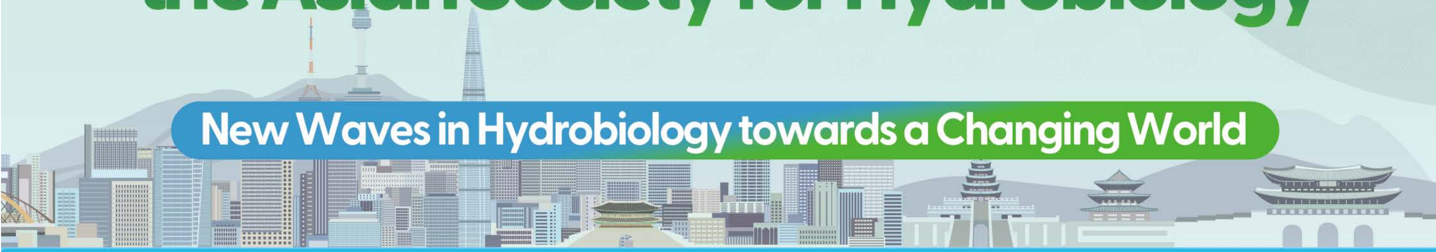
**PROGRAM & ABSTRACT BOOK**

# The 6th Biannual Conference of the Asian Society for Hydrobiology



## New Waves in Hydrobiology towards a Changing World

**July 1 (Mon) - 4 (Thu), 2024**
**Hana Square, Korea University, Seoul**

Hosted by

ASH CONFERENCE 2024
ASH Asian Society for Hydrobiology

Organized by

KOREAN ENTOMOLOGICAL INSTITUTE 1963 KOREA UNIVERSITY

OJERI @ KU Korea University OJEong Resilience Institute

KOREA UNIVERSITY BK21 FOUR R&E Center for Environmental Science and Ecological Engineering

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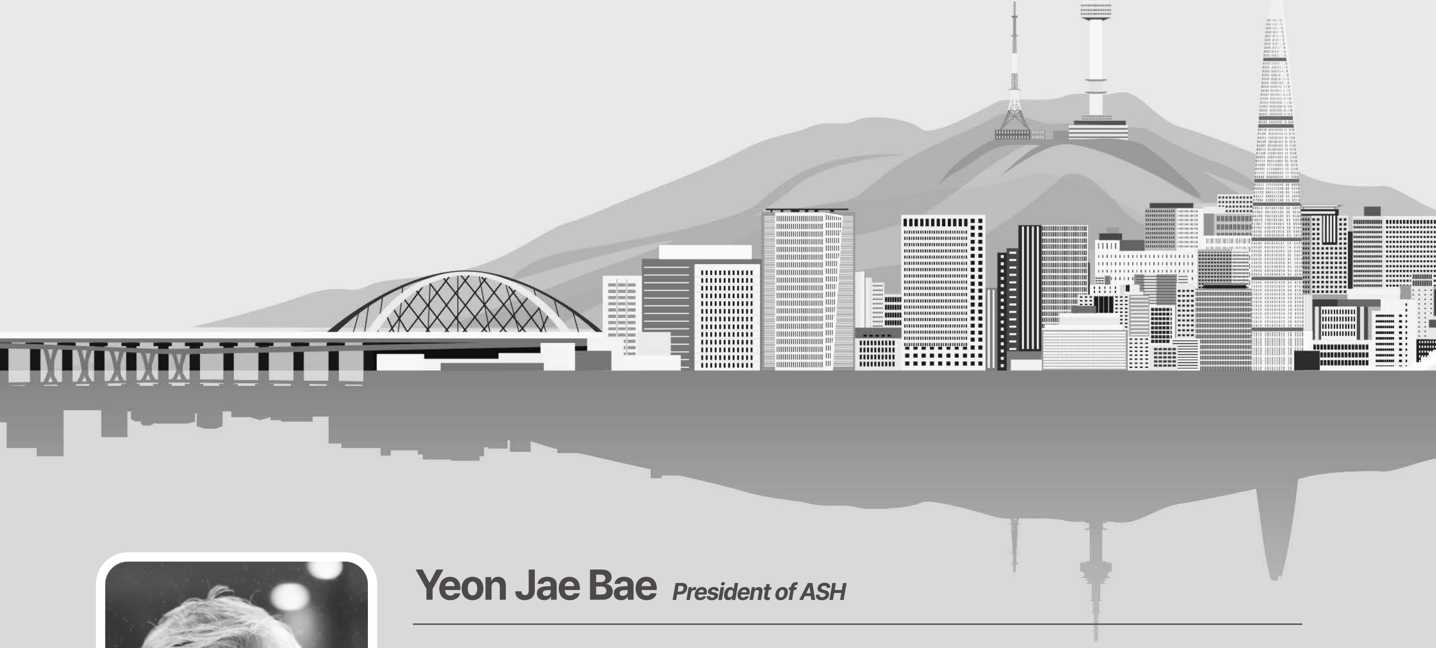

## Yeon Jae Bae *President of ASH*

ASH members, colleagues, friends, distinguished guests, and my dear students,

I am very pleased to meet you again at the 6th Biannual Conference of the Asian Society for Hydrobiology (ASH) held here at Korea University in Seoul, following the conference in Chiang Mai two years ago. On behalf of myself and all the members of the Local Organizing Committee who prepared this event, I would like to welcome presenters, invited speakers, and all other participants.

As you know, our society was founded 25 years ago to address environmental issues in aquatic ecosystems faced by Asian countries and to promote the advancement of aquatic biology. This meeting started voluntarily, and over the years, the fields and organization of the society have rapidly grown, evolving into an international society with over 500 members from 21 countries.

As we are all aware, we are facing a changing world. The rate of environmental change, including that of aquatic ecosystems in the Asian region, is progressing even faster. Advanced technologies such as big data analysis using computers, environmental DNA analysis, and artificial intelligence technologies are emerging as solutions to future environment issues in aquatic ecosystems. Against this backdrop, we have set the theme of this conference as "New Waves in Hydrobiology towards a Changing World."

During the four days of this conference, I hope that all participants will build friendships, engage in active academic exchanges, and achieve fruitful academic outcomes while having a great experience in Seoul.

With the full support of President Dong-One Kim of Korea University, together with the co-host of the conference, Director Woo-Kyun Lee of the OJEong Resilience Institute of the College of Life Sciences, and Director Jinho Jung of the BK21 FOUR R&E Center for Environmental Science and Ecological Engineering, I am honored to declare the opening of the ASH 2024 Conference!

Thank you.

## Dong-One Kim *President of Korea University*

ASH members, colleagues, guests, and students,

Welcome to the 6th Conference of the Asian Society for Hydrobiology (ASH) at Korea University in Seoul. Our theme is "New Waves in Hydrobiology towards a Changing World."

As you all know, environmental changes are threatening water resources. Asian countries, which are growing rapidly, face severe water pollution and depletion. ASH, the only academic group covering all of Asia, has advanced hydrobiology here for 25 years to confront these issues.

As your host, I am proud to present Korea University, a top research university known for excellence and innovation. Since 1905, we have been fostering critical thinking and research, constantly addressing global challenges with our state-of-the-art expertise and facilities.

Korea University actively engages in partnerships and exchanges with institutions worldwide, promoting global engagement and understanding. Thus, this conference is not only an academic exchange; it is also a venue for building lasting relationships. Together, our diverse perspectives will lead to innovative solutions and partnerships.

I would like to extend my heartfelt gratitude to the organizing committee, our sponsors, plenary speakers, and all participants for their contributions to making this conference possible.

Once again, welcome to ASH 2024 and to Korea University. May this conference be a fruitful and inspiring experience for all.

Thank you.

## Min Hwan Suh *President of the National Institute of Biological Resources*

Dear ASH members and distinguished guests,

Since its foundation in 1998, the Asian Society for Hydrobiology (also called President of ASH) has contributed significantly to the conservation of biodiversity in aquatic ecosystems and the academic advancement of hydrobiology. As such, it is a great pleasure and honor to meet Korea University President Dong-One Kim, ASH President Yeon Jae Bae and the Society's members today.

In modern times, many aquatic ecosystems are rapidly disappearing due to urbanization, the expansion of tourist sites, and development projects. Thus, organisms that inhabit these areas face significant threats. Additionally, climate change has led to frequent outbreaks of mass emergence, prompting global efforts to analyze the causes and devise countermeasures.

I strongly believe that the continuous exchange amongst hydrobiologists from numerous Asian countries, including Korea, Japan, China, Russia, Thailand, India, Mongolia, Vietnam, Nepal, and the Philippines, for the conservation of these aquatic ecosystems has great significance.

I hope that this year's ASH Conference will facilitate many beneficial discussions for the conservation of biodiversity in aquatic ecosystems. I also look forward to the Society's further development, enabling it to play a central role in hydrobiology beyond Asia.

In the future, the Ministry of Environment and the National Institute of Biological Resources will continue to maintain sustainable networks with the Asian Society of Hydrobiology and its researchers. We will strive to integrate progressive research outcomes and proposals into practical applications for the conservation and sustainable management of hydrobiology.

Thank you.

## John Morse

***Advisory Committee Chair, Professor Emeritus of Clemson University***

Dear friends, colleagues, and young hydrobiological scholars, as Chair of the Advisory Committee for the Asian Society for Hydrobiology, it is my pleasure to welcome you to this 6th Biannual Conference of the Society. Although I cannot be with you physically for this year's Conference, please be confident that I do join you in spirit and in enthusiastic commitment to our common purpose of helping to increase understanding of hydrobiological structure and function and to improve our ability to manage modern challenges against them. The theme for this year's Conference, "New Waves in Hydrobiology towards a Changing World," invites us all to focus on this common purpose. This invitation is extended regardless of whether our research pertains to chemistry, physics, biology, or combinations of these; whether our biological investigations emphasize behavior, distribution, habitats, biological interactions, functional traits, or taxonomic diversity; or whether our studies answer basic questions for any of these issues or help solve applied concerns for human health and economic prosperity.

I am especially glad to see how our discipline and the Asian Society for Hydrobiology has grown in scope and integrated the work of additional disciplines since its origins in the study of freshwater insects. As much as I have enjoyed investigating and teaching about aquatic insect diversity in many Asian countries and participating in the organization of several of ASH's founding aquatic entomological societies since the 1980s, the improved opportunity to network with colleagues in related fields and to stimulate enthusiasm for hydrobiological science among a new generation of young scientists provides confidence that the future of our science is bright.

I envy your special opportunities this week to make new friends, capture new ideas, form new research and teaching partnerships, plan new collaborative research projects, consider new possibilities for use of Artificial Intelligence and other developing technologies, and map a new vision for hydrobiological science in Asia and globally into the near and distant future.

Best wishes to you all, and especially to you younger participants for a most fruitful and stimulating experience through the rest of the meeting. As always, please feel free to call on me if I can help in any way to help answer your hydrobiological questions and advance your promising careers. I and your other mentors are proud of you and delighted to watch your emerging and blossoming success. Good luck!

# Program

| Date | 7.1 (Mon) | 7.2 (Tue) | | 7.3 (Wed) | | | 7.4 (Thu) | |
|---|---|---|---|---|---|---|---|---|
| Room | East Building, College of Life Sciences | Hana Square | | Hana Square | | | Hana Square | |
| Time | Room 109 | Auditorium (B112) | Multimedia Room (B115) | Auditorium (B112) | Multimedia Room (B115) | Seminar Room (B120) | Auditorium (B112) | Multimedia Room (B115) |
| 8:00~8:30 | | Conference Registration | | | | | | |
| 8:30~9:00 | | | | | | | | |
| 9:00~9:30 | Workshop Registration | ASH Opening Ceremony | | Session 2-1 | Session 5-1 | | Session 4-2 | Session 5-3 |
| 9:30~10:00 | Workshop Opening (Workshop Participants only) | Coffee Break | | | | | | |
| 10:00~10:30 | Students' & Researchers' Workshop (Image Analysis using A.I.) | Plenary Session 1: Dr. Javier Alba-Tercedor (Room B112) | | ASH Special Session | Symposium 3 (Dongsung Biopharm and Sahmyook University) | | | |
| 10:30~11:00 | | | | | | | | |
| 11:00~11:30 | | Plenary Session 2: Dr. Sangchul Lee (Room B112) | | | | | General Meeting & Closing Ceremony (B112) | |
| 11:30~12:00 | | | | | | | | |
| 12:00~12:30 | Lunch | Lunch | | | | | | |
| 12:30~13:00 | | | | | | | Excursion & Field Trip<br>(Excursion: Insadong, Hangang Park Sebit Island)<br>(Field Trip: Yoomyeong Mountain) | |
| 13:00~13:30 | Students' & Researchers' Workshop (Image Analysis using A.I.) | | | | | | | |
| 13:30~14:00 | | Session 1 & Session 3 | Symposium 1 (NIBR) | Session 2-2 | Symposium 4 (KDCA) | | | |
| 14:00~14:30 | | | | | | | | |
| 14:30~15:00 | | | | | | | | |
| 15:00~15:30 | | | | | | | | |
| 15:30~16:00 | | Session 4-1 | Symposium 2 (NNIBR) | Session 5-2 | Symposium 5 (OJERI) | Session 6 | | |
| 16:00~16:30 | | | | | | | | |
| 16:30~17:00 | | | | | | | | |
| 17:00~17:30 | Executive Board Meeting (Room 222) | | | | | | | |
| 17:30~18:00 | | Banquet (Samcheonggak) | | Poster Presentation (Exhibition Hall) | | | | |
| 18:00~18:30 | | | | | | | | |
| 18:30~19:00 | | | | Small Group Meeting / Dinner for ASH Honorary Presidents (Sudang Samyang Faculty House) | | | | |
| 19:00~21:00 | Executive Dinner | | | | | | | |

## Session

| | |
|---|---|
| Session 1 | New Technology in Hydrobiology: Insights & Challenges |
| Session 2 | Taxonomy, Molecular Phylogeny and Evolution |
| Session 3 | Behavior / Structural and Functional Ecology |
| Session 4 | Aquatic Biodiversity and Conservation / Climate Change |
| Session 5 | Biomonitoring and Bioassessment of Aquatic Ecosystems |
| Session 6 | Hydrobiology for Human Health / Vector Ecology |

# Contents

## I. Plenary Session

**7.2 (Tue)** 10:00-12:00 **Auditorium (B112)** **Chair :** Hyuk Je Lee (Sangji University)

## II. ASH Special Session: Future Outlook of Asian Benthology

**7.3 (Wed)** 10:00-11:50 **Auditorium (B112)** **Chair :** Beixin Wang (Nanjing Agricultural University)
**Co-chair :** Chitchol Phalaraksh (Chiang Mai University)

10:00-10:10 Welcome Remarks
Yeon Jae BAE

# III. Main Session

## Session 1 New Technology in Hydrobiology: Insights & Challenges

| **7.2 (Tue)**<br>13:30-14:45 | **Session 1**<br>**Auditorium (B112)** | **Chair :** Koji Tojo (Shinshu University) |
|---|---|---|
| **1-1**<br>13:30-13:45 | Applications of automated image recognition for marine megafauna<br>Peng ZHOU, Zhinuo ZHOU, Chenghan YANG, Chun-Sheng WANG, Hongbin SHEN, and Xiaoyong PAN | p12 |
| **1-2**<br>13:45-14:00 | Relationship between benthic macroinvertebrate communities and hydromorphology using unmanned aerial vehicle<br>Sung Hwan PARK and Yeon Jae BAE | p13 |
| **1-3**<br>14:00-14:15 | Climate-driven difference in gut microbiome community of dung beetles (Coleoptera: Scarabaeidae: Scarabaeinae, Aphodiinae) from different climate zones in Mongolia<br>Junshik BAE, Changseob LIM, Duleepa PATHIRAJA, Byeong-Hyeok PARK, Bo-Gun KIM, Jung-Joon PARK, and In-Geol CHOI | p14 |
| **1-4**<br>14:15-14:30 | Enhancing biological water monitoring through supervised machine learning: A taxonomy-free approach using macroinvertebrate eDNA metabarcoding<br>Sheng TIANJIN, Ning YESHUANG, Xu XIAOHANG, and Beixin WANG | p15 |
| **1-5**<br>14:30-14:45 | The eDNA methods outperforming the traditional method in capturing the diversity of stream benthic macroinvertebrates<br>Jinxian CHEN, Haoming ZANG, Libo HAN, Zhen DENG, and Beixin WANG | p16 |

## Session 2 Taxonomy, Molecular Phylogeny and Evolution

| **7.3 (Wed)**<br>09:00-10:00 | **Session 2-1**<br>**Auditorium (B112)** | **Chair :** Vinh Van Nguyen (VNU University of Science) |
|---|---|---|
| **2-1-1**<br>09:00-09:15 | Environmental DNA study by a newly developed DNA barcoding region (with the mtDNA 16s rRNA region): Issues with the general DNA barcoding region<br>Masaki TAKENAKA, Yuta HASEBE, Koki YANO, Seiya OKAMOTO, and Koji TOJO | p17 |
| **2-1-2**<br>09:15-09:30 | The exact morphology of a new record mayfly *Thraulus ishiwatai* Grant, 2024 from China (Ephemeroptera: Leptophlebiidae)<br>De-Wen GONG and Chang-Fa ZHOU | p18 |
| **2-1-3**<br>09:30-09:45 | Description of two new Baetidae species in Korea (Ephemeroptera)<br>Wooseong KIM and Yeon Jae BAE | p19 |
| **2-1-4**<br>09:45-10:00 | Taxonomic revision of the *Drunella ishiyamana* species group (Ephemeroptera: Ephemerellidae) in the East Palaearctic<br>Sang Kyu SHIN, Changseob LIM, Junyoung LEE, HakHyun KIM, and Yeon Jae BAE | p20 |

**7.3 (Wed)** 13:30-15:30 **Session 2-2** **Auditorium (B112)**
**Chair :** Joong-Ki Park (Ewha Womans University)
**Co-chair :** Yong-Jin Won (Ewha Womans University)

## Session 3 Behavior / Structural and Functional Ecology

**7.2 (Tue)** 14:45-15:30 **Session 3** **Auditorium (B112)**
**Chair :** Koji Tojo (Shinshu University)

## Session 4 Aquatic Biodiversity and Conservation / Climate Change

**7.2 (Tue)** 15:30-17:30 **Session 4-1** **Auditorium (B112)** **Chair :** Boonsatien Boonsoong (Kasetsart University)

**7.4 (Thu)** 09:00-11:00 **Session 4-2** **Auditorium (B112)** **Chair :** Changfa Zhou (Nanjing Normal University)

## Session 5 Biomonitoring and Bioassessment of Aquatic Ecosystems

**7.3 (Wed)** 09:00-09:45 **Session 5-1**
**Multimedia Room (B115)**
**Chair :** Christine Jewel C. Uy-Yabut (University of the Philippines Diliman)

**7.3 (Wed)** 15:30-17:45 **Session 5-2**
**Auditorium (B112)**
**Chair :** Hyuk Je Lee (Sangji University)
**Co-chair :** Changseob Lim (Korea University)

**7.4 (Thu)** 09:00-11:00 **Session 5-3**
**Multimedia Room (B115)**
**Chair :** Narangarvuu Dashdondog (National University of Mongolia)

## Session 6 Hydrobiology for Human Health / Vector Ecology

**7.3 (Wed)** **Session 6** **Chair :** Chitchol Phalaraksh (Chiang Mai University)
15:30-17:30 **Seminar Room (B120)** **Co-chair :** Nattawut Sareein (Chiang Mai University)

# IV. Symposium

## Symposium 1 (NIBR) Current status and management methods of outbreak organisms

| **7.2 (Tue)**<br>13:30-15:20 | **Multimedia Room**<br>**(B115)** | **Principle Organizer :** Sun-Jae Park (National Institute of Biological Resources)<br>**Co-organizer :** Dong Gun Kim (Sahmyook University) |
|---|---|---|

|  |  |  |
|---|---|---|
| **S1-1**<br>13:30-14:00 | **Speaker 1 - Keynote** Synchronized mass emergence and parthenogenesis of the polymitarcyid mayfly, *Ephoron shigae*<br>Kazuki SEKINE | **p76** |
| **S1-2**<br>14:00-14:20 | **Speaker 2** Lovebug outbreaks: Finding patterns of dispersal and adaptation<br>Jonghwan CHOI, Dahyun BYEON, Choongwon JEONG, and Seunggwan SHIN | **p77** |
| **S1-3**<br>14:20-14:40 | **Speaker 3** Research on factors to hypertrephied of *Metcalfa pruinose* (Hemiptera: Flatidae)<br>Sangjin KIM, Juhyeong SOHN, Yeonghyeok YU, Yeongmo KIM, Hyeban NAMGUNG, Seokho SO, Munhwan YU, and Hyojoong KIM | **p78** |
| **S1-4**<br>14:40-15:00 | **Speaker 4** Outbreak of walking-stick insects, *Ramulus mikado* (Rehn & J.A.G.) (Insecta: Phasmida), in temperate forests in relation with environmental variables<br>Jong-Kook JUNG, Seunghwan HAN, Sora KIM, In-Won JEONG, Min Jeong BAEK, and Sun-Jae PARK | **p79** |
| **S1-5**<br>15:00-15:20 | **Speaker 5** Habitat preference of *Ephemera orientalis* (Ephemeroptera: Ephemeridae) larvae in Hangang River<br>Dong Gun KIM, Hyun LEE, Tae Joong YOON, and Yeon Jae BAE | **p80** |

## Symposium 2 (NNIBR) Biological collections in the Digital Age: Challenges and opportunities

| 7.2 (Tue)<br>15:30-17:20 | Multimedia Room (B115) | **Principle Organizer :** Kyong In Suh (Nakdonggang National Institute of Biological Resources)<br>**Co-organizer :** Seung Hyun Lee (Nakdonggang National Institute of Biological Resources) |
|---|---|---|

## Symposium 3 (Dongsung Biopharm and Sahmyook University)
## Safe and healthy water resource acquisition and management strategies

| **7.3 (Wed)**<br>10:00-11:50 | **Multimedia Room**<br>**(B115)** | **Principle Organizer :** Dong Gun Kim (Sahmyook University)<br>**Co-organizer :** Pumis Thuptimdang (Chiang Mai University) |
|---|---|---|
| **S3-1**<br>10:00-10:30 | **Speaker 1 - Keynote** Applying the latest strategy to manage harmful algal blooms in waterbody<br>Heesuk LEE | **p86** |
| **S3-2**<br>10:30-10:50 | **Speaker 2** Mass production of graphene for water treatment<br>Chang-Ho CHOI, Yonghyun LEE, Si-Hyun PARK, and Chul-Woong CHO | **p87** |
| **S3-3**<br>10:50-11:10 | **Speaker 3** Investigation the stability and reactivity of chlorine dioxide ($ClO_2$) in the water treatment process<br>Kwan-Yong LEE, Se-Ra JIN, Si-Hyeon PARK, Jeong-Min CHEON, and Chul-Woong CHO | **p88** |
| **S3-4**<br>11:10-11:30 | **Speaker 4** Evaluation of chlorine dioxide disinfection ability using chironomids (Diptera: Chironomidae)<br>Jang Ho LEE, Tae Joong YOON, and Dong Gun KIM | **p89** |
| **S3-5**<br>11:30-11:50 | **Speaker 5** Effect of diclofenac on bacterial community in activated sludge from wastewater treatment plant of Chiang Mai University<br>Pumis THUPTIMDANG, Keletso CHAKALISA, and Chadarat SILATHAM | **p90** |

## Symposium 4 (KDCA) Strategies for various disease vector management

**7.3 (Wed)** 13:30-15:20 **Multimedia Room (B115)**

**Principle Organizer :** Hee il Lee (Korea Disease Control and Prevention Agency)
**Co-organizer :** Chitchol Phalaraksh (Chiang Mai University)

**S4-1** 13:30-14:00
**Speaker 1 - Keynote** Implementation of big data to control disease vector mosquitoes p91
Hyung Wook KWON

**S4-2** 14:00-14:20
**Speaker 2** Ecological feature of *Aedes albopictus* (Diptera: Culicidae) in Korea p92
Hyunwoo KIM and Hee Il LEE

**S4-3** 14:20-14:40
**Speaker 3** Predators as biocontrol agents of mosquito larvae in small and large habitats in Chiang Mai, Thailand p93
Panida RAHONG, Chotiwut TECHAKIJVEJ, Yeon Jae BAE, and Chitchol PHALARAKSH

**S4-4** 14:40-15:00
**Speaker 4** Phylogenetic analysis and distribution of Asian predaceous *Toxorhynchites* mosquitoes (Culicidae): Exploring their potential for biological control as a Nature-Based Solution (NbS) p94
Nattawut SAREEIN, Jassada SAINGAMSOOK, Changseob LIM, Ji Hyoun KANG, and Yeon Jae BAE

**S4-5** 15:00-15:20
**Speaker 5** Characterisation of acaracide resistance-conveying mutations in para-sodium channel of dog and cattle ticks, Thailand p95
Jassada SAINGAMSOOK, Celia WARRE, Nattawut SAREEIN, Patchara SRIWICHAI, and Saruda TIWANANTHAGORN

## Symposium 5 (OJERI) Optimizing water, food, energy, and ecosystem nexus in the 3rd pole region of the Mid Latitude Region

| | | |
|---|---|---|
| **7.3 (Wed)**<br>15:30-17:30 | **Multimedia Room (B115)** | **Principle Organizer :** OJEeong Resilience Institute at Korea University (OJERI@KU)<br>**Co-organizer :** Mid Latitude Region Network (MLRN) (Korea University)<br>**Moderator:** Hun Park (OJERI@KU) |

15:30-15:40 **Welcome Remarks**
Woo-Kyun Lee (Director, OJERI)

**S5-1** 15:40-16:00 **Speaker 1 - Keynote** Third pole in crisis: Opportunities, challenges, and climate actions p96
Sonam Wangyel WANG

**S5-2** 16:00-16:20 **Speaker 2** Water-Energy nexus: Turning wastewater into bioenergy p97
Sun-Mi LEE

**S5-3** 16:20-16:40 **Speaker 3** Spatial livelihood vulnerability modeling in Central Asia using various indices p98
Cholho SONG

**S5-4** 16:40-17:00 **Speaker 4** Challenges facing ecosystem resilience in Nigeria – West Africa p99
Sonigitu EKPE

17:00-17:15 **Panel discussion**
**Panelist** Munkhnasan Lamchin and Sang-Chul Lee

17:15-17:25 **General discussion**
**Moderator** Hun Park

17:25-17:30 **Concluding Remarks:** Conclusion and way forward
Woo-Kyun Lee

# V. Poster Session

## Session 2 Taxonomy, Molecular Phylogeny and Evolution

## Session 3 Behavior / Structural and Functional Ecology

## Session 4 Aquatic Biodiversity and Conservation / Climate Change

## Session 5 Biomonitoring and Bioassessment of Aquatic Ecosystems

## Session 6 Hydrobiology for Human Health / Vector Ecology

# VI. BK21 Special Session

## Poster Presentation

# I. Plenary Session

July 2, 2024 | 10:00~12:00

# Computerised microtomography (micro-CT) as a valuable tool to study and discover functionalities of little organisms

**Javier ALBA-TERCEDOR**

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Correspondence e-mail address: jalba@ugr.es

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High resolution computerised microtomography (micro-CT) is not a new technique; it is routinely used in science. Mostly, it is used as a substitute for microscopy to examine external and internal structures. However, it can also be used to clarify non-answered questions and for educational purposes. In many cases, it simply represents itself, Art.

Throughout the presentation, it will be explained in general terms what microtomography is and its differences from scanning electron microscopy. Different methods of preparation prior to fixing the samples for scanning, visualisation of the rendered images and how to proceed to visualise/separate the different structures/organs with different colours, either by their differentiation according to the degree of x-ray opacity (what it is possible by adjusting the colour transfer functions curves into the volumetric software) or by a manual outline segmentation.

The main advantage of micro-CT is that it is a non-destructive technique. However, micro-CT is not a technique that can immediately produce wonderful images. In addition to the fact that the scanning time can be long, software processing time is required to clean up the images and, if necessary, to segment/isolate structures/organs before viewing the image stack with any of the existing software packages.  However, once the whole process has been carried out, the reconstructed image dataset obtained can be stored, and at any time viewed, rotated, sliced, etc…, making it possible to study details of external and internal structures from any possible perspective.

In summary, the presentation will explain microtomography and how it works, presenting examples of different anatomical and functional studies, and presenting details of how the speaker uses a high-resolution Skyscan 1172 microtomograph to unveil “mysteries” beyond the currently known horizon, including examples with aquatic animals. The presentation will be illustrated with high-resolution images and videos to demonstrate how micro-CT makes it possible to take amazing trips inside the specimens.  “Real” models can be obtained to be visualise with mobile devices (smartphones and tablets), permitting rotations, cuts, etc.…, in an easy and interactive mode. Resulting useful for research and very attractive for students, and after that very promising for educational purposes. In any case, many of the obtained results have a nice visual artistic appearance, and attendees will understand why Micro-CT represents a tool straddling Scientist Research, Art and Education.

Keywords: Micro-CT, functional anatomy, art, education

# Predicting the vector mosquito populations in Seoul, Republic of Korea using machine learning models

**Sangchul LEE**[1,P,C] **and Hyemin JEONG**[2]

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Korea University, Seoul, Republic of Korea
[2]Department of Environmental Engineering, Graduate Student, University of Seoul, Seoul, Republic of Korea
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Mosquitoes are a biological species that serve as vectors for various infectious diseases. To suppress the spread of diseases transmitted by these mosquitoes, it is necessary to understand the causative factors. Recently, machine learning models (MLMs) have been increasingly applied across various fields. However, the impacts of landscape factors on vector mosquito populations using MLMs are largely unknown. The objective of this study is to develop MLMs to predict vector mosquito populations in Seoul, Republic of Korea, using landscape and meteorological factors. The data for this study were collected during the mosquito season from May to October each year between 2015 and 2022. Vector mosquito population data were obtained from 50 Digital Mosquito Monitoring Systems (DMS). The meteorological factors—including temperature, precipitation, wind speed, humidity, maximum temperature, minimum temperature, annual average temperature, and monthly average temperature—were derived from the Automatic Weather Stations (AWS) nearest to each of the 50 DMS sites. The landscape factors were determined through an in-situ survey of the major landscape feature types for individual DMS sites. The MLMs used in this study included Random Forest (RF), Gradient Boosting (GB), Light Gradient Boosting (LGB), and Extreme Gradient Boosting (XGB). Performance metrics, R-squared ($R^2$) and Root Mean Squared Error (RMSE), were used. The results showed that the four MLMs exhibited decent performance with an $R^2$ of approximately 0.58 and an RMSE of 18.46, while the LGB model exhibited the greatest performance. The findings from this study demonstrate the potential usefulness of MLMs in predicting vector mosquito populations.

Keywords: mosquitoes, diseases, machine learning models, landscape factors, meteorological factors

Acknowledgements: This study is supported by a Korea University Grant.

# II. ASH Special Session

## : Future Outlook of Asian Benthology

# Future outlook of asian benthology: Increasing demand for genetic marker-based studies

**Koji TOJO**[1,2,C]

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In recent years, the effects of climatic change have begun to become evident in ecosystems. In particular, rising ocean temperatures due to global warming have promoted seawater evaporation, resulting in increased precipitation and causing heavy rainfall in many parts of the world. There has also been an increase in flood damage caused by larger typhoons, hurricanes and other heavy rains. On the other hand, precipitation is becoming more concentrated, the land is becoming drier, resulting in water shortages. Such a major change in precipitation patterns poses a serious challenge to the habitat environments of freshwater organisms. Many areas in Asia are considered to be globally important hotspot regions of biodiversity, home to a rich variety of living organisms and important rare and endemic species. Climatic change is a major threat to the stable maintenance of these biota, and as such, it is important to closely monitor annual changes in each region. In particular, freshwater aquatic organisms are representative of the areas that will be most sensitively affected, with a high number of endangered species relative to the size of their habitat. It is important to steadily advance monitoring of inland water organisms, given the “nature positive” perspective and the fact that the 17 goals of the SDGs are a major source of global interest. In particular, it is of great significance to monitor genetic diversity as one of the indicators in addition to species diversity. Furthermore, improvements in environmental DNA (eDNA) analysis technology, which has been attracting attention recently, are expected to be a game changer that will bring about a paradigm shift in this field of study. In this presentation, I would like to discuss the current status of benthic diversity researches using molecular markers.

Keywords: biodiversity, climatic change, DNA structure, eDNA, genetic diversity, species diversity

# Future challenges in river ecological assessment using benthic macroinvertebrates in China

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Benthic macroinvertebrates have been used to assess river ecological condition worldwide for over 100 years. China began its water quality biomonitoring using macroinvertebrates in the 1960's and experienced significant growth in the 2000's when Chinese authority realized the importance of biomonitoring in water quality management and biodiversity conservation. Subsequently, the biotic index (BI), biological monitoring working party (BMWP), biotic integrity index (IBI) or multi-metric index (MMI) were introduced to China and used to assess water quality. After more than 20 years of development of the biotic indices, China issued modified biological water quality classifications of BI and BMWP in a national technical guideline for water ecological monitoring. However, the sensitive values of most macroinvertebrates used for the calculation of BI and BMWP were defined qualitatively rather than quantitatively. Recently, a new technology, eDNA metabarcoding method is emerged as an alternative to the traditional morphological method in routine biomonitoring programs. Therefore, we suggested that future research should focus on the quantitative definition of sensitive values of macroinvertebrate families, general and species based on the eDNA data, the establishment of regional DNA barcode libraries and standardization of eDNA biomonitoring programs.

Keywords: Biomonitoring, eDNA, Macroinvertebrates, Aquatic insects, River health

# Challenges of benthic research in Vietnam

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Vietnam’s aquatic environments include a range of habitats such as rivers, lakes, reservoirs, wetlands, coral reefs, mangroves, and seagrass beds. In Vietnam, the study and understanding of these organisms have made significant progress. However, the benthic community is currently facing many challenges such as environmental pollution, threatened habitat, climate change, as well as socio-economic development challenges. Currently and in the near future, research on benthic organisms in Vietnam will focus on the following basic topics: 1) Regarding taxonomy, it is necessary to focus research on very diverse but little-known groups of benthic organisms such as polychaetes, oligochaetes, aquatic insects, nematodes, benthic algae... 2) In ecology, the study of benthic animals’ ability to adapt to environmental changes, especially in climate change. 3) Regarding biology, continue to research, breed and cultivate benthic species that play an important role in agriculture, forestry, fishery and medicine. 4) Research on restoration and conservation of rare and endemic benthic species.  5) Another important topic related to benthic organisms is the restoration of ecosystems such as lake ecosystems, rive ecosystem, mangrove ecosystems, coral reef ecosystems in Vietnam.

Keywords: challenges, benthic, taxonomy, ecology, biology, restoration, conservation

# Present and future outlook of hydrobiological research for sustainability in Thailand

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The hydribiological research in Thailand have been dramatically developed. The number of young generation researchers are increasing every year. Regrading geographic diversity, there are various type of ecosystems in Thailand. The Northern high elevation mountainous area, North-eastern Plateau, Western Forest Complex and lowland tropical rainforest are perfect habitats for aquatic invertebrates. The main research themes of Freshwater Biomonitor Research Laboratory (Chiang Mai University) are to study aquatic insect diversity, aquatic macroinvertebrates as bioindicator for water quality monitoring, molecular ecology and the biological mosquito control. In addition, the water resource management (water footprint, water balance) are also our research theme to solve the water-based problems in some area of Thailand. One of the strongest laboratories is from Kasetsart University, Assoc.Prof. Dr. Boonsatien Boonsoong has conducted research projects on rapid bioassessment using freshwater benthic macroinvertebrates, taxonomy, and systematics of aquatic insects, especially mayflies (Ephemeroptera) and Odonata. Moreover, his research interests in post-embryonic development of Thai mayflies. Not only these examples, but there are also many strong research groups have been conducting hydrobiological researches, including from Khon Kaen University, Mahasarakham University, Prince of Songkla University, Chiang Mai Rajabhat University and so on. Our planet is changing. Global temperature is severe rising. The future trend of our research should not only “study to know”, but also “study to solve” the problems. This might be too complicate to study alone. The strong connection between laboratories are absolutely important. Together is always better and is the future outlook of our research.

Keywords: hydrobiology, Thailand, tropical area, research outlook, network

Acknowledgements: Chiang Mai University, Kasetsart University, Chiang Mai Rajabhat University, Mahasarakham University

# Eco-evolutionary and conservation research on Korean freshwater fishes using a combined analysis of ecological and genetic data

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Through this research presentation, I would like to introduce our recent, some case studies using an integrated analysis of ecological (e.g. morphological) and genetic/genomic data to better understand unresolved ecological and/or evolutionary questions of several freshwater fish species indigenous to the Korean Peninsula. From conservation genetics research on endangered freshwater bitterling and its host mussel species that are particularly rapidly declining due to ongoing human intervention and climate change, to evolutionary ecology, speciation genetics, population genetics and phylogeography of closely related freshwater fish species (e.g. Korean dark chub-dark chub, Kumkang fatminnow-Deogyu fatminnow) and also environmental DNA (eDNA)-based biomonitoring of freshwater ecosystems. For future research directions of freshwater ecosystems in Korea, genetic- and phylogenetic diversity-based assessments of aquatic biodiversity will help to inform on effective management and conservation for the ecosystems as a whole. eDNA-based biomonitoring can also be useful for assessing 'the ecosystem health' in relation to physico-chemical environmental conditions at a relatively large scale. Moreover, morphology-based taxonomic study of three dimensional (3D) models and micro CT scanned data can assist in solving the taxonomic issues of closely related, but taxonomically unresolved species groups and also to discover cryptic diversity and even a novel species.

Keywords: biodiversity, coextinction, conservation, ecosystem health, environmental DNA (eDNA), phylogenetic diversity, phylogeography

Acknowledgements: This study was supported by a National Research Foundation of Korea (NRF) grant [NRF-2020R1I1A2069837] funded by the Korean government and also by a grant from the Korea National Park Research Institute (Project Number: NPRI 2023-38).

# III. Main Session

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- **Session 1** New Technology in Hydrobiology: Insights & Challenges
- **Session 2** Taxonomy, Molecular Phylogeny and Evolution
- **Session 3** Behavior / Structural and Functional Ecology
- **Session 4** Aquatic Biodiversity and Conservation / Climate Change
- **Session 5** Biomonitoring and Bioassessment of Aquatic Ecosystems
- **Session 6** Hydrobiology for Human Health / Vector Ecology

# Applications of automated image recognition for marine megafauna

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The biodiversity of marine megafauna plays a key role through ecosystem services. To investigate and monitor the marine biodiversity, extensive surveys are being carried out, which generate huge amounts of images (still images and videos) of megafauna specimens. Manually analyzing the images, such as classification based on morphological features, is labor-intensive and time-consuming, and the expertise of domain knowledge is required. However, the number of experts is limited. Recently, artificial intelligence (AI), especially deep learning (DL) techniques, have been applied to automated image recognition, providing promising solutions to this challenge. Previously, a web application, named EchoAI, has been constructed for automated image recognition of echinoderms in global oceans. Subsequently, two more applications have been developed for automated image recognition of marine fishes and decapods. To improve the model performance, different DL backbones have been tested and evaluated, including EfficientNetV2, Vision Transformer, CLIP (Contrastive Language-Image Pre-training). The recent achievements will be presented. As the AI-based assistant tools are increasingly used in the analysis of image data and might substantially boost the robustness of the related research, there is an urgent need to establish related standards. Thereby, recommendations to enhance the reproducibility and comparability in AI-based automated image analysis will also be discussed, such as detailed documentation of biodiversity data sources and data division. Considering the progress of artificial intelligence and image database, adaptive strategies should be used for standardization to promote the integration of state-of-the-art technical advancements to achieve the best practices in automated image analysis.

Keywords: artificial intelligence, deep learning, biodiversity, automated image recognition, megafauna

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# Relationship between benthic macroinvertebrate communities and hydromorphology using unmanned aerial vehicle

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The distribution of benthic macroinvertebrates in streams is influenced by hydromorphological heterogeneity, which provides a spectrum of ecological niches for optimal growth and development. Historically, research on the impact of hydromorphology on these communities has primarily focused on macroscale hydromorphological characteristics. Meanwhile, advancements in remote sensing technology in hydrology have revolutionized watershed research, facilitating the efficient and cost-effective acquisition and utilization of hydrological information across vast areas. The popularize and improvement of unmanned aerial vehicles (UAVs) have enabled the capture of extensive river areas at a low cost and with high resolution. These acquired images can be processed to generate high-resolution 3D representations of rivers, unveiling mosaic patterns of distinct hydromorphological patches. To analyze the effect of microscale hydromorphological patches on benthic macroinvertebrate communities, we selected 61 sites in Han River basin on the central Korean peninsula. Each river was filmed 1km using a drone based on the survey point, and the acquired images were processed with the Agisoft Metashape Pro (version 2.0.4) to obtain high resolution orthophotos. Benthic macroinvertebrate monitoring data were obtained from the Water Environment Information System (water.nier.go.kr). Hydromorphological characters were identified from the orthophotos and compared with benthic macroinvertebrate communities. Our study demonstrates the potential of utilizing unmanned aerial vehicles (UAVs) to analyze the relationship between mesohabitat characteristics and benthic macroinvertebrate communities in rivers and streams.

Keywords: Benthic macroinvertebrate, Hydromorphology, Unmanned aerial vehicles, Orthomosaic photos

# Climate-driven difference in gut microbiome community of dung beetles (Coleoptera: Scarabaeidae: Scarabaeinae, Aphodiinae) from different climate zones in Mongolia

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Mongolia, characterized by its diverse landscapes ranging from desert, steppe and taiga areas, harbors a rich source of biodiversity. The dung beetle of Mongolia is found in a wide range of habitats, from arid desert regions to grassland areas with nearby freshwater environment. They interact closely with the surrounding ecosystem by working as active decomposers by their remarkable ability to break down the feces of other animals. As the field of microbiome research gains importance and interest day by day, it is yet known about the relatedness of habitat conditions with host gut microbiome structure which resides in the habitat. In this study, a total 260 gut bacterial community samples of 11 dung beetle species belonging to 6 genera, 2 subfamilies of family Scarabaeidae were examined. *Vagococcus* was identified as the common bacteria genus of Mongolian dung beetles (Max = 50%, Min = 10%), which majorly dominates subfamily Scarabaeinae. And Wolbachia (Max = 90%, Min = 40%) dominated Genus *Onthophagus* (Scarabaeinae), while *Chishuiella* (Max = 60%, Min = 15%) covered a high proportion in Aphodiinae dung beetles. Statistical techniques and clustering algorithms such as NMDS, DBSCAN and Hierarchical clustering were employed for dung beetle gut microbiome clustering, and to evaluate the clustering validity between each clustered group. Dung beetle samples from the same habitat type were closely clustered together, while samples from different habitat types did not form distinct clusters, even if they were in the same taxonomic group. Assortment of 19 Bioclimate factors which has the most impact on bringing difference between gut microbiome from each habitat type (desert and grassland) was held by Random Forest algorithm. Mean temperature of the warmest quarter was shown to be the major Bioclimate component which mostly contributes to the difference of gut bacterial community between each habitat type. Furthermore, the analysis of the Mongolian dung beetle microbiome community revealed the presence of several bacterial species commonly found in freshwater environments, including *Acinetobacter*, *Chishuiella*, and *Pseudomonas*. Additionally, several bacterial species were discovered which are known to infect freshwater inhabitants, such as *Vagococcus*. I assume the existence of (1) symbiotic key microbial taxa that contribute to the fitness of host individuals in different habitat conditions, and (2) the specific climate factor which may potentially affect the gut microbiota of residing animal fauna.

Keywords: Aphodiinae, climate, dung beetles, gut microbiome, Mongolia, Scarabaeinae

# Enhancing biological water monitoring through supervised machine learning: A taxonomy-free approach using macroinvertebrate eDNA metabarcoding

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Biomonitoring data plays critical role in the assessment of water quality, whether using traditional morphological methods or eDNA methods. However, most operational taxonomic unit (OTUs) generated from eDNA sequences cannot be accurately classified at low taxonomic level due to limited reference barcode library. Recently, the biotic index (BI) predicted by supervised machine learning (SML) using unassigned sequence data has shown better performance in water quality assessment compared to that of assigned data. Nevertheless, it remains unclear whether the predicted average score per taxon (ASPT) of macroinvertebrates can more accurately discriminate water quality classes than those calculated from assigned families. Our study addressed this issue by developing an SML predictive model based on morphological data and eDNA data collected from 233 steam and river sites in the Taihu Lake basin and Qiantang River Basin, China. We calculated ASPT using the 112 morphological families and 102 eDNA assigned families. Then we applied random forest algorithm to predict the ASPT of 3138 unassigned OTU based on morphological ASPT. Our results showed predictive ASPT performed better than ASPT derived from assigned families when comparing their linear relationship with morphological ASPT. Moreover, predictive ASPT showed remarkable correlations with measured water chemical variable than morphological and family-assigned ASPTs. Our results highlighted the potential of SML predicted macroinvertebrate index to accurately assess water quality as well as identify stressors.

Keywords: biomonitoring, supervised machine learning, environmental DNA, ASPT, benthic macorivertebrates

# The eDNA methods outperforming the traditional method in capturing the diversity of stream benthic macroinvertebrates

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Abstract: eDNA method is a promising new technology for detecting freshwater biodiversity. In addition, eDNA data can offer new insights into relationships between communities and the environment. Various active and passive eDNA sampling methods have been tested for their effectiveness in species detection. However, research on the performance of active and passive eDNA methods in detecting diversity of stream macroinvertebrates compared to morphological method is limited. This study aimed to evaluate the effectiveness of different eDNA methods and traditional methods in detecting taxonomic richness and beta-diversity, as well as the relationships between sequences and individuals of Operational Taxonomic Units (OTUs) simultaneously detected by traditional and eDNA methods. We collected eDNA and macroinvertebrate samples from six stream sites using passive eDNA method from channel and stream side, respectively, two active eDNA methods (water and ethanol) and Surber net. On average, the eDNA methods detected twice as many OTUs (295) compared to the traditional method (139). Passive methods detected an average of 361 OTUs, outperforming water (258) and ethanol (200) methods. Additionally, passive methods identified the highest unique OTUs (71) among all four methods. Nevertheless, all methods showed a consistent beta-diversity pattern with values ranging from 0.719 to 0.737, predominantly driven by the turnover component. The relative sequence abundance of families demonstrated consistent linear relationships with the individuals found in morphological samples. These findings highlighted that eDNA methods are well-suited for monitoring stream macroinvertebrate diversity, with passive methods showing higher efficiency in capturing macroinvertebrate species compared to active methods.

Keywords: eDNA, benthic macroinvertebrates, active eDNA method, passive eDNA method, biodiversity

# Environmental DNA study by a newly developed DNA barcoding region (with the mtDNA 16s rRNA region): Issues with the general DNA barcoding region

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DNA barcoding is a powerful tool for automated species identification, using standardized genetic regions. It is particularly useful for revealing the existence of cryptic species or biomonitoring. In methods employing DNA barcoding, environmental DNA (eDNA) emerges as a powerful detection tool, enabling easy and effective biomonitoring of aquatic insects. DNA barcoding and/or eDNA require the development of a universal PCR primer set. In general, the mtDNA COI region is used for insect DNA barcoding, but previous studies based on this region have not successfully detected many insect species, and there has been frequent amplification of non-target taxa (e.g., algae and diatoms). Under such circumstances, we developed new universal primer set, “MtInsects-16S”, based on the mtDNA 16S rRNA region which were designed to include a hypervariable site between highly conserved sites, and confirmed successful DNA amplification of all tested species (14 orders, 43 families, 68 species), including Apterygota and Pterygota. Also, we conducted eDNA water sampling and physical capture surveys at six sites in the Sagami-gawa River and Sakawa-gawa River, and compared eDNA analyses using both the general COI (fwhF2 and EPTDr2n) primer set and our newly developed 16S primers set. As a result, among the list of species which were collected by physical capture surveys, 74.9% were detected by the MtInsects-16S primer set, while 40.1% were detected by the COI (fwhF2 and EPTDr2n) primer set. This study demonstrated that the application of eDNA analyses using the MtInsects-16S primer set could be conducted with accuracy and reliability, provided that a corresponding reference DNA database can be established. Finally, we recommend the mtDNA 16S rRNA region as the new standard DNA barcoding region for insects.

Keywords: biomonitoring, cryptic species, freshwater, metabarcoding, mtDNA 16S rRNA, MtInsects-16S

# The exact morphology of a new record mayfly *Thraulus ishiwatai* Grant, 2024 from China (Ephemeroptera: Leptophlebiidae)

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The exact morphology of the species *Thraulus ishiwatai* Grant, 2024, which was found in Japan and Korea before, was not represented and photographed in details. Here we collected some materials of it from eastern China. They show this species can be distinguished from the other members of *Thraulus* Eaton, 1881 by the following characters: in nymphs, anterior margin of labrum with a median process, dark abdominal tergites with conspicuous pale bands and grey to dark tibiae, gills I with a lanceolate dorsal lamella and a half broadened ventral lamella; in imagos, they have similar color pattern on abdomen and dark tibiae to those of nymphs, their wings have remarkable brown pigments at base. Morphologically, this species seems have an intermediate position of the genus. This is the first record of the species *Thraulus ishiwatai* from China. And together with others, they show China has diverse *Thraulus* species.

Keywords: mayfly, aquatic insect, diversity, morphology, ecology

# Description of two new Baetidae species in Korea (Ephemeroptera)

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The family Baetidae, comprising over 900 species across approximately 100 genera, stands as the most species-rich taxon within the order Ephemeroptera. Its distribution spans all biogeographic regions, with nymphs adapting to various types of freshwater microhabitats. Due to the lack of distinct morphological characteristics among adults, species identification in Baetidae typically relies on nymphal traits. However, even among closely related species, morphological differences are subtle, prompting researchers to increasingly use molecular analyses to uncover cryptic species. Approximately 100 species of Baetidae have been recorded in geographically adjacent regions to Korea, including Mongolia, Russian Far East and Japan, but only 15 species from 8 genera have been documented in Korea, with the last record dating back to 1998. We conducted multiple surveys from 2022 to 2024 in Korean streams, particularly in high mountains and island regions, to find unrecorded Baetidae species in Korea. *COI* barcode sequences were extracted from the collected nymphal specimens. Using this data and *COI* barcodes from Baetidae species available in the NCBI database, we constructed a phylogenetic tree and discovered that two species groups, each belonging to the genera *Baetis* and *Nigrobaetis*, formed distinct clades with significant genetic distances from the other species in Korea. Structural traits of the mouthparts and abdominal segments were examined and described in detail to define the species' morphological concepts. The discovery of these species expands the diversity of Baetidae in Korea and is expected to enhance our understanding of Baetidae distribution in Northeast Asia through future comparisons with species lists from neighboring countries.

Keywords: Baetidae, DNA barcodes, Mayfly, Korea, New Species

2-1-4

# Taxonomic revision of the *Drunella ishiyamana* species group (Ephemeroptera: Ephemerellidae) in the East Palaearctic

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The mayfly genus *Drunella* is a diverse group known to show intraspecific morphological variations in their larval stage. Recent molecular analyses of *Drunella* species revealed possible cryptic diversity within several groups, including the Northeast Asian *D. ishiyamana*. To properly assess the cryptic nature of *D. ishiyamana*, past articles of this species were reviewed, and larval specimens of Japan, Mongolia, and South Korea were morphologically examined. Based on our results, a new species *D. punctata* sp. nov. is formally described based on their unnotched median anterior tubercle on their head and weakly formed pairs of abdominal submedian tubercles. Phylogenetic reconstruction using mitochondrial cytochrome c oxidase I sequences supports our findings and indicates that the *ishiyamana* species complex is much more diverse than it appears. Further morphological and phylogenetic analyses of *D. ishiyamana* specimens from other regions are needed to fully understand the biodiversity of this group.

Keywords: Biodiversity, Drunella ishiyamana, *Drunella punctata* sp. nov., Mayfly, Northeast Asia

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2-2-1

**Keynote Speaker**

# Genetic evidence of multiple freshwater invasions and genomic adaptation in the bivalve family Cyrenidae (Heterodonta: Mollusca)

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Habitat transitions in living organisms are key innovations often coupled with species diversification after their successful adaptation to new environment. The Cyrenidae is among the most well-known heterodont bivalve groups that have successfully invaded freshwater systems from brackish water environments. This family also exhibits a wide spectrum of lineage-specific reproductive modes, ranging from planktotrophic development to parental care system with maternal incubation. Given the wide array of habitat types and reproductive modes among Cyrenidae species, a robust phylogeny and genomic evidence from comparative genomic analysis are fundamental to precisely understanding their evolutionary diversification of reproductive modes and patterns of adaptive radiation into freshwater environments. Genetic evidence of multiple freshwater invasions and genomic adaptations in response to habitat transitions will be presented in detail.

Keywords: multiple freshwater invasions, mitochondrial genome phylogeny, comparative genomics, Cyrenidae, *Corbicula*

Acknowledgements: This study was supported by the National Research Foundation of Korea (NRF) grant funded by the Korean Government (MSIT) (No. 2020R1A2C2005393).

# The presence of cryptic species diversity in Korean endemic freshwater fish, *Zacco koreanus* suggests taxonomic re-evaluation with reference to its relative *Zacco temminckii*

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Allopatric speciation has long been considered as a primary mechanism for the origin of species in freshwater ecosystems as geographically disconnected/isolated rivers serve as a ‘barrier’ to gene flow among populations. *Zacco koreanus*, a widely distributed freshwater fish species endemic to Korea, was recently classified as a new species from its relative *Zacco temminckii*. However, morphological interspecific differences between the two species are often ambiguous, and genetic and morphological differences between/within species remain also unclear. In this study, using mitochondrial DNA cytochrome oxidase I (COI), control region (CR) and our developed 9 microsatellite loci, we determined the phylogenetic relationships and population genetic structure among geographically separated six river basins [Z. *koreanus*: Han River (HR), East-flowing river (ER), Geum River (GR), Nakdong River (NR), Seomjin River (SR) and Mangyeong River (MR); Z. *temminckii*: (GR)]. We also quantitatively evaluated three previously proposed ecotypes of Z. *koreanus* and compared their morphology with Z. *temminckii*. We found relatively well-separated clades evolved in each of the river basins in Z. *koreanus*. Among them, the NR/SR clade showed the farthest genetic distance from other river basins and turned out to be the most closely related to Z. *temminckii*, with some individuals showing signs of hybridization. Moreover, despite significant genetic differences, the observed morphological and body coloration disparities among the three ecotypes of Z. *koreanus* were not apparent, supporting the hypothesis of cryptic diversity evolved in Z. *koreanus*. These findings offer valuable insight into how Korean freshwater fishes evolved ecologically in response to geographically isolated river environments.

Keywords: cryptic species, ecotype, endemic species, phylogeography, speciation

Acknowledgements: This study was supported by grants from the Korea Environment Industry & Technology Institute (KEITI) through Aquatic Ecosystem Conservation Research Project funded by Korea Ministry of Environment (MOE) (2020003050004). This work was partly supported by a grant from the Korea National Park Research Institute (project number: NPRI 2022-05) and also by a National Research Foundation of Korea (NRF) grant funded by the Korean government (NRF-2020R1I1A2069837).

# Population genetic study of two endemic loach species of Korea: Paleo-drainage systems, river capture, and distribution

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Two closely related species of loach, *Iksookimia hugowolfeldi* and *I. longicorpa*, are typically found in rivers and streams that flow into the southwestern coasts of the southern Korean Peninsula. Our research on these sister species has revealed that these sister species are made up of seven distinct genetic groups, each with approximately the same genetic distance from one another and geographically subdivided. This result suggests that there has been a radiative evolution from the common ancestors. The absence of monophyly within each of the nominal species, however, has challenged the current taxonomy. The observed genetic pattern is associated with two environmental factors. Firstly, geographical isolation due to distance, land, and sea was proportional to the divergence among the fish populations. Secondly, the current drainage and paleo-drainage systems, which act as fish dispersal routes, have impacted their connectivity. During the ice ages, there were past connections between currently disconnected islands and independent small rivers through paleo-drainage systems, which influenced the genetic connectivity of fish populations between them, respectively, resulting in regional genetic groups. Finally, evidence of river capture events was found in the upper parts between the Yeongsan and Seomjin Rivers, which provides a useful hypothetical framework for studying the effects of river capture on other fish species in the future. Based on these findings, our study suggests that *I. hugowolfeldi* should be synonymized with *I. longicorpa*, and caution should be exercised when deciding on the species status of these fish without sufficient geographical sample coverage and genetic evidence.

Keywords: cobitid fish, *Iksookimia*, population genetics, climate changes, paleo-drainage systems, river capture

# Understanding speciation process of Korean endemic fat minnow (*Rhynchocypris kumgangensis)* and its close relative, Deogyu fat minnow (*Rhynchocypris deogyuensis*), based on genetic/genomic and morphological analyses

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*Rhynchocypris kumgangensis* is an endemic and cold-water adapted fish species that inhabits only the upper reaches of rivers and streams. *R. kumgangensis* inhabiting the Gucheondong Valley was classified as a novel species, *Rhynchocypris deogyuensis*, based on genetic and morphological analyses. However, whether they are separate species and the level of genetic/phenotypic divergence remain uncertain. We analyzed the population genetic structure of both species using mtDNA cytochrome oxidase I (COI) and control region (CR) sequences and eight microsatellite loci. Moreover, we quantitatively assessed the genetic differences between the two species at the whole mitogenome level. Furthermore, we quantitatively evaluated the metric/meristic and also geometric morphometric characteristics to assess the shape differences between the suggested two putative species. Results revealed higher genetic diversity in the 11 populations of R. *kumgangensis* compared to the single population of *R. deogyuensis*, with *R. deogyuensis* showing only one haplotype and high levels of inbreeding (FIS=0.25). Comparative mitogenome analysis showed approximately 2.9% divergence between *R. kumgangensis* and *R. deogyuensis*, supporting species-level divergence, Among measured traits, only four metric traits showed significant differences between *R. kumgangensis* and *R. deogyuensis*. However, geometric morphometric analysis revealed significant morphological differences between the two species. Lastly, low values in the length-weight relationship in *R. deogyuensis* are attributed to increased inbreeding and depleted genetic diversity. In conclusion, our results provide genetic/phenotypic evidence for considerable divergence between *R. kumgangensis* and *R. deogyuensis*, supporting more the hypothesis that they are distinct species, although we are still uncertain whether the speciation process is already complete.

Keywords: Allopatric speciation, endemic freshwater fish, coldwater fish, mitochondrial genome, morphological divergence

Acknowledgements: This work was supported by a grant from the Korea National Park Research Institute (Project Number: NPRI 2023-38) and the National Research Foundation of Korea (NRF- 2020R1I1A2069837).

# Faunistic study of Mycetophilidae (Diptera: Bibionomorpha) in Ulleng Island

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Ulleung Island is the largest volcanic island in South Korea located on the East Sea. The island has a unique ecosystem with a warm temperate climate caused by geological isolation from the mainland. The freshwater ecosystems of Ulleung Island act as an ecological indicator that reveals changes caused by various environmental factors. For that reason, faunistic studies on insects in Ulleung Island water systems have been conducted by several researchers. The family Mycetophilidae is one of the largest groups in the infraorder Bibionomorpha in Diptera with eight subfamilies, 230 genera, and almost 5,000 described species worldwide. They have a mycetophagous larval stage and act as pollinators in nature. Adults inhabit moist environments such as under the roots of trees and rocks covered with mosses near water systems. Mycetophilidae has a very diverse habitat environment, it has never been reported before for this region. The list of insects in Ulleung Island has 18 orders, 179 families, and 1,177 species were previously reported. However, these data were reported without Mycetophilidae due to the absence of specialists. Thus, this study aims to report the freshwater systems of Ulleung Island and the list of Mycetiophilidae in Ulleung Island. In this study, six genera and eight species of Mycetophilidae including two newly recorded species. Further investigations on this island are needed to get more accurate data on the fauna of Mycetophilidae.

Keywords: Fresh water system, Fungus gnat, Mycetophilidae, New recorded species, Species list

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# Species diversity, systematics and conservation of freshwater mussels (Bivalvia: Unionida) in China

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China is a crucial distribution centre for global freshwater mussels, particularly in East Asia. It is imperative to update the phylogenetic relationships, distribution patterns, and conservation status of Unionidae in China with taxonomic revisions and new species discoveries. This study comprehensively reviews the classification and phylogenetic research on Unionidae in China through literature compilation and analysis. The classification system proposed includes 2 families, 5 subfamilies, and 38 genera. In China, we have identified 113 valid species of Unionidae. The study summarizes diversity patterns by organizing distribution information. Our findings suggest that research has predominantly focused on the middle and lower Yangtze River, creating significant data gaps in other regions. We compares the endangered status of three Chinese Unionidae levels, revealing that most species in China are severely threatened, and there is a significant lack of data for endangered status assessments. Therefore, a nationwide survey is necessary to prioritize environmental protection, intensify scientific research, raise conservation awareness, and promote sustainable utilization.

Keywords: Unionidae; Systematic Classification; Biodiversity; Conservation; China

# The morphology of *Potamanthodes formosus* Eaton, 1892 and two related species in eastern Palaearctic (Ephemeroptera: Potamanthidae)

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The species *Potamanthodes formosus* was named originally by Eaton in 1892. The types of it were collected from Burma. From then on, a series of researchers have reported it from several countries in eastern Asia, including China, Korea and Japan. However, the updated researches and new materials show there are three different species in eastern Palaearctic, the other two are *P. yooni* and *P. japonica* sp. nov. In adults, they can be differentiated by eye size, male genitalia, color pattern on wings and venation of hindwing. In nymphs, they have different sizes of compound eyes, mandibular tusks and foretibiae. Including the *P. formosus* and *P. yooni*, China has nine *Potamanthodes* species out of total ten, and the eastern Asia is its original and diverse center.

Keywords: Ephemeroptera, taxonomy, phylogeny, diversity, China

# Local variation in the spawning patterns of a bitterling fish species and its spawning in a small Asian clam from the Nakdong River basin

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The bitterling is a temperate freshwater fish with a symbiosis with host mussels for their spawning. Freshwater mussel populations have recently been declining drastically, due to accelerating anthropogenic pressure, which can pose considerable threats to the risk of bitterling extinction cascades. The observed bitterling's spawning events in a small Asian clam, *Corbicula fluminea* from Korea were reported in our previous study, which can be attributed to increasing resource competition for mussels. Its conservation importance as a novel host mussel species for bitterling fishes further needs to be studied, particularly with regard to whether bitterling's embryo can undergo successful development inside the small mussel. The present study aimed to further explore the bitterling's spawning eggs/larvae in *C. fluminea*, and to determine whether the bitterling's embryo can emerge from *C. fluminea* through the successful development. In addition, we examined the spawning patterns of bitterling fish species in geographically separate river basins at local scales. We observed bitterling's spawning eggs/larvae in every one to four mussel species at the study sites. By using our developed genetic technique, we were able to identify number of eggs/larvae for each bitterling fish in each mussel species. In addition, we experimentally tested whether bitterling's offspring develop successfully and leave from the mussels to sustain an independent life cycle. The results of this study will provide ecologically and genetically important information for developing effective conservation, management, and restoration strategies by demonstrating the geographic differences in spawning patterns of bitterling fish species in local riverine environments.

Keywords: bitterling fishes, *Corbicula fluminea*, coextinction, competition, conservation

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# First insight on functional morphology of the mandibular tusks of the burrowing mayflies (Insecta: Ephemeroptera) using microcomputed tomography scanning and morphometrics of 3D models

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This study utilizes 3D models generated from micro-computed tomography scanning to investigate the morphometrics of mandibular tusks within the superfamily Ephemeroidea (Ephemeroptera), employing an innovative automatic landmarking method based on point cloud registration. Including at least one representative from all six families of tusked burrowing mayflies worldwide, we classify species into morphotypes based on dwelling type and substrate habitat, following Bae & McCafferty (1995). These morphotypes are characterized as: 1) flat body-shape and interstitial dwellers (IDm), 2) cylindrical body-shape and burrowing dwellers (BDm), and 3) cylindrical body-shape and hard substrate burrowing dwellers (HBDm). Discriminant and K-means analyses support these proposed morphotypes based on mandibular tusk and foreleg shape. Principal component analysis (PCA) reveals evolutionary tendencies in tusk shape, driven by curvature, size, basal cross section type, and robustness. Additionally, PCA of foreleg shape correlates with size and foretibiae development. These outcomes are used to compare and associate shape tendencies of tusks and forelegs with their respective functions (i.e., as weapons, burrowing devices, and feeding devices), providing ecological and functional-adaptation insight for these structures.

Keywords: Ephemeroptera, Ephemeroidea, burrowing mayflies, mandibular tusks, morphometrics, functional morphology

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# Spawning patterns and symbiotic relationships of eight Korean indigenous freshwater bitterling fishes (Pisces: Acheilognathinae) with host mussels

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Bitterling fishes evolve an idiosyncratic symbiosis with freshwater mussels, in which they are obligated to spawn in the gills of mussels for reproduction. The mussels may also exploit bitterling fishes as a dispersal vector by infecting their larva (i.e. glochidia) to the gills, fins, and/or body of the host at an early life history stage. The relationship between these two interacting animals has thus been considered as mutualistic or even opposite, double host-parasitic. Which interspecific interaction (mutualism, commensalism, or parasitism) fits the most the bitterling-mussel relationship remains not resolved yet. In the present research project, we had four primary objectives. First, we determined the spawning preference and phenotypic adaptation of eight Korean bitterling fish species in relation to the host body size. Second, we analyzed and compared the phylogeographic and population genetic structure of the presumed two coevolving bitterling-mussel species (*Tanakia signifer* and *Nodularia breviconcha*) to test for the symbiotic relationships. Third, we investigated the potential ecological advantages of autumn-spawning bitterling (*Acheilognathus rhombeus*) in response to different competitive pressures and also identified candidate genes involved in egg winter-diapause using transcriptomic analysis. Lastly, we tested the hypothesis of the bitterling's preference for larger host and quantitatively analyzed the egg ejection rate of the mussels through laboratory-based behavioral experiments. Our findings will provide insight into the long-standing evolutionary question of how and why symbiotic interactions in the two taxa evolve, the mechanisms underlying the evolution of reproductive traits (spawning season, egg diapause) and also the intensity of selection pressure as counter-adaptation in mussels.

Keywords: coextinction, host preference, morphological adaptation, mussel-symbiotic bitterling, coevolution, spawning strategy, diapause

Acknowledgements: This study was supported by a National Research Foundation of Korea (NRF) grant [NRF-2020R1I1A2069837] funded by the Korean government.

# Diversity of EPT group (Ephemeroptera, Plecoptera, and Trichoptera) and its implications for environmental DNA application in Thailand

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Aquatic insects play an essential role in environmental monitoring and research, particularly the EPT group, i.e., Ephemeroptera, Plecoptera, and Trichoptera, which are highly sensitive to environmental changes. According to the impacts of climate change on the quantity and quality of Asian streams, these EPT species are at risk. Recently, environmental DNA (eDNA) applications have been used for diversity studies and conservation purposes; however, the lack of available species sequences can limit their utilization. This study aims to assess the availability of both species lists and their *COI* sequences from online databases in Thailand. Moreover, a review of the applications of the EPT group in Southeast Asian countries is provided. As a result, the diversity of identified species is highest in Trichoptera, followed by Ephemeroptera and Plecoptera, respectively, while their *COI* sequence availability in the online database is most comprehensive for Ephemeroptera, followed by Trichoptera and Plecoptera, respectively. In addition, our findings indicate that the EPT group was utilized in assessing the health of rivers, tracking habitat changes due to human disturbances, and deforestation. The activities involve using the EPT index and studying the relationships between EPT taxa and physiochemical parameters, substrates, decomposition, and so on. This underscores the urgent need for enhanced data collection and further research, particularly concerning the Plecoptera group, to facilitate future environmental and ecological studies in Southeast Asia.

Keywords: EPT, bioindicator, stream, Southeast Asia, biodiversity, eDNA, Thailand

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# Predicting the current and future distribution of non-native fish in the rivers of the Western Ghats biodiversity hotspot, India using MaxEnt model

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This study presents the outcomes of a pioneering endeavor aimed at predicting the distribution of non-native fish within the Western Ghats (WG) biodiversity Hotspot using the MaxEnt model. While documenting 28 non-native fish species from the southern region of the Western Ghats, this paper focuses on 12 specific alien species, comprising seven invasive species (*Clarias gariepinus, Cyprinus carpio, Gambusia affinis, Oreochromis mossambicus, Oreochromis niloticus, Poecilia reticulata,* and *Pterygoplichthys pardalis*) and five exotic species (*Ctenopharyngodon idella, Hypopthalmichthys molitrix, Pangasianodon hypopthalmus, Piaractus brachypomus,* and *Trichopodus trichopterus*). The modeling utilized solely the presence data of these species in the water bodies of the study area, particulary at hydrobasins of the rivers. The findings indicate that the guppy *Poecilia reticulata* is projected to be the most invasive species, potentially occupying over 80% of the total area of the state by 2050 and 2070 across various climatic conditions. Additionally, *Gambusia affinis* and *Cyprinus carpio* are anticipated to pose significant challenges under future climatic conditions for both predicted years. *Oreochromis mossambicus* and *O. niloticu*s are expected to thrive in future climatic scenarios by 2050 and 2070. Conversely, the likelihood of occurrence for species such as *Ctenopharyngodon idella, Clarias gariepinus, Hypophthalmichthys molitrix, Pangasianodon hypophthalmus, Piaractus brachypomus, Pterygoplichthys pardalis*, and *Trichopodus trichopterus* is more limited under future climatic conditions. These results are contextualized within the framework of managing alien species in biodiversity hotspots rich in endemic species, emphasizing the importance of spatial modeling and prioritized conservation action.

Keywords: Bioinvasion, modelling, invasive alien species, exotic species, climate change

4-1-3

# Current and future risk of invasion posed by non-native freshwater fishes in a mega-biodiversity country (The Philippines)

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The Philippines is a mega-biodiversity country hosting a vast number of aquatic species of which most are endemic, with its 7,100 islands, boasts incredible aquatic biodiversity. Many fish species are vital to the country's ecosystems and economy. However, this rich resource is threatened. Research in the Philippines has primarily focused on marine and terrestrial ecosystems, highlighting a significant gap in the study of inland waters and its freshwater fishes. In total, 374 freshwater fish species belonging to 29 orders and 78 families have been documented in the Philippines. This large number of fish species faces high extinction risks due to various human-induced impacts including the presence of introduced species. This study investigates the risk of invasiveness of the 64 introduced species in total currently present in the Philippines. Of these species, 65.6% and 70.3% were ranked as carrying a high or very high risk of invasiveness under current and future climate conditions, respectively. The highest risk species were goldfish *Carassius auratus*, Indonesian snakehead *Channa micropeltes*, largemouth black bass *Micropterus salmoides*, pirapitinga *Piaractus brachypomus*, vermiculated sailfin catfish *Pterygoplichthys disjunctivus* and Amazon sailfin catfish *Pterygoplichthys pardalis*. Given the high conservation value of Philippine freshwater ecosystems, efforts are needed from stakeholders and environmental managers in the mitigation and prevention of the detrimental impacts of the invasive fish species already present, and preventative measures are required to counteract the introduction of any additional species.

Keywords: Introduced species, Invasive species, Risk screening, AS-ISK, Climate change

# Three undescribed species of gut-inhabiting fungi from aquatic insects in Kuji River, Ibaraki, Japan

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Harpellales and Orphellales are fungal orders, of which species attached to aquatic insects' digestive tracts by a holdfast. To date, 284 species are known in total. Fifty-three species are known from Asia, consisting of 39 from Diptera, 10 from Ephemeroptera, and 4 from Plecoptera. Exploring less investigated hosts is required to elucidate the diversity in these orders. I collected aquatic insects from 2020 to 2023, in Kuji River, Ibaraki, Japan. *Baetis* sp. (Ephemeroptera) was collected from late-September to mid-November. *Glotzia* sp. (Harpellales) was obtained from the hindgut, with the genus characteristics of two short appendages and one long appendage on a spore. This species was distinguished from known species by having a zygospore with a long appendage exceeding 150 μm. *Capnia japonica* group (Plecoptera) was collected from late October to February. Two fungal species were detected in the hindgut, and sometimes simultaneous two-species infection was observed. The first species was *Genistelloides* sp. (Harpellales), with the genus characteristics of a long pear-shaped spore with two appendages, and biconical zygospores connected perpendicularly to the zygosporophore. This species was distinguished from known species by having two types of spore generative cells: long and short. The other species was *Orphella* sp. (Orphellales), with the genus characteristics of the accessory hyphae just above the holdfast and the cyme-like sporulation. Both spores and zygospores were coiled. This species was distinguished from known species by its long zygosporophore exceeding 200 μm. Asia has a large host fauna, and a large diversity of gut-inhabiting fungi is expected.

Keywords: Baetidae, Capniidae, Harpellaceae, holdfast, Legeriomycetaceae, Orphellaceae, Trichomycetes

# Semiaquatic Heteroptera (Hemiptera) from mangroves in Thailand

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The semiaquatic heteropteran fauna in mangroves located in Thailand were examined in 2019-present. In total, 175 Thai mangroves were surveyed to study the diversity of semiaquatic heteropterans. Specifically, 125 mangroves in the Gulf of Thailand and 50 mangroves in the Andaman Sea were chosen to examine the semiaquatic heteropteran fauna in this research. Twenty-three species representing 15 genera and eighth families were discovered. The most speciose family is Gerridae with nine species, whereas Gelastrocoridae contains a single species. Records of *Nerthra* (Gelastrocoridae), *Nereivelia* (Mesoveliidae), and *Stenobates* (Gerridae) were additional records from Thailand for the first time. The taxonomic, ecological, and biological information on these semiaquatic heteropteran is presented

Keywords: semiaquatic, Heteroptera, mangroves, Thailand

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# Taxonomic and functional responses of macroinvertebrate assemblage to summer drought in subtropical small water bodies

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Climate change is supposed to intensify the seasonal drought of small water bodies and accordingly greatly influencing aquatic biodiversity. Understanding the response of different aspects of biodiversity to the seasonal drought would benefit biodiversity conservation. Nevertheless, the response to summer drought of macroinvertebrate assemblages remains unclear in Asian monsoonal region. This study aimed to examine how the taxonomic and functional diversity of stream and pond macroinvertebrates response to summer drought. Macroinvertebrate and environmental data were collected from isolated ponds (IP), stream-fed ponds (SFP), pond linked stream (PLS) and non-pond linked stream (NPLS) in spring, summer and winter at Zijin mountain, Nanjing city, China. We found the measured environmental variables were generally similar between streams and ponds. Community compositions between ponds and streams were distinctly separated into two groups in spring and winter, but not in summer. Streams were of the highest species richness in spring whereas ponds had the highest species richness in summer. Both ponds and streams displayed the highest functional richness in summer and the lowest in winter. Nevertheless, the functional redundancy revealed an increased trend from spring to winter for both ponds and streams. The volumes of trait space of IP and SFP in summer were larger compared to spring and winter, in contrast the volumes of trait space of PLS and NPLS in summer were lower compared to spring and winter. Pure spatial factors generally played greater contribution than pure environmental variables in structuring pond and stream macroinvertebrate composition.

Keywords: seasonal drought, functional diversity, climate change, functional trait

# The latitude trends of riparian leaf litter traits across China

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Many studies found that the diversity and abundance of shredders increased with latitude and result in increased macroinvertebrate-mediated LL breakdown rate with latitude at global scale. Some researchers suggested that the latitudinal patterns of shredders may be related to the increased LL quality (e.g., low toughness) with latitude. Here, we tested this hypothesis by collecting riparian LL from 28 sites (8-10 species per site) across China. We measured nine physical LL traits, and collected data of climatic, geographical, and soil characteristics. Our results showed that: (1) there were >50 folds of variations for most traits, and there were strong correlations between paired LL traits; (2) many LL traits showed clear latitudinal trends: leaf dry mass, leaf length, leaf thickness, toughness, and leaf tissue density decreased with increasing latitude; while SLA increased with latitude, however, the latitudinal trends of LL traits were contrary between evergreen and deciduous species; (3) leaf thickness, toughness, and leaf tissue density increased with both mean annual temperature (MAT) and mean annual precipitation (MAP), while SLA decreased with MAT and MAP; (4) climatic, geographical, and soil characteristics explained 3-21% of the variations in LL traits, and climatic factors generally explained higher variations in LL traits than other factors. Our results imply that LL toughness and SLA which are suitable candidates of LL decomposability did show latitudinal trends across China, indicating higher LL quality towards the poles. More, future climate warming would induce decreased LL quality by increasing toughness, leaf thickness, and leaf tissue density, and decreasing SLA.

Keywords: leaf-litter, trait, latitude, climate change

# Study of aquatic invertebrate biodiversity in nature protected area («Bastak» state nature reserve, Jewish autonomous region, Russia) for the using in freshwater biological monitoring

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Studying the state of river ecosystems in protected areas is an important part of the work when creating regional systems for bioassessment of running waters. In the Russian Far East, the development of Rapid Bioassessment Protocols for Streams and Rivers (RFE RBPs) is being carried out in the Laboratory of Freshwater Hydrobiology, Federal Scientific Center for Biodiversity FEB RAS. To adapt RFE RBPs to the conditions and fauna of the Jewish Autonomous Region, since 2018, research began on the fauna and structure of benthic communities in protected areas not affected by anthropogenic impact. In the natural territory of the "Bastak" State Nature Reserve (mountain forest zone), 12 sample sites were chosen and 44 macrozoobenthos samples were taken: 26 qualitative, 14 conditionally quantitative (D-net) samples, and 4 quantitative (Surber Sampler) samples collected according to standard procedures (Vshivkova et al., 2019). To compare the similarity of species assemblages we used Ward's method with application of the Bray-Curtis similarity coefficient, which made it possible to divide the analyzed benthic communities into two groups with a probability of 95-98%. Two types of "faunas" have been identified, corresponding to two dominant communities: type I – lotic riffle biotopes of the rithral zone (in medial), type II – pool biotopes of the rithral zone (ripal). The same division was achieved by the use of a classification procedure based on fuzzy sets using the ordination method: the "core" of these agglomerations consists of the same species assemblages (samples) that were classified by the cluster method.

Keywords: bottom communities, running waters, freshwater bioassessment

# Diversity and immature stage dynamics of mosquitoes (Diptera: Culicidae) in rice fields in Dan Phuong district, Ha Noi city

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Mosquitoes are nuisance biters and disease carriers of significant public and veterinary health importance. Knowledge about the prevalence and distribution is crucial for comprehending the risk factors associated with the diseases transmitted by these vectors. Rice fields are dominant land use in agriculture, potentially harbors diverse mosquito communities in Vietnam. However, the ecological relationship between culicine and rice fields has never been studied in the country. The present study aims to determine if the rice growing should be used as parameter in risk model that predicts the abundance of mosquito populations. Mosquito larvae were collected biweekly since January 2024 using standard dipper, counted, and reared to adults. Samples collected were identified morphologically and genetically using Polymerase Chain Reaction (PCR) assays. As of now, we have morphological identified a total of 4 species of mosquito, belonging to 2 genera, which include *Anopheles* and *Culex*. The species are *Anopheles barbirostris*, *Anopheles umbrosus*, *Culex quinquefasciatus* and *Culex tritaeniorhynchus*. Results show that *Culex* make up the majority of larvae collected at all sample points. Furthermore, we also collected multiple water parameters such as pH, conductivity, turbidity, DO, rice height, water depth, … to build the model showing correlation between mosquito populations density and environmental parameters. The final results are expected to provide the list of species composition confirmed by both morphological and genetic methods and mosquito population dynamics along the rice development in northern Vietnam, that would helpful to understand the epidemiology for the mosquito community, and to generate data for developing larval control strategies in rice ecosystems.

Keywords: Rice growth cycle, culicide larval species, rice height, water depth, larval control

# Aquatic insects diversity and water quality assessment of a freshwater stream in Phayao Thailand

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The water sources in Phayao Province found many species of aquatic insects. According to a literature review on aquatic insects, each species can live in a variety of water sources. Therefore, aquatic insects can use to evaluate water quality. This study aims to find the diversity of aquatic insects and evaluate the water quality using aquatic insects. The samples were collected between August 2023 to December 2023. The study area is Maeyien waterfall. The head of this watercourse is abundance of nature and few disturbances which suitable for being as habitat of aquatic insects. The five sampling sites were chosen and 3 replicates of sampler were collected from each site. The results consist of 2-part, that are the diversity of aquatic insects and the water quality. Aquatic insects were analysed with BMWP Thai Score and ASPT and the water quality was assessed by measuring physico-chemical parameters. The diversity of aquatic insect in habitat were found 77 families from 9 orders. The most abundance order is ephemeroptera, subordinate with trichoptera and diptera. The number in family of baetidae (ephemeroptera) were found in enormous followed by heptageniidae (ephemeroptera), chironomidae (diptera) and hydropsychidae (trichoptera) respectively. The ASPT were 6.57-6.88 which is in the range 2-3 of the surface water quality standards of Thailand. This means that it can be used in agriculture and should be treated before consumption. The water quality by determined using aquatic insects indicated that good to fair level, which was similar to water quality by measurement.

Keywords: aquatic insect, diversity of aquatic insects, water quality, Maeyien waterfall

Acknowledgements: The project of aquatic insects diversity and water quality assessment of a freshwater stream in Phayao,Thailand can be implemented because it has been well support by Dr.Chayanan Jitmanee our advisor, who provides advice, knowledge and how to solve various problems. We would like to express our deepest appreciation to our committee and University of Phayao for the place and our colleagues for their assistance in this project. Finally, we hope that this project will be useful to those interested.

# Insect diversity assessment in an urban landscape: University of the Philippines Diliman campus, Quezon city

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Insect diversity assessments are essential for understanding ecosystem dynamics and informing conservation efforts, especially in urban areas where human activities affect biodiversity. However, in the Philippines, limited taxonomic and ecological studies on insects exist due to lack of specialists and taxonomists. Most studies focus on forests or protected areas, neglecting urban environments crucial for supporting diverse flora and fauna, including insects, which play vital roles in ecosystem functioning. This study aims to assess insect diversity within the University of the Philippines Diliman campus, one of Metro Manila's largest green spaces. Extensive terrestrial and aquatic adult insect sampling, including sweeping, light traps, and Malaise traps, covers various campus zones, including both protected and non-protected areas. As of March 2024, 130 insects belonging to 13 orders (*i.g*., Lepidoptera, Coleoptera, Odonata, Mantodea, Hymenoptera, Hemiptera, Orthoptera, Blattodea, Embioptera, Neuroptera, and Ephemeroptera, Trichoptera, and Diptera) have been identified. Notably, orders Ephemeroptera, Trichoptera, and Diptera serve as significant biodindicators of water quality. The objectives of the study include generating a photographic identification guide, discovering new species or records, and informing the public about insect diversity in the urban landscape. Ongoing sampling efforts are expected to reveal a higher species count. The comprehensive sampling and taxonomic identification will provide insights into insect diversity and its ecological significance in urban settings, guiding conservation strategies to sustain urban biodiversity. Furthermore, this study highlights the importance of taxonomists in conservation efforts in the country.

Keywords: insect diversity, taxonomic study, urban environments, conservation, biodindicators, water quality

# Assessment of local stressors shaping macroinvertebrate community composition in mountain rivers of Nepal

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Rivers are primary habitats for wildlife and centers for socio-cultural practices in a landscape, however, currently, they are highly stressed by diverse types of human interventions. Unraveling the magnitude of the impacts of these stressors on aquatic macroinvertebrates is key to maintaining and restoring river ecosystems. Here, we studied the impact of water abstraction, operations of hydropower dams, gravel extractions and organic pollution on the ecological integrity of six mountain rivers (100 m to 1300 m asl) in Nepal that are located in the east, central, western, and mid-western regions of Nepal, respectively and aimed to reveal which of the stressors leads to establish a novel community, which of the parameters' changes bring seasonal changes between natural and impaired sites and which of the organisms show persistence in all types of stressors. A total of 60 sites were sampled for water quality parameters and benthic macroinvertebrates for pre-monsoon and post-monsoon seasons of a year. Twelve water quality parameters including nitrate, ammonia, and orthophosphate; and three hydrological parameters: velocity, the wetted river width, and water depth were measured for each site and in the laboratory in addition to macroinvertebrates data. Macroinvertebrates were identified to the possible taxonomic resolution mainly genus and family levels. In total 73 taxa belonging to 49 families and 17 orders for post-monsoon seasons and 78 taxa belonging to 56 families and 18 orders of macroinvertebrates were recorded for pre-monsoon season. The diversity of macroinvertebrates was not found significantly different between the impairments while abundance was significant between the impairments. Changes in macroinvertebrate community assemblage were more pronounced in the rivers influenced by organic pollution followed by the operation of hydropower dams, water abstraction, and morphological degradation. Water temperature, total hardness, nitrate, and orthophosphate were found key parameters structuring the community assemblages. Multivariate analysis indicated that taxa of Ephemeroptera, Diptera, and Coleoptera were more resistant to different levels of impairments in rivers. The outcomes of the study would help river managers to restore the ecological integrity of the disturbed rivers to healthier ones.

Keywords: Biological indicators Organic pollution, Hydrological alteration, Municipal effects, Morphological degradation

Acknowledgements: We are thankful to the International Foundation for Science, Sweden for the Individual IFS grant.

# Restore polder and aquaculture enclosure to the lake: Balancing environmental protection and economic growth for sustainable development

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In this study, we investigated the ecological restoration of shallow lakes and buffer zones in the Yangtze-Huaihe River Basin, specifically focusing on the removal of polder and aquaculture enclosure areas within the lakes. By examining data from eight shallow lakes and their corresponding buffer zones, encompassing lake morphology, water quality parameters, and land use/land cover (LULC) data spanning from 2008 to 2022, which shed light on the complex relationships involved. During the process of restoring polder and aquaculture enclosure areas, we observed a general decrease in the extent of polders and aquaculture enclosures within the lakes. Notably, the removal of aquaculture enclosures had a more pronounced effect (reduction rate of 83.37%) compared to the withdrawal of polders (reduction rate of 48.76%). Linear regression analysis revealed a significant decrease in the concentrations of seven water quality parameters, including COD, CODMn, TN, TP, $NH_3$-N, Chl-a, and F, while pH and DO factors exhibit a distinct increasing trend. The results of RDA and Pearson correlation analysis demonstrated significant correlations between the area of polders and aquaculture enclosures and the changes in lake water quality. Encouragingly, the withdrawal of polders and the removal of aquaculture enclosures had a positive impact on the lake water quality improvement. In contrast, the LULC in the buffer zones of the lakes experienced a gradual decline owing to land degradation, resulting in a reduction in ecosystem service value (ESV). These results offer valuable support for policymakers in their endeavors to restore lake water quality, mitigate the degradation of buffer zones land, and promote the sustainable development of land and water resources.

Keywords: Shallow lake; Polder; Aquaculture enclosure; Water quality; Land use/land cover; Ecosystem service value

# The Philippine groundwater research trend from 1980 to 2023: A bibliometric analysis and evaluation

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Groundwater is a practical and economically important freshwater resource in the Philippines. At present, natural and anthropogenic factors have severely impacted the quality and quantity of our groundwater resources. We conducted a bibliometric analysis to investigate the dynamics of groundwater research in the Philippines from 1980 - 2023. A total of 77 document types were retrieved and selected from the Web of Science Core Collection and Scopus databases. Bibliometric analysis and visualization were done using Microsoft Excel and the *bibliometrix* R-package Biblioshiny. The findings showed a modest rise in research since 1997 and an annual publication growth rate of 0.97%. The University of the Philippines Diliman, Mapua University, and University of Santo Tomas had the most important groundwater-related publications; the top three most relevant authors were Siringan F, Cardenas M, and De Jesus K. Furthermore, the Philippines collaborated most with the USA and Japan on groundwater studies. The Geophysical Research Letters is the most preferred journal for groundwater research publication. The primary themes of groundwater research, as indicated by keyword analysis, were heavy metal contamination, geographic information system, and submarine groundwater discharge; groundwater modeling is an emerging research area. However, there is little groundwater research value associated with groundwater biodiversity. The current review presents the gaps and trends in groundwater research, which will form the foundation for future groundwater research in the Philippines.

Keywords: groundwater research, Philippines, groundwater biodiversity, Web of Science, Scopus

# Distribution of microplastic contamination and its impact on freshwater biota in Seti and Bagmati River basins, Nepal

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Plastic pollution is a major environmental challenge of the 21st century, with global usage projected to triple by 2060, leading to a doubling of leakage to 44 million tonnes annually and a tripling of accumulated plastic stocks in rivers and oceans. Nepal, like many regions, faces alarming levels of plastic pollution. Microplastic (MP) pollution research is crucial globally, yet Nepal's freshwater ecosystems are understudied. Research reveals significant variations in microplastic contamination levels between urban and remote rivers, influenced by human activities and seasonal fluctuations. Our study examines the spatiotemporal distribution of microplastics (MP) in water and sediment, and its impact on macroinvertebrates and fish species in the Glacial-fed Seti River and spring-fed Bagmati River of Nepal, which experience varying degrees of human interference. The Seti River, a left tributary of the Trishuli River, flows through the western region (Gandaki Province) of Nepal, originating from the base of the Annapurna Mountain and merging with the Trishuli River near Devghat. With a length of 140 km, the Seti River traverses Pokhara city, the fourth largest city in Nepal. Similarly, the Bagmati River, stretching 230 km within central Nepal, originates in Bagdwar-Shivapuri Nagarjun National Park and flows into India, passing through the capital city Kathmandu. A total of 34 sites along the Seti River and its tributaries and 45 sites along the Bagmati River and its tributaries were sampled across their entire lengths. Samples were collected during both winter and summer seasons. The methodology involved the collection of 50 liters of surface water (0-30 cm depth) from four random points at each site along the shoreline, spaced a few meters apart, using stainless steel containers. The entire 50 liters of water collected underwent filtration through a 32 µm stainless steel sieve. The residue trapped in the sieve was rinsed with 100 mL of filtered distilled water and transferred to a 250 mL glass bottle with a metallic cap. To preserve the samples, 2 mL of 70% filtered ethanol was added to each bottle, which was then capped and covered with aluminum foil. The collected samples were transported to the laboratory and stored at 4°C prior to analysis. For sediment collection, a 25 cm × 25 cm quadrat was used at each location to gather the top 2 cm of shoreline sediment using a stainless steel spoon. The sediment samples were sealed in aluminum foil bags and labeled for laboratory analysis. Benthic macroinvertebrates were sampled following a multi-habitat sampling approach, with 20 sub-samples collected using a kick-net with a mesh size of 500 µm at each site, considering available micro-habitats. Fish sampling was conducted using cast-nets and electro-fishing gear. At each site, 40 casts were made within the 200-400 m river stretch. Additionally, electro-fishing gear was utilized for a duration of 30 minutes within a 200 m river stretch where possible. Standard laboratory methods were employed for processing all samples, and the resulting findings will be presented at the 6th Biannual ASH Conference in Seoul 2024.

Keywords: Himalayan rivers, plastic pollution, macroinvertebrates, fish, ecological consequences

# Review of aquatic insect research in the Philippines

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The Aquatic insects research in the Philippines are very scarce and normally associated with biomonitoring of stream health. But even for ecological studies, species identification of larva is very difficult because majority of the larva species in the Philippines are not yet known. Studies associating larva and adult whether by rearing or using molecular tool is not a common study. That is why it is very challenging for Biodiversity studies because majority of the species are still unknown. Even in Genbank, the species that were sequenced were very few, even less than a hundred. The references that Philippine researchers are using are mainly the ones published from Malaysia, Hong Kong, and the Mekong River Commission. Species that were described for aquatic insects in the Philippines was as early as 1853 by Walker followed by expedition from the 1940s. As of now, there are a total of 44 Ephemeroptera species, 398 Trichoptera species, 37 Plecoptera species, about 200 species of Coleoptera, and more than 200 species of aquatic Hemiptera had been described in the country. The order Odonata was quite exceptional because of its charismatic morphological features such as the color and its large size, this is the most species that had been described. A total of 1,239 Odonata species has been recorded so far in the Philippines. Majority of the aquatic species described in the Philippines are concentrated only on famous islands such as Palawan, Mindoro, and Panay. Therefore, it is expected that the species number will double or even triple in number if biodiversity studies including aquatic insect adults will be prioritized. In University of the Philippines Diliman (UPD), study with regards to the insect biodiversity has started. This is one of the very few remaining forests patch in the city. The collection started last January 2024 and species of sensitive groups such as Ephemeroptera and Trichoptera were observed.

Keywords: Philippine insects, aquatic insects, Biodiversity, UP Diliman

# Composition of the food spectrum of Pacific salmon in the Karymai channel of the Bolshaya River

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Based on materials collected in July 2017, an analysis of the nutrition of juvenile salmon species (sockeye salmon *Oncorhynchus nerka*, chum salmon *O. keta*, coho salmon *O. kisutch*, char *Salvelinus malma*) was carried out. The diet of coho salmon was more varied than that of other salmonids. The results obtained were compared with data on the nutrition of juvenile salmon fish in 2018. Despite the differences in hydrological conditions, the nutritional spectra of chum salmon differed only in the absence of insects and mysids in the samples of 2017. And we did not find mollusks in the stomachs of sockeye salmon, whereas in the following year, they were present in the diet of sockeye salmon in large quantities. In 2018 there was also no phytoplankton in the diet of sockeye salmon. All other food components were practically the same. In 2018, the diet of juvenile coho salmon consisted only of insects, mysids, and juveniles of other fish, while in our 2017 samples, the diet of coho salmon consisted of larvae of chironomids, mayflies, stoneflies, insects, and mysids. In 2017, the composition of the food spectrum of juvenile coho salmon was more diverse. It was noted that in 2017, during the low-water period, greater species diversity was observed in the diet of fish, since then in abnormal years the diet of juvenile salmon contains random, forced food components (mollusks and fish).

Keywords: sockeye salmon, chum salmon, coho salmon, char, food spectrum, quality composition of food

5-1-2

# Predator cues increase negative effects of a similated marine heatwave on tropical zooplankton

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Extreme temperatures caused by ocean heatwaves are one of the major causes of biodiversity loss. However, when assessing the impact of extreme temperatures on natural populations, we often overlook biological interactions such as non-consumptive predator stress on prey species, which is considered to be “the core of ecology”. To address this issue, *Pseudodiaptomus incisus*, a predominant species in the coastal areas of Central Vietnam, was exposed to temperatures (30 and 34°C) with presence or absence of predator cues. Predator cues (kairomones) was obtained from barramundi larvae. The adult size, the clutch size, hatching success, the accumulated nauplii and fecal pellets production and the adult lifespan of *P. incisus* were assessed. Results showed that at 34°C, all fitness parameters reduced strongly, particularly in nauplii production and adult lifespan. The effects of temperature were stronger in the presence of predator cues, suggesting that the presence of predator cues can increase the vulnerability of *P. incisus* to extreme temperatures. Our results shed light on how predator stress is a major factor driving the susceptibility of marine prey species to extreme events, especially when marine heatwaves are increasing in number and severity under global climate change.

Keywords: biodiversity loss, marine heatwave, predator-prey interaction, synergy

# The drift of macroinvertebrate community and the effects of community and its adaption to the high flow in streams

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Macroinvertebrate drift behaviors play an important role in structuring and stabilizing the community composition and functions of stream ecosystems. In this study, we studied the temporal-spatial patterns of drifted macroinvertebrates in the totally natural streams of the Shennongjia Reserve based on the long-term monitoring and simulation in suit. And we also evaluated the compositions of drifted macroinvertebrates accounted for totally particle organic matters fluxes at different stream orders and different seasons. The results showed that: 1) Even so the most dominant taxa of macroinvertebrate changed at different seasons, the community structures of drifted macroinvertebrates were relatively stable. The differences of community compositions of different seasons were not significant. 2) The day light intensity plays the controlling role in the day-time drift patterns of macroinvertebrate, and the hydraulic condition of flow dominated the seasonal drift patterns of macroinvertebrate. 3) The active elusion behaviors were identified during the high flow after the rain. The drifted densities were significantly lower in the rain time. 4) The drift was a kind of continual and constant behavior of macroinvertebrates, however, the drift rates were significantly different among different taxa. The different “move-in” and “move-out” rate of each taxa would change the theory of neutral theory. According to our studies, we renewed the traditional theory of drift behavior of macroinvertebrates. The probabilities of catastrophic drift are small and may be mainly happened in the flooding period, human disturbances of habitat, or occasionally pollutions. This study confirmed the importance of drifted macroinvertebrate for the stabilities of longitudinal patterns of stream ecosystems. And our studies also promoted to integrate the neutral theory and river continuum concept at the watershed scale. This study also enhanced the multidisciplinary of stream ecology, hydrological ecology and ecohydraulics.

Keywords: drift, macroinvertebrate, high flow, neutral theory

5-2-1

Keynote Speaker

# Computational methods applied to analysis of individual and group behaviours of Zebra fish in laboratory conditions

**Tae-Soo CHON[1,2], Yong-Hyeok JANG[2], Hye-Won KIM[2,3], and Chunlei XIA[1,2]**

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Analysing individual and group movements is a basis for revealing origin of social behaviour of animals. Fish behavior has been widely adopted for unravelling social interactions, providing useful information in both theoretical and practical aspects. Automatic recognition and computational methods were developed for characterizing multi-individual interactions in laboratory conditions. Movements of Zebra fish (Cyprinidae, *Danio rerio*) in different genetic strains were continuously observed across days. Dynamic positioning within arena was analysed by movement parameters, data structure analysis, machine-learning and spatio-termporal clustering. Aggregation patterns, individual interactions and group activities were objectively characterized in different genetic strains across photo- and scoot-phases in formulating group activities. Usefulness of automatic monitoring of group movement is further discussed for monitoring and genetic functioning in fish populations.

Keywords: Fish behaviour, automatic movement detection, machine-learning, crowding, individual interaction

# Effects of land use on river water quality and benthic macroinvertebrate communities in Marinduque island, the Philippines

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Rivers and streams are among the most critical ecosystems. However, in many parts of the world, such monitoring is lacking, and ecological indicators have not been defined, particularly in the Philippines, where the country lacks an established aquatic bioindicator system to evaluate stream quality and preserve biotic integrity. This study evaluated river water quality and benthic macroinvertebrate communities in response to different land use types on Marinduque Island. Sixteen streams and wadeable rivers along a disturbance gradient related to land use (forested, agricultural, recreational, and mixed) were assessed. Water physiochemistry was collected, and BMI samples were identified up to family level. Biological metrics and water quality parameters were analyzed using multivariate and ordination analyses to evaluate each element's variation and to discriminate the number of parameters. Results from the analyses revealed that forested and agricultural sites were linked with higher water quality, while recreational and mixed sites significantly have poorer water quality, likely due to multiple stressors present such as increased channel alteration, pollution, and nutrient loading. Improved water quality conditions also reflect variations in BMI, as forested and agricultural sites achieved higher biological quality scores (ASPT, BMWP, and EPT-C Index) compared to recreational and mixed sites, suggesting the effects of water quality significantly affect BMI assemblage. The results support the use of BMIs as a cost-effective alternative in synergy with water physiochemistry that the province will use to address water resource conservation and sustainable management. Improved water quality monitoring based on bioindicators leads to better management of water resources.

Keywords: average score per taxon (ASPT), benthic macroinvertebrates (BMI), disturbance gradient, ecosystem health, land use, Marinduque island, tropical stream

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# Selenium biotransformation in marine ecosystems

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The content of selenium (Se) was determined by flameless atomic absorption spectrophotometry in organisms of different trophic levels (of Peter the Great Bay, Sea of Japan) – 21 species of seaweed and herbs, 27 species of bivalves, 4 species of echinoderms, 22 species of fish, bottom sediments, and also taking into account the background Se content in sea water. There are significant differences in the accumulation of Se by organisms of different trophic levels. It was found that the coefficients of accumulation values in the Primorsky Krai coastal water are high and reach 7000 at the first trophic level (water/algae).

Activity of microorganisms is a decisive factor in transformation of essential and, at the same time, toxic Se in marine waters. It has been shown that microorganisms that reduce selenates to selenites and then to metallic selenium and oxidizing selenides play an important role in the biogenic migration of Se.

The analysis of selenium biogenic migration in marine ecosystems requires an understanding of the role of organic sediments in the transfer of an element through the food chain.

Keywords: selenium, marine waters, selenium biotransformation, Sea of Japan

# Drivers of macroinvertebrate assemblages in littoral and pelagic habitats of a large shallow lake: Implications for bioassessment

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Different measures of macroinvertebrate diversity (species diversity, functional diversity, phylogenetic diversity) and functional feeding groups (FFGs) are commonly used to assess the ecological status of large shallow lakes. In this study, biodiversity and FFGs in a large shallow lake, Lake Hongze, differed between littoral and pelagic habitats, with the spatial variability of littoral assemblages more correlated with nutrient concentrations, whilst pelagic assemblages were more associated with wave-induced disturbance (e.g., mean effective fetch). Species diversity was poorly correlated with measures of nutrient concentrations, whereas functional diversity, phylogenetic diversity and functional feeding groups are promising indicators of nutrient concentrations. Multimetric indexes (MMI) were developed for both habitats, using historical data as reference for pelagic and least disturbed conditions for littoral. Key attributes for pelagic MMI included Biological Monitoring Working Party (BMWP), mollusca taxa, and the percentage of mollusca taxa, the percentage of filter-collector taxa, the percentage of predator taxa, the percentage of gather-collector taxa, the percentage of sensitive taxa and functional dispersion, while littoral MMI incorporated the percentage of scraper abundance, mollusca taxa, Biological Pollution Index and BMWP. The results of structural equation models (SEM) indicated that sediment organic matter strongly affected pelagic health, while chlorophyll a and transparency influenced littoral health due to varying littoral utilization. The study supports using biodiversity and FFGs to understand nutrient and disturbance effects on macroinvertebrates. Furthermore, monitoring both littoral and pelagic habitats in large shallow lakes is recommended for comprehensive ecosystem assessment.

Keywords: functional feeding groups, functional diversity, species diversity, phylogenetic diversity, biomonitoring, history reference, habitat variation

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# Spatio-temporal variation in 'green tide' causing *Ulva* communities between Jeju Island and the South Sea in Korea

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In recent years, "green tide" occurrences, caused by the proliferation of green macroalgae (genus *Ulva*), more and more severely and frequently took place worldwide, attributed to accelerating climate change and human activities. In Korea, while green macroalgal blooms have persisted along the northeastern coastlines of Jeju Island since the 2000s, they have also been sporadically observed on the southern coasts of the mainland of the Korean Peninsula. What *Ulva* species primarily account for these bloom incidences and spatio-temporal variation in *Ulva* community remain largely unknown. This study examined the *Ulva* community structure on Jeju Island and the South Sea from 2019 to 2021, by conducting comprehensive phylogenetic analysis on 966 specimens from 46 sites using chloroplast barcode *tufA* gene as a molecular marker. Additionally, nuclear 5s rDNA marker was analyzed for 90 *Ulva* specimens to identify the species within a LPP (*Ulva linza-prolifera-procera*) clade. A total of 9 *Ulva* species were found on Jeju Island, with *Ulva ohnoi* (43.7%) and *Ulva pertusa* (=*Ulva australis*; 29.3%) being the predominant species throughout the year. In the South Sea, *U. pertusa* (34.8%) and *U. linza* (18.9%) were the most dominant species, with *U. linza* absent during the autumn. Considerable spatio-temporal variation in *Ulva* community structure, including seasonal variation in species composition for both regions, were observed. The findings of this study will provide crucial genetic/taxonomic information on *Ulva* species in Korean coastal areas, which will aid in the development of management strategies to mitigate the negative impacts of green tides.

Keywords: Macroalgal blooms, *Ulva species*, DNA Barcoding, Species composition, Climate change

Acknowledgements: This research is supported by the project "Improvement of management strategies on marine disturbing and harmful organisms (No. 20190518)" and "Long-term change of structure and function in marine ecosystems of Korea" funded by the Ministry of Oceans and Fisheries, Korea.

# Microplastics transfer from larvae to adults of the order Trichoptera (Hydropsychidae, *Amphipsyche meridiana*)

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Microplastics (MPs) are omnipresent contaminants present in freshwater, marine, and terrestrial habitats. Aquatic species will inevitably ingest MPs due to their abundance in aquatic systems and pass them up the food chain. To date, however, no studies have been done to ascertain if MPs can be transferred during developmental life stages that make use of distinct surroundings. We explore that MPs in water can be transferred between *Amphipsyche meridiana* life phases, specifically to the adult flying stage. The findings of this study indicate that MPs can be transferred ontogenically from a feeding life stage (larva) of 0.1 MP item/ind. to a non-feeding life stage (pupa) of 0.64 MP item/ind., and lastly to the adult terrestrial life stage (0.13 MP item/ind.). MPs ranging in size from less than 100 μm to larger than 500 μm can readily be transferred into pupae and adult stages. The move of MPs to adults suggests a potential aerial avenue for contamination of the new habitat. Therefore, a living organism that feeds on freshwater insects during their terrestrial life stage could be impacted by MPs present in aquatic habitats.

Keywords: microplastics, development, life cycle, *Amphipsyche meridiana*, food chain

5-2-7

# How sediment dredging alters phosphorus dynamics in a lowland rural river?

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China’s lowland rural rivers are facing severe eutrophication problems due to excessive phosphorus (P) from anthropogenic activities. However, quantifying P dynamics in a low-land rural river is challenging. A P dynamic model (River-P) was specifically designed for lowland rural rivers to address this challenge. This model was coupled with the Environmental Fluid Dynamics Code and the Phosphorus Dynamic Model for lowland Polders to characterize P dynamics under the impact of dredging in a lowland rural river. Based on a two-year (2020–2021) dataset from a representative lowland rural river in the Lake Taihu Basin, China, the coupled model was calibrated and achieved a model performance of $R^2 > 0.59$, RMSE < 0.04 mg/L. Our research in the study river revealed that the time scale for the effectiveness of sediment dredging for P control was ~300 days, with an increase in P retention capacity by 74.8 kg/year and a decrease in TP concentrations of 23% after dredging. Dredging significantly reduced P release from sediment by 98%, while increased P resuspension and settling capacities by 16% and 46%, respectively. The sediment-water interface plays a critical role in P transfer within the river, as resuspension accounts for 16% of TP imports, and settling accounts for 47% of TP exports. Given the large P retention capacity of lowland rural rivers, drainage ditches and ponds with macrophytes are promising approaches to enhance P retention capacity. Our study provides valuable insights for local environmental departments, allowing a comprehensive understanding of P dynamics in lowland rural rivers.

Keywords: Lowland rural river, phosphorus, macrophytes, sediment dredging, retention capacity

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# Comparison of the lethal and sublethal effects of pristine and biofouled PLA microplastics on *Daphnia magna*

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Microplastics (MPs) pose significant ecological risks, and understanding their effects on aquatic organisms is critical. This study investigates the lethal and sublethal impacts of pristine and biofouled polylactic acid (PLA) microplastics on the freshwater crustacean, *Daphnia magna*. We conducted both chronic (17-day) and acute (48-hour) exposure experiments with juvenile (4 days old) D. magna to assess mortality and oxidative stress responses. In the chronic exposure at 5.0 mg/L, pristine PLA MPs resulted in 50% mortality among *D. magna*, whereas biofouled PLA MPs caused only 20% mortality, indicating that biofouling mitigates some of the lethal effects. Conversely, acute exposure to a high concentration of 40.0 mg/L for 48 hours showed no mortality in either pristine or biofouled conditions, suggesting that *D. magna* can tolerate short-term exposure to high doses of PLA MPs without immediate lethal effects. Sublethal impacts were assessed through oxidative stress markers: superoxide dismutase (SOD), catalase (CAT), and lipid peroxidation (LPO). The results showed distinct oxidative stress responses across the treatment groups. Elevated activities of SOD and CAT, along with increased LPO levels, were observed in *D. magna* exposed to pristine PLA MPs, indicating higher oxidative stress compared to those exposed to biofouled MPs. These findings imply that biofouling may reduce the toxicity of PLA MPs, possibly due to a biofilm layer that lessens the direct interaction of MPs with the daphnids. This research highlights the importance of considering the biological aging of MPs in ecotoxicological studies.

Keywords: Polylactic acid, Microplastic, *Daphnia magna*, Biofouled, Oxidative stress

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# Philippine watersheds in the anthropocene: Cases of southern urban hydrosystem syndrome and freshwater protected area

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In the Global South countries, like the Philippines, watersheds are severely impacted by multiple stressors associated with urban development and extreme climate under the Anthropocene. As such, this paper *a)* synthesizes current efforts in bioassessment, biomonitoring, conservation, and management of lotic and lentic ecosystems and *b)* evaluates the significance of Freshwater Protected Areas (FPAs) in preserving the watersheds and their associated biodiversity in the country. Emphasis is given to the observed manifestations of Southern Urban Hydrosystem Syndrome in the Philippines and how past biomonitoring studies (i.e. 2009-2023) have been helpful in providing clarity on the status of the country's freshwater ecosystems. Efforts applied in the protected areas in the country are usually concentrated on the conservation and management of terrestrial and marine habitats. If appropriately categorized and prioritized, 59% (i.e. 143/244) of current protected areas in the Philippines would benefit from adequately conserved and managed FPAs. This is an excellent opportunity in maximizing the conservation outcomes and benefits considering that this area (i.e., 2,648,573 ha) is 36% of the total coverage of protected sites in the country. Recent case studies in Marikina Watershed and Casecnan Protected Landscape have provided clarity on how FPAs may conserve the taxonomic and functional diversity of stream benthic macroinvertebrates despite the influence of anthropogenic activities (e.g., agricultural land use). The provided database and baseline information in this study may guide researchers and policy-makers in establishing a clear framework for assessment, monitoring, conservation, and management strategies for Philippine FPAs.

Keywords: aquatic insects, bioassessment, biomonitoring, EPTC, indigenous peoples, urban stream syndrome

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5-3-2

# Monitoring on migration birds in South Korea: the activities of Yeoldurudal ecological club in Korea University

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Korea University's club, Yeoldurudal, was founded in 2012 and has contributed to public awareness about the conservation of wetland ecosystems through various activities. In this presentation, we introduce our three monitoring efforts focused on migration birds in South Korea, which also have been featured in both media and academic field. Last winter, there was a mass appearance of mandarin ducks (Aix galericulata) living in Jungnangcheon in Seoul. However, through monitoring conducted by environmental media and Yeoldurudal, it's confirmed that this is a danger signal that mandarin ducks are flocking due to habitat loss. In the process of monitoring, we came to think about the problems with inappropriate media reporting and the direction of a proper birdwatching culture. Last winter in Incheon, we observed and recorded a Black-faced Spoonbill (*Platalea minor*), which does not typically overwinter in Korea, being adopted and moving with a flock of Spoonbills. Since South Korea is an important breeding ground for the Black-faced Spoonbill, monitoring and protecting their movements is crucial. Lastly, Yeoldurudal reached further beyond Korea to Mongolia to learn more about professional field activities. Through the trip, we were able to observe biodiversity of Eugii Lake in Mongolia and understand the importance of monitoring suspended molt in migratory birds. We are also trying to participate in cooperation among organizations from neighboring countries for monitoring migratory birds. Our club will continue to contribute to wetland conservation through migratory bird monitoring as citizen scientists.

Photos that have taken by ourselves, as well as video materials that would help understanding the ecology of the birds will be presented.

Keywords: birdwatching, citizen science, student activities, water birds, wetland ecosystem

# A preliminary study on food selectivity, the effects of diet on reproduction and larval survival rate of paradise fish *Macropodus opercularis* (Linneaus, 1758) in laboratory conditions

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The paradise fish *Macropodus opecularis* (Linneaus, 1758) shows potential as a biological mosquito control agent. Hence, researches regarding food preferences, the effects of different diets on their reproductivity as well as larval survivability are crucial information for field application. We first tested the food selectivity of 52 adult paradise fish of different standard length when they are fed with live preys: 4th instar *Aedes* larvae, *Tubifex* sp., *Daphnia* sp.; and also frozen *Chironomus* larvae and 4th instar *Aedes* larvae. Results showed that when being fed with the same amount of prey each type, fish of different sizes preferred live over dead preys, and exhibited strongest preference towards live 4th instar *Aedes* larvae. *M. opercularis* prefers visual over chemical stimuli while hunting. Four breeding pairs of *M. opercularis* were then fed with four different diets to test their effects on the reproductivity of the fish. The diet consisted of only live *Aedes* mosquito larvae demonstrated the best reproductive potential and recovery for both the parents as compared to the other types of diet used during the span of our experiment. The fish larvae were fed with three different types of food available in the lab, and the survival rate of fish larvae responsed best to a diet of freshly-hatched brine shrimps (*Artemia* sp.), then *Daphnia* sp., and lastly compressed food pellets. Our study has provided insights on the possible use of the paradise fish for controlling mosquito-borne diseases, and also give a preliminary understanding on effective rearing of *M. opercularis* in laboratory conditions.

Keywords: mosquito control, biocontrol, paradise fish, reproduction, survival rate

# The species diversity and observed biology of Deuterophlebiidae and Nymphomyiidae from China (Insecta: Diptera)

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Deuterophlebiidae and Nymphomyiidae are two monotypic families of Diptera, representing the most plesiomorphic clade of the order. Although extant 21 and 10 species of them have been described respectively before 2021, their diversity, biogeography and biology were still vague. Since 2021, we have collected consciously and hard in China to find their specimens. So far, seven new species of Deuterophlebiidae have been reported from the country, and a long series of others in both two families are still awaiting to describe. Biologically, parthenogenesis, sympatric distribution and dealation were observed in field collecting. Our materials and researches show China host much more diversity of those two families than we knew before, and because of morphological variation in adults, the pupal characters and molecular evidences seem more valuable in their taxonomy.

Keywords: mountain midge, net-winged midge, torrent, origin, diversity

# Comparing the health assessment index of benthic macroinvertebrates and vegetation communities in wetland ecosystems

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Wetlands are unique ecosystems where the soil surface is either permanently or temporarily covered by freshwater, brackish water, or saltwater, consisting diverse habitats. Within these ecosystems, various organisms interact based on their unique ecological niches. Wetland vegetation found predominantly in riparian zones and benthic macroinvertebrates are representing diverse habitat functions. Assessing wetland ecosystem health relies on both groups, yet research on their interactions and the correlation between assessment indicators is pronunciation. Thus, this study aims to elucidate this relationship using health assessment indices. Field surveys, guided by the Wetland Environment Survey Guidelines (2017), targeted 15 wetlands in Korea. Health assessments employed the Korea Wetland Benthic Macroinvertebrate Index (KWBMI) for benthic macroinvertebrates, which includes indicators such as community structure, tolerance taxa, characteristic taxa, and functional diversity. Wetland vegetation health was evaluated using the Wetland Vegetation Index (WVI), considering community number, landscape structure grade, and species diversity. Correlation analyses were conducted between health assessment metrics and water quality parameters to validate associations. A correlation analysis was conducted on indicators of KWBMI and WVI, revealing a correlation between the Chironomidae spp. Individual density and the landscape structure of wetland vegetation, as well as the total score. Positive correlation was observed in the 2023 spring survey, while negative correlation was observed in the autumn survey of 2023. However, when the surveys (autumn and spring) were analyzed totally, a negative correlation was confirmed. In this study, although a distinct correlation was not identified, it was observed that wetlands with rich riparian vegetation tended to have higher diversity of benthic macroinvertebrates community.

Keywords: Wetland, Benthic Macroinvertebrate, Vegetation, Assessment index

# Back to the idea of creating the international rapid bioassessment protocols for streams and rivers in the ENEA region: The path through public biomonitoring

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Unification of freshwater biomonitoring systems is important for East Asian countries. The USA, European countries, Australia and some Asian regions such as Hindu Kush-Himalayan Region have established own bioassessment systems by XXI century. East Asian countries are still on the way. For Russian Far East (RFE) creating of international rapid bioassessment protocols is the important task, because the RFE as well as adjacent countries are developing very quickly last years and bring serious impacts to the environment. The idea of combining initiatives to create a unified freshwater bioassessment system in East Asia firstly was proposed by a group of scientists under leading of Dr. J.C. Morse in 2007. Then it was discussed at the 1st BSA Symposium in Matsumoto (Japan) in 2011. However, due to different complexities, this idea did not reach the necessary development. Nevertheless, this call led to the emergence of several international scientific projects connected with this issue. Although the idea never received proper development at the state and academic levels, it began to be implemented within the international programs aimed at developing public environmental initiatives. Thus, since 2022, the program “Biomonitoring of Freshwater Ecosystems” initiated by Environmental Subcommittee of the Association of Regional Administrations of Northeast Asian Countries was started. Express bioassessment protocols were developed for beginners (students and schoolchildren) and were used in some ENEA countries in 2022–2023. The result have showed that specially trained youth are able to assess the stream quality at a high level, comparable to the work of professionals.

Keywords: public freshwater biomonitoring, Rapid Bioassessment Protocols, running waters

# The structure of bottom communities as an indicator of the small stream ecological state (based on examples of Primorye streams, Russian Far East)

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Problems of conservation of freshwater resources require a well-developed system of monitoring and control from both the state and public sides. This is especially true for small water bodies, which are the most defenseless in the face of growing urbanization. This problem can be solved with widespread public involvement in saving small rivers. In the Primorsky Territory, the movement to save small reservoirs and study their ecological state using bioindication methods is becoming increasingly popular. To this end, scientific institutes of the Far Eastern Branch of the Russian Academy of Sciences and the Scientific & Public Coordination Center "Clean Water Center" are developing methodological guidelines and manuals for public experts – simple, but adequately reflecting the ecological state of streams. Since 2020, we have been included in a group of schoolchildren and students to assess the ecological state of small rivers in Primorye based on macrozoobenthos under the auspices of the NGO "Clean Water Center".

Goal: To study the composition and structure of benthic communities of macrozoobenthos in 6 streams of the Primorsky Territory, located in riverbeds with varying degrees of anthropogenic impact, in order to identify trends in changes in communities during environmental disturbances.

Objectives:

1. To identify the taxonomic composition of macrozoobenthos in selected watercourses.
2. Describe the species structure of benthic communities based on abundance indicators.
3. Analyze the main metrics of biodiversity and calculate biotic indices in 6 studied areas of selected watercourses.
4. Determine the water quality of the studied sites based on aquatic invertebrates, and identify patterns of changes in the bottom community structures under anthropogenic impact.

Results.

1. The taxonomic composition of macrozoobenthos has been revealed: the list includes 42 species from 23 genera, 22 families, 11 orders and 5 types of animals.
2. The species and trophic structure of macrozoobenthos communities is described and 3 types of communities formed under different levels of anthropogenic impact are identified; a) clean water communities (rivers located into or at the protected areas); b) communities of clean streams, but with mechanical disturbances of the channel and deforestation (small streams of the All-Russian Children's Center "Ocean", here hydrochemical indicators were normal, but the structure of the communities was sharply different from the natural one (in comparison with the situation before deforestation); c) communities of polluted waters in conditions of chronic pollution and deforestation cover (the Second Rechka River, located in the city center and has been in a zone of strong anthropogenic impact from many sources of pollution for more than 80 years).
3. Water quality was assessed using 9 biotic indices: Goodnight-Whitley, Woodiwiss, EPT, HT (High Taxa Index), FBI (Family Biotic Index), BMWP (FE Biological Monitoring Working Party Index), FE ASPT (Average Score Taxon Index), SO (Sensitive Organism Index), FE PSI (Far East Proportion of Sediment-Sensitive Invertebrates Index).
4. It has been shown that the deforestation and mechanical disturbance of the stream channel in the water protection zone causes fundamental changes in the structure of benthic communities: in our example, before deforestation, crustacean *Gammarus koreanus* Ueno, 1940 dominated (shredders), which is typical for undisturbed south far eastern forest streams; and after deforestation, dipterans Chironomidae (collector-gatherers) and Simuliidae (collector-filterers) became dominants.

# Water quality profiles at Caldera Lakes in Bali – Indonesia

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Lake Buyan and Lake Tamblingan are located at an altitude of 1,200 m above sea level in the central part of Bali Island and have the potential to be developed for the tourism industry as well as being a residential and agricultural area with fertile land. Landscapes with such basins are called “endorheic basins” or closed lake basins. The history of the formation of those lakes is a powerful eruption that destroyed most of the ancient volcanic peak and left only the slopes of the mountain which now forms an oval-shaped caldera with a diameter of 12 × 7 km. The objective of this study is to analyze the trend of water quality of caldera tropical lakes represented by Lake Buyan and Lake Tamblingan related to their land use changes. Lake Buyan area is 3.67 km$^2$ with a maximum depth of 79.5 m. Meanwhile, Lake Tamblingan has an area of 1.15 km$^2$ with a maximum depth of 40 m. The average depth for Lake Buyan and Lake Tamblingan lakes are 31.7 m and 23.5 m, respectively. The water surface height in both lakes is 1,214 m above sea level. A survey at both lakes was conducted in 2012 and 2023. The results showed that the degradation of water quality followed the land use changes. It could be explained that the activities surrounding both lakes influenced the water quality conditions. It suggested also that the potential impact of agriculture and tourism contributed to pollutant concentration in those lakes. To maintain the sustainability of the lakes, basic ecological information is necessary for the next study.

Keywords: Water quality, Lake Buyan, Lake Tamblingan, Caldera, Basin

# Preliminary study of hydrodynamics of Saguling resevoir, West Java Indonesia using Water Ecology Tools (WET)

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Saguling reservoir is one of cascade reservoir that have function to produce electricity for Java and Bali. In addition, this reservoir is also used for fish cage culture, sometime the phenomenon of fish mortality was often. For this reason, it is necessary to study hydrodynamics characteristic of Saguling river to explain fish mortality phenomenon. Therefore, this research aims to determine the temporal pattern of changes in water column temperature, and its relationship with the incidence of fish mortality in floating net cage. This study uses the Water Ecology Tools model (1-dimension) as tools to analyse hydrodynamics process in water column especially temperature process. In addition, we used data recorded using water level sensors and CTD Rinko ASTD102 instrument. This research was conducted from December 2021 to October 2022 in Saguling reservoir, West Java Province, Indonesia. The result show that there were as isothermal phenomena in water column that explain the mixing process and made the water mass from the bottom that lack oxygen rise to the top, and causes fish mortality. For the future research, it possible to use 3-dimension model to better understand the hydrodynamic process.

Keywords: Hydrodynamic, Reservoir, 1-Dimensional model

# Mosquito flight behavior based on time after capture within an experimental chamber

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The world's deadliest animals listed in WHO are mosquitoes of the order Diptera. The mosquito family Culicidae is well known as vectors of dengue fever, Japanese encephalitis (JEV), malaria and Zika virus. Yet, control of these vector has always posed a challenge. Contemporarily, new methods of identification of mosquitoes utilizing animal acoustics and artificial intelligence (AI) have been discussed extensively. Yet the task of detecting mosquitoes from the sound of their wing beats is challenging due to the difficulty in collecting and standardizing such recordings in the field. To address this, experiments regarding mosquito flight behaviors within a prototype monitoring chamber would aid in improving methods for future mosquito monitoring and identification. This study aims to establish a general linear model (GLM) relationship between mosquito flight duration and time period after capture based on recordings of mosquitoes in free flight, which would serve as preliminary data for structuring future monitoring methods. For this study, a total of 24 hours of mosquito flight audio recordings have been labeled from eight mosquito species (*Aedes albopictus, Ae. togoi, Ae. aegypti Culex pipiens, Cx. tritaeniorhynchus, Anopheles cracens, An. minimus, An. sinensis*). The result suggest that the species have differing tendencies of flight behavior once captured and that individual capture is not sufficient for the acoustic monitoring method.

Keywords: Acoustics, *Aedes albopictus*, artificial intelligence, mosquito flight behavior, vector monitoring

Acknowledgements: This work was supported by the Institute for Information & Communications Technology Promotion (IITP) grant funded by the Korea government (MSIT) (No.RS-2023-00262750).

# Forgotten predators: The untold story of *Mesocyclops* in Vietnam's battle against Dengue mosquitoes

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Biological control using *Mesocycles*' as predators of *Aedes aegypti* larvae was proved as an effective and eco-friendly strategy and offers the possibility of functioning without the frequently repeated applications necessary for pesticides. However, the method did not receive serious consideration when the chemical control strategy collapsed in recent decades in Vietnam. Mosquito-control professionals and public health officials relied heavily on chemical pesticides because of its convenience and quick response. Since the resistance of *Aedes aegypti* for arrays of pesticides in Vietnam, the pesticide-based strategy of the last thirty years has been a failure. As a result, ecological management promises to be more effective and sustainable and the copepod *Mesocyclops* provides a great example. In this research, we reviewed the history of using *Mesocyclops* in Vietnam to find the key factors leading to success and to discuss what have not been studied yet. We also quantify the predatory impact of the most abundant *Mesocyclops* species named *Mesocyclops thermocyclopoides* towards four different larval instars of *Aedes aegypti* prey across a water depth gradient. Consumption rates were interspecific differences in responses to variations in water depth. Adult *Mesocyclops* were able to handle *Ae*. *aegypti* prey across all instar stages, yet predation rates were remarkably higher towards early as opposed to late instar prey. Further, efficient predation rates of this species were most pronounced in medium water collum compared to shallow and deep. Our results further demonstrate predatory impacts of *Mesocyclops* predators towards Dengue mosquitoes and quantify biotic and abiotic context-dependencies which modulate their impact. The research findings on the effectiveness of *Mesocyclops* predators and their impact on *Aedes aegypti* larvae provide valuable insights into sustainable and eco-friendly strategies for controlling mosquito populations.

Keywords: *Mesocyclops, Aedes aegypti*, biological control, Vietnam

# Occurrence of predacious mosquito species in association with mosquito larvae in residential areas of Chiang Mai, Thailand

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Mosquitoes transmit the deadliest diseases to humans and appear to increase their survival rate by easily adapting to rapid changes in climate and the environment. *Toxorhynchites* mosquitoes, natural predators of vector mosquitoes, possess a unique characteristic: their larval stage consumes other mosquito larvae, while their adult stage is non-hematophagous. This highlights the critical role of *Toxorhynchites* as a biocontrol agent capable of controlling mosquito. To better understand the relationships between mosquito larvae and *Toxorhynchites* larvae in residential areas of Chiang Mai, Thailand, the densities of *Toxorhynchites* and mosquito larvae were observed at three suburban and three urban study sites during both the wet season (October to November 2023) and the dry season (December 2023 to February 2024). As a result, the number of *Toxorhynchites* larvae and other mosquito larvae was higher in the wet season than in the dry season. Moreover, the abundance of *Toxorhynchites* and mosquito larvae was observed to be higher in urban study sites than in suburban study sites. Additionally, the abundance of artificial mosquito habitats was observed in urban areas, whereas the abundance of natural mosquito habitats was observed in suburban areas. Only in urban mosquito habitats during the wet season, the occurrence of *Toxorhynchites* larvae was negatively correlated with mosquito larvae significantly, while the correlations were insignificant in other cases. These results imply the possibility of using *Toxorhynchites* species as biocontrol agents in the regions, especially during the rainy season.

Keywords: biological mosquito control, Chiang Mai, predaceous mosquito, Thailand, *Toxorhynchites*, vector mosquito

Acknowledgements: We thank the Korea Disease Control and Prevention Agency (KDCA) for the KDCA-DVP 2024 Grant Program (R67EX00317) and for partial support from Chiang Mai University (R000032014).

# Preference assessment of new mosquito attractants in varied outdoor environmental conditions

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This study aimed to develop effective attractants to enhance the eco-friendly physical control of disease-vector mosquitoes. The experiment was conducted from January 9 to March 10, 2024, across residential, waterfront, and forested areas in Mueang Chiang Mai District, Chiang Mai Province, Thailand. Six key mosquito breeding sites were selected in each area. Five attractants were tested: two commercially available ones (A and B) and three newly synthesized ones (Type 1, Type 2, and Type 3). A control group used no attractant. Different traps were used for each attractant, with BG-Sentinel traps (BG trap, Biogent) employed for mosquito collection. To exclude the influence of study sites on attractant efficacy, traps were rotated clockwise among sites every 24 hours. Mosquitoes were collected over six days per attractant at each site, repeated three times. Collected specimens were sorted into mosquitoes and other species, and counted. Results showed that attractant A had the highest average daily collection rate at 27.39 ± 2.37%, followed by Type 1 at 21.81 ± 2.34%, attractant B at 18.22 ± 1.73%, Type 2 at 17.58 ± 2.04%, and Type 3 at 15.01 ± 1.73%. Statistical analysis revealed no significant difference between the collection rates of the existing attractant A and the newly synthesized attractant Type 1, both demonstrating higher capture rates compared to the other attractants.

Keywords: Mosquito, Attractant, Compound, Preference, Physical control, Eco-friendly Control

# Characterization of overwintering habitat of *Haemaphysalis longicornis* (Acari: Ixodidae), a major vector of severe fever with thrombocytopenia syndrome (SFTS) virus

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Ixodid ticks are the global vectors of pathogens causing various tick-borne diseases, such as babesiosis and Lyme disease. In particular, *Haemaphysalis longicornis* (Acari: Ixodidae) is a major vector of the severe fever with thrombocytopenia syndrome (SFTS) virus, which has been newly reported as a fatal pathogen in East Asia. Although empirical data are scant, understanding the overwintering ecology of *H. longicornis* is crucial for the effective vector management. In this study, a nationwide field survey was conducted across eight different provinces in South Korea to characterize the overwintering habitats of *H. longicornis* and their harboring rates of the SFTS virus. In each province, four different landscapes were surveyed: grassland, shrub, coniferous forest, and deciduous forest. A total of 679 samples were collected from ground (leaf litter, soil surface, and topsoil layer), rocks, and dead trees. From the survey, 85 overwintering ixodid ticks were collected, and *H. longicornis* was the most dominant species (93%). Among the collected *H. longicornis*, the nymphal stage was the main developmental stage of overwintering (94%), and the ticks were exclusively found from ground samples, especially from the top soil layer (77%). In addition, *H. longicornis* were mainly found from herbaceous landscapes, such as grassland (43%) and shrub (54%). Finally, among the 49 tick pools tested for the SFTS virus, the three positive pools were detected from nymphal *H. longicornis* yielding the minimum infection rate as 4%. The results of this study provide baseline information for the development of management programs for the vectors of tick-borne diseases.

Keywords: hard tick, longhorned tick, tick surveillance, vector biology, overwintering biology

# Functional analysis of predatory mosquito larvae, *Lutzia vorax* (Diptera: Culicidae) in South Korea

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Larvae of genus *Lutzia* Theobald (Diptera: Culicidae) are ferocious predator of container habitats in forest areas. Due to their consumption of various invertebrates including mosquito larvae, they are important main predators of container habitat and potential biological agents of mosquitoes. Two species, *L. vorax* Edwards and *L. fuscana* (Wiedemann) have been reported in South Korea which are widely distributed species in Oriental, east Palearctic regions, and Australia. However, regards of *L. vorax* of South Korea, it is often erroneous recorded as related species, *L. halifaxii* Theobald in some literatures and catalogues. Also, general biology and ecology of *L. vorax* have been barely studied, although it is common species, found broadly throughout South Korea. In this study, comprehensive identification of larvae of *L. vorax* using cytochrome c oxidase subunit I barcoding and morphological examination were conducted to confirm the distribution of *L. vorax* in South Korea. Also, seasonal occurrence pattern of *L. vorax* larvae in South Korea is reported for the first time, through field monitoring surveys in three different deciduous forests (Gangneung, Gangwon-do; Hanam, Gyeonggi-do; Pocheon, Gyeonggi-do) using tire larvae-traps in 2023. According to community structure analysis of the monitoring data, using permutational analysis of variance (PERMANOVA) and non-metric multi-dimensional scaling (NMDS), presence of *L. vorax* larvae is significant factors (P value <0.05) for mosquito larvae community structure in the three sites, suggesting that *L. vorax* may be valuable as a both potential mosquito control agent and key predator species of container habitat in forest areas of South Korea.

Keywords: *Lutzia*, Culicidae, mosquito predator, community ecology, DNA barcoding

Acknowledgements: This work was supported by the National Research Foundation of Korea (NRF) grant funded by the Korean Government (MSIT) (2022R1A2C1009024).

# The effect of $PM_{2.5}$ solutions on *Aedes aegypti* mosquito survivorship

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While $PM_{2.5}$ exposure is known to induce adverse effects in environmental organisms, the variation in toxicity in particular vector mosquito associated with differing $PM_{2.5}$ concentrations remains poorly understood. To investigate the impact of concentration on $PM_{2.5}$ ecotoxicity, the present study employed seven $PM_{2.5}$ concentrations ranging from 12.5 to 150 mg/L, alongside control groups of deionized (DI) water and rainwater. These exposures were conducted on two insecticide-resistant strains of *Aedes aegypti* (PMD and PMD-R), as well as a field-collected strain (LML) of *Ae. aegypti*, all subjected to the same conditions of $PM_{2.5}$ exposure. The physiological aspects, specifically survivorship, as well as chemical parameters including ion solubility, pH, and conductivity, were examined. Exposure to the highest concentration of $PM_{2.5}$ at 150 mg/L led to notable adverse impacts on larval survival rates, with reductions of 55.6%, 42.2% and 50.0% for PMD, PMD-R, and LML, respectively. Additionally, pupal survival rates experienced similar adverse effects, demonstrating reductions of 50.0%, 40.0%, and 42.2% for PMD, PMD-R, and LML, respectively. Notably, PMD exhibited the highest survival rate, followed by LML. PMD-R displayed the lowest survival rate among the investigated species. According to the findings, it was observed that the concentration of major ionic species ($SO_4^{2-}$, $NO_3^-$, and $NH_4^+$) exhibited a positive correlation with increasing $PM_{2.5}$ concentration. Moreover, a concurrent increase in acidity of pH levels and electrical conductivity was noted with escalating $PM_{2.5}$ levels. The study revealed a negative correlation between $PM_{2.5}$ concentration and survival rates across all *Ae. aegypti* mosquito.

Keywords: *Aedes aegypti*, Survivorship, $PM_{2.5}$

# A multidisciplinary approach for two morphotypes of *Aedes koreicus* (Diptera: Culicidae) in South Korea

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Concerning the role as vectors of diseases such as dengue fever, the genus *Aedes* Meigen, 1818 stands out as the notable genera within the family Culicidae. Recent attention has been drawn to *Aedes koreicus* Edwards, 1917, along with its sibling species *Aedes japonicus* Theobald, 1901 because of their invasion into Europe and the Nearctics. In South Korean *Ae. koreicus*, there is a Jeju form found only on Jeju Island, and a Korean Peninsula form found throughout the rest of the country and research on these two morphotypes remains unexplored within the native country. Along several morphological differences, the color of hind tarsomere V scales is used as criteria for form classification and tracking invasion routes. Considering that the key character between *Ae. koreicus* and *Ae. japonicus* is the color of hind tarsomere IV scales, a morphological examination of the two morphotypes of *Ae. koreicus* and *Ae. japonicus* was conducted in this study, along with molecular analysis using COI and ND4 molecular markers. The results revealed that the two morphotypes of *Ae. koreicus* are not based on regional populations, and the known morphological differences are merely variations within the species. Additionally, through analysis of the wingbeat sounds of the two species was conducted, and the average frequency and frequency range within each species showed sexual differences. The wingbeat data of two species are presented for the first time and hold potential application in sound-based species identification, trap or monitoring systems.

Keywords: mosquitoes, *Ochlerotatus*, variation, invasive species, sibling species

# IV. Symposium

- **Symposium 1** (NIBR)

  **July 2, 2024 | 13:30-15:20**

  Current status and management methods of outbreak organisms

- **Symposium 2** (NNIBR)

  **July 2, 2024 | 15:30-17:20**

  Biological Collections in the Digital Age: Challenges and Opportunities

- **Symposium 3** (Dongsung Biopharm and Sahmyook University)

  **July 3, 2024 | 10:00-11:50**

  Safe and healthy water resource acquisition and management

- **Symposium 4** (KDCA)

  **July 3, 2024 | 13:30-15:20**

  Strategies for various disease vector management

- **Symposium 5** (OJERI)

  **July 3, 2024 | 15:30-17:30**

  Optimizing water, food, energy, and ecosystem nexus in the 3rd pole region of the Mid Latitude Region

# Synchronized mass emergence and parthenogenesis of the polymitarcyid mayfly, *Ephoron shigae*

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*Ephoron shigae*, a polymitarcyid mayfly, is widespread in Far East Asia and causes synchronized mass emergence in Japan. The mass emergence is observed during a few days in early fall, and positive phototaxis causes large numbers of individuals to swarm to streetlights. Their individuals and carcasses accumulate under streetlights on riverbanks and bridges, often causing car accidents and media attention. Although the emergence have been observed throughout Japan since the late 1970s, one of the older emergence is described in detail in the literary work “Honen-mushi,” written by Naoya Shiga in 1928. The adult life span is very short, up to 2 hours, during which time the female mates and lays eggs. The emergence period is concentrated in a short period in September, and the mass emergence is probably related to the unique nature of this mayfly.

In addition, all-female parthenogenetic reproduction is known only in Japanese *E. shigae*, in which bisexual and unisexual populations are distributed throughout Japan with no clear biogeographic boundaries. Population genetic structure studies show that the parthenogenetic origin occurred only once in western Japan and then spread throughout Japan. In some rivers in eastern Japan, both reproductive types coexist, and parthenogenetic females occupied the population within 20 years after the unisexual introduction. Furthermore, slight differences in emergence times between both reproductive types have been found.

Keywords: geographic parthenogenesis, reproduction, molecular phylogeny, river, Ephemeroptera

# Lovebug outbreaks: Finding patterns of dispersal and adaptation

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Lovebugs (Diptera: Bibionidae: *Plecia*), named for their behavior of flying in copulation, can become serious nuisance pests when massive swarms occur in the living space of humans. A well-known example is the case of *P. nearctica*, with excessive outbreaks and dispersal along the US Gulf Coast recorded since the mid-20th century. Recently, the newly introduced *P. longiforceps* in the Seoul Metropolitan Area of Korea is drawing public attention, as intensive swarming in residential and commercial areas raise concerns on their risk on public health and the environment; however, control measures are limited due to the lack of comprehension on the ecology, threats, and origin of this species. To address the demand for biological information on this urban pest, we summarize previous and current investigations on lovebug outbreaks, with emphasis on patterns of range expansion and establishment in urban habitats. The pest status and outbreak history of *P. nearctica* are reviewed, and experimental studies on swarming behavior are briefly revisited. We introduce our ongoing research on *P. longiforceps*, including systematics, population genomics, and ecological observations. Through the combination of novel information based on up-to-date techniques with insight from a matching history, we hope our findings contribute to a better understanding of outbreak phenomena in urban environments, and provide fundamental data for effective pest management.

Keywords: march flies, *Plecia, Plecia longiforceps*, population genomics, range expansion, urban pest

Acknowledgements: This work was supported by a grant from the National Institute of Biological Resource (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202410201).

# Research on factors to hypertrephied of *Metcalfa pruinose* (Hemiptera: Flatidae)

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The *Metcalfa pruinosa* was first discovered in 2009 and has since spread nationwide after 2011 in Korea. With a broad host range encompassing 74 plant species, including black locust, grapevine, and apple trees, this insect inflicts damage on crops by feeding on both nymph and adult stages, resulting in deterioration of crop quality and the development of sooty mold due to honeydew secretion. Their phototactic behavior leads to frequent occurrences even in well-lit urban areas, and coupled with various contributing factors, the population density is on the rise, heightening the risk of hypertrephied. To address this pressing issue, research efforts aim to investigate the preferred environmental conditions such as temperature, humidity, and altitude, along with identifying host plant and Presence of natural enemies. Additionally, we aim to elucidate the correlation through genetic analysis among populations of the M. pruinose.

Keywords: invasive species, pest management, agricultural impact, population getentics

Acknowledgements: This work was supported by a grant from the National Institute of Biological Resource (NIBR), funded by Ministry of Environment (MOE) of the Republic of Korea (NIBR202410201).

# Outbreak of walking-stick insects, *Ramulus mikado* (Rehn & J.A.G.) (Insecta: Phasmida), in temperate forests in relation with environmental variables

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The walking-stick insects, Ramulus mikado, are outbreaks in several mountainous areas in central Korea since 2020, but knowledges about outbreak pattern of the insects is still lacking. In this study, we studied the host tree preference in natural conditions, the ecological stoichiometry of major host trees, the hatch rate of overwintered eggs, and the infection rate of R. mikado by entomophathogenic fungi during growing season. In addition, we analyzed the genetic divergence of their populations in COI gene. The population density of the insects was steadily declined from 2022 to 2024 due to environmental changes in addition to the pest management program by local governments. In 2022, the host tree preference in natural conditions was change according to seasonal changes and the developmental change of the insects but following two years did not showed specific host tree preferences. In terms of the ecological stoichiometry of major host trees, the ratio of nitrogen in leaves was significantly higher in Robinia pseudoacacia rather than Prunus serrulate var. pubescens and Quercus mongolica. The hatch rate of overwintered eggs of R. mikado was significantly declined with elevation increased in both 2023 and 2024. The infection rate of R. mikado adults by entomophathogenic fungi, Metarhizium phasmatodea, during 2022 to 2023 was rapidly increased since rainy season started and thus most adult insects were died. The genetic diversity showed that the R. mikado population at the Mt. Cheonggye Ridge #1 has had the greatest impact on the formation of the other populations at the lower elevation of the Mt. Cheonggye area and the nearby Mt. Suri and Mt. Geumam populations. In conclusion, we found relationships between the outbreak of R. mikado and various environmental variables, such as host characteristics, winter temperature, and relative humidity during summer season.

Keywords: phasmids, forest insect pests, plant-insect relationship, plant nutrition, entomophathogenic fungi, genetic divergence

Acknowledgements: This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202405104).

# Habitat preference of *Ephemera orientalis* (Ephemeroptera: Ephemeridae) larvae in Hangang River

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The outbreak intensity of *E. orientalis* in the Hangang River is increasing, and the need for research on the ecological characteristics of *E. orientalis* larvae inhabiting the Hangang River is also growing. This study investigated the habitat preferences of *E. orientalis* larvae in the Hangang River through quantitative analysis and comparison of their number of larvae, water depth, and substrate types. Samples were collected in April 2024 at 10 different sites between Paldang Bridge and Hannam Bridge. At each site, the survey was repeated three times at five locations: Left bank, Left center, Center, Right center, Right bank. Larvae and substrate were collected using a surber net (50cm × 50cm), and the substrate was categorized into 9 different particle sizes using a sieve (64, 31.5, 16, 8, 4, 2, 1, 0.5mm). The number of larvae, water depth, and weight of substrate by particle size were compared with correlation analysis. Where composed only of substrate with particle sizes ranging from 0 to 0.5mm, showed low number of larvae. The number of larvae showed a positive correlation with substrate diversity and water depth. Additionally, the riparian zone showed low substrate diversity and the number of larvae, whereas the central zone showed high substrate diversity and the number of larvae. These results suggest that *E. orientalis* larvae prefer substrate of various particle sizes, indicating a preference for inhabiting the central zone of the Hangang River where the substrate diversity is higher compared to the riparian zone.

Keywords: *Ephemera orientalis*, Substrate, Hangang River, Habitat preference

S2-1

Keynote Speaker

# Digital conservation and utilization of biological collections: Focusing on the NNIBR cases

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Biological specimen collections require permanent preservation as tangible evidence of the morphology, genetic traits, and distribution records of organisms that existed at specific times and locations. Also, Specimen data are essential for analyzing shifts in freshwater species and community changes due to climate change, necessitating their continuous collection and management. Recently, many biological specimen repositories, including leading natural history museums worldwide, have been pursuing the digitization of specimens to achieve the permanent preservation of morphological data and to establish data portals for global access and information sharing.

The Nakdonggang National Institute of Biological Resources (NNIBR), a freshwater biological research institution in Korea, manages seven specimen storage facilities. These facilities are organized by specimen type and are equipped with constant temperature and humidity controls as well as specialized fire extinguisher systems, capable of holding over 6 million specimens. Currently, they house a total of 544,560 specimens. Since 2021, NNIBR has been developing digital imaging technologies suitable for various biological taxa and has established a photography studio for specimen digitization. To date, using various equipment such as microscopes, DSLR cameras, and X-ray CT, it has captured and archived high-resolution multifocal 2D images and 3D digital images of 965 species of freshwater organisms. Additionally, NNIBR has created various digital content from these images for research, exhibition, education, and outreach, making them available online for public access and sharing.

Keywords: Biological collections, specimen digitization, multifocal 2D images, X-ray CT, digital contents

# Beyond technology: Perseverance and imagination as pillars of success in microtomography

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Over the past 14 years, I have undergone a significant change in my scientific career, becoming completely enamoured with the possibilities of computed tomography (micro-CT). Not only has it allowed me to conduct anatomical studies, but it has also enabled me to answer several questions. I have been particularly drawn to the intrinsic beauty of the images I have obtained, which has led me to dedicate countless hours, almost obsessively, to improving the results.

Initially, the process was very disappointing, as I could barely obtain images with minimal detail, blurry, and far from what I had envisioned with my entomological background. Gradually, however, I discovered small but impactful tricks in sample preparation, scanning, and software use, which transformed those initial struggles into achievements that surpassed my initial dreams.

Over the years, I have received numerous inquiries and requests to improve the scans of colleagues from around the world, many of whom have access to much more sophisticated technology than I possess. However, I have found that the key is not to have the latest equipment but to know how to handle it and the associated software effectively. This requires perseverance and imagination to propose new challenges and achieve successful results.

In this brief talk, I will share my experiences and insights gained from completing the presentation scheduled for Plenary Session 1.

Keywords: Micro-CT, disappointments, tricks, attitudes, success

Acknowledgments: We thank the staff of Bruker SkyScan in Kontich (Belgium) for their effectiveness, fast support, constant improvements to the software, and implementation of the requested new features. In this respect, we are especially indebted to Alexander Sasov (now at NeoScan www.neoscan.com), Stephan Boons, Xuan Liu, Phil Salmon, and Vladimir Kharitonov.

# Status and further direction of biological specimen digitization

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Biological specimens have been actively manufactured since the 17th century and has had a great influence on the establishment and development of biology, especially taxonomy. Recently, biological specimens are being used as basic materials in various scientific fields. Digitized specimens can break down the physical barriers to specimens access and can be a countermeasure against the risk of real specimens loss. Furthermore, they are usable for materials of the research, the education, and the digital content business. In the case of the London Natural History Museum, which calculated the value obtained by digitizing biological specimens, they are estimated that it will create £2 billion. DiSSCo is one of the largest projects of biological specimen digitization and more than 170 institutions from 23 European countries are involved. Through this project 73 million specimens have been digitized. iDigBio, which involves universities and natural history museums across the U.S.A, links more than 130 million records of specimens with 53 million media information. These projects allow us to understand trends in digitization technologies and data distribution systems. In Korea, it is estimated that the number of specimens held by major institutions is about 5 million and digitization is approximately 10 percent. For the efficient digitization and use of the products, several things should be considered. One is the application of high-quality standards that take into account technological advancements. Another is to establish IT platform that supports information integration and distribution. In addition, continuous financial resources, training of digitization personnel, and information standardization are necessary.

Keywords: biology, digitization, natural history, specimen, taxonomy

# The case of the National Science Museum of Korea's digital archive in Natural History

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The National Science Museum of Korea conducts the science and culture promotion through collection, preservation, research, exhibition, and education based on scientific collections. As of 2024, the museum has more than 800,000 specimens includes plants, animals and other taxonomic groups, which are preserved and managed in 15 storages (1,370.1 $m^2$) under constant temperature and humidity.

The digital archives in the field of natural history at the National Science Museum can be broadly divided into 2D images and 3D printed data. There are a total of 55,561 2D image data (150,617 photos) of plants, insects, invertebrates, and other taxonomic groups, accounting for about 6.9% of the total, and 2,717 3D printed data. In addition to the primary data that uses the images themselves, 2D data is also available as a secondary data that excavates the stories contained in the collection and provides them as photo and video stories. In addition, 3D printing data is produced in various file formats such as jpg, ply, stl, icf, and wrl for the convenience of users. The digital archive materials are available for anyone to check through the National Science Museum Archive (www.col.science.go.kr) and download and use according to the copyright usage guidelines in order to increase information accessibility and utilization of the collections.

In this study, we introduced the current status of the digital archive in the field of natural history, which is being built at our Science Museum and discussed cases of its utilization.

Keywords: national science museum of KOREA, digital archive, natural history, scientific collections, 2d images, 3d printed data

# Combined use of electron microscopy and X-ray micro-computed tomography of biological resources

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Electron microscopy (EM) has been a powerful tool for biologists to observe specimens since the invention in the 1930s. With a superb sub-angstrom spatial resolution, EM allows irreplaceable documentation of cellular features at the nanoscale level. First commercialized in human medicine in the 1960s, X-ray-based imaging methods, including X-ray micro-computed tomography (XCT), have been employed in various disciplines of science to reveal three-dimensional (3D) structures without time-consuming mechanical sectioning. A combined use of EM, X-ray microanalysis, and XCT offers high-resolution images, elemental composition, and virtual sections, respectively. Recent specimens encompassed wood blocks with calcium oxide crystals, leaf trichomes colonized with fungal hyphae, and beetle dorsal cavities for storing fungal spores. These observations indicate the availability of a versatile imaging platform for 3D digital archiving of biological resources with compositional details.

Keywords: digital archiving, electron microscopy, X-ray microanalysis, X-ray micro-computed tomography

S3-1

**Keynote Speaker**

# Applying the latest strategy to manage harmful algal blooms in waterbody

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The occurrence of harmful algae in summer is accelerating every year due to climate change, and the intensity and duration of harmful algae occurrence are becoming longer due to the inflow of non-point pollutants containing high concentrations of nutrients into water bodies. In order to prevent harmful algae blooming occurs every year, efforts are continuously made to reduce pollutants occurring upstream of dams and reduce the inflow of non-point pollutants. In addition, in waterbody where eutrophication is already in progress due to the high concentration of nutrients, the introduction of new technologies to control harmful algal blooming should be actively promoted.

In the field, areas where habitually occurring harmful algae are likely to cause secondary problems are designated as “harmful algae management priority areas,” and efforts are being made to prevent the harmful algal blooming. In addition, through active connection between technologies in the harmful algae, such as harmful algae monitoring, harmful algae forecast and prediction, and harmful algae control, harmful algae technology development and development from an integrated perspective so that the results of each harmful algae technology field can be used for the other stage of harmful algae management. To this end, there is a need to develop an integrated harmful algae management platform and advance the latest technologies such as digital twins in the water environment to increase technical completeness, and to ensure that the results of field application of the technology can be derived from harmful algae control and efficient harmful algae management strategies in water bodies.

Keywords: Harmful algae, Technology, Strategy

# Mass production of graphene for water treatment

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Graphene, a single layer of carbon atoms arranged in a hexagonal honeycomb lattice, has captivated researchers for over two decades due to its remarkable properties and wide range of potential applications.

Here, we introduced a selective edge oxidation (SEO) method for efficient production of few-layer graphene suited for industrial applications. Our protocol focuses on selectively oxidizing graphite edges, which simplifies the process by reducing the use of oxidants and shortening washing steps, leading to high-throughput production. Unlike conventional methods that degrade the entire graphene basal plane, our approach preserves intrinsic properties by maintaining most of the basal plane intact. SEO effectively weakens Van der Waals forces between layers, achieving a 100% yield of few-layer graphene through mild sonication. The resulting SEO-graphene (SEOG) demonstrates exceptional dispersibility in various solvents without dispersants and scalability, with 400 grams produced per batch. Furthermore, this method supports scalability by simply increasing the number of production units. As a potential application of SEOG, the removal of environmental harmful substances was investigated to demonstrate its applicability for the water treatment process.

Keywords: Graphene, Water treatment, Scalability, Two-dimensional carbon material

# Investigation the stability and reactivity of chlorine dioxide ($ClO_2$) in the water treatment process

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Disinfectants such as chlorine ($Cl_2$), ozone ($O_3$), sodium hypochlorite (NaOCl), and chlorine dioxide ($ClO_2$) are used to eliminate phthogenic viruses and small organisms in the water treatment process. Because these disinfectants are directly related to human health, it is necessary to check their reactivity and stability. Among the disinfectants, $ClO_2$, which has good disinfection efficiency, has not been sufficiently studied in Korean water purification systems. Therefore, in this study, we experimentally analyzed the stability and reactivity of $ClO_2$ in water treatment environments. In the experiments, the effects of light and temperature on the stability of $ClO_2$ were analyzed, and its reactivity with organic and inorganic substances was analyzed. The concentration of $ClO_2$ was determined using the analysis method suggested by the U.S. Environmental Protection Agency (EPA), and quantities of the organic and inorganic substances and by-products of $ClO_2$ after its reaction were analyzed using ion chromatography, liquid chromatography-mass spectrometers, and inductively coupled plasma-mass spectrometry. It is believed that the results obtained through this experiment can be used as the basic data needed to properly utilize $ClO_2$ in water treatment plants in Korea.

Keywords: Chlorine dioxide, Water treatment, Stability, Reactivity

# Evaluation of chlorine dioxide disinfection ability using chironomids (Diptera: Chironomidae)

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The recent appearance of chironomids in tap water has raised significant concerns about the safety and hygiene of tap water. Accordingly, there has been increased scrutiny of disinfection efficiency in water purification plants. However, toxicological studies on the use of disinfectants to control small organisms are still insufficient. Traditional disinfectants used in water purification plants require high concentrations or prolonged exposure times to control small organisms, which can result in increased by-products, a known side effect of chlorine-based compounds. In contrast, chlorine dioxide ($ClO_2$) is effective at lower concentrations and shorter exposure times, producing fewer by-products and being an unstable compound with a low boiling point. This study compares the insecticidal effects of $ClO_2$ and sodium hypochloride (NaOCl) on chironomids. Wild types from the Wangsuk-cheon Stream *Glyptotendipes tokunagai* (4th instar larvae rearing at laboratory) were used. Three types of $ClO_2$ and one type of NaOCl were tested at concentrations of 1, 2, 4, 8, 16, 32, 64, and 128 ppm, with a control group treated with distilled water. Five individuals were used per group, and mortality was assessed for 24 hours at specific intervals. Probit analysis estimated the median lethal time ($LT_{50}$) for each concentration. Results showed that $LT_{50}$ decreased with increasing concentration for all disinfectants. The type of $ClO_2$ did not significantly affect mortality rates at the same concentration. All types of $ClO_2$ showed lower $LT_{50}$ values at lower concentrations compared to NaOCl. This indicates that $ClO_2$ has the potential to be an effective method for chironomid management.

Keywords: Chironomids, Chlorine dioxide, Sodium hypochloride, Lethal time

# Effect of diclofenac on bacterial community in activated sludge from wastewater treatment plant of Chiang Mai University

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Diclofenac (DCF) is a widely consumed drug that is found in treated wastewater and environment. This is because DCF may not completely be removed in a municipal wastewater treatment plant (WWTPs), which commonly relies on activated sludge (AS) process and is not designed to remove an emerging contaminant like DCF. This study aimed to determine the effect of DCF on the bacterial community in AS taken from Chiang Mai University wastewater treatment plant, Chiang Mai, Thailand. The 14-day exposure experiments were conducted in flasks containing AS and wastewater sample with 0, 1, and 10 mg/L of DCF. All flasks were shaken at 150 rpm and 30°C. After 14 days, the AS samples were analyzed for bacterial community by 16s rRNA analysis. The results showed that the bacterial community in 14-day AS at 10 mg/L of DCF significantly changed compared with that of the 0-day AS at 0 mg/L of DCF. Some bacteria such as *Gemmatacese* appeared to be benefitting from DCF exposure while *Pseudomonas*, which was found being able to degrade DCF by previous studies, did not show an increase in the abundance in this study. Findings from this study can help elucidating the positive and negative effects of DCF on bacteria that could be important to wastewater treatment of drugs such as DCF in an AS system.

Keywords: activated sludge, bacterial community, biodegradation, diclofenac, wastewater treatment

# Implementation of big data to control disease vector mosquitoes

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Vector-borne infectious diseases are among the most significant concerns for humans and animals due to climate change and increased global trade and travel. Dengue and Zika viruses are becoming more prevalent in many regions, and the recurrence of malaria cases has been continually reported in some countries. These diseases are all transmitted by mosquitoes, whose larval stages are heavily dependent on aquatic environmental changes. Consequently, the rapid and precise detection of vector mosquito species such as *Aedes*, *Anopheles*, and *Culex* is crucial for implementing effective vector control measures. In this presentation, the brief history and importance of handling big data, such as genome information and AI-based on-site inference for monitoring mosquito population fluctuations, will be discussed. This approach represents a new direction for controlling disease vectors in environmentally friendly ways.

S4-2

# Ecological feature of *Aedes albopictus* (Diptera: Culicidae) in Korea

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*Aedes albopictus* is an invasive mosquito that can be found on all continents and considered as vector of many infectious disease such as dengue fever, chikungunya fever and also Zika virus. Although the case of autonomous dengue fever has not been reported in Korea, it leaves rich possibility of influx of this disease because vector mosquito are present and many imported dengue cases was reported annually. In this study, we evaluated the larval growth time with temperature, activity time, and also bloodmeal source. The larval growth time was reduced as the temperature increased: it takes 242.3 h, at 26□ and 209.1 h at 28□. *Ae. albopictus* showed a very aggressive daytime biting. Its peak active times are during the early morning (8:00-9:00) and late afternoon (16:00-17:00). However, in a deep forest reduced light intensity, the peak activity showed a single broad peak in the afternoon. The bloodmeal source of *Ae. albopictus* were mammalian 71% followed by avian 26%, amphibian 2% and fish 1%. Most of blood source of mammalian was human (86%). This result showed that *Ae. albopictus* feed almost exclusively anthrophophylic. Therefore, it is urgently need to survey throughout the country for making control strategy.

Keywords: *Aedes albopictus*, daily activity, bloodmeal source, Dengue fever, chikungunya fever, Zika virus

# Predators as biocontrol agents of mosquito larvae in small and large habitats in Chiang Mai, Thailand

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Controlling mosquito-borne disease is a major global challenge due to the rise of insecticide-resistant mosquitoes. In response, we conducted a study in Chiang Mai Province, Thailand, which is one of the largest and the most popular cities for tourists in Southeast Asia, to explore the potential of local species as biological control agents for mosquito larvae. Mosquito larvae and aquatic predators were sampled from large and small habitats, while relevant physico-chemical parameters were measured. The study identified 560 predators and 1,572 mosquitoes, with most mosquito species belonging to the genus *Culex*. Additionally, the study identified 16 predator taxa, including four fish taxa and 12 taxa of predatory aquatic insects belonging to four orders: Coleoptera, Hemiptera, Odonata, and Diptera. The study found that several locally occurring predator species, namely *Poecillia, Laccophilus, Lutzia, Toxorhynchites splendens, Agrionoptera,* and *Pseudarion*, shared habitats with mosquitoes, indicating their potential as effective biological control agents for mosquito control. Conductivity, dissolved oxygen, and pH were the important physico-chemical parameters that affect both predators and mosquito larvae. Consequently, promoting native predators and reducing mosquito larvae through habitat management would be a sustainable and ecologically friendly approach in large habitats where it is not possible to remove mosquito oviposition sites. In smaller habitats, releasing local aquatic predators and removing oviposition sites may be a suitable strategy.

Keywords: biocontrol, mosquito larva, predator, habitat, Chiang Mai, suburban area

Acknowledgements: Thanks to the Faculty of Science, Chiang Mai University, Thailand for financial support.

# Phylogenetic analysis and distribution of Asian predaceous *Toxorhynchites* mosquitoes (Culicidae): Exploring their potential for biological control as a Nature-Based Solution (NbS)

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*Toxorhynchites* mosquitoes possess a unique predatory behavior: their larvae consume other mosquito larvae, while they are non-hematophagous in adult stage. As one of top predators in mosquito habitats, their predatory behavior makes them a promising biocontrol agent for mosquito control. Despite its limitations, biological mosquito control is eco-friendly when effectively implemented. However, the molecular phylogeny, ecology, and distribution of *Toxorhynchites* species in East and Southeast Asia have not been fully investigated. To better understand species in this genus as biological control agents, we analyzed *COI* sequences of four species (*Toxorhynchites sunthorni, Tx. splendens, Tx. gravelyi*, and *Tx. towadensis*) from the East and Southeast Asia region. The analysis revealed that the average intraspecific genetic distances of *Tx. sunthorni* and *Tx. splendens* (from tropical areas) are 0.17% and 0.45%, respectively, whereas *Tx. gravelyi* and *Tx. towadensis* from subtropical and temperate areas showed slightly higher intraspecific genetic distances (0.59% and 0.95%, respectively). The genetic distances between *Tx. sunthorni* and *Tx. splendens* were lower than those between *Tx. gravelyi* and *Tx. towadensis* (2.70% and 12.15%, respectively). The *Tx. sunthorni-Tx. splendens* clade has a widespread distribution, with most of their larvae found in artificial habitats (e.g., used tires). In contrast, the *Tx. gravelyi-Tx. towadensis* clade has a more limited geographic range, and their larvae are normally found in natural habitats (e.g., tree holes). Understanding the taxonomy, molecular phylogeny, and genetic diversity of *Toxorhynchites* mosquitoes, along with their distribution, will enhance sustainable biological control as a nature-based solution (NbS), especially as urban mosquitoes develop insecticide resistance.

Keywords: biological mosquito control, larvicide, nature-based solution, predaceous mosquito, Thailand, *Toxorhynchite*s

Acknowledgements: We thank the Korea Disease Control and Prevention Agency (KDCA) for the KDCA-DVP 2024 Grant Program (R67EX00317) and for partial support from Chiang Mai University (R000032014).

# Characterisation of acaracide resistance-conveying mutations in para-sodium channel of dog and cattle ticks, Thailand

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Dog and cattle ticks of the genus *Rhipicephalus* are widespread disease vectors and agricultural parasites worldwide. Pyrethroid acaricides are commonly used to treat tick infestations, leading to knock-down resistance (*kdr*) development. In this study, we aimed to survey the prevalence of *kdr* mutations in field-collected tick populations across northern, northeastern, eastern, western and central Thailand. Preliminary PCR-based assays of domain II (DMII) and domain III (DMIII) of the para-sodium channel gene were used to confirm the presence of resistance-conferring mutations. We then investigated phenotypic resistance to pyrethroids (permethrin and deltamethrin) in two tick populations collected from northern Thailand through larval packet discriminating dose testing. Genotypic analysis of tick samples was then used to establish a correlation between phenotypic and genotypic resistance. Our study detected 5 *kdr* mutations associated with pyrethroid resistance in *R. sanguineus*: T170C, C190A, C190G, G215T in DMII and T2134C in DMIII. At least one resistance SNP was present in 82.5% of all samples. In *R. microplus*, we detected corresponding mutations G215T and T2133C in 2 individuals. Larval packet testing revealed high levels of permethrin resistance in both populations and slightly lower levels of deltamethrin resistance. Our analysis revealed a potential correlation between larval packet survivorship and resistance alleles. These data provide a preliminary report of synthetic pyrethroid resistance in four out of five regions of Thailand and identify *kdr* mutations in Thai ticks for the first time. This suggests a need for further resistance surveying across Thailand for a full picture of pyrethroid resistance.

Keywords: dog tick, cattle tick, *Rhipicephalus sanguineus, Rhipicephalus microplus*, pyrethroid, acaricide resistance, larval packet test, *kdr*

S5-1

Keynote Speaker

# Third pole in crisis: Opportunities, challenges, and climate actions

**Sonam Wangyel WANG**[P,C]

Research Professor, OJERI & Coordinator for Mid Latitude Region Network

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The High Mountains of Asia (HMA), known as the Third Pole (TP), hold the world's third-largest reserve of freshwater, critical for over 2 billion people. Its ecosystem benefits are globally enjoyed. The TP, comprising vast glaciers, permafrost, and diverse ecosystems, is increasingly vulnerable to climate change. Recent studies highlight rapid warming, accelerated glacier melt, and permafrost degradation, which disrupt hydrological processes and reduce water availability, impacting both biodiversity and human livelihoods. The rise in glacial lake outburst floods (GLOFs) and habitat loss endangers species like the snow leopard, while intensifying resource conflicts among alpine communities and wildlife.

Research led by Dr. Sonam Wangyel Wang and the OJEong Resilience Institute aims to address these challenges by enhancing understanding of climate impacts and developing science-based adaptation strategies. Preliminary findings indicate severe threats from climate-induced disasters and changing biodiversity dynamics, necessitating robust research, multilevel partnerships, and education to foster resilience. Addressing these issues is further complicated by socio-economic factors and limited adaptive capacity, underscoring the urgency for comprehensive climate actions to safeguard the TP's environmental and social well-being.

# Water-Energy nexus: Turning wastewater into bioenergy

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Bioenergy presents a sustainable and economical alternative to fossil fuels, playing a crucial role in mitigating climate crisis. With its diverse applications in heat, electricity, and transportation, bioenergy serves as a fundamental supplier of carbon-neutral energy sources. The sustainable supply of organic materials is pivotal consideration, given that bioenergy is derived from such materials. Initially, food crops were used as 1st-generation feedstocks for bioenergy production. Subsequently, non-edible lignocellulosic biomass and microalgae were employed as 2nd and 3rd-generation feedstocks, respectively. More recently, there has been a surge of interest in bioenergy production from wastewater, which integrates waste-management with renewable energy supply. This strategy is aligned with the water-energy nexus, facilitating the attainment of sustainable development goals. This presentation aims to impart a foundational understanding of bioenergy production from various feedstocks, with a particular emphasis on Sustainable Aviation Fuels (SAF). SAF represents a pragmatic avenue for renewable and carbon-neutral alternatives in air transportation. Leveraging the inherent potential of bio-oil production capacity in yeast enables sustainable SAF production. Furthermore, this presentation will delve into recent advancements in bio-oil production utilizing wastewater, while also addressing synthetic biology and metabolic engineering approaches for optimizing the potential of bio-oil for SAF production.

S5-3

# Spatial livelihood vulnerability modeling in Central Asia using various indices

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With the increasing vulnerability assessments in the Central Asian Third Pole, many studies relied on local community surveys. However, transferring all indicators from community surveys into spatial data is very difficult but necessary to develop proper Third Pole nexus management plans. Therefore, this study tried to develop the Spatial Livelihood Vulnerability Index (SLVI) and focused on some issues for spatial analysis of SLVI. First off, possible 3rd pole boundaries, which is the target region of applying SLVI, were defined in various ways considering different spatial data. The most common approach was using physical status like elevations based on the Digital Elevation Model (DEM), but considering administrative boundaries and river basins had advantages in holistic analysis. Second, the development of SLVI required spatial data for climate, sensitivity, and adaptive capacity. In addition, aggregation methods for different indices were required. Therefore, we checked the current LVI framework and found other integrated indices assessment concepts based on the Intergovernmental Panel on Climate Change (IPCC) and the Mediterranean Desertification and Land Use (MEDALUS) approach. As the third step, data for physical, social, human, and natural assets were gathered as much as possible using proxy data. Within this step, the global availability of open-source data was checked. Finally, SLVI was analyzed as the primary assessment. In this assessment, the Pamir Plateau and Hindu Kush Himalaya region were the most vulnerable areas. There were some gaps between the original LVI and SLVI, but spatial assessment is somewhat meaningful it because illustrates vulnerable areas. In addition, quality gaps between globally available data and detailed local data based on the current LVI framework were observed. Through this quick application of SLVI, the downstream area of the high mountains was extremely vulnerable in this assessment. Therefore, the development of local spatial datasets as well as consideration of the possibility of proxy data will be important in future studies.

# Challenges facing ecosystem resilience in Nigeria – West Africa

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The economic structure of Nigeria, a country that produces around six quadrillion of Btu oil per year, heavily relies on energy production. Oil accounts for about 90% of total exports and 75% of the government's revenue. After oil, agriculture holds the most significant share of the economy, which holds one fourth of the country's GDP. Both oil and agricultural production is the most significant revenue sources for the country and the main threats to biodiversity. Oil spillage, gas flaring, and pipeline explosions in Nigeria are the most known ones. However, some other threats endanger biodiversity's such as hunting, overfishing, and some local groups' sacred habits. These complex interaction between humans and nature has led Nigeria to be one of the most debated biodiversity loss cases in the world. The 5000 km$^2$ Oban-Korup Forest block, which covers large parts of Cross River State (CRS) and continues into Cameroon, represents more than 50% of Nigeria's remaining tropical high forest and is considered one of Africa's most important biotic reserves. Already experiencing rapid degradation from an ever-expanding agricultural frontier, a resurgent plantation economy serves to amplify pressures on forest resources. Poor law enforcement on biodiversity has occasionally caused embarrassment for the Government and people of Nigeria.' It was further noted that poor institutional cohesion, low capacity of States to manage varied biodiversity related portfolios, the lack of commitment to and investments in the departments responsible for biodiversity matters characterize the biodiversity conservation policy of most States in Nigeria - Biodiversity related legislations such as the wildlife and forestry laws are obsolete, non-implementable and are totally ignored by the customary, sharia and other courts. So far evaluation has shown that the global treaties and conventions has not helped countries in the global south to be more resilience in the face of land degradation, deforestation, and land, water and air pollution among others. Knowing that Land is by far the most important resource necessary for subsistence. Evaluation showed that abuse of environmental laws without commensurate penalties, weak enforcement, corruption on the part of the staff of the regulatory agency, poor public enlightenment education had led to the huge challenges confronting Nigeria ecosystem resilience.

# V. Poster Session

Posters will be posted during the conference and poster presentation will be at **5:30 pm on July 3rd.**

# Taxonomy of the genus *Epeorus* Eaton, 1881 (Ephemeroptera, Heptageniidae) in Thailand

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The genus *Epeorus* Eaton, 1881 has been reviewed in Thailand. Seven known species of *Epeorus* (*E. aculeatus* Braasch, 1990; *E. bifurcatus* Braasch & Soldán, 1979; *E. inthanonensis* Braasch & Boonsoong, 2010; *E. khayengensis* Braasch & Boonsoong, 2010; *E. martinus* (Braasch & Soldán), 1984; *E. thailandensis* Boonsoong & Braasch, 2013 and *E. unicornutus* Braasch, 2006) have been recorded from Thailand. Five subgenera (*Belovius, Caucasiron, Iron, Proepeorus,* and *Siniron*) of Thai *Epeorus* were reported from Thailand. Two new putative species (*E. (Caucasiron)* sp. and *E.* (*Proepeorus*) sp.) are recognized. The subgenus *Caucasiron* is the first record from Thailand. Biological data and distribution of known Thai species are also provided. The imaginal stages of *E.* (*Belovius*) *khayengensis* Boonsoong & Braasch, 2010 are presented for the first time. Moreover, the delimitation of six species is verified by mitochondrial COI sequences for the first time.

Keywords: *Epeorus*, Heptageniidae, Thailand, DNA barcoding

# The Thai cryptic species of Ephemerellidae (Insecta; Ephemeroptera) revealed by the integrative taxonomy

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The family Ephemerellidae has been observed in around fifteen species across six genera, although the exact number of species remains uncertain and may be underestimated. Therefore, the diversity of the family Ephemerellidae in Thailand has been studied using integrative taxonomy. Samples of Ephemerellidae have been collected throughout Thailand between 2016 and the present. The data collected includes both morphology and COI sequence data. The genera were assigned into six as same as previously. The COI phylogeny indicates the presence of two major clades comprised of *Cincticostella* and the clade with polytomy of the genus *Teloganopsis* and *Torleya*. In addition, the genus *Serratella* was placed within the genus *Teloganopsis*. The morphology and COI phylogeny suggest a higher number of species than the previous report which there are at least 21 species. Even though, the COI region was unsuccessful amplified from some morphospecies. Based on our results, further study is necessary to revise the family Ephemerellidae using multiple markers and comparison with morphological data to accurately determine the genus and species of Ephemerellidae in Thailand.

Keywords: *Cincticostella*, COI, DNA barcoding, Mayfly, *Teloganopsis, Torleya*

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# A DNA barcoding reference library for the family Baetidae of Thailand

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Baetidae is one of the numerous and diverse mayfly family but there were few studies of this family in Thailand were conducted previously. Due to their small body size, the members are often ignored and left unnamed in as Baetidae sp. by the former Thai researcher. After a subject for a decade, there were 15 genera including 32 species reported officially in Thailand and only six genera were including the DNA data. Here, we are reported the DNA barcoding reference library of the family Baetidae in Thailand. COI sequences were produced, freshly specimens were selected from the collection of the six years mass survey in Thailand by four teams from CMU, BU, KU and KKU. These data were combined with retrieving published COI sequences in databases (GenBank and BOLD). Based on genetic evidence, a species of the Baetidae family from Thailand shows that 16 genera are divided into 42 species. Remarkably, there are more species based on genetic evidence than there were in earlier reports from Thailand. This study provides the first Baetidae DNA reference database, allowing future species identification accuracy by combined data of morphology and molecular for the purposed of improving knowledge of water monitoring taxa and the reference collection will be accessible for future studies on metabarcoding and environmental DNA.

Keywords: COI, Mayfly, Southeast Asia, Integrative taxonomy, GenBank, BOLD

Acknowledgments: This project is funded by National Research Council of Thailand (NRCT) contract Number N42A660904 and the faculty of Veterinary Medicine (Chiang Mai University).

# Genetic structure of *Macrobrachium nipponense* (Decapoda, Palaemonidae) in Yatsunuma, traditional agricultural reservoirs in the Hiki Hills

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The oriental river prawn, *Macrobrachium nipponense*, is widespread in East Asia including Japan. These juveniles generally grow up in the sea and then migrate to upper rivers. However, if migration to the sea is interrupted, the prawns may complete its life cycle in freshwater. “Yatsunuma” has been traditionally used for agricultural reservoirs in the Hiki Hills of Saitama Prefecture, Japan. These reservoirs have long history that the oldest its since around 600 AD, and this agricultural system was already completed 1600 AD. Water releases from these reservoirs are regulated by agricultural progress and season, presumably limiting the migration of these prawns.

In this study, we revealed the genetic structure of this species inhabiting the reservoirs of the Hiki Hills and the surrounding rivers, Ara-kawa River using the *COI* gene and the D-loop region of mitochondrial DNA. As a result, a common haplotype was detected in the *COI* gene from all sites. On the other hand, common haplotypes were detected in the *D-loop* region in the middle reaches of the Ara-kawa River and in the reservoirs, but these haplotypes were different from the lower reaches of the river. This suggests that the individuals inhabiting Yatsunuma maintain their populations only in freshwater areas such as reservoirs and surrounding rivers, without migrating to downstream or to the sea.

Keywords: traditional pond, genetic endemism, genetic diversity, oriental river prawn, long-armed palaemonid prawn, freshwater prawn, Palaemonidae

# Checklist of the gomphid larvae (Odonata, Gomphidae) in Thailand

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The family Gomphidae is a well known group of dragonflies comprising about 60 species in Thailand. Nevertheless, the knowledge of gomphid larvae such as species diversity, distribution, and larval morphology are paucity. The gomphid larvae exhibit several apparently characteristics, such as 2-segmented fore tarsi and middle tarsi and 4 segmented antennae with antennomere 4 minute or vestigial. In this study, diversity of gomphid nymphs were investigated. The gomphid larvae were collected from 34 sampling sites. The gomphid larvae of 25 species of the 21 genera that occur in Thailand are keyed and diagnosed (*Amphigomphus somnuki, Asiagomphus xanthenatus, Burmagomphus divaricatus, B. williamsoni, Gomphidia abbotti, Gomphidictinus perakensis, Heliogomphus selysi, Ictinogomphus decorates, Lamelligomphus castor, Macrogomphus kerri, Megalogomphus sumatranus, Merogomphus pavici, Microgomphus ferrelli, M. svihleri, Nepogomphus walli, Nychogomphus duaricus, Onychogomphus louissiriusi, Orientogomphus minor, Paragomphus capricornis, Phaenandrogomphus asthenes, P. dingavani, P. thokinicus, Sieboldius nigricolor, Stylogomphus malayanus, S. thongphaphumensis*). In addition, a checklist of the gomphid larvae in Thailand is presented in this study including notes on occurrence, habitat types, and environmental monitoring. Additionally, we have gained a better understanding of distribution, and updated the DNA barcoding to reflect the relative abundance of species more accurately.

Keywords: Gomphidae, Thailand, larvae, diversity, distribution, microhabitat

# Revisiting the taxonomy of Korean *Ischnochiton* species (Ischnochitonidae: Polyplacophora) based on a combined analysis of morphological and molecular data

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Chiton species belonging to the genus *Ischnochiton* J. E. Gray, 1847 are commonly found in intertidal rocky shores worldwide, with the exception of the northern Atlantic and Arctic oceans. *Ischnochiton* species are characterized by imbricate girdle scales that are uniform in size, rounded, sculpted with striae, or occasionally smooth. However, their species-level taxonomy is complicated due to the high variation in their shell microstructures. Despite more than a hundred species are reported worldwide, the taxonomic studies of this group remain relatively unexplored in Korean waters, with only a few species recorded to date. In this study, we compared the microstructural characters (tegmentum sculpture, girdle scales, and radula) of external and internal morphology of four Korean *Ischnochiton* species using high-resolution microscopic images and a scanning electron microscope (SEM). Along with mtDNA *cox1* sequence comparison, a comprehensive analysis of their morphology revealed that *I. hayamii* Owada, 2018 was identified for the first time in Korean waters. This species is morphologically distinguished by its small body size of adults, smooth lateral areas on valves, and small perinotum scales sculptured with weak longitudinal ribs. The present study provides detailed morphological comparison of microstructures of shell plates, girdle scales, and radula for four Korean *Ischnochiton* species. Phylogenetic analysis of the mtDNA cox1 sequence also provides distinct resolution at the species level, but interrelationships among Ischnochiton species remain unresolved. Results from the morphological and molecular analyses provided in this study offer valuable taxonomic information that can be used for accurate species identification among closely related *Ischnochiton* species.

Keywords: *Ischnochiton*, Ischnochitonidae, microstructures, shell plate, perinotum scales, radula, SEM, mtDNA *cox1*, Korea

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# On marine midges (Diptera: Chironomidae): Morphological description of four Korean species

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Intertidal habitats support a wide variety of marine life, but harbor only a few insect diversity due to their high salinity and various invertebrate predators including crustaceans and mollusks. Despite these environmental challenges, relatively a few insect groups such as beetles and flies can be successfully adapted to the intertidal environments. The family Chironomidae, commonly known as  non-biting midges, is a diverse group of aquatic insects belonging to the Diptera, playing an important role as a prey in the intertidal ecosystems, including salt marshes, supra-littoral splash pools, tidal pools, sublittoral sediments and even the pelagic zone. More than a hundred marine chironomid species from 30 genera of three subfamilies have been described worldwide. In this study, we report the discovery of four marine midge species from intertidal habitats along the Korean sea coast for the first time: *Clunio tsushimensis* Tokunaga, 1933, *Pseudosmittia bifurcata* (Tokunaga, 1936), *Semiocladius endocladiae* (Tokunaga, 1936) (Orthocladiinae), and *Telmatogeton japonicus* Tokunaga, 1933 (Telmatogetoninae). We provide their taxonomy, habitat, and detailed morphological information along with illustration of diagnostic characters of the male genitalia.

Keywords: Marine chironomids, *Clunio*, *Pseudosmittia*, *Semiocladius*, *Telmatogeton*

Acknowledgements: This work was supported by a grant (NIBR202402202) from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea.

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# Mitochondrial Genome phylogeny of Patellogastropoda (Gastropoda: Mollusca)

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Patellogastropoda, commonly known as “true limpets”, is considered one of the most primitive gastropod groups that has diversified into different zonal habitats, ranging from deep-sea environments to intertidal habitats. Nevertheless, they are most abundantly found on intertidal rocky shores worldwide. Although they are characterized by having a conical shell and a muscular foot, phylogenetic relationships among their family members from the morphology and previous molecular analyses still remains unresolved. To reconstruct phylogenetic relationships among the families of Patellogastropoda, we conducted a phylogenetic analysis for the nucleotide and amino acid sequences of the 13 protein-coding genes of the complete mitochondrial genome. Our dataset comprises 44 complete mitochondrial genome sequences, including 13 newly sequenced mitochondrial genomes of limpet species, 25 from GenBank, and six from non-Patellogastropoda. Resulting phylogenetic trees, inferred from maximum likelihood and Bayesian methods, for both nucleotide (NT) and amino acid (AA) sequence datasets, consistently revealed that the Patellogastropoda species were subdivided into two clades with high branch supporting values: Clades I comprise Nacellidae, Pectinodontidae, Lepetidae, Acmaeidae, Rhodopetalidae, Neolepetopsidae, and Patellidae, whereas Clades II contains Lottiidae and Eoacmaeidae. The discrepancy in topology between the two datasets was found within Clade I. In the NT tree, Lepetidae is positioned as the sister group of Pectinodontidae, whereas in the AA tree, Lepetidae occupies the basal position among the other five families. The NT and AA datasets indicate different inter-relationships among family members. Along with comparison of gene arrangement data, we present and discuss the phylogenetic relationships among the family members pf Patellogastropoda.

Keywords: Mollusca, Gastropoda, Patellogastropoda, Mitochondrial genome, Molecular Phylogeny, Gene Arrangement

# Phylogeography of the Siberian stone loach *Barbatula toni* (Cypriniformes: Nemacheilidae): Back dispersal from Hokkaido to Sakhalin

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The Quaternary geohistory affected the geographic distribution patterns of organisms and dispersal routes of their populations. The formation of land bridges during glacial periods in the Pleistocene played important roles in migration of organisms between continents and their surrounding remote islands. We investigated the phylogeographic pattern and genetic structure of the Siberian stone loach *Barbatula toni* in East Asia, using the sequencing data of the mitochondrial DNA cyt *b*, COI, 12S rRNA and D-loop regions. As the result of our phylogenetic analyses, the populations of the Hokkaido and Sakhalin islands consisted a derived lineage within the species, and they formed a monophyletic group: it was detected two major clades (North and South clades) consisting of six subclades. Regarding estimation of divergence times, it was indicated the divergence between the two major clades was estimated 2.2 Ma, and the divergence times between the six subclades was approximately 1 Ma. Thus, these clades' divergence periods were accorded to the "glacial-interglacial cycles" during the Pleistocene. Ancestral area reconstruction indicated the two major clades originated in Hokkaido. The results of neutrality tests and "Extended Bayesian Skyline Plots" analysis indicated recent population expansions in subclades which are distributed in Hokkaido and Sakhalin. These findings strongly suggested the "back dispersal" phenomenon of *B. toni* from Hokkaido to Sakhalin, provided vital insights into the role of the island system in creation and maintenance of biodiversity.

Keywords: Phylogeography, Back dispersal, Glacial-interglacial cycles, Pleistocene, land bridge

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# Mitochondrial genome phylogeny of the genus *Ceratostoma* (Gastropoda: Muricidae) revealed amphi-Pacific origins of their diversification

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The genus *Ceratostoma* (Gastropoda: Muricidae), commonly known as “the rock shells”, a group of marine gastropods belonging to the family Muricidae. They are most abundantly found in rocky intertidal and subtidal habitats and distinguished by its wing-like structures (called ‘varix’) and spiny or knobbed shell sculpture. They are distributed in coastal regions of both the Western Pacific (WP) and Eastern Pacific (EP) seas: *C. rorifluum*, *C. burnetti*, and *C. fournieri* are widely distributed in the WP, while *C. nuttalli* and *C. foliatum* are found in the EP. However, the spatio-temporal patterns of diversifying origin in the Ceratostoma species still remain unexplored, emphasizing the need for further study to understand how and when their speciation events occurred, ultimately leading to their amphi-Pacific distribution. For comprehensive understanding of their speciation, and dispersal origin(s), we performed a phylogenetic analysis for the complete mitochondrial genome sequences of five *Ceratostoma* species representing the two amphi-Pacific species (*C. rorifluum*, *C. burnetti*, *C. fournieri* [WP]; *C. nuttalli* and *C. foliatum* [EP]), with three ocenebrine species as outgroups. Phylogenetic trees from maximum likelihood and Bayesian inference methods show a well-supported dichotomy of the two monophyletic clades, each representing Western Pacific (*C. fournieri* (*C. rorifluum* + *C. burnetti*) and Eastern Pacific species (*C. nuttalli* + *C. foliatum*). The inferred phylogenetic relationships provide significant insights into evolutionary history and biogeographic patternns of the genus *Ceratostoma*, thereby enhancing our understanding of the diversification origins of marine invertebrate species in the northern Pacific.

Keywords: Gastropoda, *Ceratostoma*, Mitochondrial genome phylogeny, Diversification origin, Amphi-Pacific distribution, Northern Pacific

# Genome assembly and population genomic data of a pulmonate snail *Ellobium chinense*

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*Ellobium chinense* is an airbreathing, pulmonate gastropod species that inhabits saltmarshes in estuaries of the northwestern Pacific. Due to a rapid population decline and their unique ecological niche in estuarine ecosystems, this species has attracted special attention regarding their conservation and the genomic basis of adaptation to frequently changing environments. Here we report a draft genome assembly of *E. chinense* with a total size of 949.470 Mb and a scaffold N50 of 1.465 Mb. Comparative genomic analysis revealed that the GO terms enriched among four gastropod species are related to signal transduction involved in maintaining electrochemical gradients across the cell membrane. Population genomic analysis using the MSMC model for 14 re-sequenced individuals revealed a drastic decline in Korean and Japanese populations during the last glacial period, while the southern Chinese population retained a much larger effective population size ($N_e$). These contrasting demographic changes might be attributed to multiple environmental factors during the glacial–interglacial cycles. This study provides valuable genomic resources for understanding adaptation and historical demographic responses to climate change.

Keywords: *Ellobium chinense*, Pulmonata, WGS, Evolutionary adaptation, Demographic history

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# Genetic structure based on the mitogenomic and genome-wide population analysis of the East Asian *Stenopsyche marmorata* populations

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Rivers contain a variety of environments that change longitudinally, and organisms adapt to each habitat. Therefore, these environmental changes in rivers are thought to have significant impact on the diversity of biological communities within the freshwater ecosystem. In the river ecosystem with diverse environments, understanding which regions contribute to the creation and maintenance of diversity is an important issue for considering freshwater population dynamics. In this study, we focused the caddisfly, *Stenopsyche marmorata*, which has the most widespread distribution and biomass among aquatic insects in Japanese rivers. Furthermore, this study focused on the Kano-gawa River system, which contains a spring-origin tributary with a flow of over 1 million tons of water per day and an artificially created discharge channel that runs directly from the middle of the river to the sea. With these features, the Kano-gawa River system has a great habitat complexity and is a suitable place for this study. First, we conducted the low-coverage whole genome sequencing (lcWGS) and assembled mitogenome to reveal the phylogenetic relationships of East Asian populations of *S. marmorata*. After the wide-regional phylogeographic relationships within this species were confirmed, genome-wide SNPs analysis using GRAS-Di method was conducted to reveal population structure and geneflow within the Kano-gawa River system. This study shows that the populations within the spring-origin tributary is slightly independent from other populations (main river and other tributaries), however the entire populations of the river system can be treated as a "metapopulation".

Keywords: phylogeography, population genetics, cryptic species, mtDNA

# Cryptic species diversity in Korean benthic populations of golden tide seaweed, *Sargassum horneri*, based on phylogeographic and population genetic analyses

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The sea basins of the Northwest Pacific (NWP) were geologically separated from the land during the Pleistocene, which largely influenced the current geographic distributions of many marine species that were able to survive in the glacial refugia in those regions. The NWP coastal ecosystem harbors a high regional biodiversity with abundant seaweed populations, which is perhaps attributed to complex and dynamic oceanic current system. In recent years, individuals of some *Sargassum* species detached from the substrate drift on the sea surface and form enormous patches, sometimes causing severe damage to coastal ecosystems globally. In particular, *Sargassum horneri* often brings about ‘golden tide’ blooming in the NWP open ocean. However, *S. horneri* is commonly distributed along the NWP coast and its benthic populations play an important role in the marine ecosystem by forming marine forests. In this study, we determined the phylogeographic distributions and current population genetic structure of benthic S. horneri on the entire coasts of South Korea including Jeju Island, the East Sea, South Sea, and West Sea using mitochondrial (mt) DNA and seven microsatellite markers. Comparisons of the four sea areas revealed that populations from Jeju Island, the West Sea, and the East Sea were divided into two distinct clades, while the South Sea represent admixture of two clades. The analyses of $F_{ST}$ statistics showed limited genetic connectivity among populations in general. Based on the results of network analysis, we identified two unique lineages showing potential as cryptic species complex evolved in *S. horneri*. This study is the first to evaluate the population genetic features of Korean benthic *S. horneri* and provide the genetic information on two well-separated lineages with a rather deep evolutionary divergence. This study will further help to manage non-indigenous and floating *S. horneri* effectively in Korean waters.

Keywords: cryptic diversity, Korean benthic population, Northwest Pacific (NWP), phylogeography, population genetics, *Sargassum horneri*

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# Population genetics and ecological niche modeling of massively emerging burrowing mayfly species, *Ephemera orientalis* (Ephemeroptera: Ephemeridae), in South Korea

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The widespread phenomenon of mayflies mass emergence involves the synchronized appearance of a large number of individuals in specific seasons, often triggered by temperature and photoperiod. A notable example is the burrowing mayfly *Ephemera orientalis* (Ephemeroptera: Ephemeridae), which is widely distributed across South Korea and has become a significant nuisance due to repeated mass emergence in urban areas. In the present study, we investigated the level of genetic diversity and population genetic structures of 18 *E. orientalis* populations to better understand the genetic attributes of mass-emerging populations. Additionally, we performed ecological niche modeling (ENM) using data from National Ecosystem Survey to identify the major environmental drivers to these mass emergence patterns. We indicated that a higher level of genetic diversity in mass-emerging populations, suggesting a larger effective population size (Ne) compared to other populations. STRUCTURE analysis revealed only two distinct genetic clusters of *E. orientalis* with significant genetic differentiation within Korea. Our ENM analysis identified key environmental attributes influencing the mass emerging populations. These findings offer valuable insights into the genetic, ecological, and environmental factors that influence mass emergence patterns in the mayfly system.

Keywords: COI, Ecological niche modeling (ENM), Ephemeroptera, Mass emergence, Microsatellites, Population genetic structures

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# Development and characterization of novel microsatellite markers for *Nemoura geei* (Plecoptera: Nemouridae)

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Stoneflies (Plecoptera) are known for being sensitive to water pollution and are used as bioindicators for evaluating water quality. Among them, Nemouridae, especially the genus *Nemoura*, which are commonly referred to as winter stoneflies, can be found around streams even during the cold winter months. *Nemoura geei* Wu, 1929, among them, was originally described from Beijing and is widely distributed in Korea, China, Japan, and the Russian Far East. Here, we report the development and characterization of new functional microsatellite markers of *N. geei* using high-throughput sequencing technology. A total of 80,661 microsatellite loci were identified with a total length of 1,801,591 bp. The average length was 22.34 bp, and microsatellites occupied 0.42% of the entire sequence. The novel 20 microsatellite markers developed in this study can be usefully applied to the population genetics analyses as important genetic resources for understanding the ecological and evolutionary characteristics of a stonefly species at the population level in Korea.

Keywords: high-throughput sequencing, microsatellite, population genetics, stonefly

*These authors contributed equally to this work.

# X-ray micro-computed tomography of the subterranean amphipod *Pseudocrangonyx hwanseonensis* (Crustacea: Amphipoda: Pseudocrangonyctidae)

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X-ray micro-computed tomography (Micro-CT) is a non-destructive method that provides three-dimensional(3D) imaging information. In this study, 3D morphological analysis was performed on *Pseudocrangonyx hwanseonensis* using micro-CT. This species is a subterranean amphipod found in the groundwater of limestone cave (Hwanseon Cave) in South Korea. Micro-CT enabled the observation of both internal and external morphological characteristics of *P. hwanseonensis.* This is the first report of 3D reconstruction using micro-CT for morphological analysis of *P. hwanseonensis*.

Keywords: *Pseudocrangonyx hwanseonensis*, Micro-CT, subterranean amphipod, morphological analysis

Acknowledgements: This work was supported by a grant from the Nakdonggang National Institute of Biological Resources (NNIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NNIBR20242102).

# Note on substrate colonization of the polymitarcyid burrowing mayfly (Ephemeroptera, Polymitarcyidae) from Thailand

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Six known species of the family Polymitarcyidae, belonging to three genera were reported in Thailand. The nymphs of these species burrow tunnels in submerged wood, inhabit aquatic plants, and live in inorganic sediment. However, the polymitarcyid burrowing mayfly includes both wood-boring and rock-boring species. Through a series of taxonomic studies on Thai mayflies and citizen science in Thailand, the substrate colonization of polymitarcyid mayflies has been observed and reported. The nymphs of Thai *Languidipes* and *Povilla* were identified as wood-boring mayflies. Specifically, the nymphs of *Povilla heardi* Hubbard, 1984, colonized in *Typha angustifolia* rhizomes, plastic floating solar panels, and the shells of captive snapping turtles (*Chelydra serpentina*). Additionally, the nymphs of *Languidipes taprobanes* Hubbard, 1984, inhabited and bored into submerged electric cables of fountains in urban areas. This study highlights how the colonization of polymitarcyid substrate-boring nymphs can directly and indirectly affect humans. Note on substrate colonization of the polymitarcyid boring species will contribute information to solve causing problems with human structures submerged in water. Further research on the biology and life history of polymitarcyid burrowing mayflies is necessary for future investigations.

Keywords: mayfly, burrower, colonization, wood-boring

# Post embryological development of external characters of *Povilla heardi* Hubbard, 1984 (Ephemeroptera: Polymitarcyidae)

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*Povilla heardi* Hubbard, 1984 is a common species of mayfly in urban area of Thailand. The nymphs were found in lentic habitats, whereas the adults were found near electric light after sunset. However, the developmental biology of *P. heardi* is poorly studied. To understand post-embryonic developmental stages, female adults were collected and induced to oviposition. After 10 days of hydration, embryonic nymphs were hatched and investigated for size and external morphology for 20 days. Head width (HW) and body length were significantly correlated ($r=0.856$, $p<0.01$) from the analysis of 213 nymphs. The nymphal instars were divided into four instar stages, each characterized by specific external features. In the first instar (HW < 0.17 mm.), abdominal gills were absent. In the second instar, nymphs developed six pairs of unbranched gills, and the basal segment of the antenna and caudal filament elongated. In the third instar, abdominal gills continue to increase branch number, while forelegs exhibit numerous fine setae. In the third instar, mouthparts were well developed, especially the mandibular tusks, which were more elongated and conspicuous in the fourth instars. These structures are sensitive indicators of molting changes. By unraveling the post-embryological development, we can gain deeper insights into the complex interplay between the form and function of this species. This information will contribute to understanding the life history patterns and applications in toxicity testing.

Keywords: Post-embryonic, development, *Povilla heardi*, mayfly

# The study of macrobenthic community in the South-east Sea of Korean Peninsular using SIMPER analysis

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The macrobenthic fauna on the southeastern coast of the South Sea of the Korean Peninsula was studied through community analysis and SIMPER analysis to determine microbenthic community habitats in the seabed. Surveys were conducted seasonally eight times at 43 stations for two years. Smith-McIntyre grab was used to collect some sediments twice at each station for animal identification and substrate analysis for particle size and TOC measurements. Macrobenthic animals identified at the species level formed four or more animal communities through cluster analysis using the Bray-Curtis similarity index and showed similar regional characteristics. These could generally be divided into groups A, B, C, and D, and their selection was based on the peak group to which they belonged more than four times during a total of 8 survey periods. As a result, cluster A in the area furthest from the coast. It could be divided into Cluster B, the middle area of the survey area; Cluster C, which includes the South Sea coast and Yokjido; and Cluster D, the coast of Geoje Island. The macrobenthic animals representing each community were determined as the species that contributed most to forming the community through SIMPER, and representative species of the community were selected with a contribution rate of more than 5% in each community, and the appearance of the contributing species more than five times during the eight survey periods. Accordingly, the amphipods *Ampelisca brevicornis* and *Haploops tubicola* and the annelid *Nothria shirikishinaiensis* were selected for group A, *A. brevicornis* and *Heteromastus filiformis* for group B, *H. filicormis* and *A. brevicornis* for group C, and no species that appeared more than five times in group D. There were four occurrences of *A.brevicornis*, three of *Photis longicaudata*, and three of *H. filiformis*. Group A maintains a very stable community despite being away from the coast, while group D varies so much that it is difficult to define it as a specific community.

Keywords: Macrobenthic community, South-east Sea of Korea, Cluster Analysis, SIMPER Analysis

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# Seasonal variation in the species composition of larval fish assemblages in the coastal waters of Ongjin, West Sea of Korea

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The West Sea of Korea is semi-enclosed by land. The water depth is relatively low, and its temperature can drop below 5℃ in winter and rise above 25℃ in summer. The distribution of plankton varies significantly over time and space by environmental conditions. Fish mortality rates are high in the larval stages of their life cycle due to harsh environmental conditions, making their survivability dependent on these conditions. Therefore, we conducted a distribution survey of larval fish to determine their species composition and seasonal fluctuations in the coastal waters of Ongjin-gun located in West Sea of Korea.

A survey was conducted seasonally in the coastal waters of Ongjin-gun over 12 times from 2020 to 2022. Water temperature and salinity were measured to determine the seasonal environmental characteristics of the surveyed sea area. The larval fish were collected at an angle using an RN100 net. The seasonal species diversity index of the larval fish was calculated. The cluster analysis with the Bray-Curtis similarity index was performed.

During the survey period, 7 orders, 13 families, and 18 species of larval fish appeared, of which four families and 9 species belonging to the Scorpaeniformes accounted for 50.0% of the total taxa. *Hexagrammos otakii* was the most dominant during the survey, accounting for 65.7% of the total appearance with 9,396 ind./1,000 $m^3$. Species richness and diversity were highest in spring. The cluster analysis with the Bray-Curtis similarity index showed two groups by season.

Keywords: Larval Fish, Ongjin-gun, Scorpaeniformes, *Hexagrammos otakii*

# Decoration of a portable case by a caddisfly larva: Its functional role in prey-catching and intra-population variation

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Many invertebrates “decorate” themselves or their structures by attaching foreign materials (e.g. plant litter, carcasses, and sand) on their exterior. Although invertebrate decoration appears to function in protection against predators or abiotic forces in many species, adaptive consequences and variations among individuals have rarely been demonstrated.

The larvae of stream caddisfly *Limnocentropus insolitus* construct unique portable cases with a stalk attached to streambed. They are predacious and catch drifting invertebrates while floating in water column. Some individuals decorate their cases by actively attaching small plant materials such as pieces of needle litter. There is large variation in amounts of pieces used for decoration among individuals even in a single population. We hypothesized that the case decoration facilitates prey-catching success. We also hypothesized that the amount of decoration varies according to flow environments in which larvae live because flow should influence the prey-catching behavior.

We conducted a laboratory experiment examining the influence of artificially removing the case decoration on larval prey-catching in a channel with both low- (13-15 cm $s^{-1}$) and high-velocity (42-44 cm $s^{-1}$) sections. Mean prey-catching success decreases by 10% after the removal of decoration under low-velocity condition. By contrast, the change in mean prey-catching success after the removal did not be detected under high-velocity condition. We collected last instar larvae from diverse habitats in a 200-m stream section. The amount of decoration was greater in larvae attached in substrate with lower velocity. This can be explained by adaptive significance of decorating behavior only under low-velocity condition.

Keywords: Decorating behavior, Predation, Variation, Adaptation, Aquatic insect

# Reproductive strategies in paternal care and unprecedented "male brood parasitism" in insects

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Reproductive strategy is extremely important for organisms because it directly affects their evolutionary fitness. "Parental care" is one of the strategies adopted by organisms to improve their fitness. However, even in the case of parental care, females often bear a large burden in raising offspring, and it is extremely rare for males to care for offspring alone ("paternal care"). Offspring care results in fewer re-mating opportunities, resulting in a greater reproductive cost for males. Under such conditions, paternal care has evolved in males of several animal taxa. However, how paternal care behaviour has evolved and how is it maintained, remains uncertain. In this study, we conducted a free-choice mating experiment in a laboratory for a typical paternal care exhibiting insect, *Appasus japonicus*, commonly known as the giant water bug. We also developed sensitive molecular markers for the giant water bug and conducted a "paternity testing" experiment using the markers we developed. The results of the "paternity testing" experiment showed unexpectedly low actual paternity rates. Moreover, "brood parasitism" between males was evident. This discovery is the first of its kind in the insect world, and constitutes a significant finding regarding the evolution of reproductive strategies.

Keywords: Belostomatidae, male brood parasitism, maternity, mating experiment, SSR marker

# Ecology and ecomorphology of larvae and pupae of the phytotelmata-dwelling *Tipulodina koreana* Baek and Bae '고려각다귀' (Diptera: Tipulidae)

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*Tipulodina koreana* is a phytotelmata-dwelling specialist that spends its immature stages in small water pools of tree holes. However, the ecology of the immature stage of *T. koreana*, which is currently unknown, was newly observed and recorded in this study. Diagnostic characters for eggs, larvae, and pupae of the Korean *Tipulodina* crane-fly are described, photographed and illustrated based on specimens collected from water pools of the hollow formed between Y-shaped trunks of a *Carpinus laxiflora* tree in Mt. Baekwoon-san, Gwangyang, Junla nam-do, Republic of Korea. Twenty larvae were reared in plastic containers to confirm the periods of egg, larva, and pupa. As results, the average length of egg period of *T. koreana* was five days, the larval period thirty five days, and the pupal stadium five days.

Keywords: Korean Tipulodina, crane-fly, phytotelmata-dwelling, ecology, water pool

# Diet analysis and risk screening of native and introduced fish species in Angat Watershed Forest Reserve: Implications on protection and management

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Diet analysis coupled with risk screening of fish communities in freshwater ecosystems provide timely information on trophic interactions and ecosystem health. As such, this study aimed to assess dietary composition and analyze invasiveness potential through gut content analysis and risk screening of introduced and one endemic fish species in the Angat Watershed Forest Reserve (AWFR). Water quality assessment was performed at six sampling sites around the dam, where four introduced species, *Amphilophus trimaculatus* (three-spot cichlid), *Clarias batrachus* (walking catfish), *Coptodon zillii* (redbelly tilapia), and *Cyprinus carpio* (carp); and one endemic species, *Arius manillensis* (Manila Sea catfish), were collected and analyzed. Results revealed fish components and *Chironomus* sp. as the dominant prey categories in all five fish species based on frequency of occurrence (%F), numerical composition (%N), and index of relative importance (IRI). Risk screening using the Aquatic Species Invasiveness Screening Kit (AS-ISK) provided information on the possibility of all four introduced species becoming invasive (i.e., high risk based on Basic Risk Assessment and Climate Change Assessment) and causing harm to native and economically important fish species in AWFR. Overall, this study underscores the reliability of dietary analysis and risk screening informing policies and encouraging actions that stakeholders can revisit and establish towards conservation efforts through management and prevention of high-risk introduced species in AWFR.

Keywords: Angat Watershed Forest Reserve, Dietary Habits, Gut Content, Invasiveness Risk Assessment

# The subtropicalization of temperate costal ecosystems in the East China Sea: Tropical rabbitfish and temperate kelp alga, *Ecklonia cava*

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Kelp forests mainly consisted in Ecklonia cava, a perennial endemic alga, serve ecologically diverse habitats and are economically important ecosystems in Korea and Japan. However, we confirmed the active feeding activity of tropical fish herbivores (*Siganus canaliculatus*) from the southern coast of the Korean peninsula in November 2023, and found E. cava with severe damage (i.e., bite marks and reduced lateral blade length) as a result. Sea surface temperature (SST) is a strong indicator of global climate change and an unusually higher SST in 2023 was observed in the northern East China Sea. The mean SST value in the study area from August to November 2023 were found to be 1.1°C higher than the SST anomaly averaged over the last two decades. Because *S. fuscescens* has been reported to have the highest feeding rate at water temperatures of 26-29°C, the critical damage of the E. cava population cause by the high activity and intensive grazing of rabbitfish is likely to be associated with the significant increase in SST in 2023. Furthermore, the timing of the year was been identified as the only annual reproductive peak for the temperate kelp species. Eventually, the decline of endemic *E. cava* in Korean waters is likely to be accelerated by the combined effects of global warming and invasive herbivory.

Keywords: *Ecklonia cava, Siganu*s, tropical fish herbivory; sea surface temperature, East China Sea

Acknowledgements: This research was funded by the Korea Institute of Ocean Science and Technology (KIOST), grant numbers PEA0206 (project title: Development of marine environment change management system at Southern sea and Jeju coast). This research was also supported by ‘regional innovation mega project’ program through the Korea Innovation Foundation funded by Ministry of Science and ICT (Project Number: 2023-DD-UP-0007).

# Diversity and distribution of Odonata nymphs in Nakhon Nayok and Rachaburi provinces, Thailand

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The diversity and distribution of Odonata nymphs were investigated during November-December 2023 at streams (10 sampling sites) of Ratchaburi and Nakhon Nayok provinces, Thailand. The nymphs were collected using the multihabitat approach. Selected physio-chemical parameters and visual-based habitat assessment were measured. A total of 462 individuals were identified as 10 families, 22 genera, 22 species, the most abundant species consist of *Pseudagrion rubriceps, Neurobasis chinensis* and *Rhyothemis plutonia*, while *Diplacodes trivialis* and *Hydrobasileus croceus* were found only one sampling site. The family Libellulidae made up the most dominant family collected with 27.92% of total individuals recorded, followed by Gomphidae (27.06%), Coenagrionidae (14.29%). PCA analysis revealed that *Gomphidia abbotti, Megalestes kurahashii, Euphaea decorate, Lamelligomphus castor, Orientogomphus minor*, and *Zygonyx iris* were strongly correlated with the reference sites. Bank structure, stream bottom and riparian vegetation were strongly associated with the distribution of tentative indicator species and reference sites. Disturbance of the stream condition is causing a decline in Odonata species diversity, community composition, and structure.

Keywords: Diversity, Distribution, Odonata, Anisoptera, Zygoptera

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# Temporal change in genetic structure of *Brachymystax lenok tsinlingensis* in Korean national parks: Conservation implications for endangered species

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An important consideration in the implementation of species management is the conservation of genetic variation, which is essential for the long-term persistence of populations and the adaptive potential of species. In particular, monitoring the demography and genetics of reintroduced populations is necessary to assess the success of reintroductions. The Manchurian trout, *Brachymystax lenok tsinlingensis*, is of high conservation concern in South Korea due to a decline in both distribution and abundance, and is now an endangered species. Conservation strategies for this species include protection of source populations (i.e. Hongcheon), introduction and reintroduction. Understanding changes in genetic diversity, fine-scale temporal genetic structure, and gene flow of *B. l. tsinlingensis* is critical for developing successful conservation strategies for this endangered species. In this study, we used mtDNA control region sequences and eight microsatellites from samples collected from two sites in Korean national parks, Chiaksan (introduced population) and Taebaeksan, to determine how genetic variation is structured over 10 years and the extent to which population structure has been influenced by introductions. We suggested that introductions for restoration should consider the genetic composition of regional populations and highlighted that assessing effective population size plays an important role in adaptability to future environmental change.

Keywords: translocation, freshwater fish, genetic drift, population genetics, threatened species

Acknowledgements: This research was supported by grant from the Korea National Park research Institute, Korea National Park Service in South Korea.

# Macroinvertebrate communities of some springs and oases of Trans-Altai Gobi Desert

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The Trans-Altai Gobi is the largest protected area in Asia and the fifth largest one in the world which a combination of unique Central Asian deserts, among which the highly continental and arid climate have no analogs anywhere in Asia. In addition to its unique ecosystems, the Trans-Altai Gobi Desert contains many oases that support as wildlife habitats and supports a unique biodiversity and provide vital ecosystem services. This study aimed to determine the diversity of benthic macroinvertebrates in springs and oases in the Trans-Altai Gobi Desert of Mongolia. We collected benthic macroinvertebrates using D-net in June and September between 2015-2021. A total of 7085 individuals were identified from 59 sampling sites, classified into 16 orders (Myodocopida, Diptera, Amphipoda, Anomopoda, Mollusca, Oligochaeta, Ephemeroptera, Nematoda, Trichoptera, Coleoptera, Cyclopoida, Odonata, Hemiptera, Gastropoda, Arachnida, Plecoptera), 52 families, and 59 genera. Myodocopida (31%), Diptera (29%) and Amphipoda (20%) were dominant groups among the macroinvertebrate assemblages. Macroinvertebrate community defined using taxon richness, relative abundance, the values of Pielou evenness index (E) and Shannon diversity index (H´). The highest taxon, richness, and diversity (H´=1.07) occurred at the Olon bulag spring, which provided the most heterogeneous habitats to the benthic fauna. In contrast, the lowest diversity was found at the Toli bulag spring. Benthic macroinvertebrate communities are relatively evenly distributed in the Khalzan us and Uyench river (E=0.99). Bray Curtis's similarity index was calculated using Primer 6 software. The result indicated that macroinvertebrate assemblage groups of Sukhantiin dund us and Sukhantiin us springs are revealed, related to a heterogeneous habitat. Therefore, it is essential to determine the environmental factors that influence the distribution and community patterns in Gobi springs and oases.

Keywords: Gobi water resources, benthic macroinvertebrates, diversity

# Prediction of the potential habitat of two non-native tilapia species, including the first record of a feral population of *Oreochromis aureus* (Steindachner, 1864) in South Korea

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It is challenging to undertake morphological species identification of the different species of tilapia that have been introduced and that inhabit many areas of the world. Most tilapia species farmed today are based on hybrid combinations of *O. niloticus, O. aureus* and *O. mossambicus*, and species identification requires both morphological and genetic analyses. Also, most tilapia species farmed is an invasive species that has become widely distributed around the world. Recently, in relation to climate change, many studies have been conducted to predict the potential habitat area and distribution range of tilapia and the suitability of habitat for each species. In the present research project, we had two primary analyses. First, we investigate an invasive tilapia population in the Dalseo Stream in Korea and conduct a morphological-based species identification and using the cytochrome oxidase (COI) gene, molecular-based phylogenetic species identification. Second, we predicted habitable areas and the possibility of spreading of habitat ranges of tilapia (*Oreochromis* spp.) inhabit domestic streams. The results of the morphological and genetic species identification analyses found that two species, *O. aureus* and *O. niloticus*, inhabit the Dalseo Stream. Thus, we observed for the first time that a new invasive species, *O. aureus*, inhabits a stream in Korea. In addition, in the case of tilapia, which lives in tributaries of the Geumho River, Dalseo Stream, and the Nakdong River, its range of habitat is expected to expand to the middle and lower of the Nakdong River system. Our study will aid in the management of invasive tilapia.

Keywords: morphological identification, molecular identification, invasive species of tilapia, first record in Korea, distribution of tilapia habitat

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# Forest types influence on stream fish community characteristics in South Korea

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Freshwater ecosystems are influenced by both intra-aquatic interactions and interactions with external ecosystems. Fish are key members of stream ecosystems and represent the health of streams. Forests, which are important resource contributors to streams, are generally known to be associated with healthy stream conditions. However, the effect of forest types on fish community characteristics in streams has not been sufficiently studied. Therefore, in this study, we analyzed differences in the characteristics of fish community according to forest types. In terms of fish community index, guild characteristics, and functional diversity, fish community showed various taxonomic and guild characteristics depending on forest type. Among the forest types, deciduous coniferous forests showed a negative correlation with the fish diversity index, omnivore rate, and pollution-tolerant species rate, and a positive correlation with the fish dominance index, invertivore rate, and sensitive species rate. Also, broadleaf forests, mixed coniferous and broadleaf forests, and Korean pine (*Pinus koraiensis*) forests showed similar trends to them. On the other hand, pitch pine (*Pinus rigida*) forests and chestnut forests showed the opposite trend to deciduous coniferous forests in terms of community structure. The study results provide ecological insight into the relationships between forest types and fish communities. These foundational data are expected to be useful in ecosystem management considering both forest and freshwater ecosystems.

Keywords: Fish community, community index, guild characteristics, Forest type, Freshwater ecosystem

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# Effects of photoperiods on the development and predatory capacity of the giant water bug *Diplonychus rusticus* Fabricius, 1871: Application in studying the effects of light pollution

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The giant water bug *Dyplonychus rusticus* shows high potential in consuming mosquito vectors transmitted diseases in urban areas. However, light pollution in these areas nowadays has critical effects on insects such as altering species interactions and changes insects communities. To evaluate the effects of light pollution on *D. rusticus*, a series of experiments were performed in laboratory conditions including (1) an acute test to evaluate the effects of full-time exposure on the predatory capacity of adult bugs; (2) the effects of photoperiods on the development, survival and fertility of the bugs. The predatory capacity of adults under the 24-hour lighting condition showed a slight increase on the first day, followed by a significant decrease on the third day. However, it gradually recovered in the subsequent days. The development time of the bugs from the first instar to adulthood is positively correlated with the duration of light exposure whereas photoperiods did not affect the morphometry (length and width) at any life stages. Further, survival and fertility of adult bugs is considerably decreased under the conditions of prolonged lighting. The lighting condition may therefore strongly affect to aquatic bug *D. rusticus* by increase the developmental rate but could have downstream consequences for the reproduction and fitness of adults.

Keywords: *Diplonychus rusticus*, light pollution, biological conservation

# *Diplonychus esakii* in Korea: Genetic diversity and foundational data for conservation efforts

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*Diplonychus esakii*, a water bug belonging to the family Belostomatidae, is a crucial species in freshwater ecosystems across East Asia, particularly in Korea. This study aims to assess the genetic diversity and population structure of *D. esakii* to provide foundational data for conservation efforts. We collected 318 specimens from 27 sites across the Korean peninsula and conducted genetic analyses using *COI* sequencing. The results show significant genetic diversity in Changnyeong-gun, Gyeongsangnam-do, especially near the Upo Wetland ($h=0.857$, $\pi=0.181$), which is a Ramsar-protected area. However, low genetic diversity and restricted gene flow were observed in Jeju-do populations, indicating potential genetic bottlenecks. These findings highlight the necessity for targeted conservation strategies, such as treating each population as an independent management unit and enhancing habitat connectivity. Continuous monitoring, especially in geographically isolated regions like Jeju-do, is essential to ensure the long-term viability of *D. esakii* populations.

Keywords: *COI* sequencing, conservation biology, *Diplonychus esakii*, genetic diversity, mitochondrial DNA, population structure

Acknowledgements: This work was supported by a grant from the National Institute of Biological Resources (NIBR), funded by the Ministry of Environment (MOE) of the Republic of Korea (NIBR202402108).

# Biodiversity of zoobenthos of a cold spring in the south of Primorsky Krai and its role as a winter refuge for terrestrial arthropods

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The first data on crenobiota of the Southern Russian Far East is presented. Fauna of invertebrates associated with a small cold spring located on the Muravyov-Amursky Peninsula is examined. The water temperature at the source of the spring, classified as helocrene, remained at about 8°C throughout the year. Macrobenthos of the spring includes at least 30 species from 25 genera and 20 families in Arthropoda, Platyhelminthes and Annelida. Main groups of benthos – amphipods, planarians, larvae of dipterous insects, caddisflies and stoneflies. The larvae of mayflies, riffle beetles, adult minute moss beetles and oligochaetes are characterized by lower numbers. The ecological groups of benthos are also diverse. Here, rheo- and stagnophiles, highly specialized stygo- and crenobionts, and species with wide ecological niches, sharply differing in their preferences, closely coexist. Hydrophilic springtails, limoniids *Chionea*, rove beetles *Gabrius*, woodlouse *Hyloniscus riparius*, as well as spiders Lycosidae and Amaurobiidae, were found at subzero air temperature near the helocrene. This indicates a significant influence on the fauna of terrestrial invertebrates by the microclimate around not only thermal, but also cold springs.

Keywords: cold spring, crenobiota, macrobenthos, aquatic and hydrophilic invertebrates, microclimate

# The role of aquatic plants in restoring river ecosystems

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Abstract: This study was conducted to find a restoration method suitable for typical polluted rivers in Jiangsu Province, China. A segment of a representative river in Tianmu Lake Basin was selected as a research area through previous investigation, and the polluted river was cleaned and restored by three methods, i.e., artificial wetland, floating island type wetland and purification floating island. Floating plants (*Hydrocotyle verticillata* and *Myriophyllum spicatum*) showed better restoration effects than emergent aquatic plants (*Iris wilsonii, Arundo donax*, reed, water-cultured *llex chinensis* and *Lythrum salicaria*). The two types of plants showed the removal rates of total nitrogen (TN) of 43.2% and 40.7%, respectively, the removal rates of total phosphorous (TP) of 85.5% and 78.9%, respectively, the removal rates of chemical oxygen demand (COD) of 83.9% and 81.2%, respectively, the removal rates of ammonia nitrogen (AN) of 81.7% and 82.9%, respectively, and the removal rates of suspended solid (SS) of 67.1% and 71.3%, respectively. Among the three methods of river restoration, the purification floating island technique exhibited the best removal effect, with the removal rates of 82.4%, 75.5%, 95.9%, 87.7% and 79.6% for TN, TP, COD, AN and SS, respectively. The nitrification and denitrification rates of bottom mud and water samples in the engineering segment were remarkably higher than those in the reference segment by 14% and 22%, respectively. Overall, the purification floating island has a significant restoration effect on typical polluted river in Jiangsu.

Keywords: river ecosystems, removal rates, aquatic plants, Tianmu Lake Basin

Acknowledgements: This study was financially supported by the Youth Innovation Promotion Association, Chinese Academy of Sciences (2020317).

# The study of marine aggregate dredging impact on macro-benthic assemblage in the open sea

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The impact of marine sediment extraction on macro-benthic fauna was investigated. The investigation was carried out over two years at 12 points, including 4 points in the harvested area and 8 points in the non-collection area. We investigated the differences between non-excavating areas and attempted to reveal the specific effects of marine sand extraction. In addition, sea sand was collected during the 1st to 4th survey periods and not during the 5th to 8th survey periods. However, in Cluster analysis and MDS analysis using the Bray-Curtis similarity index, there were differences between the extraction area and surrounding areas in most surveys. Nevertheless, there were no statistical differences in the number of species, density, biomass, and ecological indices (abundance, evenness, diversity, dominance), and BPA (Benthic Polychaete Amphipoda) Analysis, which is sensitive to physical disturbance, was also collected. Although the frequency of midges was high in the area, statistical significance was shown only in the third survey. Looking at the species that contributed to community formation in the collection area, indirect influence area, and control area through SIMPER analysis, *Heteromastus filiformis*, a polychaete that frequently appears on fine-grained substrates, was found to be the main contributed species and dominant species in the collection area during the sea sand collection period. In the indirectly affected area and the control group, the amphipod *Ampelisca brevicornis* appeared as a significant contributing and dominant species. After the extraction of sea sand stopped, not only *A. brevicornis*, the dominant species in the non-extraction area, but also various species appeared as contributing and dominant species in the extraction area.

Keywords: Marine Aggregate Dredging Impact, Macro-benthic Assemblage, BPA Analysis, SIMPER Analysis

# Benthic macroinvertebrate communities reveal urban river health in Batangas and Zambales, Philippines: Does protection status influence urbanized watersheds?

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Influence of protection efforts in streams situated in urban settings remain unclear in the Philippines. Despite the ecosystem services provided by urban lotic environments, these are exposed to pollution and overexploitation. In this study, we aimed to assess the response of benthic macroinvertebrate communities to the gradients of environmental conditions in protected and unprotected watersheds in Batangas and Zambales, Philippines. Benthic macroinvertebrates and physicochemical parameters were sampled across 10 sites categorized by land use and protection status. Multivariate analyses compared water quality between protected and unprotected streams and its influence on benthic community responses. Diversity indices, including the key invertebrate index (EPTC), Shannon Diversity Index and Evenness, were calculated to assess macroinvertebrate community structures. On one hand, protected streams in Batangas with lower dissolved oxygen and higher total dissolved solids and conductivity were correlated with the abundance of pollution-tolerant species (e.g., *Chironomus* sp., *Lamprodrilus* sp.). On the other hand, unprotected streams with better water quality in the headwaters of Zambales are associated with pollution-sensitive taxa (e.g., *Acentrella* sp., *Hydropsyche* sp.). Regardless of these specific responses, both watersheds were observed to have degrading water quality, increased nutrient inputs (i.e., ammonia, phosphate), and low macroinvertebrate diversity. Despite contrasting protection statuses, urban watersheds in Batangas and Zambales revealed poor stream ecosystem conditions affected by anthropogenic activities. This study underscores the need for immediate and definitive measures for Philippine urban streams and to implement mitigation and management strategies toward watershed governance and rehabilitation.

Keywords: Amphipod, Aquatic insect, CCA, EPTC, Multivariate Analysis, Ecosystem Management

# Macroinvertebrates of the riverbank of Eg and Delgermurun rivers, Mongolia

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Macroinvertebrates are abundant in riverbanks than the middle parts of the river depending on the seasonality. Studies of macroinvertebrates in riverbanks often ignored in our country. Within the framework of the “MACRO” project, a study along the Eg and Delgermurun rivers provides an opportunity to make research comparison between the riverbanks and the middle parts of the rivers.

We collected macroinvertebrate samples from 12 sites in Delgermurun and Eg rivers in September 2017, by using d-frame net with 10 jabs within the 200 meter reaches of riverbanks. A total of 6857 individuals of invertebrates belonging to 61 genera of 36 families, 15 classes were identified from this collections. We used previous identified macroinvertebrate data (riffle, run and pool habitats) from middle parts of rivers in order to compare the riverbanks and middle parts. Chemical and physical parameters were measured by portable equipment, riparian vegetation and substrate compositions were recorded by researcher’s sight evaluation. Measures of biodiversity include species richness, abundance, the diversity index, the dominance index, evenness and the similarity index. There are no differences of macroinvertebrate abundance and richness among the riffle, run, pool and riverbanks. However, they differ in diversity index and evenness among these four habitats. Macroinvertebrates of riverbanks were less diverse and less abundant than other three habitat types. According to the similarity index, the most similar habitats are the communities of riffle and run habitats, with 62% similar. On the riverbank, order Trichoptera are more abundant than other groups compared to the other habitat types.

We checked the most influencing factors of environmental parameters to the macroinvertebrates of river banks. Boulders are negatively affecting the macroinvertebrate richness, while grazing intensity of riparian vegetation negatively affect the macroinvertebrate evenness.

Keywords: macroinvertebrate diversity, riverbank, habitat types, environmental parameters

# Longitudinal patterns of macroinvertebrate functional feeding groups in the Loei River, Thailand

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The Loei River is a tributary of the Mekong River located in northeast Thailand, known for its high biodiversity of fauna and flora species. This study was conducted to investigate whether functional feeding groups (FFGs) of macroinvertebrates could be used as surrogates for biological assessments. The research involved surveying the composition and abundance of macroinvertebrate assemblages' FFGs along the Loei River in Loei Province, in November 2023 and January 2024. Three replicates were collected from five sampling sites using a D-flamed net (450 micrometers). The abundance of the macroinvertebrates was calculated using Shannon-Wiener, Simpson, and Pileou's evenness indices. A total of 3,755 individuals were collected from 9 orders, 30 families, and 64 genera. Ephemeroptera (8 families, 22 genera), Hemiptera (7 families, 19 genera), and Trichoptera (3 families, 8 genera) were the most diverse orders. According to the FFG proportions, collector-gatherers were the most abundant in the Loei River and represented 52.9% of the macroinvertebrate assemblages, while shredders were absent in all sampling sites. The FFG ratios indicated that most of the five sites were strongly heterotrophic, and streams received additional energy sources from leaf litter and other organic matter, with fine particulate organic matter (FPOM) being the primary food source. FPOM was found in suspended aggregates and as sedimentary deposits. Also, the study found that site L3 in Kut Pong Subdistrict, Mueang District, had the most stable substrate. Therefore, using FFGs can be a valuable tool for assessing and monitoring anthropogenic impacts on the river.

Keywords: Macroinvertabrates, Functional feeding groups, Loei River, Mekong River tributary

Acknowledgements: Mahasarakham University Development Fund

# Research on collecting methods for benthic macroinvertebrates in Korean sedimentary ecosystems

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Sediment, one of the factors that make up sedimentary ecosystems formed under the substrate of streams and lakes, varies in particle size from small to large, and contains heavy metals and various ions between particles. In particular, fine-grained sediment primarily limits the oxygen supply of sedimentary ecosystems to create an anoxic state, and as a secondary result, toxic chemicals such as ammonia and hydrogen sulfide are generated, which is likely to directly or indirectly affect benthic macroinvertebrate community. Although continuous research is needed to deeply understand the relationship between fine-grained sediment and benthic macroinvertebrates, research related to sedimentary ecosystems is insufficient in Korea, and no collecting method has been proposed, so related research is absolutely necessary. A core sampler was used to present standards for collecting methods for the study of sedimentary ecosystems, and four factors (inner diameter size, number of collections, collecting depth, the pore size of sieve) deemed appropriate for Korea were selected among the methods studied in existing literature. As a result, standards for research methods suitable for Korea were presented for each factor, based on the abundance and species numbers of the benthic macroinvertebrate.

Keywords: sediment, collecting method, benthic macroinvertebrate, core sampler

Acknowledgements: This work was supported by Korea Environmental Industry & Technology Institute (KEITI) through Aquatic Ecosystem Conservation Research Program, funded by Korea Ministry of Environment (MOE) (2022003050006).

# Freshwater species biomass calculation: Introduction of R package

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Freshwater ecosystems, where various organisms live and are essential for human life, require biomonitoring and health assessments to conserve their biodiversity and environments. Because traditional assessment based on species abundance and richness is dependent on numbers, it does not reflect the characteristics of various species and taxa well. Therefore, in addition to assessment methods based on population and species numbers, energy flow defined by species biomass should be considered. However, the calculation of biomass is very complicated and time-consuming. As an alternative, we developed an R package to support the calculation of species biomass, taking into account the morphological traits of various species and taxa. This package consists of a database that helps convert to biomass using morphological characteristic information of various taxa (phytoplankton, zooplankton, fish, and benthic macroinvertebrates), and some functions that automatically and quickly calculate species biomass. In this package, biomass is calculated by entering the abundance and information of species. With this package, researchers can now easily obtain biomass information and enhance their research for health assessment and environmental conservation in freshwater ecosystems.

Keywords: Aquatic Ecosystem, Biomass Calculation, Freshwater Organisms, Morphological Database

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# Impact of drought severity on benthic macroinvertebrate communities and functional diversity: A mesocosm wetland experiment

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This study conducted a mesocosm wetland experiment to compare the effects of drought severity on benthic macroinvertebrate (BMI) communities and functional diversity. Mesocosm wetlands were categorized into control, short-term drought (spring drought), and long-term drought (winter-spring drought) conditions. Based on colonization stages, the wetlands were further divided into early colonization (1 phase), mid-colonization (2 phase), and late-colonization (3 phase), resulting in a total of nine comparison groups. The average temperature during the study period was 12.5±10.5°C, and the total rainfall was 3,015 mm, reflecting monsoon climate characteristics. Dissolved oxygen, water temperature, pH, and electrical conductivity showed no significant differences among the control, short-term and long-term drought conditions, although water depth was higher in the control conditions compared to the short-term drought conditions. BMI community analysis revealed differences before and after drought, and NMS analysis differentiated BMI communities based on drought severity. Cluster analysis using the Bray-Curtis similarity grouped the nine conditions into three clusters at 70% similarity: Group A (early colonization), Group B (only control condition), and Group C (short-term and long-term conditions). Comparisons of community indices and functional diversity indices among the nine groups showed significant differences in abundance and functional richness between the long-term drought conditions 1 and 3 phase. Functional diversity indicated that species with various biological traits survived and adapted differently under drought conditions. Specifically, diverse functional traits were observed under drought stress, confirming that BMI communities were affected by drought stress.

Keywords: Mesocosm, wetland, benthic macroinvertebrate, functional diversity, drought, biological trait

# Evaluating ecosystem integrity: Benthic macroinvertebrate multimetric index for Veli-Akkulam Lake, south India

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The Macroinvertebrate Index of Biotic Integrity (M-IBI) was developed to assess the ecosystem integrity of Veli-Akkulam Lake in south India over a two-year period. Sampling of macroinvertebrates across multiple habitats was conducted at both Veli-Akkulam Lake and the nearby reference site, Vellayani Lake, resulting in the collection of 246 taxa spanning 113 families. Evaluation of more than twenty-five benthic metrics revealed significant correlations (Pearson's correlation, $p<0.01$) between key water quality parameters such as dissolved oxygen, biochemical oxygen demand (BOD), chemical oxygen demand (COD), phosphate, and seventeen benthic metrics. Sensitivity analysis distinguished test and reference sites, while redundancy among metrics was examined ($r>0.85$). From this assessment, nine metrics were selected for the development of the macroinvertebrate index, including the number of intolerant taxa, Average Score Per Taxon (ASPT), percentage of tolerant individuals, Biological Monitoring Working Party (BMWP) score, and percentages of Ephemeroptera, Plecoptera, and Trichoptera (EPT), dipteran taxa, gatherers-collectors (GC), grazers-scrapers (GR-SC), and clinger taxa. The resulting Index of Biotic Integrity (IBI) ranged from 11 to 19 for the polluted Veli-Akkulam Lake, contrasting with the reference site's score of 45, indicating superior ecosystem integrity and water quality. The findings were juxtaposed with those derived from the Ecopath model developed for the system. The study underscores the efficacy of employing the macroinvertebrate biotic integrity index as a pragmatic tool for conducting long-term assessments of lacustrine ecological integrity. Consequently, the outcomes have catalyzed urgent actions by authorities towards restoring the health of Veli-Akkulam Lake through targeted eco-restoration endeavours aimed at pollution abatement.

Keywords: Biomonitoring, macroinvertebrates, Index of Biotic Integrity, water quality, Ecopath model

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# Analysis of mass emergence in the common burrowing mayfly *Ephemera orientalis* (Ephemeroptera: Ephemeridae) in Namyangju City, South Korea

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This study was conducted to analyze the patterns of mass emergence events of *Ephemera orientalis* in Namyangju City by examining their ecological environment and monitoring their life cycle in 2023. The study areas were selected based on locations along the Han River where residents suffer significant damage due to massive *Ephemera orientalis* outbreaks. The tributaries of the Han River (WS) and the main stream of the Han River (GC) were chosen, with Dangjeong Island in Hanam (DJ) included as a control site. The basic data for ecological environment analysis showed that the average air temperature was 21.8°C for WS and 22.1°C for both GC and DJ, indicating a similar trend. The average water temperature was 22.4°C for WS, 21.5°C for GC, and 21.4°C for DJ, with the tributaries showing slightly higher temperatures compared to the main stream and control site. The average density of larvae (individuals/$m^2$) was 21 for the WS, 71 for GC, and 117 for DJ. The average density of adults (individuals/times) was 5,253.5 for WS, 14,533.6 for GC, and 10,587 for DJ, confirming that *Ephemera orientalis* were more prevalent in the main stream of the Han River than in its tributaries. The peak density of adult *Ephemera orientalis* was observed on September 11 for WS, on September 2 for GC, and on May 19 for DJ. During the survey period, the mess emergence occurred once in its tributaries and twice in the main stream of the Han River.

Keywords: *Ephemera orientalis*, Mass emergence, Life cycle, Ecological analysis, Han River, Namyangju City

Acknowledgements: Namyangju City, Namyangju City Health Center

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# Identification of medically and forensically relevant flies using a decision tree-learning method

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Flies play an important role in medical and forensic entomology worldwide. Each developmental stage (eggs, larvae, pupae, and adults) can be utilized as evidence at crime scenes. Accurate species identification is an essential prerequisite for the subsequent analysis of the respective stage. In general, there are two common identification methods using morphological and molecular approaches; however, some limitations have been reported. Since machine learning already plays a major role in many areas of daily life, such as education, business, industry, science, and medicine, its use as a classification tool for insects is interesting. Morphometric analysis of wing shape is widely used to identify flies. This data could be analysed in more detail using machine learning, as shown in this study, which applies the decision tree method to construct a model for distinguishing adult flies of three families [Calliphoridae, Sarcophagidae, Muscidae] and seven species [*Chrysomya megacephala* (Fabricius), *Chrysomya rufifacies* (Macquart), *Chrysomya* (*Ceylonomyia*) *nigripes* Aubertin, *Lucilia cuprina* (Wiedemann), *Hemipyrellia ligurriens* (Wiedemann), *Musca domestica* Linneaus, and *Parasarcophaga* (*Liosarcophaga*) dux Thomson]. One hundred percent overall accuracy was obtained at family level, followed by 83.33% at species level. Interestingly, the interaction of wing morphometric data with morphology (body and gena color) improved species' model performance up to 99.05%. The results of this study provide non-taxonomists and/or –entomologists a tool for identifying adult flies from a field survey or a crime scene and the results of this work might be useful in the future to develop an application that is easy to use.

Keywords: decision tree, wing morphometric, identification, forensic entomology

# Occurrence of microplastics in life-cycle of mosquitoes from Chiang Mai urban wetlands

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Plastic pollution significantly affects freshwater and biota ecosystem health nowadays. Mosquitoes, as link of aquatic and terrestrial environments, have potential as indicators of microplastic (MPs) contamination across ecosystems. This study aim to investigate the occurrence of MPs in surface water, sediment, and mosquito larvae in two different sites were chosen of Mae Kha Canal, Chiang Mai, Thailand. Then physico-chemical parameters were characterized. Sampling was conducted during wet and dry seasons in 2023, there were surface water, sediment at depths of 0-30 cm and a total of 20 mosquito larvae individuals were collected then examined MPs. Three replicates of surface water samples were digested by Wet Peroxide Oxidation, and three replicates of sediment and mosquito larval samples were chemically digested. Subsequently, they were quantified MPs under a stereo microscope. Then Fourier transforms infrared spectrophotometer (FTIR) was using to classify plastic types. The results during wet season showed MPs lengths in sediment were 100–250 and 250–500 µm, whereas more than 1,000 µm in surface water and mosquito larval. The occurrence of MPs was found to be more prevalent in sediment samples during the dry season than during the wet season, but in wet season MPs greater found in mosquito larval. MPs in the surface water presence of slightly fluctuated about 5 to 12 pieces. Copolymer PP/PE and polyester were the most common types of MPs found. This study increases knowledge regarding MPs in Mae Kha canal and mosquitoes for pollutant management in the future.

Keywords: microplastic, mosquitoes, Chiang Mai, wetlands

Acknowledgements: Freshwater Biomonitor Research Laboratory (FBRL) and Environmental Science Research Center, Chiang Mai University, Thailand

# Effects of air pollution-contaminated water on life cycle of urban mosquito larvae *Aedes aegypti* and *Culex quinquefasciatus* (Culicidae)

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*Aedes aegypti*, a vector for dengue fever, is becoming more prevalent due to climate change, while *Culex quinquefasciatus*, a carrier of arboviruses and parasites, shows strong adaptability to urban environments. Moreover, the air pollution situation becomes more severe during the dry season in Chiang Mai, Thailand, and this might contaminate water resources, including urban mosquito habitats, such as those of *Ae. aegypti* and *C. quinquefasciatus*. This study aimed to observe the survival rate and developmental time of *Ae. aegypti* and *C. quinquefasciatus* reared in both uncontaminated and air pollution-contaminated water. To prepare uncontaminated conditions free from air pollution, deionized water and rainwater from the non-haze season 2022 were used for rearing larval mosquitoes. Meanwhile, during the haze season of 2023, air pollution-contaminated water was prepared by placing empty plastic containers on the rooftop of the Faculty of Science (SCB1), Chiang Mai University, for 21 days (average range of PM2.5, 106 μg/m$^3$), then filling them with deionized water to create artificial mosquito habitats. First instar *Ae. aegypti* and *C. quinquefasciatus* were reared in 300 ml of water with 30 larvae per condition, i.e., deionized water, rainwater, and air pollution-contaminated water. As a result, the highest survival rate was observed in *Ae. aegypti* reared in air pollution-contaminated water. The developmental time of *Ae. aegypti* and *C. quinquefasciatus* in air pollution-contaminated water was significantly shorter than in deionized water and rainwater. The study suggests that air pollution-contaminated habitats affect the life cycles and developmental times of both *Ae. aegypti* and *C. quinquefasciatus*.

Keywords: *Aedes aegypti*, air pollution, Chiang Mai, *Culex quinquefasciatus*, life cycle, Thailand

Acknowledgements: We thank the Korea Disease Control and Prevention Agency (KDCA) for the KDCA-DVP 2024 Grant Program (R67EX00317) and Fundamental Fund fiscal year 2023, under project “Source Characterization and Health Impacts Assessment of Size Fractionated PM and Air Pollution in Northern Thailand” for financial support partially.

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# Development of harmful cyanobacteria adsorption materials utilizing biologically derived substances

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In recent years, the occurrence of algal blooms has been on the rise due to factors such as climate change and nutrient influx. In response, this study aimed to develop and apply an adsorption-based method for controlling the proliferation of harmful cyanobacteria, which frequently occurs during the summer season, contributing to algal blooms. Microcystis aeruginosa, a predominant harmful cyanobacterium associated with algal blooms, was selected as the target organism in our research. Harmful cyanobacteria are known to carry a positive surface charge in aquatic environments. In this study, we harnessed positively charged adsorption materials to effectively control these organisms. Additionally, we modified chitosan, a biopolymer, and cellulose from cotton, focusing on compounds such as urea and benzylamine that have a known interaction with cyanobacteria to induce their aggregation on material surfaces, facilitating effective adsorption. We verified the potential of these materials for controlling harmful cyanobacteria at the laboratory scale, utilizing surface analysis and SEM imaging to gain insights into the material surface properties and confirm cyanobacterial aggregation after adsorption. These materials hold promise for practical applications in the field to counteract the proliferation of harmful cyanobacteria, thus addressing the issue of algal blooms.

Keyword: Harmful algal blooms, cyanobacteria, adsorption

# Machine learning prediction and interpretation of the impact of microplastics on soil properties

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The annual microplastic (MP) release into soils is 4–23 times higher than that into oceans, significantly impacting soil quality. However, the mechanisms underlying how MPs impact soil properties remain largely unknown. Soil-MP interactions are complex because of soil heterogeneity and varying MP properties. This lack of understanding was exacerbated by the diverse experimental conditions and soil types used in this study. Predicting changes in soil properties in the presence of MPs is challenging, laborious, and time-consuming. To address these issues, machine learning was applied to fit datasets from peer-reviewed publications to predict and interpret how MPs influence soil properties, including pH, dissolved organic carbon (DOC), total P, $NO_3^-$-N, $NH_4^+$-N, and acid phosphatase enzyme activity (acid P). Among the developed models, the gradient boost regression (GBR) model showed the highest $R^2$ (0.86–0.99) compared to the decision tree and random forest models. The GBR model interpretation showed that MP properties contributed more than 50% to altering the acid P and $NO_3^-$-N concentrations in soils, whereas they had a negligible impact on total P and 10–20% impact on soil pH, DOC, and $NH_4^+$-N. Specifically, the size of MPs was the dominant factor influencing acid P (89.3%), pH (71.6%), and DOC (44.5%) in soils. $NO_3^-$-N was mainly affected by the MP type (52.0%). The $NH_4$+-N was mainly affected by the MP dose (46.8%). The quantitative insights into the impact of MPs on soil properties of this study could aid in understanding the roles of MPs in soil systems.

Keywords: UN SDGs, Sustainable Waste Management, ESG, Sustainability, Artificial Intelligence, SDG15 Life on Land

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# Adsorbent reinforced with bacterial-based compounds enhances the efficiency and selectivity of controlling harmful cyanobacteria, *Microcystis aeruginosa*

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In the present study, algicidal bacteria cultivated aqueous medium was applied as surface modification agent to develop efficient adsorption for the removal of *Microcystis aeruginosa*. The modification gave positive effect enhancing *M. aeruginosa* cell removal efficiency. In addition, it was confirmed that the introduction of bio-compound secured removal specificity against *M. aeruginosa*. It was further confirmed that the adsorption process using developed adsorbent might be environmentally safe through subsequent cyanotoxin release and acute toxicity tests. Furthermore, the practical feasibility of adsorptive removal of *M. aeruginosa* was confirmed through the cell removal tests using developed adsorbent, performed in scaled-up reactor (50 L and 10 tons). In this test, the effects of adsorbent application type, water temperature, and initial cell concentration on the adsorption efficiency against *M. aeruginosa* was evaluated. Our results will newly suggest the valorization strategy of biological algicides recreating as adsorbents and provide the practical operational data of developed adsorbents for effective *M. aeruginosa* removal process in scaled-up conditions.

Keywords: Harmful algal blooms, Adsorption, Surface modification, selectivity, practical application

# Application of biochar in concrete

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The continuous rise in global temperatures is an evidence of climate change. $CO_2$ emissions have caused major problems owing to its contribution to climate change. In particular, the construction industry has a considerable carbon footprint. Therefore, investigations into climate change mitigation are indeed a priority. All steps in the construction process, from raw materials preparation to cement production, contribute to $CO_2$ emissions. This can be mitigated to a certain extent by incorporating bio-based constituents into construction materials. However, bio-based materials may negatively affect cement reaction and structural performance, despite their positive environmental impacts. Biochar, a carbon-rich product of biomass pyrolysis, is considered a potential substitute for cement replacement that can enhance structural properties if used in appropriate amounts. Although biochar has conventionally been used as a soil amendment in the agricultural industry, researchers have recently investigated its applicability in concrete. Importantly, the results thus far have reported its contribution to the enhancement of the mechanical, thermal, and physical properties of cement. This review provides a comprehensive overview of the physicochemical properties of biochar added cementitious materials, including the fresh and hardened properties of biochar-cement mixtures considering both environmental and economic aspects.

Keywords: Biochar, Cement composites, Construction materials, Pyrolysis, Climate change mitigation, Carbon neutrality

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# Towards more nature-positive outcomes: A review of corporate disclosure and decision-making on biodiversity

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Biodiversity loss and nature degradation are pressing issues with huge impacts on our society and economy. Businesses, investors, and regulators have a growing focus on corporate efforts to support biodiversity and positive actions for nature. Here we provide a comprehensive overview of the topics ranging from, the importance of biodiversity in corporate settings, the materiality of biodiversity and the role of mandatory and non-mandatory regulations on corporate environmental reporting and sustainability disclosure frameworks. We also provide descriptive information on the evolution of sustainability frameworks with a comparison of key factors of the most prominent sustainability frameworks with a special focus on the materiality approach and biodiversity-related disclosure recommendations. Further, it provides suggestions for more holistic approaches to improve the future sustainability frameworks focusing on the biodiversity impact while showing the necessity of more focus on the decision-making paradigm. Further research on measuring biodiversity impact is required, as well as innovative trends in sustainability reporting to better reflect nature-positive outcomes in corporate sector businesses.

Keywords: ESG, UN SDGs, Sustainability Report, Corporate Environmental Reporting, Resilience

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# Role of microplastics in cyanobacteria toxicity to Daphnids: A critical review

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Cyanobacterial blooms have been increasing worldwide due to anthropogenic factors and the growing issue of climate change. Cyanotoxin production increases the threat posed by cyanobacteria to aquatic organisms. Microplastics are not only an environmental problem in themselves but also have the potential to promote the growth of cyanobacteria and act as vectors for the uptake of cyanotoxins by aquatic organisms, especially *Daphnia*. However, knowledge of the effects of microplastics in promoting cyanobacteria growth and associated toxicity to *Daphnia* is still limited. Therefore, this study aims to reveal the significance of this issue. First, the effects of cyanobacteria on *Daphnia*, including growth and reproduction inhibition, physical interference, and reactive oxygen species generation are comprehensively reviewed. Second, the overall effect of microplastics on *Daphnia* and contributory factors are reviewed. Third, cyanobacteria/microplastics interactions and their potential to increase toxicity are discussed. This review provides insight into the ecotoxicological risks of microplastics as cyanotoxin vectors.

Keywords: Microplastics, cyanotoxins, daphnids, combined toxicity, aquatic ecosystems

# A review of biodegradation and formation of biodegradable microplastics in the soil and freshwater environment

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Plastic pollution is a critical environmental concern, as conventional plastics (CPs) endure in ecosystems for centuries. In response, biodegradable plastics (BPs) have emerged as a potential solution to limit plastic waste accumulation. However, the fate of BPs in the environment, particularly their degradation and the subsequent generation of biodegradable microplastics (BMPs), remains poorly elucidated. This review aims to provide a comprehensive insight into the process of biodegradation and the formation of BMPs across soil and freshwater systems. The degradation of BPs involves intricate interactions among physical, chemical, and biological elements. Microorganisms play a pivotal role in this process by dismantling polymer chains into smaller particles. Factors influencing the rate of biodegradation encompass polymer composition, environmental conditions (e.g., temperature, ultraviolet radiation, pH), and the presence of additives. Nonetheless, incomplete degradation can lead to the creation of BMPs, potentially perpetuating their presence in the environment and posing risks to ecosystems and organisms. Understanding the mechanisms governing biodegradation and the genesis of BMPs is imperative for evaluating their environmental consequences and devising effective strategies for managing plastic waste. This review consolidates existing knowledge on biodegradation processes and the formation of BMPs, pinpointing knowledge gaps and charting paths for future research.

Keywords: Biodegradable plastics, Biodegradation, Soil, Freshwater, Microplastics

# The sublethal toxicity (48 h) for Diflubenzuron on water flea, *Daphnia magna*

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Diflubenzuron (DFB), an insect growth regulator pesticide extensively utilized in agricultural practices, poses a considerable threat to aquatic ecosystems due to its pervasive contamination. DFB have mode of action related to inhibition of the formation of chitin, the epidermal tissue of insect bodies, and has a mechanism to inhibit the hatching of insect eggs, so it can be toxic to water fleas such as crustaceans. In this study, we conducted a thorough eco-toxicity to elucidate the sublethal effects of DFB on *Daphnia magna*, a keystone species in freshwater ecosystems. DFB were exposed to *D. magna* during the acute period (48 hour), mortality and immobilization were assessed, and oxidative stress was analyzed. Gene expression and chitin will be analyzed. For acute toxicity was exposed in concentration from 6.5 to 200 ng $L^{-1}$ for 48 hours. The EC50 value for DFB in *D. magna* was 51.76 ug $L^{-1}$ (CI: 45.65-58.70) which revealed a concentration-dependent increase in immobilization and mortality. In previous study, DFB's interference with the molting process in *D. magna* emerged as a critical factor contributing to growth and developmental disruptions. This study provides comprehensive insights into the sublethal toxicity of DFB, encompassing acute mortality, sublethal effects, and disturbances to vital life history traits. The findings underscore the urgent imperative for holistic risk assessments of pesticides to safeguard the health and resilience of aquatic ecosystems, emphasizing the pressing need for stringent regulatory measures to mitigate the adverse effects of DFB contamination on non-target organisms and ecosystem functioning.

Keywords: Pesticides; Diflubenzuron; Acute toxicity; Sublethal toxicity; Gene expression; Mortality; Daphnids

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# Cyanobacterial biomass: An eco-friendly energy resource and potential algal growth promoter

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Microalgae biomass, particularly for cyanobacteria, is considered as promising resources rich in proteins, carbohydrates, and substances with high value. The study aimed to screen and analyze the novel native cyanobacterium possessing these potentials. In addition, to achieve economic feasibility, we attempted to establish an additional new approach for cheap cyanobacterial cultivation. To this end, algal-blooming cyanobacteria *Microcystis aeruginosa* biomass was obtained and subsequently lipid was extracted for biodiesel production. The residual *M. aeruginosa* biomass was then transformed to be a component of medium for the cultivation of microalgae including *Ochromonas danica* and *Euglena gracilis*. Taken together, our study provides potentials of the native cyanobacteria previously unidentified and a possible cost reduction strategy in microalgal cultivation with waste biomass including lipid extracted residue of *M. aeruginosa.*

Keyword: Microalgae, *Microcystis aeruginosa*, cost reduction

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