, and higher *Aedes aegypti* abundance in lower socio-economic status neighborhoods.

Our ongoing data collection for the KAP survey targets 4,304 homeowners, with a current participation rate of 1.8%. Preliminary results suggest a lack of disparities in residents' knowledge and practices but reveal significant differences in their perceptions of mosquitoes, mosquito-borne diseases, and mosquito control measures. These findings underscore the complex interplay between socioeconomic factors, mosquito abundance, and public attitudes toward vector-borne diseases, providing valuable insights for targeted intervention strategies.

Tick- host dynamics underpinning transmission of Kyasanur Forest Disease in degraded forest landscapes in India: insights from MonkeyFeverRisk

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Vector-borne zoonoses pose a major threat to livestock and human health. Rodents are significant wildlife reservoirs of zoonotic pathogens, that adapt to human induced landscape change. Kyasanur Forest Disease (KFD) is a tick-borne, potentially fatal viral zoonosis occurring in the Western Ghats region of India. Human cases of KFD are increasing in number, with recent spread to previously disease-free Indian states. The transmission cycle of KFD is complex, involving multiple vector and host species, with small mammals thought to be important disease reservoirs. However, the ecology of KFD has not been studied since the 1970s, and transmission was considered to be dependent on vectors and hosts associated with forests. Given extensive human modification and fragmentation of the Western Ghats forests, an updated understanding of pathogen-vector-host dynamics, ecological hazard and reservoir roles is critical. To address this, within our MonkeyFeverRisk project (www.monkeyfeverrisk.ceh.ac.uk), rodent and tick sampling was undertaken at 40 study sites stratified across dominant habitats, including forests, and where recent human KFD cases had been reported or where the disease was absent. Ticks were collected from the environment and rodents, and screened for KFD virus. Molecular and morphological methods were used to identify vectors and hosts. Tick vectors and rodents and shrew species previously implicated in KFD transmission were found across habitats, including in close proximity to houses. KFD infected ticks were found predominantly on shrews in plantations as well as on house rats (*Rattus rattus*). Vector and host community composition differed qualitatively from surveys during the last century. Difficulties in morphological identification and the cryptic nature of some vector and host species suggests that molecular methods are needed to understand pathogen-vector-host distributions and human spillover risk in conjunction with data on human behaviour and vulnerability. We discuss the implications of our findings on ecological hazard for disease management strategies.



S3-P14

Assessing the potential impact of anthropization in the forest ecosystem in Southern Cameroon on vector communities and the risk of zoonotic virus emergence

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The accelerating anthropization of the Southern Cameroons forest increases the risk of transmission and emergence of wildlife-borne viruses, especially those transmitted by mosquitoes. This increase in infectious diseases is attributed to human activities that bridge sylvatic and anthropic environments. Mosquito-borne viruses such as Zika, yellow fever, dengue, and chikungunya viruses are a major health burden. Human activities promote the establishment of anthropic mosquito species, changing the composition of native vector communities and increasing the risk of pathogen transferring from wildlife to humans. My PhD project aims to assess the impact of anthropization on mosquito communities in forest ecosystems and how this may translate into an increased risk of novel arboviruses emerging in humans. More specifically, my project aims to characterize mosquito communities and assess the impact of anthropization on their composition and diversity, as well as to delineate the existing and theoretical links (and bridges) between wildlife and humans through mosquitoes. Fieldwork will be conducted in two study sites in Southern Cameroon, each with different anthropization contexts: the Autonomous Port of Kribi (PAK) and the CAMVERT agro-industrial complex. In each area, mosquitoes will be surveyed and collected using BG Sentinel Pro traps along an anthropization gradient to characterize mosquito communities (alpha, beta diversity analysis). Subsequently, the trophic preference of mosquitoes will be studied by determining the blood origin using blood fed females (amplification, sequencing and blast analysis of 16S gene) and double netting devices to study host preference using different host sources (human and animal). Collected arthropods will be screened to search for enzootic virus infections using an NGS method. All results obtained will be analyzed using modeling approaches to explore relationships between anthropization and the risk of new enzootic viruses emerging in humans.

The distribution of *Ixodes ricinus* ticks and their infection with *Borrelia* spp., *Anaplasma phagocytophilum* and *Neorlichia mikurensis* in urban green spaces in Lithuania

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The transformation of natural ecosystems into urban areas becomes a major problem because it could affect the circulation of tick-borne pathogens in cities. Urban green areas such as urban forests, parks, and open grasslands used for recreational activities have recently been identified as high-risk environments for exposure of the human population to infected ticks. Although the distribution of ticks and their infection with pathogens have been studied in Lithuania in natural habitats, the situation in the urban and suburban areas remains largely unknown. This study aimed to investigate the distribution of Ixodid ticks and their infection with *Borrelia* spp., *Anaplasma phagocytophilum* and *Neorlichia mikurensis* in urban green spaces in Lithuania.

A total of 3485 *Ixodes ricinus* ticks were collected from 28 urban parks in Lithuania during 2021-2023. Ticks were screened for the presence of pathogens using triplex real-time PCR assay. Positive samples were further analyzed by PCR and sequencing using different targets of the bacteria genomes. *Borrelia* DNA was detected in 25,6% of collected ticks. Five *Borrelia* species *B. garinii*, *B. burgdorferi* s.s., *B. afzelii*, *B. lusitaniae* and *Borrelia miyamotoi* were identified. *A. phagocytophilum* and *N. mikurensis* were present in 4.9% and 7.7 % of the samples, respectively. Double and triple co-infections with different pathogens were detected. The prevalence of infection differed among years, urban and peri-urban parks, and it was higher in adults than in nymphs. The present study highlights the risk of tick-borne diseases in urban green spaces in Lithuania. *N. mikurensis*, a newly emerging pathogen of significant public health importance, was detected in Lithuania for the first time.

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Variation in the spatial distribution of *Aedes aegypti* and *Aedes albopictus* in Madagascar

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Aedes aegypti and *Aedes albopictus*, both vector of arbovirus, co-exist in Madagascar. *Ae. aegypti* has recently been reported to have originated from the islands of the southwest Indian Ocean, while *Ae. albopictus* is native to south-east Asia. Their geographic distribution and the epidemiology of the diseases they transmit are affected by variable climatic conditions. Changes in the distribution of these two species have been noticed in recent decades. This study aims to assess the dynamics of both species and identify the factors driving their distribution. For that, available unpublished and published data on the *Aedes* of Madagascar from 1904 to 2016 were compiled. Data from literature based-review, plus mosquito database from the Institut Pasteur de Madagascar and personal communications from close collaborators in Institut de Recherche pour le Développement were compiled. Furthermore, data from the longitudinal survey on adult and immature stages of *Aedes* (*Stegomyia*) carried out from February to December 2019 in 25 districts of five bioclimatic domains of Madagascar were also used. The relationship between these entomological data and climatic, environmental, topographical and demographic data was analysed using statistical models. Our results demonstrated that low vegetation cover significantly determines the presence of *Ae. Aegypti* in Madagascar, with a slight variation in its climatic suitability over time. On the other hand, *Ae. albopictus* tends to colonise all five bioclimatic domains, and changes in relative humidity and rainfall over time influence its distribution. Human population density has no effect on neither presence nor absence of both species. Both species are climate-sensitive, but the adaptation speed does not seem to be the same. Joint species analyses need to be carried out in order to study the niche dynamics and niche overlap between them, as well as to compare their rate of invasion according to changes in their distribution area.

Identifying socio-ecological drivers of tick-borne zoonosis impacts for disease management: insights from a OneHealth systems approach to Kyasanur Forest Disease in India

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Kyasanur Forest Disease (KFD) or Monkey Fever, is a tick-transmitted haemorrhagic viral infection that can be fatal to humans. Endemic to the Western Ghats mountain region in southern India, the disease has undergone considerable range expansion since 2012. Like many other neglected zoonoses, KFD disproportionately affects resource-poor communities that rely on the increasingly degraded local forest ecosystems for their livelihoods, and up to 500 cases are reported each year. The KFD disease system is highly complex, with numerous tick, and vertebrate host species suspected to play a role in the virus transmission cycle. How these, and human priorities and behaviour contribute to zoonotic disease risk is poorly understood. Our Indo-UK collaboration, MonkeyFeverRisk (www.monkeyfeverrisk.ceh.ac.uk) took an interdisciplinary, stakeholder-focused One Health approach to identify critical knowledge gaps and needs of local communities and disease managers alike. We gathered empirical data both on the habitat associations and seasonal abundance of potential KFD vectors and hosts, and why, how, and when these habitats are frequented and used by the local communities. We could demonstrate for the first time that KFD risk extends beyond forests, into agricultural areas and villages, and incriminated known and new tick and small mammal species being implicated in KFD virus transmission. Our social surveys and knowledge holder and policy mapping exercises identified which seasonal livelihood activities render certain groups amongst the local communities vulnerable to increased tick contact rates, and enabled us to identify barriers to uptake of adaptive measures against disease risk. Disentangling the socio-ecological mechanisms driving disease exposure was vital to co-produce guidance and decision support tools with stakeholders from the public health, animal health and forestry sectors, enabling them to design improved disease intervention strategies and helped design tailored, health-critical information material to empower local communities avoid exposure to the KFD virus (<https://doi.org/10.1079/onehealthcases.2023.0011>).

West Nile virus monitoring in Flanders (Belgium) during 2022-2023 reveals endemic Usutu virus circulation in the wild bird population

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Introduction and Objectives

In recent years, Europe has seen a rise in West Nile virus (WNV) and Usutu virus (USUV) cases. Wild birds, serving as key reservoirs for WNV and USUV, often act as crucial indicators for the introduction and spread of these viruses. Currently, there is no durable large scale monitoring for WNV in Belgium, and specific monitoring for USUV is lacking. In Flanders, passive WNV monitoring in wild birds has been in place for many years, while active monitoring efforts started in 2022. Here, we present the results of a limited study conducted during the vector seasons of 2022 and 2023 in Flemish wild bird populations to actively and passively monitor the prevalence of WNV and additionally assess the presence of USUV.

Methods and Results

Several RT-qPCR's were employed for virus detection, revealing the absence of WNV-RNA during both vector seasons. Conversely, USUV-RNA was identified in 2022 through active surveillance, affecting two (5.5%) out of 36 birds (*Corvus corone*) and in passive surveillance, impacting eight (72.7 %) out of 11 birds (*Turdus merula* (6) and *Rhea pennata* (2)). WGS and phylogenetic analysis of the virus in the positive *Rhea pennata* indicated its placement with the Africa 3 lineage. In 2023, active surveillance identified 16 (7.2 %) USUV-RNA positive birds (*Buteo buteo* (1), *Turdus merula* (14) and *Athene noctua* (1)) out of 222 examined birds, while passive surveillance detected two (7.1 %) positive birds (*Turdus merula*(1), and *Larus marinus* (1)) out of 28.

Discussion and Conclusion

This restricted WNV monitoring effort in Flanders did not reveal WNV presence, but found indications of an endemic USUV circulation in Belgium. It is crucial to intensify monitoring efforts for WNV in the coming years, considering its endemic status in several European countries and its expanding geographical range in northern Europe.

S3-P19

Urban adaptation, host behavior, and distribution affects the presence of *Dirofilaria* parasites: a phylogenetic comparative study of wild carnivores

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Dirofilariosis is an emerging mosquito-borne disease among domestic carnivores, wildlife, and humans. Our understanding of its ecological drivers, geographical distribution, and prevalence in sylvatic hosts is limited. We explored several factors that potentially drive the presence of *Dirofilaria* spp. in 70 species of wild carnivores in the Palearctic region, using published presence/absence infection data and phylogenetic comparative analysis. We found a positive relationship between mammals exploiting urban habitats and infection presence, additionally we show that diurnal species exhibit a lower likelihood of infection when compared to nocturnal species. Lastly, larger geographical distribution range was positively associated with parasite presence. Our results suggest that urban habitats can be important reservoirs of emerging infectious diseases. By understanding how urbanization affects wildlife health, we can better manage and mitigate potential disease transmission between wildlife, domestic animals, and humans, reducing the risk of zoonotic diseases.



S3-P20

***modis2fast* : an R package for efficient access to satellite-based MODIS (and similar) data used in landscape eco-epidemiology**

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Satellite-based data from the MODIS sensor, such as temperature or vegetation indices, are increasingly being used in landscape epidemiology research (especially in data-poor areas), e.g. to model the spatio-temporal distribution of vectors or to better understand their bio-ecology. However, due to their size and spatio-temporal granularity, these data can be difficult to access and process, especially when large time series are required. Here we present *modis2fast*, an R package that provides a simple and efficient way to download MODIS data (as well as VIIRS and GPM data) in R. Built upon the robust open-source *OPeNDAP* framework, *modis2fast* allows the user to subset any MODIS product directly at the download stage, using spatial, temporal, and band/layer filters ; thus reducing the downloaded data and disk usage to the strictly necessary. In addition, *modis2fast* supports parallel downloads. This package is therefore particularly useful for retrieving and processing large MODIS time series at their finest spatial and temporal resolution. In this poster, we also present some practical examples of the use of *modis2fast* for research on mosquito-borne diseases. *modis2fast* naturally promotes digital sobriety in our research. The package is available at <https://github.com/ptaconet/modis2fast>.

Studying the effects of *Aedes* invasive and native mosquito species on the circulation of Filarial nematodes in a hungarian southwestern city

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Introduction and Objectives

Dirofilaria immitis and *Dirofilaria repens* are mosquito-transmitted Filarial nematodes with increasing human infections that may be linked to the successful spreading of invasive mosquito species like *Aedes koreicus* and *Aedes albopictus* in Europe. In Hungary, these mosquitoes have been continuously monitored since their emergence. Thus, the aim of our study was to monitor *Dirofilaria* infection in mosquitoes in an area known for human infections and to understand the relationship between mosquito infestation, seasonality, and species.

Materials and Methods

Mosquitoes were collected in Pécs, Hungary from May to October in 2022 and 2023. After trapping and identifying mosquitoes, they were tested using real-time PCR for *Filaria* screening. Positive pools were further examined using conventional PCR, gel electrophoresis and Sanger sequencing to identify the *Filaria* species.

Results

4444 specimens in 1015 pools of mosquitoes were processed, including AIM and native species. A total of 30 pools belonging to different genera, with *Aedes vexans* and *Culex pipiens* being the most common, tested positive for *Filaria* spp.. The majority of positive samples occurred in June-July in both 2022 and 2023. Our results so far have confirmed 2 cases of *D.repens* (*Ae.vexans* and *Culiseta annulata*), one case of *D.immitis* (*Cx.pipiens*), while the rest of the positive samples were infected with *Setaria tundra* and *S.labiato-papillosa*.

Discussion and Conclusion

Our study found a seasonal pattern in *Filaria* infection but no clear link between *Aedes* species and *Dirofilaria* infestation in mosquitoes. The dry season in 2022 may have affected mosquito activity and *Filaria* presence in 2023. The presence of *S.tundra* near human cases requires further investigation. Our focus was on the prevalence of *D.repens* and *D.immitis* as emerging zoonotic pathogens. More research is needed to understand the relationship between mosquito species and *Filaria* prevalence and we are committed to ongoing surveillance and investigating human infections.

Influence of climate change and infection on thermal preferences of mosquitoes

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The impact of temperature on vector-borne disease dynamics has been studied intensively, especially to model the effect of climate change. However, these studies used macroclimatic data and did not consider whether vectors select specific microhabitats. With this project, the preferred temperatures of mosquitoes were investigated in different setups. The results show that mosquitoes behaviourally thermoregulate, preferring cooler resting temperatures than their physiological optimum proposed in the literature. Interestingly, *Ae. aegypti* infected with *D. immitis* showed a cold preference at day nine after infection, which presumably is a pathogen-induced stress response against the migration of the larvae 3 of the pathogen into the proboscis. Another set of thermal preference experiments was performed with *Ae. aegypti* lines selected for 20 generations at 24°C or 30 °C and acclimated each at these temperatures. Cold selected and warm acclimated mosquitoes showed a higher abundance in the warmest temperature zone. Since mosquitoes select microhabitats by thermal preference, and this preference is adaptable, it is important to include thermal preference with its adaptability in future models.



S4-P01

Microbiome and host resistance to a trematode parasite: the case of a vector snail

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The expansion of *Pseudosuccinea columella* snails worldwide has boosted *Fasciola* transmission. Most *P. columella* are susceptible to *F. hepatica* but a few field-occurring populations are resistant to the parasite in association with a potent immune response possibly based on gene-for-gene interactions. The microbiota plays a critical role in training the host's immune system, while the immune system orchestrates the maintenance of key features of host-microbe symbiosis. Hence, we hypothesized that a distinct microbiome signature exists in resistant *P. columella* in relation to its genetic/phenotypic identity that is maintained across ecological contexts and that adds host fitness for resistance. The microbiome, the infection and the genetics of resistant and susceptible populations were characterized at G0 (field generation; occurring in sympatry or allopatry) and G2 (2nd lab-generation). Overall, the microbiome composition of resistant snails was site-specific but the microbiome of resistant differs from susceptible snails, even in sympatry. A clearer genotype-wise segregation was observed in lab-G2. Spatio-temporal shifts (G0-G2) reduced microbiome diversity, homogenizing variability and conserving population/phenotype identity. Dysbiosis after experimental exposure to *F. hepatica* was observed, following similar kinetics but still completely segregated phenotype wise. Parasite exposure also resulted in specific microbiota signatures that varied as infection/exposure progresses. An association of resistant snails with certain genera (*Pirellula*, *Luteobacter*, *Roseomonas*) and Phyla (Genmetimonadota, Halanaerobiaeota) was depicted. To answer if the microbiome has a significant effect on host resistance to *F. hepatica* infection in *P. columella*, experimental approaches of microbiome transfer from one phenotype to the other, in cross-combinations, are ongoing.

Virulence and transmission vary between Usutu virus lineages in *Culex pipiens*

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Usutu virus (USUV) is a zoonotic arbovirus infecting mainly wild birds. It is transmitted by ornithophilic mosquitoes, mainly of the genus *Culex* from birds to birds and to several vertebrate dead-end hosts. Several USUV lineages, differing in their virulence have emerged in the last decade and now co-circulate in Europe, impacting human populations. However, their relative transmission and effects on their mosquito vectors is still not known. We thus compared the vector competence and survival of *Culex pipiens* mosquitoes experimentally infected with two distinct USUV lineages, EU2 and EU3, that are known to differ in their virulence and replication in vertebrate hosts. Infection rate was variable among blood feeding assays but variations between EU2 and EU3 lineages were consistent suggesting that *Culex pipiens* was equally susceptible to infection by both lineages. However, EU3 viral load increased with viral titer in the blood meal while EU2 viral load was high at all titers which suggest a greater replication of EU2 than EU3 in mosquito. While their relative transmission efficiencies tended to be similar, positive correlation between transmission and blood meal titer was observed for EU3 only. Contrary to published results in vertebrates, EU3 induced a higher mortality to mosquitoes (i.e. virulence) than EU2 whatever the blood meal titer. Therefore, we found evidence of lineage-specific differences in vectorial capacity and virulence to both the vector and vertebrate host which lead to balanced propagation of both viral lineages. These results highlight the need to decipher the interactions between vectors, vertebrate hosts, and the diversity of arbovirus lineages to fully understand transmission dynamics.

Unraveling Mechanisms of ISV-Arbovirus Interactions in Mosquito Cells

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Mosquitoes are important vectors for many arthropod-borne viruses (arboviruses), such as Chikungunya (CHIKV) and Zika Viruses (ZIKV). In addition to arboviruses, many insect-specific viruses (ISVs) have been discovered in mosquitoes in the last decade. ISVs, in contrast to arboviruses transmitted by mosquitoes, cannot replicate in vertebrate cells. ISVs are considered a potential intervention strategy to reduce the arboviral burden due to their possible interference with arbovirus infection leading to a reduction of arbovirus transmission. The mechanisms of ISV interference with arbovirus infections are not yet well understood. Here, we investigated the interactions of two different ISVs (Eilat Virus-EILV and Agua-Salud alphavirus-ASALV) with different arboviruses (CHIKV and ZIKV) in mosquito cell lines. Both EILV and ASALV interfere with CHIKV but facilitate ZIKV infection only when these ISVs were persistently infecting the cells. However, no interference was observed in acutely infected cells. Using this model with different interference phenotypes, we investigated the mechanisms of interference using transcriptomics, small RNA sequencing and functional analyses. Our results showed differences in EILV -host RNAi pathways interactions between acute and persistent infections. Furthermore, we identified several transcripts that were only differentially expressed in the arbovirus-infected cells, which showed the interference phenotype. Silencing experiments were used to determine the importance of the identified targets regarding the observed interference phenotype on CHIKV. Overall, our results underscore the complexity of virus-host interactions in mosquito cells and highlight the pivotal role of ISVs in modulating arboviral transmission dynamics. By investigating the molecular mechanisms of ISV-mediated interference, our study provides crucial insights into harnessing the potential of ISVs as novel intervention strategies against arboviral diseases.



S4-P04

Mosquito infection dynamics by Rift valley fever virus

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Over the past three decades, one third of (re-)emerging viral diseases have been caused by arboviruses, a group of pathogens transmitted from arthropod vectors to vertebrate hosts during blood feeding. Rift Valley fever virus (RVFV) is a mosquito-borne arbovirus that was first described in 1930 in the Rift Valley, East Africa. RVFV is widespread in Africa and has caused outbreaks outside the African continent, in the Arabian Peninsula (2000) and in Mayotte (2018). For one hundred years, RVFV has caused hundreds of human deaths, significant losses in livestock and major socio-economic impacts in exposed countries, making RVFV one of the top ten priority pathogens of World Health Organization. Moreover, RVFV can be transmitted by a wide range of mosquito vectors, particularly *Aedes* and *Culex* genus, putting a large part of the world at risk, including Europe.

Many gaps remain in our understanding of RVFV transmission potential, which depends on the complex interplay between host, vector and virus parameters under the influence of environmental factors. We notably have a limited understanding of mosquito vector competence for RVFV, defined as the mosquito's ability to become infected and transmit the virus. The aim of my PhD project is to assess the impact of key virus features (*e.g.*, cellular origin of virions, infectious doses) on mosquito vector competence for RVFV. To this end, mosquitoes were exposed to an artificial blood meal containing different doses of RVFV derived from different cell lines, then mosquito infection, dissemination and transmission rates were measured overtime by infectious titration assays to estimate RVFV intra-vector dynamics in mosquitoes.

Our results highlight the importance of each of the parameters tested on vector competence and guide future research on the main determinants of RVFV transmission by mosquitoes as well as their impact on RVFV epidemiology.



S4-P05

First detection of Crimean–Congo haemorrhagic fever virus in *Hyalomma marginatum* ticks, southern France

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Ticks, which are vectors of several zoonotic pathogens, represent an important and increasing threat for human and veterinary health. *Hyalomma marginatum*, one of the main tick vectors of the Crimean-Congo Haemorrhagic Fever (CCHF) virus, has been present in Corsica for decades. Given the recent establishment of this tick species in continental France, it was crucial to evaluate the epidemiological situation of CCHFV in France. Transmission of the CCHFV to humans occurs predominantly via bites of *Hyalomma* ticks, especially *H. marginatum* and *H. lusitanicum* in Europe, or via exposure to infected blood or tissues from viraemic animals or humans. As ticks are the only known natural reservoirs of CCHFV, we focused on field collection of ticks. We collected ticks from horses, which are considered to be the preferred hosts of *H. marginatum* and on cattle, which are considered as good amplifiers of CCHFV and thus enhance local virus circulation. We collected ticks in the Pyrénées-Orientales department where antibodies against CCHFV were identified in 2021-2022 from cattle. Cattle farms with the highest within-herd seroprevalences were selected, as well as a few seronegative farms in the same areas. In addition, farms with horses, located in the neighbourhood of the seropositive cattle farms were also visited. The ticks *H. marginatum*, analyzed molecularly, revealed the presence of CCHFV in this department, in proportions ranging from 3.1% to 55.8% of infected ticks across positive sites. All CCHFV isolates sequenced in this study were highly identical and belonged to the same genotype (genotype III). This finding confirms for the first time the transmission of CCHFV in France and highlights the need for close monitoring of *H. marginatum* in areas where the tick is already established, and further investigations into its probability of geographic expansion.



S4-P06

Deciphering the role of skin bacterial odours in the attraction of mosquitoes using *in vivo* and *in vitro* studies

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When arriving at a short-range from potential blood hosts, body odours play a significant role in the final host selection of mosquitoes. Interestingly, many of these body odours released from the skin are produced by the bacterial communities present on the skin. It has been shown in various systems that mosquitoes are more attracted to blood hosts with bacterial communities that have a low(er) diversity and a high(er) bacterial density. In addition, the presence of several specific bacterial species or strains may increase or decrease their preference. Furthermore, recent studies show that certain (components of) skin bacteria can play a significant role in determining the severity of a mosquito-borne infection. We aim to unravel the impact of host skin bacteria on the attraction to mosquitoes and the establishment of a mosquito-borne virus infection in a mammalian host. We will steer these interactions by a selective killing of bacteria with bacteriophage-encoded lysins as novel tool. In this presentation I will give an overview of the larger project and I will present data on behavioural experiments in which we studied the preference of *Ae. aegypti* towards skin bacterial volatiles in a newly developed two-choice setup with video tracking.



S4-P07

Vector competence of Belgian *Culex pipiens* and *Anopheles plumbeus* mosquitoes for West Nile Virus

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An increased circulation of West Nile virus (WNV) has been reported in Europe during the last decade. Although there have thus far been no reported cases of WNV infections in Belgium, several neighboring countries reported WNV cases in animals and humans, making risk assessments of high importance with regard to a preparedness plan. Therefore, we determined the vector competence of field-collected *Culex pipiens* and *Anopheles plumbeus* mosquitoes for WNV. Both species could significantly contribute to WNV transmission upon an introduction of the virus due to their proven competence in other countries (*Cx. pipiens*) and their biting behavior (*An. Plumbeus*). Mosquitoes were exposed to a blood meal containing 7.13×10^7 PFU/mL WNV (strain Israel 98, lineage 1) and incubated for 14 days at a constant temperature of 25°C, 80% relative humidity, and 16L:8D cycle. Subsequently, infection rate (IR), dissemination rate (DR) and transmission rate (TR) were determined by detection of viral RNA via qRT-PCR in abdomen, head-wings-legs and saliva, respectively. Results indicated that *Culex pipiens* exhibited an IR of 20%, DR of 55%, and TR of 50%, while *Anopheles plumbeus* showed an IR of 31%, DR of 42%, and TR of 13%, with survival rates of 93% (54/58) and 43% (62/143), respectively. These findings suggest that both mosquito species are competent vectors for WNV at 25°C, with overall transmission efficiencies of 5.6% and 1.6%, respectively. Virus isolations are ongoing to confirm the presence of infectious virus in the collected samples while further investigations will explore the impact of temperature gradients on vector competence, reflecting current summer conditions in Belgium (gradient of 15/25°C). These efforts will provide valuable insights into the dynamics of WNV transmission and inform the development of effective preparedness and control strategies to mitigate the risk of WNV outbreaks in Belgium.



S4-P08

The West Nile virus lineages circulating in Spain can be transmitted by *Culex pipiens* reared in two different types of water

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West Nile virus (WNV) is a mosquito-borne orthoflavivirus that is kept in an enzootic cycle mainly between birds and mosquito vectors. Occasionally, it affects humans and equines causing encephalopathies. *Culex pipiens* (Linnaeus, 1758) is one of the main vectors of WNV in Europe. Recently, some studies have pointed out that vector's microbiota may influence in arbovirus transmission. Rearing water is one of the major sources of microorganisms belonging to mosquito microbiota. Therefore, our main goal was to assess the influence of two distinct types of rearing water on the vector competence of *Cx. pipiens* for two WNV lineages circulating in Spain. *Cx. pipiens* field-collected larvae from Barcelona province were reared in the laboratory under summer environmental conditions, split into two groups, one reared with dechlorinated laboratory water and the other with water from a stream. Ten-to-12 days old females from these groups were fed with chicken blood doped with WNV lineages 1 and 2 (7 log₁₀ TCID₅₀/ml). Engorged females were sacrificed at 3, 7, 14, 21 and 28 days post-exposure (dpe), and body, head and saliva were extracted to assess infection, dissemination and transmission rates and transmission efficiency. *Cx. pipiens* was experimentally confirmed as a competent vector for WNV (both lineages 1 and 2), with no significant differences between the two rearing conditions. Both lineages were able to infect and disseminate within *Cx. pipiens* mosquitoes, and they were able to be transmitted since infectious virus was isolated from the saliva of disseminated individuals. Transmission efficiency was null in early time-points. From 14 dpe onwards, it ranged from 2.94% to 30% in WNV-1 and from 2.33% to 13.51% in WNV-2. Overall, *Cx. pipiens* showed to be competent to transmit WNV, only restricted significantly by the time of exposure to the virus.

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S4-P09

Worldwide distribution of *Candidatus Borrelia capensii* in ticks associated with seabirds

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Introduction and Objectives

Relapsing fever spirochetes (RF, *Borrelia*) are known pathogens for humans, often transmitted by soft ticks. Compared to their sister group, the Lyme disease spirochetes (*Borrelia*), our understanding of their transmission cycle, natural reservoirs, spatial scale of circulation, wildlife role and impact, and vectors is less comprehensive. Our work focuses on the circulation of *Borrelia* in seabirds and their associated ticks. Sporadic studies have reported phylogenetically similar isolates in seabird species from South Africa and Eastern Asia, in association with an ill bird, and present in a soft tick. Its association with a human clinical case (then named *Borrelia* sp. K64) and the ecological proximity between seabirds and humans raise particular interest in the context of zoonotic disease emergence.

Material and Methods

We investigated the distribution of *Borrelia* in ticks associated with seabirds. We screened 1,944 individual tick samples of the *Ornithodoros capensis* species complex, collected globally from nests of 22 seabird species, using PCR and qPCR assays. Additionally, we examined *Borrelia* circulation by testing avian blood samples in a colony where tick infestation is high.

Results, Discussion and Conclusion

Our findings confirmed the presence of *Borrelia* in 19 locations, spanning diverse geographic areas up to 7,600 km apart, with 62 positive samples (3.19%). No blood samples were positive. Further characterization was performed using a multi-locus typing approach. Most samples, associated with 5 seabird genera, clustered with the *Borrelia* sp. K64 isolate. Genetic distance analysis with known RF bacteria revealed that these isolates represented a distinct species, leading us to propose the species *Candidatus Borrelia capensii*. Studies are now needed to validate tick vector competence and examine the impact of this *Borrelia* species on seabirds. Additional colony-level surveys are also called for to evaluate the risk of exposure for human populations that live near to nesting seabirds.



S4-P10

Tissue tropism convergence among tick major nutritional symbionts: insights from preliminary results

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Introduction

Nutritional symbiosis is crucial for tick growth, with many tick species associating with intracellular bacteria providing them B vitamins, such as *Coxiella*-like endosymbionts (CLE) or *Francisella*-like endosymbionts (FLE). Understanding the tissue tropism of these endosymbionts is vital for unraveling the complex relationships between ticks and their microbial partners.

Objective

This study examines tissue tropism convergence among CLE and FLE endosymbionts across various soft and hard tick species: *Ornithodoros moubata*, *Ornithodoros maritimus*, *Dermacentor marginatus*, and *Dermacentor reticulatus*.

Material and Methods

Both male and female specimens of these tick species were dissected to analyze their Malpighian tubes, testes or ovaries, salivary glands, midgut, and body. Subsequently, qPCR was used to assess CLE and FLE distribution in organs,

Results

Preliminary findings reveal a convergence in tissue tropism, particularly notable in the Malpighian tubes and ovaries, where CLE and FLE endosymbionts were consistently abundant regardless of the tick species.

Discussion

Ovarian infection indicates maternal transmission of endosymbionts to developing oocytes, leading to a typically higher quantity of CLE or FLE in females compared to males. The high density in Malpighian tubules supports their nutritional role, potentially in B vitamins biosynthesis.

Conclusion

Further research is required to elucidate the significance of these organs in nutritional symbiosis.



S4-P11

Spatio-temporal re-colonization of top-predators modulates zoonotic infections in ticks

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Introduction and Objectives

Predator community can change abundance and composition of their preys, affecting emergence and spread of tick-borne infectious diseases for which some preys (e.g., ungulates, rodents) are often common hosts. We aim to evaluate if sites with gradual temporal re-colonization of a top-predator, the wolf (*Canis lupus*), exhibit different infection prevalence in ticks.

Materials and Methods

We identified four study areas (NV = Non Valley, PP = Paneveggio-Pale di San Martino Natural Park, LV = Laghi Valley and GV = Giudicarie Valley) located in the Autonomous Province of Trento (Italian Alps), each characterized by a temporal gradient of re-colonization from east-to-west of wolf in recent years. Specifically, in NV the species is considered stable since 2017, in PP since 2019, in LV since 2021, while yet sporadic in GV. During 2023 in each study area, we collected host-seeking ticks in forested habitats by dragging the vegetation. PCR-based methods were used to detect infection of *Borrelia burgdorferi* s.l., *Anaplasma* spp. and *Babesia* spp. in ticks.

Results, Discussion and Conclusion

During this first year, we collected 394 *Ixodes* spp. ticks across the study areas. We preliminary obtained higher prevalence of *Borrelia* spp. in areas historically colonized by wolves (NV: 28.72%; PP: 15.48%), compared to where wolf is sporadic (LV: 13.11%; GV: 11.70%) and the opposite for *Anaplasma* spp. (NV: 1.06%; GV: 4.25%). *Babesia* spp. prevalence rates did not show any specific pattern. We speculate that the established presence of wolf may interfere differently with the circulation of tick-borne pathogens, both indirectly affecting *Borrelia*-competent rodent hosts' predators, such as foxes, or directly by hunting *Anaplasma*-competent hosts, such as ungulates.

These preliminary findings suggest that predator community may have a crucial role in modulating zoonotic disease circulation by affecting preys local presence or abundance that will be incorporated in our forthcoming analysis.

Blood Source Impact on WNV lineage1 transmission dynamics in *Culex pipiens*

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Introduction

West Nile virus (WNV), a mosquito-transmitted orthoflavivirus, has increased in prevalence across Europe, with a northward expansion. Birds, both resident and migratory, are primary reservoirs, with other vertebrates, including humans and equids, being incidental dead-end hosts susceptible to infection, occasionally with fatal outcomes. Vector competence (VC) experiments are instrumental in understanding virus transmission in mosquito vectors, although the results can be influenced by experimental design variables. This study examines the potential influence of different vertebrate blood sources on WNV VC in *Culex pipiens* mosquitoes.

Materials and Methods

Female mosquitoes ($n=20$) were offered an infected blood meal containing WNV (strain NY99, 3.11 x 10⁷ PFU/ml), utilizing four blood sources per group: horse (defibrinated), sheep (defibrinated), chicken (antibiotic treated), and rabbit (antibiotic treated). Fourteen days post-infection (DPI), surviving females were dissected, and body, wing/legs, and saliva samples were collected and processed individually. Viral RNA was detected using a WNV RT-qPCR.

Results, Discussion and Conclusion

Bloodfeeding rates varied from 84.2% (17/20) for chicken blood to 95% (19/20) for horse blood, but were not significantly different (p -value, 0.7705). Survival rates (SR) at 14-DPI were 94.7% (18/19), 66.6% (12/18), 82.3% (14/17) and 94.7% (18/19) for horse, sheep, chicken and rabbit groups respectively, showing slightly significant difference (p -value, 0.0486). Mosquitoes that ingested chicken and rabbit blood exhibited a significantly higher infection rate (IR) (42.8% and 22.2%, respectively), compared to those that ingested horse and sheep blood (5.5% and 8.3%, respectively) (p -value, 0.0425). Although antibiotic treatment for some blood sources could be an important factor, the SR variability and IR difference between chicken and rabbit groups, both treated, suggest that further intrinsic characteristics of the blood may influence biological responses in mosquitoes.



S4-P13

A virus-induced gene silencing (VIGS) approach to study plant-geminivirus-insect vector interactions and virus transmission

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Geminiviruses (family *Geminiviridae*) are plant viruses with small circular single-stranded DNA genomes (mono- or bipartite) encapsidated in geminate virions and transmitted by insect vectors in a persistent circulative manner, causing devastating crop diseases globally. In the Mediterranean, the monopartite geminivirus Tomato yellow leaf curl virus (TYLCV, genus *Begomovirus*), and invasive recombinants, all transmitted by the whitefly *Bemisia tabaci*, severely impact tomato and vegetable yields. Recently, bipartite begomoviruses like Tomato leaf curl New Delhi virus (ToLCNDV), prevalent in India and East Asia, have also been detected in the Mediterranean. In a collaborative project funded by the Indo-French foundation CEFIPRA and the Occitanie foundation RIVOC, we investigate mechanisms of circulative transmissions of TYLCV and ToLCNDV by invasive whitefly species, Middle East Asia Minor 1 (MEAM1) and Mediterranean (MED). Effective control of geminiviruses is targeted to their insect vectors, necessitating a thorough understanding of virus circulation and potential replication within the vectors. Our research employs Nanopore sequencing, transcriptomics, and small RNA-omics to study viral DNA replicative forms, viral transcripts, and small RNAs in MED and MEAM1 whiteflies transmitting monopartite versus bipartite begomoviruses. We use a virus-induced gene silencing (VIGS) approach, in which begomovirus betasatellite-based VIGS vectors were designed carrying inserts of the candidate plant and insect genes. Plants infected with helper begomovirus and betasatellite VIGS constructs targeting plants and insect genes are used as source plants for virus acquisition by the whitefly, followed by transmission tests. Initial tests confirmed the efficient replication and transmission of empty VIGS constructs. Subsequently silencing the plant PCNA gene led to severe developmental abnormalities, indicating its role in DNA replication. On-going experiments, aim to silence the whitefly PCNA gene and two other genes implicated in begomovirus transmission. The ultimate goal is to develop new strategies to control viral disease spread at the transmission step without affecting insect viability.

Occurrence of tick-borne pathogens in ticks collected from wildlife and domestic animals in northeastern Italy

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Northeastern Italy is considered endemic for tick-borne pathogens (TBPs) and wildlife and domestic hosts play an important ecological role in their maintenance. The aim of this study was to investigate the occurrence and prevalence of TBPs in ticks removed from wildlife and domestic animals and assess the ecological and reservoir role of these hosts.

Ticks (adults, nymphs and larvae) were collected from both wildlife and domestic hosts through passive surveillance in endemic areas of northeastern Italy during 2019–2023. Nucleic acids extraction was performed automatically and TBPs detection (i.e., TBE virus, *Anaplasma phagocitophilum*, *Babesia* spp., *Borrelia* spp. and *Rickettsia* spp.) was carried out with Real Time PCRs followed by sequencing. Ticks were removed from 98 animal hosts belonging to 13 species. A total of 537 ticks were collected: 312 *Ixodes ricinus* (58.1%), 147 *I. hexagonus* (27.3%), 1 *I. canisuga* (0.18%), 1 *D. marginatus* (0.18%), 1 *Rhipicephalus sanguineus* (0.18%), 2 *Ixodes* spp. (0.37%) and 2 *Dermacentor* spp. (0.37%).

None tick sample resulted positive for TBE virus, whereas 12 TBPs were detected in 110 out of 466 pools (420 adults, 40 nymphs and 4 larvae). *Ixodes ricinus* was found positive for at least one TBP in 99/318 samples (31.1%): *A. phagocitophilum* 60 pools (12.9%); *R. helvetica* 31 pools (6.6%); *R. monacensis* 14 pools (3%); *Borrelia* spp. 11 pools (10%; 7 *Borrelia miyamotoi*, 3 *B. afzelii* and 1 *B. burgdorferi* s.s.) and 4 pools for *Babesia* spp.

One *Dermacentor* spp. was found positive for *R. slovaca* and seven *I. hexagonus* for *Rickettsia* spp. According to these data, the main tick species in this study was *I. ricinus* and one third of these resulted positive at least for one pathogen. Moreover, in the study area there is a high occurrence of wild species underlining their role in the epidemiology of TBDs.



S4-P15

Insecticide resistant *Anopheles* from Ethiopia but not Burkina Faso show a microbiome composition shift upon insecticide exposure

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Malaria remains a key contributor to mortality and morbidity across Africa, with the highest burden in children under 5. Insecticide based vector control tools, which target the adult *Anopheles* mosquito are the most efficacious tool in disease prevention. Due to widespread use of these interventions, insecticide resistance is now ubiquitous across Africa. Understanding the underlying mechanisms contributing to this phenotype is necessary to both track the spread of resistance and to design new tools to overcome resistance. Here, we show that the microbiome composition of insecticide resistant populations of *An. gambiae*, *An. coluzzii* and *An. arabiensis* originating in Burkina Faso from field caught populations and colonies across two countries show no changes in microbial composition compared to their susceptible counterparts. In contrast, *An. arabiensis* from Ethiopia demonstrates clear differences in microbiome composition in those dying and surviving insecticide exposure. To further understand resistance in the *An. arabiensis* population from Ethiopia, we performed RNAseq on field caught, exposed mosquitoes. Taken together, these results shed light on the contribution of the microbiome to insecticide resistance in some settings.



S4-P16

Abstract Title: Unveiling Tahyna Virus Transmission: Vector Competence of Mosquitoes from Europe

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Introduction and Objectives

Tahyna virus (TAHV) was the first isolated arthropod-borne virus (arbovirus) in Europe, and has subsequently been found across Asia and Africa. Infections are mostly asymptomatic but can cause “valtice fever”, characterized by influenza-like symptoms, and primarily experienced by children. In severe cases, neurological symptoms may manifest. Studies conducted in the 1970s revealed a high seroprevalence in endemic areas, reaching up to 90% among an elderly population in former Czechoslovakia. The virus is maintained in an enzootic cycle between small mammals and mosquitoes, with *Aedes vexans* presumed to be the vector in Europe. However, recent and comprehensive studies on the vector competence for TAHV are limited.

Materials and Methods

Therefore, we investigated a panel of mosquito species: *Aedes aegypti*, *Ae. albopictus*, *Ae. japonicus japonicus*, *Ae. koreicus*, *Culex pipiens* biotype *pipiens*, and *Cx. torrentium*. The mosquitoes were orally infected and subjected to a salivation assay 14 days post-exposure.

Results

We demonstrated that TAHV can infect all tested species. However, the infection rate is generally higher in the genus *Aedes*, with the highest rate of 70% observed in the invasive species *Ae. albopictus*. This species is also able to transmit the virus at a transmission efficiency (positive saliva per mosquito) of 3.3%. Furthermore, our study revealed that a decrease in temperature reduces the infection and transmission rate.

Discussion and Conclusion

These results suggest transmission of TAHV by *Ae. albopictus* in Europe, which is possible to increase by the ongoing geographical expansion of this mosquito species.



S4-P17

Interactions of insect-specific viruses (*Bunyavirales*) with the mosquito host and Rift Valley Fever Virus

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Mosquitoes are not only important vectors for arboviruses but also carry insect-specific viruses (ISVs). ISVs naturally infect mosquitoes but in contrast to arboviruses, cannot infect vertebrates. Some ISVs have the potential to be used in vector or arbovirus transmission control, as they can be pathogenic to mosquitoes or interfere with arbovirus infection in mosquitoes. While many ISVs have been discovered in the last decade, our understanding of their interactions with their hosts is very limited. ISVs can influence mosquito vector ecology through modulating lifespan, reproductive success, and susceptibility to arboviruses. Recent research suggests that ISVs phylogenetically closely related to arboviruses can inhibit the replication of these viruses. The order *Bunyavirales* encompasses numerous ISVs and arboviruses, such as Rift Valley Fever Virus (RVFV). RVFV is spread by different *Aedes* and *Culex* mosquitoes. In this study, we characterized the growth kinetics and interactions of three ISVs from this order in *Aedes* and *Culex*-derived cells, followed by their infection characteristics in mosquitoes. In all tested mosquito cells, Herbert and Gouleako viruses replicated to high titers. Additionally, the Herbert virus interferes with RVFV infection in AF5 cells. We further investigated the dissemination and infection of ISVs in *Aedes aegypti* mosquitoes. Badu and Gouleako viruses can infect *Aedes aegypti* through oral feeding and intrathoracic inoculation. We further investigated their dissemination patterns in *Aedes* for insights into their transmission routes and maintenance in nature, as well as their interactions with arboviruses in mosquito organs. Overall, our results show interference between ISVs from the *Bunyavirales* order and RVFV for the first time and shed light on the previously unknown host range and dissemination pattern of these ISVs.

Arbovirus infection modulates deltamethrin susceptibility of *Culex pipiens*

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Several flaviviruses outbreaks, such as Dengue, West Nile and Usutu viruses, have become more worrying due to extensive circulation, a greater associated mortality in animals, and frequent neuroinvasive infections in humans. Because of the lack of protective vaccines, targeting the mosquito vectors with insecticides is often the only affordable measure to control transmission. Several studies evidenced that insecticides exposure can modulate vector-pathogen interactions and impact the control of disease transmission. As the number of approved insecticides for public health is very limited in France and Europe, the increased risk of flavivirus transmission has become serious enough to warrant evaluation of the efficacy of insecticides. We have investigated the interactions between deltamethrin exposure and infection by Usutu virus in *Culex pipiens*. We evaluated whether Usutu infection influences the susceptibility of mosquito to deltamethrin. After feeding mosquito with USUV infectious blood, they were exposed to impregnated paper using WHO test tubes 7 and 13 days later. Feeding on infectious blood did not affect deltamethrin susceptibility at 7 days however it reduced the deltamethrin associated mortality at 13 days. This reduction occurs late in the extrinsic incubation period when the virus reaches the mosquito saliva and the transmission to the next host may be possible. We further analyzed the presence of Usutu virus in mosquito head and salivary glands and no difference were found between dead or surviving females suggesting. These findings indicated that feeding on blood containing USUV rather than the infection itself may have induced physiological changes that reduced deltamethrin susceptibility. Further research is ongoing to investigate the mechanisms behind the protective effect of infection against the insecticide effect and we discussed the impact on the risk to human and animal health.

First evidence of flavivirus circulation in Greater Mouse-eared bats *Myotis myotis* (Borkhausen, 1797) in Slovakia

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Introduction and Objectives

Bats have recently received increased attention as natural hosts and reservoirs of many pathogens, and have been shown to host zoonotic pathogens. In addition, bats biological characteristics such as their ability to fly, their social behaviour, their immunity, and their migration or hibernation increase their capacity as pathogen reservoir. The main objective of this study was to provide a serologic screening of possible presence of antibodies to West Nile virus (WNV) and Usutu virus (USUV) in Slovak bat population.

Materials and Methods

The Greter Mouse-eared bats included in the study originated from a natural colony located in Drienovská Cave (Slovak Karst National Park). The bats were captured and handled by a licensed chiropterologist (LK) under the Permission No. 3051/219-6.3 from the Act. No. 543/2002 from Act. No. 543/2002 of the code on nature and landscape protection, granted by the Ministry of Environment of the Slovak Republic. At all, we examined 64 bat serum samples using competitive ELISA test (Ingezim WNV Compac[®]) and by microtitration virus neutralization test.

Results, discussion and Conclusion

Using ELISA test, we found 1 dubious sample for the presence of antibodies to WNV. The virus neutralisation test performed on bat serum samples confirmed 1 positive bat on the presence of antibodies to USUV. This is the first time USUV infection was detected in bats in Slovakia. If our study is the first to look for flaviviruses in bats from the Slovak Karst, some previous studies has been conducted at this locality, demonstrating presence of WNV, USUV, tick-borne encephalitis virus and Tribeč virus in wild birds (Csank T et al. 2019, Korytár et al. 2020) and mosquitoes (Peňazziová et al. 2021).

Acknowledgments

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Characterizing the bacterial microbiome of the invasive vector *Aedes albopictus* in Hungary: a pilot study using Oxford Nanopore sequencing

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Introduction and Objectives

The microbiome of mosquitoes plays a critical role in vector competence, influencing their capacity to transmit pathogens. Understanding the bacterial communities within vector species is essential for developing strategies to control vector-borne diseases. In this pilot study, we aimed to investigate *Aedes albopictus* mosquitoes' bacterial microbiome and reveal dominant bacterial genera with their potential impact on vector competence in the Hungarian populations.

Methods

In the analysis, we used 12-12 mosquito specimens originating from two different populations in the country's southwestern part. To profile the bacteria composition, Oxford Nanopore sequencing was performed using a standard 16S rRNA-based metagenome approach. Sequence data were classified with Kraken2 software using the Silva database.

Results

Our findings revealed a diverse bacterial landscape, with distinct dominant genera in mosquitoes from each location: Members of *Wolbachia* (71%) and *Acinetobacter* (18%) genera dominated in individuals from one area, while *Zymobacter* (42%) was also represented in the other population. Besides, *Escherichia-Shigella*, *Stenotrophomonas*, *Delftia*, and *Halomonas spp.* were detected (less than 5% ratio).

Discussion and Conclusion

This research marks the first comprehensive profiling of the bacterial community structure in Hungarian *Ae. albopictus* populations. Although *Wolbachia* is the most studied bacterial symbiont in this species, the occurrence of further bacteria in hematophagous groups is poorly documented. Our study highlights the complexity and local variation in the microbiome of mosquitoes, which may influence their vectorial capacity and interactions with pathogens. As a follow-up, we plan to analyse the microbial interaction network to reveal co-exclusionary patterns.

Understanding interactions between humans, mosquitoes, waters and plastics: an interdisciplinary approach in Montpellier, France

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Today, plastic pollution has become ubiquitous. Fragmentation and degradation of macroplastics create microplastics, known for their direct hazards but also for their capacity to serve as carriers of contaminants and microorganisms. This persistent pollution threatens numerous organisms across ecosystems, potentially impacting also disease vectors such as mosquitoes. In this context, our research project aims to untangle the intricate connections between microplastic pollution, environmental contexts (spanning urban, peri-urban, and 'natural' settings), and the life history traits and microbiota of mosquitoes - a crucial factor influencing their vectorial capacity.

From this project, an interdisciplinary initiative encompassing ecology and anthropology has emerged, studying and focusing on the dynamic interplay between humans, mosquitoes, plastics, and waters in Montpellier (France). To achieve this, 20 semi-structured interviews were conducted with a diverse array of stakeholders, primarily associated with the research project. They were complemented by participant observations of various stages of the project, both in the field and in laboratory settings. These methodologies aim to yield qualitative insights into participants' perceptions regarding mosquitoes, waters, and plastics, as well as the intricate links, or lack thereof, between these elements, associated practices, and the attendant challenges.

These interviews have unveiled multifaceted perspectives on mosquitoes, encompassing concerns about their biting nuisances, potential for disease transmission, alongside recognizing their ecological significance within food chains. However, perception of plastic pollution was more homogeneous among the surveyed individuals. Another pivotal objective of the project was to shed light on the actors of the project, processes, and methodologies employed in generating scientific findings, tracing the trajectory of water and mosquito samples from the field to the lab. Our interviews unveiled that stakeholders possess a nuanced comprehension of the research project, influenced by their specific roles within it. Consequently, enhancing communication seems essential for research initiatives involving a consortium of individuals from diverse institutes.



S4-P22

The social construction of *Aedes albopictus* as a public problem: views cross between daily nuisances and public health issues in Occitania

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The proliferation of the tiger mosquito in Occitania has become a major issue, perceived as both a daily nuisance and a public health problem. This presentation examines how citizens and institutions approach this issue, based on a qualitative study of forty interviews and a questionnaire aimed at the region's citizens. These surveys revealed a divergence between the perceptions of authorities, who view the tiger mosquito as an infectious risk, and citizens, who see it more as a nuisance. This thesis highlights two parallel forms of public action: formal institutional actions and more informal citizen actions, occurring in domestic spaces through adaptive behaviors. These behaviors result from a three-stage process established by citizens: identifying the problem, assessing the risk, and implementing solutions. The qualitative survey revealed four types of coping behavior: fight, live with, avoidance, and disinterest, illustrating the variety of citizen responses to the tiger mosquito problem. Analysis of these adaptation forms tends to refute the idea that public policy is rationally transferred from decision-makers to citizens. However enlightened, a policy becomes public when citizens engage with it or are affected by it. Citizens' choices and adjustments are not merely direct, rational outcomes of decisions made by policy-makers. Consequently, these formal (institutional) and informal (citizen) public actions address the same problem, with differing definitions but potentially coinciding solutions. This approach helps us better understand the interactions between individual and collective actions and institutional strategies. By examining how citizens define and respond to the tiger mosquito issue, the study sheds light on the social dynamics surrounding its management in Occitania. It also questions the processes implemented by citizen.



S4-P23

Non-native game birds released for recreational shooting amplify Lyme disease risk via spillback.

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Biological invasion is a potentially important driver of zoonotic disease emergence. Spillback, when invaders act as reservoirs for endemic pathogens, is considered a key mechanism by which invaders amplify zoonoses. However, few studies directly measure spillback (i.e. changes in pathogen prevalence in native hosts or disease vectors) associated with invasions. Furthermore, invasions are typically correlated with other potential disease-promoting factors, such as anthropogenic disturbance, making it difficult to disentangle the two processes. Here we capitalise on a quasi-experimental, replicated release of common pheasants (*Phasianus colchicus*) for recreational shooting to quantify the impact of this non-native invader on Lyme disease risk. *Borrelia* sp. prevalence in questing *Ixodes ricinus* ticks was almost 2.5 times higher in pheasant-release woods compared to control woods, whereas nymph abundance was unaffected. Our study provides the strongest evidence to date that biological invaders can amplify zoonotic disease risk via spillback in an ecological relevant context.

Ticks and flies as vectors of *Bartonella* spp., *Mycoplasma* spp. and *Rickettsia* spp. pathogens in cats

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The presence of companion animals alongside humans can introduce feline ectoparasites such as ticks and fleas and the associated feline vector-borne pathogens they carry into our shared environment. Domestic cats, being vulnerable to infestation by these ectoparasites, can serve as potential transmitters of diseases to humans. Despite their asymptomatic nature, feline vector-borne pathogens warrant urgent investigation, often being overlooked as diagnosis typically relies on presumptive assessments based on the animals' exposure to ectoparasites. Monitoring the prevalence of these pathogens can serve as an early warning system for outbreaks, facilitating timely intervention.

This study assessed the infection of domestic cats and their ectoparasites with vector-borne pathogens - *Bartonella* spp., *Mycoplasmaspp.* and *Rickettsia* spp., identifying strains and genetic diversity through molecular techniques. This study marks the first identification of two causative agents of cat scratch disease, *B. henselae* and *B. clarridgeiae* and ruminant-associated *Bartonella* sp., haemotropic *Mycoplasma* bacteria - *M. haemofelis* and 'Ca. *M. haematominutum*' and diverse *Rickettsia* pathogens - human pathogenic *R. helvetica*, *R. conorii* subsp. *raoultii* and *R. felis*, in domestic cats and their ectoparasites in Lithuania.

Microbiota-nutrition-physiology interactions in *Aedes* mosquitoes: treat and trick?

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The interrelationships between microbiota, nutrition, and host physiology have gained significant attention in biomedicine. Although these interactions are complex, it is now clear that the “right” gut microbiota contributes to a balanced diet by influencing the nutritional requirements, nutrients allocation and metabolism, which in turn ensures optimal juvenile growth, immunity and lifespan. Hence, fully understanding these interactions in biomedically relevant species could inform the design of novel strategies to limit disease burden. To date, however, the microbiota-nutrition-host interactions have been mostly studied in model organisms. My laboratory has focused on the mosquito *Aedes aegypti*, the main vector of many human pathogenic viruses such as dengue [DENV], chikungunya [CHIKV], and Zika [ZIKV] viruses, which are global public health threats and economic burdens. We uncovered a crucial role of sugar feeding in mosquito antiviral immunity which in turn decreases vector competence for arboviruses. Since *Ae. aegypti* can almost exclusively feed on blood in some natural settings, our findings suggested that this lack of sugar intake could increase the spread of mosquito-borne arboviral diseases. In addition, we showed that the magnitude of the sugar-based protection is limited by the microbiota, shedding light on the importance of nutrition and microbiota interactions on mosquito vector competence. I will present recent data suggesting that the microbiota may limit sugar response to benefit the mosquito female’s physiology, which in turn may be detrimental to humans by increasing arbovirus transmission rate.

Potential Vectors of Epizootic Hemorrhagic Disease (EHD) in Europe: Insights from Italy

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Introduction and Objectives

Epizootic Hemorrhagic Disease (EHD) is a *Culicoides*-borne viral disease affecting ruminants. In 2022, EHDV-8 made its first appearance in Italy. To understand which *Culicoides* spp. could potentially transmit the virus, a thorough entomological field survey was conducted on affected farms in Sardinia. In 2023, vector competence studies with EHDV-8 were also carried out on Italian populations of *Culicoides imicola* and *Culicoides obsoletus/scoticus*.

Materials and Methods

During the field survey, *Culicoides* were collected using blacklight traps at four outbreaks sites, identified, and sorted in pools for real-time RT-PCR testing. For competence studies, live *Culicoides obsoletus/scoticus* from the Abruzzo region and *C. imicola* from the Sardinia region were collected and fed under laboratory conditions. After the extrinsic incubation period, surviving females were individually tested using real-time RT-PCR. A multiplex PCR based on internal transcribed spacer 2 ribosomal DNA sequences (ITS2) was used to distinguish between *C. obsoletus* and *C. scoticus*.

Results

During the field survey, a total of 411 pools (comprising 5,721 midges) were tested. Out of these, ten pools of *C. imicola*, four of *C. obsoletus/scoticus*, two of *C. pulicaris*, one of *C. newsteadi*, and one of *C. bysta* tested positive for EHDV-8. Following the extrinsic incubation period, 122 *C. obsoletus/scoticus* and 61 *C. imicola* survived. Among them, four *C. scoticus*, one *C. obsoletus*, and six *C. imicola* tested positive for EHDV-8.

Discussion and Conclusion

According to our findings, EHDV seems to use similar transmission patterns as BTV: *C. imicola* may act as main vector in the Mediterranean region, while *C. obsoletus* and *C. scoticus* could potentially spread the virus in Northern Italy and Europe. During the outbreaks, EHDV-8 was also detected in *C. newsteadi*, *C. pulicaris*, and *C. bysta*. The discovery of *C. bysta* as a potential vector of orbiviruses represents a novel finding, necessitating further confirmation through additional research.



S4-P27

The vector competence of *Synopsyllus fonquerniei* and *Xenopsylla brasiliensis* fleas from Madagascar to transmit the plague bacillus

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Xenopsylla cheopis and *Synopsyllus fonquerniei* are the two species of flea recognized as vectors of human plague in Madagascar, where the disease is still present. Another species, *X. brasiliensis*, a plague vector in continental Africa, is also present. Although *X. cheopis* is widely studied, the vector competence of *S. fonquerniei* and *X. brasiliensis* for the transmission of the plague bacillus is limited, if not unknown. Two transmission mechanisms are known: proventricular blockage and Early Phase Transmission. This blocking capacity has been observed in *X. cheopis*, but has not yet been demonstrated in the other two species present in Madagascar. Here, we aimed to study the transmission mechanisms of *Yersinia pestis* by these two species. To this end, fleas from the insectary were allowed to take an infected blood meal using an artificial feeding device, then fed on uninfected mouse blood three times a week for one month. After each uninfected meal, we monitored the flea mortality and proventriculus blockage (a process important for the transmission of the agent of plague). We found that the mortality and blockage rates were similar between both species, 30% vs 39% and 13% vs 14% for *S. fonquerniei* and *X. brasiliensis* respectively. Therefore, our data suggest that *S. fonquerniei* and *X. brasiliensis* display the same vector competence in our experimental conditions. This information enables us to better define the risk of plague in Madagascar.

Impact of timing on co-infection dynamics of bluetongue virus serotypes 13 and 17 in *Culicoides* biting midge cells

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Bluetongue (BT) is a vector-borne viral disease of ruminants with an estimated annual loss of 3 billion US\$ worldwide. BT is caused by bluetongue virus (BTV), which has a segmented, double-stranded RNA viral genome. To date there are 29 recognized BTV serotypes. Reassortment is an important driver of BTV evolution and emergence as multiple serotypes circulate in susceptible animal populations every vector season. The primary BTV vectors are *Culicoides* spp. biting midges. Midges blood feed every 3-4 days over their lifespan and may ingest more than one serotype either while feeding on a co-infected animal in a single blood meal or ingest different serotypes over time. To determine the impacts of co-infection timing on viral dynamics and progeny genetics in a *Culicoides* midge cell line (W8), we first simultaneously infected cells (MOI=1) with BTV-13, BTV-17, or BTV-13 & BTV-17. We then tested the impact of staggered timing by infecting W8 cells with one serotype then a second serotype at 3 days post-infection. Infectious virus titers from simultaneously co-infected cells were similar to cells infected with the serotypes individually, but only RNA from the BTV-17 VP2 gene was detected, suggesting that it outcompeted BTV-13. During staggered co-infection, cells infected with BTV-17 followed by BTV-13 had higher virus titers compared to cells infected with BTV-13 followed by BTV-17. BTV-13 VP2 RNA was significantly lower than BTV-17 when cells were infected with BTV-17 followed by BTV-13. However, cells infected with BTV-13 followed by BTV-17 showed no significant difference in serotype-specific VP2 detection. Thus, initial infection with BTV-17 enhanced viral infection and suggests that co-infection dynamics in cell culture are dependent on both timing and serotype. A whole genome analysis of progeny viral sequences will provide insight into the impact of co-infection timing and serotype on potential reassortment.



S4-P29

***Aedes aegypti* vector competence for two genetically divergent Kenyan Rift Valley fever virus isolates**Victoria SY (victoria.sy@pirbright.ac.uk), Philie HOLLINGHURST, Simon GUBBINS, Isabelle DIETRICH*The Pirbright Institute, Woking, United Kingdom*

Rift Valley fever (RVF) is a zoonotic vector-borne disease affecting mainly ruminants and humans, with mosquitoes of the genus *Aedes* and *Culex* acting as main vectors. Rift Valley fever virus (RVFV) is endemic in sub-Saharan Africa and the Arabian Peninsula but has significant potential to spread to other regions. The 2006 RVF outbreak in Kenya caused an unprecedented number of cases and high human fatality rate. Several RVFV isolates have been recovered from this outbreak. The aim of this study was to compare the mosquito vector competence for one of the Kenya-RVFV-2006 isolates to that of an ancestral isolate, Kenya-RVFV-1983. We hypothesize that genetic differences in the virus genome could impact vector competence and could have contributed to the 2006 outbreak magnitude. To test this, we used two genetically modified viruses differing in the M segment of the genome: RVFV-M-1983 and RVFV-M-2006, to infect *Aedes aegypti* via blood meal. Blood-fed mosquitoes were incubated for 7 and 12 days and 30 mosquitoes per virus isolate were sampled at each timepoint. Legs and wings were dissected from the rest of the body and all samples were tested for virus infection. Infection and dissemination rates were calculated and used to compare vector competence. We found that the infection rate was significantly higher in mosquitoes infected with GM-virus RVFV-M-1983 compared to RVFV-M-2006. However, there were no significant differences in dissemination rate between viruses. Thus, differences in viral replication dynamics in mosquitoes are not likely to account for the high human case load and fatality rate seen during the Kenya 2006 outbreak, with the limitation that we were not able to determine transmission rate for the two GM-viruses. Vector competence data generated in this study together with data generated from other in vitro and in vivo studies will be used to model RVFV transmission dynamics.



S4-P30

Exploring the interaction between Tick-Borne Encephalitis Virus (TBEV) and *Ixodes ricinus* ticks after infection via an Artificial Blood Feeding system

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Tick-Borne Encephalitis (TBE) is an emerging zoonotic tick-borne disease in Europe, caused by TBE virus (TBEV). The main vector in Europe are *Ixodes ricinus* ticks. Ticks can become infected at each life stage when feeding on a host, mostly by co-feeding. Upon infection, TBEV is maintained in the tick for the rest of its life and the virus can be transmitted to a naive host during the subsequent blood meal.

The TBEV replication cycle has been mainly studied in mammalian cells, while the replication, spread and maintenance in ticks has not been fully elucidated. This research aims to study TBEV interactions with *I. ricinus* ticks.

The first objective was to develop an artificial blood feeding system that would allow to infect ticks *in vitro* under controlled conditions. A feeding method was optimized using a membrane mimicking the host skin and tested with field-collected ticks. After optimization of several parameters like membrane thickness, blood origin, blood additives and feeding stimuli, the *in vitro* blood feeding improved and resulted in an attachment rate of adult ticks up to 95% (19/20) after 5 days of feeding with no mortality. The feeding was not fully finished but led to the laying of eggs by two females.

After optimization of the blood feeding system, TBEV was added to the blood in order to infect the ticks. The addition of TBEV reduced the attachment rate but did not affect the feeding of the attached ticks, although this needs to be evaluated with more ticks. Initial qPCR analysis of ticks that were attached to the membrane for 5 days revealed the presence of TBEV.

The optimized feeding system and the successful TBEV infection of ticks are the start of additional experiments that will be performed to study virus replication, spread and maintenance in tick organs over time.



S4-P31

First detection and molecular characterization of *Hepatozoon canis*, in *Rhipicephalus sanguineus* sensu lato ticks collected from dog shelters in central Morocco

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Introduction and Objectives

Rhipicephalus sanguineus s.l. commonly known as the brown dog tick, is the most dominant ectoparasite in dogs worldwide, which is involved in the transmission of various pathogens, including those of zoonotic interest. The aim of this study was to molecularly investigate the occurrence of pathogens in ticks collected from heavily infested shelter dogs in Morocco.

Materials and Methods

Ticks individuals were collected between April and October 2023 from 314 dogs in three dog shelters (DS) located in urban (DS1, Rabat), peri-urban (DS2, Ras El-Ma), and rural (DS3, Shoul) areas in central Morocco of which 10% of the unengorged specimens were screened for bacteria belonging to *Anaplasmataceae* family and piroplasmids by conventional PCR targeting the *16s rRNA* and *18s rRNA* genes respectively.

Results

A total of 5363 *Rhipicephalus sanguineus* s.l. were collected. The seasonal pattern of ticks exhibited three peaks of activity occurred in April, August and September for adults, as opposed to nymphs, which peaked in August. Almost all specimens were collected in DS3. *Hepatozoon* spp. DNA was detected in 36.4% of the pools tested (20/55), four of which were successfully sequenced and showed 99–100% identity with *Hepatozoon canis* (7.3%). However, All DNA pools from ticks were negative for *Ehrlichia* spp. and *Anaplasma* spp.

Discussion and Conclusion

The high tick infestation is related to poor sanitary conditions and lack of antiparasitic treatments in the three DS. The circulation of *H. canis* is reported for the first time in Morocco, corresponding to the clinical signs observed in dogs, such as anemia, fever and lethargy. A study is underway on carried pathogens in dogs from the three DS (n=144).



S5-P01

The effect of pyrethroid bednets on lifelong blood feeding and egg laying success in mosquitoes carrying the malaria transmission-blocking symbiont *Microsporidia* MB

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Introduction

New mosquito control strategies that mitigate the threat of insecticide resistance are needed for malaria elimination. The microsporidian symbiont *Microsporidia* MB has revealed the potential for an additional control measure as it naturally spreads and blocks malaria transmission in *Anopheles arabiensis* mosquitoes. Here we assess whether the use of insecticide-treated nets (ITNs) maximize the impact of such strategy or prevent MB dissemination.

Methods

The lifelong blood feeding performance of microsporidia MB-positive mosquitoes is compared to MB-negative mosquitoes (both colonies derived from the field) after a sublethal exposures to a standard ITN (PermaNet 2) and a new-generation dual-ITN (PermaNet 3). Using custom-made-2-way tunnels, mosquitoes are tested for their capacity to avoid the presence of insecticide, to blood feed and lay eggs multiple times in life. Blood feeding success, blood meal size, number of eggs laid per female, and offspring size are measured daily.

Results

The PermaNet 3 kills all exposed mosquitoes regardless of resistance status and microsporidia infection. The PermaNet 2 reduces lifelong blood feeding successes, blood meal size, egg laying episodes and the total number of eggs. No substantial difference is found between MB-positive and MB-negative mosquitoes regarding the blood feeding successes and number of eggs laid. The presence of microsporidia is associated with the overexpression of GSTs (GST 1-1 and GSTd5) which can protect against oxidative stress and insecticide exposure, a potential competitive advantage for MB-positive mosquitoes in nature (for integrated release programs).

Discussion

The dissemination of MB-positive mosquitoes in areas with high insecticide coverage seems a viable option to control malaria transmission. But confirmation is needed from semi-field work where the sublethal effect of insecticide on mating, flying patterns and foraging is analysed. As resistance mutations carry fitness costs associated with reproduction and survival, the role of the microsporidia MB in conferring resistance needs further investigation.



S5-P02

Optimisation of tiger mosquito control in Barcelona

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Tiger mosquito (*Aedes Albopictus*) is a well-known vector of multiple diseases, such as dengue, Zika or chikungunya. Due to this, on top of being a nuisance to humans, it poses a threat to public health in urban areas. Therefore, mosquito control is of great importance to both citizens and authorities.

In this work, we propose and study a presence-absence model for mosquito larvae in Barcelona. The model consists of a system of ordinary differential equations, the parameters of which are either taken from the literature or fit using real presence-absence data of water drains that have been periodically visited by the Agència de Salut Pública de Barcelona in the years 2019-2023. The model incorporates real climatological data from various weather stations, along with the geographic locations of the breeding sites.

Water drains are distributed in 8 risk zones. The model aims at assessing the likelihood of tiger mosquito infestation in these zones based on the zone characteristics, such as the size and distribution of the water drains. Using numerical optimization algorithms, optimal visiting schedules are determined for each risk zone. These schedules balance treatment efficacy with resource allocation, maximizing the interval between visits to minimize the burden on the surveillance agency.

While the aim is to develop a useful tool for effective mosquito control management in the city of Barcelona, by allowing cheaper surveillance and better resource allocation, the findings of this study have the potential to mitigate the health risks associated with mosquito-borne diseases also in other urban environments. The modeling framework and methodologies employed here can be extrapolated to other contexts and geographic locations.



S5-P03

Development of dPCR and NGS methods for improved surveillance and detection of new mosquito densoviruses

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The use of mosquito densoviruses (MDVs) is considered a promising way to control mosquitoes through insecticide/MDV combinations or in combination with the sterile male technique (boosted SIT). Existing scientific literature mostly focuses on prevalence studies in mosquito populations from limited geographical regions, or on experimental studies evaluating larvicidal efficacy of specific MDV strains, and overall some gaps remain about the diversity, biological as well as geographical distribution of MDV worldwide. To date, only a few MDV candidates have been identified and tested as effective control tools, and methods for monitoring MDV and its spread in wild mosquito populations or persistence in the environment are still lacking. The lack of such tools limits the ability to monitor MDV dynamics and evaluate its efficacy as a control measure in real time. We have developed two complementary methods to detect MDV in wild populations. First, we screened field mosquito populations collected to detect the presence of MDV using an NGS approach based on Illumina sequencing of targeted viral sequences. Second, we developed a pan-MDV monitoring tool using dPCR to detect viruses belonging to the two genera known to infect mosquitoes (*Protoambidensovirus* and *Brevihamaparvovirus*). Although dPCR allows accurate virus detection, it currently does not allow sequencing of positive droplets, limiting our ability to identify viruses based on sequence analysis. To overcome this limitation, we investigated the possibility of using double emulsion PCR, which allows droplet recovery, positive sorting and sequencing. I will present the results obtained using both NGS and dPCR approaches on mosquito collections from Gabon, Camargue and Mexico.

Production and supply of *Aedes albopictus* sterile males

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Aedes albopictus (Skuse) is one of the most dangerous invasive mosquito species with a high vectorial capacity for several arboviruses. It is therefore necessary to control and reduce the diffusion of this species but traditional control methods are not effective enough. The Sterile Insect Technique is a promising method which consists in the production and release of a large number of sterile males, which mate with wild virgin females that subsequently lay infertile eggs.

The SIT mass rearing facility established at Centro Agricoltura Ambiente in Crevalcore, Italy, has the capacity to produce 1 million males per week, and in the summer 2023 produced more than 700,000 sterile males a week.

Larvae are reared in racks with 50 trays each and pupae collected after 160 hours from egg seeding, male pupae are separated from the females by means of an automatic sex-sorter and X-ray irradiated. The male pupae are left to emerge inside emergence tubes with netted top and bottom placed into trays filled with water. Sugar solution is provided in each tube for the adult nourishment. The tubes are maintained for two/three days to allow complete adult emergence.

In case of short distance transportation sterile males can be released directly from the tubes by car, while for long distance transportation the tubes are stacked into a refrigerated cabinet ($8\pm 1^{\circ}\text{C}$) for about 15 minutes to cold shock anesthetize the males for easy packaging and shipping by express courier. To maintain the temperature in the range $10\text{-}12^{\circ}\text{C}$ over 24 h transportation, adequate quantity of phase changing materials (PCM gel packs) are included in the package.

In 2023 sterile males were supplied to Portugal, Croatia, Serbia (supported by IAEA), Switzerland (SUPSI and ISIDORE), Greece (Benaki Phytopathological Institute), Germany (ICYBAC), Zagabria (Institute of Public Health) and to Italy (supported by the Municipality of Bologna).

Biting Back! Wild harvested mosquitoes as an alternative protein ingredient in broiler feed: Insights from the first pilot study

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Introduction

Approximately 3.5 billion people are at-risk of mosquito-borne diseases each year. Disease outbreaks and nuisance have intensified, particularly in regions of heightened agricultural activity, where conditions favor massive mosquito population production. Significant resources are allocated globally towards mosquito control (mostly insecticides and repellents), costing billions of dollars annually. Sustainable mosquito management solutions are needed that do not rely solely on insecticides. Mechanical removal of mosquitoes via mass trapping could contribute to reducing mosquito populations and the costs associated with chemical control. Wild-caught mosquitoes can be upcycled back into the agricultural industry as a nutritional source for animal feed. The exploration of such alternative sources has become important due to the increasing environmental toll of current feed production systems. To start exploring this new feed paradigm we performed a series of studies aiming to a) assess the nutritional composition and safety of wild-caught mosquitoes, b) investigate the effect of a mosquito-based diet on poultry health, meat quality and consumer acceptance, and c) design a mass-trap device for insect harvesting.

Materials and Methods

Wild-harvested mosquitoes (*Culex*, *Aedes*, *Anopheles*) trapped from rice-fields were used in all microbiological/nutritional assessments. Mosquitoes were incorporated into a standard broiler's (Cobb hybrid broilers) diet (10% soybean meal substitution). Chickens were randomly placed in a control and a mosquito-fed group.

Results and Discussion

The mosquito-based diet exhibited an overall highly nutritious profile. Chicken meat quality attributes and animal health were comparable between the control and the treatment group, while the organoleptic evaluation showed that the thighs from the mosquito fed group had the highest overall consumer acceptance. Based on this pilot study mosquitoes showed potential as a protein source for broiler feed. Additional research is warranted to investigate further the nutritional value and safety of wild-harvested insect pests and optimize mass trapping technologies (for maximum insect collection efficiency).

Partial clearance of pre-established *Plasmodium falciparum* infection in mosquitoes by feeding on patients treated with Artemether+Lumefantrine + Atovaquone-Proguanil

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Background

Effective treatment of malaria involves targeting both the disease-causing stages and the transmissible sexual stages to impede further transmission and prevent drug resistance. The present study relies on a clinical trial testing the benefits of combining Atovaquone Proguanil (AP) with Artemether-lumefantrine (AL) in the treatment of uncomplicated malaria. AP treatment has been shown to block *Plasmodium falciparum* transmission by affecting gametocyte carriage and transmission to mosquitoes. Additionally, exposure to atovaquone has been found to impact pre-established *P. falciparum* infection in mosquitoes. Here, we aimed to assess the impact of meal intake on blood from patients treated with AL versus AL+AP for pre-infected mosquitoes on the further development of parasites until sporozoites.

Methods

We collected infectious blood meals from naturally infected donors in Burkina Faso and fed them to laboratory-reared female *Anopheles gambiae*. Four days later, these mosquitoes were exposed to a second blood meal containing plasma from patients treated with either AL+placebo or AL+AP, collected at eight time points from Day 0 to Day 28.

Results

A total of 9,259 mosquitoes were dissected, with 4,850 at 7 days and 4,409 at 14 days post-infection (dpi). Our preliminary analysis shows that in *P. falciparum*-infected mosquitoes, a blood meal with plasma from AL+AP treated patients significantly inhibits parasite development. This inhibition includes a decrease in infection prevalence, reduced infection intensity, and slower sporogonic development. Plasma from patients treated with AL alone had no effect. The impact of AL+AP was significant for plasma samples collected between 5 hours and 5 days post-treatment. Further analysis, including correlation with drug dosage and sporozoite quantification in salivary glands, is underway.

Conclusion

These findings underscore the benefits of triple ACT containing AP, as treated patients may help reduce the vector competence of the local mosquito population and contribute to partial community protection against malaria transmission.



S5-P07

Insecticidal activity of essential oils of *L. alba* harvested in Burkina Faso and Côte d'Ivoire against mosquito vectors

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Mosquito resistance to chemical insecticides and their persistence in the environment promoted the research on natural plant extracts to determine their potential as alternative tools for mosquito control. A study carried out by our team showed that the essential oil of *L. alba* harvested in Abidjan had insecticidal effect against mosquito vectors. As the biological activity of an essential oil may vary according to its chemotype, the aim of this study was to determine the phytochemical composition of essential oils of *L. alba* harvested in Bobo-Dioulasso (LA-BF) and Abidjan (LA-CI) and their insecticidal activity on adults of susceptible and resistant strains of *Anopheles gambiae* (KIS, KdrKIS, Tiass) and *Aedes aegypti* (SBE, KdrSBE and Abdj). The essential oils were obtained by distillation of the dry leaves using an alembic and a clevenger. Data on the chemical composition of the oils were obtained by GC-MS and analysed using MzMine 2.53 software. The insecticidal effect of the oils was assessed at 0.1 and 1% using the method of WHO test tubes. The main compounds in LA-BF were methyl geranate (16.9%), hepten-2-one (6-methyl-5-) (11.4%), geranial (11.3%) and lavandulyl acetate (9.1%). Those of LA-CI were methyl geranate (14.8%), geranial (9.9%), hepten-2-one (6-methyl-5-) (8.4%) and ocimene (e-beta-) (7.8%). The mortality rates produced by both 0.1% oils on all strains were lower than the 14% achieved by LA-BF on KIS. At 1%, LA-BF and LA-CI induced 100% and 84.9% mortality for KIS, 89.8% and 98.1% for KdrKIS, and 85% and 29% for Tiass, respectively. 100% mortality of SBE strain also observed with LA-CI at 1%. Our study showed that LA-BF and LA-CI were toxic against adults of *An. gambiae* and *Ae. aegypti*. Plants have been used for protection against insects since ancient times and remain potential alternatives for the development of natural tools to control mosquitoes.



S5-P08

Impacts of prolonged use of unilateral mosquito control methods in Hungary: Evaluating pyrethroid resistance in disease vectors *Culex pipiens* and *Aedes albopictus*

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Introduction

Mosquito-borne diseases are a global concern, often managed by pyrethroid-based adulticidal interventions. Despite the known ecological harm and the risk of insecticide resistance, it is widely used in Hungary but its long-term effects are less-researched. For this reason, in this study, we aim to highlight the development of resistance in local mosquito populations by detecting genetic knockdown mutations in *Culex pipiens* and *Aedes albopictus*, primary vectors of the West Nile and Dengue viruses.

Methods

The investigated *Ae.albopictus* (n=100) and *Cx.pipiens* (n=150) mosquitoes were collected with CO₂ baiting traps in 2023, from 3 different regions of Hungary. An allele-specific PCR followed by gelelectrophoresis was used to detect the resistance and/or susceptible allele.

Results

Our analysis revealed the presence of the allele responsible for the pyrethroid resistance in 61% of the investigated *Cx.pipiens* individuals and 93% of these specimens showed heterozygous genotype. In the case of *Ae. albopictus*, all the investigated specimens showed the susceptible genotype, the resistant allele was not present.

Discussion

Our results confirmed the expectations that *Cx.pipiens* has been able to develop resistance against pyrethroids since it is one of the most common endemic mosquito species in Hungary and it is constantly targeted by chemical control. Although the investigated specimens of *Ae.albopictus* showed the susceptible genotype, the presence of the resistance allele in this species in the neighbouring countries emphasizes the importance of preventative measures. In Hungary, *Ae.albopictus* was first detected in 2014. It suggests, that it may not have had sufficient time to develop resistance.

Conclusion

In light of our findings, persisting with pyrethroid-based methods could not only worsen ecological damage but also fail to control mosquito populations effectively. Therefore, transitioning to alternative control strategies and integrated management is essential for sustainable vector management in Hungary.



S5-P09

Study of the chemotype of Caribbean plants in the vector control of the *Aedes Aegypti* mosquito

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According to the World Health Organization, mosquitoes are the most deadly animal in the world in terms of disease transmission. Mosquitoes are responsible for more than 725,000 deaths every year. For years, scientists have been looking for healthy ways to reduce the impact of the *Aedes aegypti* mosquito without harming the skin or the environment.

Given Guadeloupe's rich flora, this study is investigating the potential of Caribbean plants to modify mosquito behaviour and even kill them. Based on traditional Guadeloupean knowledge, an ethnobotanical survey was carried out among the Guadeloupean population in order to select the most appropriate plants and identify those known to have 'anti-mosquito' uses.

The results of the ethnobotanical survey provided valuable insights into the plants traditionally used against mosquitoes in Guadeloupe. The study succeeded in identifying and collecting the most six relevant plants and processing them to obtain various extracts by hydrodistillation (i.e. lyophilized powder, essential oil and hydrolates).

Through mosquito tests (behavioural mosquito tests and insecticide tests) this study was conducted to determine the toxic effects of the extracts on female mosquitoes and understand their behavioural responses.

Thanks to the statistical analysis of these tests, we have already identified three insecticidal extracts and five capable of modifying the behaviour of the *Ae. aegypti* mosquito.

These interesting results justify the development of future green and local cosmetic formulations.

This research project aims to contribute to the development of new integrated vector control strategies that are more effective, specific and environmentally friendly. By exploring the anti-vector potential of Caribbean plants, the study will provide a basis for the discovery of new active ingredients that can be used in pest control. The knowledge gained from this research can help in the fight against mosquito-borne diseases in the Caribbean and may have wider implications for global vector control efforts.

Mosquifen: are mosquito traps an effective tool in the battle against mosquitoes?

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Mosquito traps have been used for longtime as mosquito monitoring tools in research or mosquito control program. If the first light-traps targeted nocturnal mosquitoes and others insects, then, more sophisticated and mosquito focused devices were developed using carbon-dioxide and animal specific odours(kaironomes). With the increasing concern of environmental impact and mosquito resistance to insecticide-based methods to control larval and imaginal mosquito population, traps have been tested and introduced as complementary tools in mosquito control strategies. In this study, we tested the effectiveness of mass trapping in barrier design with carbon dioxide baited traps against salt-marsh mosquitoes, as a complementary tool in larviciding controlled area. This experiment was conducted in three campsites along the french Mediterranean wet-coast between summer and autumn 2022. The targeted species were floodwater mosquito species with anthropophilic preferences like *Aedes caspius* and *Aedes detritus* who represents a year-long nuisance. Mosquito Magnet traps (MM) were set in a barrier design around campsites to avoid mosquitoes from entering and were active 24h/24h, whereas BG-Protector traps (BGP) traps were set within the campsites, close to human, with the aim of diverting mosquitoes away from humans at peak activity during 3 hours at sunrise and 3 hours at sunset. During the 12 weeks of monitoring, over 210,000 mosquitoes of 11 species from 4 genera were collected by traps across treatment campsites, with no significant differences in mosquito community samplings between BGP and MM traps. Traps were effective at trapping *Ae. caspius*, reducing total mosquito abundance in two of the three study sites by 34% and 55%. This study provides valuable insights into the efficacy and feasibility of using mass trapping barriers as a complementary control strategy for mosquito species in floodwater it also highlighted the costs and operational limits of this kind of devices.

S5-P11

Application of sterile male of *Aedes albopictus* as an integrated control measure in Switzerland: Results of two consecutive release seasons

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The exotic invasive tiger mosquito, *Aedes albopictus*, appeared in Canton Ticino (southern Switzerland) in 2003. The spread of the mosquito has been surveyed constantly since then, and an integrated vector management, focused on the juvenile stages of the insect, has been implemented to control its numbers. Despite the system has proven its ability to contain tiger mosquitoes the densities in the territory of *Ae. albopictus* still present a risk factor for disease transmission. In addition, there is a growing practice by citizens to utilize automatic dissemination systems of products against mosquito adults thus running the risk of facilitating resistance processes. The aim of this project is to test whether the application of the Sterile Insect Technique (SIT) can bring these densities below the risk threshold, reduce the nuisance at the climatic and urban conditions present in the region. The aim of the project is to test whether the sterile male technique is an effective and sustainable tool to be ported into the integrated control measures for the tiger mosquito already in place. Both the results of the release seasons that took place between May and September in the years 2023 and 2024 and the future developments this technique might have at the level in Switzerland will be presented here. The project carried out by SUPSI is linked to the TDR Sterile Insect Technology Project in partnership with CDC, IAEA and NTD/WHO global project that is precisely about testing whether the application of SIT can effectively and sustainably reduce the incidence of diseases related to *Ae. albopictus* and *Ae. aegypti*, such as dengue, chikungunya and Zika. The field outcomes collected in the Canton Ticino setting will therefore made available to the wide vector control community to contribute to the overall evaluation of this technique when applied to invasive *Aedes*.

S5-P12

Assessing the impact of a low-cost door-to-door strategy on *Aedes albopictus* populations

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Aedes albopictus, one of the world's most invasive species, is now widely established in France, where it's a potential arbovirus vector and a major nuisance. The door-to-door strategy is one of the tools that local authorities can implement to reduce the number of larval breeding sites in private properties, with agents who can also be involved in communication and social mobilisation. The lack of evidence on the cost/benefit of such interventions still limits its implementation by municipalities. The objectives of the study were to realise a door-to-door campaign with limited resources and to evaluate its impact on mosquito populations.

The intervention was carried out in Saussan, a small municipality near Montpellier, covering an area of 12 hectares (150 properties). Four door-to-door sessions of 6 days each were carried out by 2 agents between June and September 2022, with the support of 5 neighbourhoods' ambassadors. Mosquito abundance was monitored weekly using BG-Sentinel traps, which ran continuously for 5 months in the intervention site and a nearby control site (10 traps/site).

Agents were able to visit 79% of gardens: 44% were positive at least once and 137 breeding sites were inactivated, including 22% of water storage containers that accounted for 33% of the total estimated larval production. The average number of adult females caught weekly per trap before the intervention was 31.3 ± 3.9 in the intervention site and 29.2 ± 5.4 in the control site. During the intervention period the seasonal increase was significantly lower in the intervention site (51.9 ± 2.7) than in the control one (78.3 ± 3.7), resulting in an overall corrected population reduction of 38%.

The evaluated strategy achieved encouraging results, showing a promising cost-benefit ratio if this action is done at the municipality scale as part of a preventive control programme.



S5-P13

Defining repellent properties and electrophysiological responses of essential oils against the Lone Star tick, *Amblyomma americanum*

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Introduction and Objectives

The Lone Star tick, *Amblyomma americanum* is relevant to human health due the diseases resulting from pathogens transmitted while feeding. Personal protectants are advised to prevent tick bites, despite the limited chemical diversity of available repellents. Further, development of novel repellents is challenged by poor translation of repellency data from the lab to field. The goal of this study was to test natural and synthetic molecules for repellent activity against ticks in spatial, contact, and fingertip bioassays.

Materials and Methods

We compared 16 natural oils and 1R-trans-chrysanthemic acid (TCA, from natural pyrethrins I) for repellency activity to standard tick repellents nootkatone and DEET, by using 3 different repellent assays.

Results

The contact assay repellency rank order for top 5 oils was clove, geranium, oregano, cedarwood, and thyme. TCA showed a concentration to repel 50% of ticks (RC50) of 5.9µg/cm², which was 5- and 7-fold more active than DEET and nootkatone, respectively. In the spatial bioassay, the most active oils were thyme and cassia where 10µg/cm² repelled nearly 100% of ticks within 15min exposure time, and with TCA were approximately 2-fold more active than DEET and nootkatone. Finally, TCA was highly repellent in the fingertip bioassay alongside DEET and nootkatone, whereas patchouli was the only oil that significantly repelled ticks.

Discussion and Conclusion

The repellency correlation between the assays described a negative correlation between the fingertip and contact assay and between the spatial assay and the contact assay. This suggests that repellents active in one trial may not work in another and emphasize that the development of new tick repellents must consider the method of use in the field to improve translation between laboratory and field studies. Active compounds in oils and electrophysiological responses to chemosensory sensilla have been identified. This data will inform downstream tick repellent development.

Using the ectocide Fluralaner administered *per os* to *Rattus rattus* to control the flea *Xenopsylla cheopis*, vector of *Yersinia pestis* in Madagascar: the Ectopeste project

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In Madagascar, plague remains a major public health concern. The causative agent (bacterium *Yersinia pestis*) is mainly vectored by *Xenopsylla cheopis*, a flea commonly associated to black rats, *Rattus rattus*, that play a central role in plague eco-epidemiology. Fleas are becoming resistant to the molecules routinely used for their control, enhancing the need of alternative strategy for disease control.

The EctoPeste project aims at exploring an innovative approach to control *X. cheopis* using fluralaner administered *per os* for a systemic toxic effect on blood-feeding fleas. Fluralaner is effective for up to three months against pet fleas. Its mode of action being different from other insecticides, this would help managing the flea resistance issue. Fluralaner in fleas and rodent droppings would amplify deleterious effect at population level. *Per os* administration would enable focused distribution to control for environmental toxicity.

We will characterize insecticidal efficacy and duration of fluralaner treatments administered *per os* to wild *R. rattus* on laboratory-reared *X. cheopis*. Colonies of fleas susceptible and resistant to insecticides will be fed on control and fluralaner-treated rodents (6 per arm) during direct skin-feeding experiments at different times post-treatment (2 days to 4 months). Blood fed fleas' mortalities will be followed-up. Rodent feces will be analyzed for fluralaner concentrations for proxy-characterization of lethal concentrations provoking 50 and 90% flea mortalities (LC50 and LC90).

Ectopeste experiments will start in May 2024, therefore, we expect to present the following results: (i) the induced mortalities of blood-fed fleas on treated rodents for each time point post-treatment (ii) the treatment efficacy duration (iii) the fluralaner concentration vs. induced mortalities relationship to determine LC50 and LC90 values.

Our results will be discussed on the light of the approach efficiency in different contexts of Madagascar, especially urban ones, and potential community-based interventions.

S5-P15

Citizen action for sustainable control of dengue vectors in Abidjan, Côte d'Ivoire: a cluster-randomised control trial

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Transmitted by day biting, highly anthropophilic invasive *Aedes* mosquitoes, dengue is a life-threatening disease that has dramatically increased in incidence globally in recent years. While vector control interventions are the mainstay in dengue control programmes, in sub-Saharan Africa such programmes are largely inexistent or mainly follow a top down approach with sporadic insecticide spray campaigns targeting adult mosquitoes. However, these interventions have only a very limited, short-term impact and are environmentally not sustainable, since *Aedes* mosquitoes breed in small water containers that are ubiquitous in urban areas. Together with local residents, we designed a community-based intervention to reduce *Aedes* breeding sites in Abidjan, the largest city in Côte d'Ivoire. To measure the intervention's efficacy, we implemented a cluster-randomised control trial. In the trial, we are also testing whether mass trapping with simple traps targeting egg-laying females reduces mosquito densities, either alone or in combination with the community-based larval source management. The trial consists of four study arms, including larval source management (LSM), mass trapping using Biogents *Aedes* gravid traps (BG-GAT), the combination of LSM and BG-GAT, and a control arm without any of the two interventions. In each study arm, we included 10 clusters adding to 40 clusters in total. At the meeting, we will present the trial design and preliminary results from the trial.



S5-P16

Ecological alternatives to insecticides for controlling vector-borne diseases: effects of caffeine on mosquito ageing

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Background

The search for new ways to control mosquito-borne diseases constitutes a major public health challenge due to the emergence and spread of resistance to insecticides.

Natural products, usually secondary metabolites produced by bacteria, plants, or fungi, offer promising alternatives to synthetic insecticidal compounds. In this study, we explored the potential of caffeine, an alkaloid of the methylxanthine family, commonly present in many plants and known for its psychotropic stimulant and toxic effects, as an eco-friendly alternative to conventional insecticides.

Methods

We exposed adult mosquitoes to a diet of caffeine dissolved in a 10% sucrose solution at concentrations varying from 128 to 1024 ppm (in the case of *Aedes aegypti*), or from 50 to 400 ppm (in the case of *Anopheles arabiensis* and *An. gambiae*) and followed their survival for up to a month.

Results

A significant negative correlation between caffeine dose and mosquito longevity was found in both *Anopheles* and *Aedes* (log-rank test $P < 0.0001$ in all cases). The addition of caffeine did not impact feeding rates on the treated sucrose solution compared to the control.

Significance

These results reveal that caffeine is a potential target for mosquito control research aiming at sustainable intervention tools. Its sub-lethal, dose-dependent, cumulative effects on longevity during the course of the mosquito lifespan could help prevent the transmission of pathogens and mitigate the appearance of genetic resistance. Consequently, caffeine represents a promising natural alternative to synthetic insecticides, aiming to reduce the vectorial capacity of mosquito populations without their elimination.

Keywords: *Aedes aegypti* - *Anopheles arabiensis* - *Anopheles gambiae* - Survival - Caffeine

Ecological, insecticide free, simple, efficient and sustainable mosquito trapping-bednet against malaria: a new approach eliciting users' acceptability

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Failure to mitigate selection and spread of Anopheles insecticide resistance is the cause, at least in part, of increasing malaria incidence currently witnessed in Sub-Saharan Africa. The repeated acknowledgment of rapidly evolving mosquitoes' populations to adapt to insecticides calls for breaking this mainstream trend. While an erosion of bednet (LLIN) adherence and use is also occurring, it is therefore urgent to propose new complementary approaches.

Taking advantage of the natural interactions of Anopheles with the bednets which are occurring for 80% on the roof, a mosquito trapping tool (T-Net) was designed to be retro-fitted on the roof of any bednet, hence adding mechanical trapping-killing function to insecticide property. Human odor plume represents the lure. Trapped mosquitoes die from desiccation in the device. From experimental hut proof-of-concept studies in areas where Anopheles are highly resistant to insecticides in Côte d'Ivoire, we demonstrated (i) that insecticide-free LLIN mounted with T-Net killed 4 times more wild pyrethroid resistant mosquitoes than pyrethroid-only LLIN, and (ii) that killing performance of Permanet 2.0-T-Net was increased by 31-58.5% when compared to PBO- and IG2-LLINs, the new generation of bi-treated nets.

Hence, mass-trapping of malarial mosquitoes using T-Net, elicits bednets' efficiency even in contexts of high insecticide resistance. Given the fact that users would actually witness trapped mosquitoes, we believe our device should win greater usage. Our innovative approach will be evaluated, in communities, in the framework of a 4-arms cluster randomized controlled trial to be run in 40 villages of Korhogo district, Côte d'Ivoire (TrapNet project). Interventions are mass distributions of trapping or classical bednets. Primary endpoints are *Plasmodium falciparum* prevalence and entomological inoculation rate. Users' acceptability will be scrutinized. We believe that our insecticide free trapping bednet concept should represent a game-changer in malaria vector control, and could constitute a future sustainable WHO recommendation.



S5-P18

Exploring Wolbachia and Usutu virus interactions- a potential control strategy?

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Introduction and Objectives

Wolbachia pipientis is an intracellular bacterium which has been used as a novel vector control strategy with great success in field trials, mainly targeting *Aedes aegypti* and dengue transmission. Usutu virus is an emerging enzootic virus maintained primarily in a bird-mosquito cycle, which can spill over and cause human infections. Beyond causing malaise and neurological symptoms in humans, Usutu virus causes mortality in European bird species, notably common blackbirds (*Turdus merula*).

The primary vector of Usutu virus in Europe is *Culex pipiens s.l.*, all of which are naturally infected with *Wolbachia*. Despite this, no study to date has looked at how Usutu and *Wolbachia* interact. The objective of this study is to determine the effect *Wolbachia* has on Usutu virus replication. Additionally, we examine whether the *Wolbachia*-Usutu interaction is influenced by viral concentration or viral strain.

Materials and Methods

Here we examine how three strains of Usutu virus (ENT MP 1626, SAAR 1776, and London 2020) interact with *Wolbachia* strain *wAlbB* in-vitro within C6/36 cells. Cells were independently infected with virus at two concentrations (MOI 0.1 & 1). Samples were taken every 24 hours up to 96 hours post-infection and viral load was quantified via plaque assay.

Results, Discussion and Conclusion

The *Wolbachia* strain *wAlbB* showed complete blocking of Usutu virus strain ENT MP 1626 within C6/36 cells 72-96 hours post infection. However, when *wAlbB* C6/36 cells were infected with Usutu strain SAAR 1626, it did not block viral replication completely. This differential blocking of Usutu virus is the first-time virus strain-dependent differences in *Wolbachia* blocking has been observed. Critically, we discuss further analysis of this finding and the potential contribution to understanding *Wolbachia*-viral interactions.

S5-P19

Development of a Broad-Spectrum Antiviral System Using Flavivirus-Inducible Gene Expression to Control Mosquito-Borne Diseases

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Incidence of dengue virus (DENV) and Zika virus (ZIKV), two mosquito-borne flaviviruses, is increasing in large parts of the world. Vaccination and medication for these diseases are unsatisfactory. Here, we developed a novel antiviral approach, using a virus-inducible gene expression system, to block virus replication and transmission. Constructs containing the smallest replication units of dengue virus serotype 2 (DENV2) with negative-stranded DENV2 artificial genomes and genes of interest were established in an *Aedes aegypti* cell line, resulting in expression of target genes after DENV2 infection. Green fluorescent protein (GFP) assays confirmed the system was virus-inducible. When we used one of two apoptosis-related genes, *A. aegypti* michelob_x (*AaMx*) and inhibitor of apoptosis (IAP)-antagonist michelob_x-like protein (*AaIMP*) instead of GFP, the production of viral RNA and proteins were inhibited for all five viruses tested (DENV1–4 and ZIKV), and effector caspase activity was induced. The system thus inhibited the production of infectious virus particles *in vitro*, and in mosquitoes it did so after DENV2 infection. This is a novel broad-spectrum antiviral approach using a flavivirus-inducible gene-expression system, which could lead to new avenues for mosquito-borne disease control.

S5-P20

An innovative method of using *Agrobacterium tumefaciens* to screen for AMPs against insect-transmitted plant vascular diseases: demonstrated with *Xylella fastidiosa*

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Xylella fastidiosa (*Xf*) is a fastidious, gram-negative bacterium that can infect the xylem of a wide range of cultivated, ornamental, and wild host plants and is transmitted by plant vascular feeding Hemipteran insects. *Xf* is the causal agent of Pierce's disease in grapevine in North America, citrus variegated chlorosis in South America, coffee and almond leaf scorch diseases, and olive quick decline syndrome, notably in Southern Italy. *Xf* is responsible for significant economic losses in regions like the United States, Italy, and Brazil. The current approaches to control *Xf* epidemic mainly rely on eradication of infected plant to reduce inoculum source, the use of insecticides to control the vector populations, and the use of healthy plant propagation material. Therefore, there is a need for new and safe biological solutions for *Xf* disease management. The use of antimicrobial peptides (AMPs) represents an interesting approach due to their great diversity and broad spectrum of activity against microorganisms, low environmental impact, and limited evolution of resistance. This project aims at adapting an innovative approach combining the use of AMPs and a delivery system based on *Agrobacterium tumefaciens* developed at the U.S. Horticultural Research Laboratory in Fort Pierce, Florida, to combat citrus greening disease. To this end, an *in vitro* method with plant extracts from *Agrobacterium*-infiltrated leaves is being developed at EBCL for the screening of AMPs against *Xf*. The reduction of *Xylella* titer in host plants would mitigate symptoms but also decrease the spread of the disease via the insect vector. In addition, this approach could be used to target the bacterial symbionts of the insect vectors as an insect control method to reduce their vectorial capacity and transmission of *Xylella*. This screening method allows rapid screen AMPs in an easily performed plant-based bioassay adaptable for either antimicrobial or insecticidal activities.



S5-P21

Repellent activity evaluation of Cumin Essential Oil and its components against *Phlebotomus papatasi* (Diptera: *Psychodidae*) via a high-throughput screening bioassay

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Introduction and Objectives

Leishmaniasis, a vector-borne disease prevalent among both humans and animals, is transmitted through the bite of infected female phlebotomine sand flies. Currently, among the primary personal protection measures against sand fly bites is the use of topical repellent compounds. Despite the development of numerous synthetic insecticides and repellents, their widespread application has led to reduced efficacy and the emergence of resistance. Moreover, chemical control methods have adverse effects on both human health and the environment. Consequently, there is a pressing need for novel ecologically sound alternative compounds with repellent activity, to improve the efficacy of sand fly control and to overcome potential resistance phenomena resulting from prolonged reliance on synthetic repellents. In this context, the present study aimed to evaluate the repellent efficacy of cumin essential oil and its major constituents against adult sand flies in comparison with commonly used synthetic repellents. A cost-effective, and easily decontaminated apparatus facilitated rapid high-throughput screening of the compounds.

Materials and Methods

The repellency potential of Cumin Essential oil and its main components was evaluated against adult female *P. papatasi* sand flies. DEET and transfluthrin, two commonly used, synthetic insect repellents, were also tested. EC₅₀ estimation, knockdown activity and assessment of sand flies' avoidance behavior in response to the tested repellents were performed.

Results

Cumin essential oil and cumin aldehyde were effective repellents against *P. papatasi*, with results comparable to those of DEET and transfluthrin.

Discussion and Conclusion

The results indicate a promising new group of natural compounds for sand fly control. Within an integrated pest management framework, environmentally friendly sand fly control programs could be developed. Further research is needed to elucidate the mode of action of the specific chemical components, to assess their environmental impact on non-target organisms, and to improve their efficacy and stability through formulation improvements.

THE CLIMOS PROJECT: Towards better understanding the risk of sand fly-borne diseases in the changing climate and environment

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CLIMOS - Climate Monitoring and Decision Support Framework for Sand Fly-borne Diseases Detection and Mitigation with Cost-benefit and Climate-policy Measures – is a research projects that includes 30 partners across 16 countries, forming a unique consortium of researchers, technology platform designers, public health representatives, veterinarians and at-risk communities. It aims to address an important public health and veterinary concern of sand fly-borne diseases, caused mainly by parasitic protozoans of the genus *Leishmania* and various sand fly-borne viruses, conducting basic and applied research to better prepare for current and future impacts of climate and environmental changes on sand fly-borne pathogens that affect human and animal health.

CLIMOS studies climatic, environmental, demographic, and epidemiologic characteristics associated with longitudinal datasets on sand fly presence and abundance and animal infection rates at different geographical scales across Europe and neighbouring countries, using historical datasets of previous research projects (EDEN, EDENext, VBORNET, and VectorNet), as well as active sand fly field surveillance in 14 countries of Europe and adjacent regions in a coordinated and standardized effort, followed by meticulous screening of sand fly-borne pathogens (*Leishmania* sp. and phleboviruses). This data feeds into epidemiological-climatic predictive mathematical models of realistic human-induced climatic changes scenarios to help develop an early warning system of infection and disease designed with the input of partner public health ministries for public use. The project develops novel technologies to monitor and mitigate human-sand fly contact and implements a comprehensive scientific and public dissemination policy and outreach programme to communicate project results to local communities at risk of sand fly-borne diseases.

The CLIMOS consortium is co-funded by the European Commission grant 101057690 and UKRI grants 10038150 and 10039289. CLIMOS is one of the six Horizon Europe projects, BlueAdapt, CATALYSE, CLIMOS, HIGH Horizons, IDAlert, and TRIGGER, which form the Climate Change and Health Cluster.

Developing systematic guidelines for Mosquito Control in Built-Up Areas in Europe

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There is a growing risk of mosquito-borne diseases (MBDs) worldwide, including in the European region. To address this problem, a dedicated group of experts from the European Mosquito Control Association (EMCA), with support from WHO/TDR, set out to establish clear guidelines and recommendations for effective surveillance and control measures against mosquitoes of public health concern. In the absence of systematic clinical studies assessing the impact of vector control interventions on the epidemiology of human MBDs, it was agreed to use entomological endpoints as proxy for the biting activity of mosquitoes (and thus the risk of disease transmission) instead.

A scientific review revealed a very heterogeneous landscape of various entomological endpoints supported by generally poor evidence as suggested by the GRADE system, which is meant to evaluate the quality of evidence and strength of recommendations. This reduces the strength of our recommendations for or against certain mosquito control measures. However, some interventions might be recommended in specific settings, such as larviciding, or chemical control of adult mosquitoes under specific conditions (outbreak of disease, high infestation with adult mosquitoes).

There is an urgent need to evaluate the efficacy of mosquito control interventions in a systematic fashion that would also include epidemiological endpoints. However, given the relatively low current numbers of MBD cases in Europe, this would be unrealistic. Therefore, future studies should encompass data from various regions worldwide. We further recommend standardizing the data collection process for future field studies by providing technical guidance to build-up a solid and exploitable database related to vector control.

These guidelines represent the first attempt to implement the GRADE system, which had originally been developed for clinical trials, to evaluate the efficacy of mosquito control method in urban settings in Europe in a systematic and rigorous manner.

S6-P03

VECTOBOL: An Initiative Investigating Vector Biodiversity in Bolivia that could inspire European laboratories

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Bolivia, a megadiverse country, is home to over 410 species of pathogen-vector insects, including Culicidae (malaria and arbovirus vectors), Triatominae (Chagas disease vectors), and Phlebotominae (Leishmaniasis vectors). Despite extensive field data collection by government health agencies, much of this valuable data hasn't been preserved, either in data files with field annotations or as preserved specimens. Consequently, Bolivia's entomological heritage is at risk.

VECTOBOL, a multilab initiative, aims to address this issue by preserving essential data on vector insect samples and encouraging the preservation of collected specimens in laboratory collections. It relies on a shared database established using REDCap (Research Electronic Data Capture), a web-based application developed by Vanderbilt University for database creation. REDCap is versatile and can collect various types of data in any setting, with a primary design to facilitate online and offline data capture for research studies using computers or cell phones. It boasts high-security measures and a user-friendly interface, enabling groups of researchers to work on their own independent datasets in their laboratories as well as using the shared database.

The REDCap server for VECTOBOL, initially a grassroots initiative located in Cochabamba, Bolivia, is now hosted at IRD (Institut de Recherche pour le Développement) in France and is compliant with European PHI (Protected Health Information) regulations.

The VECTOBOL database contains basic field survey information, such as unique sample codes, GPS coordinates, and ecological conditions, captured offline in the field. Species identifications are added later in the laboratory, along with field trip data. Data can be filtered and downloaded to generate standard files for statistical analysis or GIS manipulation, enabling the creation of distribution maps among other applications. Currently, the VECTOBOL database contains more than 1200 records and continues to grow. For more details, visit the dedicated website: <https://vectobol.ird.fr>.

S6-P04

The INOVEC Project: a pan-European partnership for enhancing the surveillance and control of mosquito vectors of emerging arboviruses

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Over the past 10 years, arboviral diseases like dengue, Zika, chikungunya and yellow fever, have (re) emerged presenting heightened prevalence and severity. In the absence of effective vaccines and treatments, preventing these diseases at the global scale continues to depend largely on controlling mosquito vector populations, and interrupting human–vector contact or both. However, recent outbreaks worldwide underscore the inadequacies of current vector control measures in preventing epidemics and reducing disease incidence. Novel, cost-effective, scalable vector control measures and community-based interventions are urgently needed to prevent the introduction, spread and establishment of *Aedes*-borne diseases.

Addressing this pressing need, the INOVEC project, funded by the European Commission (MSCA Staff Exchanges 2023-2026), aims to develop, optimize and promote integrated approaches and innovative tools and technologies for the surveillance and control of mosquito vectors of emerging arbovirus diseases. Structured around five interconnected work packages and 18 tasks, the INOVEC project is led by 26 partners from academia and the private sector. This collaborative framework promotes cross-sectoral and international synergy in vector-oriented research, social sciences, and product development, thereby maximizing its impact on both scientific advancement and societal well-being. Moreover, INOVEC endeavors to facilitate the transfer of knowledge and technologies to regions facing escalating risks of arboviral diseases.

In its inaugural year, through implementation of secondments, the INOVEC project made a significant contribution to research and innovation in the field of vector control through the organization of workshops and conferences, the dissemination of scientific findings, and the facilitation of knowledge transfer and career development opportunities.

Looking ahead, INOVEC will contribute to international efforts to improve global health and human welfare by reducing the burden of vector borne diseases.

S6-P05

VECTOCLIM PROJECT. Assessing the impact of climate change on two vector-pathogen pairs: *Aedes albopictus* & *Culicoides* midges in Occitania, France and Europe

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The VECTOCLIM project aims at assessing the impact of recent and future climate change on the risk related to major vector-borne diseases for the European temperate ecosystem. The project will focus on two vectors of interest for public and animal health: the invasive tiger mosquito, *Aedes albopictus*, vector of viruses to humans, and *Culicoides* biting midges, vectors of viruses to livestock. Through the use of mathematical modelling and large scale and longitudinal entomological data, we will evaluate the influence of global warming on the geographic and seasonal distribution of these vectors according to different IPCC scenarios. Medium and long-term projections of vector distribution will be produced and compared. Epidemiological modelling will be used to assess risk factors, identify levers of control, and produce dynamic risk maps of the studied areas for three arboviruses (dengue, chikungunya and Zika viruses) and two orbiviruses (bluetongue and epizootic haemorrhagic disease viruses).

Four reference research institutes in the fields of infectious diseases and climate sciences collaborate in the implementation of this study: the University of Montpellier, CIRAD, IRD and ICTP. Regular consultations will be organised with public and veterinary health professionals in order to guide research objectives and provide the best possible support to the current and future challenges facing stakeholders. The results will be analysed and discussed with experts from different disciplines such as entomologists, economists and sociologists, and disseminated to policy-makers and stakeholders.

VECTOCLIM is a 2-years research project (2024-2025) funded by the French Occitania region through the grand challenge RIVOC (*Infectious Risks and Vectors in Occitania*). It is an operational research project which aims to support health stakeholders at different scales: regional (Occitania), France, Europe, in a One Health context. Supported by various international reference institutes and networks, the project aims to be interdisciplinary and intersectoral.

S6-P06

VectorByte - a hub for biological trait (VecTraits) and abundance data (VecDyn)Samuel RUND (srund@nd.edu)*University of Notre Dame, Notre Dame, IN, USA*

The VectorByte project aims to address the challenges associated with vector-borne diseases (VBDs) by establishing a centralized open-access data platform. VBDs, transmitted by vectors such as arthropods, pose significant threats to human, animal, and plant health, as well as global economic welfare. Despite their impact, efforts to prevent and predict VBD outbreaks have been hindered by isolated and disparate vector data, making integration and analysis difficult. VectorByte seeks to overcome these barriers by creating a standardized repository of vector trait and population abundance data, facilitating easier sharing and utilization within the disease ecology community and beyond. Additionally, the project will develop tools for data analysis and modeling, along with educational materials and workshops to train early career scientists. By providing accessible and standardized data, VectorByte aims to enhance research, mathematical modeling, and understanding of VBD dynamics, ultimately enabling the development and assessment of effective control strategies. The platform's user-friendly interface and global scope have the potential to transform VBD research by promoting FAIR (Findable, Accessible, Interoperable, and Reusable) data standards, facilitating comparison across VBD systems, and advancing open and testable models of VBD dynamics.

S6-P07

The Global Vector Hub - building entomological capacity worldwide and improving epidemic preparedness

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The Global Vector Hub (GVH) is an exciting new online platform currently under development at the London School of Hygiene & Tropical Medicine (LSHTM), focusing on control of arthropod disease vectors globally. An early version was launched in summer 2020 in the context of the COVID-19 pandemic to address the urgent need for accurate and up-to-date information and resources on vector-borne diseases and vector control interventions. Following on from this success, a revised version was released in 2023.

The aims of the GVH are to assist in **capacity building** for vector control globally, establish a **community of practice** for vector control interventions, and enable stakeholders to make **evidence-based decisions**. The main audiences of the GVH are **public health officials, vector control agents** and **vector researchers**. The GVH consists of a community-led, online, **open-access** resource to provide comprehensive information on vector control and vector biology. This includes geo-tagged entomological **data** and epidemiological data, a searchable registry and worldwide **network** of vector researchers and vector controllers, and a comprehensive **resource** database of training and educational materials, vector control guidelines and research tools.

In addition, the Special Programme for Research and Training in Tropical Diseases (TDR) and the GVH have developed a web-based **global directory of medical entomology courses** as a new resource for strengthening the capacity of scientists combating neglected tropical diseases and other vector-borne diseases. The directory currently lists a total of 169 medical entomology courses offered both on-campus and through distance learning in 38 countries across all WHO regions, covering seven languages. The freely available directory was developed in collaboration with the GVH and Arctech Innovations, following the mapping of courses available globally. For each course, session dates, course outline, fees, language of instruction and responsible managers are listed.



S7-P01

Comprehensive Surveillance of Mosquitoes and Sandflies in Slovenia: Implications for Public Health and Emerging Pathogens

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The global dissemination of infectious diseases constitutes a significant risk to public health, as novel pathogens emerge in previously unaffected areas. These threats primarily include arboviruses and parasites like *Leishmania* and *Plasmodium*, which have a significant potential to spread due to the increasing prevalence of their vectors. It's therefore critical to research new pathogens with a harmonized approach and to establish sensitive surveillance systems.

Our targeted research, conducted between 2020 and 2022, focused on mosquitoes (Diptera: Culicidae) and sandflies (Diptera: Psychodidae), the key vectors of human diseases. Our aim was to catalogue the mosquito and sand fly species in Slovenia, describe the distribution and abundance of both autochthonous and invasive species, and compile data on the presence or absence of viral and parasitic pathogens transmitted by these vectors. We implemented standardized monitoring protocols for mosquitoes and sand flies in accordance with European guidelines. Over a three-year period, we surveyed vectors at 226 sites across Slovenia. We captured and identified over 85,000 adult mosquitoes belonging to 41 species, including seven species newly documented in the Slovenian mosquito fauna. During the sampling, we detected various microorganisms, including Usutu and Sindbis viruses and the parasite *Dirofilaria repens*. By developing predictive maps, we have delineated regions with an increased probability of harboring medically significant mosquitoes.

During sand fly surveillance, we caught over 1,200 individuals and identified four species, with the highest population density observed in the Primorsko-Notranjska and Goriška regions. Notably, no *Leishmania* species were detected. The collected data on the distribution of vectors and the emergence of novel microorganisms have proven to be crucial for assessing the risk associated with emerging pathogens and identifying locations with increased likelihood of mosquito and sand fly presence. These results contribute significantly to improving our comprehension of epidemiological drivers and the establishment of effective surveillance strategies.

S7-P02

A predictive model of *Aedes albopictus* density in Emilia-Romagna region (Italy) based on ovitrap and meteorological data

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Aedes albopictus, the Asian tiger mosquito, has become a prevalent pest in Italy, causing severe nuisance and posing a threat for transmission of arboviruses introduced by infected travelers. We propose a predictive model on the seasonal population density of *Ae. albopictus* in the Emilia-Romagna region based on ovitrap data and meteorological parameters.

A Bayesian approach was employed to identify the best meteorological predictors of species trend, using the eggs collected monthly from 2010 to 2023 by the Emilia-Romagna regional monitoring network. The findings show that the winter–spring period (January to May) plays a crucial role on the size of the first generation and the following seasonal dynamic. As an example we show the summer density population prediction for 2024.

The data collected confirmed the impact of climate parameters on the *Ae. albopictus* population's seasonal trend.



S7-P03

Historical review of insecticide resistances of *Anopheles* in Madagascar: Origin and spatio-temporal dynamics

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Synthetic insecticides have been key actors for malaria control in Madagascar since 1949, with the first use of DDT. Nonetheless insecticide resistances rapidly appeared and spread across Madagascar. We carried out an historical review of all the studies monitoring the resistance in *Anopheles* in order to understand the spatio-temporal dynamics. Between 1965 and 2023, a total of 2,285 resistance tests were performed on four different *Anopheles* species at 131 sites across Madagascar.

Our results showed that insecticide resistances are very common in *Anopheles gambiae* and *An. funestus* populations for most of the insecticides families, while *An. mascarensis* and *An. coustani* remain susceptible. Resistances are widely distributed across the island, which means that there are not geographical barriers to their spread. Interestingly, susceptible status is restored in some locations through time. Isolated from the continent, Madagascar offers a compelling opportunity to understand the origin and evolution of insecticide resistance in *Anopheles*. The reemergence of malaria in the island during the last decade, requires to better understand the underlying genetic and physiological mechanisms involved in the resistance.



S7-P04

Prevalence of *Borrelia burgdorferi* genospecies in *Ixodes ricinus* ticks: models for surveillance and prevention based on the French citizen science program CiTIQUE

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Vector-borne diseases pose a growing global health threat, exacerbated by factors such as habitat modification and climate change. Lyme borreliosis, the most common arthropod-borne disease in Europe, is primarily transmitted by the tick *Ixodes ricinus*. With rising human exposure and no available vaccine, understanding the ecology and epidemiology of the *Borrelia burgdorferi* species complex is urgently needed for effective control and prevention. However, traditional monitoring approaches face challenges in acquiring large-scale data. Here, we utilize a citizen science initiative, the French CiTIQUE program, to gather georeferenced ticks and *Borrelia* samples on a broad scale. We investigate the relationship between *Borrelia* distribution and various factors including vector suitability, climate, ecology, and human activity. Our analysis identifies vector suitability as the primary determinant of *Borrelia* prevalence. Moreover, we reveal significant spatial heterogeneity in species distribution within the *Borrelia burgdorferi* complex. We discuss these findings within the context of the current understanding of tick ecology and pathogen dynamics and highlight implications and future challenges for Lyme disease surveillance and prevention.

The Asian tiger mosquito - Current situation and surveillance programs in Austria

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The potentially invasive Asian tiger mosquito (*Aedes albopictus*) was able to establish itself rapidly in southern Europe after its arrival. Rising temperatures in Central Europe as a result of global warming and generally higher temperatures in urban areas favor its survival and allow it to spread further north. As the Asian tiger mosquito is a competent vector for many pathogens, monitoring programs have been set up in Austria to record the occurrence and spread of this species. This allows new populations to be identified at an early stage for the monitoring of the development of existing populations.

Two complementary monitoring programs are being carried out in Austria to monitor the tiger mosquito. As part of a scientific program, ovitraps are set up at around 50 locations nationwide and sampled weekly from May to October. In addition, there is also a citizen science program in which citizens can report possible tiger mosquitoes using the «Mosquito Alert» app.

While tiger mosquitoes were only found sporadically in 2020, the number of findings is steadily increasing. In 2022, tiger mosquitoes were detected in all federal states for the first time. This species is now common in parts of the urbanized areas of Vienna and Graz, so it can be assumed that stable populations have already established here. We can show that the two monitoring programs complement each other well. The scientific monitoring provides data that can be compared both spatially and temporally and includes absence data as well. Further, it is also an important method for monitoring points of entry. The citizen science program provides a much more precise distribution of tiger mosquitoes within cities. Further, several findings of tiger mosquitos in new areas have been reported through the “Mosquito-Alert” app.

Infrared spectroscopy: a stop shop technology for vector borne disease surveillance?

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Introduction and Objectives

Surveillance of vector populations is essential for controlling vector-borne disease transmission. However, measuring key demographic traits such as mosquito species composition, age structure, infection prevalence, blood meal preference and insecticide resistance status is often challenging, given the lack of accurate and scalable methods, resources and high costs. Our team aims to develop a technology to measure several vector key traits simultaneously, using a high-throughput, accurate and cost-effective technology based on infrared spectroscopy.

Materials and Methods

Using *Anopheles gambiae s.l.* mosquitoes as a study system, we have been measuring >50,000 mosquito spectra using an ATR-FTIR mid-infrared spectrometer (MIRS) from different laboratories and field sites in Africa. The biochemical information obtained with MIRS was then used to train machine learning (ML) algorithms to predict key traits in individual mosquitoes.

Results, Discussion and Conclusion

MIRS-ML was able to correctly classify *Anopheles gambiae s.l.* cryptic species, age classes, *Plasmodium* infection, insecticide resistance status and blood meal host species with high accuracy under laboratory conditions. Field validation using wild mosquito collections confirmed that the technology can be used under natural conditions, although with the need of local calibration. We are currently developing a generalisable, holistic approach to simultaneously predict all key traits from different ecological settings. Preliminary results from other taxa (Culicine mosquitoes, tsetse flies, blackflies) suggest that MIRS-ML can be applicable to different vectors and become a stop shop technology for vector-borne disease surveillance.



S7-P07

First findings from “Rapportera Fästing” – a citizen science tool for reporting ticks in Sweden

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Introduction and Objectives

Tick-borne diseases are an important public health concern, but tick surveillance remains challenging and resource-consuming. Climate change can lead to the expansion of tick habitats and the emergence of novel tick species and pathogens in previously free areas. Citizen science approaches can offer cost-effective and rapid surveillance strategies to monitor tick populations and detect possible introductions. *Ixodes* spp., a vector of the tick-borne encephalitis virus, is endemic in Sweden. However, the introduction of *Hyalomma marginatum* was first detected in July 2018 thanks to an initial citizen science program focused on sending ticks to the Swedish Veterinary Agency (SVA).

Material and Methods

In May 2023, a web-based tool named ‘Rapportera Fästing’, was launched. This mobile-friendly website enables users to report tick findings and, through the recognition of image patterns, direct them towards different genera of endemic and exotic ticks. Users are encouraged to submit photos/videos using the tool. If an exotic tick pattern is selected (i.e. *Dermacentor*, *Hyalomma*), the user is requested to send the specimen to SVA. The information available for each report includes the municipality, the selected photo/pattern, and the host species/environment of collection.

Results, Discussion and Conclusion

As of March 1, 2024, 20,404 reports were received from all Swedish municipalities. Most reports came from the counties of Västra Götaland, Stockholm, and Skåne. Nearly 96% of reports came from dogs, cats, and humans. Assessment of the received photos has identified nearly 40 tick observations not belonging to the genus *Ixodes*. Notably, we received reports of *Dermacentor* spp. (n=4) and *Hyalomma* spp. (n=5). Tick species belonging to the Swedish tick fauna but looking different from *Ixodes* spp. were also reported: *Carios vespertilionis* (n=22) and *Haemaphysalis punctata* (n=5). *Rhipicephalus sanguineus*, another exotic tick species, was also reported (n=4), despite not being available as a choice in the tool.

Modelling the distribution of tsetse flies and African animal trypanosomosis in Côte d'Ivoire to guide livestock disease control

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Introduction

Detailed information is available on sleeping sickness and tsetse in the endemic foci in Côte d'Ivoire, with elimination of transmission on the horizon. Much less is known about the distribution of tsetse at national level and of African animal trypanosomosis (AAT) despite the heavy burden for smallholder farmers. As the country aims to improve livestock production, with a focus on the northern cotton belt, information on AAT risk is badly needed. To fill this gap, we predicted AAT and tsetse distribution at the national level.

Methods

We developed species distribution models for the two main tsetse species (*Glossina palpalis gambiensis* and *G. tachinoides*), and AAT (*Trypanosoma brucei brucei*, *T. congolense* and *T. vivax*) at national level. We used presence only (Maximum Entropy; Maxent) and presence-absence models (Generalized Linear Model; GLM and Random Forest; RF) trained with data from a national atlas of tsetse and AAT currently under development (2012-22). We used readily available environmental variables on vegetation, temperature, rainfall, soil moisture, rivers and topography, human and livestock densities and distance to protected areas.

Results

Our tsetse distribution models directly aided the Ivoirian Direction of Veterinary Services in designing targeted field sampling within the cotton belt, prioritizing areas critical for tsetse elimination efforts. Population genetics studies are underway to investigate potential isolation of tsetse populations, which may be linked to key environmental factors identified by our models, i.e. the distance to rivers and classified forests. This understanding, for example, helps in explaining tsetse absence in the South-West region. These combined insights are crucial to inform future control.

Conclusion

Our tsetse and AAT distribution models inform policy decisions and help target surveys to fill data gaps as Côte d'Ivoire is in the process of developing its Progressive Control Pathway for AAT.



S7-P09

A proof-of-principle for a Cost-efficient User-friendly LAMP-enabled Xenosurveillance (CULEX) approach

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Introduction and Objectives

Xenosurveillance has proven to be a successful approach to detecting vector-borne and non-vector-borne pathogens in blood meals of biting insects. However, the most common molecular methods used in this approach are costly and require a proper infrastructure that might not be available in areas of high risk. Non-instrumental nucleic acid amplifications, such as isothermal amplifications, are a great alternative to address these challenges. The aim of the project was therefore to develop multiple LAMP assays using mosquitoes as “flying syringes” that can be deployed in the field for pathogen detection.

Materials and Methods

Aedes aegypti, *Anopheles coluzzii*, and *Culex pipiens* mosquitoes were blood-fed with human, cow, and chicken blood. A chemically treated filter paper for nucleic acid capture was used to preserve the DNA/RNA of the mosquito's blood meal. The chemically treated paper was used as the template source for nucleic acid amplification to explore simple sample preparation steps in the field. Different time points post-feeding were tested. The mosquitoes were collected, knocked down by cold, and crushed in the chemically treated filter paper. LAMP assays will be optimized for blood-fed mosquitoes as a proof-of-principle that isothermal amplification assays can be used for xenosurveillance. Limit of detection, sensitivity, and specificity will be analyzed for the LAMP assays. The reactions will be performed on a real-time PCR machine as well as using a phase-change material to mimic conditions in the field without the use of laboratory equipment to detect host blood meal and pathogens through a user-friendly detection system.

Results, Discussion and Conclusion

We expect that amplification through LAMP assays can be optimized and be detected by the naked eye, or through simple cost-efficient, and user-friendly devices.

Investigation of tularemia in Shida Kartli (Georgia) with One Health approach

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Introduction and Objectives

Tularemia is a multi-host, contagious, vector-borne zoonosis caused by *Francisella tularensis*, a bacterium with a broad range of hosts that include invertebrates, mammals, and birds. It is a zoonosis of complex epidemiology, and knowledge of reservoir hosts for this pathogen is still incomplete. Tularemia can be transmitted to humans through contact with animals or contaminated environments, and vectors.

Shida Kartli is a landlocked administrative region in the eastern part of Georgia, which is an endemic region for tularemia. The region is characterized by moderate humid climate, with moderate cold winters and warm long summers. With an objective of better understanding the disease in Shida Kartli, One Health approach was utilized during investigations in 2023.

Material and Methods

For detection of human tularemia-specific antibodies, enzyme-linked immunosorbent assay classic *F. tularensis* IgG and IgM tests were utilized, together with in-house microagglutination test. After identifying human cases, rodents, vectors, and environmental samples were collected for testing with real-time PCR using outer membrane coding sequences - fopA and tul4.

Results

A total of four human cases were confirmed. Investigations were initiated with One Health approach in all instances - 215 rodents (*Apodemus uralensis*, *Apodemus witherbyi*, *Microtus socialis*, and *Mus musculus* species), 1089 vector (*Dermacentor marginatus*, *Haemaphysalis parva*, *Haemaphysalis inermis*, *Hyalomma marginatum*, *Rhipicephalus sanguineus*, *Ixodes ricinus*, *Rhipicephalus annulatus*, *Rhipicephalus bursa*, *Rhipicephalus sanguineus*) pools, and four water samples were collected. Results showed fopA and tul4 genes in only one vector pool.

Discussions and Conclusions

Utilizing a One Health approach, investigations extended to rodents, vectors, and environmental samples, providing a comprehensive understanding of disease dynamics. Overall, this study provides valuable insights for the purpose of developing One Health strategies in tackling zoonotic diseases in Shida Kartli.

Entomological and diagnostic surveillance of West Nile and Usutu viruses in Piedmont and Liguria during the period 2021-2022

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Globalization and climate change in Europe increase the risk of Mosquito-Borne Diseases (MBDs) spread. Since 2020, the National Plan for Prevention, Surveillance, and Response to Arboviruses 2020-2025 (PNA) has been active in Italy, incorporating measures to prevent pathogen transmission including risk communication, training, environmental measures, vector control, and blood and blood component donation monitoring. This study reports the 2021-2022 surveillance results of West Nile (WNV) and Usutu viruses in northwestern Italy.

The regional territory (below 600m altitude) of Piedmont and Liguria was divided into 15x15 km grids for trap placement based on risk criteria. CDC-CO2 light, BG-Sentinel and gravid traps were used. Collections occurred every 15 days from May to November, followed by morphological identification and molecular testing of female mosquitoes for WNV and USUV.

In 2021, 13,971 mosquitoes (1,105 pools) were analyzed in Piedmont and 3,502 (341 pools) in Liguria. Piedmont had 3 WNV-positive and 1 USUV-positive pools; Liguria had no positives. In 2022, in Piedmont were captured 39,337 mosquitoes (1,320 pools) with 42 WNV-positive and 9 USUV-positive pools; in Liguria 7,839 mosquitoes (405 pools) were analyzed with one USUV-positive pool. The dominant mosquito shifted from *Culex pipiens* to *Ochlerotatus caspius*, increasing from 36% to 54% in Piedmont. Moreover, molecular investigations revealed 6 WNV-positive pools in *Oc. caspius* in 2022 compared to 4 in 2014-2020.

In conclusion, these findings underscore the critical role of vector surveillance in analyzing risk factors for both human and animal health, as well as in characterizing the distribution patterns of both native and newly introduced mosquito species.

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West Nile and Usutu surveillance in Abruzzo and Molise regions (Central Italy): entomological approach and data management

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Introduction and Objectives

Within the West Nile (WN) and Usutu (USU) surveillance system in Italy, mosquito collections are performed to early detect virus circulation in both “high-risk” and “low-risk” areas. In Abruzzo and Molise regions, categorised as low-risk areas, the surveillance has been ongoing at selected sites since 2019. Real-time results are disseminated to stakeholders via a dashboard. We hereby present the findings concerning mosquito species distribution and virus detection, as well as their visualization and accessibility.

Materials and Methods

Suction traps baited with light and CO₂, operating overnight, were used for mosquito collections. Over five seasons (2019-2023), 60 collection sites were sampled weekly or fortnightly. Mosquitoes were identified to species level, and females were sorted into pools of up to 50 specimens and tested for WNV and USUV by real time RT-PCR. A web-based interactive GIS dashboard, developed using ESRI technology and hosted on the ArcGIS Online cloud platform, displays spatial distribution, frequency, and identified species in the entomological sites.

Results

In total, 50,498 mosquitoes (from 2,572 collections) were identified, representing 22 species and seven genera. Female mosquitoes were sorted into 4,214 pools. All pools tested negative for WNV, while USUV was detected in 36 pools. The dashboard is freely accessible, offers versatile filtering options and illustrates:

- Distribution and abundance of species in different years, regions, and provinces;
- Species composition at each site;
- Positive pools for WNV or USUV, including Minimum Infection Rate (MIR) per site/year.

Discussion and Conclusion

This approach provides an immediate overview of entomological surveillance in the Abruzzo and Molise regions, continuously updated. The automated dissemination of laboratory results (including virus detection) through a daily scheduled data pipeline enhances its effectiveness. This methodology could be applied to other regions and vectors.

Tick infestation on migratory birds and spread of different *Borrelia* and *Rickettsia* species: a seasonal study in Lombardy region (Northern of Italy)

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Several tick species are known to infest birds, which can disperse ticks over short, medium and long distances. These ticks can be infected by tick-borne pathogens. Only a few studies have been conducted in Italy to investigate the tick fauna in birds and the harboured pathogens.

Ticks were collected from migratory birds captured at the Passo Spino bird observatory, Lombardy region, Italy, from August to November 2023. Ticks were morphological and molecularly identified, and the presence of bacteria (*Borrelia* spp., *Anaplasma* spp., *Rickettsia* spp., *Coxiella burnetii*) and viruses (TBEV, CCHF, WN, USUTU, Phlebovirus) was determined by quantitative real-time PCR.

A total of 7577 bird individuals of 63 species were examined for ticks; 87 bird individuals of 10 species were infested with at least one tick specimen.

A diverse range of tick species was collected from the infested birds, with the majority (95.3%) identified as *Ixodes ricinus* (112 larvae, 9 nymphs, 43 females). The remaining ticks included *Ixodes arboricola* (0.6%), *Ixodes acuminatus* (0.6%), and *Ixodes caledonicus* (3.5%).

Most ticks were found on the *Fringilla coelebs* and *Fringilla montifringilla* (46% and 23%, respectively). Of all ticks analysed, 16% were positive in real-time PCR for *Borrelia* spp., and 11% were positive for *Rickettsia* spp. All 172 ticks were negative in real-time PCR for *Anaplasma* spp, *Coxiella burnetii*, TBEV, CCHF, WN, USUTU, and Phlebovirus. Three different *Borrelia* and two different *Rickettsia* species were identified: *B. burgdorferi* s.l. was the predominant species and detected in 37% of all *Borrelia*-containing ticks, followed by *B. afzelii*, 26% and *B. miyamotoi*, 7.4%. The rickettsiae were identified as *R. helvetica*, 29.2% and *R. monacensis*, 3.4%.

These findings underline the crucial role of migrating birds in the long-distance dispersal of human-pathogenic or potentially pathogenic *Borrelia* and *Rickettsia*, shedding light on the transmission dynamics of tick-borne diseases.

Use of mobile applications and Geographic Information Systems for enhanced characterization of *Aedes albopictus* risk areas in urban environments

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Identification of *Aedes* hotspots in urban settings empowers the implementation of effective surveillance and control strategies for vector-borne diseases such as Dengue, Chikungunya, and Zika.

To achieve this objective, the MediLabSecure network, funded by the European Commission, organized a training in Tirana (Albania) in September 2023 with the support of Avia-GIS, the French National Research Institute for Sustainable Development (IRD) and the Institute of Public Health in Albania, as the local focal point.

The training was structured into two parts: the initial session held in Tirana, followed by remote follow-up sessions to reinforce skills and generate maps. Twelve trainees from eight countries participated, selected through entomological focal points within beneficiary institutes of MediLabSecure Network, spanning Albania, Armenia, Bosnia and Herzegovina, Georgia, Kosovo*, Montenegro, Palestine, and Turkey.

Field activities were carried out in an urban park and three areas in Tirana, each with a different degree of urbanization. They encompassed mosquito collection using a range of tools, including human landing catches, ovitraps, and identification of larval breeding sites. These activities were supplemented by spatial mapping using VECMAP mobile app with additional environmental data such as site characteristics, temperature, and wind conditions.

The mosquito species collected in the field were subsequently identified in the laboratory. The eggs retrieved from the traps were quantified, and the collected larvae were raised using equipment provided by the Institute of Public Health, under the guidance of expert entomologists.

The following online training focused on mastering the basics of QGIS for generating maps depicting the distribution areas of *Aedes albopictus* within the different districts of Tirana with the relevant metadata. The skills and expertise acquired can constitute a solid foundation for strengthening *Aedes* control efforts in the various countries of the MediLabSecure network, while reinforcing the implementation of the «One Health» approach.

S7-P15

Why do tiger mosquitoes spread in certain areas of the city of Vienna faster than in others? The role of tree bags and tree holes for alien *Aedes* species

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Introduction and Objectives

In 2020 tiger mosquitoes (*Aedes albopictus*) were first reported in Vienna in an allotment garden in the Prater (a large public park). An ovitrap-based monitoring from 2022 indicates establishment and spread of the tiger mosquito but *Ae. japonicus* and *Ae. koreicus* have also been found in the entire Prater. The latter mainly consists of renaturation areas where a lot of dead trees and tree holes are present. However, the highest egg abundances were documented in the locations between the main highway in Vienna and a railway bridge, as well as in a forested dog area, where almost no artificial breeding sites are present.

Methods

To evaluate the importance of tree holes as breeding sites in a non-native urban environment, more than 20 tree holes of various tree species (e.g. maples and lindens) were examined for the presence of mosquito larvae during the season 2023. Moreover, a few tree bags were included in 2023. In 2024 the focus is set on the role of tree-bags (n= approx. 400) in i) Prater area (an area where tiger mosquitoes spread rapidly), ii) in the neighbouring urban area (where tiger mosquitoes were first observed in 2023), iii) in an urban area where tiger mosquitoes were observed since 2020 in low abundance, and iv) nearby park and urban areas where tiger mosquitoes have not been observed yet. Additionally, ovitraps were set up in this study area and checked weekly. Mosquito larvae and eggs were identified morphologically and further analysed with molecular techniques to species level when necessary.

Results and Discussion

Aedes albopictus immatures were found in tree holes but in a much lower abundance than in tree bags (July-October). Moreover, other alien *Aedes* spp. were detected. Results of this currently ongoing study are presented in terms of diversity and abundance.

S7-P16

Identifying ecological factors mediating the spread of three invasive mosquito species: citizen science informed prediction and evaluation

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Due to their potential role in pathogen transmission, invasive mosquitos pose considerable threats to human and animal health. Several studies have identified the most important ecological drivers mediating the establishment and spread of the key species (e.g. *Aedes aegypti*, *Ae. albopictus*), and made predictions for future distribution. We evaluated the effect of an exhaustive list of environmental predictors on distribution of three invasive species in Hungary (*Ae. albopictus*, *Ae. japonicus*, and *Ae. koreicus*) by using the same standards for data collection based on citizen science observations. Current distribution maps of these species were generated from a 5-year survey, then were compared with various predictor maps reflecting climate, habitat type, food supply, traffic, interspecific competition by using a Boosted Regression Trees approach that resulted in a subset of variables with the strongest impact. The best predictor sets were used to predict the probability of occurrence of the focal species on regions, where no citizen science reports were available. These predictions were evaluated against the results of a recent field surveillance focusing on previously unsampled areas. We found that different predictor sets were selected for the three different species, and only predictions for *Ae. albopictus* could be validated with direct trapping data. Therefore, citizen science informed distribution maps can be used to identify ecological predictors that determine the spread of invasive mosquitos, and also to draw risk map based on the predicted distribution. However, we uncovered species-specific patterns highlighting that extrapolating on unsampled areas based on observed presence/environment relationships may sometimes be unrealistic.

20 years of entomological surveillance of *Culicoides imicola* on the French Mediterranean littoral

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Culicoides imicola, an Afrotropical biting midge species, is historically known to transmit orbiviruses of veterinary importance, including bluetongue virus (BTV), epizootic haemorrhagic disease virus, and African horse sickness virus. Distributed around the Mediterranean basin, *C. imicola* was first recorded in France in September 2000 on the Mediterranean island of Corsica, shortly before the emergence of BTV serotype 2 in October 2000. Three years later, individuals were collected on the French mainland in the Var and Alpes-Maritimes departments. Unexpectedly, a small population was also observed in the Pyr  n  es-Orientales department near Spain between 2008 and 2012.

Since its initial detection on mainland France, annual entomological surveillance using OVI traps has been conducted at the end of summer to (i) investigate the spatial distribution along the French Mediterranean coast and (ii) monitor abundance peaks over time.

Twenty years of surveillance data, associated with results of genetic studies, reveal passive dispersal of *C. imicola* individuals from Corsica to mainland, and that *C. imicola* maintains a stable yet sparse distribution on the French Mediterranean coast, in contrast to its higher abundance in Corsica. The population of the Pyr  n  es-Orientales department appears to have collapsed after 4 years of limited distribution and low abundance. As the northernmost known population, it is hypothesized that the French continental *C. imicola* population has struggled to adapt to local environmental and climatic conditions, thus limiting until now its potential for further northern expansion.

Sand flies sampling and pathogens detection in Northeastern Italy

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Sand flies are vectors of pathogens as *Leishmania* spp. and Phlebovirus; phleboviruses are causative agents of summer fevers and among these, Toscana Virus (TOSV) is associated with neurological disorders. To date, there is still a lack of knowledge on TOSV epidemiology and no specific surveillance plans are applied. In the present study, we report a survey for sand flies presence/abundance and pathogens detection in Veneto region. Eight sites were selected in 2022 and two in 2023 according to geographical distribution of reported TOSV infection in human.

Sand flies were trapped using one CD-CO₂-light trap and ten sticky traps running for one night every two weeks per site. Thus, phlebotominae were morphologically identified, counted, sexed and pooled for *Leishmania infantum* and *Phlebovirus* spp. molecular investigation.

In 2022 3287 *P. perniciosus* (79.7%), 74 *P. neglectus* (1.79%), 8 *P. perfiliewi* (0.19%), 53 *Sergentomya minuta* (1.28%) and 700 *Phlebotomus* spp. (16.9%) were collected. Twenty-two pools resulted positive for *L. infantum* (18 in *P. perniciosus*, 3 in *S. minuta* and 1 in *P. neglectus*), and one pool of 35 males of *P. perniciosus* was positive for TOSV. In 2023 779 *P. perniciosus* (48.1%), 21 *P. neglectus* (1.29%), 41 *P. perfiliewi* (2.53%), 1 *S. minuta* (0.06%) and 777 *Phlebotomus* spp. (47.9%) were collected. Pools of *P. perniciosus* were positive for *L. infantum* (one pool) and Phleboviruses (one pool for TOSV, one for Punique virus, and one for *Phlebovirus* spp.).

This study demonstrated the presence of several phlebotominae species in Veneto region and *P. perniciosus* resulted the predominant species and infected with phleboviruses and *L. infantum*. Molecular investigations revealed the first detection of TOSV in *P. perniciosus* in Veneto and the first evidence of Punique virus in Italy and Europe. These findings should promote specific monitoring activities on sandflies as a preventing factor for public health.

Nextgen surveillance tools to monitor mosquito vector populations at a fine temporal scale

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Introduction and Objectives

The use of smart-traps for automated remote mosquito surveillance is on the vanguard of the entomological research. These new approaches offer a promising perspective for the future of the field by reducing the time and costs involved in traditional surveillance and providing high-quality data for research with an unprecedented time-resolution. The objective of the present study was to develop a practical example of how to take profit from the nextgen mosquito monitoring tools to obtain fine-scale data, key for a comprehensive understanding of vector ecology.

Materials and Methods

In this work, an automated entomological monitoring station, based on an optical sensor coupled to the entrance of a conventional mosquito suction trap, was used to monitor mosquito activity dynamics for three consecutive years (2021-2023) at different temporal scales (from seasonal to sub-daily) in a botanical garden in the province of Girona (Catalunya, Spain).

Results

A total of 9165 flying insects entered the trap between 2021-2023, from which only 12.3% (N=1448) were mosquitoes. The sensor discriminated *Aedes* and *Culex* mosquitoes from other insects with a mean balanced accuracy (BA) of 96%; and classified their genus and sex with 86% of BA. The daily activity of *Aedes albopictus* and *Culex pipiens* exhibited two pronounced peaks. A certain degree of overlap was observed among them, with *Ae. albopictus* being more diurnal and *Cx. pipiens* being more crepuscular. These activity peaks had a seasonal variation according to mosquito phenology. Mosquito and human activity at the botanical garden showed a temporal segregation.

Discussion and Conclusion

Smart-traps can be powerful tools to remotely and automatically classify mosquitoes in real-time. They can offer a unique opportunity for vector surveillance and for an early-warning response to different epidemiological scenarios; but they are also interesting tools to explore vector behaviour and ecology at daily and sub-daily scales.

S7-P20

From research to operational: Feedback from mosquito surveillance in southern France, from an environmental to a vector surveillance approach

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Since 2008, EID Méditerranée, the public operator responsible for mosquito control in the south of France, has set up a network of traps to monitor the biodiversity and abundance of mosquitoes in the 220 communes where it operates. The network is used to monitor the hatching dynamics of the fifteen or so main biting mosquitoes, from among the 50 species of mosquito present on the French Mediterranean coast. No fewer than thirty-six CO₂ traps are deployed every week between April and October in the 60,000 hectares of wetlands monitored by the EID-Med.

The extension of this network not only for the study of mosquito biodiversity, but also for mosquito-borne diseases (MVD) such as West Nile Virus (WNV) over such a long area, remains unexplored and requires coordination and protocols standardisation to offer reliable results. In this study, we show the results of implementing a low-cost device for molecular xenomonitoring (MX) in our trap network (L'Ambert et al 2023). We demonstrate the feasibility and usefulness of using this approach in field activity for the surveillance of MBD, as well as the vigilance points to be considered for this type of real-time monitoring of a large area.

By sharing these results, we hope to facilitate the establishment of extensive surveillance networks, which will increase the collection of reliable data to contribute to a better understanding of the epidemiology of WNV and the early detection of virus circulation.

arbocartoR: a simulation tool for decision support in the implementation of surveillance and control of mosquito-borne diseases

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The recent expansion of the invasive *Aedes* mosquitoes give rise to serious concerns about the risk of the emergence of arboviruses in new territories (eg. dengue, Zika and chikungunya). Although strongly affected by unpredictable random effects, the emergence and spread of vector-borne viruses depend on vector and host abundances and movements, as well as the introduction scenario and vector control implementation. Modeling offers powerful tools to understand the underlying mechanisms of emergence and explore epidemiological scenarios. arbocartoR is a modeling tool developed at CIRAD as part of the H2020 MOOD project. The underlying model is a compartmental model built on a mechanistic approach based on observation and description of biological and behavioral processes. It integrates both deterministic and stochastic algorithms. arbocartoR can be used to spatially simulate and explore (i) the population dynamics of two *Aedes* species: *albopictus* & *aegypti*, considering local conditions (precipitation, temperature, breeding sites), and (ii) the transmission dynamics of *Aedes*-borne viruses following introduction events and taking into account the responsiveness and effectiveness of control strategies. It provides key figures to guide vector control and surveillance in order to better understand, anticipate, and prepare for the risk of mosquito-borne disease emergence and spread.

Citizen science for surveillance of tick bites on humans and tick-borne pathogens in ticks in Belgium

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Introduction

In Belgium, ticks are present countrywide with *Ixodes ricinus* being the most common species biting humans. *Ixodes ricinus* is known to be a carrier of several tick-borne pathogens (TBP), posing a potential health risk to humans. In order to understand better the risk of tick bites on humans in time and space, TekenNet, a citizen surveillance platform was launched in 2015. The robust collection of data through citizen science allowed us to conduct additional research: risk maps were created and the prevalence of TBPs in ticks was studied.

Methods

Citizens report tick bites via TekenNet (web/app) including, among others, information about the geographical location and activity. Every four years, citizens can send the tick that bit them to Sciensano where it is analyzed for TBP. For the development of the risk maps, Bayesian disease mapping of tick bite reports was performed taking into account environmental predictors (hazard and exposure).

Results

Over eight years, citizens reported 62 492 tick bites during 40 212 reporting events. Ticks were mainly active between March and September with a peak in June. Yearly variations were observed. Citizens mainly report tick bites that occurred close to their home, in the garden or forest, while doing leisure activities. The most prevalent pathogens were *Borrelia burgdorferi* in 2017 and *Rickettsia helvetica* in 2021. Public health relevance of the latter is expected to be limited. The risk map classified all municipalities within one of three risk classes.

Conclusion

The presence of a continuous and stable surveillance system for human tick bites allowed us to better understand the tick bite risk and prevalence of TBP in Belgium. Risk factors in time and space could be identified for the Belgian population. The risk maps are a tool to support the implementation of local prevention measures by municipalities.



S7-P23

Surveillance and temporal population dynamics of *Culex pipiens* s.l. in Scotland

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Mosquito-borne diseases pose a growing threat in northern Europe as environmental and anthropogenic changes enable vector species and pathogens to emerge in previously disease-free areas. In Britain, the distribution and temporal dynamics of *Culex pipiens* s.l. are major risk factors for the establishment of zoonotic avian diseases such as West Nile Virus (WNV) and Usutu Virus. Surveillance of native vector species is therefore key to monitoring and predicting the risk of disease emergence. The «Mosquito Scotland» project aims to assess this risk in Scotland, where historically there has been a considerable gap in mosquito surveillance. Here, we present findings from our targeted surveillance of *Cx. pipiens* s.l. populations. The primary objective of this work was to characterise the seasonal population dynamics of *Cx. pipiens* s.l. within Scotland. Sampling was conducted at three sites across a latitudinal gradient between April and October 2024. Three buckets filled with hay infusion were set up at each site to attract ovipositing *Cx. pipiens* s.l. females. Buckets were monitored for egg rafts and larvae 2-3 times per week throughout the season. Adults were collected up to twice per week using Biogents BG Pro traps baited with carbon dioxide and a BG lure. Adult wing measurements were taken as a proxy for body size. Further to the field surveillance, an online citizen science tool was developed to enable public reporting of mosquitoes across Scotland, with a secondary aim of identifying populations of *Cx. pipiens molestus* via reports of nuisance biting in residential areas. Initial findings indicate that native vector species are widely distributed throughout mainland Scotland, suggesting that vector distributions may not be a limiting factor in mosquito-borne disease risk. The data collected here will be used to build the framework for an epidemiological model of zoonotic virus emergence in Scotland.

S7-P24

Comprehensive Surveillance of Mosquitoes and Sandflies in Slovenia: Implications for Public Health and Emerging Pathogens

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The global dissemination of infectious diseases constitutes a significant risk to public health, as novel pathogens emerge in previously unaffected areas. These threats primarily include arboviruses and parasites like *Leishmania* and *Plasmodium*, which have a significant potential to spread due to the increasing prevalence of their vectors. It's therefore critical to research new pathogens with a harmonized approach and to establish sensitive surveillance systems.

Our targeted research, conducted between 2020 and 2022, focused on mosquitoes (Diptera: Culicidae) and sandflies (Diptera: Phlebotominae), which are the primary vectors of human diseases. Our aim was to catalogue the mosquito and sand fly species in Slovenia, describe the distribution and abundance of both autochthonous and invasive species, and compile data on the presence or absence of viral and parasitic pathogens transmitted by these vectors. We implemented standardized monitoring protocols for mosquitoes and sand flies in accordance with European guidelines. Over a three-year period, we surveyed vectors at 226 sites across Slovenia. We captured and identified over 85,000 adult mosquitoes belonging to 41 species, including seven species newly documented in the Slovenian mosquito fauna. During the sampling, we detected various microorganisms, including Usutu and Sindbis viruses and the parasite *Dirofilaria repens*. By developing predictive maps, we have delineated regions with an increased probability of harboring medically significant mosquitoes.

During sand fly surveillance, we caught over 1,200 individuals and identified four species, with the highest population density observed in the Primorsko-Notranjska and Goriška regions. Notably, no *Leishmania* species were detected. The collected data on the distribution of vectors and the emergence of novel microorganisms have proven to be crucial for assessing the risk associated with emerging pathogens and identifying locations with increased likelihood of mosquito and sand fly presence. These results contribute significantly to improving our comprehension of epidemiological drivers and the establishment of effective surveillance strategies.

CANCELLED

Spatial distribution of pyrethroid resistance-associated mutations in *Aedes albopictus* populations across southern France

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Introduction and Objectives

Preventing major *Aedes*-borne diseases heavily relies on the use of insecticides. As the first signs of pyrethroids resistance in *Aedes albopictus* have been detected in Europe, a cost-effective molecular method is needed to large scale studies of pyrethroid resistance-associated mutations.

Materials and Methods

In this study, we screened for the spatial distribution of knockdown resistance (*kdr*) mutations in *Ae. albopictus* across southern France using a two-step approach based on multiplexed amplicon sequencing, realized in pool DNA from adults or larval instars collected in 95 sites.

Results, Discussion and Conclusion

Our approach efficiently screened for *kdr* mutations in multiple sites at low cost. Subsequent validation through individual mosquito DNA sequencing validated these findings while giving access to *kdr* allele prevalences and genotypes. Initial pooled mosquito DNA sequencing revealed the presence of the V1016G allele in 14 distinct sites, notably distributed in the south-eastern part of France, close to the Italian border. Isolated occurrences of the V1016G allele were also detected in south-western area near Bordeaux and Marmande. Remarkably, all identified mosquitoes carrying the V1016G mutation were found to be heterozygous, suggesting a lack of phenotypic resistance to pyrethroid insecticides. Furthermore, our investigation identified four additional mutations within our targeted genomic sequence: I1532T, M1006L, M1586L, and M995L. Through the sequencing of a segment of the maternally inherited mitochondrial genome, we confirmed that the colonization of *Ae. albopictus* in France originated from founders with haplogroup A1. These findings contribute to the broader understanding of resistance dynamics of *Ae. albopictus* populations, which will become an issue for the future of vector control in Europe.



S7-P26

Field evaluation of four commercially available mosquito traps for collecting mosquitoes in Scotland

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Introduction and Objectives

As vector-borne diseases (VBDs) increasingly affect northern Europe, monitoring temperate mosquito populations becomes crucial for assessing disease risk. In Scotland, where mosquitoes are less common and understudied, comparing trap types is essential for identifying effective surveillance methods. This study marks the first assessment of various commercially-available mosquito traps in Scotland, aiming to evaluate their effectiveness in estimating mosquito abundance and species diversity.

Methods

This study conducted two experiments in an urban marsh in Glasgow City, known for its mosquito presence, during August 2023. Firstly, three trap types—Biogents BG-Pro (BGP), Biogents BG-Sentinel (BGS), and CDC miniature light traps (CDC)—were compared over 15 days using a 3 x 3 Latin square design. A subsequent experiment incorporated the Mosquito Magnet (MM) trap, comparing it with the initial three traps over 12 days using a 4 x 4 Latin square design.

Results

Over the 93 trap nights, 150 mosquitoes were collected from eight species. The most common species collected was *Anopheles claviger* (n= 63), followed by *Culiseta morsitans* (n= 28). In experiment one, the mean no. of mosquitoes per trapping period was 1.02 (SE=6.71), with no significant difference between the trapping methods. In the second experiment, the MM caught significantly more mosquitoes than any other method, with a mean of 7.5 per (SE=0.53) trapping period ($p = <0.01$). The MM also caught the greatest number of species (6), predominantly *Anopheles claviger*, followed by the CDC (4), BGP(3) and lastly, the BGS(1).

Discussion and Conclusions

The MM caught significantly more mosquitoes than other methods and the greatest number of species. This suggests that this trap type could be optimal for sampling Scottish mosquito populations. The data here will inform future trapping protocols to enhance ongoing mosquito surveillance in Scotland.



S7-P27

Advancing arbovirus surveillance in the Netherlands: Field evaluation of FTA cards and citizen science integration

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Introduction and Objectives

Understanding the circulation of arboviruses like USUV, WNV, and SINV is critical for public health and wildlife health in the Netherlands. Current surveillance scheme for mosquito and arboviruses includes 11 bird ringing locations across the Netherlands. However, risk-targeted surveillance faces logistical and time limitations, such as mosquito processing and laboratory capacity. Our study evaluates the field deployment of FTA-cards (Flinders Technology Associates) for mosquito saliva and excreta collection aiming to enhance current arbovirus surveillance methods. Integrating FTA cards in current surveillance and further use within citizen science initiatives while leveraging postal services for transportation, could increase surveillance capacity and response during arbovirus detection.

Materials and Methods

Initial mosquitoes trapping was conducted in Leiden over 24h period at 12 sites. CO₂-baited BG-Pro traps (Bio-gents AG, Germany) were used. FTA cards (3cmx3cm cut-outs) with honey-water 3:1 ratio (0.3g blue dye) were placed inside the traps. Mosquitos were sorted, identified and their feeding status determined. FTA-cards were stored at -80°C until further testing. In parallel, validation of the MX adapter (L'Ambert et al, 2023) for mosquito excreta collection was carried-out within the same framework. Next, to facilitate implementation by citizen scientists honey-baited cards were prepared, frozen and distributed to designated bird-ringing locations across the Netherlands, where surveillance for mosquitoes and arboviruses has been conducted over the past four years. Subsequently, participating birders returned the cards via postal services.

Results, Discussion and Conclusion

Preliminary findings show a high attraction rate of mosquitoes to honey-baited cards, with an average 75% feeding rate and similar rates in *Culicoides spp.* Besides, 4% of mosquitoes had a prior blood-meal. USUV detection on FTA cards was less sensitive than in mosquito pools. Our results demonstrate that FTA cards serve as a cost-effective and straightforward supplement to existing monitoring schemes, potentially expediting the near-real-time detection of arboviruses.



S7-P28

Strong resistance with cuticular thickening in the diflubenzuron-resistant phenotype of *Culex pipiens* raises new concerns for mosquito control

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The selection of insecticide-resistant phenotypes in disease vector species is an increasingly common problem, hindering the effectiveness of control actions worldwide. In recent decades, several resistance cases have been documented against Chitin Synthesis Inhibitors (CSIs), insecticides that are safe for most non-target organisms and represent one major tool in Integrated Pest Management (IPM). Resistance to the CSI diflubenzuron (DFB) has been recently detected in some European populations of the mosquito vector *Culex pipiens*, and it is determined by three different alleles (I1043M, I1043L, and I1043F), resulting from aminoacidic substitutions at position 1043 of the chitin-synthase gene (*chs-1*). Although the genetic mechanism underlying this resistance has been defined, its effects on the phenotype are still unclear.

Here, by comparing susceptible and resistant laboratory colonies established from field samples, we explored the molecular and phenotypic characteristics of a DFB-resistant strain of *Cx. pipiens* homozygous for the I1043M mutation. Performing DFB bioassays on mosquito larvae, we revealed a Resistance Ratio of 9,006, which suggests extremely high resistance levels. Moreover, through molecular analysis and scanning electron microscopy, we found that resistant mosquitoes have an 11-fold constitutive over-expression of the *chs-1* gene and a modified cuticle, characterized by increased thickening and chitin content.

The above findings have interesting implications for resistance management in the field and the planning of alternative control strategies. Firstly, since the I1043M has the broadest distribution and the greatest relative frequency among the three resistant alleles, the evidence of such high resistance levels poses a serious threat regarding the future use of DFB as a control tool. Secondly, the observed cuticle modifications may establish cross-resistance mechanisms, reducing the penetration of other insecticidal molecules, as well as they may have an impact on individual fitness. Further studies will be needed to investigate these aspects.

Strengthening the control of invasive mosquitoes at Points of Entry: a longitudinal monitoring in port and airports of Northeastern Italy

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The ecology of mosquito borne diseases has changed significantly during the last decades, primarily due to anthropic impact and globalization. Human transport of eggs and adult mosquitoes has facilitated the dispersal of different *Aedes* species, expanding globally the distribution range of pathogens they can transmit. Vector surveillance and control at Points of Entry (PoE) has become an essential issue to prevent the spread of vectors in new areas. In Italy, a specific monitoring program was established at PoE of northeastern region following the introduction of the Asian *Aedes albopictus*, *Aedes koreicus*, and *Aedes japonicus*. Specifically, a pilot survey (2018) and an entomological monitoring (2019-2023) were carried out at port and airports of Veneto region (Treviso airport, Venice port and airport), using ovitraps and BG-sentinel (BG-S) working weekly from June to October. A total of 58,414 eggs and 24,355 adults were collected. Eggs belonged all to *Ae. albopictus* except one egg of *Ae. koreicus* found in Venice airport during the pilot survey (2018). Thirteen species were collected with BG-S traps and the most represented were *Culex pipiens* (47.3%) and *Ae. albopictus* (50.7%). Two adults of *Ae. koreicus* were identified in 2019 at Treviso airport and one at Venice airport in 2020. Although this species was already reported in Veneto region, this was the first finding in Venice province. Since its discovery in Venice province, *Aedes koreicus* has been found yearly in the municipality of Mirano (18 km from the Venice airport) and it is possible that the species was introduced via airport and then dispersed to another site. Although a direct link cannot be established, this finding suggests the need to strengthen the controls at PoE to prevent the rapid spread of invasive mosquitoes.

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VectorNet-Spain: giving support to European and national vector surveillance via national experts

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VectorNet is a collaborative European network funded by the European Centre for Disease Prevention and Control (ECDC) and the European Food Safety Authority (EFSA) to improve preparedness and response to vector-borne diseases. The network supports the collection of vector and pathogen-vector data related to both animal and human diseases.

VectorNet was run during two periods 2014-2018 and 2019-2023 and recently, the new period (starting in 2024) includes a «One Health» approach to promote the development of a database on the presence and distribution of vectors throughout Europe and the Mediterranean basin.

Since 2020, an unofficial national collaborative network of medical and veterinary entomologists, called **VectorNet-Spain**, was established. The network at national level supported the network at European level by responding to questionnaires and/or providing expert information on vectors. To date, VectorNet-Spain includes 84 members, of which 73% are linked to research, 16% to management (administration) and 11% to public or private companies. As for the expertise, 39% are experts in mosquitoes (Culicidae), 25% in sandflies (Psychodidae), 16% in ticks (Ixodidae and Argasidae), 11% in biting midges (Ceratopogonidae) and 8% in other groups.

To establish the new One-Health VectorNet Entomological Network (OHVEN) at European level, both ECDC and EFSA requested the designation at national level of two Operational Contact Points (OCPs) for entomological matters in the country. The Spanish OCPs are therefore the liaison between the national network VectorNet-Spain and the national and European authorities. Future roles of VectorNet-Spain within the new «One Health» framework would be to establish and strengthen a collaborative and operational framework among national entomologists, to integrate this network as part of the National Plan for the Prevention, Surveillance and Control of vector-borne diseases and to consolidate the transfer of entomological surveillance data related to human and animal health at national and European level.



S7-P31

Potentials and limitations in the application of Convolutional Neural Networks for mosquito species identification using wing images

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Mosquito-borne diseases pose a significant global health challenge, particularly in tropical and subtropical regions, exacerbated by factors such as global warming. Accurate identification of mosquito species is crucial for entomological surveillance programs. However, molecular, and morphological identification methods are costly and require specialized expertise.

Recent advances in Convolutional Neural Networks (CNNs) hold promise for accurately identifying mosquito species from images alone. Previously, we presented a classification model capable of identifying 14 different mosquito species with high accuracy using wing images. However, the efficacy of CNN models is often limited to controlled environments. Traditional CNN pipelines, while effective in specific contexts, often fail to perform adequately when faced with the challenges of real-world deployment. In this study, we investigate the performance of CNNs for mosquito species classification based on wing images, addressing the challenges of real-world deployment, including the use of different imaging devices.

We constructed a dataset of over 14,000 wing images of female mosquitoes from 68 species across 9 genera, captured by 3 different imaging devices. Our analysis revealed the impact of imaging devices and corresponding biases on CNN performance in an out-of-distribution context. Based on our findings, we identify strategies to improve model reliability in different settings. We demonstrate that the selective use of augmentation and pre-processing techniques can be used to steer CNN models toward a more reasonable classification decision, producing reliable results on multiple devices. Finally, we achieved a balanced accuracy of over 90% to classify 24 mosquito species based on wing images. Our results underscore the potential of CNNs for insect identification, but also highlight the need to address real-world challenges. This study provides valuable insights into the development of robust CNN-based classification systems for species identification, particularly for vector surveillance and research.

Stop-Aedes, a collaborative board game to promote collective action to control Aedes vectors

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The tiger mosquito, *Aedes albopictus*, is the vector of several major arboviruses, including dengue, chikungunya and Zika viruses. The vector-borne diseases caused by these viruses are increasing worldwide. In Europe, autochthonous cases of dengue fever are regularly recorded and have recently increased. In addition, arbovirus epidemics occurs in French overseas departments such as La Réunion, for over twenty years. To be effective, the control of *Aedes albopictus* requires a strategy that combines and integrates several control methods, including chemical and biological control, mechanical destruction of breeding sites and social mobilization. Social mobilization is a major factor in effective mosquito control. Although a number of awareness-raising tools exists, and the general public is familiar with good practices for the control of *Aedes albopictus*, it is still necessary to move on to concrete action and get people committed to it. This calls for the development of new tools to raise public awareness of their role and the importance of their actions in the collective fight against the tiger mosquito. Thus, we developed the collaborative board game *Stop Aedes* aiming to (1) promote good practices in social mobilization to control *Aedes albopictus*, (2) initiate dialogue between stakeholders, including citizens, and (3) reinforce collective action. Following an iterative process including development and field-testing phases between 2022 and 2023, a version of the board game adapted to the context of La Réunion has been developed. The aim of the game is to eliminate all potential breeding sites in courtyards, gardens and streets of a neighbourhood. A version adapted to the European context is currently under construction, and the game will also be adapted to other contexts, such as the control of *Aedes aegypti* in Brazil.

Tick-borne encephalitis in the Slovak Republic

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Introduction and Objectives

Tick-borne encephalitis (TBE) is a viral disease affecting humans and warm-blooded animals. Incubation period is 7 to 28 days; the disease usually has a two-phase clinical course. TBEV has its reservoirs in rodents and it is transmitted by ticks. TBEV can also be transmitted via the alimentary route through milk from infected animals.

Materials and Methods

Human blood samples, along with blood samples collected from sheep and goats were used for detection of TBEV RNA using the RT-PCR method. Tick samples were collected directly from vegetation and tested by RT-PCR. Sera were used for detection of antibodies by serological methods.

Results, Discussion and Conclusion

Over the past 5 years, 643 cases of TBE were reported in Slovakia. The highest morbidity was in the Žilina, Banská Bystrica and the Trenčín region. The most common cause was tick bite; the probable factor of transmission in each case being unpasteurized sheep's milk and products made from it. A total of 1113 milk samples were tested with a 1,8 % positivity rate. Antibody tests were carried out on 421 serum samples from sheep and goats and reported positive results in 8.36 % of cases; 393 samples of blood from wild birds were collected, from which 8 blackbirds (2%) were seropositive. By RT-PCR method, 11 samples of native blood and organs of goats were examined with a negative result; 2787 tick samples collected directly from vegetation were tested; positive results were confirmed in 9,44 % of cases (BMC Institute of Virology at the Slovak Academy of Sciences). In 2022, a fatal case of a man was reported; the patient was unvaccinated (RUVZ, Banská Bystrica).

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S7-P34

Detection of Usutu Virus in *Culex pipiens* Mosquitoes Collected in Greece

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Usutu virus (USUV) is an arbovirus closely related to endemic in Greece West Nile virus (WNV). It is a member of the flaviviridae family, which belongs to the Japanese encephalitis complex. Since USUV emergence in Europe, a number of studies, regarding its circulation have been performed. In nature, its transmission cycle involves mosquitoes, mainly of the *Culex* genus and wild birds acting as reservoirs for viral proliferation. Moreover, USUV has been isolated from humans, horses and other mammals. This is the first study performed in Greece revealing the presence and circulation of USUV in mosquito population. We conducted molecular screening, applying a sensitive and specific Reverse transcription real-time -PCR method in 1.329 RNA samples isolated from *Culex pipiens* mosquito pools. Each pool contained from 2 to 70 adult female mosquitoes collected for the time-period 2020 – 2023, from the region of Attica and other parts of the country. Out of the 1.329 samples, USUV presence was detected in four samples, one from the year 2022 and the other 3 from 2020. RT real time PCR results have been further verified by conventional PCR and confirmed by Sanger Sequencing. These findings suggest co-circulation of USUV along with WNV in Greece, in areas with annual records of a substantial number of WNV human cases. Several studies have revealed co - circulation of the two viruses, consistent with the present study since one out of the four USUV positive pools was also positive for WNV. Our preliminary results suggest a need to carry out further studies and research in the field of USUV in Greece, both in mosquitoes but also in humans and other hosts. A One Health approach could improve the monitoring of its spread in Europe and assist public health authorities to implement surveillance and control measures.

Leishmaniasis: investigating the current extent of the epidemiological risk in Switzerland

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Introduction and Objectives

In Europe, the vector-borne disease leishmaniasis is endemic in Mediterranean countries, with the protozoan species *Leishmania infantum* (Kinetoplastida: Trypanosomatidae) causing canine (CanL) and zoonotic visceral (VL) and cutaneous (CL) leishmaniasis. In the last 30 years, the disease has gained importance in Central Europe, due to the gradual northward expansion of the vectors, namely, the phlebotomine sand flies of the genus *Phlebotomus* (Diptera: Phlebotominae), and the moving of infected dogs between endemic and non-endemic regions. In Switzerland, the sand fly *Ph. perniciosus*, vector of *L. infantum*, has been recorded south of the Alps, in the canton Ticino, where cases of imported CanL are also present. Therefore, the autochthonous transmission of *L. infantum* is a potential hazard that needs to be addressed. We investigated the extent of the epidemiological risk in Ticino and in the cantons Basel-Stadt and Basel-Landschaft close to the border with Germany, where populations of phlebotomine sand flies are present.

Materials and Methods

At the vector level, distribution and density of phlebotomine sand flies, as well as presence of *L. infantum* in female sand flies, was investigated at potentially suitable sites. At the host level, a serosurvey was carried out on 101 domestic dogs residing in Ticino.

Results, Discussion and Conclusion

No sand flies were captured in the cantons Basel-Stadt and Basel-Landschaft. In Ticino, 16 out of 20 sites surveyed were positive for phlebotomine sand flies. *Phlebotomus perniciosus* was the most abundant species, with site densities ranging from 0.1 to 5.3. No *L. infantum* was detected in female sand flies. A *L. infantum* seroprevalence of 3.0% (95% CI: 0.6-8.4%) was observed in the tested dogs. These results, along with the worsening epidemiological situation in neighbouring northern areas of Italy, emphasise the need for an integrated surveillance and prevention measures.

Updated distribution of kdr mutations associated with resistance to pyrethroids in European populations of *Aedes albopictus*

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Introduction and Objectives

Resistance to pyrethroids (PR) - the only insecticides authorized in case of autochthonous arboviruses transmission in Europe - has been reported in *Aedes albopictus* populations from Italy and Spain. Mutations at positions 1016, 1532 and 1534 of the voltage-gated sodium-channel (VGSC) gene are the most strongly associated with target-site PR in the species. Results of a European-wide investigation revealed a widespread presence of allele 1016G in *Ae. albopictus*, with frequencies up to 45% in Italy. Although four alleles at positions 1532 (T) and 1534 (C/L/S) have been described worldwide, in Europe *Ae. albopictus*, monitoring of 1534C was carried out only for *Ae. albopictus* from Italy and Greece, and the allele was found at frequencies up to 68% in the latter. Here, we show the results of a European-wide effort to map 1532 and 1534 mutations in *Ae. albopictus*.

Materials and Methods

Genotyping was performed by Allelic-Specific PCR (AS-PCR) in 19 European *Ae. albopictus* populations. Results were validated by sequencing in a sub-sample of specimens.

Results, Discussion and Conclusion

Among the 1,731 specimens genotyped, allele 1534C was found in Spain, Georgia, Romania, and Albania at frequencies <5%, in Greece at frequencies up to 47%, as well as in the recently colonized Cyprus (84%), suggesting the origin of the invasive population from Greece. Allele 1534L was only found in Rome (3.1%). Allele 1532T was detected in Spain, France, Italy, Greece, Albania and Turkey at 4%-25% frequencies. The high frequencies of allele 1016G in Italy and 1534C in Greece and the intense connections between the two countries open the possibility of cross-over between haplotypes carrying the two alleles resulting potentially in a strongly enhanced resistance phenotype, a risk which stresses further the need to carefully monitor resistance genotypes.



S7-P37

A mechanistic model to predict spatial-temporal patterns of *Culex pipiens s.s./Cx. torrentium* in Germany

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Mosquitoes are well known for their ability to transmit pathogens, including various arthropod-borne viruses (arboviruses) that are of veterinary and medical interest. The threat of (re-)emerging arboviruses in Europe is increasing due to globalization and climate warming. This also applies to temperate regions, where the transmission of viruses is becoming possible due to an increase of the ambient temperature, i.e. shortening of the extrinsic incubation period. *Culex pipiens s.s./Culex torrentium* are the primary vectors of Usutu virus and West Nile virus in Europe and are commonly found in and around human settlements. The prediction of spatial-temporal abundance allows for the early assessment of arbovirus transmission risk and the organization of intervention methods, such as vector control. A mechanistic model was developed to predict the spatial-temporal occurrence of *Cx. pipiens s.s./Cx. torrentium* in Germany. The model output is driven by local rainfall and temperature data provided by the European Climate Assessment & Dataset. Evaluated with field data from 87 sampling sites, the mechanistic model is capable of pinpointing the peak in abundance. The mean offset of the simulated and observed peak is -6.59 days (CI: -14.26-1.09) and does not significantly differ from zero ($t_{(110)} = -1.7$, $p=0.09$). One third of the sampling sites showed a significant positive Pearson correlation between simulated and observed mosquito abundance and there is an overall significant linear relationship ($R^2= 0.25$, $F_{(1, 4794)}=1622$, $p<0.001$). The model is also capable of predicting population dynamics for entire Europe, as tested for Moldavia, Ireland, Sweden and Portugal. This mechanistic model can be used to simulate vector control measurements or the impact of increasing temperatures in cause of climate warming. Most importantly, it can be used to implement a near real-time risk assessment tool for emerging mosquito borne diseases.



S7-P38

Household Insecticide Use and Flea Resistance in Madagascar: Implications for Public Health

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In Madagascar where bubonic plague, a flea-borne disease, is endemic, there is a gap in knowledge regarding household pest control that may impact health and insecticide resistance in vectors. In this study, we investigated 254 households distributed in seven villages within three districts in plague-endemic areas of Madagascar to understand the pattern of insecticide use in homes and to assess rat flea (*Xenopsylla cheopis*) susceptibility to insecticides. Here we present the results from household surveys and insecticide bioassays. We found that insecticide was the most common means for domestic pest control for 91.66% of respondents, primarily targeting the house flea (80.11%). Most applied liquid formulations (78.41%), and 62.50% used insecticides without dilution. Bedrooms (90.90%) and floors (89.78%) were primarily treated. Only 46.02% of respondents were satisfied with the treatment results. Products were bought from outdoor market vendors (38.64%) or agricultural stores (31.25%). Retailer advice (55.68%) and neighbors' opinions (21.02%) guided insecticide selection. Retailers provided most (84.65%) information on insecticide usage. Yet, 39.54% of respondents could not recall the commercial name of the insecticide used, given that only 26.13% bought labeled products. Laboratory insecticide bioassay revealed widespread resistance to DDT (mortality rate, MR: 0 - 7.50%) and deltamethrin (MR: 0 - 42.50%) among rat fleas from these villages. Flea populations were tolerant (MR: 82.8 - 95.10%) and susceptible (MR: 97.60 - 100%) to fenitrothion, with only fleas from one village showing resistance (MR: 45%). The significant reliance on insecticide to control household pests and limited product knowledge pose a risk for health and may contribute to the development of resistance in non-target insects such as the plague vector, *X. cheopis*. Our findings suggest that interventions focused on retailers (i.e., education, incentives) would equip households with accurate pesticide knowledge. Addressing these challenges is imperative for safeguarding public health and mitigating insecticide resistance.

National Vector Surveillance Programme for Ticks in Austria

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Introduction and Objectives

Ticks are vectors for multiple pathogens causing disease in humans and animals. *Ixodes ricinus* is the most common tick species in Austria. Within this tick species bacteria of the *Borrelia burgdorferi* sensu lato complex are the most frequently found pathogens. These spirochetes are the causative agent of Lyme Borreliosis which is the most common vector-borne disease in Central Europe.

In the last few years autochthonous tick species such as *Hyalomma marginatum* are occasionally found which can transmit Crimean-Congo-Haemorrhagic-Fever virus (CCHFV), leading to serious disease with a high fatality rate. Those ticks are most likely introduced by migratory birds and can persist when weather conditions are suitable, hence, posing a high health risk to humans.

The aim of this study is to monitor the tick vector landscape of Austria. Moreover, selected ticks will be analysed for the presence of *Borrelia* and CCHFV. The surveillance project launched in January 2024 and will continue for the next few years.

Materials and Methods

Ticks are collected by a citizen science approach and additional flagging events. Morphological identification of ticks is followed by molecular screening by real-time PCR for *B. burgdorferi* sensu lato for endemic hard tick species. In the case of *H. marginatum* ticks the screening is focused on detection of CCHFV.

(Preliminary) Results

So far, the following tick species were identified ($n = 97$): *I. ricinus* complex (88.7%), *Ixodes* spp. (7.2% - not further identifiable), *D. reticulatus* (2.1%), *D. marginatus* (1.0%) and *A. reflexus* (1.0%).

Discussion and Conclusion

Monitoring the vector landscape by a nation-wide surveillance programme can aid as early response system when it comes to new exotic diseases. Moreover, knowledge about endemic and imported tick vectors including their pathogens is of high epidemiological importance.

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Sand flies surveillance in Emilia-Romagna, Italy, during 2022-2023

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Phlebotomine sandflies (genus *Phlebotomus*) are insects that contribute to the burden of arthropod borne diseases. Mainly known as *Leishmania* vectors, they harbour several pathogens important in human and animal health, particularly Phleboviruses. Their ideal habitat consist of sheltered moist areas in woods, caves or animal pens, in rich organic matter soils. At our latitudes, their distribution is located in hilly zones covered in vegetation, seldom found in open plain areas.

During the hot – June to September - seasons 2022-2023, a phlebotomine sandflies surveillance was performed using CDC light and CO₂ traps through the Emilia-Romagna territory (northern Italy). Traps were placed in hilly areas as well as in lowland, taking advantage of the ongoing WNV regional surveillance plan. Traps work usually for one night every two weeks from 5:00 pm to 9:00 am, caught individuals were kept refrigerated and transported to the laboratory for the analyses. Female sandflies were pooled in 50 individuals' 1,5 ml vials and processed for pathogen screening while the males were identified at species level using morphological characters.

During the period considered we captured almost 99,000 sandflies, 44,496 in 2022 and 54,390 in 2023. Females' pathogens screening investigated the presence of Phleboviruses, including Toscana and Fermo viruses and *Leishmania* parasite. We analyzed 1,711 pools in total. Pools tested positive mainly to Fermo virus -317- and *Leishmania* parasite -214-, together with 74 positive Toscana virus pools; further, different pools tested positive to more than one pathogen. We also identified other less investigated Phleboviruses, like Ponticelli, Corfou and Punique viruses. The sandflies sampled belong mostly to *Phlebotomus perfiliewi* and to a lesser extent to *Phlebotomus perniciosus* species. The insects were mainly found in areas known as Leishmaniasis foci. Interestingly, though, several individuals come from captures in plain land, an environment considered not particularly suitable for sandflies.



S7-P41

Invasion history and genetic population structure of *Aedes koreicus* mosquitoes in Italy

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Aedes koreicus is an invasive mosquito species, native to Far East of Asia and recorded in Europe since 2008. Its invasiveness success is mostly evident in Italy, where populations of *Ae. koreicus* are not confined to small areas near the site of first introduction, as has instead been described for other European countries. Despite the increasing number of reports, possible entry points and dispersion pathways have been poorly investigated. In the present study, population genetic analyses were carried out to assess the genetic structure of populations of *Ae. koreicus*, with a focus on Italian populations. The understanding of the genetic relatedness among individuals from different locations is fundamental to draw hypothesis on invasion patterns and dispersal routes of this mosquito in Northern-Italy, and possibly in other European countries.

Ten microsatellite markers were developed and used for the study of populations of *Ae. koreicus* sampled in different localities in the pre-alpine area of Italy, in the neighbouring Slovenia, and in the native area (South Korea). Basic and Bayesian population genetic analyses were performed to evaluate patterns of genetic variation, genetic structure, and demography of mosquito populations.

Intra- and inter-genetic variation among examined individuals reveals a structuring of the populations of *Ae. koreicus* from Italy and Slovenia. However, variation among individual mosquitoes is limited, even between different populations. On the contrary, the population of *Ae. koreicus* from South Korea presents a clear differentiation from the examined European populations. These results support the expansion of a single population propagule of *Ae. koreicus* in Italy and Slovenia, even though the occurrence of multiple introductions cannot be excluded and will require the study of further populations. These data, associated with continuous monitoring, would in prospect permit the discovery of geographic origins and entrance pathways of invasive populations of *Ae. koreicus*, and then prevent/limit further introductions.



Assessing the Short-Term Impacts of Mosquito Control on Vector Populations in Attica Region, Greece, Using a Distributed Lag Nonlinear Model

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Introduction and Objectives

With climate change expanding the range and season of vector-borne disease (VBD) transmission, there is a growing need to invest in mosquito control programs. In Attica Region, Greece, there is an integrated vector management (IVM) program which could provide a model for other regions with less experience in VBD prevention and control. The objective of this study is to evaluate the effect of the Attica IVM program under real-world conditions.

Materials and Methods

We obtained mosquito collection and intervention occurrence data from 2021-2023 at 56 trap locations throughout Attica. We conducted a time series analysis using a distributed lag nonlinear model (DLNM) to assess the short-term lagged effect of mosquito control interventions on the one-week rate of change in *Culex pipiens* counts, the primary vector for West Nile virus. This was fitted in a spatio-temporal generalized additive model (GAM) following a zero-inflated Poisson distribution. Meteorological covariates were considered through forward selection using the AIC as the selection criterion.

Results, Discussion and Conclusion

After adjusting for seasonality and the one-week change in both precipitation and mean temperature, intervention was associated with an immediate decline in *Cx. pipiens* counts in the concurrent week compared to sites without an intervention. One-week lag was not associated with significant change in counts, but two- and three-week lags were both associated with a significant decline in *Cx. pipiens* counts compared to no intervention. The effect had receded by the fourth week post-intervention. The significance of the concurrent week lag indicates that adult mosquitoes were immediately affected while the significance of the two- and three-week lags suggests an interruption to the breeding cycle, although we were unable to dissociate the specific intervention target due to small group sizes. Further analyses will focus on the invasive *Aedes albopictus* to investigate whether the same patterns hold.

Human-biting ticks and associated pathogens: a 5-year report (2019-2023) in Piedmont, Northwest Italy

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The escalating incidence of tick bites in Northwest Italy raises significant concerns due to the role of these arthropods in transmitting Tick-Borne Diseases (TBDs). The emergence of TBDs poses a threat to temperate regions, driven by factors like increased global travel and climate change. Surveillance is crucial to monitor the introduction of new invasive tick species and Tick-Borne Pathogens (TBPs) and to ensure public health. Here, we report the prevalence of tick bites and TBPs in humans in Piedmont, northwest Italy, during the period 2019–2023.

Ticks collected from bitten residents from urban and rural areas were identified at the species level, with a focus on at-risk populations such as children under 14, adults over 65, and symptomatic or frail patients. Biomolecular analyses were employed to detect pathogens including *Anaplasma* spp., *Borrelia* spp., *Rickettsia* spp., and TBE virus, with positive samples confirmed through sequencing.

Over the study period, 1176 ticks were collected and stored for analysis, with varying numbers collected each year. Most ticks belonged to the *Ixodes* genus, predominantly *Ixodes ricinus*. Notably, in 2023, a non-native *Amblyomma* tick was identified in a patient who had recently traveled to Kenya.

Ticks collected from at-risk individuals were tested for various pathogens, revealing an overall infection prevalence of 19.85%. *Rickettsia* spp. was found in 5/387 ticks (1,29%); *Borrelia* spp. in 12/386 samples (3,11%) and *Anaplasma* spp. in 65/386 samples (16,84%). 36 samples were tested for TBE virus infection, but no cases were detected.

Our findings align with prior research in the Piedmont region and emphasize the importance of surveillance in detecting the introduction of new tick species and TBPs. Continued monitoring is crucial for promptly addressing potential public health threats posed by TBDs.

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S7-P44

Irish biting midges: an update

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Introduction

Biting midges, particularly *Culicoides* species, can act as vectors for a range of animal pathogens. While some midge-borne pathogens such as Schmallenberg virus (SBV) are endemic in Ireland, others such as Bluetongue virus (BTV) or Epizootic Haemorrhagic Disease virus (EHDV) are absent, although they have been reported from continental Europe. Having a good knowledge of *Culicoides* species in the country, as well as their seasonality, is key to monitoring and controlling possible future outbreaks of midge-borne pathogens.

Objectives

- Update *Culicoides*' Irish species list using morphological and molecular techniques.
- Update the geographical and temporal distribution of *Culicoides* species in Ireland.

Materials and Methods

Culicoides midges were trapped in 13 sites across the country under the NetVec Ireland project between May 2022 and December 2023. Professionals and members of the public were involved in the collection. Specimens were manually separated from the bycatch and morphologically identified based on the wing patterns. PCR amplification of the COI locus followed by Sanger sequencing was used to confirm identification in a subset of specimens.

Results and Discussion

Around 190,000 *Culicoides* were collected in total and 18,368 specimens were morphologically identified. In addition to the *Obsoletus* complex (members of which cannot be morphologically differentiated through wing patterns) 10 species were recorded including species with vector capacity (i.e the *Obsoletus* complex and *Pulicaris* complex) made up 85% of the *Culicoides* captured. By-catch varied from site to site, but the biting midge proportion was small in all cases. Midges were collected throughout the year, including the official 'midge-free' period (November to March).

First detection of West Nile Virus RNA in field-collected mosquitoes in Croatia

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Introduction and Objectives

West Nile virus (WNV) infections have increased recently and WNV has become one of the most widespread arboviruses in the world, threatening both human and animal health. The ability to appropriately manage this threat is dependent on early detection. Outbreaks of West Nile fever have been reported in Italy, Greece, France, Romania, Hungary, Serbia, and Spain, with further spread expected. Most outbreaks in Western Europe have been due to infection with WNV lineage 1. In Eastern Europe, WNV lineage 2 has been responsible for human and bird mortality. The objective of this study was to determine prevalence of WNV infection in potential mosquito vectors and detect the lineage.

Materials and Methods

Mosquito traps (CDC dry ice baited traps and BG sentinel traps) were placed in rural and urban sites, collected specimens were morphologically characterized and pooled by date, location, and species. Mosquito pools containing *Culex pipiens* and *Aedes albopictus* were tested by conventional PCR for Flaviviruses in a heminested system. The Flavivirus-positive samples were further tested with a specific real-time PCR designed to detect the genetic lineage 1 and 2 of WNV.

Results, Discussion and Conclusion

A total of 226 mosquito pools were tested. The first positive pool was detected in a sample from Osijek-baranja county containing *Cx. pipiens* mosquitoes. Due to the low amount of virus RNA in this case we could not detect the lineage. The second positive pool is from Vukovar-srijem county, also containing *Cx. pipiens*. From this pool, sequencing of amplicons revealed WNV lineage 2 partial NS5 gene sequences. These results present the first detection of WNV RNA in mosquitoes in Croatia and confirm that human cases in this region are most likely infections transmitted by the local *Culex* mosquitoes infected with lineage 2 of WNV.

S7-P46

Decision making for operation control action based on surveillance data for justification at Anastasia Mosquito Control District, St. Augustine, FloridaRui-De XUE^{1,2} (xueamcd@gmail.com)¹ Anastasia Mosquito Control District, St Augustine, FL, USA ; ² University of Miami Miller School of Medicine, Miami, Florida, USA

Anastasia Mosquito Control District (AMCD), St. Augustine, Florida, USA established since 1948. AMCD is a special independent district and taxes residents for mosquito control only. Currently, AMCD provides the service to more than 340,000 residents and cover 609 square miles. There are five programs under three major catalogues that guide AMCD daily activity, such as operation (customer and professional service, surveillance, operation control), research, and education. Service requests and answering services are based on the strong surveillance and operation control programs. Annually AMCD received/answered about 5,000 service requests. The services include regular mosquito-borne diseases (especially arbovirus) through the collaboration with Department of Health, animal diseases through Department of Agriculture, sentinel chickens and mosquito pools and mosquito population surveillance in house. The operation control actions include ground and aerial applications for larviciding and adulticiding. The pesticide applications are one of major methods/tools for control of the disease vectors and nuisance mosquitoes. The decision making thresholds for operation control action by using pesticides have been based on the justification of surveillance data. They are: any human (travel related and local-acquired) and animal cases, positive sentinel chickens and mosquito pools, high number (> 20) of service requests from a specific/hot area (about 5 square miles), hurricane damaged and flooded area, positive human landing rate counts, more than 25 adult mosquitoes caught by a CDC light trap/night, and a special event/activity planned... For ground application, AMCD uses different larvicides for control of larval mosquitoes, thermal fogging and barrier spraying for adult mosquito control in vegetated areas at daytime, and ULV spraying by trucks and helicopter for adult mosquito control at night. After many year experiences and practices at the AMCD, St. Augustine, Florida, the application of the method/strategy showed successful prevention and control of the potential outbreaks of mosquito-borne diseases in the area.

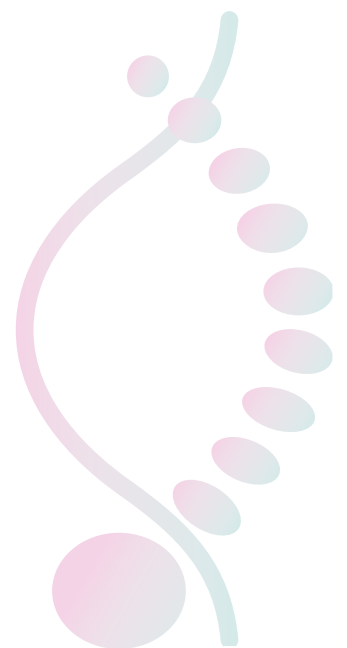


ONE HEALTH IN ACTION

14-17th October 2024
MONTPELLIER, FRANCE



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