2022

ISCE-APACE

Joint Meeting of the 37th Annual Meeting of the International Society of Chemical Ecology and the 11th Asia-Pacific Association of Chemical Ecologists Conference in Kuala Lumpur, Malaysia

Abstract Book

Managing sustainability in challenging times

Asia-Pacific Association of Chemical Ecologists

About the Joint Meeting Logo

The logo was inspired by a morning serendipitous observation of a wild fruit fly orchid, *Bulbophyllum cheiri* in full bloom that was attracting males of the pest Oriental fruit fly, *Bactrocera dorsalis* in a house garden. The orchid is known to produce one of the most potent male insect attractant ever-methyl eugenol. As methyl eugenol attracts male *B. dorsalis* in nanogram quantities that are ingested, and biotransformed to male sex pheromones, it has also been shown to mediate the interaction of orchid and the fly as the pollinator. The logo depicts the male fly landing on the lip of the fruit fly orchid containing methyl eugenol. Volatiles containing methyl eugenol are shown in the picture as molecular structures in the background. A precarious balance between being a pest and pollinator supporting the fragile ecosystem.

References:

Nishida R, Tan KH, Wee SL, Hee AKW, Toong YC (2004) Phenylpropanoids in the fragrance of the fruit fly orchid, Bulbophyllum cheiri, and their relationship to the pollinator, Bactrocera papayae. Biochemical Systematics and Ecology 32: 245–252. https://doi.org/10.1016/S0305-1978(03)00179-0

Tan KH, Nishida R (2012) Methyl eugenol: Its occurrence, distribution, and role in nature, especially in relation to insect behavior and pollination. Journal of Insect Science 12. https://doi.org/10.1673/031.012.5601

Tan KH, Nishida R, Toong YC (2002) Floral synomone of a wild orchid, Bulbophyllum cheiri, lures Bactrocera fruit flies for pollination. Journal of Chemical Ecology. https://doi.org/10.1023/A:1016277500007

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SYNTECH

ISCE-APACE 3RD JOINT MEETING | 2022 | August 8–12 | Kuala Lumpur, Malaysia

WELCOME TO THE ISCE-APACE 3rd JOINT MEETING !

Ass. Prof. Dr. Alvin Hee

Dear friends and colleagues,

It is a great honour for me to represent the Organising Committee in welcoming you as participants of our 3rd Joint Meeting of the International Society of Chemical Ecology (ISCE) and the Asia-Pacific Association of Chemical Ecologists (APACE). This third meeting in Kuala Lumpur, Malaysia aspires to follow the successful footsteps of the first Joint Meeting that took place in Melbourne, Australia in 2013, and the second in Kyoto, Japan in 2017.

The idea to have a Joint Meeting in Southeast Asia for the first time emerged in 2014, while I was a young APACE councilor. Following the success of the 2nd APACE Meeting in 2001 in Penang, my dream was to host a Joint Meeting in Malaysia. Alex Il'ichev, the APACE President at that time, and the APACE executive committee were supportive of this idea. In 2016, Malaysia was chosen to host the 2021 ISCE-APACE Joint Meeting. Six years of preparation and the Covid-19 did not dampen the enthusiasm of seeing this meeting in Kuala Lumpur becoming a reality. Kuala Lumpur, as Malaysia's nation capital, was an obvious choice as it is the face of Malaysia, Truly Asia - A capital of food and culture.

We are happy to introduce 14 exciting symposia featuring research of world-class chemical ecologists plus 5 plenary lectures delivered by various prominent scientists from different regions. A number of ISCE and APACE awards will be conferred and presented by the awardees. Students are also highlighted for their travel awards.

In this Joint Meeting, we will have, for the first time, a symposium on Frontiers in Chemical Ecology that will be made public in efforts to democratize science and discover new talents. Young rising stars in chemical ecology are highlighted. What's more, for the first time since the establishment of the Wittko Francke's Daaks-Chemicals Fund, we will be having a public lecture to acknowledge the support from the public.

This Joint Meeting is the first hybrid conference in the history of chemical ecology, enabling participants to attend in-person or virtually. Thus, we thank you for choosing this Joint Meeting of ISCE-APACE as the platform to showcase your work and wish everyone a productive meeting and enjoyable stay in Kuala Lumpur. Do soak up the vibrant atmosphere of KL as Kuala Lumpur is affectionately known!

Selamat datang ke Kuala Lumpur! Welcome to Kuala Lumpur!

Alvin Kah-Wei Hee - Organising Chair

WELCOME TO THE ISCE-APACE 3rd JOINT MEETING !

Dear colleagues and friends,


Prof. Dr. Stefano Colazza

It is a great pleasure to welcome you to the 3rd joint meeting of the International Society of Chemical Ecology (ISCE) and the Asia-Pacific Association of Chemical Ecologists (APACE).

This congress has represented a challenge to our chemical ecology community due to the great difficulties created by the COVID-19 pandemic. The challenges have been particularly difficult for the meeting organizers, and we are very grateful for all their hard work in making this meeting happen, despite all the difficulties that they have faced. We should also try and turn apparent disadvantages into opportunities. Therefore, we are looking forward to holding this year's congress in a hybrid format, that combines "live" in-person sessions with a "virtual" online component. This allows us to expand our participants and welcome past and new attendees from all over the world. It also allows speakers who could not attend in person due to covid or other problems, to still be able to present their work to an international audience. The hybrid format also will allow more attendees to engage directly with speakers during the Q&A session after the presentations.

Efforts have been made to prepare a program suited for in-person and virtual audience that is sensitive to this new era, in which exploring deeply the "language of nature" will help to prevent pandemic emergencies such the one that we are currently enduring. The program committee and symposium organizers have developed a diverse and excellent program, and the networking opportunities will be indeed outstanding. The backdrop of the beautiful city of Kuala Lumpur and the warm welcome that we will receive will add to our enjoyment of the meeting and provide lasting memories and friendships beyond the science.

I wish to thank *in primis* Dr. Alvin KW Hee for the efforts to organize the congress in a safe and efficient way, according to all current standards, the Scientific Committee for their work in developing the program, and the Local Committee for making all the arrangements for us to have a memorable conference and visit to Malaysia.

I look forward to numerous chats and discussions with all of you over the next few days as we all enjoy the conference and the social events.

Stefano Colazza - ISCE President

WELCOME TO THE ISCE-APACE 3rd JOINT MEETING !

Dear colleagues and friends,

Prof. Dr. Guirong Wang

It is my pleasure and honor to join Professors Stefano Colazza and Alvin Hee to welcome you all to the 3rd joint meeting of the Asia-Pacific Association of Chemical Ecologists (APACE) and the International Society of Chemical Ecology (ISCE) in Kuala Lumpur, Malaysia. The first joint meeting took place almost ten years ago in Melbourne, Australia in 2013. Both of APACE and ISCE members met for the first time in the world-famous tourist city and international metropolis. And then for the 2rd joint meeting in 2017 in the city of Kyoto, Japan with the unforgettable historical relics and Japanese cuisine. A lot of friends and colleagues from the whole world can attend the first two joint meetings on site. We had exchanged scientific issues and made friends face to face, leaving infinite happiness.

With the great efforts of the organizers, even if the Covid-19 pandemic brings us great difficulties at the moment, we still have been able to bring together of chemical ecology researchers to present and discuss new progress and findings in Kuala Lumpur, an international metropolis with great influence on culture, education, sports, commerce and finance. We have innovated the form of mutual communication in the joint meeting either on site or virtual, that makes more friends and colleagues have the opportunity to attend the meeting.

I am truly delighted that we have a wonderful day here in Malaysia. To meet our old and new friends and colleagues, I bet you will enjoy both of the beautiful scientific program that was put together by our Chair Alvin Hee as well as magnificent culture and cuisine. The program committee and symposium organizers will provide outstanding platform for you to present and share your excellent ideas and new data as well as also exchange and discuss the questions that you are interested in.

Finally I would like to take this opportunity to offer my heartfelt thanks to Dr. Alvin Hee and his team for the efforts to organize the meeting and the most generous hospitality for hosting us during this meeting. I want to give special thanks to our sponsors for their kind support.
Please enjoy every minutes!

Guirong Wang - APACE President

STUDENT TRAVEL AWARDS

In support of our young generation of chemical ecologists, we wish to congratulate our student travel award winners as follows:

ISCE Student Travel Award Winners
Aswathi Sasidharan
Bashiru Adams
Cao Li
Gälle Ramiaranjatovo
Jeremy Chan
Munir Mostafiz
Natália de Souza Ribas
Peter Biwer
Quoc Hung Le
Rajarajan Ramakrishnan
Vignesh Venkateswaran

Bedoukian Applied Semiochemical Research Travel Award
Manish Kumar

ISCE-ALAEQ Travel Award
Amanda C. Túler

APACE Student Travel Award Winners
Shuting Chen
Yuki Chiba

PROGRAM OVERVIEW

Color code	Onsite	Onsite and Livestreamed	Virtual only

Time (UTC+8)	MONDAY, AUGUST 8th
7:50 - 15:00	Registration (EQ Kuala Lumpur Hotel)
15:00	APACE Executive Committee Meeting
16:00	ISCE Executive Committee Meeting
19:00	Welcome reception in the Evening (EQ Kuala Lumpur Hotel)

Time (UTC+8)	TUESDAY, AUGUST 9th (Onsite venue: Conservatory)	
7:50-8:00	**Opening** Welcoming address by the Organising Chair, **Alvin KW Hee**	
8:00-8:50	**ISCE Silver Medal Lecture** Moderator: Jerry (Junwei) Zhu	**Christer Löfstedt** Production of pheromones in plant and cell factories for sustainable pest control
8:50-9:40	**APACE Lifetime Achievement Lecture** Moderator: Alvin KW Hee	**Alexandre Il'ichev** Multi-species area-wide mating disruption with mass-trapping of mated females for sustainable and environmentally-friendly pest control in orchards.
9:40-10:30	**ISCE Silverstein-Simeone Lecture** Moderator: Jerry (Junwei) Zhu	**Le Kang** Secret of locust plagues: The simultaneous emission of PAN and 4VA by locusts maintains aggregation and large-scale swarm
10:30-10:50	**Coffee Break**	

Time (UTC+8)	TUESDAY, AUGUST 9th (Onsite venue: Conservatory)	
10:50-11:40	**ISCE Applied Chemical Ecology Lecture 1** Moderator: Andrés Gonzalez	**Fatma Kaplan** Nematode pheromone extracts act as boosters for entomopathogenic nematodes' efficacy to control agricultural pests in the soil
11:40-12:30	**ISCE Applied Chemical Ecology Lecture 2** Moderator: Stefano Colazza	**Aijun Zhang** Discovery of insect attractant and repellent/pesticide based on chemical stimuli from apple
12:30-13:30	**Lunch & Posters**	
13:30-14:20	**Wittko Francke's Daaks-Chemicals Memorial Lecture** Moderator: Jeremy McNeil	**Stefan Schulz** The impact of Wittko Francke on modern research in chemical ecology
14:20-15:10	**APACE Applied Chemical Ecology Lecture** Moderator: Aijun Zhang	**Jerry (Junwei) Zhu** From basic understanding of chemical ecology to advance practical applications with developed semiochemical-based technologies
15:10-15:30	**Coffee Break**	
15:30-16:20	**APACE Young Scientist Lecture** Moderator: Alex Il'ichev	**Po-An Lin** Plant-insect interactions in changing environments and the crucial roles of water and light
16:20-17:10	**ISCE Early Career Lecture 1** Moderator: Ted Turlings	**Maryse Vanderplanck** Nutritional resilience in bumblebees
17:10-18:00	**ISCE Early Career Lecture 2** Moderator: Nicole van Dam	**Shuqing Xu** Function and evolution of plant specialized metabolites - looking backward to move forward

Time (UTC+8)	WEDNESDAY, AUGUST 10th (Onsite venue: Conservatory)	
8:00-9:00	**Plenary 1** Moderator: Alvin KW Hee	**Walter S Leal** Skatole- and indole-sensing odorant receptors with reciprocal specificity mediated by a single amino acid residue
09:00 - 09:05	**SYMPOSIUM 1** **Frontiers in Chemical Ecology (1)** Moderator: Walter Leal	**Wendell Roelofs** Preface
09:05 - 09:30		**Greg Pask** Ant Chemosensory Sensilla Microstructures Support a Dual Function in Detecting Both Volatile and Contact-Mediated Cues
9:30-9:55		**Nicoletta Faraone** Host detection by ticks in the context of repellent exposure
9:55-10:20		**Erika Machtinger** The curious response of the blacklegged tick (Ixodes scapularis) to odors of sympatric rodent hosts
10:20-10:40	**Coffee Break**	
10:40-11:05	**SYMPOSIUM 1** **Frontiers in Chemical Ecology (1)** Moderator: Walter Leal	**Trevor Sorrells** A persistent behavior state in mosquitoes for pursuing humans
11:05-11:30		**Chloe Lahondere** Personal care products alter mosquito-host interactions
11:30-11:55		**Ali Afify** Species-specific differences in the mosquito olfactory response to repellents
11:55-12:20		**Joel A. Butterwick** Structural basis of olfactory receptor activation by an insect confusant
12:20-12:30		**May Berenbaum** Closing remarks

Time (UTC+8)	WEDNESDAY, AUGUST 10th (Onsite venue: Conservatory)	
12:30 - 13:30	**Lunch & Posters**	
13:30-14:15	Plenary 2 Moderator: Kye Chung Park	**Andrea Clavijo McCormick** Chemical ecology: supporting biosecurity in Aotearoa New Zealand
14:15-15:00	Plenary 3 Moderator: Baldwyn Torto	**John Pickett** New chemical routes to sustainable food production from chemical ecology
15:00 - 15:20	**Coffee Break**	
	SYMPOSIUM 2 (Onsite venue: Conservatory) **APPLICATION OF SEMIOCHEMICALS IN INSECT PEST MANAGEMENT (1)** Moderator: Aijun Zhang	**SYMPOSIUM 3** (Onsite venue: Cluster 2) **PLANT METABOLOMICS AND CHEMICAL DEFENSES (1)** Moderator: Po-An Lin
15:20-15:40	**Qing-He Zhang** RESCUE!® Semiochemical-based Fly Traps for Consumer Market	**Hampus Petrén** Measuring chemodiversity considering biochemical and structural properties of compounds with the R package chemodiv
15:40-16:00	**R Andrew Hayes** Is the symbiotic yeast Kodamaea ohmeri playing a role in the aggregative behaviour of the honeybee pest *Aethina tumida* (Coleoptera: Nitidulidae)?	**Mary V Clancy** Terpene chemotypes in wild cotton (*Gossypium hirsutum*) from the Yucatan peninsula
16:00-16:20	**Anna Jirošová** Infestation of Norway spruces by tree-killing bark beetles *Ips typographus* was prevented by anti-attractant baits consisting of newly added trans-4-thujanol	**Jamin Ali** Plant-plant signalling affects the performance of Myzus persicae aphids on potato

Time (UTC+8)	WEDNESDAY, AUGUST 10th	
16:20-16:40	**Aijun Zhang** Development of a Push & Pull Strategy for Spotted Wing Drosophila Control in Blueberry Orchard	**Carlos Bustos-Segura** Variation in airborne communication between plants among wild cotton genotypes
16:40-17:00	**Junyong Song** Effect of soybean volatiles on the behavior of the bean bug, Riptortus pedestris	**Baldwyn Torto** The chemical ecology of plant-root knot nematode interactions
17:00-17:20	**Dangsheng Liang** Identification of bed bug probing stimulants from human skin for bait development	**SYMPOSIUM 4** (Onsite venue: Cluster 2) **PHEROMONES AND CHEMOPERCEPTION (1)** Moderator: Björn Bohman
		Rajarajan Ramakrishnan Investigating pheromone biosynthesis de novo from Eurasian spruce bark beetle – *Ips typographus*.
17:20-17:40	**Arnaud Costa** Effectiveness of Wota-T traps baited with sex pheromone to control the Sweet Potato Weevil *Cylas formicarius* (Fabr.) in Vietnam	**Marcelo Gustavo Lorenzo** Characterization of the sensory gene repertoire of *Triatoma infestans* and the effect of blood-feeding on antennal expression profiles
17:40-18:00	**Bao-Jian Ding** Release of pheromone compounds from *Nicotiana benthamiana* leaf glandular trichome upon transient expression of biosynthetic genes	**Björn Bohman** Unsaturated hydrocarbons - structural elucidation and function as fungus gnat pheromones and orchid pollinator attractants

Time (UTC+8)	THURSDAY, AUGUST 11th					
8:00-8:45	**Plenary 4** (Onsite venue: Conservatory) Moderator: Renee Borges		**Kazushige Touhara** Chemical communication in animals: pheromone, receptor and behavior			
	SYMPOSIUM 5 (Cluster 2) **FUTURE GENERATIONS OF CHEMICAL ECOLOGISTS (1)** Moderator: Andrés Gonzalez	**SYMPOSIUM 6** (Conservatory) **CHEMICAL AND MOLECULAR ECOLOGY OF PLANT-HERBIVORE-ENEMY INTERACTIONS (1)** Moderator: Nicole van Dam		**SYMPOSIUM 7** **CHEMICAL COMMUNI-CATION AND BIOLOGICAL INVASION (1)** Moderator: Lilin Zhao	**SYMPOSIUM 4** **PHEROMONES AND CHEMO-PERCEPTION (2)** Moderator: Yang Liu	**SYMPOSIUM 8** **ANTHROPOGENIC CHANGES AND APPLIED SOLUTIONS (1)** Moderator: Hajime Ono
8:45-9:05	**Manish Kumar** An untapped plant defense: novel biopesticide and plant-mediated RNAi combination for the pest management	**Nicole M. van Dam** Same problem, different solutions. Isothiocyanate detoxification in two related root herbivore species	8:45-9:00	**Lilin Zhao** Chemical communications between pinewood nematode and its vector insect	**Hisashi Ômura** Male wing substances acting as sex pheromones in the yellow butterfly	**Hongmei Li-Byarlay** Pesticide residues in the hive products and their potential risks to honey bees

9:05-9:25	**Bashiru Adams** Identification of non-host plant volatiles for the management of the invasive tomato leafminer, *Tuta absoluta*	**Betty Benrey** Bottom-up and top-down induced plant responses in wild lima bean plants and their consequences for plant fitness and trophic interactions	9:00-9:15	**Jun-Bo Luan** Lysine provisioning by horizontally acquired genes facilitates the fitness of the invasive whitefly	**Tianzi Gu** Identification and homologous analysis of odorant receptor genes in three Notodontidae species	**Nicoletta Faraone** What ticks don't like: formulations and deployment of novel natural product-based repellents
9:25-9:45	**Gaëlle Ramiaranjatovo** Fruit flies olfactory systems specialize in discriminating their various hosts	**Marine Mamin** Constitutive and herbivore-induced volatiles emitted by wild cotton populations	9:15-9:30	**Dmitry Kurenshchikov** Gypsy moth (*Lymantria dispar*) on the Far East of Russia	**Li Xu** Two odorant receptors tuned to a host plant volatile regulate oviposition behavior of *Bactrocera dorsalis*	**Caitlin Rering** Drought stress modulates floral chemistry with consequences for pollinator attraction
9:45-10:05	**Aswathi Sasidharan** Host plant quality vs. enemy-reduced space: explaining oviposition preference & larval performance in *Plutella xylostella*	**Radhika Venkatesan** Understanding host-parasitoid interactions for application in integrated pest management	9:30-9:45	**Jacob D. Wickham** Rapid assessment of cerambycid beetle biodiversity in a tropical rainforest using a multicomponent pheromone lure	**Longlong Sun** Anatomy and neuronal element identification of gnathal ganglion in larval *Helicoverpa armigera*	**SYMPOSIUM 3** **PLANT METABOLOMICS AND CHEMICAL DEFENSES (2)** Moderator: Hajime Ono

10:05-10:25	**Li Cao** Chemical communication in a highly specialized plant-pollinator interaction	**Tarikul Islam** Can plant silicon defences play a part in biocontrol of herbivorous arthropods?	9:30-9:45			**Shunta Sakamoto** β-Tyrosine accumulates in developing rice leaf via jasmonic acid-induced long-distance transport
10:25-10:45	**Coffee break**					
	SYMPOSIUM 5 (Cluster 2) **FUTURE GENERATIONS OF CHEMICAL ECOLOGISTS**(1) Moderator: Andrés Gonzalez	**SYMPOSIUM 2** (Conservatory) **APPLICATION OF SEMIOCHEMICALS IN INSECT PEST MANAGEMENT (2)** Moderator: R Andrew Hayes	9:45-10:00	**Xiangbo Kong** Monoterpenoid signals and their transcriptional responses to feeding and juvenile hormone regulation in bark beetle *Ips hauseri*	**Xiaolan Liu** Correspondence between input and output information in the macroglomerular complex in male *Helicoverpa armigera* (Hübner)	**Chengcheng Yao** Stemborer-induced rice plant volatiles boost direct and indirect resistance in neighboring plants
10:45-11:05	**Jeremy K Chan** Nocturnal NOx pollution impacts hawkmoth visitation in the field by degrading *Oenothera pallida* scent	**Saravan Kumar Parepally** Volatile cues of beehives aids *Galleria melonella* locate its host	10:00-10:15	**Sufang Zhang** RNA interference of key olfactory and visual genes during adult stage of *Agrilus planipennis*	**Baiwei Ma** Morphological identification of the neurons control Aphid cornicle muscle	**Jin Zhang** Competing beetles attract egg laying in a hawkmoth

11:05-11:25	**Vignesh Venkateswaran** Olfactory perception in elevated ozone: the role of ozone reaction products	**Mo Cui** Developing a new pathway for chemical synthesis of the sex pheromone of the fall web worm to facilitate the mating disruption strategy	10:15-10:30	**Xun Zhang** Screening of Potential RNAi Targets and Improving Interference Effect in *Hyphantria cunea*	**Yaning Li** Sex differential expression analysis on antennae transcriptome of *Hyphantria cunea*	**Tugcan Alinc** Role of plant-growth promoting fungi in mediating response of an egg parasitoid to plants induced by stink bug egg deposition
11:25-11:45	**Amanda C. Túler** Pathogenic fungus enhances dispersal through alteration of insect olfactory behavior	**Le Van Vang** Mating disruption by synthetic sex pheromones reduces damage of the beet armyworm on shallot fields in the Mekong Delta of Vietnam	10:30-10:45	**Xiaofeng Chen** Identification of odorant binding proteins which bind with methyl eugenol in *Bactrocera dorsalis*	**Xiaxuan Zhang** A female specific odorant receptor tuning to egg-surface odorants mediates oviposition aversion in *Helicoverpa armigera*	**Dun Jiang** Cd exposure through *Hyphantria cunea* pupae reduces the fitness of the parasitic wasp *Chouioia cunea*: A potential risk to the biocontrol efficiency
11:45-12:05	**Yuki Chiba** Sexual dimorphism of cuticular wax mediates male mating behavior in the strawberry leaf beetle, *Galerucella grisescens*	**Duong Kieu Hanh** Synergistic effect of volatiles from newly wounded leaf sheathes of mangrove palm	10:45-11:00	**Hao Guo** Functional analysis of pheromone receptor repertoire in the fall armyworm, *Spodoptera frugiperda*	**Yongliang Fan** Molecular regulatory mechanisms of contact sex pheromone biosynthesis in the German cockroach, *Blattella germanica*	**Jiaxing Fang** Metabolic exploitation of phytochemical signals mediates Ips typographus dispersal and species interaction in spruce forests

<table>
<tr><td rowspan="2">12:05-12:25</td><td rowspan="2">Peter Biwer
Secondary metabolites of amphibian skin-associated bacteria</td><td rowspan="2">(Nypa fruticans) on the aggregate attraction of the lesser coconut weevil (Diocalandra frumenti) inhabiting Mekong Delta of Vietnam</td><td>11:00-11:15</td><td>Yishu Geng
Electroantennographic and field responses of male Hyphantria cunea to synthetic sex pheromone components</td><td>Q&A Discussion</td><td>Q&A Discussion</td></tr>
<tr><td>11:15-12:30</td><td>Q&A Discussion</td><td>Q&A Discussion</td><td>Q&A Discussion</td></tr>
<tr><td>12:30</td><td colspan="5">Lunch</td></tr>
<tr><td></td><td colspan="5">Kuala Lumpur City Excursion/Free time</td></tr>
<tr><td>19:00</td><td colspan="5">Gala Dinner in the Evening (EQ Kuala Lumpur Hotel)</td></tr>
</table>

Time (UTC +8)	FRIDAY, AUGUST 12th					
8:00-9:00	**Plenary 5** (Onsite venue: Conservatory) Moderator: Dong Cha	**Suk-Ling Wee** An update on dacine fruit fly-male lure interactions				
	SYMPOSIUM 9 **Venue:** Cluster 2 **FRUIT FLY ATTRACTANTS, REPELLENT, AND HOST INTERACTIONS (1)** Moderator: Dong Cha	**SYMPOSIUM 11** **Venue:** Conservatory **EVOLUTIONARY CHEMICAL ECOLOGY** (1) Moderator: Jan Buellesbach		**SYMPOSIUM 10** **CHEMICAL DEFENSE IN VERTEBRATES (2)** Moderator: Adriana M Jeckel		**SYMPOSIUM 1** **FRONTIERS IN CHEMICAL ECOLOGY (2)** Moderator: Walter S Leal
8:45-9:05	**Matthew Sidehurst** Development of Synthetic Food-Odor Lures for *Bactrocera* Species	**Jan Buellesbach** Unraveling evolutionary conserved signaling elements in chemical profiles of insects	8:45-9:00	**Grégory Genta-Jouve** Another evidence regarding the dietary source for skin alkaloids of poison frogs	9:00-9:05	**Walter S Leal** Introductory Remarks,

9:05-9:25	**Alvin Kah-Wei Hee** Assessing non-methyl eugenol-responding lines of Oriental fruit fly males on their lure response	**Weizhao Sun** Decoding cuticular hydrocarbon-mediated female sexual attractiveness in the parasitoid wasp *Nasonia vitripennis*	9:00-9:15	**Takato Inoue** New insights into dietary toxin metabolism: diversity in the ability of the natricine snake to convert toad-derived bufadienolides	9:05-9:25	**Rick Fandino** Applying reverse genetics to evaluate complex sensory mechanisms in Lepidoptera
9:25-9:45	**Kye Chung Park** Olfactory roles of antenna and maxillary palp in long-range attraction of *Bactrocera* fruit flies	**Quoc Hung Le** Beyond cuticular hydrocarbons: Development of SPME on-fiber derivatization methods to investigate volatile organic surface compounds in parasitoid wasps	9:15-9:30	**Lauren O'Connell** Diet and genetics influence population differences in poison frog alkaloid profiles	9:25-9:45	**Mengbo Guo** A neural pathway for coding a significant floral scent component in the cotton bollworm *Helicoverpa armigera*
9:45-10:05	**Soo Jean Park** Plant-based attractant, zingerone and its analogs reveal structural requirements for attraction of Jarvis's fruit fly.	**Renee Maria Borges** Changing scents: how pollinator-attractive odors in *F. racemosa* vary across distance in Asia	9:30 - 9:45	**Mabel Gonzalez** Non-lethal methods open new possibilities to perform a metabolite profiling of skin secretions from endangered and endemic amphibians	9:45-10:05	**Karen Menuz** A conserved anoctamin regulates olfactory neuron firing

10:05-10:25	**Pradeepa Hewa Ranaweera** Attraction of Oriental fruit fly to β-caryophyllene	**Darren Wong** Molecular mechanisms underlying flower colour and volatile variation in a deceptive orchid (*Glossodia major*)	9:45-10:00	**John Dumbacher** Autoresistance to toxins in poisonous Pitohui birds is not rooted in sodium channel mutations	10:05-10:25	**Chen-Zhu Wang** Mutagenesis of the odorant receptor co-receptor (orco) reveals severe olfactory defects in a crop pest moth, *Helicoverpa armigera*
			10:00-10:25	Q&A session		
10:25-10:45	Coffee Break					
	SYMPOSIUM 9 **Venue:** Cluster 2 **FRUIT FLY ATTRACTANTS, REPELLENT, AND HOST INTERACTIONS (1)** Moderator: Dong Cha	**SYMPOSIUM 11** **Venue:** Conservatory **EVOLUTIONARY CHEMICAL ECOLOGY** (1) Moderator: Jan Buellesbach		**SYMPOSIUM 2** **APPLICATION OF SEMIOCHEMICALS IN INSECT PEST MANAGEMENT (3)** Moderator: Jian Chen		**SYMPOSIUM 1** **FRONTIERS IN CHEMICAL ECOLOGY (2)** Moderator: Walter S Leal
10:45-11:05	**Greg Loeb** Discovery and deployment of repellent compounds for managing *Drosophila suzukii*	**Jibin Johny** Functional Evolution of Odorant Receptors in Termites	10:45-11:00	**Vibina Venugopal** Efficacy of pheromone kairomone blends to trap coconut red palm weevil in India	10:45-11:05	**Ani Agnihotri** Refolding of odorant-binding proteins (OBPs) and a new binding assay method

<table>
<tr><td>11:05-
11:25</td><td>Dong Cha
Identification of female attractants for improved monitoring of oviposition-impending Bactrocera dorsalis</td><td>Chaymae Fennine
Mapping out olfactomes to unravel tephritid olfactory ecology and evolution</td><td>11:00-
11:15</td><td>Jian Chen
Two saturated C12 oxo fatty acids from the male alates of red imported fire ants and their behavioral effects</td><td rowspan="3">11:05-
11:25</td><td rowspan="3">Amber Crowley-Gall
Olfactory variation among closely related cactophilic Drosophila species</td></tr>
<tr><td rowspan="3">11:25-
11:45</td><td>SYMPOSIUM 8
Venue: Cluster 2

ANTHROPOGENIC CHANGES AND APPLIED SOLUTIONS (2)

Moderator:
Radhika Venkatesan</td><td>SYMPOSIUM 12
Venue: Conservatory

CHEMICAL ECOLOGY IN BIOSECURITY AND CONSERVATION (1)

Moderator:
Kye-Chung Park</td><td rowspan="2">11:15-
11:30</td><td rowspan="2">Jerome Niogret
Long-lasting monitoring system for the cocoa pod borer, development & application</td></tr>
<tr><td>Magali Proffit
Acute ozone exposure impairs detection of floral odor, learning, and memory of honey bees</td><td>Yuko Ohata
Dual defense system induced by pest damage in on-tree apple fruit</td></tr>
<tr><td>11:45-
12:05</td><td>Wilson R Valbon
Activation of odorant receptor Or31 contributes</td><td>Chaymae Fennine
Application of chemical ecology to pre-release</td><td>11:30-
11:45</td><td>Nalini Thiyagarajan
First report on analysis of volatile compounds of Oecophylla smaragdina from India</td><td>11:25-
11:45</td><td>Dan Peach
Floral attraction of mosquito pollinators</td></tr>
</table>

	to pyrethrum repellency in *Aedes aegypti* mosquito	risk assessments for the intentional introduction of biological control agents	11:45-12:00	**Fu Liu** Identification and field bioassays of the sex pheromone of the winter moth *Erannis ankeraria* Staudinger (Lepidoptera: Geometridae)	11:45-12:05	**Zain Syed** What drives a tick to seek a host and/or a habitat?
12:05-12:25	**SYMPOSIUM 10** **Venue:** Cluster 2 **CHEMICAL DEFENSE IN VERTEBRATES (1)** Moderator: Radhika Venkatesan	**SYMPOSIUM 13** **Venue:** Conservatory **INSECT BEHAVIOR AND EVOLUTION (1)** Moderator: Kye-Chung Park				
	Adriana M. Jeckel Sequestration timeframe and systemic distribution of alkaloids in a Dendrobatid poison frog	**Jeremy McNeil** *Drosophila melanogaster* Stress Odorant is an interspecific alarm cue	12:00-12:45	Q&A Discussion	12:05-12:25	**Walter S Leal** Closing Remarks
			12:45-13:15	Lunch		
12:25-12:45	**SYMPOSIUM 3** **Venue:** Cluster 2 **PLANT METABOLOMICS AND CHEMICAL DEFENSES (1)**	**Samuel Boff** The impact of environmental stressors on mating behaviour of wild bees	13:15-13:30	**SYMPOSIUM 12** **CHEMICAL ECOLOGY IN BIOSECURITY AND**		

	Moderator: Radhika Venkatesan			**CONSERVATION (2)** Moderator: **Andrea Clavijo McCormick**		
	James Perkins Floral Notes from Underground: The Scent of the Underground Orchid (*Rhizanthella speciosa*)			**Naoko Yoshinaga** Oviposition inhibitory activity of calcium carbonate wettable powder against *Carposina sasakii*		
12:45-13:30	Lunch				Lunch	

13:30	**SYMPOSIUM 9** **FRUIT FLY ATTRACTANTS, REPELLENT, AND HOST INTERACTIONS (2)** Moderator: Anat Levi-Zada	13:30	**SYMPOSIUM 13** **INSECT BEHAVIOR AND EVOLUTION (1)** Moderator: Kye-Chung Park	13:30	**SYMPOSIUM 11** **CHEMICAL ECOLOGY IN BIOSECURITY AND CONSERVATION (2)** Moderator**:** Andrea Clavijo McCormick	13:30	**SYMPOSIUM 14** **ENVIRONMENTAL IMPACTS OF PLANT-INSECT INTERACTION** Moderator: Wen-Po Chuang

13:30-13:45	**Kamala Jayanthi PD** Stake a claim: microbes facilitate niche partitioning in tephritid fruit flies	13:30-13:50	**Nisansala Perera** Identifying livestock dung headspace volatiles that drive the preference of the dung beetle Bubas bison	13:30-13:45	**D. Paul Barret** Could metabolomic profiling of host plant biochemistry improve the effectiveness and safety of classical weed biocontrol	13:30-14:00	**Christelle Robert** Plant responses to the third trophic level: an underestimated interplay shaping multitrophic interactions
13:45-14:00	**Vivek Kempraj** Salicylate alters the volatile profile of mango thereby affecting the behavior and development of the Oriental fruit fly, *Bactrocera dorsalis*	13:50-14:05	**SYMPOSIUM 13** **INSECT BEHAVIOR AND EVOLUTION (2)** Moderator: Koji Noge	13:45-14:00	**Evans Effah** Impact of invasive plants on native plants and herbivores: the role of volatile organic compounds	14:00-14:15	**Wen-Po Chuang** Environmental impacts on the insect resistance in rice
14:00-14:15	**Farzana Yesmin** Insect-Plant interaction: A new approach of *Bactrocera* fruit flies on the Cape Jasmine flower (Gentianales: Rubiaceae)		**Etya Amsalem** Social behavior and colony trajectory in bumble bees are shaped by the brood	14:00-14:15	**Aditi Mishra** The innate floral template and its unlearning in hoverflies	14:15-14:30	**Jessica Kansman** Water stress effects on arthropod communities and plant-aphid interactions: Investigating the impact of water stress intensity

	SYMPOSIUM 7 **CHEMICAL COMMUNICATION AND BIOLOGICAL INVASION (2)** Moderator: Anat Levi-Zada	14:05-14:20	**Sarah K Spence** Multimodal signaling by queens regulates mating behavior in the bumble bee *Bombus impatiens*		**SYMPOSIUM 6** **CHEMICAL AND MOLECULAR ECOLOGY OF PLANT-HERBIVORE-ENEMY INTERACTIONS (3)** Moderator: Andrea Clavijo McCormick	14:30-14:45	**Boon Huat Cheah** Copper Elemental Defense of Rice against *Cnaphalocrocis medinalis*
14:15-14:30	**Ruixu Chen** Identification of an alarm pheromone in the destructive invasive species: the red-necked longhorn beetle, *Aromia bungii*	14:20-14:35	**Valeria Palma-Onetto** The defensive eversible organ in papilionids: source of volatiles, exposure of toxic hemolymph and aposematic warning	14:15-14:30	**Carla C. M. Arce** The polyvalent sequestration ability of an economically important beetle	14:45-15:00	**Po-An Lin** Low water availability enhances volatile-mediated direct defenses but disturbs indirect defenses against herbivores
14:30-14:45	**Shouyin Li** Tolerance to host specific terpenoid defences in an emerging insect pest, *Pagiophloeus tsushimanus*	14:35-14:50	**Hajime Ono** Chemical substances underlying different host utilization between host races in the leaf-mining moth *Acrocercops transecta*	14:30-14:45	**Ricardo A.R. Machado** Molecular and chemical regulators of foraging behavior of root herbivores		

<table>
<tr><td rowspan="2">14:45-
15:00</td><td>SYMPOSIUM 5

FUTURE GENERATIONS OF CHEMICAL ECOLOGISTS (2)

Moderator:
Andrés Gonzalez</td><td rowspan="3">14:50-
15:30</td><td rowspan="3">Q&A Discussion</td><td rowspan="3">14:45-
15:30</td><td rowspan="3">Q&A Discussion</td><td rowspan="3">15:00-
15:30</td><td rowspan="3">Q&A Discussion</td></tr>
<tr><td>Shuting Chen
The frass excreted by the striped stem borer Chilo suppressalis induces defense responses in rice.</td></tr>
<tr><td>15:00-
15:30</td><td>Q&A Discussion</td></tr>
<tr><td>15:30-
16:00</td><td colspan="7">Coffee Break</td></tr>
<tr><td>16:00</td><td colspan="7">Business Meeting (Venue: Conservatory)
& Closing</td></tr>
</table>

SOCIAL PROGRAM

Monday, August 8, 2022

Welcome Reception

Venue: Nipah Restaurant, EQ Kuala Lumpur

7:00 pm

Thursday, August 11, 2022

Kuala Lumpur Excursions *(at own cost)*

2:00-5:00 pm

Gala Dinner *(tickets purchased during registration)*

7:00 pm

Venue: The Conservatory, EQ Kuala Lumpur

Featuring special string quartet led by one of the most acclaimed violinists in the region, Joanne Yeoh. Joanne is multi-talented being a violinist with 3 own albums, a PhD academician, and an examiner. She has performed with superstars and appeared with famous personalities. She's currently an Associate Professor with Universiti Putra Malaysia.

https://new.joanneyeoh.com.my

SYMPOSIUM LIST AND LINKS

AWARD LECTURES

Tuesday, August 9th
8 am - 6 pm

ISCE Silver Medal Lecture - *Moderator*: Jerry (Junwei) Zhu

Production of pheromones in plant and cell factories for sustainable pest control
Christer Löfstedt

APACE Lifetime Achievement - *Moderator*: Alvin KW Hee

Multi-species area-wide mating disruption with mass-trapping of mated females for sustainable and environmentally-friendly pest control in orchards
Alexandre IL'ICHEV

ISCE Silverstein-Simone Lecture - *Moderator*: Jerry (Junwei) Zhu

Secret of locust plagues: The simultaneous emission of PAN and 4VA by locusts maintains aggregation and large-scale swarm
Le Kang

ISCE Applied Chemical Ecology 1 - *Moderator*: Andrés Gonzalez

Nematode pheromone extracts act as boosters for entomopathogenic nematodes' efficacy to control agricultural pests in the soil
Fatma Kaplan, David Shapiro-Ilan, Edwin Lewis

ISCE Applied Chemical Ecology 2 - *Moderator*: Stefano Colazza

Discovery of insect attractant and repellent/pesticide based on chemical stimuli from apple
Aijun Zhang

Wittko Francke Daaks-Chemicals Memorial Lecture - *Moderator*: Jeremy McNeil

The impact of Wittko Francke on modern research in Chemical Ecology
Stefan Schulz

APACE Applied Chemical Ecology Lecture - *Moderator*: Aijun Zhang

From basic understanding of chemical ecology to advance practical applications with developed semiochemical-based technologies
Junwei Jerry Zhu

APACE Young Scientist Lecture - *Moderator*: Alex Il'ichev

Plant-insect interactions in changing environments and the crucial roles of water and light
Po-An Lin

ISCE Early Career Lecture 1 - *Moderator*: Ted Turlings

Nutritional resilience in bumblebees
Maryse Vanderplanck

ISCE Early Career Lecture 2 - *Moderator*: Nicole van Dam

Function and evolution of plant specialized metabolites: looking backward to move forward
Shuqing Xu

ISCE Silver Medal Lecture

Production of pheromones in plant and cell factories for sustainable pest control

Christer Löfstedt

Lund University, Department of Biology, Lund, Sweden

Biological production of insect pheromones has evolved rapidly as an alternative to conventional synthetic pheromone production. Recent success to produce moth pheromones in cell and plant factories build on research on pheromone biosynthesis that took off in the early 1980s. The explosion of molecular tools and "omics" from the end of the 1990s facilitated characterization of genes involved in biosynthesis and metabolic engineering of plants and microbes opened up for large-scale biological production of pheromones. This strategy is environmentally friendly and has become a cost-effective way of producing pheromones with high purity and a minimum of waste. So far, the production of C_{16} and C_{14} monounsaturated fatty acyl precursors and pheromone components have been targeted, which in many ways represent "low-hanging fruit"; a sufficient pool of saturated C_{16} or C_{14} precursors already exist in the plant and yeast platforms or can be easily created by metabolic engineering. Production of pheromones with shorter chain length and with multiple double bonds is an outstanding challenge but the prospects of success are bright enough to attract investment. Fermentation of moth pheromone compounds in *Yarrowia lipolytica* developed as part of the research project OLEFINE and resulted in the biotech start-up company BioPhero. The research project Oil Crops for the Future gave rise to SemioPlant that in partnership with ISCA aims for production of pheromones from precursors produced in the oilseed crop Camelina. Biological production of insect pheromones works, the rest is a matter of upscaling, investments, and market penetration.

Keywords: Moth pheromones, Biological production, pest control

Löfstedt C, Xia YH (2021) pp. 89–121 in GJ Blomquist, RG Vogt (eds) Insect Pheromone Biochemistry and Molecular Biology. Second Edition. Academic Press
Wang HL, Ding BJ, Dai JQ, Nazarenus TJ, Borges R, Mafra-Neto A, Cahoon EB, Hofvander P, Stymne S, Löfstedt C (2022) Nat Sustain (*accepted*)

APACE Lifetime Achievement

Multi-species area-wide mating disruption with mass-trapping of mated females for sustainable and environmentally-friendly pest control in orchards

Alexandre IL'ICHEV

(Agriculture Victoria Research – retired), Tatura, Australia

Mating disruption (MD) is an important tool of sustainable, effective and environmentally-friendly Integrated Pest Management (IPM) in Horticulture. The use of hand-applied MD dispensers in only individual orchard blocks and known host-plants for control of oriental fruit moth (OFM) or codling moth (CM) have been standard practice in Australian orchards. An increased OFM damage on the borders of MD treated and untreated blocks stimulated area-wide application of MD program, with more than 1,100 ha of 40 contiguous orchards with MD dispensers applied to all fruit trees in the Cobram region of Victoria, and during three consecutive years substantially improved protection against OFM damage. Later, local growers continued area-wide MD program as self-sufficient community approach and re-established MD across the whole Cobram region. The ability of OFM and CM to migrate between orchards, quickly invade new host-plants and together damage the same variety stimulated grower's need to cater for both pests in their IPM strategies. The area-wide MD program to control both OFM and CM has been successfully established in Greater Shepparton and Invergordon regions of northern Victoria. During our collaboration with Michigan State University (late Prof. Larry Gut) we also evaluated new multispecies MD dispensers in counter seasonal replicated trials in Victoria (Australia) and Michigan (USA). Also, we demonstrated that attractiveness of pear-derived kairomone, on pears under MD may be synergised by host-plant volatiles, making the lures even more attractive for CM males and, most importantly, mated females. The real goal of MD is to reduce the female's ability to reproduce, therefore mass-trapping of CM mated females was an effective way to compliment MD by reducing the pest populations to levels controllable by MD. Our results demonstrated that mass-trapping significantly reduced CM populations and provided proof of concept for mass-trapping of mated CM females in MD treated pears.

Keywords: Mating disruption, Mated females, Mass-trapping

ISCE Silverstein-Simone Lecture

Secret of locust plagues: The simultaneous emission of PAN and 4VA by locusts maintains aggregation and large-scale swarm

Le Kang

Institute of Zoology, Chinese Academy of Sciences, Beijing, China

Locust plagues continue to pose a major threat to agriculture, economy and environment. Since 1970s, scientists recognized that aggregation pheromones may be the most critical factor for locust swarming. However, none of these compounds can meet all the criteria for aggregation pheromone. Recently, we identified two chemicals released by gregarious locusts, phenylacetonitrile (PAN) and 4-vinylanisole (4VA). We reveal that PAN acts as an olfactory aposematic signal and precursor of hypertoxic hydrogen cyanide (HCN) to protect gregarious locusts from predation. PAN biosynthesis from phenylalanine is catalyzed by a cytochrome P450 enzyme in gregarious locusts. When locusts are attacked by birds, PAN is converted to HCN, which causes food poisoning in birds. 4-vinylanisole is identified for the first time as an aggregation pheromone of locusts. Both gregarious and solitary locusts are strongly attracted to 4VA, regardless of age and sex. Although, it is emitted specifically by gregarious locusts, 4VA production can be triggered by aggregation of four to five solitary locusts. It elicits responses specifically from basiconic sensilla on locust antennae. We also identified OR35 as a specific olfactory receptor of 4VA. Knockout of OR35 using CRISPR-Cas9 markedly reduced the electrophysiological responses of the antennae and impaired 4VA behavioural attractiveness. Finally, field trapping experiments verified the attractiveness of 4VA to experimental and wild populations. These results indicate that simultaneous emission of PAN and 4VA by locusts maintains individual aggregation and large-scale swarm, resulting in the outbreaks of locust plagues. These findings provide insights for the development of novel control strategies for locusts.

Keywords: Phenylacetonitrile, 4-vinylanisole, Aggregation Pheromone

[1] Wei JN, Shao WB, Cao MM, Ge J, Yang PC, Chen L, Wang XH, Kang L (2018) Science Advances, 4: DOI: 10.1126/sciadv.aav5495.
[2] Guo XJ, Yu QQ, Chen DF, Wei JN, Yang PC, Yu J, Wang XH, Kang L (2020) Nature, 585(7822):584-588.

ISCE Applied Chemical Ecology 1

Nematode pheromone extracts act as boosters for entomopathogenic nematodes' efficacy to control agricultural pests in the soil

Fatma Kaplan[1], David Shapiro-Ilan[2], Edwin Lewis[3]

[1]*Pheronym, Inc., Davis, USA.* [2]*USDA-ARS, Byron, USA.* [3]*University of Idaho, Moscow, USA*

Nematode pheromones, ascarosides, stimulate dispersal, which is a key nematode behavior to find a new food source and to be a successful biocontrol agent. Currently, commercially available nematodes may not disperse sufficiently when they are applied to a field. Since the insect target is mobile, nematodes need to be actively moving and seeking an insect host. We hypothesized that pheromone extracts increase the dispersal of EPN infective juveniles (IJs) leading to increased insect encounter and efficacy. First, we determined whether pheromone extracts improved IJ movement/dispersal in 35 cm long soil columns baited with *Tenebrio molitor* larvae. We found that pheromone extracts induced higher numbers of *Steinernema carpocapsae* and *Steinernema feltiae* IJs to move towards *T. molitor* larvae in the bottom of the column compared to IJs treated with infected cadaver macerate and water, positive and negative controls, respectively. Furthermore, the number of *S. carpocapsae* IJs that invaded *T. molitor* larvae was higher for the pheromone extract treatment than for the controls. *S. feltiae* IJs that were pretreated with pheromone extracts and macerate (positive control) infected *T. molitor* at the same rate but the invasion was superior to IJs that were treated with water. Consistent with the soil column tests, both *S. carpocapsae* and *S. feltiae* IJs treated with pheromone extracts performed better in killing larvae of two economically important insect larvae, pecan weevil and black soldier fly in greenhouse tests compared to IJs treated with water. We demonstrated pheromone-mediated behavioral manipulation of a biological control agent to enhance pest control potential. Next, we tested the efficacy of IJs exposed to pheromones prior to field application in the peach and pecan orchards in the spring as curative treatment and in the fall as a preventative treatment. The finding from the field trials will be presented.

Keywords: Entomopathogens, Pheromones, Ascarosides, Efficacy

ISCE Applied Chemical Ecology 2

Discovery of insect attractant and repellent/pesticide based on chemical stimuli from apple

Aijun Zhang

USDA ARS, Beltsville, Maryland, USA

Plants use chemical stimuli in diverse ways, attracting pollinators for pollination or attracting herbivores that damage the plants. In addition, plants can also defend themselves against herbivores with their toxic and deterrent scents. Apple is one of the most consumed fruits worldwide. More than 300 volatile aroma compounds have been recognized from apple. Previously, we identified 5 esters from apple as an attractive blend for apple maggot, *Rhagoletis pomonella*. It was significantly more attractive than the previous identified blend and has been used with commercial apple maggot monitoring spheres. The spotted wing drosophila (SWD), *Drosophila suzukii*, is a new invasive fruit-infesting fly to the United States. Since the rotted apple is incredibly attractive to SWD, the apple cider vinegar has been used by farmers as the population monitoring tool. However, it is not selective and not very efficient at attracting flies prior to fruit injury and farmers cannot properly time protective measures. We compared headspace volatiles collected from fresh and fermented apple juices and identified 5-component blend that could selectively attract *D. suzukii* in the early stages of infestation, prior to fruit injury. Therefore, it has been used to develop an efficient, safe, and environmentally friendly SWD control method to manage SWD population levels under the economic threshold and help growers meet the market demands. Beside attractants, we also found a compound, methyl benzoate (MB), from fermented apple juice that exhibited significant repellent/toxic activities against SWD and other insect pests, including the brown marmorated stinkbug, diamondback moth, gypsy moth, some stored product insect pests, red imported fire ant, bed bug, and mosquito. Discovery of MB and its analogs provided the opportunities for development of more efficient, safe, and environmentally friendly green repellents/pesticides for farmers, reducing the possible adverse effects of synthetic pesticides on human health and the environment.

Keywords: -

Wittko Francke Daaks-Chemicals Memorial Lecture

The impact of Wittko Francke on modern research in Chemical Ecology

Stefan Schulz

TU Braunschweig, Braunschweig, Germany

Wittko Francke was one of the driving forces in Chemical Ecology during his whole career. Although his research was deeply rooted in the identification and synthesis of insect semiochemicals of insects, he had profound entomological knowledge. This enabled him to be a key interdisciplinary scientist combining both worlds, being inspirational to a broad network Chemical Ecologists. I will concentrate in the lecture on his impact on modern Chemical Ecology. In the lecture specific scientific topics highly influenced by Wittko will be discussed. The power of mass spectrometry is of fundamental importance in Chemical Ecology still today. I will show how his approach in deciphering new compound classes, e. g. mass spectrum interpretation, microreactions, and synthesis can be further developed into an integrated system including other data such as IR data and computer calculations into a unifying system for structure elucidation, if NMR spectroscopy is not an option. It can be envisioned that in the future the potential for full or at least partly automatized structure elucidation is possible. Also emphasized will be that correct identification of nature's compounds, one of Wittko's hallmarks, is essential to arrive at biologically meaningful results. Given the easy access to results with modern widespread GC/MS usage it is still surprising how many often obviously wrong identifications are published. A thorough understanding of multi-component compound bouquets using biosynthetic knowledge as advocated by Wittko is certainly needed. This approach will be illustrated in the lecture using *Heliconius* butterfly pheromone communication we investigate. I will also discuss the identification of structurally unique compounds described by Wittko and their influence today. These include spiroacetals, the striped cucumber beetle pheromone, and the unique microbial volatile sodorifen.

Keywords: GC/MS, Database, Mass spectrometry, Open data, Community effort

APACE Applied Chemical Ecology Lecture

From basic understanding of chemical ecology to advance practical applications with developed semiochemical-based technologies

Junwei Jerry Zhu

USDA-ARS, AMRU, Lincoln, USA

Chemical Ecology studies the roles of chemicals that mediate interactions between organisms and organisms and to their environments. It is highly interdisciplinary, which involves behavior, ecology, physiology, biochemistry, microbiology, neurobiology, genomics, and chemistry. Among these, olfaction is acting as one of the most important factors that most insects perceive and interact with the biotic world. Consequently, the outcome from studying insect chemical ecology has provided tremendous knowledge base being used to develop further into various practical control technologies for managing pests in agriculture, as well as in other fields such as environmental and human protection. In the present talk, I will briefly summarize some of research work that have been accomplished in my USDA lab and other labs through my chemical ecology careers. This includes 1). Development of the attractant lures for enhanced biological; 2). The use of identified fruit odorants for nuisance fly trapping; 3). The discovery of plant defensive compounds acting as both aphid deterrents and their natural enemies' attractants; 4). Biosensors using insect sensory organs for detecting land mines and other hazardous sources; 5). Natural product based repellent discovery and development for animal and human protection and 6). The first attractant impregnated adhesive material and tape trap development. All these case studies have shown great promises with already-developed and proto-type pest management products and technologies with great potentials for future commercial product development, which have collaborated with professionals from industries and scientists of universities and other institutions that will benefit agricultural crop producers and pest control product manufactures and professionals.

Keywords: Applied chemical ecology, Practical control, Pest management, Semiochemicals, Green technologies

APACE Young Scientist Lecture

Plant-insect interactions in changing environments and the crucial roles of water and light

Po-An Lin

National Taiwan University, Taipei, Taiwan

Water and light are two essential factors for plant growth and development. While increasing evidence has suggested their importance in plant biotic interactions, how these factors influence plant-insect interactions remained unclear. The first part of my work investigates the impact of water availability on plant-insect interactions. We discovered that drought stress leads to changes in the defense strategies of plants, including enhanced resistance and decreased tolerance against herbivory. Drought stress also compromised plant indirect defenses indicated by lower numbers of natural enemies and the emission of repellent volatiles under drought. Furthermore, we documented the ability of insect herbivores to induce drought-like responses (e.g., stomatal closure) using their salivary protein. Stomatal closures were further linked to inhibition of important defense-related HIPVs, similar to HIPV changes caused by water deficit. The findings suggest that herbivores might utilize drought-like responses of plants for their own benefits and show the links between stomatal behavior and HIPV emission. The second part of my work investigates the role of light in the host range evolution of lepidopteran herbivores. We found that higher plant volatile emission is closely associated with host plant specialization across the lineage of Lepidoptera. In contrast, lower levels of volatile emission at night are linked to diet generalization. Based on these results, we proposed the Salient Aroma Hypothesis that predicts the evolutionary pattern of host plant specialization according to diel changes in plant volatile signals. Our work contributes to a better understanding of the impact of water and light on the ecology and evolution of plant-insect interactions.

Keywords: -

ISCE Early Career Lecture 1

Nutritional resilience in bumblebees

Maryse Vanderplanck

CEFE, Univ Montpellier, CNRS, EPHE, IRD, Montpellier, France. Laboratory of Zoology, Institute for Biosciences, Univ Mons, Mons, Belgium

It is common knowledge that bees predominantly collect pollen to satisfy their nutritional and physiological requirements, pollen being essential for reproduction, individual health and larval development. Pollen is a complex chemical mixture that contains both central metabolites, which are vital for plant survival, and specialized metabolites, which play a key role in the interaction of the plant with its environment (insect attraction or deterrence). Pollen composition is highly variable among plant species so bees face a high degree of variation in pollen quality, with some pollen types being unsuitable for some bee species. This implies that even generalist bees cannot forage randomly on all available resources but must display selective foraging. Wild bees have therefore evolved a sophisticated ability to discriminate among plants and forage selectively on resources that allow them to achieve their nutritional optimum. Evidence is that such host discrimination may be guided by the chemical composition of pollen. While consumption of inadequate resources can lead to reduced survival, decreased immunity, and increased susceptibility to pathogens; consumption of adequate resources could provide bees with resilience facing environmental challenges. Nutritional resilience could be one of the key mechanisms determining the capacity of organisms to cope with living in a changing world. As such, there is a clear need to understand how pollen metabolites could contribute to the resilience of bees to global change. This proposed journey through some of my recent studies aims to demonstrate whether nutritional resilience occurs in bumblebees to face environmental stressors such as changes in floral resources, heat waves and parasites using field observations and laboratory experiments. A better understanding of the mechanisms underlining such bee nutritional resilience could greatly improve pollinator conservation programs by proposing nature-based solutions, such as promoting a medicinal flora.

Keywords: Environmental stressors, Decline, Pollinators, Pollen chemicals, Nutritional resilience

ISCE Early Career Lecture 2

Function and evolution of plant specialized metabolites - looking backward to move forward

Shuqing Xu

Department of Evolutionary Plant Sciences, Institute of Organismic and Molecular Evolution, Johannes Gutenberg University Mainz, Mainz, Germany

Plants produce highly diverse specialized metabolites (SM) that mediate fitness-relevant ecological interactions, such as defense against herbivores. While the evolution of plant SM has been considered mainly to be driven by herbivores, our recent studies in illustrating the biosynthesis and function of plant SM suggested that genome evolution, functional pleiotropy and autotoxicity can also shape the evolutionary trajectory of plant SM (1-4). In this talk, I will discuss these findings in a historical context. I will also share some of my thoughts on future directions in studying plant SM evolution.

Keywords: Plant specialized metabolites; Herbivore; Pleiotropy; Genome evolution; Autotoxicity

[1] Li, J. C. et al. Controlled hydroxylations of diterpenoids allow for plant chemical defense without autotoxicity. Science 371, 255-260 (2021).
[2] Xu, S. et al. Wild tobacco genomes reveal the evolution of nicotine biosynthesis. Proc Natl Acad Sci USA 114, 6133-6138 (2017).
[3] Zhou, W. et al. Tissue-specific emission of (*E*)-alpha-bergamotene helps resolve the dilemma when pollinators are also herbivores. Curr Biol 27, 1336-1341 (2017).
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PLENARY LECTURES

Plenary Lecture 1 - Wednesday, August 10th - 8 am

Moderator: Alvin KW Hee

Skatole- and indole-sensing odorant receptors with reciprocal specificity mediated by a single amino acid residue

Flavia Franco, Pingxi Xu, Brandon Harris, Vladimir Yarov-Yarovoy, Walter Leal

Plenary Lecture 2 - Wednesday, August 10th - 1.30 pm

Moderator: Kye Chung Park

Chemical Ecology: supporting biosecurity in Aotearoa New Zealand

Andrea Clavijo-McCormick

Plenary Lecture 3- Wednesday, August 10th - 2.15 pm

Moderator: Baldwyn Torto

New chemical routes to sustainable food production from Chemical Ecology

John Pickett

Plenary Lecture 4 - Thursday, August 11th - 8am

Moderator: Renee Borges

Chemical communication in animals: pheromone, receptor and behavior

Kazushige Touhara

Plenary Lecture 5 - Friday, August 12th - 8 am

Moderator: Dong Cha

An update on Dacine Fruit Fly-Male Lure interactions

Suk-Ling Wee

Plenary Lecture 1

Skatole- and indole-sensing odorant receptors with reciprocal specificity mediated by a single amino acid residue

Flavia Franco, Pingxi Xu, Brandon Harris, Vladimir Yarov-Yarovoy, Walter Leal

University of California-Davis, DAVIS, USA

Gravid female mosquitoes that already had a blood meal might carry viruses and pathogens that cause human disease. Traps baited with oviposition attractants are crucial for surveillance, i.e., identifying emerging and endemic viruses circulating in an area. The southern house mosquito, *Culex quinquefasciatus*, utilizes two ORs, CquiOR10 and CquiOR2, narrowly tuned and well conserved among mosquito species to detect the oviposition attractants indole and skatole. These receptors were formerly named CquiOR21 and CquiOR10 and showed high specificity to skatole and indole, respectively. Surprisingly, specificity is determined by only one out of more than 370 amino acid residues. Specifically, Ala-73 in CquiOR10 and Leu-74 in CquiOR2 are determinants of the reciprocal specific responses to skatole and indole. Our data show that CquiOR10A73L behaved like CquiOR2, whereas CquiOR2L74A recapitulated CquiOR10 specificity. In this presentation, we will discuss our swapping domain approach coupled with molecular modeling (RoseTTA and AlphaFold) and docking to unravel the structural features of these oviposition attractant-sensing receptors.

Research supported by the National Institute of Allergy and Infectious Diseases (NIAID) of the NIH under award number R01AI095514.

Dedicated to the late Professor James H. Tumlinson III

Keywords: *Culex quinquefasciatus*, CquiOR2, CquiOR10, Rosetta, RosettaLigand

Plenary Lecture 2

Chemical ecology: supporting biosecurity in Aotearoa New Zealand

Andrea Clavijo-McCormick

Ministry for Primary Industries, Wellington, New Zealand. Massey University, Palmerston North, New Zealand

Increased human migration, global trade, and climate change are contributing to the spread and establishment of invasive species worldwide. The unique New Zealand/Aotearoa biota (with an estimated 80,000 endemic species) is particularly vulnerable to biological invasions with many invasive taxa outnumbering local species (e.g., mammals and vascular plants). The New Zealand Biosecurity system is faced with the challenging task of stopping pests at the border and managing those already in the country, in a manner that is sustainable, socially acceptable, and preserves New Zealand's cultural and biological heritage. To achieve this aim, the system engages with a wide variety of stakeholders including Māori (Aotearoa indigenous people). Māori are full partners with the Crown in the biosecurity system and take an active role in protecting Aotearoa from unwanted organisms in alignment with their worldview. The Māori worldview (Te Ao Māori) understands the environment in a holistic sense where everything is interconnected (whakapapa) and has an internal energy or life force (māuri), which permeates living and non-living things and maintains a healthy environment. Although, Western understanding of what an "invasive species" is may differ from that of Māori, it is agreed that widespread invasive species can disrupt whakapapa and damage māuri. Furthermore, Māori are also committed to the protection of treasured species (Taonga tuku iho), the sustainable management of resources for future generations (Te Ao Tūroa) and to exercise guardianship over natural resources (kaitakitanga). Chemical Ecology is increasingly becoming a tool to support the biosecurity system in New Zealand, enabling the early detection and management of invasive species, and a better understanding of their ecological impacts. In this talk, we will explore the contribution of Chemical Ecology to the New Zealand Biosecurity system, discuss its alignment with Te Ao Māori, and explore future opportunities for transdisciplinary research.

Keywords: Biosecurity, Indigenous Knowledge, Pest Management, Biological Control, Metabolomics

Plenary Lecture 3

New chemical routes to sustainable food production from chemical ecology

John Pickett

Cardiff Univertsity, Cardiff, United Kingdom

Although we have extensive awareness of the un-sustainability of current food production, there is a paucity of effective technologies to achieve sustainability in terms of eliminating the currently mostly seasonal inputs involved in food production. Way back in 2009, a noble, but unsuccessful, attempt was made to direct new resources towards essential sustainable solutions. There are opportunities which can be developed now but these require the discipline of chemical ecology for realistic deployment. Exemplification will be around the crucial goal for developing sustainable agricultural production: the replacement of annual crops and cropping systems with perennial cultivars. In applied chemical ecology, the advantages of perennial crop plants have long been known and not least, the value of longer-term investment in plant roots and the wider rhizosphere ecosystems over those for annual planting systems. To exploit perennialisation, entry into the crop must be eliminated, except for harvest, thereby requiring crop management by sentinel plants signaling to the main crop stand, for example for the induction of defence embedded genetically in the initial perennial planting material by GM and GE. This is a challenge that chemical ecology can meet. Plant priming and induction signal related promoter sequences are available and related inducible indirect defense is already exploitable by GM in global crop plants. We must now expand our research plans to provide the chemical ecologically based tools necessary to take forward the already current work towards new perennial crop cultivars.

Keywords: Chemical Ecology, Sustainable agriculture, Perennial crops, Defense signaling, Sentinel plants

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[2] Pickett JA (2016), Nature Plants 2: 16078
[3] Pickett JA & Khan ZR (2016), New Phytologist (Tansley Review), 212: 856
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Plenary Lecture 4

Chemical communication in animals: pheromone, receptor and behavior

Kazushige Touhara

The University of Tokyo, Tokyo, Japan

The sense of olfaction plays a major role in regulating various behaviors including feeding behavior and socio-sexual interaction in many animals. Identification of specific chemical cues including pheromones and their receptors has provided a useful model to study how sensory inputs are converted into certain behavioral outputs. In this talk, I will show our work regarding chemical communication in insect and mouse; 1) identification of pheromones, receptors, and neural circuits that elicit sexual and social behavior in mice and 2) elucidation of mechanisms underlying odorant, pheromone, and taste sensing via olfactory and gustatory receptors that regulate sexual and feeding behavior in Bombyx mori. I will also talk about some of our recent work addressing whether chemical communication is utilized in primates including humans.

Keywords: -

Plenary Lecture 5

An update on Dacine Fruit Fly-Male Lure interactions

Suk-Ling Wee

Department of Biological Sciences and Biotechnology, Faculty of Science and Technology, Universiti Kebangsaan Malaysia, Selangor, Malaysia

Certain male Bactrocera and *Zeugodacus tephritid* fruit flies are known to be actively foraging for plant-derived semiochemical lures (hereafter male lures) that consist primarily of phenylbutanoids such as raspberry ketone, RK; and phenylpropanoids such as methyl eugenol, ME. Male flies' responses to these male lures are so specific that they were regarded as either ME-responding or RK-responding species. This has become a feature of diagnostic tool in fruit fly species identification. Hitherto, this intricate relationship between male lures and Dacine fruit flies discovered over a century ago continues to excite researchers with new findings. In a new paradigm shift, male flies' attraction to male lures is no longer confined to just ME-responding or RK-responding species. There are phenylbutanoids or phenylpropanoids shown to attract both ME- and RK-responding species, while some previously weakly or non-responsive species to ME/RK show specific and better responses to phenylbutanoids/phenylpropanoids other than ME/RK. Some non-phenylbutanoids/phenylpropanoids are demonstrating good to high attractancy to some dacine fruit fly species. Nevertheless, we found that the fly-lure interactions are still unique and species-specific phenomena. While males' response to lures increases concomitant with sexual maturity attainment, these interactions vary between fruit fly species and lures. Lure feeding by males also invariably enhances male mating performance by increasing male sexual signalling and/or increased attractiveness of sexual signals. Recent detailed studies involving other *Bactrocera* and *Zeugodacus* species have revealed that age at lure administration, lure amount, and lure type affect the outcome of physiological consequences of the lure-fed male fruit flies. The ecological significance of phytochemical lures in the plant-fruit fly relationship leading to new insights that implicate area-wide control programmes against those pestiferous fruit flies will be discussed.

Keywords: -

Symposium 1: Frontiers in Chemical Ecology (1)

Wednesday, August 10th
9 am - 12:30 pm

Session moderated by Walter S Leal
Session sponsored by BedoukianBio

Microstructures of ant chemosensory sensilla support a dual function in detecting both volatile and contact-mediated cues
Hannah Gellert, Daphné Halley, Zackary Sieb, Jody Smith, Gregory Pask

Host detection by ticks in the context of repellent exposure
Nicoletta Faraone, N. Kirk Hillier

The curious response of the blacklegged tick (*Ixodes scapularis*) to odors of sympatric rodent hosts
Erika Machtinger, Feizollah (Arash) Maleki, Karen Poh, James Tumlinson, III

A persistent behavior state in mosquitoes for pursuing humans
Trevor Sorrells, Anjali Pandey, Adriana Rosas, Leslie Vosshall

Personal care products alter mosquito-host interactions
Morgen VanderGiessen, Anaïs Tallon, Bryn Damico, Vinauger Clément, Chloé Lahondère

Species-specific differences in the mosquito olfactory response to repellents
Ali Afify, Joshua Betz, Olena Riabinina, Chloé Lahondère, Christopher Potter

Structural basis of olfactory receptor activation by an insect confusant
Joel Butterwick

S1-O137

Microstructures of ant chemosensory sensilla support a dual function in detecting both volatile and contact-mediated cues

Hannah Gellert, Daphné Halley, Zackary Sieb, Jody Smith, Gregory Pask

Middlebury College, Middlebury, USA

Ants and other eusocial insects emit and receive chemical signals to communicate important information within the colony. In ants, nestmate recognition, task allocation, and reproductive distribution of labor are largely mediated through the detection of cuticular hydrocarbons (CHCs) that cover the exoskeleton. With their large size and limited volatility, these CHCs are believed to be primarily detected through direct contact with the antennae during behavioral interactions. Here we use scanning electron microscopy to investigate the unique morphological features of CHC-sensitive basiconic sensilla of two ant species, the black carpenter ant *Camponotus pennsylvanicus* and the Indian jumping ant *Harpegnathos saltator*. These basiconic sensilla possess an abundance of small pores typical of most insect olfactory sensilla, but also have a large concave depression at the terminal end. Basiconic sensilla are enriched at the distal segments of the antennae in both species, further supporting their proposed role in contact chemosensation. A survey of these sensilla across other ant subfamilies shows varied microstructures at their tips, but each possess surface textures that would also increase sensory surface area. These unique ant chemosensory sensilla represent yet another example of how specialized structures have evolved to serve the functional requirements of eusocial communication.

Keywords: Ants, Antenna, Sensilla, SEM Imaging, Cuticular Hydrocarbons

S1-O124

Host detection by ticks in the context of repellent exposure

Nicoletta Faraone[1], N. Kirk Hillier[2]

[1]*Department of Chemistry, Acadia University, Wolfville (NS), Canada.* [2]*Department of Biology, Acadia University, Wolfville (NS), Canada*

Ticks are obligate blood-feeding ectoparasites of many hosts and are second only to mosquitoes as the most common arthropod pathogen vectors. Ticks use many different strategies to detect suitable hosts for a successful blood meal. Understanding tick olfaction and chemoreception has relevant implications in animal and human health, particularly for the development of novel repellent technologies. The effect of long-term exposure to repellent compounds on tick chemosensory systems are unknown, and no studies have studied potential long-term disruption of tick host detection. We investigated whether exposure to repellents subsequently impacted function of the tick chemosensory system. We recorded the electrophysiological response of adult *Ixodes scapularis* females to a known attractant and host volatile (i.e., butyric acid), pre- and post-exposure to selected known repellents (i.e., lemongrass, DEET). The behavioural effect of such exposure was further assessed through Y-tube bioassays. Ticks exhibited a reduced response to butyric acid post-exposure to lemongrass essential oil, indicating that some repellents may inhibit tick's ability to detect host volatiles and result in a decrease in sensitivity. These results provide new insights regarding tick olfaction and important information on mode of action of tick repellents.

Keywords: Electrophysiology, Tick, Host volatiles, Repellent exposure, Y-tube bioassays

[1] Faraone N, Light M, Scott C, MacPherson S, Hillier NK (2020) Insects 11: 502
[2] Faraone N, MacPherson S, Hillier NK (2019) Exp Appl Acarol 79: 195

S1-O155

The curious response of the blacklegged tick (*Ixodes scapularis*) to odors of sympatric rodent hosts

Erika Machtinger, Feizollah (Arash) Maleki, Karen Poh, James Tumlinson, III

Pennsylvania State University, University Park, USA

Lyme disease, caused by the bacteria *Borrelia burgdorferi* and vectored by *Ixodes scapularis* is the most reported vector-borne illness in the US. Host use by blacklegged ticks is highly heterogeneous which suggests more than random host encounters drive *I. scapularis* parasitism. We have observed extreme differences in tick burdens with two common sympatric hosts, *Peromyscus leucopus* (white-footed mouse) and *Microtus pennsylvanicus* (meadow vole) where tick burdens were>4 ticks per mouse, but <0.5 ticks per vole over a period of two years. Field research has failed to identify biological characteristics that are consistent drivers of this parasitism dichotomy and limited studies have been conducted on tick chemical ecology. While arrestment pheromones and sex pheromones have been explored for *I. scapularis*, attraction of blacklegged ticks to other compounds involved in host selection has not. Our major goal was to better understand the process of host selection from tick host choice to successful parasitism and identify candidate volatile organic compounds that may influence host attraction. We have established bioassays to evaluate tick responses to odors associated with mice and voles and collected headspace volatiles to assess chemical profiles. While rodents are considered important hosts for this tick species, tick responses to host odors were not as we had hypothesized. Current behavioral results will be discussed as well as a summary of odors from the two rodent species. Understanding these mechanisms behind host-parasite interactions and host selection is necessary to find novel ways to affect tick ecology for better control methods.

Keywords: Ticks, Hosts, Behaviour, Ecology

[1] Centers for Disease Control and Prevention (CDC) (2018) Illness on the rise from mosquito, tick, and flea bites. Vital Signs, May
[2] Allan SA, Sonenshine DE (2002) Evidence of an assembly pheromone in the black-legged deer tick, Ixodes scapularis. Journal of Chemical Ecology 28: 15–27

S1-O129

A persistent behavior state in mosquitoes for pursuing humans

Trevor Sorrells, Anjali Pandey, Adriana Rosas, Leslie Vosshall

Rockefeller University, New York, USA

Predatory animals use sensory cues such as scent to detect their prey then pursue them over time and distance to obtain a meal. It is critical for predators to persist in the chase even when prey is difficult to detect but also to abandon pursuit if enough time has elapsed that the prey is no longer present. We studied prey detection and pursuit in the mosquito *Aedes aegypti*, a micropredator of humans. Mosquitoes detect humans through sensory cues such as carbon dioxide in breath and body heat from human skin. To study how initial prey detection influences the duration of pursuit, we developed optogenetic tools to induce a brief fictive sensation of carbon dioxide and used machine learning-based classification of behavior to investigate how mosquitoes respond to subsequent human cues. We found that a 5-second optogenetic pulse of fictive carbon dioxide induced a persistent behavioral state in female mosquitoes that lasted for more than 10 minutes. This state is highly specific to females searching for a blood meal and was not induced in recently blood-fed females or in males, who do not feed on blood. We show that the persistent state triggered by detection of fictive carbon dioxide enabled females to engorge on a blood meal mimic offered up to 14 minutes after the initial stimulus. Our results demonstrate that a persistent internal state allows female mosquitoes to integrate multiple human sensory cues over long timescales, an ability that is key to their success as an apex micropredator of humans.

Keywords: -

S1-O157

Personal care products alter mosquito-host interactions

Morgen VanderGiessen, Anaïs Tallon, Bryn Damico, Vinauger Clément, Chloé Lahondère

Virginia Tech, Blacksburg, USA

Female mosquitoes predominantly rely on their sense of olfaction and use volatile organic compounds (VOC) emitted by plants and human hosts to identify and locate food resources essential to their survival and reproduction. In line with the principle of chemical parsimony, these resources can overlap in their chemical composition and an important layer of additional information resides in the relative abundance of VOCs in the headspace composition of each resource. Interestingly, since the Mesopotamian and Egyptian antiquity, humans have developed the habit of using plant-derived fragrances, first as part of religious rituals and later on for cosmetic purposes. Nowadays, the use of personal care products (PCPs) such as soaps, shampoos, deodorants, and perfumes, which add plant-related VOCs to the composition of their olfactory signature, is highly common. Despite strong epidemiological relevance, how and to what extent, this phenomenon affects mosquitoes' host-seeking behavior has been largely overlooked. Here, using headspace sampling and gas-chromatography mass-spectrometry, we analyzed the extent to which human body odor is modified by the application of soap. Quantifying *Aedes aegypti* mosquitoes' responses to soap-washed human hosts, we then showed that the application of soap significantly alters host selection processes, with some soaps increasing the attractiveness of the host and some soaps reducing it. Finally, analytical methods allowed us to identify the main chemicals associated with either enhanced or reduced attraction to create artificial mixtures recapitulating the observed behavioral effects. These mixtures could potentially be used as baits or repellents for control purposes.

Keywords: *Aedes aegypti*, host seeking behavior, personal care products, mosquito control

S1-O123

Species-specific differences in the mosquito olfactory response to repellents

Ali Afify[1,2], Joshua Betz[3], Olena Riabinina[4], Chloé Lahondère[5], Christopher Potter[1]

[1]*Johns Hopkins University School of Medicine, Baltimore, USA.* [2]*Drexel University, Philadelphia, USA.* [3]*Johns Hopkins Bloomberg School of Public Health, Baltimore, USA.* [4]*Durham University, Durham, United Kingdom.* [5]*Virginia Polytechnic Institute and State University, Blacksburg, USA*

The mode of action for mosquito repellents is not well understood. This is due to the difficulty in monitoring how the mosquito olfactory neurons respond to odors. Here, we engineered transgenic *Anopheles coluzzii* mosquitoes to enable activity-dependent Ca^{2+} imaging in their olfactory neurons. This system allows neuronal responses to common insect repellents to be directly visualized in living mosquitoes from all olfactory organs including the antennae. We found that man-made repellents such as DEET do not activate *Anopheles* olfactory receptor neurons (ORNs). In contrast, natural repellents such as lemongrass oil strongly activated small numbers of ORNs in the *Anopheles* mosquito antennae. We present evidence that man-made repellents reduce the volatility of human-derived odorants leading to a decreased olfactory response towards these odorants. This suggests that man-made repellents disruptively change the chemical profile of human scent signatures on the skin surface rendering humans invisible to *Anopheles* mosquitoes. To test our findings, we developed a behavioral assay in which repellents can be tested directly, independent from human odors. We found that DEET directly repels *Aedes* and *Culex* mosquitoes but not *Anopheles* mosquitoes, while lemongrass oil was repellent for all three species. This suggests that confusion in the field about DEET's mode of action is partially due to species-specific differences in the mosquito responses to DEET. Overall, these findings will help guide decisions on choosing the right repellent for each mosquito species and will help develop safer alternatives to currently used mosquito repellents.

Keywords: Olfaction, Mosquitoes, Repellents, Human Odorants, Calcium Imaging

S1-O181

Structural basis of olfactory receptor activation by an insect confusant

Joel Butterwick

Yale University, New Haven, USA

The insect odorant receptor co-receptor (Orco) is a special member of the odorant receptor (OR) family. Orco is part of each olfactory receptor complex but remains silent so that odor identity can be encoded by the activity of odorant-binding OR subunits. Here, we leverage a structural approach to identify a mechanism that impedes Orco from interacting with odorants and discover how the synthetic agonist VUAA4 overcomes this obstruction to activate olfactory receptors via Orco. Using single-particle cryo-electron microscopy (cryo-EM), we visualized Orco embedded in a membrane-like environment (nanodisc) and observed a phospholipid situated similarly to where ORs bind odorants. Lipid binding likely prevents spurious Orco activation by occupying the ligand-binding site and blocking odorants from interacting with Orco. VUAA4 does not bind in the canonical odorant-binding pocket, but instead bypasses the phospholipid barrier and directly contacts the channel pore. Through rational mutagenesis, we created a modified Orco with higher affinity for VUAA4. Our work reveals the structural basis for insect olfactory receptor activation by small molecules that act as sensory confusants.

Keywords: Olfaction, Olfactory receptor, Sensory confusant, Cryo-EM

Symposium 1: Frontiers in Chemical Ecology (2)

Friday, August 12th
9 am - 12:30 pm

Session moderated by Walter S Leal
Session sponsored by BedoukianBio

Applying reverse genetics to evaluate complex sensory mechanisms in Lepidoptera
Richard A. Fandino

Molecular and neural basis of phenylacetaldehyde detection in the Cotton Bollworm *Helicoverpa armigera*
Mengbo Guo, Baiwei Ma, Yang Liu, Guirong Wang

A conserved anoctamin regulates olfactory neuron firing
Pratyajit Mohapatra, Darya Task, Joshua Raji, Johannes Reisert, Christopher Potter, Karen Menuz

Mutagenesis of the odorant receptor co-receptor (orco) reveals severe olfactory defects in a crop pest moth, *Helicoverpa armigera*
Xiao-Bin Fan, Bao-Tong Mo, Guo-Cheng Li, Ling-Qiao Huang, Hao Guo, Xin-Lin Gong, Chen-Zhu Wang

Refolding of odorant binding proteins (OBPs) and a new binding assay method
Aniruddha Agnihotri, Wei Xu

Olfactory variation among closely related cactophilic Drosophila species
Amber Crowley-Gall, Aaron Hamrick, Lucinda Lawson, John Layne, Stephanie Rollmann

Floral attraction of mosquito pollinators
Dan Peach, Regine Gries, Elton Ko, Adam Blake, Gerhard Gries

What drives a tick to seek a host and/or a habitat?
Zainulabeuddin Syed

S1-O159

Applying reverse genetics to evaluate complex sensory mechanisms in Lepidoptera

Richard A. Fandino

Cornell University, Ithaca, USA

Insects interact with their environment through sensory behaviors that impede our wellbeing or support ecological diversity. These behaviors are often elicited through sensory mechanisms that integrate multiple cues or signals through coordinated expression of sensory genes and metabolic states. In the past our understanding of these sensory mechanisms has been limited to a select number of genetic model organisms. The fundamental relationship between Lepidoptera and plants, however, provides an avenue to understand deep evolution of behavioral traits that affect ecosystem stability or agriculture. CRISPR-Cas9 allows a semi-feasible route for in vivo mechanistic insight into gene, gene function, and behavior in a number of traditional non-genetic model Lepidoptera species. For example, we applied a reverse genetic approach using CRISPR-Cas9 to generate a line of partially anosmic hawkmoths and measure the importance of odorant sensory reception in relation to the full stimulus of the host plant. We found that natural behaviors were differentially disrupted suggesting different modalities of sensory integration directing mating, foraging and oviposition behaviors. In this talk I will briefly discuss the feasibility of reverse genetics in Lepidoptera as well as offer a glimpse into an integrative approach that examines the relative importance of different sensory modalities and mechanisms that guide important plant seeking behaviors in butterflies and moths.

Keywords: CRISPR-Cas, Lepidoptera, Olfaction, Neurobiology, Genetics

S1-O117

Molecular and neural basis of phenylacetaldehyde detection in the Cotton Bollworm *Helicoverpa armigera*

Mengbo Guo[1,2], Baiwei Ma[2], Yang Liu[2], Guirong Wang[1,2]

[1]*Agricultural Genomics Institute at Shenzhen, Chinese Academy of Agricultural Sciences, Shenzhen, China.* [2]*Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China*

Olfactory cues are crucial in guiding multiple ecological behaviors of insects for both survival and reproduction. It is critical for the olfactory system to recognize ecologically relevant molecules from complex background odors. The olfactory signal is perceived and processed by a highly efficient and precise detection system that detects odor molecules using olfactory receptors (ORs) expressed in the dendritic membrane of olfactory sensory neurons (OSNs) at the periphery, from where the neural signals are conveyed to the olfactory processing center of the brain. To deepen our understanding of the interactions between insects with their surrounding environment, we need to address the questions of which receptor is employed by a particular insect species to detect a specific ecologically important odor? Moreover, what coding strategy is operated by the olfactory system? Lepidoptera represents a critical insect order with highly diverse organisms that play important ecological roles as pollinators and herbivores. However, little functional data about the Lepidoptera OR repertoire involved in plant-volatile detection is available. We profile the functions of the OR repertoire in the *Helicoverpa armigera*, and then conducted a large-scale comparative analysis to demonstrate that the function of a particular OR (OR42) for sensing the common flower volatile phenylacetaldehyde (PAA), a floral scent component common to most angiosperms and attracts multiple moth and butterfly species, is highly conserved across lepidopteran species. By conducting loss-of-function studies combined with neuroethology in *H. armigera*, we showed that HarmOR42 underlies the molecular basis of the attractiveness of PAA, which is coded by a sensitive and specific neural pathway.

Keywords: Olfactory perception, *Helicoverpa armigera*, Phenylacetaldehyde, Neural pathway

S1-O114

A conserved anoctamin regulates olfactory neuron firing

Pratyajit Mohapatra[1], Darya Task[2], Joshua Raji[2], Johannes Reisert[3], Christopher Potter[2], Karen Menuz[1]

[1]*University of Connecticut, Storrs, USA.* [2]*Johns Hopkins University School of Medicine, Baltimore, USA.* [3]*Monell Chemical Senses Center, Philadelphia, USA*

Insect olfactory systems are key targets in the development of agents to prevent the negative impact of insect behavior on human health and economy. The past few decades have witnessed an explosion of research into the odorant response profiles and behavioral significance of most receptors in the widely-used model organism *Drosophila melanogaster*. However, these receptors are poorly conserved amongst insect species, and few other molecules that contribute to olfactory signaling have been identified. We previously used a bioinformatics screen to detect several highly conserved candidate genes with potential roles in insect olfaction. Here, we have studied one candidate gene, CG6938, a member of the anoctamin family. In Drosophila, this gene is selectively expressed in chemosensory neurons, including all classes of olfactory neurons. It has similar broad olfactory neuron expression in the antennae of Anopheles mosquitoes. In Drosophila, CG6938 protein is localized to olfactory neuron dendrites, consistent with a functional role in sensory transduction. Our functional data indicate that CG6938 is a calcium-activated chloride channel, like vertebrate orthologs. Mutations in CG6938 prevent the spiking response of olfactory neurons from closely tracking the level of fluctuating odorants, likely due to a role in signal termination. Such effects are seen in multiple combinations of olfactory neurons and their cognate odorants. Together, our data suggest that CG6938 may play a conserved role in regulating olfactory signal transduction in insects.

Keywords: Olfaction, Neuron, Ion channel, Calcium, Chloride

S1-O111

Mutagenesis of the odorant receptor co-receptor (Orco) reveals severe olfactory defects in a crop pest moth, *Helicoverpa armigera*

Xiao-Bin Fan[1,2], Bao-Tong Mo[1,2], Guo-Cheng Li[1,2], Ling-Qiao Huang[1], Hao Guo[1,2], Xin-Lin Gong[1,2], Chen-Zhu Wang[1,2]

[1]State Key Laboratory of Integrated Management of Pest Insects and Rodents, Institute of Zoology, Chinese Academy of Sciences, Beijing, China. [2]CAS Center for Excellence in Biotic Interactions, University of Chinese Academy of Sciences, Beijing, China

Odorant receptors (ORs) are ligand-gated nonselective ion channels that are necessary for olfactory detection in insects while there are other types of olfactory receptor proteins, such as: ionotropic receptors (IRs), some gustatory receptors (GRs) also involved in insect olfactory system. Orco (the odorant receptor coreceptor) is an obligatory subunit of insect OR ion channels and highly conserved, thus providing an opportunity to systematically evaluate OR-dependent olfactory responses. Herein we successfully established the *orco*$^{-/-}$ mutant of *Helicoverpa armigera*, a notorious crop pest, using CRISPR/Cas9 gene editing technique, and compared the changes in olfactory response characteristics of adults and larvae. At first, we found the female lifespans of the *orco*$^{-/-}$ mutant were longer than that of the wild type, and the mutant was infertile when self-crossed. Although, some ORs in adult antennae were up- or down-regulated in *orco*$^{-/-}$ mutant moths, the expressions of most conventional ORs and IRs were not obviously affected. In addition, there was no change of neuro-anatomical phenotypes in *orco*$^{-/-}$ mutant moths at the level of antennal lobes (including the male MGC region). Using EAG and SSR techniques, we discovered that the electrophysiological responses of the *orco*$^{-/-}$ mutant moths to sex pheromone components and many host plant odorants were abolished. Meanwhile, the mutant males completely lost the upwind flying behavior to the sex pheromone in the wind tunnel, the mutant females exhibited severely impaired oviposition preferences to the host plant green pepper, and the mutant larvae lost their chemotaxis to green pepper. In conclusion, our study confirms that the olfactory signal pathway mediated by ORs (OR-dependent olfactory response) is central for *H. armigera* to find a mate and select host plants, providing an important target for behavior regulation and novel pest control strategies.

Keywords: Olfaction, Odorant receptor co-receptor, CRISPR/Cas9, *Helicoverpa armigera*

S1-O40

Refolding of odorant binding proteins (OBPs) and a new binding assay method

Aniruddha Agnihotri, Wei Xu

Murdoch University, Murdoch, Australia

Insect behaviours are guided by olfactory systems, in which odorant binding proteins (OBPs) are one of the crucial components and play an essential role in transporting the hydrophobic volatile odorant molecules to the olfactory receptors. Most insect OBPs were investigated by using recombinant technology in bacterial cells because it is a fast, cost-effective, and high production system. However, one of the major concerns of bacterial expression system is that the protein is frequently expressed in an unfolded state in inclusion bodies (IBs), which requires further *in vitro* protein refolding step to make the protein biologically active. While doing this, there are always high chances of protein misfolding which results in soluble or insoluble protein aggregation. Thus, it is highly important to confirm the efficiency of each refolding method, used for OBP refolding, in terms of getting the correctly folded structure of the target protein. Unfortunately, it was neglected in many previous studies, resulting in doubts on various functional studies of insect OBPs. In this study, three *Helicoverpa armigera* OBPs, HarmOBP2, HarmOBP5, and HarmGOBP2, were selected as model proteins to compare the different protein refolding strategies in producing correctly folded recombinant OBPs. Along with that, a novel pH-dependent method for protein refolding was developed, which was demonstrated as a more efficient and productive approach for selected HarmOBPs' refolding compared to other used methods. Further, a novel reverse chemical ecology method was developed to isolate and identify the candidate natural ligands from host plants for HarmOBPs. This study points out a crucial but largely ignored step of insect OBP research, protein refolding and the loopholes associated with it in previous studies, which will improve our understanding of insect olfaction and help develop more efficient and environmentally friendly insect control strategies.

Keywords: Odorant Binding Protein, OBP, Refolding, Reverse Chemical Ecology

S1-O185

Olfactory variation among closely related cactophilic Drosophila species

Amber Crowley-Gall[1], Aaron Hamrick[2], Lucinda Lawson[2], John Layne[2], Stephanie Rollmann[2]

[1]*University of California Davis, Davis, USA.* [2]*University of Cincinnati, Cincinnati, USA*

Chemical cues are important for a wide range of tasks such as host plant identification and localization, oviposition site selection, and mate recognition. Insects use volatile cues emitted from plants when navigating toward an appropriate host and divergence in odor detection has been shown to result in shifts in host use between populations and in some cases reproductive isolation between populations and eventual speciation. A comparative phylogenetic approach can determine whether variation in the olfactory system is linked to shifts in host plant use and is a means to determine the influence of olfactory tuning on divergence between species. A useful model to examine this is the *Drosophila repleta* species group, a radiation of flies specializing on cacti, that exhibits three types of host use: 1) *Opuntia* specialists, 2) columnar specialists, and 3) "generalists" on both. *Opuntia*, a flat leaf cactus, is hypothesized to be the ancestral host, and the use of the more chemically complex columnar cactus is believed to be an acquired trait. *Columnar cacti* contain elevated levels of secondary compounds that can be toxic to flies and affect the volatile headspace flies are exposed to when choosing a suitable host plant. This study examined the extent to which odor tuning has diverged along with the repeated shifts in host plants within the *Drosophila repleta* species group. We characterized odor response profiles from select sensillar subtypes across multiple species within the *repleta* group as well as the outgroup *D. melanogaster*. Variation in both sensitivity and specificity to odors was observed, with some ORNs exhibiting variation associated with host cactus use. This study is the first in-depth analysis of the olfactory system across the *repleta* group and provides the opportunity to test for conserved mechanisms in the olfactory system underlying divergence and host shift.

Keywords: Olfaction, Speciation, *Drosophila*, Cactophilic

S1-O170

Floral attraction of mosquito pollinators

Dan Peach[1,2], Regine Gries[2], Elton Ko[2], Adam Blake[2,3], Gerhard Gries[2]

[1]*University of Georgia, Athens, USA.* [2]*Simon Fraser University, Burnaby, Canada.* [3]*The University of Washington, Seattle, USA*

Floral nectar is the primary food of adult male and female mosquitoes. Heretofore, the role of sugar-foraging mosquitoes as pollinators was not clear and how mosquitoes are attracted to sources of floral nectar has only been partially elucidated. Working with flowers of *Tanacetum vulgare* and *Hieracium lachenalii*, and mosquitoes such as *Culex pipiens*, and *Aedes aegypti*, we tested the hypotheses that (H1) mosquitoes provide pollination services; and (H2) the entire inflorescence Gestalt of olfactory and visual cues is more attractive to sugar-foraging mosquitoes than floral odorants alone. Testing H1, we demonstrate that *Cx. pipiens* readily carries pollen and induces significant seed set in *T. vulgare*. Testing H2, we demonstrate that (i) olfactory and visual *T. vulgare* inflorescence cues play a role in mosquito attraction and (ii) UV light-reflecting and absorbing patterns of *H. lachenalii* inflorescences affect mosquito foraging decisions. Future research on mosquito pollination and the relation between floral odor and vertebrate host-preference is discussed.

Keywords: Floral odor, Mosquitoes, Mosquito sugar-feeding, Mosquito Pollination

[1] Foster, W. (1995) Mosquito sugar feeding and reproductive energetics. Annu Rev of Entomol 40:443-474.
[2] Peach, D.A.H., and Gries, G. (2020) Mosquito phytophagy – sources exploited, ecological function, and evolutionary transition to haematophagy. Entomol Exp Appl 168: 120-136.

S1-O162

What drives a tick to seek a host and/or a habitat?

Zainulabeuddin Syed

Department of Entomology, University of Kentucky, Lexington, USA

Ticks display robust olfactory driven behaviors and have a remarkable biology that drives these behaviors. They survive the longest period of starvation among arthropods, have limited mobility, are adapted to feed on multiple host types within a single lifetime, and demonstrate exceptionally low metabolic rates. A confluence of these factors triggers a unique host-seeking strategy termed as ambushing. This strategy – originally described as a 'waiting attitude' while the tick is in the alert mode (with forelegs extended) on a vertical vegetative structure such as a grass blade, and later defined as 'questing' – leads a tick to exploit a host for blood-feeding. A distinct apparatus called Haller's organ is located on the foretarsal region of the first pair of legs and serves as the only olfactory organ in ticks. Blacklegged tick, *Ixodes scapularis*, are equipped with only ca. 20 olfactory sensilla in and around the Haller's organ that detect and define the complete olfactory landscape. I will present results from our ongoing Gas-chromatography-coupled single sensillum recordings (GC-SSR) and behavioral observations that are leading us towards the development of innovative vector management strategies.

Keywords: Haller's organ, Single unit recordings, Circadian behavior, Saturation deficit, Pheromones

[1] Josek, T., Sperrazza, J., Alleyne, M. and Syed, Z. Neurophysiological and behavioral responses of blacklegged ticks to host odors. Journal of insect physiology 128, 104175 (2021).
[2] Syed, Z and O'Dell, K.L. Jr. Finding a repellent against ticks: neurophysiological and behavioral approaches" in Advances in Arthropod Repellents. (Elsevier, 2022), pp. 131-140.

Symposium 2: Application of semiochemicals in insect pest management (1)

Wednesday, August 10th
3:20 pm - 6 pm

Session moderated by Aijun Zhang

RESCUE!® Semiochemical-based fly traps for consumer market
Qing-He Zhang

Is the symbiotic yeast *Kodamaea ohmeri* playing a role in the aggregative behaviour of the honeybee pest *Aethina tumida* (Coleoptera: Nitidulidae)?
Brogan Amos, Michael Furlong, Bronwen Cribb, Diana Leemon, R Andrew Hayes

Infestation of Norway spruces by tree-killing bark beetles *Ips typographus* was prevented by anti-attractant baits consisting of newly added *trans*-4-thujanol.
Anna Jirošová, Rastislav Jakuš, Roman Modlinger, Fredrik Schlyter

Development of a push & pull strategy for spotted wing Drosophila control in blueberry orchard
Aijun Zhang, Nick Larson, Cody Gale, Simon Zebelo, Tigist Tolosa

Effect of soybean volatiles on the behavior of the bean bug, *Riptortus pedestris*
Junyong Song, Gisuk Lee, Jinkyo Jung, Jung-Kyung Moon, Sang-Gyu Kim

Identification of bed bug probing stimulants from human skin for bait development
Chen Zha[1], Jose Pietri, Dangsheng Liang

Effectiveness of Wota-T traps baited with sex pheromone to control the sweet potato weevil *Cylas formicarius* (Fabr.) in Vietnam
Arnaud Costa, Sivapragasam Annamalai, Hanh Thi My Tran

Release of pheromone compounds from *Nicotiana benthamiana* leaf glandular trichome upon transient expression of biosynthetic genes
Bao-Jian Ding, Hong-Lei Wang, Christer Löfstedt

S2-O13

RESCUE!® Semiochemical-based fly traps for consumer market

Qing-He Zhang

Sterling International, Inc., Spokane, USA

Sterling International, Inc., is a semiochemical-based company manufacturing RESCUE!® brand traps/attractants and repellents for pestiferous social wasps, filth flies, Japanese/Oriental beetles, stink bugs, vinegar fruit flies, pantry moths, mosquitoes and spiders for the consumer retail market. Unlike agricultural, commercial, and professional pest control operator (PCO) markets, the consumer market consists primarily of household consumers who buy semiochemical-based pest control products as a "Do It Yourself" (DIY) trapping technology for individual or family consumption. Our RESCUE!® traps, repellents and ant baits are available at all major retail stores in the United States and Canada, such as Wal-Mart, Home Depot, Lowes, ACE Hardware, and Costco, plus online sites like Amazon. The massive scale of these retailers provides tremendous opportunities for sale revenues and profits; but this marketplace is also extremely competitive for companies such as ours competing for limited shelf space. This year we are celebrating the 40th anniversary of the company. In this presentation, I will discuss the history, current successes, and future challenges of RESCUE!® semiochemicals-based fly traps targeting at the common nuisance/filth flies (e.g. house flies, blow/bottle flies and flesh flies) for the consumer market.

Keywords: Attractant, Trap, House fly, Blow fly, Flesh fly

[1] Schneidmiller, RG (1987) U.S. Patent # 4,638,592; issued on January 27, 1987

S2-O17

Is the symbiotic yeast *Kodamaea ohmeri* playing a role in the aggregative behaviour of the honeybee pest *Aethina tumida* (Coleoptera: Nitidulidae)?

Brogan Amos[1,2], Michael Furlong[1], Bronwen Cribb[1], Diana Leemon[3], R Andrew Hayes[4]

[1]*The University of Queensland, St Lucia, Australia.* [2]*James Cook University, Cairns, Australia.* [3]*Queensland Department of Agriculture and Fisheries, Brisbane, Australia.* [4]*University of the Sunshine Coast, Brisbane, Australia*

The small hive beetle *Aethina tumida* (Coleoptera: Nitidulidae) is a major pest of western honeybee hives in the USA and Australia. The beetle vectors a symbiotic yeast into hives, which is responsible for fermentation of hive products. The beetle displays aggregative behaviour and previous research has identified an aggregation pheromone. The aggregative behaviour of the beetle has implications for transmission of this yeast between beetles. We used laboratory assays to investigate the conditions under which this aggregation occurs and the purpose of this aggregation, beyond its reported role in assisting reproduction, and to understand the role of the symbiotic yeast *Kodamaea ohmeri* (Ascomycotina: Saccharomycotina) in the aggregation. We investigated clustering of mated and unmated male and female conspecifics, as well as clustering with dead conspecifics. We also examined the beetle's ability to form clusters without the associated yeast. Aggregation under laboratory conditions appears to be sex neutral, only occurs in response to other live *A. tumida*, and is facilitated by the presence of *K. ohmeri*. We determined that beetles reared without *K. ohmeri* took longer to form clusters (= aggregation of five or more beetles) and the clusters formed were smaller. We determined that horizontal transmission of *K. ohmeri* is possible and appears to be facilitated by the beetle's aggregation behaviour. Following the differences in behaviour of *K. ohmeri*-free beetles, hexane cuticular extraction analysis showed differences in the cuticular compounds of *K. ohmeri*-free beetles and untreated beetles. This work extends our current understanding of *A. tumida* aggregative behaviour and conspecific communication, which may have potential in control and management strategies for this pest.

Keywords: Small hive beetle, Aggregation, Insect-fungal interactions, CHC

S2-O24

Infestation of Norway spruces by tree-killing bark beetles *Ips typographus* was prevented by anti-attractant baits consisting of newly added *trans*-4-thujanol.

Anna Jirošová[1], Rastislav Jakuš[1,2], Roman Modlinger[1], Fredrik Schlyter[1,3]

[1]*EXTEMIT-K, ETM, Faculty of Forestry and Wood Sciences, Czech University of Life Sciences Prague, Prague, Czech Republic.* [2]*Institute of Forest Ecology, Slovak Academy of Sciences, Zvolen, Slovakia.* [3]3*Department of Plant Protection Biology, Chemical Ecology, Swedish University of Agricultural Sciences, Alnarp, Sweden*

The Eurasian spruce bark beetle (*Ips typographus*; Coleoptera) is currently the most spruce-destroying insect in Central Europe. Conifer bark beetles use both attractant and anti-attractant semiochemicals to find suitable host trees. Previously, for the protection of spruce trees against *I. typographus* was tested the anti-attractants mixture consisting of non-host tree substances *trans*-conophthorin, 1-hexanol, 1-octene-3-ol and 1-octanol, and from the old-host bark compound - verbenone. Here, we report the activity of the improved mixture with the addition of *trans*-4-thujanol, a newly discovered potent anti-attractant and with the replacement of verbenone by the 1,8-cineole, a spruce defense compound. In the field trapping experiment, we found the strong inhibition activity of the complete mixture, which in combination with pheromone, lowered beetle catches by 100% in comparison to pheromone itself. We established the experiment to study the individual components' synergistic effect in the mixture. Consequently, we conducted two experiments for the protection of spruce trees. First, we placed the baits on trunks of spruces in the two rows of fresh forest edges, the first beetles' target. We recorded the first attacked trees at 40 m and their high density at 70 m far from baited trees. The second experiment evaluated the protection of young spruce forest fragments. Traps with pheromone or pheromone in combination with anti-attractants were placed in the vicinity of twelve fragments. The eighteen trees close to the pheromone itself were infested, and the trees close by the pheromone, in combination with the anti-attractant, remained protected.

Keywords: Eurasian spruce bark beetles, 1,8-cineol, Coleoptera: Curculionidae: Scolytinae, Repellent, Integrated pest management

[1] Schiebe C, Blazenec M, Jakus R, Unelius CR, Schlyter F (2011) J Appl Entomol, 135: 726.
[2] Jirošová A, Kalinová B, Modlinger R, Jakuš R, Unelius CR, Blaženec M and Schlyter F (2022), Pest Manag Sci, 78: 1992

S2-O27

Development of a push & pull strategy for spotted wing Drosophila control in blueberry orchard

Aijun Zhang[1], Nick Larson[1], Cody Gale[1], Simon Zebelo[2], Tigist Tolosa[2]

[1]*USDA ARS, Beltsville, USA.* [2]*University of Maryland Eastern Sore, Princess Anne, USA*

The Spotted Wing Drosophila (SWD), *Drosophila suzukii* Matsumura (Diptera: Drosophilidae), is a fruit fly native to Southeast Asia. Since its introduction in 2008, this invasive species has become a devastating pest of soft-skinned fruit crops that causes huge economic losses in the United States and many European countries. Significant efforts have been made for development of reliable and sustainable control tools for this pest. In our previous study, SWD attractants and repellents/toxicants have been identified from apple juices. In this study, the efficacy of push & pull strategy using previous identified SWD repellent, methyl benzoate, as a push agent and attractant as a pull agent was evaluated in two blueberry orchards in Maryland state. Our preliminary results in 2020 indicated that 1) SWD population was not significantly reduced by the push & pull system and 2) although the SWD emergency rates from blueberries were reduced from push & pull treatments, they are not significantly different from blank control treatment. More research, testing a broader range of repellent loadings, different experimental design with big buffer zones to reduce the interferences between blocks, and different fruit damaging evaluation method will be conducted in 2022 to develop the push & pull strategy into an efficient, safe, and environmentally friendly SWD-control method.

Keywords: Fruit Fly, Semiochemicals, Filed tests, Pest Management

[1] Feng Y, Bruton R., Park A, Zhang A (2018) J Pest Sci 91: 1251-1267
[2] Zhang A, Feng Y (2020) US Patent 10638754
[3] Feng Y, Zhang A (2017) Sci Rep 7: 42168
[4] Zhang A, Feng Y (2017) US Patent 9629362
[5] Zhang A, Feng Y, Chen, J (2019) WO Patent 2019143506A1

S2-O29

Effect of soybean volatiles on the behavior of the bean bug, *Riptortus pedestris*

Junyong Song[1], Gisuk Lee[1], Jinkyo Jung[2], Jung-Kyung Moon[3], Sang-Gyu Kim[1]

[1]Korea Advanced Institute of Science and Technology, Daejeon, Republic of Korea. [2]National Institute of Crop Science, Suwon, Korea, Republic of. [3]National Institute of Crop Science, Wanju, Republic of Korea

The bean bug, *Riptortus pedestris*, is a major pest of soybean (*Glycine max*), feeding on various leguminous plants. Although its economic importance, little is known about how the insect locates and colonizes soybean fields. In this study, we examined the electroantennographic and behavioral responses of the adult *R. pedestris* to soybean volatiles. *R. pedestris* adults were attracted more to their host-plant soybean, even when physical contact was absent than to air or a non-host plant. Accordingly, we hypothesized that *R. pedestris* can recognize soybean through a plant's volatile organic compounds (VOCs). Five VOCs were identified from intact soybean plants at the vegetative stage: (*Z*)-3-hexen-1-ol, (*Z*)-3-hexenyl acetate, 4-ethylbenzaldehyde, α-farnesene, and methyl salicylate. Response spectra of the antennae to these volatiles clearly showed that both male and female *R. pedestris* can detect soybean volatiles. The adult bean bugs did not show behavioral orientation to any individual compounds but showed significant orientation to a particular blend of synthetic soybean volatiles when tested under laboratory conditions. In the field, this soybean volatile blend did not significantly attract the bean bugs, but it did interact synergistically with the aggregation pheromone by significantly increasing the attraction of the adult bean bugs. These results highlight the role of host plant volatiles in the sensory ecology of *R. pedestris* and help explain the colonization pattern of the bean bugs in soybean fields.

Keywords: Glycine max, Host-plant volatiles, Riptortus pedestris, Olfactory response, Aggregation pheromone

[1] Song J, Lee G, Jung J, et al. (2022) Journal of Chemical Ecology 48:207-218

S2-O41

Identification of bed bug probing stimulants from human skin for bait development

Chen Zha[1], Jose Pietri[2], Dangsheng Liang[1]

[1]*Apex Bait Technologies, Inc., Santa Clara, USA.* [2]*University of South Dakota, Vermillion, USA*

The common bed bug, *Cimex lectularius* L. (Hemiptera: Cimicidae), is a blood-feeding ectoparasite which experienced world-wide resurgence during recent decades. Previous studies have focused on long distance attractants such as aggregation pheromone and host cues and their findings have significantly improved the effectiveness of traps. However, for the development of bed bug baits, it is critical to understand the close range behavior after bed bugs have located their hosts. In this study, we developed a novel probing stimulant bioassay and examined the bed bug probing stimulants from human skin. We have identified several compounds that showed significant probing stimulation effects. We formulated a chemical blend that showed similar probing stimulating effects to human skin swabs.

Keywords: Bed Bug, Bait, Probing Stimulant, Blood Feeding, Skin Swab

S2-O83

Effectiveness of Wota-T traps baited with sex pheromone to control the sweet potato weevil *Cylas formicarius* (Fabr.) in Vietnam

Arnaud Costa[1], Sivapragasam Annamalai[1], Hanh Thi My Tran[2]

[1]*Centre for Agriculture and Bioscience International (CABI) South East Asia, Serdang, Malaysia.* [2]*Southern Horticultural Research Institute (SOFRI), My Tho, Vietnam*

The Sweet Potato Weevil (SPW), *Cylas formicarius*, (Fabr.) is a major pest of sweet potato worldwide, being able to reduce up to 80% of the yield. In Vietnam, sweet potato is grown in large monocultures often prone to SPW damage. Insecticide treatments are by far the most common control method used by farmers. In previous studies conducted in other regions, SPW female-produced sex pheromone, (Z)-3-dodecen-1-ol (E)-2-butenoate [1], has been found to be very attractive to SPW males. We conducted trials in Vinh Long province (Vietnam), a key area for sweet potato production, to compare the use of SPW pheromone to traditional farmers methods (e.g., chlorantraniliprole). We compared the use of SPW pheromone to insecticide treatment and recorded the abundance of SPW males using Wota-T-water traps set at a density of 16 to 20 traps/acre. The economic impact (tuber marketability) was also assessed. Results showed that SPW pheromone tested in combination with Wota-T traps was highly attractive to SPW males (N= 392.4 ± 32.6, P< 0.001). For small holes created by SPW larvae, the use of pheromone was found as effective as conventional insecticide treatment (P = 0.183). However, larger holes, made by immature of the sweet potato tuber moth (SPTM) *Nacoleia* sp. (Lepidoptera: Crambidae) were recorded in SPW pheromone treatment (N= 19.0 ± 1.6 holes compared to N= 2.3 ± 0.3 in insecticide treatment, P< 0.0001), creating a significant reduction (32.8%) in high quality tubers (P< 0.001). In Vietnam, SPW sex pheromone used with Wota-T traps can be a promising alternative to repeated insecticide use, but further studies will investigate how to integrate it better when other local pests also affect tuber marketability.

Keywords: Sweet potato weevil, Sex pheromone, Mass trapping, Integrated pest management, Vietnam

[1] Heath RR et al. (1986) Journal of Chemical Ecology 12: 1489-1503

S2-O93

Release of pheromone compounds from *Nicotiana benthamiana* leaf glandular trichome upon transient expression of biosynthetic genes

Bao-Jian Ding, Hong-Lei Wang, Christer Löfstedt

Department of Biology, Lund University, Lund, Sweden

Sustainable agricultural practices rely on reduced inputs while maintaining productivity. As a major alternative to the use of pesticides insect pheromones can be used for mass trapping and mating disruption of many serious pests. Pheromones are species specific, limited negative impact on beneficial non-target organisms such as pollinators and non-toxic to human. Unfortunately, synthetic insect pheromones remain expensive, and their synthesis may result in environmentally hazardous by-product. Plant-based production of insect sex pheromones has been proposed as an innovative strategy to increase the sustainability of pest control in agriculture. Pheromone compounds made by the plant can be released upon expression of heterologous biosynthetic genes, may serve as live dispensers in the field. The aim of this study is to develop pheromone-releasing plants through genetic engineering. We first characterized a few trichome-specific promoters to direct the expression of pheromone biosynthetic genes to glandular trichomes to avoid undesirable effects on plant growth and development of using 35S promoter to produce potentially toxic compounds. Optimal arrangement of promoters and genes are obtained after testing many combinations of promoters and biosynthetic genes by transient expression in *N. benthamiana* leaves. In the end, we confirmed that Z11-16: OH/Ald/OAc is released from the healthy-looking plant. This study represents a significant stride forward for using genetically modified plants as natural dispensers of insect pheromones for integrated pest management.

Keywords: Moth pheromone biosynthesis, Trichome-specific expression, Pheromone release, Biosustainablility

[1] Xia YH, Ding BJ, Wang HL, Hofvander P, Jarl-Sunesson C, Löfstedt C (2020), J Pest Sci 93: 1333-1346
[2] Wang HL, Ding BJ, Dai JQ, Nazarenus TJ, Borges R, Mafra-Neto A, Cahoon EB, Hofvander P, Stymne S, Löfstedt C (2022) Nat Sustain: under revision
[3] Xia YH, Ding BJ, Dong SL, Wang HL, Hofvander P, Löfstedt C (2022) BMC Biol 20: 80

Symposium 2: Application of semiochemicals in insect pest management (2)

Thursday, August 11th
10:45 am - 12:30 pm

Session moderated by Andrew Hayes

Volatile cues of beehives aids *Galleria melonella* (Lepidoptera: Pyralidae) locate its host
Saravan Kumar Parepally, Divija SD, Gandham Krishnarao, Vivek Kempraj, Kamala Jayanthi PD

Developing a new pathway for chemical synthesis of the sex pheromone of the fall web worm, *Hyphantria cunea*, to facilitate the environmentally friendly mating disruption strategy
Yinzhong Cui

Mating disruption by synthetic sex pheromones reduces damage of the beet armyworm (*Spodoptera exigua*) on shallot fields in the Mekong Delta of Vietnam
Le Van Vang, Nguyen Chi Cuong, Duong Kieu Hanh, Trieu Phuong Linh, Huynh Kim Dinh, Kamei Kaeko, Ando Tetsu

Synergistic effect of volatiles from newly wounded leaf sheathes of mangrove palm (*Nypa fruticans*) on the aggregate attraction of the lesser coconut weevil (*Diocalandra frumenti*) inhabiting Mekong Delta of Vietnam
Trieu Phuong Ling, Nguyen Hong Ung, Duong Kieu Hanh, Ando Tetsu

S2-O95

Volatile cues of beehives aids *Galleria melonella* (Lepidoptera: Pyralidae) locate its host

Saravan Kumar Parepally[1,2], Divija S D[3], Gandham Krishnarao[1], Vivek Kempraj[4], Kamala Jayanthi P D[1]

[1]*Indian Institute of Horticultural Research, Bengaluru, India.* [2]*Jain University, Bengaluru, India.* [3]*University of Agricultural Sciences, Bengaluru, India.* [4]*Komohana Research Center, Hawaii, USA*

The greater wax moth, *Galleria mellonella* (Linn.) (Lepidoptera: Pyralidae) is a ubiquitous pest of honeybees and one of the primary reasons to the declining honeybee populations. Although, several management measures have been developed to combat wax moths, the control of wax moths remains a challenge. An alternative management measure is the use of host semiochemicals as attractants, however, the semiochemicals involved in the wax moth's attraction to its host are unknown. In the present study, we aim to understand the host location behavior of *G. melonella* and decipher the attractant cues involved. Headspace volatiles from intact beehives with bees (BB), beehives without bees (BW), and beehives with conspecific damage (BC) were collected and offered to *G. mellonella* in Y-tube attraction assays followed by GC-EAD/MS. The Y-tube results revealed *G. mellonella* preferred BB and BW over BC. GC-EAD/MS results revealed several volatile cues from BB (13 EAD active compounds) and BW (14 EAD active compounds) that were attractive to gravid female *G. mellonella*. The synthetic blends of EAD active cues of BB and BW were significantly attractive to gravid female *G. mellonella* in olfactometer assays. We demonstrate, for the first time, that the volatile cues of beehives may be utilized to develop a kairomone-based lure for trapping female *G. mellonella*.

Keywords: Greater wax moth, honeycombs, GC-EAD/MS, Synthetic blends, Lures

S2-O110

Developing a new pathway for chemical synthesis of the sex pheromone of the fall web worm, *Hyphantria cunea*, to facilitate the environmentally friendly mating disruption strategy.

Yinzhong Cui

Pherobio Technology Co., Ltd, Beijing, China

Invasive forestry insect, the fall web worm *Hyphantria cunea* (Lepidoptera:Erebidae), is a severe defoliator of forest and shade trees and has always been a serious insect pest since it was settled in China. Although, its pheromone [(Z,Z)-9,12-octadecadienal,(Z,Z,Z)-9,12,15-octadecatrienal, and (Z,Z)-3,6-cis-9,10-epoxyheneicosadiene] has already been identified and was widely used for infestation detection and population monitoring, application of mating disruption strategy to control this pest has never been tried in China because the complexity of the pheromone chemical structures with high cost of production. Here we report a cost-effective chemical synthetic pathway for (Z,Z,Z)-9,12,15-octadecatrienal), and all the works trying to develop a single-component mating disruption product to reduce chemical sprays in Urban community.

Keywords: -

S2-O151

Mating disruption by synthetic sex pheromones reduces damage of the beet armyworm (*Spodoptera exigua*) on shallot fields in the Mekong Delta of Vietnam

Le Van Vang[1], Nguyen Chi Cuong[1], Duong Kieu Hanh[1], Trieu Phuong Linh[1], Huynh Kim Dinh[2], Kamei Kaeko[3], Ando Tetsu[4]

[1]College of Agriculture, Can Tho University, Can Tho, Vietnam. [2]Vinh Long Provincial Plant Protection Department, Vinh Long, Vietnam. [3]Kyoto Institute of Technology, Kyoto, Japan. [4]Graduate School of Bio-Applications and Systems Engineering, Tokyo University of Agriculture and Technology, Tokyo, Japan

The beet armyworm, *Spodoptera exigua* (Lepidoptera: Noctuidae) is one of the most destructive insect pests of onion, both shallot and green onion, in the Mekong Delta of Vietnam. Effective control of this species by spray of insecticides is difficult to obtain because of its highly insecticide resistance. In order to utilize sex pheromones as tool for alternation or replacement of insecticides in sustainable management programs for *S. exigua*, the efficacy of mating disruption by synthetic sex pheromones was examined at shallot fields in Soc Trang province. Attraction tests at green onion and shallot fields in Vinh Long and Soc Trang provinces affirmed the high attraction of synthetic pheromones lure (a rubber septum impregnated with 0.8 mg of Z9, E12-14: OAc and 0.2 mg of Z9-14: OH; ISOMATE-BAW, Shin-Etsu Chemical Co.) to *S. exigua* males (averagely 75.5 males/trap/week). At a density of 400 dispensers/ha, polyethylene pheromone dispensers (20-cm-long red polyethylene tubes containing 80 mg of the 8:2 ratio mixture of Z9, E12-14: OAc and Z9-14: OH) disrupted successfully the attraction of lure as well as those of a virgin female traps to *S. exigua* males in field with the relative disruption >95% until 8 weeks. Besides, the bush and leaf damage ratios, the densities of egg masses and density larvae in the mating disruption plot (ha) were significantly lower than those in 5 - 8 insecticide sprays plots. Additionally, the hatching rates of egg masses which were collected from tethered virgin females and on fields in the plot with dispensers were also lower than hatching rates of egg masses in the plots without dispensers. Mating disruption by sex pheromones can alternate or replace the use of insecticides to manage the damage of *S. exigua* on shallot fields in the Mekong Delta.

Keywords: -

S2-O175

Synergistic effect of volatiles from newly wounded leaf sheathes of mangrove palm (*Nypa fruticans*) on the aggregate attraction of the lesser coconut weevil (*Diocalandra frumenti*) inhabiting Mekong Delta of Vietnam

Trieu Phuong Ling[1], Nguyen Hong Ung[2], Duong Kieu Hanh[1], Ando Tetsu[3]

[1]College of Agriculture, Can Tho Univiversity, Can Tho, Vietnam. [2]Department of Agriculture and Aquaculture, Tra Vinh University, Tra Vinh, Vietnam. [3]Graduate School of Bio-Applications and Systems Engineering, Tokyo University of Agriculture and Technology, Tokyo, Japan

The lesser coconut weevil (*Diocalandra frumenti*) was considered as one of the most harmful pests of coconut in the Mekong Delta of Vietnam. In order to utilize semiochemicals as tool for monitoring the adult population, effect of volatiles from newly created wounds on the coconut and mangrove palm leaf sheathes on the attraction to *D. frumenti* adults was investigated. Experiments were at first performed in laboratory (T0: 29±10C; RH: 65±5%) by using an aligned glass olfactometer system, and then examined by net-house and field tests. Odors emitted from the male adults attracted aggregately other conspecific individuals (both sexes). Besides, newly created wounds on the leaf sheathes of coconut and mangrove palm (*Nypa fruticans*) showed high attraction to *D. frumenti* adults (the EPI values were 0.67 and 0.94, respectively). Furthermore, the bait which was made by the combination of newly created wound with alive *D. frumenti* male adults attracted significantly more *D. frumenti* adults than those of the wounds or the alive *D. frumenti* males alone. In field examinations, diurnal *D. frumenti* adults were attracted mainly to the newly created wounds on mangrove palm sheathes from 6:00 am to 18:00 pm. The monthly counting numbers of *D. frumenti* adults which were attracted by newly created wounds on mangrove palm sheathes in mangrove palm fields at three districts in Ben Tre province showed that the beetle adults presented throughout the year with two peaks of densities appeared at the early (May-June) and late (October-November) of the rainy season in the Mekong Delta.

Keywords: -

Symposium 2: Application of semiochemicals in insect pest management (3)

Friday, August 12th
10:45 am - 12:30 pm

Session moderated by Jian Chen

Efficacy of pheromone kairomone blends to trap coconut red palm weevil *Rhynchophorus ferrugineus* (Coleoptera: Dryophthoridae) in India
Vibina Venugopal, Subaharan Kesavan

Two saturated C12 oxo fatty acids from the male alates of red imported fire ants and their behavioral effects
Jian Chen

Long-lasting monitoring system for the cocoa pod borer, development & application
Jerome Niogret, Aijun Zhang

First report on analysis of volatile compounds of *Oecophylla Smaragdina* from India
Selvam Kulanthaivelu, Nalini Thiyagarajan

Identification and field bioassays of the sex pheromone of the winter moth *Erannis ankeraria* Staudinger (Lepidoptera: Geometridae)
Fu Liu

S2-O43

Efficacy of pheromone kairomone blends to trap coconut red palm weevil *Rhynchophorus ferrugineus* (Coleoptera: Dryophthoridae) in India

Vibina Venugopal[1], Subaharan Kesavan[2]

[1]ICAR-Central plantation crop research institute, kasargod, India. [2], ICAR- Nation bureau of agricultural insect resource, Bengaluru, India

Red Palm Weevil (RPW), *Rhynchophorus ferrugineus* is a lethal pest of palms in India and globally. Current control method includes injecting the trunk with carbamate or organophosphates, which has severe side effects on environment and humans, hence we need an alternative eco-friendly method. Synergism of aggregation pheromone (4-methyl-5-nonanol and 4-methyl-5-nonanone 9:1) when used in tandem with food baits has been established in the field as an effective method to trap higher number of weevils. The need for the traps to be serviced with fresh food baits at frequent intervals coupled with replacement of pheromone lures once in three months is a hurdle in its adoption as its laborious and expensive. We attempted to develop a kairomone blend by characterizing the volatiles emanating from food baits (fermenting banana, pineapple, coconut petiole and neera) that elicit antennal and behavioural response in the RPW adults by GC-MS-EAD and windtunnel respectively. The volatiles from food baits viz., ethyl acetate, ethyl propionate, acetoin, ethyl butyrate, isoamyl acetate, phenol and guiacol caused electrophysiological behavioural response in RPW adults. Three blends viz., A, B, C were formulated and used in tandem with RPW aggregation pheromone. The blends were evaluated for its efficacy in field weevil trapping. The results revealed that blend A performed higher than the standard method pheromone trap aided with food baits while other blends B, C performed better than individual compounds which has been in synergizing the pheromone. Our results could be used as a module in integrated pest management as a strong semio-chemical yet ecofriendly tool to mass trap the lethal pest.

Keywords: Red palm weevil, Pheromone, Kairomone, GC-MS-EAD, Multi-component lure

[1] Faleiro JR. (2006) Int J Trop Insect Sci 26:135–154
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S2-O73

Two saturated C12 oxo fatty acids from the male alates of red imported fire ants and their behavioral effects

Jian Chen

USDA-ARS, Stoneville, USA

Red imported fire ant, *Solenopsis invicta*, is an important invasive pest. Bait is one of major tools used in controlling *S. invicta*. Attractants and feeding stimulants can be used to improve bait efficacy and enhance the bait selectivity by facilitating the finding and acceptance of the bait by ants. As a social insect, a greater diversity of pheromones are used by fire ants in coordinating their colonial activities. Some of pheromones function as attractants in a broad spectrum of behavioral contexts. In this study, two oxo fatty acids, 10-oxo-dodecanoic acid and 11-oxo-dodecanoic acid were identified in male alates of the red imported fire ants. Behavioral bioassays using synthetic compounds showed that both oxo fatty acids are attractants to *S. invicta* workers. Fire ant workers also preferentially fed on soybean oil containing 10-oxo-dodecanoic acid or 11-oxo-dodecanoic acid over the untreated vegetable oil, indicating they may be feeding stimulants to fire ants. Since soybean oil is a common food source used in most fire ant bait products, these two compounds can potentially be used to improve the efficacy of existing fire ant bait products and develop new fire ant bait products.

Keywords: Oxo fatty acid, Red imported fire ant, Rale alate, 10-oxo-dodecanoic acid, 11-oxo-dodecanoic acid, Attractant, Feeding stimulant

S2-O90

Long-lasting monitoring system for the cocoa pod borer, development & application

Jerome Niogret[1,2], Aijun Zhang[3]

[1] *Integrated Pest Management, Mars Wrigley, Smithfield, Australia.* [2]*Centre for Tropical Environmental & Sustainability Science, James Cook University, Australia, Smithfield, Australia.* [3]*Invasive Insect Biocontrol and Behavior Laboratory, USDA, ARS, Beltsville, USA*

In Southeast Asia, the Cocoa pod borer *Conopomorpha cramerella* (Snellen) (Lepidoptera: Gracillariidae), is a major economic pest of cocoa. Female sex pheromone and kairomone lures were evaluated for their long-lasting male attraction on Indonesian cocoa plantations. The 1.0 mg pheromone lure did not show any higher attraction than the 0.1 mg pheromone lure during the first month but was significantly attractive for CPB males for a 7-month period in field conditions. The first formulation of our kairomone lure significantly traps more males than the unbaited trap, with similar field longevity to the 1 mg pheromone lure. Once the kairomone formulation was optimized, the lure longevity reached over a year-long. No additive or synergistic effects were observed when the two lures were combined. Such extended longevity offers new mitigation perspectives like mass-trapping and mating disruption technologies. These attractant-based mitigation technologies would benefit from the extensive attractant longevity, which is usually the limiting factor. If coupled with cost-effective synthetic pathways, the steady CPB male capture for up to a year may bring those control methods economically viable for environmentally friendly cocoa production.

Keywords: Cocoa pod borer, Pheromone lures, Kairomone lures, Longevity

S2-O99

First report on analysis of volatile compounds of *Oecophylla Smaragdina* from India

Selvam Kulanthaivelu[1], Nalini Thiyagarajan[2]

[1]*SRS Institute of Agriculture and Technology, Dindigul, India.* [2]*Annamalai University, Chidambaram, India*

Volatile compounds of *Oecophylla smaragdina* were identified by GC-MS. For that colonies of *O. smaragdina* were collected from mango trees. The ants anaesthetized were removed from the dissected nest. Approximately 600-800 worker ants were placed individually on Petri plate. Each worker ant was dissected into head, thorax, and abdomen. Each portion was extracted for 20 minutes at room temperature in ethyl acetate. The extract was placed in screw cap vials and stored at -15°C until analysis (Modified after Igwe and Eze, 2015). The extract was analyzed with Perkin Elmer Clarus SQ8C (PerkinElmer, Waltham, USA) gas chromatograph coupled with an unique Clarifi™ detector. From the head of *O. smaragdina*, 312 compounds were extracted. Eleven classes of chemicals were identified namely hydrocarbon, carboxylic acid, organo phosphorous, ester, alcohol, amine, amide, glucose, amino glycoside, nitrile and aldehyde. Organo phosphorous, amide, amine, glucose, amino glycoside and nitrile were the additional six major functional groups newly found in head which were not reported earlier. This is the first study to report on chemical compounds from the thorax of *O. smaragdina*. From the thorax of *O. smaragdina*, 211 compounds were extracted. Seven classes of chemicals were identified namely carboxylic acid, amine, hydrocarbon, ester, alcohol, nitrile and ketone. From the abdomen of *O. smaragdina*, 361 compounds were extracted. Nineteen classes of chemicals were identified namely hydrocarbon, sugar acids, ester, hexadioic acid, dioctyl ester, sulfonyl fluoride, benzoic acid, ester, alcohol, phthalate ester, diaryl methane, benzothiophene, corticosteroid, glucosinolates, alpha acid, phyto sterol, isoxazolinones, vitamin, beta bitter acid, peptide and unidentified compounds.

Keywords: Volatile compounds, *Oecophylla smaragdina*, Head, Thorax, Abdomen

[1] Igwe OU, Eze PN (2015). Chemical analyses of volatile compounds from cuticular and non-cuticular abdominal glands of African weaver ants (*Oecophylla longinoda*). International Journal of Chemical Sciences 5: 304-312.

S2-O194

Identification and field bioassays of the sex pheromone of the winter moth *Erannis ankeraria* Staudinger (Lepidoptera: Geometridae)

Fu Liu

Ecology and Nature Conservation Institute,Chinese Academy of Forestry, Beijing, China

The winter-flying moth *Erannis ankeraria* (Staudinger) (Lepidoptera: Geometridae), a rare endangered geometrid moth in Europe, appeared at damaging levels in the Saihanba and inner Mongolia region of China. However, sex pheromones generated by *E. ankeraria* females have not been identified. The present study detected four male-antennally active components, namely (3*Z*,6*Z*,9*Z*)-3,6,9-nonadecatriene (*Z*3*Z*6*Z*9-19: Hy), (3*Z*,9*Z*)-3,9-*cis*-6,7-epoxy-nonadecadiene (*Z*3*Z*9-6,7-*epo*-19:Hy), (6*Z*,9*Z*)-6,9-*cis*-3,4-epoxy-nonadecadiene (*Z*6*Z*9-3,4-*epo*-19:Hy), and (3*Z*,6*Z*)-3,6-*cis*-9,10-epoxy-nonadecadiene (*Z*3*Z*6-9,10-*epo*-19: Hy), in female pheromone gland extracts by using gas chromatography coupled with mass spectrometry and gas chromatography coupled with electroantennographic detection (EAD). *Z*3*Z*6*Z*9-19: Hy elicited the strongest EAD response, whereas *Z*6*Z*9-3,4-*epo*-19: Hy and *Z*3*Z*6-9,10-*epo*-19: Hy exhibited similar EAD responses, *Z*3*Z*9-6,7-*epo*-19: Hy induced a weak EAD response. Field trapping confirmed that a quaternary mixture of nonadecatriene and three epoxynonadecadienes in a ratio of 1:0.5:1.2:1.3, (approximate ratio of females emitting pheromone), was optimal for catching *E. ankeraria* males. *Z*3*Z*6*Z*9-19: Hy exhibited a synergistic effect on the attraction of three epoxynonadecadienes to males. The 9,10-epoxide component was initially reported as an attractant component in the *Erannis* (*Agriopis*) species. Finally, *Z*3*Z*6*Z*9-19: Hy, *Z*3*Z*9-6,7-*epo*-19: Hy, *Z*6*Z*9-3,4-*epo*-19: Hy, and *Z*3*Z*6-9,10-*epo*-19: Hy are confirmed as vital sex pheromone components of *E. ankeraria* and offer a beneficial foundation for optimising a practically applicable lure for monitoring this pest.

Keywords: *Erannis ankeraria*, Sex pheromone, Synergistic influence, Field bioassay.

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Symposium 3: Plant metabolomics and chemical defenses (1)

Wednesday, August 10th
03.20 pm - 05:00 pm

Session moderated by Po-An Lin

Measuring chemodiversity considering biochemical and structural properties of compounds with the R package *chemodiv*
Hampus Petrén, Tobias G. Köllner, Robert R. Junker

Terpene chemotypes in wild cotton (*Gossypium hirsutum*) from the Yucatan peninsula
Mary V Clancy, Marine Mamin, Galien Flückiger, Teresa Quijano-Medina, Biiniza Pérez Niño, Luis Abdala-Roberts, Ted CJ Turlings, Carlos Bustos-Segura

Plant-plant signalling affects the performance of *Myzus persicae* aphids on potato
Jamin Ali, Toby Bruce

Variation in airborne communication between plants among wild cotton genotypes
Carlos Bustos-Segura, Mary V Clancy, Marine Mamin, Wenfeng Ye, Luis Abdala-Roberts, Ted CJ Turlings

The Chemical Ecology of plant-root knot nematode interactions
Baldwyn Torto, Ruth Kihika-Opanda, David Tchouassi, John Beck

S3-O20

Measuring chemodiversity considering biochemical and structural properties of compounds with the R package *chemodiv*

Hampus Petrén[1], Tobias G. Köllner[2], Robert R. Junker[1,3]

[1]*Philipps-University Marburg, Marburg, Germany.* [2]*Max Planck Institute for Chemical Ecology, Jena, Germany.* [3]*University of Salzburg, Salzburg, Austria*

Plants produce large numbers of phytochemical compounds, which influence antagonistic and mutualistic interactions between plants and their biotic and abiotic environment. Mixtures of phytochemicals form a complex phenotype, and relatively little is known about what dimensions of this phenotype are most important for its function, such as herbivore protection or pollinator attraction. Recently, chemodiversity has attracted considerable attention as an ecologically and evolutionary meaningful way to quantify mixtures of phytochemical compounds. However, currently used measures of phytochemical diversity, and related measures of phytochemical dissimilarity, generally only consider the number and relative abundances of compounds, and do not take their structural or biosynthetic properties into account. Such properties may contribute to shaping the function of the phytochemicals and should therefore be included in diversity calculations. We have developed the R package *chemodiv*, which provides functions for calculating and visualizing chemical diversity and dissimilarity for phytochemicals and other types of compounds. Our package enables calculations of diversity that take the richness, relative abundance and – most importantly – structural and/or biosynthetic dissimilarity of compounds into account. In this way, phytochemical diversity is quantified in a more comprehensive way than has been done previously, analogous to measures of functional diversity. We demonstrate how such measures provide a more complete picture of the diversity of phytochemicals found in plants. By providing an R package as a tool for comprehensive measures of chemodiversity, we hope to facilitate investigations on how chemodiversity varies across levels of biological organization, and its importance for interactions between plants and other organisms.

Keywords: Chemodiversity, Phytochemicals, Plant defence, R package

S3-O25

Terpene chemotypes in wild cotton (*Gossypium hirsutum*) from the Yucatan peninsula

Mary V Clancy[1], Marine Mamin[1], Galien Flückiger[1], Teresa Quijano-Medina[2], Biiniza Pérez Niño[2], Luis Abdala-Roberts[2], Ted CJ Turlings[1], Carlos Bustos-Segura[1]

[1]*University of Neuchâtel, Neuchâtel, Switzerland.* [2]*Universidad Autónoma de Yucatán, Merida, Mexico*

Cultivated cotton plants (*Gossypium hirsutum*) consistently emit low levels of volatile organic compounds, primarily mono- and sesquiterpenoids, which are produced and stored in the pigment glands characteristic of cotton. In this study, we evaluated the terpene profiles of wild cotton plants to explore the scope of intraspecific chemotypic diversity in a crop wild relative of farmed cotton. Under controlled environmental conditions, plants were grown from sixteen *G. hirsutum* populations collected along the coastline of the Yucatan peninsula, which is its likely centre of origin. We found high levels of intraspecific diversity in the terpene profiles of the plants. Two chemotypes were identified; one chemotype contained higher levels of the monoterpenes γ-terpinene, limonene, α-thujene, α-terpinene, terpinolene, and p-cymene, while the other chemotype was distinguished by higher levels of α- and β-pinene. The distribution of chemotypes followed a geographic gradient. Following concurrent analysis of maternal plants, we also present data showing that chemotypes in wild cotton are highly heritable.

Keywords: Chemotypes, Terpenes, Chemical ecology, *Gossypium hirsutum*

S3-O31

Plant-plant signalling affects the performance of *Myzus persicae* aphids on potato

Jamin Ali, Toby Bruce

Keele University, Newcastle Under Lyme, United Kingdom

Herbivore-infested plants release a different blend of volatiles compared to undamaged plants or artificially damaged plants. These herbivores induced plant volatiles can induce defense in neighboring plants. The current study was designed to test the effect of neighboring herbivore infested plants on subsequent interactions of receiver plants with aphid *Myzus persicae* and parasitoid *Diaeretiella rapae*. Receiver plants were exposed to VOCs from neighboring emitter plants. Four treatments of cultivated potato, *Solanum tuberosum* cv. 'Desiree', and a wild accession of *Solanum stoloniferum* were made to investigate the effect of neighboring plants on receiver plants. The treatments were (1) Desiree exposed to VOCs from uninfested Desiree (DD); (2) Desiree exposed to VOCs from aphid infested Desiree (DID); (3) Desiree exposed to VOCs from wild potato (DW); (4) Desiree exposed to VOCs from aphid infested wild potato (DIW). Performance and behavioral bioassays were performed to investigate responses of insects to receiver plants. A clip-cage aphid performance bioassay showed a significant reduction in the number of surviving aphids and larviposited nymphs on treatments where receiver plants were exposed to VOCs from infested plants. In olfactometer bioassays, *M. persicae* aphids spent a significantly longer in the olfactometer zone treated with volatiles collected from DD and a significantly shorter time in the olfactometer zone treated with volatile samples collected from DW treatment. *D. rapae* parasitoids spent a significantly longer time in olfactometer zones treated with volatile samples collected from DID and DIW treatments. The current study showed that volatiles released by plants exposed to treatments containing infested Desiree and wild plant volatiles enhance the defense responses in the receiver plants. Identification of volatile compounds responsible for this induced change could be used to manipulate the plant defense system against pests.

Keywords: Plant-plant communication, Induced defence, Solanum tuberosum Desiree, *Myzus persicae*

S3-O38

Variation in airborne communication between plants among wild cotton genotypes

Carlos Bustos-Segura[1], Mary V Clancy[1], Marine Mamin[1], Wenfeng Ye[1], Luis Abdala-Roberts[2], Ted CJ Turlings[1]

[1]*University of Neuchâtel, Neuchâtel, Switzerland.* [2]*Universidad Autónoma de Yucatán, Merida, Mexico*

Cotton plants (*Gossypium hirsutum*) defend themselves against insect herbivores via secondary metabolites such as gossypol, and the release of herbivore induced plant volatiles (HIPVs) that attract natural enemies. In addition, HIPVs can induce resistance in neighboring cotton plants, a phenomenon that could potentially be used in pest management. However, plant-plant communication could be weakened in present day cotton cultivars that were mainly selected for yield traits. We aimed to look at the variation in communication between plants using wild genotypes of cotton which have been under constant natural selective pressure, including insect herbivory. To this end, we evaluated gene expression, phytohormone levels and herbivore leaf consumption in receiver plant genotypes exposed to HIPVs in lab conditions. We compared responses of different plant genotypes using a crossed design among four wild cotton populations from the Yucatan peninsula in Mexico. We discovered considerable variation in plant responses among wild genotypes associated with both emitter and receiver genotypes. The gene expression induced by exposure to HIPVs was influenced by the identity of the receiver; one receiver genotype from a western population was the most strongly induced. This induction did not depend on the emitter genotype. However, the identity of emitter genotype was associated with differences in herbivore consumption on receivers, suggesting that HIPVs could play a direct role in protection of neighboring plants. Meanwhile, we did not find evidence that variation in the terpene profile in both receivers and emitters impacted the effectiveness of communication between plants. Overall, these results demonstrate that variation in plant communication in wild cotton is present at both receivers and emitters and that such variation can be associated to genetic factors. In summary, we obtained evidence for intraspecific variation in responses to odor or odor reception in cotton plants.

Keywords: Gene expression, HIPVs, Induced defenses, Cotton, Chemotypes

S3-O102

The chemical ecology of plant-root knot nematode interactions

Baldwyn Torto[1], Ruth Kihika-Opanda[1], David Tchouassi[1], John Beck[2]

[1]*ICIPE, Nairobi, Kenya.* [2]*USDA/ARS, Gainesville-Florida, USA*

Phytochemicals play an important role in intra- and inter-specific interactions in the rhizosphere, by influencing growth of neighboring plants via kin recognition or allelopathy, microbial richness, and survival of pests including plant parasitic nematodes (PPNs) such as root knot nematodes (RKNs). Root-knot nematodes (*Meloidogyne* species) are sedentary plant endoparasites that suppress plant growth by forming giant cells which inhibit plant water and nutrient uptake. They infect the roots of a host plant to feed and reproduce. Globally, they cause crop production losses of over US $100 billion each year. Economic losses in Africa due to root knot nematode damage are scarce, but typically, in small holder farms, crop loses may range between 40−100%. RKNs develop through six stages; an egg, the first juvenile stage (J1) which develops inside the egg, a second juvenile stage (J2), which emerges from the egg and must find host root to establish a feeding site. Once established at a feeding site they molt into two additional juvenile stages (J3 and J4), then become an adult. RKNs are difficult to control, but synthetic nematicides have been found to be effective against them. However, because of their effects on the environment and non-target organisms, synthetic nematicides are not recommended. They are also expensive for smallholder farmers, who rely mainly on cultural approaches (crop rotation with non-host crops) and use of botanicals. RKN infestation may persist in the soil despite these management approaches. Recent studies in our laboratory have shown that semiochemicals mediate the interaction between crops and root knot nematodes. Examples of how these semiochemicals can be used in the management of RKNs will be emphasized in this presentation.

Keywords: Plant parasitic nematode, Meloidogyne, Rhizosphere, Semiochemicals

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Symposium 3: Plant metabolomics and chemical defenses (2)

Thursday, August 11th
09.45 am - 10:25 am

Session moderated by Po-An Lin

β-Tyrosine accumulates in developing rice leaf via jasmonic acid-induced long-distance transport
Shunta Sakamoto, Takanori Yoshikawa, Masayoshi Teraishi, Naoko Yoshinaga, Naoki Mori

Stemborer-induced rice plant volatiles boost direct and indirect resistance in neighboring plants
Chengcheng Yao, Yunhe Li

S3-O119

β-Tyrosine accumulates in developing rice leaf via jasmonic acid-induced long-distance transport

Shunta Sakamoto[1], Takanori Yoshikawa[2], Masayoshi Teraishi[2], Naoko Yoshinaga[1], Naoki Mori[1]

[1]*Division of Applied Life Sciences, Graduate School of Agriculture, Kyoto University, Sakyo, Kyoto, Japan.* [2]*Division of Agronomy and Horticultural Sciences, Graduate School of Agriculture, Kyoto University, Sakyo, Kyoto, Japan*

We previously identified (3*R*)-β-tyrosine, a nonproteinogenic β-amino acid detected only in *japonica* rice cultivars (*Oryza sativa* L. ssp. *japonica*) in plant kingdom. The β-tyrosine content in the *japonica* cultivar Nipponbare is elevated by jasmonic acid (JA) treatment, thus β-tyrosine has likely defensive function as well as other defensive nonproteinogenic amino acids, such as L-canavanine and *m*-tyrosine. However, physiological and ecological functions of β-tyrosine remain unclear. Since β-tyrosine is overrepresented in the *japonica* cultivars compared to other cultivars, the elucidation of its functions will probably lead to understanding of intraspecific diversity of environmental adaptation in *Oryza sativa*. Therefore, the purpose of this study is to clear the β-tyrosine dynamics in *japonica* cultivars under stress conditions as the first step to understand the functions of β-tyrosine. LC/MS analysis of JA-treated Nipponbare leaves revealed that β-tyrosine accumulated in developing leaf. However, *in vitro* activity measurement of OsTAM1, enzyme for β-tyrosine biosynthesis, showed that developing leaf was scarcely able to produce β-tyrosine by itself. Therefore, we hypothesized that long-distance transport of β-tyrosine is induced by JA. Stable isotope study demonstrated that the efficiency of β-tyrosine transport into developing leaf was increased under JA treatment. Furthermore, time-course experiments showed that β-tyrosine accumulation in distal mature tissues preceded its accumulation in developing leaf, indicating that JA-induced β-tyrosine biosynthesis preceded JA-induced β-tyrosine transport. Our current study revealed predictable functional location of β-tyrosine in Nipponbare under stress conditions and underlying mechanism of its accumulation there.

Keywords: β-tyrosine, Jasmonic acid, Long-distance transport, *Japonica* rice

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S3-O120

Stemborer-induced rice plant volatiles boost direct and indirect resistance in neighboring plants

Chengcheng Yao, Yunhe Li

State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China

Herbivore-induced plant volatiles (HIPVs) are known to be perceived by neighboring plants, resulting in induction or priming of chemical defenses. There is little information on the mechanisms that are involved in these plant-plant interactions and the phenomenon has scarcely been studied in rice. Using chemical and molecular analyses in combination with insect behavioral and performance experiments we studied how volatiles emitted by rice plants infested by the striped stem borer (SSB) affect defenses against this pest in conspecific plants. Compared to rice plants exposed to the volatiles from uninfested plants, plants exposed to SSB-induced volatiles showed enhanced direct and indirect resistance to SSB. When subjected to caterpillar damage, the HIPV-exposed plants showed increased expression of jasmonic acid (JA) signaling genes, resulting in JA accumulation and higher levels of defensive proteinase inhibitors. Moreover, plants exposed to SSB-induced volatiles emitted larger amounts of volatile compounds and were more attractive to the parasitoid *Cotesia chilonis*. This novel insight into the mechanisms involved in HIPV-mediated defense priming in rice plants, reveals a key defensive role for proteinase inhibitors. It paves the way for novel rice management strategies to enhance the plant's resistance to one of its most devastating pests.

Keywords: Herbivore-induced plant volatiles (HIPVs), Jasmonate (JA), Proteinase inhibitor (PI), Defense priming, *Chilo suppressalis*

[1] Ye M, Glauser G, Lou YG, Erb M, Hu LF (2019) Plant Cell 31: 687-698
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Symposium 3: Plant metabolomics and chemical defenses (3)

Friday, August 12th
12.25pm - 12:45 pm

Session moderated by Po-An Lin

Floral notes from underground: the scent of the underground orchid (*Rhizanthella speciosa)*
James Perkins, Rod Peakall

S3-O86

Floral notes from underground: the scent of the underground orchid (*Rhizanthella speciosa)*

James Perkins, Rod Peakall

ANU, Canberra, Australia

The Orchidaceae is well known for its exceptional diversity of floral forms and pollination strategies. Amongst this diversity are many examples of remarkable and strange flowers, but there are perhaps none so enigmatic as the Australian underground orchids (*Rhizanthella* spp.). These orchids are leafless and achlorophyllous, and even their flowers remain at or just beneath ground level, buried in leaf litter. Because of this unusual habit, very little is known about the abundance, distribution, pollination biology, and ecological interactions of these rare orchids. Given the hidden nature of their flowers, we predicted that these orchids rely heavily on scent rather than visual cues to attract pollinators from beneath the leaf litter. We tested this hypothesis in *Rhizanthella speciosa*, a recently described species noted for its unusual scent to humans. Our volatile analysis commenced in the field with headspace trapping and solvent extractions of floral tissue. Subsequent GC-MS analysis detected two compounds in floral headspace samples, both 5-pentylresorcinol (olivetol) derivatives. These olivetol derivatives were also found in solvent extracts of floral tissue, confirming they originate from the orchid. The identification of olivetol derivatives was confirmed by co-injection of synthetic compounds. Several other compounds were identified from solvent extracts, including common floral benzenoids and p-cresol, which may contribute to the flower's unique bouquet. Sniffer dogs are being trained on the scent of the orchids, in the hope that they can be used to find other populations of the orchids. Intriguingly, in one preliminary test, one olivetol derivative caused the dog to indicate the presence of an orchid, suggesting the compound may be a promising replacement for orchid tissue used to train detection dogs. Extensive field tests with synthetic blends of the olivetol derivatives and other compounds in the solvent extracts are planned for the forthcoming field season.

Keywords: Orchid, Flower, Volatile, *Rhizanthella*, Pollination

Symposium 4: Pheromones and chemoperception (1)

Wednesday, August 10th
5:10 pm - 6 pm

Session moderated by Björn Bohman

Investigating pheromone biosynthesis de novo from Eurasian spruce bark beetle – *Ips typographus*
Rajarajan Ramakrishnan, Amit Roy, Jaromír Hradecký, Marco Kai, Ales Svatos, Anna Jirošová

Characterization of the sensory gene repertoire of *Triatoma infestans* and the effect of blood-feeding on antennal expression profiles
Jose Manuel Latorre Estivalis, Lucila Traverso, Marcelo Gustavo Lorenzo

Unsaturated hydrocarbons-structural elucidation and function as fungus gnat pheromones and orchid pollinator attractants
Björn Bohman, Tobias Hayashi, Gavin Flematti, Rod Peakall

S4-O60

Investigating pheromone biosynthesis de novo from Eurasian spruce bark beetle – *Ips typographus*

Rajarajan Ramakrishnan[1], Amit Roy[1], Jaromír Hradecký[1], Marco Kai[2], Ales Svatos[2], Anna Jirošová[1]

[1]*Czech University of Life Sciences, Prague, Czech Republic.* [2]*Max Planck Institute of Chemical Ecology, Jena, Germany*

The Eurasian spruce bark beetle *Ips typographus* is known for its devasting attack on host tree *Picea abies*, a common conifer in Europe. The beetle uses various pheromone components (2-methyl-3-buten-2-ol and cis-verbenol) for mass aggregation to overcome the tree defense mechanism. Though the role of pheromones in the aggregation behaviour of the beetle is known for a few decades now, gene-level understanding behind the biosynthesis of these compounds is still unknown. Our recent findings have paved the way for understanding the *de novo* aggregation pheromone biosynthesis and its possible candidate gene families from the gut tissue from various life stages of *I. typographus* . Interestingly, the approach of applying Juvenile hormone as a treatment on the beetles has also helped in inducing the specific pheromone biosynthesis irrespective of the beetle's life stage. Thus, in this study, we have performed the method of JHIII application over the beetle and analyzed the pheromone biosynthesis and its respective candidate gene expression behaviour from the gut tissue of the beetle. Our extensive analysis revealed *isoprenyl di phosphate synthase* (IPDS) as a potential candidate gene for functional characterization. The identified IPDS is suspected to be directly involved in 2-methyl-3-buten-2-ol, a vital aggregation pheromone compound of *I. typographus*. We are using classical gene characterization methods with bacterial cell line (*E. coli* expression system) and RNA- interference for the IPDS gene. This approach would lead us to elucidate the molecular basis of pheromone biosynthesis and the derived knowledge from this study would lead to eco-friendly pest management for this aggressive pest.

Keywords: *Ips typographus*, Bark beetle, Pheromone biosynthesis, *de novo*, Gene characterization

[1] Ramakrishnan, R., Hradecký, J., Roy, A., Kalinová, B., Mendezes C. R., Synek, J., Bláha, J., Svatoš, A., Jirošová, A. (2022). Insect Biochem. Mol. Biol. 0965-1748. https://doi.org/10.1016/j.ibmb.2021.103680.

S4-O105

Characterization of the sensory gene repertoire of *Triatoma infestans* and the effect of blood-feeding on antennal expression profiles

Jose Manuel Latorre Estivalis[1], Lucila Traverso[2], Marcelo Gustavo Lorenzo[3]

[1]IFIBYNE, UBA, Buenos Aires, Argentina. [2]CREG-CONICET, La Plata, Argentina. [3]IRR-FIOCRUZ, Belo Horizonte, Brazil

Triatoma infestans is the main vector of *Trypanosoma cruzi*, the etiological agent of Chagas disease. Control programs lost efficiency because bugs became resistant to pyrethroids. Behavioral manipulation represents an alternative for developing sustainable control methods. Behavioral state-dependency allows insects to adjust their responsiveness to sensory cues depending on development or nutritional status. Here, we sequenced a transcriptome in order to characterize the set of genes expressed in the antennae of *T. infestans*. Subsequently, we evaluated the impact of blood-feeding by comparing gene expression profiles in the antennae of unfed and fed insects. Antennae of 5th instar larvae (20 per sample; 6 replicates *per* condition) were collected from 34-day-old larvae (antennae of fed bugs excised 4 days after engorging). Illumina NovaSeq6000 sequencing using 150 bp paired-end reads, and 20 million reads *per* sample were used. A de novo assembly was performed using Trinity. DEseq2 was used to detect differentially expressed transcripts. Chemosensory receptor candidates (as well as other relevant sensory-related gene families) were identified using sequences of *R. prolixus* and other hemipterans, and pfam domain seeds as queries in BLASTp searches. Sensory genes found were: 127 ORs, 38 IRs, 11 GRs, 43 OBP, 20 CSP, 17 TRP, 13 PPKs, three SNMP, and three ammonium transporters. Six OR expansions and an expanded number of IR75 paralogues were identified in *T. infestans* and *R. prolixus* when compared to other hemipteran species. Blood ingestion induced a great modification of the expression profile of *T. infestans* antennae, with 2,122 transcripts significantly changing their abundance (s-values <0.05 and absolute fold-change threshold >1.5). This set of genes showing significantly altered expression after feeding includes representatives of diverse sensory gene families. The results will be discussed from the perspective of behavioral state-dependency induced by blood-feeding.

Keywords: Transcriptome, Antennae, Olfaction, Odorant receptors, Modulation

S4-O34

Unsaturated hydrocarbons-structural elucidation and function as fungus gnat pheromones and orchid pollinator attractants

Björn Bohman[1, 2, 3], Tobias Hayashi[3, 2], Gavin Flematti[2], Rod Peakall[3, 2]

[1]*The Swedish University of Agricultural Sciences, Lomma, Sweden.* [2]*The University of Western Australia, Crawley, WA, Australia.* [3]*The Australian National University, Canberra, ACT, Australia*

Fungus gnats (Diptera) are believed to pollinate hundreds of sexually deceptive orchids, yet unlike orchids pollinated by bees and wasps, the chemistry of fungus gnat-pollinated orchids remained unknown until recently. Furthermore, despite the importance of fungus gnats as pollinators and pests, and evidence for sex pheromones for over 50 years, no structure of any fungus gnat sex pheromone had been confirmed previously. Here, we present the identification of a mixture of five hydrocarbons shared between the orchid *Pterostylis orbiculata* and females of the male pollinator, a *Mycomya* sp. (Mycetophilidae) fungus gnat. The hydrocarbon mixture included three alkanes, a C23 diene, and a novel C23 triene. The triene was synthesized and confirmed as (6Z,9Z)-1,6,9-tricosatriene. In field bioassays, a blend of the five hydrocarbons elicited strong attraction and sexual behavior from male gnats. Moreover, the triene alone elicited attraction and low levels of sexual behavior, while the blend without it was unattractive, suggesting that this compound is a key component of orchid pollinator attraction and a component of the female fungus gnat sex pheromone. The isolation by preparative GC from a natural floral extract, structural elucidation, and the confirmation of double bond positions of this novel triene will be described.

Keywords: Fungus gnat, Orchid, *Pterostylis*, Pollination, Unsaturated hydrocarbons

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Symposium 4: Pheromones and chemoperception (2)

Thursday, August 11th
8:45 am - 12:30 pm

Session moderated by Yang Liu & Wei Xu

Male wing substances acting as sex pheromones in the yellow butterfly *Eurema mandarina*
Kento Yoshimori, Chika Okuda, Shinji Ohta, Hisashi Ômura

Identification and homologous analysis of odorant receptor genes in three Notodontidae species
Tianzi Gu, Jingkang Song, Chang He, Longwa Zhang

Two odorant receptors tuned to a host plant volatile regulate oviposition behavior of the pregnant flies in *Bactrocera dorsalis*
Li Xu

Anatomy and neuronal element identification of gnathal ganglion in larval *Helicoverpa armigera* (Lepidoptera: Noctuidae)
Long-Long Sun, Xiao-Lan Liu, Ya-Nan Wang, Bente Gunnveig Berg, Gui-Ying Xie, Wen-Bo Chen, Yang Liu, Gui-Rong Wang, Xin-Cheng Zhao, Qing-Bo Tang

Correspondence between input and output information in the macroglomerular complex in male *Helicoverpa armigera* (Hübner)
Xiao-Lan Liu, Xi Chu, Long-Long Sun, Ya-Nan Wang, Gui-Ying Xie, Wen-Bo Chen, Yang Liu, Bente Gunnveig Berg, Shi-Heng An, Gui-Rong Wang, Xin-Ming Yin, Xin-Cheng Zhao

Morphological identification of the neurons control Aphid cornicle muscle
Baiwei Ma, Guirong Wang, Bingzhong Ren

Sex differential expression analysis on antennae transcriptome of *Hyphantria cunea*
Yaning Li, Shanchun Yan

A female specific odorant receptor tuning to egg-surface odorants mediates the oviposition aversion behavior of *Helicoverpa armigera*
Xiaxuan Zhang, Mengbo Guo, Dongdong Sun, Yang Liu, Guirong Wang

Molecular regulatory mechanisms of contact sex pheromone biosynthesis in the German cockroach, *Blattella germanica*
Yongliang Fan, Sheng Li, Tong-Xian Liu

S4-O15

Male wing substances acting as sex pheromones in the yellow butterfly *Eurema mandarina*

Kento Yoshimori[1], Chika Okuda[2], Shinji Ohta[1,2,3], Hisashi Ômura[1,2,3]

[1]*Graduate School of Biosphere Science, Higashihiroshima, Japan.* [2]*School of Applied Biological Science, Hiroshima University, Higashihiroshima, Japan.* [3]*Graduate School of Integrated Sciences for Life, Hiroshima University, Higashihiroshima, Japan*

The common grass yellow *Eurema mandarina* has a characteristic patch (sex brand) on the ventral surface of male forewing. This structure is male-specific and is thought to release volatiles that induce female abdominal bending as a characteristic behavior to accept mating. However, no study has demonstrated male wing substances function as its sex pheromones. Here we report the identification of male sex pheromones of *E. mandarina*. Chemical analyses using GC-MS revealed that 6,10,14-trimethylpentadecan-2-one (TMP) and (*E*/*Z*)-3,7,11,15-tetramethylhexadec-2-enal [(*E*/*Z*)-phytal] were abundant in certain regions of the male forewings of *E. mandarina*. TMP was most abundant in the sex brand, whereas (*E*/*Z*)-phytal was localized in the anal cell (cell 2A). The amounts of TMP and (*E*/*Z*)-phytal were low or absent in males immediately after emergence but increased as males aged. In bioassays, virgin females displayed a posture of abdomen-bending in response to stimulation by fresh male forewings. However, solvent-washed male wings did not induce such female responses, suggesting that some compounds from the male wings serve as triggers. When we applied authentic compounds to solvent-washed male wings and examined female responses to them, TMP and (*E*/*Z*)-phytal alone showed little activity, but the mixture elicited abdomen-bending responses from one-third of the females. Therefore, TMP and (*E*/*Z*)-phytal were found to act synergistically as aphrodisiac pheromones in *E. mandarina* females, although these activities were weak. Microscopic examinations revealed that the male sex brand had morphologically distinctive structures: specialized wing scales (androconia) and wing intermembranous cells. These structures might be involved in the emanation of TMP. On the other hand, the anal cell lacked such structures, suggesting that (*E*/*Z*)-phytal might be transferred from the hindwing anterior regions, which contained the second highest amount of it, by rubbing the wings.

Keywords: Butterfly, Sex Pheromones, Mating, GC-MS, Volatiles

S4-O56

Identification and homologous analysis of odorant receptor genes in three Notodontidae species

Tianzi Gu, Jingkang Song, Chang He, Longwa Zhang

Anhui Agricultural University, Hefei, China

Clostera restitura, *Clostera anachoreta* and *Micromelalopha sieversi* are oligophagous defoliators and causing severe damage to poplar plantation in China. Three pests belong to Notodontidae and share similar morphology and biology characteristics, they usually occurred together but no hybrids were observed. As we all know, moths can locate mates accurately due to sex pheromone specific recognition. To explore how the three closely-related species distinguish their own mates, firstly, we identified odorant receptor (ORs) genes of the three species based on RNAseq. 64, 41 and 22 OR genes with relatively complete ORF were found in *C. restitura*, *C. anachoreta* and *M. sieversi* respectively. Sequence alignment and phylogenetic analysis were conducted to identify candidate homologous PR genes in three related species. As a result, CresOR55, MsieOR18/20/25 and CanaOR7 gathered into Classical PR family, among which CresOR55 and MsieOR18 formed a branch with 1000 bootstrap value. CresOR33/45/56, MsieOR22/30, CanaOR6/9/10 gathered with Novel PRs identified in previous study. CresOR56、CanaOR10 and MsieOR22, CresOR33 and CanaOR6, MsieOR30 and CanaOR9 gathered in distinct branches with high bootstrap values, we can infer that these are homologous PR genes among three species, which might play important roles in intro- or inter-specific chemosensory process. Gene expression analysis based on transcriptome sequencing showed that CresOR55/56, MsieOR22 and CanaOR7/10 which were enriched in male antennae may participate in male mate-searching. CresOR33/45, MsieOR18 and CanaOR6/9 might be related to host selection during female oviposition since they had higher expression level in female antennae. In conclusion, several candidate homologous PRs were identified from three closely-related species in Notodontidae, offering the foundation for further study on intro- and inter-specific pheromone communication.

Keywords: Notodontidae, Homologous pheromone receptor, Phylogenetic analysis, Gene expression analysis

[1] Yang, K., Wang, C. Z. Review of pheromone receptors in heliothine species: expression, function, and evolution[J]. Entomol Exp Appl, 2020, 169(2): 156-171.

S4-O62

Two odorant receptors tuned to a host plant volatile regulate oviposition behavior of the pregnant flies in *Bactrocera dorsalis*

Li Xu

Southwest University, Chong Qing, China

The Oriental fruit fly, *Bactrocera dorsalis* (Hendel), is one of the most destructive and invasive pests, causing huge economic losses to the world fruit and vegetables. Currently, the most economic and effective way to control *B. dorsalis* is olfaction-based trapping. The main component of the widely used commercial attractant is methyl eugenol (ME) which shows strong attraction to the male flies. However, oviposition inside of the fruit is the major reason for fruit damage. Therefore, it would be ideal to develop novel female attractant based control method. 1-octen-3-ol have been shown to have the potential to attract females of this fly, especially the gravid females. However, the molecular basis for the perception of 1-octen-3-ol in this fly remains unknown. We identified odorant receptors (ORs) that respond to 1-octen-3-ol using two heterologous expression system comprising *Xenopus* oocytes and HEK293 cells. The screened ORs were then knocked out using CRISPR/Cas9 system, and the oviposition behavior of the mutant flies were observed. We annotated 73 ORs in the genome wide of *B. dorsalis*. Among them, OR7a-6 and OR13a were screened out for 1-octen-3-ol perception based on voltage clamp and calcium imaging assays. After knocked out these two ORs, the EAG response of mutants to 1-octen-3-ol were reduced, and the oviposition of the *OR7a-6*$^{-/-}$ and *OR13a*$^{-/-}$ mutants significantly decreased under 1-octen-3-ol exposure. OR7a-6 and OR13a are the key ORs for 1-octen-3-ol perception in *B. dorsalis*, and they could regulate the oviposition behavior of *B. dorsalis* under the 1-octen-3-ol exposure.

Keywords: *Bactrocera dorsalis*, 1-octen-3-ol, Odorant receptor, Oviposition

S4-O112

Anatomy and neuronal element identification of gnathal ganglion in larval *Helicoverpa armigera* (Lepidoptera: Noctuidae)

Long-Long Sun, Xiao-Lan Liu, Ya-Nan Wang, Bente Gunnveig Berg, Gui-Ying Xie, Wen-Bo Chen[1], Yang Liu[3], Gui-Rong Wang[3], Xin-Cheng Zhao[1], Qing-Bo Tang[1]

[1]Henan International Joint Laboratory of Green Pest Control, College of Plant Protection, Henan Agricultural University, Zhengzhou, China. [2]Chemosensory laboratory, Department of Psychology, Norwegian University of Science and Technology, Trondheim, Norway. [3]State Key Laboratory for Biology of Plant Disease and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China

The gustatory reception plays an essential role in identifying nutrients and avoiding toxic substances. The peripheral coding of how to detect host plants has been studied in many insect species, but the morphology and physiology of gustatory neuron of the central taste center, gnathal ganglion (GNG), is not yet clear. In the present research, we investigated the architecture of GNG first by immunostaining and then characterized the morphology and physiological functions of its gustatory neuron in the cotton bollworm *Helicoverpa armigera* (Lepidoptera: Noctuidae) larvae by utilizing the intracellular recording/staining technique. The results show that the GNG could be divided into 3 areas, 6 sub-areas and 32 small areas based on the location of nerves, commissures and bundles. There were 133 stained neurons among 300 samples can be classified into five types of gustatory neurons in GNG including the gustatory sensory neuron (GSN), the local neuron (LN), the projection neuron (PN), the motor neuron (MN) and the descending neuron (DN). In addition, we have been successfully labelled 64 functional neurons including 12 GSNs, 23 GLNs, 22 GPNs, 4 GMNs, and 3 GDNs. The innervating areas showed no obviously distinct projection pathways between the sweet and the bitter central neurons based on the morphology and electrophysiology of the limited investigated gustatory neurons, which indicates that the projection patterns of different central gustatory neurons may not related to the responding properties themselves, but more investigations should be carried out in the near future.

Keywords: Sensory neuron, Interneuron, Motor neuron, Gnathal ganglion, Taste coding model

S4-O113

Correspondence between input and output information in the macroglomerular complex in male *Helicoverpa armigera* (Hübner)

Xiao-Lan Liu[1,2], Xi Chu[3], Long-Long Sun[4], Ya-Nan Wang[4], Gui-Ying Xie[4], Wen-Bo Chen[4], Yang Liu[2], Bente Gunnveig Berg[5], Shi-Heng An[4], Gui-Rong Wang[2], Xin-Ming Yin[4], Xin-Cheng Zhao[4]

[1]*Henan International Joint Laboratory of Green Pest Control, College of Plant Protection, Henan Agricultural University, ZhengZhou, China.* [2]*State Key Laboratory for Biology of Plant Disease and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China.* [3]*Chemosensory laboratory, Department of Psychology, Norwegian University of Science and Technology, Trondheim, Norway.* [4]*Department of Entomology, College of Plant Protection, Henan Agricultural University, ZhengZhou, China.* [5]*Chemosensory laboratory, Department of Psychology, Norwegian University of Science and Technology, Beijing, China*

The male-specific macroglomerular complex (MGC) in the antennal lobe receives the input from the femaleproduced sex pheromones via olfactory sensory neurons (OSNs) housed in sensilla on the antenna. Second order projection neurons (PNs) convey the output information from the MGC to the higher olfactory center in the protocerebrum. In the study presented here, we performed electrophysiological and morphological characterization of the input- and the output neurons of the MGC in *Helicoverpa armigera*. By utilizing the techniques of single-sensillum recording and tracing and intracellular recording and staining we could identify individual OSNs and PNs. Our results show that OSNs localized in Type A trichoid sensilla responded to the major pheromone component, Z11-16:Ald, and their axonal terminals projected to the cumulus (Cu) of MGC. The OSNs in Type B trichoid sensilla responded to the behavioral antagonist, Z9-14:Ald, and their axonal terminals projected to the anterior dorsomedial (DMA) unit of the MGC. In Type C trichoid sensilla, there were two OSNs: one that responded to the antagonists, Z9-14:Ald and Z11-16:OH, with axon terminals projecting to the DMA, and another that responded to the minor components Z9-16:Ald and Z9-14:Ald, with axonal terminals projecting to the posterior dorsomedial (DMP) unit of the MGC. Correspondingly, three groups of uniglomerular PNs, each of which responded to 1) Z11-16:Ald, 2) Z9-14:Ald and Z11-16:OH, and 3) Z9-14:Ald and Z9-16:Ald extended their dendrites in Cu, DMA, and DMP, respectively. In conclusion, we found that Cu is responsible for the major pheromone, DMA and DMP are responsible for the secondary pheromones and antagonists, respectively.

Keywords: Olfactory sensory neuron, Projection neuron, Macroglomerular complex, Single sensillum recording, Intracellular recording.

S4-O133

Morphological identification of the neurons control Aphid cornicle muscle

Baiwei Ma[1,2,3], Guirong Wang[2], Bingzhong Ren[1,3]

[1]*Jilin Provincial Key Laboratory of Animal Resource Conservation and Utilization, School of Life Sciences, Northeast Normal University, Changchun, China.* [2]*State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China.* [3]*Key Laboratory of Vegetation Ecology, MOE, Northeast Normal University, Changchun, China*

Aphid cornicles are abdominal appendages that secrete volatile and nonvolatile compounds with diverse ecological functions. Until now, very few articles researched the structure of cornicle and found all aphids possessed a muscle originating on the venter bellow the cornicle and inserting on the cornicle valve. But no research on the morphologies of neurons control the cornicle muscle. In order to identify these neurons, we used neuronal tracer to backfill the neurons from transverse incision of unilateral cornicle, after dissection of the central nervous system (CNS), fixed in the formaldehyde solution and a series of immunohistochemical procedures, we scanned the CNS in confocal laser scanning microscope. Finally, we made three dimensional reconstructions of these neurons in software. We find that there are only motor neurons contact with the cornicle, their somas and dendrite distribute in the end of the ventral nerve cord.

Keywords: Aphid, Cornicle, Motor neuron

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S4-O136

Sex differential expression analysis on antennae transcriptome of *Hyphantria cunea*

Yaning Li[1,2], Shanchun Yan[1,2]

[1]*School of Forestry, Northeast Forestry University, Harbin, China.* [2]*Key Laboratory of Sustainable Forest Ecosystem Management-Ministry of Education, Northeast Forestry University, Harbin, China*

Hyphantria cunea (Drury) is an invasive quarantine pest, and its host is generally adaptable, which gravely endangers forestry safety in China. The sense of smell is the basis of insect behaviors such as feeding, courtship, mating, host location, looking for spawning sites, and avoiding enemies, which is crucial for the survival and reproduction of its species. Insect recognition of chemical signals mainly involves three kinds of proteins, namely odor binding protein (OBP), odor receptor (OR) and odor degrading enzyme. Therefore, the identification of olfactory receptors may provide a new strategy for the scientific control of *Hyphantria cunea*. In this study, the antennae of male and female *Hyphantria cunea* were sequenced, splined and assembled by high-throughput sequencing technology. Common odor receptors were screened and differential genes were found by principal component analysis and differential analysis. Quantitative real-time PCR (qRT-PCR) was used to study the expression of common odor receptors, and the sequencing results were verified to study the gene expression differences between male and female antennae tissues of *Hyphantria cunea*. A total of 3923 sex-related differential genes were screened, including 1620 up-regulated genes in males and 2303 up-regulated genes in females. The GO annotation includes three categories and 41 subcategories, among which the proportion of genes involved in biological processes is large. A total of 11059 genes were annotated by KO enrichment analysis, including 475 differential genes. Based on the third generation transcriptomic data, 17 common odor receptors were annotated. Real-time quantitative PCR was conducted to verify the transcriptomic differential analysis results, and four genes with gender differential expression were found, among which Isoform5835 and 20145 were female preferentially expressed, and Isoform19497 and 27520 were male preferentially expressed.

Keywords: *Hyphantria cunea*, Antennae transcriptome, Differentially expressed genes, Common odor receptor

S4-O146

A female specific odorant receptor tuning to egg-surface odorants mediates the oviposition aversion behavior of *Helicoverpa armigera*

Xiaxuan Zhang[1,2], Mengbo Guo[1], Dongdong Sun[2], Yang Liu[2], Guirong Wang[1,2]

[1]*Agricultural Genomics Institute at Shenzhen, Chinese Academy of Agricultural Sciences, Shenzhen, China.* [2]*Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China*

Oviposition is one of the most important aspects of female behaviors and finding a suitable oviposition site is a very challenging task for insects. Oviposition-deterrent pheromones (ODPs) usually released from egg surface or larvae are vital for gravid females to find the appropriate oviposition sites. However, the molecular mechanisms underlying detection of ODPs remain open. Here, we showed that egg surface volatiles including methyl oleate (MO), methyl palmitate (MP), and methyl stearate (MS) deter *Helicoverpa armigera* oviposition. We further characterized that *OR56* exclusively expressed in female antennae and are specifically tuned to the three ODPs (MO, MP, and MS). Our study also firstly showed that neurons expressing *OR56* housed in a class of sensilla trichodea on female antennae can be activated by ODPs. To test this, we disrupted the *OR56* in *H. armigera* using CRISPR/Cas9 and found that OR56 mutant females are not avoided to MO, MP, and MS. In addition, single sensillum recording (SSR) experiments showed that OR56 mutants lost SSR response to three ODPs. In sum, OR56 is necessary for *H. armigera* to detect ODPs. We hence conclude that oviposition aversion behaviors of *H. armigera* is governed by *OR56* specifically expressed in female antennae. Our research showed that how insects detect odors from eggs and will provide considerable insight into the recognition of ODPs in insets.

Keywords: *Helicoverpa armigera*, Oviposition-deterrent pheromones, Olfaction, Odorant receptor, Oviposition

S4-O148

Molecular regulatory mechanisms of contact sex pheromone biosynthesis in the German cockroach, *Blattella germanica*

Yongliang Fan[1], Sheng Li[2], Tong-Xian Liu[1]

[1]*Northwest A&F University, Yangling, China.* [2]*South China Normal University, Guangzhou, China*

In the German cockroach, *Blattella germanica*, females produce a blend of hydrocarbon-derived nonvolatile contact sex pheromone (CSP) components to elicit close-range male courting behavior, however, the molecular mechanisms on the regulation of CSP biosynthesis are mostly unknown. With mainly the available genome, transcriptome and using RNAi technology, we have delineated the specific functional fatty acid synthase (BgFas1), fatty acid elongases (BgElo12, BgElo24), and a P450 decarboxylase (CYP4G19) participating in hydrocarbons biosynthesis. Molecular mechanism on the regulation of sexually dimorphic cuticular hydrocarbon profiles in *B. germanica* has been revealed. We found that the differential expression of BgElo12 between the sexes is modulated by sex differentiation pathway. Male-specific doublesex (BgDsx) inhibits the transcriptional expression of BgElo12 in males.Further investigation led the revelation of the molecular mechanisms on the regulation of CSP biosynthesis in B. germanica. It has long been proposed that contact sex pheromone in *B. germanica* is controlled by JH and a sex-specific expression of P450 hydroxylase converting hydrocarbons to the corresponding methyl ketone sex pheromones. However, the proposed P450 gene has not been confirmed until now. Using RNA-SEQ technology we recently nailed down this elusive hydroxylase CYP4PC1. Surprisingly spatio-temporal gene expression of found that CYP4PC1is most highly expressed in the antennae. Our results showed that CYP4PC1 and CSP is regulated by the coordinated and reciprocal action of sex differentiation genes and JH. In females JH and Bgtra stimulate CYP4PC1 expression and regulate CSP production, while lack of JH and the male-specific splicing variant of Bgdsx repress CYP4PC1 expression. By removing the inhibition of BgDsx on CYP4PC1 in males we observed the male-male sexual courting behavior. This work provides a staging ground for future research that aims to understand the evolution of CSP production pathways.

Keywords: Sex pheromone, Hydrocarbon, Hydroxylase, Sexual dimorphism, P450

Symposium 5: Future generations of chemical ecologists (1)

Thursday, August 11th
8:45 am - 12:25 pm

Session moderated by Andrés Gonzalez

An untapped plant defense: novel biopesticide and plant-mediated RNAi combination for the pest management
Manish Kumar, Umesh KP, Animesh Anand, Sagar Pandit

Identification of non-host plant volatiles for the management of the invasive tomato leafminer, *Tuta absoluta*
Bashiru Adams, Abdullahi Ahmed Yusuf, Baldwyn Torto, Fathiya Khamis

Fruit flies (Diptera: *Tephritidae*) olfactory systems specialize in discriminating their various hosts
Gaëlle Ramiaranjatovo, Maud Charlery de la Masselière, Pierre-François Duyck, Vincent Jacob

Host plant quality vs. enemy-reduced space: explaining oviposition preference and larval performance patterns in *Plutella xylostella*
Aswathi Sasidharan, Enakshi Ghosh, Radhika Venkatesan

Chemical communication in a highly specialized plant-pollinator interaction: kinetic of emission of plant VOCs and genes involved in their production
Li CAO, Anne-Geneviève Bagnères, Denis Saint-Marcoux, Aurélie Bony, Valentin Vrecko, Joris Huguenin, Martine Hossaert-McKey, Sylvie Baudino, Jean-Louis Magnard, Magali Proffit

Nocturnal atmospheric NOx pollution impacts hawkmoth visitation in the field by degrading *Oenothera pallida* scent
Jeremy K Chan, Joel A Thornton, Jeffrey A Riffell

Olfactory perception in elevated ozone: the role of ozone reaction products
Vignesh Venkateswaran, Jerrit Weißflog, Markus Knaden, Bill Hansson

Pathogenic fungus enhances dispersal through alteration of insect olfactory behavior
Amanda C. Túler, Arodí P. Favaris, Flávia P. Franco, Diego Z. Gallan, Felipe G. Gonçalves, Maria Fernanda G. V. Peñaflor, Walter S. Leal, Daniel S. Moura, Marcio C. Silva-Filho

Sexual dimorphism of cuticular wax mediates male mating behavior in the strawberry leaf beetle, *Galerucella grisescens* (Coleoptera: Chrysomelidae)
Yuki Chiba, Masatoshi Hori

Secondary metabolites of amphibian skin-associated bacteria
Peter Biwer, Molly Bletz, Miguel Vences, Stefan Schulz

S5-O109

An untapped plant defense: novel biopesticide and plant-mediated RNAi combination for the pest management

Manish Kumar, Umesh KP, Animesh Anand, Sagar Pandit

Agricultural Biotechnology and Chemical Ecology Laboratory, Biology Department, Indian Institute of Science Education and Research, Pune, Pune, India

Insect pests are a major threat to farmers and cause severe crop losses globally. Synthetic pesticides are injudiciously used to control these pests and cause environmental and health hazards. We explored secondary metabolite-rich *Solanum melongena* L. (eggplant), one of the host species of *Spodoptera litura* Fabricius (armyworm; a polyphagous and multi-insecticide-resistant lepidopteran pest) as a biopesticide source. By integrating non-targeted metabolomics and reverse genetics approaches, we identified a secondary metabolite from eggplants with insecticidal potential. This candidate metabolite negatively correlates with the occurrence and performance of *S. litura* larvae among different eggplant varieties. It caused a three-fold mass reduction and two-fold mortality increase in larvae; pupation and eclosion also reduced (upto 1.3-fold and 1.4-fold, respectively) when fed through an artificial diet. Nutritional indices revealed that ingestion of this candidate metabolite by *S. litura* hampered larval digestion and metabolism, leading to slower larval growth and delayed pupation and eclosion. Further, upon silencing the biosynthesis gene of this metabolite, eggplants were rendered two-fold armyworm-susceptible than controls. We also discovered the counter-adaptation mechanism and gene used by *S. litura* larvae against this metabolite to ensure survival. By silencing this gene using plant-mediated RNAi (PMRi), we generated a loss-of-function phenotype, against which our candidate metabolite was highly effective and showed 3.5-fold mass reduction and 4-fold mortality increase in larvae. Foliar application of dsRNA on eggplant showed similar effects when fed to the larvae. This candidate metabolite also showed no negative effects on armyworms' natural enemies, the biocontrol agents. Considering our findings, we propose a novel pest management strategy combining plant-based candidate insecticide(s) and PMRi (or foliar spray of dsRNA) mediated silencing of counter-adaptation genes to combat infestations of *S. litura* and other lepidopteran pests in agronomically important plants.

Keywords: Biopesticide, Eggplant, *Spodoptera litura*, Metabolomics, Plant mediated RNAi

S5-O5

Identification of non-host plant volatiles for the management of the invasive tomato leafminer, *Tuta absoluta*

Bashiru Adams[1,2], Abdullahi Ahmed Yusuf[2], Baldwyn Torto[1,2], Fathiya Khamis[1]

[1]*International Centre of Insect Physiology and Ecology, Nairobi, Kenya.* [2]*University of Pretoria, Hatfield, South Africa*

The tomato leafminer, *Tuta absoluta* (Meyrick) is a destructive invasive pest of cultivated tomato (*Solanum lycopersicum*) and other Solanaceae plants. Management of this pest is a challenge due to its high biotic potential and resistance to many insecticides. Olfactory cues mediate host finding in *T. absoluta*, hence biological control, especially use of repellent plants is an environmentally safe option to manage it. Previous work has demonstrated that wild tomato species intercropped with tomato negatively impacts on *T. absoluta* infestation. Additionally, studies have demonstrated that Asteraceae plants contain phytochemicals that exhibit insecticidal and pharmacological properties against pests and diseases. Therefore, we tested the hypothesis that odours from some of these known non-host plants can disrupt the host finding process of the pest. We used Y-tube olfactometer assays to screen the effects of odours of three of these non-host plants including (wild tomato (*Lycopersicon esculentum*) (Solanaceae) and the Asteraceae plants blackjack (*Bidens pilosa*), marigold (*Tagetes minuta*), and sunflower (*Helianthus annuus*) against adult females of *T. absoluta*. Females avoidance/repellence varied with the non-host plants when tested alone or in combination with the host plants cultivated tomato and the related plant species giant nightshade (*Solanum scabrum*). Chemical analysis by coupled gas chromatography-mass spectrometry showed that the host and non-host plants differed in their levels of volatiles, mainly monoterpenes and sesquiterpenes. While our olfactometer assays showed that the host plant attractive volatiles comprised a blend of monoterpenes, the non-host plant avoidance/repellent volatile blend was dominated by sesquiterpenes. Our findings suggest that monoterpenes and sesquiterpenes shape the attraction/avoidance/ repellent dynamics of *T. absoluta*, which can be exploited in a 'push-pull' strategy for its management.

Keywords: *Tuta absoluta*, Non-host plants, Terpenes, Attraction, Repellents

[1] Mwamba S, Kihika-Opanda R, Murungi LK, Losenge T, Beck JJ, Torto B (2021) J Agric Food Chem 69: 15145–15156

S5-O23

Fruit flies (Diptera: *Tephritidae*) olfactory systems specialize in discriminating their various hosts

Gaëlle Ramiaranjatovo, Maud Charlery de la Masselière, Pierre-François Duyck, Vincent Jacob

CIRAD, UMR PVBMT, La Réunion, France

Host volatile compounds play an essential role in locating egg-laying sites for insects. In this study, we aim to understand the sensory mechanisms used by polyphagous *Tephritidae* fruit fly species to recognize their host-fruits. For generalist species, it is generally thought that among the various compounds emitted by the wide range of hosts, insects rely primarily on those shared by most hosts. To challenge this hypothesis, we collected volatile emissions from 28 host-fruit species from intact and sliced fruits using Tenax traps (n=7). The samples were analyzed with thermal desorption, gas chromatography (GC) and mass spectrometry. We identified through retention index and mass spectrum 511 volatile compounds from intact fruits and 665 from sliced fruits. Phylogenetic principal component analysis highlighted the compounds specific to phylogenetic groups of fruits, while sharedness among fruit emissions, irrespectively of phylogeny, was estimated by an α-diversity index. Next, we assessed the olfactory sensitivity of 8 fruit fly species to 37 synthetic compounds by GC coupled with a triple electroantennogram detector (n=8 per species). We found for each species a significant negative correlation between the olfactory sensitivity of fruit flies and α-diversity. Accordingly, a behavioral bioassay showed that at low concentration *Bactrocera dorsalis* prefers compounds specifically emitted by few fruits species than compounds which are shared among fruits. This unexpected result supports a new hypothesis: *Tephritidae* species are not only specialized in detecting their hosts, but also in discriminating them. Optimization of host-discrimination would have resulted in low sensitivity threshold for specific compounds and wider dynamic range for shared compounds. This new concept implies many selective advantages, including adaptation to the local background, and should also drive the search for more effective attractants.

Keywords: Attractants, *Bactrocera dorsalis*, Evolution, GC-EAD, Kairomone.

S5-O82

Host plant quality vs. enemy-reduced space: explaining oviposition preference and larval performance patterns in *Plutella xylostella*

Aswathi Sasidharan[1], Enakshi Ghosh[2], Radhika Venkatesan[1,3]

[1]*Indian Institute of Science Education and Research Kolkata, Mohanpur, India.* [2]*Colorado State University, Fort Collins, USA.* [3]*National Centre for Biological Sciences (NCBS), Tata Institute of Fundamental Research (TIFR), Bangalore, India*

Oviposition behavior by herbivorous insects depends on the nutritional quality of host plants and the risk of attack by their natural enemies. We report the oviposition preference of a specialist herbivore and its offspring's performance in a tri-trophic system constituting bottom-up and top-down factors. We observed that oviposition preference and performance are not correlated in *Plutella xylostella* - plant interactions. The adult preference was more towards secondary metabolite (SM) -rich host plant though the performance of the offspring and the life span was compromised. We hypothesize that plant defensive chemicals provide an enemy-reduced space for the herbivore to escape from its natural enemies. Feeding on an SM-rich host provided better larval cellular immunity in terms of higher hemocyte counts and phenoloxidase activity. This elevated immunity also increased larval survival against the entomopathogens and generalist parasitoid wasp. We further observed a positive correlation of larval immunity with the concentration of SMs. However, the enhanced immunity did not influence the parasitization by a specialist larval parasitoid wasp. We have also characterized close-range volatile cues of larval origin that impact parasitoid oviposition. Interestingly, parasitoids host preference guided by these volatile cues correlates with its performance. In summary, our results show that the oviposition of herbivores and parasitoids is highly context-dependent and can be understood better only by studying multiple fitness parameters and interactions with different natural enemies.

Keywords: Oviposition preference, Secondary metabolite, Cellular immunity, Parasitoid wasp, Enemy-free space

[1] Videla M, Valladares GR, Salvo A (2012) Oecologia 169:743– 751.
[2] Ghosh E, Sasidharan A, Ode PJ, Venkatesan R (2022) J Chem Ecol. https://doi.org/10.1007/s10886-022-01363-5

S5-O30

Chemical communication in a highly specialized plant-pollinator interaction: kinetic of emission of plant VOCs and genes involved in their production

Li CAO[1], Anne-Geneviève Bagnères[1], Denis Saint-Marcoux[2], Aurélie Bony[2], Valentin Vrecko[1], Joris Huguenin[1], Martine Hossaert-McKey[1], Sylvie Baudino[2], Jean-Louis Magnard[2], Magali Proffit[1]

[1]CEFE, CNRS/University of Montpellier/EPHE /IRD, Montpellier, France. [2]LBVpam, Université de Lyon, Université de Saint-Etienne/ UMR 5079, CNRS, Saint-Etienne, France

Several studies have described the Volatile Organic Compounds (VOCs) emitted by flowers and involved in plant/pollinator interactions. Nevertheless, there is a lack of information linking these VOCs to the enzymes involved in their biosynthesis and in turn to the genes coding these enzymes. Using a model system for specialized pollination interactions, those between the dioecious Mediterranean fig tree, *Ficus carica* and its pollinator, *Blastophaga psenes*, we combined VOC and RNA-seq analyses of receptive and pollinated figs to uncover the genes underlying chemical signaling by the plant to its pollinator, and the pathways of VOC biosynthesis. Knowing that *B. psenes*, is attracted by the mixture of four VOCs (Benzyl alcohol, S-Linalool, trans-Linalool oxide and cis-Linalool oxide) we sought to characterize the function of some genes potentially responsible for the synthesis of these VOCs. PTR-ToF-MS and GC-MS analyses of the VOCs emitted by *F. carica* allowed us to characterize the rate of emission of these compounds throughout the day and to characterize the changes occurring after pollination. Comparative transcriptomics between receptive and pollinated figs allowed us to highlight some genes whose expression is down-regulated after pollination. Expression in *Escherichia coli* and *Nicotiana benthamiana* of two genes coding for terpene synthases (TPS) revealed that a monoterpene synthase belonging to the TPS-b clade, converts GPP to S-Linalool, whereas a sesquiterpene synthase of the TPS-a clade catalyzes the formation of Germacrene D. This study provides new insights into plant/pollinator interactions and understanding of the molecular mechanisms of terpene biosynthesis.

Keywords: VOCs, *Ficus carica*, Plant/pollinator interactions, Terpene synthases

[1] Dudareva N, Klempien A, Muhlemann JK, Kaplan I (2013) New Phytologist 198: 16-32
[2] Proffit M, Lapeyre B, Buatois B, Deng XX, Arnal P, Gouzerh F, Carrasco D, Hossaert-McKey M (2020) Scientific Reports 10: 10071

S5-O115

Nocturnal atmospheric NOx pollution impacts hawkmoth visitation in the field by degrading *Oenothera pallida* scent

Jeremy K Chan, Joel A Thornton, Jeffrey A Riffell

University of Washington, Seattle, USA

Floral scent signals are used by many pollinators to locate flowers. These scent signals are vulnerable to degradation in polluted atmospheres by ozone and nitrogen oxides (collectively, NOx). Previous studies have shown that air pollution impacts the ability of pollinators to recognize floral scent signals by either degrading or masking these signals. These studies have examined the impacts of ambient VOCs and ozone oxidation, but a major part of the atmosphere's oxidizing capacity is from radical species like hydroxy and nitrate radicals whose impacts have yet to be studied. Using the hawkmoth-pollinated flower, *Oenothera pallida*, in the sagebrush steppes of eastern Washington, we examined how NOx pollution impacts moth behavior and floral visitation. Scent signals are particularly important for these moths due to the lack of visual signals at night. We used GCMS and GC-EAD to identify antennographically active floral scent chemicals, and a VOCUS high resolution time-of-flight mass spectrometer to analyze floral scent degradation by NOx. Analysis of floral scent degradation under simulated high NOx levels revealed that nitrate radicals degrade floral scent compounds significantly more than ozone alone, and that the monoterpene component of the scent is heavily affected. Field behavioral experiments show that this impacts the ability of hawkmoths to locate the scent sources. Next, we used the GEOS-Chem global chemical model to show that NOx pollution around the world might significantly decreases floral scent recognition distances by up to 90% over much of the northern latitudes and southern Africa. Together, our results show that atmospheric NOx pollution degrades floral scent chemicals, particularly the monoterpene components. This impacts pollinator olfactory navigation to the flowers, and global chemical modeling shows that there are significant global impacts of NOx on pollinator olfactory navigation.

Keywords: Air pollution, Scent degradation, Olfactory navigation, Pollination, Global model

S5-O51

Olfactory perception in elevated ozone: the role of ozone reaction products

Vignesh Venkateswaran, Jerrit Weißflog, Markus Knaden, Bill Hansson

Max Planck Instiutute for Chemical Ecology, Jena, Germany

Oxidative degradation of volatile organic compounds can destroy olfactory blends mediating insect olfactory perception in nature. Little is known about the role of the reaction products in olfactory perception. These products may change olfactory perception by increasing in the background, thereby disrupting the behavioural responses of organisms. Using *Drosophila melanogaster* as a model organism, we targeted nearly 600 odourants listed in the DoOR2.0 fly database (a database of odorant-receptor responses to various odorants) and predicted their oxidative breakdown with ozone. We generated a ranked list of the most consistently occurring ozone byproducts and targeted the top ten most commonly occurring products to investigate their behavioural and neurophysiological relevance. We tested the intrinsic valence of flies to each of these compounds, both independently and in the context of the background. We also investigated the single sensillum responses to these odourants. All products were perceived by one of the three tested sensilla associated with foraging. All products varied in behavioural responses from being attractive to neutral, but not aversive. When their concentrations were increased in the background, no change in the intrinsic attraction to a blend of balsamic vinegar (BV) was observed. However, when given a simultaneous choice between BV and BV+product, flies were significantly attracted or repelled by the BV product mixtures in a product specific manner. Our results strongly suggest that the byproducts of oxidative breakdown of VOC bouquets disrupt olfactory decisions. Increased ozone concentrations in the anthropocene are expected to impact olfactory sensory perception in insects which will negatively impact crucial ecosystem services. Our study provides an important foundation for accounting for the byproducts of oxidative degradation of odours by air pollution for olfactory mediated behaviours of insects.

Keywords: Ozone, Insects, Olfaction, Anthropocene, Behaviour

[1] Agathokleous, Evgenios, et al. Science Advances 6.33 (2020): eabc1176

S5-O125

Pathogenic fungus enhances dispersal through alteration of insect olfactory behavior

Amanda C. Túler[1], Arodí P. Favaris[1], Flávia P. Franco[2], Diego Z. Gallan[2], Felipe G. Gonçalves[1], Maria Fernanda G. V. Peñaflor[3], Walter S. Leal[4], Daniel S. Moura[5], Marcio C. Silva-Filho[2]

[1]Department Entomology and Acarology, "Luis de Queiroz" College of Agriculture, University of São Paulo, Piracicaba, Brazil. [2]Department of Genetics, "Luis de Queiroz" College of Agriculture, University of São Paulo, Piracicaba, Brazil. [3]Department of Entomology, Federal University of Lavras, Lavras, Brazil. [4]Department of Molecular and Cellular Biology, University of California, Davis, Brazil. [5]Department of Biological Sciences, "Luis de Queiroz" College of Agriculture, University of São Paulo, Piracicaba, Piracicaba, Brazil

Vector-borne plant pathogens often change host traits to manipulate vector behavior to favor their spread. Different from vector-borne microorganisms, opportunistic pathogens presumably do not have an intimate association with the insect vector, although damage caused by an herbivore may facilitate infection. Herein we establish a new role for the insect-fungus-sugarcane association. It has long been assumed that *Fusarium verticillioides* is an opportunistic fungus where it takes advantage of the openings left by *Diatraea saccharalis* caterpillar attack to infect the plant. This study addressed the chemical identification of plant volatile organic compounds (VOCs) induced by fungus infection; then, we conducted electrophysiological (EAG) and olfactory preference assays to test whether the fungus influences the olfactory-based behavior of *D. saccharalis* adults. Additionally, the fungus transmission in sugarcane plants by *D. saccharalis* was investigated by tracking the fungus using a mutant *F. verticillioides* (Fv:DsRed) strain. We show that *D. saccharalis* moths orient toward VOCs emitted by fungus-infected plants to lay their eggs. Once they become adults, the fungus spreads to healthy plants by *D. saccharalis* vertical transmission through its contaminated offspring. Remarkably, the host preference changes if the insects are carriers of the fungus. Instrumentalized by the VOCs, females not carrying the fungus prefer to lay their eggs on fungus-infected plants, while females carrying the fungus prefer to lay their eggs on mock lants. Altogether, our data demonstrate that the fungus manipulates both the host plant and insect herbivore across the life cycle to promote its infection and dissemination.

Keywords: VOCs, Olfaction, Tri-trophic interaction

S5-O9

Sexual dimorphism of cuticular wax mediates male mating behavior in the strawberry leaf beetle, *Galerucella grisescens* (Coleoptera: Chrysomelidae)

Yuki Chiba, Masatoshi Hori

Graduate School of Agricultural Science, Tohoku University, Sendai, Japan

The surface of the insect body is covered with a waxy layer, called cuticular wax, which provides desiccation resistance to insects. Apart from functioning as an anti-desiccation agent, the cuticular wax is a source of semiochemicals. In Chrysomelidae, it has been reported that sex pheromones can be found on the female cuticular wax [1]. However, little is known on how the chemical information in the cuticular wax is processed by the peripheral nervous system, and how this affects male mating behavior. Here, we report that the males of the strawberry leaf beetle, *Galerucella grisescens*, can discriminate between sexes using the cuticular wax. Behavioral bioassays showed that both female and male cuticular wax elicited the male mounting behavior, while only female wax induced the following aedeagal insertion. GC/MS analysis showed that the profile of the cuticular hydrocarbons (CHCs), major components of the cuticular wax, was qualitatively different between sexes. Although more than 90% of male CHCs consisted of C24–C28 alkanes, the major components of female CHCs were C29–C33 alkanes. These indicate that a clear sexual dimorphism in CHCs enables males to distinguish females in this species. We also conducted morphological observations using electron microscopes and electrophysiological studies to evaluate how sexual differences of the cuticular wax influence the response of male chemosensory neurons. Through these experiments, we found that hair-like gustatory sensilla are one of the chemosensory organs sensing the cuticular wax. Moreover, some of the targeted sensilla elicited significantly different responses between receiving the female or male cuticular wax. Based on our findings, we discuss the importance of the sexual dimorphism of the cuticular wax for mate discrimination in the context of the insect chemosensory system.

Keywords: Cuticular wax, Chrysomelidae, Sexual dimorphism, Sensilla

[1] Sugeno W et al. (2006) Appl Entomol Zool 41: 269-276

S5-O36

Secondary metabolites of amphibian skin-associated bacteria

Peter Biwer[1], Molly Bletz[2], Miguel Vences[3], Stefan Schulz[1]

[1]*Institute for Organic Chemistry, Technische Unviersität Braunschweig, Braunschweig, Germany.* [2]*Department of Biology, University of Massachusetts, Boston, USA.* [3]*Institute for Zoology, Technische Unviersität Braunschweig, Braunschweig, Germany*

Chytridiomycosis is a severe skin disease caused by *Batrachochytrium dendrobatidis* (Bd) currently leading to a worldwide decline in the amphibian population. Prior studies suggested that amphibian skin microbiota play a key role in mediating resistance mechanisms against Bd infection by secretion of secondary metabolites. Chemical investigation of amphibian skin-associated bacteria might reveal new antagonists. Our work focuses on the organic compounds released by skin-associated bacteria isolated from German and Madagascan amphibians. Volatile and some non-volatile organic compounds were analyzed by GC/MS and HPLC/MS. Identification of metabolites was carried out by comparison of mass spectra and retention indices with data obtained from databases and authentic (synthesized) samples, as well as analysis of genome data. Among albaflavenone and violacein, both known for their biological activities, a plethora of terpenoids, ketones and sulfur volatiles were identified. Our results show that compounds isolated from skin-associated bacteria might be involved in regulating pathogen attack and structuring bacterial diversity. Furthermore, they add to the specific smell of the respective amphibian host and might influence the behavior of con- and heterospecifics.

Keywords: *Batrachochytrium dendrobatidis*; Chytridiomycosis; GC/MS; Pathogens; Volatile organic compounds

[1] Fisher MC, Garner, TW, Walker SF (2009) Annu. Rev. Microbiol. 63: 291-310.
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[3] Woodhams DC, LaBumbard BC, Barnhart KL, Becker MH, Bletz MC, Escobar LA, Flechas SV, Forman ME, Ianetta AA, Joyce MD, Rabemananjara F, Gratwicke B, Vences M, Minbiole KPC (2018). Microb. Ecol. 75: 1049-1062.
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Symposium 5: Future generations of chemical ecologists (2)

Friday, August 12th
3:10 am - 3:30 pm

Session moderated by Andrés Gonzalez

The frass excreted by the striped stem borer *Chilo suppressalis* induces defense responses in rice.
Shuting Chen, Miaofen Ye, Shiyun Jing, Peng Kuai, Na Lin, Yonggen Lou

S5-O6

The frass excreted by the striped stem borer *Chilo suppressalis* induces defense responses in rice.

Shuting Chen, Miaofen Ye, Shiyun Jing, Peng Kuai, Na Lin, Yonggen Lou

State Key Laboratory of Rice Biology & Ministry of Agriculture Key Lab of Agricultural Entomology, Institute of Insect Sciences, Zhejiang University, Hangzhou, China

The striped stem borer (SSB, *Chilo suppressalis* (Walker)), whose larvae bore into and feed on rice stems and excrete frass inside and outside holes, is the most damaging recurrent pest on rice (*Oryza sativa*) in China. It has been well documented that SSB infestation in rice induces the biosynthesis of a variety of phytohormones, including jasmonic acid (JA), salicylic acid (SA), and ethylene, which, in turn, regulate the production of defensive compounds, such as volatile organic compounds (VOCs) and trypsin protease inhibitors. However, whether the frass excreted by SSB larvae is involved in the SSB larva-induced defenses in rice remains unknown. In this study, we observed that treating plants with SSB frass solution in a phosphate buffer (pH 7.4) could rapidly activate the mitogen-activated protein kinase (MPK) cascades, an early event in herbivore-induced plant defenses, and the jasmonic acid (JA) signaling pathway. Moreover, treatment with SSB frass solution induced the production of phenolamides (PAs) and VOCs and enhanced the activity of trypsin protease inhibitors in rice, which subsequently inhibited the growth of SSB larvae. We also found that the transcription level of *OsMPK3* and *OsAOS1*, two marker genes for MPK cascades and JA signaling pathway, respectively, in rice plants treated with SSB frass solution digested by proteinase K was significantly lower than that in plants treated with SSB frass solution without proteinase K. These results suggest that SSB frass, especially proteins in it plays a vital role in SSB larva-induced defenses in rice.

Keywords: Frass, Striped stem borer (*Chilo suppressalis* (Walker)), Plant defense, Defensive compounds, Rice (*Oryza sativa*).

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Symposium 6: Chemical and molecular ecology of plant-herbivore-enemy interactions (1)

Thursday, August 11th
8:20 am - 10:30 am

Session moderated by Nicole van Dam

Same problem, different solutions. Isothiocyanate detoxification mechanisms in larvae of two related root herbivore species, *Delia radicum* and *D. floralis*
Nicole M. van Dam, Rebekka Sontowski, Axel Touw, Alexander Weinhold, Yvonne Poeschl, Cervin Guyomar, Daniel G. Vassão

Bottom-up and top-down induced plant responses in wild lima bean plants and their consequences for plant fitness and trophic interactions
Betty Benrey, Maximilien Cuny, Carlos Bustos-Segura

Constitutive and herbivore-induced volatiles emitted by wild cotton populations
Marine Mamin, Mary V Clancy, Galien Flückiger, Teresa Quijano-Medina, Biinza Pérez Niño, Luis Abdala-Roberts, Ted CJ Turlings, Carlos Bustos-Segura

Understanding host-parasitoid interactions for application in integrated pest management
Radhika Venkatesan, Enakshi Ghosh, Aswathi Sasidharan

Can plant silicon defenses play a part in biocontrol of herbivorous arthropods?
Tarikul Islam, Sidra Anwar, Christopher Cazzonelli, Ben Moore, Scott Johnson

S6-O39

Same problem, different solutions. Isothiocyanate detoxification mechanisms in larvae of two related root herbivore species, *Delia radicum* and *D. floralis*

Nicole M. van Dam[1,2], Rebekka Sontowski[1,2], Axel Touw[1,2], Alexander Weinhold[1,3], Yvonne Poeschl[1,2], Cervin Guyomar[4], Daniel G. Vassão[5]

[1]German Centre for Integrative Biodiversity Research (iDiv) Halle-Jena-Leipzig, Leipzig, Germany. [2]Friedrich Schiller University, Jena, Germany. [3]Friedrich Schiller University, Leipzig, Germany. [4]GenPhySE, Université de Toulouse, INRAE, ENVT, Castanet Tolosan, Germany. [5]Max Planck Institute for Chemical Ecology, Jena, Germany

Glucosinolates (GSLs) are metabolites specific to species belonging to the Brassicaceae. Upon herbivore damage, GSLs are enzymatically hydrolyzed, yielding isothiocyanates (ITCs) and other defensive compounds. Several aboveground herbivores can cope with ITCs by employing detoxification mechanisms that prevent GSL hydrolysis, divert GSL conversion to less toxic products or detoxify ITCs. Larvae of the cabbage root fly, *Delia radicum*, and the turnip root fly, *D. floralis*, likely deploy similar mechanisms. Brassica roots contain high levels of GSLs, yet are heavily damaged by these root herbivores. We hypothesized that both *Delia* species possess similar GSL-ITC detoxification mechanisms, considering that they are congeners and have overlapping host ranges. We also considered the role of the gut bacterial community (GBC), which may contain bacteria with ITC detoxification enzymes. Larvae were fed on artificial diets spiked with different ITCs. Chemical analyses yielded glutathione conjugates and amines as breakdown products. This suggests that both species detoxify ITCs via the mercapturic acid pathway also found in aboveground herbivores, and the hydrolytic pathway commonly observed in microbes. Using the genome of *D. radicum*, we analyzed the larval transcriptomes. This revealed that the two *Delia* species activate different gene sets upon ITC exposure. Via 16S amplicon sequencing, we identified that the two species have different GBCs. When the larvae were fed ITC-enriched diets, their GBC composition shifted. Thus, we concluded that these specialist larvae and their GBC combine general and compound-specific ITC detoxification mechanisms with differing efficacies and substrate preferences. Further studies should reveal the relevance of the GBC detoxification capacity for larval performance, and whether other GSL-related defenses, such as glucobrassinins, also play a role in *Delia* resistance.

Keywords: Root defense, Host plant adaptation, Brassicaceae, Genomics, Transcriptomics

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S6-O71

Bottom-up and top-down induced plant responses in wild lima bean plants and their consequences for plant fitness and trophic interactions

Betty Benrey[1], Maximilien Cuny[2], Carlos Bustos-Segura[1]

[1]*University of Neuchatel, Neuchatel, Switzerland.* [2]*Wageningen University, Wageningen, Netherlands*

It is increasingly recognized that plants play a pivotal and active role in determining the performance and community structure of associated organisms, in particular through the production of inducible defense compounds. By causing chemical changes in plants even organisms that never physically encounter one another can influence each other's fitness. We investigated the wider consequences of leaf and seed herbivores and their parasitoids on the performance of lima bean plants and the consequences for different insect guilds and trophic levels. In wild populations, early induced changes in the plants altered the quality of seeds and affected late season herbivores and parasitoids and these effects are carried over across plant generations. In turn, we found that seeds often can survive the attack by pre-dispersal seed beetles, but their performance and defense chemistry is compromised. Yet, some of these effects were dramatically attenuated by parasitism, resulting in a net increase of the number of viable seed offspring. Our findings confirm the long-lasting consequences of indirect plant-mediated interactions in a community-wide ecological context and the importance of parasitoids for plant fitness.

Keywords: Plant-mediated interactions, Parasitoids, Induced defenses, Plant fitness, Multitrophic interactions.

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S6-O70

Constitutive and herbivore-induced volatiles emitted by wild cotton populations

Marine Mamin[1], Mary V Clancy[1], Galien Flückiger[1], Teresa Quijano-Medina[2], Biinza Pérez Niño[2], Luis Abdala-Roberts[2], Ted CJ Turlings[1], Carlos Bustos-Segura[1]

[1]University of Neuchâtel, Institute of Biology, Fundamental and Applied Research in Chemical Ecology, Neuchâtel, Switzerland. [2]Departamento de Ecología Tropical, Campus de Ciencias Biológicas y Agropecuarias, Universidad Autónoma, Mérida, Mexico

Upland cotton (*Gossypium hirsutum*) constitutively stores volatile terpenes in glands present on all its tissues. These stored compounds are emitted at relatively low rates by healthy plants, however herbivory results in an abrupt release of their contents and triggers an increase of their concentration in leaves. Moreover, herbivory induces the delayed systemic emission of another pool of terpenes and other classes of compounds, which are synthetized *de novo*. Although VOC emissions have been characterized in cultivated cotton, less is known about wild cotton populations and their intraspecific variation. Such insight may help in the development of future pest-management tools for cotton crops. In this study, we explored how emissions are influenced by variation in the stored VOCs of wild cotton and how herbivory by the beet armyworm (*Spodoptera exigua*) alters both emitted and stored profiles. Plantlets were grown in a common environment from seeds collected in 16 wild populations distributed along the Yucatán coast (Mexico), which is where *G. hirsutum* was domesticated. Stored VOCs were analyzed using leaf extracts from all plants and emitted VOCs by sampling the headspace of a subset of individuals. By comparing the two analyses, we could identify which compounds were likely released from storage, and which were likely synthesized *de novo*. Most of the variation in the composition of VOC emissions from wild cotton was related to stored compounds characterized by particularly low inducibility. In contrast, the highly inducible compounds, found among those likely *de novo* synthesized accounted for less variation. Our results highlight how the scope of intraspecific variation in emitted compounds depends on the pools from which the VOCs are emitted.

Keywords: *Gossypium hirsutum*, Wild cotton, VOC emissions, HIPVs, Intraspecific variation

S6-O189

Understanding host-parasitoid interactions for application in integrated pest management

Radhika Venkatesan[1,2], Enakshi Ghosh[3], Aswathi Sasidharan[1]

[1]*Indian Institute of Science Education and Research Kolkata, Mohanpur, India.* [2]*National Centre for Biological Sciences, Tata Institute of Fundamental Research, Bangalore, India.* [3]*Graduate Degree Program in Ecology and Department of Agricultural Biology, Colorado State University, Fort Collins, CO 80523-1177, USA, Fort Collins, USA*

Insect parasitoids are fascinating systems to study ecological interactions and are of high economic importance as biocontrol agents. Though free-living as adults, parasitoids need to feed on a single host to reach adulthood. For this, they develop either inside or outside the body of the host that is first paralyzed and ultimately killed. Ecto-parasitoids inject maternal factors that cause permanent paralysis before oviposition making them effective natural enemies for pest management. *Bracon brevicornis* (Wesmael) (Hymenoptera: Braconidae) is a polyphagous, gregarious parasitoid and is known to attack many insect larvae. Despite reports on the biology, larval parasitoids are rarely used in pest management owing to the difficulty in rearing as well as existing knowledge gaps about their behavior and physiology. Half of the Noctuid genus *Spodoptera* (Lepidoptera) are considered serious pests worldwide, commonly referred to as 'armyworms'. *S. litura* (Fabricus), is widely distributed throughout the tropics. *S. frugiperda* is native to North America and later spread to Africa and Asia. It is known to attack different crop plants with a preference for maize. Monophagous insects like *Plutella xylostella* are also serious pests due to their ability to detoxify plant defenses. Due to the high resistance, chemical control of these pests is seldom successful highlighting the need for more environment-friendly biocontrol strategies. Here, a comprehensive study on parasitoid biology is presented, with potential application in pest management.

Keywords: Larval parasitoid, Insect host, Generalist herbivore, Specialist herbivore, *Bracon brevicornis*

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S6-O127

Can plant silicon defenses play a part in biocontrol of herbivorous arthropods?

Tarikul Islam[1,2], Sidra Anwar[1], Christopher Cazzonelli[1], Ben Moore[1], Scott Johnson[1]

[1]*Hawkesbury Institute for the Environment, Western Sydney University, Penrith, NSW, Australia.* [2]*Department of Entomology, Faculty of Agriculture, Bangladesh Agricultural University, Mymensingh, Bangladesh*

Silicon (Si) accumulation in plants is considered a potent physical defense against herbivorous arthropods, particularly against chewing insects. Si deposition makes plant tissues harder and abrasive, decreasing their masticability and digestibility to chewers. Recently, Si has been shown to influence plant indirect defenses (i.e., those involving the natural enemies of herbivores), although the underlying mechanism behind this remains uncertain. Furthermore, it remains to be investigated how feeding on Si-supplemented plants impacts insect susceptibility to their natural enemies. Using French bean, *Phaseolus vulgaris*, and the two-spotted spider mite, *Tetranychus urticae*, we demonstrated that Si supplementation of plants altered the emission of herbivore-induced plant volatiles and thus promoted the attraction of the predatory mite, *Phytoseiulus persimilis*. Furthermore, using the model grass, *Brachypodium distachyon*, and the global insect cotton bollworm (*Helicoverpa armigera*), we provide novel evidence that plant Si defences could affect the anti-predator defences of the cotton bollworm, including its integument resistance and cryptic colouration. Precisely, feeding on Si-supplemented plants disabled larval cryptic colouration by inhibiting carotenoid sequestration in the haemolymph. We suggest that the lower integument resistance of insects when feeding on Si-supplemented plants could contribute to their vulnerability to natural enemies and that potential reduction in the degree of crypsis can make insects more detectable to visually hunting predators (e.g., birds, spiders) and enhance their likelihood of being attacked and killed. Thus, our results indicate that plant Si defenses could play a role in pest biocontrol by promoting natural enemy attraction and by increasing insect susceptibility to their natural enemies.

Keywords: Silica, Natural enemy attraction, Tri-trophic interactions, Insect anti-predator defenses, Insect crypsis

Symposium 6: Chemical and molecular ecology of plant-herbivore-enemy interactions (2)

Thursday, August 11th
10:45 am - 12:30 pm

Session moderated by Ricardo Machado

Competing beetles attract egg laying in a hawkmoth
Jin Zhang

Role of plant-growth promoting fungi in mediating response of an egg parasitoid to plants induced by stink bug egg deposition
Tugcan Alinc, Antonino Cusumano, Salvatore Guarino, Ezio Peri, Livio Torta, Stefano Colazza

Cd exposure through *Hyphantria cunea* pupae reduces the parasitic fitness of parasitic wasp *Chouioia cunea*: A potential risk to the biocontrol efficiency
Dun Jiang, Shanchun Yan

Metabolic exploitation of phytochemical signals mediates *Ips typographus* dispersal and species interaction in spruce forests
Jiaxing Fang, Huicong Du, Fu Liu, Sufang Zhang, Zhen Zhang, Xiangbo Kong

S6-O50

Competing beetles attract egg laying in a hawkmoth

Jin Zhang

Nanjing Agricultural Univeristy, Nanjing, China

In nature, plant-insect interactions occur in complex settings involving multiple trophic levels, often with multiple species at each level. Herbivore attack of a host plant typically dramatically alters the plant's odor emission in terms of concentration and composition. Therefore, a well-adapted herbivore should be able to predict whether a plant is still suitable as a host by judging these changes in the emitted bouquet. Although studies have demonstrated that oviposition preferences of successive insects were affected by previous infestations, the underlying molecular and olfactory mechanisms remain unknown. Here, we report that tobacco hawkmoths (*Manduca sexta*) preferentially oviposit on Jimson weed (*Datura wrightii*) that is already infested by a specialist, the three-lined potato beetle (*Lema daturaphila*). Interestingly, the moths' offspring do not benefit directly, as larvae develop more slowly when feeding with *Lema* beetles. However, one of *M. sexta*'s main enemies, the parasitoid wasp *Cotesia congregata*, prefers the headspace of *M. sexta*-infested plants to that of plants infested by both herbivores. Hence, we conclude that female *M. sexta* ignores the interspecific competition with beetles and oviposit deliberately on beetle-infested plants to provide their offspring with an enemy-reduced space, thus providing a tradeoff that generates a net benefit to the survival and fitness of the subsequent generation. We identify α-copaene, emitted by beetle-infested *Datura*, plays a role in this preference. By performing heterologous expression and single sensillum recordings, we show that odorant receptor (Or35) is involved in α-copaene detection.

Keywords: *Manduca sexta*, Odorant, Parasitoid, Oviposition, Behavior

[1] Zhang J, et al. Curr. Biol 32, 861 - 869

S6-O63

Role of plant-growth promoting fungi in mediating response of an egg parasitoid to plants induced by stink bug egg deposition

Tugcan Alinc[1], Antonino Cusumano[1], Salvatore Guarino[2], Ezio Peri[1], Livio Torta[1], Stefano Colazza[1]

[1]*Department of Agricultural, Food and Forest Sciences, University of Palermo, Palermo, Italy.* [2]*Institute of Biosciences and Bioresources (IBBR), National Research Council of Italy (CNR), Palermo, Italy*

During the host location process, egg parasitoids rely on various chemical stimuli such as oviposition induced plant volatiles (OIPVs) which are emitted by plants in response to egg deposition by herbivore insects. To date, research has mainly focused on the underlying mechanisms of this chemical communication in the context of tritrophic interactions involving the plants, herbivores and associated egg parasitoids. Nonetheless, no information so far is available on how beneficial soil microbes modulates egg parasitoids' attractions towards plants induced by insect oviposition. To fill this gap, we investigated the effect of plant growth promoting fungi, *Trichoderma harzianum* strain T22 in multitrophic interactions by using a model system consisting of tomato plants, southern green stink bug, *Nezara viridula* and an egg parasitoid, *Trissolcus basalis*. The results showed that the egg parasitoid behavior was influenced by *T. harzianum* T22 inoculation as *T. basalis* did show a significant attraction towards OIPVs emitted from inoculated plants over OIPVs emitted by non-inoculated plants. Chemical analyses of volatiles are being conducted to check for differences on chemical blend of OIPVs between differently treated tomato plants in the attempt to link plant VOC emission with parasitoid attraction. The results will be discussed in the context of chemical perspective for better understanding the synergic interactions between stink bugs and beneficial soil microbe on the behavior of egg parasitoids.

Keywords : *Trichoderma harzianum*, Multitrophic interactions, *Nezara viridula*, *Trissolcus basalis*

S6-O132

Cd exposure through *Hyphantria cunea* pupae reduces the parasitic fitness of parasitic wasp *Chouioia cunea*: A potential risk to the biocontrol efficiency

Dun Jiang, Shanchun Yan

Northeast Forestry University, Harbin, China

Heavy metal pollution can be regarded as an environmental variable affecting the biological control efficiency of pests. However, the previous studies mainly focus on entomopathogenic microorganisms and insect predators, and lack of studies on the parasitic fitness of parasitic wasps under heavy metal stress. In the present study, the effect of Cd exposure through the pupa of *H. cunea* on the parasitic fitness of *C. cunea* was investigated. Furthermore, we also explored the mechanism by which Cd exposure affects the interaction between *H. cunea* and *C. cunea* from the perspective of innate immunity in host insect and oxidative damage in the parasitoid offspring. Our results found that the Cd could be transferred from the *H. cunea* pupae to the parasitoid offspring, and the transfer coefficient was a kind of biological amplification effect. There was no significant difference in the rate of parasitism success and offspring emergence between un-treated and Cd-accumulated environments. However, after parasitizing the pupae with Cd accumulation, the parasitic fitness of offspring wasps was decreased significantly. Cd exposure significantly reduced the innate immunity of host insect, as evident by a significantly decrease of the total hemocyte count, phagocytic activity and melanization in pupae of *H. cunea*. Compared with the control, the H2O2 content of parasitoid offspring in the Cd-treated group was significantly increased. Cd exposure significantly inhibited the SOD activity in parasitoid offspring, but the contents of ASA and GSH in parasitoid offspring were significantly increased by Cd stress. Taken together, Cd exposure reduces the cyclic utilization efficiency of *C. cunea* on *H. cunea* pupae. The direct adverse effects of Cd exposure on parasitoid offspring, such as oxidative damage, were the main reason for its reduction in parasitic fitness to Cd-accumulated *H. cunea* pupae.

Keywords: Heavy metal, Parasitic wasp, Parasitic fitness, Oxidative damage

S6-O192

Metabolic exploitation of phytochemical signals mediates *Ips typographus* dispersal and species interaction in spruce forests

Jiaxing Fang, Huicong Du, Fu Liu, Sufang Zhang, Zhen Zhang, Xiangbo Kong

Key Laboratory of Forest Protection of National Forestry and Grassland Administration, Ecology and Nature Conservation Institute, Chinese Academy of Forestry, Beijing, China

The closely related Ips bark beetles, *I. typographus* and *I. subelongatus*, are major pests of coniferous forests in eastern North China. In mixed *Picea spp*. and *Larix spp*. forests, *I. subelongatus* can attack non-historic host trees such as *P. koraiensis*, but a stable population of *I. subelongatus* cannot establish in spruce forests. We hypothesized that phytochemicals might drive the complex and interspecific interactions in the spruce forest ecosystem. We found *Ips subelongatus* is more aggressive than *I. typographus* because it develops well in both spruce and larch, whereas *I. typographus* damages only spruce but not larch. The release of the key host α-pinene is higher in spruce than in larch, which has a positive effect on *I. typographus* and negative effect on *I. subelongatus*. The metabolic profiles of enantiomeric α-pinene in *I. typographus* were identified by the isotope tracking method, and the juvenile hormone III may increase the efficiency of α-pinene conversion in vivo. The ecological functions of key α-pinene metabolites were investigated for intraspecific and interspecific regulations of *I. typographus*, *I. subelongatus*, and *Thanasimus substriatus*. The key metabolites S-(–)-cis-verbenol and R-(–)-trans-verbenol showed significant repellent effects on *I. subelongatus*, suggesting that the native species *I. typographus* may use host monoterpenes to disrupt invasion of *I. subelongatus* in spruce forests. The clerid predator *T. substriatus* uses aggregation pheromones from two Ips bark beetles as prey signals, suggesting that *I. subelongatus* cannot escape the clerid predator in its new habitat. Our results show the cascading transmission of host phytochemical signals in the "spruce-*I. typographus* and *I. subelongatus*- clerid predator" system mainly mediates the spread of native species and suppresses the invasion of aggressive species, which improves our understanding of the driving forces of chemical signals in the process of coadaptation interactions.

Keywords: Ips bark beetle, Phytochemical signal, Sympatric species, Cascading effect, Ecological interactions

Symposium 6: Chemical and molecular ecology of plant-herbivore-enemy interactions (3)

Friday, August 12th
2:30 pm - 3:10 pm

Session moderated by Ricardo Machado

The polyvalent sequestration ability of an economically important beetle
Carla C. M. Arce, Ricardo A. R. Machado, Marine Mamin, Gaetan Glauser, Pamela Bruno, Betty Benrey, Matthias Erb, Christelle Robert, Ted C. J. Turlings

Molecular and chemical regulators of foraging behavior of root herbivores
Ricardo A. R. Machado

S6-O21

The polyvalent sequestration ability of an economically important beetle

Carla C. M. Arce[1], Ricardo A. R. Machado[2], Marine Mamin[1], Gaetan Glauser[3], Pamela Bruno[1], Betty Benrey[4], Matthias Erb[5], Christelle Robert[6], Ted C. J. Turlings[1]

[1]*University of Neuchâtel, Institute of Biology, FARCE laboratory, Neuchâtel, Switzerland.* [2]*University of Neuchâtel, Institute of Biology, Experimental Biology Group, Neuchâtel, Switzerland.* [3]*University of Neuchâtel, Neuchâtel Platform of Analytical Chemistry, Neuchâtel, Switzerland.* [4]*University of Neuchâtel, Institute of Biology, E-vol laboratory, Neuchâtel, Switzerland.* [5]*University of Bern, Institute of Plant Sciences, BIN group, Bern, Switzerland.* [6]*University of Bern, Institute of Plant Sciences, Chemical Ecology group, Bern, Switzerland*

Many herbivorous insects sequester defensive secondary metabolites from their host plant and use them against natural enemies. While this phenomenon is well documented, it is still unknown whether polyphagous herbivores are able to sequester different defensive metabolites from their varying host plants, and whether or not, they can transfer them to eggs as a defensive mechanism. We studied this for the beetle *Diabrotica virgifera virgifera* feeding on maize, cucumber, cabbage, or beans, which respectively produce benzoxazinoids, cucurbitacins, glucosinolates, and cyanogenic glucosides as a chemical defense. Chemical analyses revealed that *D. v. virgifera* beetles sequester and transfer benzoxazinoids, cucurbitacins and glucosinolates, but not cyanogenic glucosides, to their eggs, showing the extraordinary polyvalent sequestration abilities of this insect pest. To test whether the sequestered toxins protect eggs against predators, we fed *D. v. virgifera beetles* with toxin-free or toxin-containing plants and offered their eggs to the predators *Dalotia coriaria* and *Orius laevigatus*. In choice experiments, both predators consumed more toxin-free eggs than toxin-containing eggs. Moreover, survival assays confirmed the toxic effects of benzoxazinoid-containing and glucosinolate-containing eggs on the predators, but, surprisingly, cucurbitacins had no apparent effect. Our results reveal a unique ability of *D. v. virgifera* to use multiple plant defensive chemicals against higher trophic levels, which may in part explain the extraordinary success of this invasive pest.

Keywords: Biological control, Maize, Predators, Progeny; Western Corn Rootworm.

S6-O35

Molecular and chemical regulators of foraging behavior of root herbivores

Ricardo A. R. Machado

Experimental Biology, Institute of Biology, University of Neuchâtel, Neuchatel, Switzerland

Belowground herbivore insects use different olfactory and gustatory chemical cues to locate their host plant and optimize their foraging decisions in complex soil environments. The molecular mechanisms on how root herbivores perceive such chemical cues is poorly understood. In this study, we identified odor and gustatory receptors of *Diabrotica virgifera virgifera*, the western corn rootworm, silenced their expression through RNA interference and conducted behavioral experiments to determine the importance of these receptors for host location and foraging. Our results uncover two genes, the putative carbon dioxide group 2 receptor (*DvvGr2*) and the putative gustatory sugar-receptor 43a (*DvvGr43a*), as important regulators of western corn rootworm foraging behavior. *DvvGr2*-silenced larvae do not behaviorally respond to plant-derived carbon dioxide. By consequence, their capacity to locate their host plants was reduced. *DvvGr43a*-silenced larvae do not respond to sugars as phagostimulants. By consequence, they had a reduced capacity to recognize maize roots as a food source and to discriminate between nutritious crown roots and less nutritious primary roots, thus demonstrating the importance of sugar perception for *D. v. virgifera* foraging success. Taken together, our study uncovers that the capacity to detect different root volatile- and non-volatile metabolites is important for host location and foraging of a specialist root herbivore. Modulating olfactory and gustatory perception through environmental RNAi is a promising approach to understand how essential plant metabolites that cannot be manipulated *in planta* determine foraging decisions by belowground herbivores and plant-herbivore interactions.

Keywords: Foraging behavior, Root herbivores, Carbon dioxide, Sugars, RNAi

[1] Arce CCM, Theepan V, Schimmel BCJ, Jaffuel G, Erb M, Machado RAR (2021) eLife 10, e65575.
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Symposium 7: Chemical communication and biological invasion (1)

Thursday, August 11th
8:45 am - 12:30 pm

Session moderated by Lilin Zhao

Chemical communications between pinewood nematode and its vector insect
Lilin Zhao

Lysine provisioning by horizontally acquired genes facilitates the fitness of the invasive whitefly
Jun-Bo Luan

Gypsy moth (*Lymantria dispar*) on the Far East of Russia
Dmitry Kurenshchikov

Rapid assessment of Cerambycid beetle biodiversity in a tropical rainforest in Yunnan Province, China, using a multicomponent pheromone lure: implications for invasive species monitoring
Jacob D Wickham, Rhett D Harrison, Wen Lu, Lawrence M Hanks, Jocelyn G Millar

Monoterpenoid signals and their transcriptional responses to feeding and juvenile hormone regulation in bark beetle *Ips hauseri*
Xiangbo Kong, Jiaxing Fang, Sufang Zhang, Fu Liu

RNA interference of key olfactory and visual genes during adult stage of *Agrilus planipennis*
Sufang Zhang, Zhizhi Fan, Xiangbo Kong, Zhen Zhang, Xun Zhang

Screening of potential RNAi targets and improving interference effect in *Hyphantria cunea*
Xun Zhang, Zhizhi Fan, Sufang Zhang, Xiangbo Kong, Fu Liu, Zhen Zhang

Identification of odorant binding proteins which binding with methyl eugenol and their function research in *Bactrocera dorsalis*
Xiaofeng Chen

Functional analysis of pheromone receptor repertoire in the fall armyworm, *Spodoptera frugiperda*
Hao Guo, Xin-Lin Gong, Guo-Cheng Li, Bao-Tong Mo, Nan-Ji Jiang, Ling-Qiao Huang, Chen-Zhu Wang

Electroantennographic and field responses of male *Hyphantria cunea* (Drury) to synthetic sex pheromone components
Yishu Geng

S7-O134

Chemical communications between pinewood nematode and its vector insect

Lilin Zhao

State Key Laboratory of Integrated Management of Pest Insects and Rodents, Institute of Zoology, Chinese Academy of Sciences, Beijing, China

Pinewood Nematode (PWN) causes little damage to pine trees in North America. However, upon introduction to non-native habitats, first to Japan in 1905, then to China in 1982, South Korea in 1988, Portugal in 1999, and Spain in 2008, PWN has caused high mortality of the native pine trees. PWN engages in a symbiotic partnership with its insect vector, the *Monochamus* beetle, as well as associated bacteria and ophiostomatoid fungi, in order to successfully infect and kill its host pine trees. Here we focused on the interspecific communication between PWN and its associated partners, and the potential role of this communication in promoting pathogenicity and invasiveness of PWN. The study of communication between PWN and its vector involved methods of field investigation, simulation experiment in laboratory, metabolites and pheromones analysis with GC-MS and LC-MS/MS, 13C stable isotope labeling experiment, bioassays and gene analysis. The complex interspecific interactions within the PWD system, as well as the availability of the PWN genome, make this system a good model for the study of invasive species coevolution with associated biota. With the increasing number of identified chemical signals, further discoveries regarding chemical communication may enable the development of methods to disrupt the development and behavior of PWN and its vector in the field. We describe the chemical and molecular signals positively influencing the survival, reproduction, and spread of PWN. Knowledge of these signals could potentially be used to interfere with the proliferation and dispersal of PWN.

Keywords: Pine wilt disease, Ascarosides, Monochamus beetle, symbiotic microorganism

[1] Zhao, L., M. Mota, P. Vieira, R. A. Butcher and J. Sun (2014) Trends in Parasitology 30(6): 299-308.

S7-O190

Lysine provisioning by horizontally acquired genes facilitates the fitness of the invasive whitefly

Jun-Bo Luan

Shenyang Agricultural University, Shenyang, China

Horizontal gene transfer is widespread in insects housing intracellular symbionts. However, the role of horizontally transferred genes (HTGs) in insect-symbiont interactions remains largely unknown. We found symbiont *Hamiltonella* maintains a nearly complete lysine synthesis pathway, while *Portiera* and *Rickettsia* require the complementation of whitefly HTGs for lysine synthesis in *Bemisia tabaci* MEAM1. We demonstrated that *Hamiltonella* did not alter the titers of *Portiera* and Rickettsia or lysine gene expression of *Portiera*, *Rickettsia* and whiteflies. *Hamiltonella* also did not impact on lysine levels or protein localization in whiteflies. Silencing whitefly lysA in whiteflies harboring or lacking *Hamiltonella* reduced lysine levels, adult fecundity and titers of *Portiera* and *Rickettsia*. Therefore, we demonstrated an essential amino acid lysine synthesized through HTGs is important for whitefly reproduction and fitness of both obligate and facultative symbionts, and it illustrates the mutual dependence between whitefly and its two symbionts. We further found that the vector 2mDNA1, an engineered begomovirus transmitted by *B. tabaci*, was effective for silencing *B. tabaci* MED lysine HTGs in the lab. The 2mDNA1-silencing lysine HTG reduced levels of lysine, titers of *Portiera* in both F0 and F2 whiteflies as well as the survival, fecundity, and population increases of whiteflies. We also revealed that the vector 2mDNA1 can be used to silence whitefly HTGs and inhibit whitefly performance in the greenhouse. Thus, we demonstrate that repressing the expression of insect lysine HTGs though a modified virus is feasible for the control of phloem feeding insect pests.

Keywords: Horizontally transferred genes, Lysine, Symbiosis, Whitefly, Invasion

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S7-O138

Gypsy moth (*Lymantria dispar*) on the Far East of Russia

Dmitry Kurenshchikov

Institute of Water and Ecology Problems of the Far Eastern Branch of the Russian Academy of Sciences, (IWEP FEB RAS), Khabarovsk, Russian Federation

The gypsy moth (*Lymantria dispar* L.) is one of the most aggressive defoliators in the temperate natural zone of the northern hemisphere. Species shows differences in biology in different parts of its areal. In the Russian Far East, caterpillars hatching from their eggs in early May, at the same time as Mongolian oak (*Quercus mongolica*), the preferred fodder species, leaf appear. Gypsy moth females are capable of active flight and after mating, the female lays one laying of eggs, which is placed on the underside of the leaves of deciduous tree species. Together with such leaves, egg layings fall into the forest litter, where they spend the winter. The flight of insects begins in the first decade of July and ends at the end of August. Long-term studies of gypsy moth populations using disparlure pheromone traps have shown that the maximum flight activity of males occurs on July 23-27. During outbreaks, about 1,000 males fall into one pheromone trap. The populations of the Far East are characterized by 3-4 years outbreak. The main factor in decreasing population is the nuclear polyhedrosis virus, which forms a viral (less often bacterial-viral) epizootic. Death from virosis occurs mainly in caterpillars of the second-fourth instars. *Lymantria dispar* L. (Asian race) is included in the List of quarantine objects: Part 2. Quarantine objects limitedly distributed on the territory of the Russian Federation.

Keywords: Gypsy moth, *Lymantria dispar*, Epizootic, Pathogens, Pheromone traps.

[1] Kurenshchikov DK, Imranova EL, Martemyanov VV (2020) Features of the Far Eastern Gypsy Moth (*Lymantria dispar* L.) Population Outbreak. Contemporary Problems of Ecology. Vol. 13. №2: 172-179.

S7-O152

Rapid assessment of Cerambycid beetle biodiversity in a tropical rainforest in Yunnan Province, China, using a multicomponent pheromone lure: implications for invasive species monitoring

Jacob D Wickham[1,2,3], Rhett D Harrison[4], Wen Lu[5], Lawrence M Hanks[6], Jocelyn G Millar[7]

[1]A.N. Severstov Institute of Ecology and Evolution, Russian Academy of Sciences, Moscow, Russian Federation. [2]Institute of Zoology, Chinese Academy of Sciences, Beijing, China. [3]Department of Entomology, Rutgers University, New Brunswick, USA. [4]World Agroforestry, East and Southern Africa Region, Lusaka, Zambia. [5]Department of Plant Protection, College of Agriculture, Guangxi University, Nanning, China. [6]Department of Entomology, University of Illinois Urbana-Champaign, Urbana, USA. [7]Department of Entomology, University of California, Riverside, USA

The Cerambycidae, commonly known as longhorned beetles, comprise a large family of woodborers with >35,000 described species in eight subfamilies. The endophytic larvae develop in woody plants, and while some species have become globally important as invasive forest pests, they generally perform valuable ecosystem services by initiating the degradation of woody plants and can be indicators of forest ecosystem health. The purpose of this study was to examine the effectiveness of a generic lure as a potential monitoring tool for both biodiversity assessment and potential invasive species. Working in a subtropical forest in southwest China, we set traps baited with generic lures at ground level (1 m) and canopy height (~18 m) across 22 randomly located forest plots (12 regenerating forest, 10 mature forest). Three stations were established per plot and each plot was trapped for 7 days in May–June 2013. In total, 4541 beetles of 71 species were caught, including 26 species with 10 or more individuals. We used Hierarchical Modeling of Species Communities (HMSC) to analyze the data and produced informative models for 18 species, showing that trap height, slope, elevation, and leaf-area index were important determinants of cerambycid distribution. Our results demonstrate the potential for using generic lures to detect and monitor cerambycid populations, both for regulatory purposes and for the study of cerambycid beetle ecology. Further research should focus on refining lure blends, and on repeated sampling to determine temporal and spatial dynamics of cerambycid communities.

Keywords: Pheromone, Cerambycidae, Invasive species monitoring, Biodiversity survey

[1] Wickham JD, Harrison RD, Lu W, Hanks LM, Millar, J.G. (2021). Rapid Assessment of Cerambycid Beetle Biodiversity in a Tropical Rainforest in Yunnan Province, China, Using a Multicomponent Pheromone Lure. Insects 12: 277

S7-O193

Monoterpenoid signals and their transcriptional responses to feeding and juvenile hormone regulation in bark beetle *Ips hauseri*

Xiangbo Kong, Jiaxing Fang, Sufang Zhang, Fu Liu

Ecology and Nautre Conservation Institute, Chinese Academy of Forestry, Beijing, China

Hauser's engraver beetle, *Ips hauseri*, is a serious pest in spruce forest ecosystems in Central Asia. Its monoterpenoid signal production, transcriptome responses and potential regulatory mechanisms remain poorly understood. The quality and quantity of volatile metabolites in hindgut extracts of *I. hauseri* were found to differ between males and females and among three groups: beetles that were newly emerged, those with a topical application of juvenile hormone III (JHIII) and those that had been feeding for 24 h. Feeding males definitively dominated monoterpenoid signal production in *I. hauseri*, which uses (4S)-(−)-ipsenol and (S)-(−)-cis-verbenol to implement reproductive segregation from *Ips typographus* and *Ips shangrila*. Feeding stimulation induced higher expression of most genes related to the biosynthesis of (4S)-(−)-ipsenol than JHIII induction, and showed a male-specific mode in *I. hauseri*. JHIII stimulated males to produce large amounts of (−)-verbenone and upregulated the expression of several CYP6 genes, to a greater extent in males than in females. The expression of genes involved in the metabolism of JHIII in females and males was also found to be upregulated. Our results indicate that a species-specific aggregation pheromone system for I. hauseri, consisting of (4S)-(−)-ipsenol and S-(−)-cis-verbenol, can be used to monitor population dynamics or mass trap killing. Our results also enable a better understanding of the bottom-up role of feeding behaviors in mediating population reproduction/aggregation and interspecific interactions.

Keywords: Bark beetle, Chemical communication, Species interactions, (4S)-(−)-Ipsenol, Transcriptome analysis

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[2] Seybold SJ, Quilici DR, Tillman JA, Vanderwel D, Wood DL, Blomquist GJ (1995) PNAS 92: 8393-8397
[3] Zhang QH, Song LW, Ma JH, Han FZ, Sun JH (2009) Chemoecology 19: 203-210

S7-O188

RNA interference of key olfactory and visual genes during adult stage of *Agrilus planipennis*

Sufang Zhang, Zhizhi Fan, Xiangbo Kong, Zhen Zhang, Xun Zhang

Ecology and Nature Conservation Institute, Chinese Academy of Forestry, Beijing, China

The emerald ash borer, *Agrilus planipennis* (Coleoptera: Buprestidae) is an international quarantine wood-boring pest. The larvae feed in phloem, cambial regions and shallow xylem of the ash trees, which is highly concealed. It is difficult to touch and kill the larvae with conventional insecticides, so the adult stage is critical for their control. Both olfactory and visual related genes play a decisive role in feeding, mating and oviposition process of adult *A. planipennis*. In this work, the olfactory and visual genes were identified in the adult *A. planipennis*, and the functions of key olfactory and visual genes were studied using RNAi technology, to explore the molecular prevention and control strategies. First, this study compared the head (including antennae) tissue transcriptomes of newly emerged and sexually mature *A. planipennis*. According to the sequencing results, the olfactory and visual related genes were screened and identified, and the expression levels of these chemosensory genes and opsin genes were analyzed between different growth stages (newly emerged stage and sexually mature stage) and different genders (male and female). Then, some of the differentially expressed genes were selected for RNAi. Real-time quantitative PCR was used to determine the expression change of the related genes after interference, so as to clarify the RNAi effect of the target genes. Color sensitivity tests were performed on adults after RNAi of visual genes with significant interference effects. This research explained the important roles of chemosensory and opsin genes in the life of *A. planipennis* from a molecular perspective, which laid a foundation of using RNAi technology to control *A. planipennis* in the future and provided a new idea for the control of other non-model insects.

Keywords: -

S7-O195

Screening of potential RNAi targets and improving interference effect in *Hyphantria cunea*

Xun Zhang, Zhizhi Fan, Sufang Zhang, Xiangbo Kong, Fu Liu, Zhen Zhang

Chinese Academy of Forestry, Beijing, China

The fall webworm, *Hyphantria cunea*, belonging to Arctiidae of Lepidoptera, is a worldwide invasive pest causing serious ecological and economic damage. The control of *H. cunea* is challenged by insecticide resistance, expensive sex pheromone traps, and negative impact on ecology. In recent years, RNAi-based pest management has been increasingly studied as a novel insect control strategy. However, the identification of efficient RNAi target genes remains a major challenge as genomic tools and RNAi efficiency is limited in most Lepidoptera insects impeding the RNAi-based pest management in *H. cunea*. In the present study, we identified three highly efficient RNAi targets (HcvATPase, HcIap and HcRop) from 20 randomly screened genes by dsRNA injection method or feeding *Escherichia coli* expressing dsRNAs. Results show that HcvATPase caused 74.07% and 66.07% mortality rate on *H. cunea* larvae after injection and ingestion of dsRNA, respectively. HcIap and HcRop respectively led to 82.14% and 65.48% mortality rate on *H. cunea* larvae after dsRNA ingestion. Our results indicate that these genes could be excellent candidate genes for RNAi-mediated pest management of *H. cunea*. In addition, to improve the interference effect in *H. cunea*, we identified and characterized four new dsRNA nuclease (dsRNase) genes from *H. cunea*, which were named HcdsRNase1, HcdsRNase2, HcdsRNase3, and HcdsRNase4. Silencing HcdsRNase3 or HcdsRNase4, or co-knockdown HcdsRNase3 and HcdsRNase4 could significantly increase RNAi efficiency. These results demonstrate that the RNAi efficiency is certainly impaired by nuclease activity in *H. cunea*. Thus, screening of highly effective target genes and overcome obstacles such as dsRNases are expected to help implement RNAi-based technologies for *H. cunea* control.

Keywords: RNAi, *Hyphantria cunea*, dsRNase, Pest control

S7-O61

Identification of odorant binding proteins which binding with methyl eugenol and their function research in *Bactrocera dorsalis*

Xiaofeng Chen

Key Laboratory of Entomology and Pest Control Engineering, College of Plant Protection, Chongqing, China. Academy of Agricultural Sciences, Chongqing, China

Olfaction is important in many crucial behaviors in insect, such as finding food, hosts, mates and avoiding predators. Odorant binding proteins (OBPs) binding hydrophobic odorants belongs to the first step of olfactory system. *Bactrocera dorsalis*, is one of the most destructive fruit-eating pest causing enormous economic loss to the fruit and vegetable industry. Methyl eugenol (ME) has been used as male attractant to monitor and eradicate *B. dorsalis* populations for 70 years, but the molecular basis of its activity remains largely unclear. Here, we want to find out the OBPs which are involved in ME perception of *B. dorsalis*. We used microscale thermophoresis to confirm that BdorOBP56f-2 and BdorOBP69a directly bind ME with strong affinity in vitro. We then used CRISPR/Cas9 to knock out the two genes, allowing us to establish homozygous mutant *B. dorsalis* lines. The electroantennogram response and behavioral attraction to ME were significantly reduced in the mutant, providing in vivo evidence that BdorOBP56f-2 and BdorOBP69a are necessary for efficient ME perception. Our results offer insight into the molecular mechanism of ME perception in *B. dorsalis* and provide a theoretical basis for the functional analysis of other OBPs.

Keywords: Odorant-binding protein, Oriental fruit fly, Genome editing, Methyl eugenol

S7-O118

Functional analysis of pheromone receptor repertoire in the fall armyworm, *Spodoptera frugiperda*

Hao Guo, Xin-Lin Gong, Guo-Cheng Li, Bao-Tong Mo, Nan-Ji Jiang, Ling-Qiao Huang, Chen-Zhu Wang

Institute of Zoology, CAS, Beijing, China

The fall armyworm, *Spodoptera frugiperda* (J. E. Smith), is a polyphagous moth species spreading all around the globe. It uses (Z)-9-tetradecenyl acetate (Z9-14: Ac) and (Z)-7-dodecenyl acetate (Z7-12: Ac) (100:3.9) as essential sex pheromone components. However, our understanding of the molecular basis of pheromone detection of *S. frugiperda* is still incomplete. Herein, we identified six PRs, i.e., SfruOR6, 11, 13, 16, 56, and 62, by transcriptome sequencing. Subsequently, we heterologously expressed them in *Drosophila* OR67d neurons and determined their response spectra with a big panel of sex pheromones and analogs. Among them, SfruOR13-expressing neurons strongly respond to the major sex pheromone component, Z9-14: Ac, but also comparably to (Z, E)-9,12-tetradecadienyl acetate (Z9, E12-14: Ac) and weakly to (Z)-9-dodecenyl acetate (Z9-12: Ac). Both SfruOR56 and SfruOR62 are specifically tuned to the minor sex pheromone component, Z7-12: Ac with varying intensities and sensitivities. In addition, SfruOR6 is activated only by Z9, E12-14: Ac, and SfruOR16 by both (Z)-9-tetradecenol (Z9-14: OH) and (Z)-9-tetradecenal (Z9-14: Ald). However, the OR67d neurons expressing SfruOR11 remain silent to all compounds tested, a phenomenon commonly found in the OR11 clade of Noctuidae species. Next, using single sensillum recording, we characterized three sensilla types on the antennae of males, namely A, B, C, and D types that are tuned to the ligands of PRs, thereby confirming that *S. frugiperda* uses both SfruOR56 and SfruOR62 to detect Z7-12:Ac. Last, using wind tunnel assay, we demonstrate that both Z9, E12-14: Ac and Z9-14: OH act as antagonists to the sex pheromone. We have deorphanized five PRs and characterized four types of sensilla responsible for the detection of pheromone compounds, providing insights into the peripheral encoding of sex pheromones in *S. frugiperda*.

Keywords: Pheromone receptors, Sex pheromones, *Drosophila* OR67d neurons, Wind tunnel, Single sensillum recording

S7-O139

Electroantennographic and field responses of male *Hyphantria cunea* (Drury) to synthetic sex pheromone components

Yishu Geng

Nanjing Forestry University, Nanjing, China

Hyphantria cunea (Drury) (Lepidoptera: Arctiidae) is a highly polyphagous insect worldwide and is now a dominant pest in China. Although sex pheromone components of *H. cunea* have been identified, the weak field attraction of synthetic sex pheromone components has hindered the application of sex pheromone-based lures in controlling this pest. In this study, the electroantennographic and field responses of *H. cunea* to three synthetic sex pheromone components, including (9Z,12Z,15Z)-Octadecatrienal(C18:3Ald), (3Z,6Z)-3,6-9,10-epoxyheneicosadiene (C21-2Epo) and (3Z, 6Z)-1, 3, 6-9, 10-epoxyheneicosatriene (C21-3Epo) were evaluated, and the antennae of adult males showed a significant dose-dependent response to three individual components and their compounds. The younger and virgin male moths showed more significant electrophysiological activity to synthetic compounds, male moth antennae were significantly less sensitive to sex pheromones after mating. The field trapping test revealed that the single- and two-component mixtures did not show significant trapping activity. Instead, the highest number of conspecific males can be caught in traps baited with the ternary blend. In addition, the field trapping indicated that the green rubber septum could be a practical dispenser for synthesized compounds. The Unitraps with green rubber septum exhibited more excellent trapping than the cotton wick. Finally, the green rubber septum baited with ternary blend monitored the dynamics of *H. cunea* occurrence and detected emergence period overlapped with the commercially available synthetic lure. In general, the bioactivity of synthetic sex pheromone in *H. cunea* was validated. It could capture *H. cunea* effectively, be a part of an IPM program, and be used to monitor and control *H. Cunea*.

Keywords: *Hyphantria cunea,* Sex pheromone, Electroantennogram, Field trapping

[1] Tariku-T Edosa, Jo Yong-H, Keshavarz Maryam, et al (2019) Current status of the management of fall webworm, *Hyphantria cunea*: Towards the integrated pest management development. Journal of Applied Entomology 143(1-2): 1-10.

Symposium 7: Chemical communication and biological invasion (2)

Friday, August 12th
2:30 pm - 3:10 pm

Session moderated by Lilin Zhao

Identification of an alarm pheromone in the destructive invasive species: the red-necked longhorn beetle, *Aromia bungii*
Ruixu Chen, Cong Chen, Xudong Zhao, Li Chen, Tian Xu, Dejun Hao

Hormetic response and coexpression of cytochrome P450 and cuticular protein reveal the tolerance to hostspecific terpenoid defenses in an emerging insect pest, *Pagiophloeus tsushimanus* (Coleoptera: Curculionidae)
Shouyin Li, Hui Li, Dejun Hao

S7-O131

Identification of an alarm pheromone in the destructive invasive species: the red-necked longhorn beetle, *Aromia bungii*

Ruixu Chen[1,2], Cong Chen[1], Xudong Zhao[1], Li Chen[3], Tian Xu[1], Dejun Hao[1]

[1]Nanjing Forestry University, Nanjing, China. [2]Jiangsu Vocational college of Agriculture and Forestry, Zhenjiang, China. [3]Hebei University, Baoding, China

The red-necked longhorn beetle, *Aromia bungii*, is an invasive wood-boring insect that has invaded into Europe and Japan, seriously damaging stone fruit trees in both native and invaded regions. cis- and trans-Rose oxides and (R)-(+)-citronellal were previously found in volatiles produced by *A. bungii* adults and are both commercially available with low prices. For developing efficient pheromone-based trapping technique to control this pest, the potential synergistic effect of the commercially purchased rose oxide and (±)-citronellal on the field attraction of major male-produced aggregation pheromone (E)-2-cis-6,7-epoxynonenal was evaluated. However, the addition of these chemicals significantly reduced the trap catch number, indicating a repellent effect on the beetles. Y-tube olfactory assays revealed that rose oxide, at a high concentration, was significantly repellent to both sexes, while (±)-citronellal didn't show either attraction or repellency to the beetles, suggesting that rose oxide has a role as alarm pheromone in *A. bungii*. A greater amount of rose oxide was released from the beetles stimulated by shaking, compared to that released from non-stimulated beetles. Rose oxide was further determined to be original from a white-colored secretion which was immediately sprayed by the beetles that were hand captured or squeezed by tweezers. To our knowledge, this was the first identification of an alarm pheromone component in Cerambycidae, which may facilitate the development of novel control strategies, such as push-pull strategy, for *A. bungii*.

Keywords: *Aromia bungii*, Alarm pheromone, Rose oxide, Defensive behavior, Invasive species

[1] Xu T, Yasui H, Teale SA, et al (2017) Identification of a male-produced sex-aggregation pheromone for a highly invasive cerambycid beetle, *Aromia bungii*. Sci Rep 7: 7330.
[2] Wei JR, Liu XB, Niu YL, et al (2003) Identification of volatiles released from the living adult *Aromia bungii* Faldermann. Forest Pest and Disease 32: 8-10.

S7-O130

Hormetic response and coexpression of cytochrome P450 and cuticular protein reveal the tolerance to hostspecific terpenoid defenses in an emerging insect pest, *Pagiophloeus tsushimanus* (Coleoptera: Curculionidae)

Shouyin Li, Hui Li, Dejun Hao

Nanjing Forestry University, Nanjing, China

Camphor oil (EO) and its main component (i.e., D-camphor) form specific terpenoid defenses in camphor trees, *Cinnamomum camphora*. However, an emerging insect pest, *Pagiophloeus tsushimanus* (Coleoptera: Curculionidae) has recently caused serious damage to this intractable plant species, which is largely elusive. To investigate the mechanism underlying the tolerance of this weevil to host-specific terpenoid defenses, we carried out the bioassays, RNA-seq, and RT-qPCR analysis based on a simulated diet environment with EO or D-camphor exposure. First, a hormetic response (a highly generalized dose-response phenomenon in toxicology) in the larval overall performance was observed in terpenoid-feeding individuals. Then, genes encoding cytochrome P450 (CYP450) and cuticular protein (CP) were induced by both EO and D-camphor exposures using comparative transcriptome, suggesting that this weevil could deploy the dual mechanism (i.e., CYP450-mediated metabolic resistance and CP-mediated cuticular resistance) to counter host terpenoid defenses. Furthermore, temporal expression of CYP450 and CP genes under a low dose of d-camphor exposure indicated that there was a trade-of relationship between the inductions of CYP450 and CP genes. We speculate that the dual mechanism of terpenoid tolerance in this specialist is an essential precondition for the hormetic response in larval growth and development, ultimately contributing to its successful colonization on camphor trees. Additionally, we found a few genes related to glucose transport, juvenile hormone, and odorant binding were fine-tuned under terpenoid exposures. Taken together, our study will open new avenues for understanding insect-plant coevolutionary adaptation and developing durable control strategies for this insect pest.

Keywords: Terpenoid exposure, Hormetic response, RNA-seq, Cytochrome P450, Cuticular protein, Insect-plant coevolutionary adaptation

Symposium 8: Anthropogenic changes and applied solutions (1)

Thursday, August 11th
8:45 am - 9:45 am

Session moderated by Hajime Ono

Pesticide residues in the hive products and their potential risks to honeybees
Xiaolin Wen, Feiran Wang, Changsheng Ma, Shudong Luo, Hongmei Li-Byarlay

What ticks don't like: formulations and deployment of novel natural product-based repellents
Nicoletta Faraone, N. Kirk Hillier

Drought stress modulates floral chemistry with consequences for pollinator attraction
Caitlin Rering, José Franco, Rachel Mallinger

S8-O205

Pesticide residues in the hive products and their potential risks to honeybees

Xiaolin Wen[1], Feiran Wang[1], Changsheng Ma[2], Shudong Luo[1], Hongmei Li-Byarlay[3]

[1]Institution for Apicultural Research, Beijing, China. [2]Institute for Apicultural Reserach, Beijing, China. [3]Central State University, Wilberforce, USA

Research evidence suggests that pesticide residues are one of the leading potential causes of the decline in pollinators, especially during vulnerable periods such as foraging in the early springtime. It is important to quantify pesticide residues in the nectar and pollen of honey bee colonies during the early field season and examines the potential risks and toxicity of pesticides to honey bees. This talk will highlight two projects investigating 1) the pesticide residues in oilseed rape in the years 2017 and 2018 of bee population in China, and 2) residues of pesticides in bee bread and honey of Apis cerana cerana in China. The risk of detected residues of pesticides to honey bees was evaluated with hazard quotient (HQ) and BeeREX. Additional chronic and acute risks to humans according to the dietary exposure were also addressed. Our results suggest that the pesticide residues detection ratio (25.4% for beebread and
2.8% for honey) and the concentrations of these residues is lower than previously reported. Among all identified pesticides, only thiamethoxam raises the concern for further risk assessment in the risk evaluation of honey bee colonies and thiamethoxam was safe for colonies in higer tier studies Our results indicated that further investigation of nearly half of the tested compounds is needed because their PHQ or NHQ values are more than 50. Especially cyfluthrin and carbofuran need advanced tier assessment due to their maximum RQ (risk quotient) values exceeding the level of concern. These results provide valuable guidance for protecting bees and other pollinators.

Keywords: -

S8-O140

What ticks don't like: formulations and deployment of novel natural product-based repellents

Nicoletta Faraone[1], N. Kirk Hillier[2]

[1]*Department of Chemistry, Acadia University, Wolfville (NS), Canada.* [2]*Department of Biology, Acadia University, Wolfville (NS), Canada*

Ticks, such as blacklegged ticks (*Ixodes scapularis*) and American dog tick (*Dermacentor variabilis*), are vectors of several pathogens that negatively impact animal and human health. In recent years, global warming has increased the threat of disease transmission significantly, resulting in an increased demand for environmentally safe, tick repellent and acaricidal products. Natural products, such as essential oils, are prospective alternatives to manage these pests. To protect and enhance their properties, different eco-friendly approaches have been explored, such as encapsulation of the active ingredients with food-grade coating materials. Here we report an overview of different encapsulation techniques that have been studied in our lab to effectively deploy botanical active ingredients with tick repellent and acaricidal activities. Cyclodextrins, nanoemulsions, inert carriers and spray drying have all been explored as formulation technologies to deploy naturally derived acaricidal and repellent materials (i.e. basil essential oil, garlic oil, yarrow essential oil and others). Formulations have subsequently been assayed for efficacy in behavioural trials with *I. scapulars* and *D. variablis*. Botanical tick repellent and acaricides associated with nanotechnology offer great potential for vector control, without posing any significant risk to the environment and human health.

Keywords: Essential oils, Tick, Nanoencapsulation, Repellent, Acaricide

[1] Hogenbom J, Jones A, Wang HV, Pickett LJ, Faraone N (2021) Polymers 13: 1892
[2] Wang HV, Pickett LJ, Faraone N (2022) Exp Appl Acarol 86: 583-598

S8-O184

Drought stress modulates floral chemistry with consequences for pollinator attraction

Caitlin Rering[1], José Franco[2], Rachel Mallinger[3]

[1]USDA-ARS, Gainesville, FL, USA. [2]USDA-ARS, Booneville, AR, USA. [3]University of Florida, Gainesville, FL, USA

Due to climate change, droughts are predicted to worsen in severity and frequency in the coming years. Therefore, an improved understanding of the effects of drought on the plant-pollinator system is needed, including impacts on the chemicals that pollinators use to evaluate forage quality, such as volatile cues like floral scent and the chemical composition of floral rewards. To evaluate the effects of drought on floral traits and pollinator responses, we subjected buckwheat plants to either drought or control (well-watered) treatments. Between these treatments we compared nectar quantity and chemical composition, pollen quantity, floral volatile emissions, pollinator visitation rates, and seed yield. Drought-stressed plants produced fewer flowers and less nectar per flower, though pollen quantity per flower was unaffected. Nectar from drought-stressed plants was depleted of sucrose relative to total sugars. Floral volatile compositions differed between treatments, with drought-stressed plants having higher emissions of four volatiles: (Z)-3-hexenol, the ubiquitous green leaf volatile emitted from plants in response to stress, and three volatiles which are known byproducts of amino acids, isobutyraldehyde, 2-methylbutanal, and 3-methylbutanal. Since amino acids accumulate in floral tissues during drought, we hypothesize that these volatiles may be biomarkers of stressed flowers, though this requires further study. Drought-stressed plants received significantly fewer visits by bumble bees, honeybees, and flies. Finally, drought stress negatively affected seed yields, both by directly reducing growth and by reducing pollinator visits to flowers. Our results show that drought can have significant effects on floral traits and pollinator attraction, reducing plant reproductive success and the resources available to pollinators.

Keywords: Bees, Volatiles, Nectar, Agriculture, Pollen

Symposium 8: Anthropogenic changes and applied solutions (2)

Friday, August 12th
11:25 am - 12:05 pm

Session moderated by Radhika Venkatesan

Acute ozone exposure impairs detection of floral odor, learning, and memory of honeybees
Fabien Démares, Laëtitia Gibert, Pierre Creusot, Candice Dubuisson, Benoit Lapeyre, Magali Proffit

Activation of odorant receptor Or31 contributes to pyrethrum repellency in *Aedes aegypti* mosquito
Wilson R Valbon, Felipe Andreazza, Feng Liu, Qiang Wang, Peng Xu, Elizabeth Bandason, Mengli Chen, Bo Feng, Genki Takamatsu, Makoto Ihara, Kazuhiko Matsuda, Eugenio E Oliveira, Yuzhe Du, Ke Dong

S8-O143

Acute ozone exposure impairs detection of floral odor, learning, and memory of honeybees

Fabien Démares, Laëtitia Gibert, Pierre Creusot, Candice Dubuisson, Benoit Lapeyre, Magali Proffit

CEFE, Univ Montpellier, CNRS, EPHE, IRD,, Montpellier, France

Air pollution affects many ecosystems where plants and animals interact. Emissions of air pollutants such as tropospheric ozone (O_3) are harmful to plants and human health; yet, few reports studied the direct effects of O_3 on pollinators' physiology and behavior. Ozone can affect pollinators' floral preference and foraging via the modification of their perception abilities. Volatile organic compounds (VOCs) released by plants constitute olfactory signals used by pollinators for host plant recognition; detecting these VOCs shapes the plant-pollinator interactions. Here, we used the honeybee *Apis mellifera* as a pollinator model, and exposed individuals to different field-relevant O_3 concentrations (up to 200 ppb) to characterize two crucial aspects: 1) the ability of O_3-exposed honeybees to detect VOCs; and 2) the impact of O_3 on olfactory learning and memory processes of honeybees. We measured increasing antennal responses with increasing O_3 concentrations. In parallel, olfactory learning was not severely disturbed by O_3 concentrations, but lead to a higher generalization rate (i.e. difficulty to discriminate between VOCs). All in all, these results suggest a link between O_3-related oxidative stress and olfactory coding disturbance in the honeybee brain, a formal link we will discuss. If ozone affects the olfaction of pollinators, selective foraging behaviors may be modified, eventually leading to a possible long-term harmful effect on pollination services.

Keywords: Air pollution, Volatile organic compounds, Olfaction, generalization, *Apis mellifera*, Plant-pollinator interactions.

[1] Démares F, Gibert L, Creusot P, Lapeyre B, Proffit M (2022). Science of The Total Environment 827: 154342.

S8-O144

Activation of odorant receptor Or31 contributes to pyrethrum repellency in *Aedes aegypti* mosquito

Wilson R Valbon[1,2,3], Felipe Andreazza[1,2,3], Feng Liu[2], Qiang Wang[2], Peng Xu[2], Elizabeth Bandason[2], Mengli Chen[2], Bo Feng[2], Genki Takamatsu[4], Makoto Ihara[4], Kazuhiko Matsuda[4], Eugenio E Oliveira[2,3], Yuzhe Du[2], Ke Dong[1,2]

[1]*Department of Biology, Duke University, Durham, USA.* [2]*Department of Entomology, Michigan State University, East Lansing, USA.* [3]*Department of Entomology, Federal University of Viçosa, Viçosa, Brazil.* [4]*Department of Applied Biological Chemistry, Faculty of Agriculture, Kindai University, Nara, Japan*

Vector control is a pivotal strategy to mitigate human-transmitted diseases such as Dengue, Zika, and Chikungunya viruses around the globe. Among current tools available to combat mosquito populations, insecticides and repellents remain the most common. Pyrethrum extract from dried flowers of *Chrysanthemum* species (to be more specific *Tanacetum cinerariifolium*) exert its insecticidal effect by acting on voltage-gated sodium channels in insects. Furthermore, pyrethrum extract has been used as insect repellent against many biting arthropods for centuries. Although many compounds, such as pyrethrum, pyrethroids, and DEET, evoke spatial (i.e., non-contact) repellency, the molecular basis of odorant receptor (Or)-mediated avoidance in mosquitoes was not well-understood. In this study, we conducted behavioral, electrophysiological, and molecular genetics assays in *Aedes aegypti* mosquito to investigate whether pyrethrum repellency is Or-mediated. We found that pyrethrum extract elicits spatial repellency and activates two specific types of ORNs in mosquito antennae. A minor (< 2%) component of pyrethrum, (*E*)-β-farnesene (EBF), activates stb-1A neurons; and the major components of pyrethrum, pyrethrins, activates sst-1A neurons. We identified the specific Or activated by EBF as the *Ae. aegypti* odorant receptor 31(AaOr31), which is also activated by (±)-citronellal and geranyl acetate. Interestingly, knockout of AaOr31 significantly reduced pyrethrum and EBF repellency and also (±)-citronellal and geranyl acetate repellency. Excitingly, Or31 is conserved in all major human disease-transmitting mosquito species. Our study uncovered a conserved mosquito odorant receptor (Or31) that mediates repellency in mosquitoes.

Keywords: Spatial Repellency, Mosquito Olfaction, Olfactory Receptor Neurons

Symposium 9: Fruit fly attractants, repellents, and host interactions (1)

Friday, August 12th
8:45 am - 11:25 am

Session moderated by Dong Cha

Development of synthetic food-odor lures for *Bactrocera* species
Stefano De Faveri, Theo Yoder, Nicole Miller, Dong Cha, Jodie Cheesman, Matthew Siderhurst

Assessing non-methyl eugenol-responding lines of Oriental fruit fly males on their lure response
Alvin Kah-Wei Hee, Mandanayake A.R.A. Mandanayake

Olfactory roles of antenna and maxillary palp in long-range attraction of *Bactrocera* fruit flies
Kye Chung Park, Seon Ah Jeong, Hyun-Woo Oh

Plant-based attractant, zingerone and its analogs reveal structural requirements for attraction of Jarvis's fruit fly.
Soo Jean Park, Benjamin Hanssen, Stefano De Faveri, Jodie Cheesman, Jane Royer, Donald Cameron, Joanne Jamie, Ian Jamie, Phillip Taylor

Attraction of Oriental fruit fly to β-caryophyllene
Pradeepa Hewa Ranaweera, Alvin Kah-Wei Hee

Discovery and deployment of repellent compounds for managing *Drosophila suzukii*
Greg Loeb, Dong Cha, Binita Shrestha

Identification of female attractants for improved monitoring of oviposition-impending *Bactrocera dorsalis*
Dong Cha, Gwang-Hyun Roh, Paul Kendra

S9-O69

Development of synthetic food-odor lures for *Bactrocera* species

Stefano De Faveri[1], Theo Yoder[2], Nicole Miller[2], Dong Cha[3], Jodie Cheesman[1], Matthew Siderhurst[2]

[1]Department of Agriculture and Fisheries, Mareeba, Australia. [2]Eastern Mennonite University, Harrisonburg, USA. [3]USDA ARS PBARC, Hilo, USA

The oriental fruit fly (OFF), *Bactrocera dorsalis* (Hendel) and the Queensland fruit fly (Qfly), *B. tryoni* (Froggatt), are serious pests of tropical agricultural crops, attacking many different varieties of fruits and fruiting vegetables. Lures that attract OFF and Qfly are important for both detection and control of these pests. While strongly attractive male lures exist for both species, chemicals that attract female flies are generally limited to food-type attractants, such as fermenting sugars, hydrolyzed protein, and yeast. Unfortunately, these liquid lures often lack potency, have limited field life, are difficult to handle, and attract non-target species. On the upside, these liquid lures also offer the promise of killing immature or protein-deprived flies as well as females thereby preventing future offspring. In this study we first identified volatiles from over half a dozen liquid protein lures using headspace volatile sampling and GC-MS analysis. Major volatile groups identified include small organic acids, alkylpyrazines, arenes, and alcohols. Additionally, ammonia evolving from each lure was analyzed using a solid-state sensor. Rotating olfactometer bioassays were used to assess OFF attraction to both single compounds and blends. Interestingly, several closely related compounds showed different sex-biased trap captures in the bioassay. However, this sex-biased was not seen in limited testing with *Zeugodacus cucurbitae* (Coquillett). Bioassays with Qfly are ongoing. These results are promising for developing a synthetic protein-lure mimic that attractants both OFF and Qfly.

Keywords: Tephritid fruit flies, Protein lures, Rotating olfactometer, Sex-bias attraction

S9-O85

Assessing non-methyl eugenol-responding lines of Oriental fruit fly males on their lure response

Alvin Kah-Wei Hee[1], Mandanayake A.R.A. Mandanayake[1,2]

[1]*Universiti Putra Malaysia, Serdang, Malaysia.* [2]*Field Crops Research and Development Institute, Mahailluppallma, Sri Lanka*

The Oriental fruit fly, *Bactrocera dorsalis* is amongst the most destructive and invasive pests of fruits, making it one of the most targeted insects for biosecurity control in the world. Currently, the sequential male annihilation technique, followed by the sterile insect technique has been used to significantly reduce the population of those feral males. However, issues with sterile males being killed by going to male annihilation traps have reduced the efficacy of this approach. The availability of males that are non-methyl eugenol-responding would eliminate this problem and increase the efficacy of both approaches. Therefore, when sterile male flies' release programs are combined with male annihilation techniques, the use of sterile males may significantly improve the effectiveness of such approaches by reducing the loss of these males to lure traps. Our results suggest that it may be possible to establish lines of reduced responder males to be used for sterile male releases. We report here on the assessment of males from those lines in terms of methyl eugenol response and mating ability.

Keywords: Oriental fruit fly, *Bactrocera dorsalis*, Methyl eugenol, Non-responsiveness, Behaviour

S9-O80

Olfactory roles of antenna and maxillary palp in long-range attraction of *Bactrocera* fruit flies

Kye Chung Park[1], Seon Ah Jeong[2,3], Hyun-Woo Oh[2]

[1]Bioprotection, New Zealand Institute for Plant and Food Research, Christchurch, New Zealand. [2]Core Facility Management Center, Korea Research Institute of Bioscience & Biotechnology, Daejeon, Republic of Korea. [3]Department of Biological Sciences, Hannam University, Daejeon, Republic of Korea

Olfactory communication system is well developed in *Bactrocera* fruit flies. Currently, male attractants such as methyl eugenol and cue lure and protein-based generic attractants are widely used for a number of *Bactrocera* species. Like many other insects, well-developed chemosensilla are present on the antennae in *Bactrocera* fruit flies. Naturally, studies have mainly focused on the olfactory role of the antennae in fruit flies, whereas much less attention has been paid on the olfactory role of the maxillary palps although the involvement of the maxillary palps in olfactory communication has been suggested in various studies. Our recent studies through electron microscope observation, electrophysiological recording, and behavioral screening to understand the olfactory communication of two *Bactrocera* species, *B. depressa* and *B. scutellata,* exhibited that the olfactory sensory system is species-specific, in which not only the antennae but also maxillary palps are involved in long-range olfactory attraction through separate sensory processes. Scanning electron microscope observation indicated that the morphological types of olfactory sensilla are clearly different between the antennae and the maxillary palps in these species, and GC-EAD recordings exhibited that the peripheral olfactory response profiles of the maxillary palps are distinct from those of the antenna. GC-EAD and field trapping tests suggested that the strong behavioural attraction of male *B. scutellata* to cue lure and raspberry ketone is achieved through the peripheral detection of these compounds by the maxillary palps, not by the antennae. Our findings provide new insight into how complex olfactory signals are perceived in *Bactrocera* fruit flies, suggesting complete long-range behavioral attraction in fruit flies is achieved through species-specific and combinational olfactory sensory channels of antennae and maxillary palps.

Keywords: Antenna, Fruit fly, GC-EAD, Maxillary palp, Olfactory communication

S9-O153

Plant-based attractant, zingerone and its analogs reveal structural requirements for attraction of Jarvis's fruit fly.

Soo Jean Park[1], Benjamin Hanssen[1], Stefano De Faveri[2], Jodie Cheesman[2], Jane Royer[3], Donald Cameron[1], Joanne Jamie[1], Ian Jamie[1], Phillip Taylor[1]

[1]*Macquarie University, Sydney, Australia.* [2]*Department of Agriculture and Fisheries, Mareeba, Australia.* [3]*Department of Agriculture and Fisheries, Brisbane, Australia*

The flowers of *Passiflora maliformis* and *Semecarpus australiensis* are known to attract *Bactrocera jarvisi* males. Zingerone, a phytochemical, is known to attract several *Bactrocera* species, including *B. jarvisi*. We investigated the chemistry of the flowers of the two plants for presence of zingerone and elucidated the functional groups of zingerone involved in the attraction of *B. jarvisi*. The flowers of *P. maliformis* and *S. australiensis* were analysed by solvent extraction and gas chromatography-mass spectrometry (GC-MS). Zingerone analogs with side-chain, hydroxy, and methoxy group modifications were synthesised and tested in the field. The flowers of both plants contained zingerone. Although *P. maliformis* is an introduced plant, zingerone *S. australiensis* is native and sympatric with *B. jarvisi*, suggesting a possible co-evolutionary relationship between the plant and insect. Field trials showed that the attractiveness of zingerone has limited tolerance to structural modification. The most attractive analogs were alkoxy derivatives, with isopropoxy being the most attractive, followed by ethoxy and trifluoromethoxy analogs. The phenolic esters tested were also attractive with the response typically decreasing with increasing size of the ester. Results indicate that the carbonyl group, methoxy group, and phenol of zingerone are key sites for the attraction of *B. jarvisi* and identify some constraints on the range of structural modifications that can be made to zingerone without compromising attraction. These findings are important for future work in developing and optimizing novel male chemical lures for fruit flies.

Keywords: Passion fruit flowers, Native cashew, *Bactrocera jarvisi*

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[2] Hanssen BL; Park SJ; Royer JE; Jamie JF; Taylor PW; Jamie IM (2019), Sci. Rep, 9(1), 19332.

S9-O106

Attraction of Oriental fruit fly to β-caryophyllene

Pradeepa Hewa Ranaweera, Alvin Kah-Wei Hee

Universiti Putra Malaysia, Serdang, Malaysia

Methyl eugeinol (ME) is a well-known highly potent attractant for males of the Oriental fruit fly, *Bactrocera dorsalis*. However, ME only traps male flies thus, limiting the possibility of controlling entire fly population or trapping females. The sesquiterpene compound, β-caryophyllene (CP) has been recorded as a strong male attractant for the guava fruit fly, *B. correcta* and CP attraction was also reported for females of *B. correcta* and *B. dorsalis*. In this work, the attraction of male and female *B. dorsalis* to BCP was investigated to ascertain effects of sexual maturity on their attraction to CP. We also investigated why traps containing CP had fail to attract *B. dorsalis* unlike that of *B. correcta* in the field. We discovered that, like previous *B. dorsalis* attraction to ME that is a function of sexual maturity, the attraction of males to CP is also concomitant with attainment of sexual maturity albeit at older age. Males are attracted by the scent of CP and would fly excitedly to land on the source of the CP before feeding on it. Gravid females however, when attracted to CP, do not feed on it unlike those males. Cage and wind tunnel bioassays further demonstrated that the attraction of *B. dorsalis* to CP is of short range only. Interestingly, we also demonstrated through combinations of antennal and maxillary palpal ablation, that both organs are required for optimal fly response to CP.

Keywords: β-caryophyllene, Attraction, Wind tunnel, Antenna, Maxillary palp

S9-O165

Discovery and deployment of repellent compounds for managing *Drosophila suzukii*

Greg Loeb[1], Dong Cha[2], Binita Shrestha[1]

[1]*Cornell University, Geneva, USA.* [2]*USDA ARS, Hilo, USA*

Drosophila suzukii (SWD) is a serious economic threat to soft-skinned fruit such as berries and cherries. Insecticides are the main tool to control SWD, although their repeated use comes with associated economic and environmental costs. We have been investigating candidate repellents to manipulate the behavior of SWD to augment the use of insecticides. We individually evaluated antennally active volatiles produced from fermenting bread dough and discovered several compounds that reduced attraction to a bait, with 2-pentylfuran (2PF) causing the largest reduction. Dose-response assays indicated 2PF at 3-5 mg/h decrease attraction in the lab. Using sachets made from polyethylene to control release of 2PF, we showed release rates of 10 mg/h significantly reduced infestation of raspberry fruit under field conditions. Using small plots of raspberries (three rows x 6m), we found a modest decrease in infestation associated with the highest release rate. During 2022 field season we will test the efficacy of 2PF released from aerosol puffers releasing at 8.5 mg/h every ten minutes. In addition, we are exploring the potential of combining three other compounds found to decrease attraction in our original assay with 2PF to test for additive or potentially synergistic effects. In addition to our work with 2PF, we will report on research investigating odors produced from the fruit-infecting fungus Botrytis cinerea as potential repellents. Our longer-term goal is to identify behaviorally active compounds and develop them into practical tactics to manage.

Keywords: Repellent, Spotted wing drosophila, Pest management

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S9-O167

Identification of female attractants for improved monitoring of oviposition-impending *Bactrocera dorsalis*

Dong Cha[1], Gwang-Hyun Roh[1], Paul Kendra[2]

[1]USDA-ARS, U.S. Pacific Basin Agricultural Research Center, Hilo, Hawaii, USA. [2]USDA-ARS, Subtropical Horticulture Research Station, Miami, Florida, USA

There is a clear need for a new and more potent attractant for surveillance of female oriental fruit fly (OFF). Worldwide, OFF is one of most destructive invasive pests of fruit that can become a serious trade-barrier once established. Current OFF surveillance programs in the US rely on a large number of traps (10,000 traps/yr in CA) baited with methyl eugenol and liquid protein bait (torula yeast, TY). Although methyl eugenol is highly potent, it only attracts males and cannot monitor OFF females. TY-baited traps can attract both sexes and thus have been used for detection of female OFF. However, TY is not strong enough to meet program needs as a standard female attractant; furthermore, TY traps are difficult to maintain, attracts many non-target insects, and changes attractiveness with time. The lack of suitable sensitivity of TY traps for OFF females appears to be related to its inability to attract females ready for oviposition. In our recent study using cohorts of 14~16-day old mated OFF females, some preferred traps baited with TY while some preferred traps baited with host fruit. Interestingly, the mated females that preferred host fruit odor had 2X more mature eggs in their ovaries and laid 2X more eggs than mated females that preferred TY odor, suggesting great potential for using host fruit odor-based lures to monitor "oviposition-ready" mated females and improve the efficacy of OFF detection. In this talk we will discuss the identification of a 16-component chemical lure that is as attractive to female OFF as a preferred host fruit (guava) and more effective than TY in catching oviposition-impending females in the field; and our plans for further optimization of this lure.

Keywords: Female attraction, Food choice, Host choice, Physiological status

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Symposium 9: Fruit fly attractants, repellents, and host interactions (2)

Friday, August 12th
1:30 pm - 2:30 pm

Session moderated by Anat Levi-Zada

Stake a claim: microbes facilitate niche partitioning in tephritid fruit flies
Kamala Jayanthi P D, Saravan Kumar Parepally, Meenal Vyas

Salicylate alters the volatile profile of mango thereby affecting the behavior and development of the Oriental fruit fly, *Bactrocera dorsalis*
Vivek Kempraj, Ravindra Aurade, Kamala Jayanthi Pagadala Damodaram

Insect-plant interaction: A new approach of Bactrocera fruit flies (Diptera: Tephritidae) on the Cape Jasmine flower (Gentianales: Rubiaceae) in Bangladesh
Farzana Yesmin, Md Nazim Uddin, Md. Hasanuzzaman

S9-O154

Stake a claim: microbes facilitate niche partitioning in tephritid fruit flies

Kamala Jayanthi P D, Saravan Kumar Parepally, Meenal Vyas

ICAR-IIHR, BENGALURU, India

Niche is a compartment that specifies boundaries in the ecological system to a certain biological entity. Herbivore insects have been known to behave within their ecological niches that may be defined in different ways. Of major interest is a reproductive niche created by existing populations so that they thrive and co-exist. Therefore, a niche partition exists among different organisms as a measure of resource distribution to provide sustainability. The reproductive isolation in general is brought about by reproductive incompatibility or geographical separation. Ovipositional niches exist as an additional level of compartmentalization that prevents population disruption among closely related insect communities. Studies in a few biological systems have revealed that microbes play an important role in creation of such biological niches. Our studies with tephritid fruit flies namely Oriental fruit fly, *Bactrocera dorsalis* and guava fruit fly, *Bactrocera correcta* populations have helped in understanding this interaction with a semiochemical perspective. Olfactometer behavioural assays suggest that the volatile profile of a fruit infested with larvae of one population is an ovipositional deterrent for the other. This was then confirmed to be affected by the microbes that are associated with eggs. The Microbes such as *Klebsiella oxytoca*, *Enterobacter cloaece*, *Citrobacter* species and Serratia marcescens were isolated from B. dorsalis infested fruits while, Elizabethking anopheles, *Klebsiella oxytoca* and *Enterobacter bugandensis/Klebsiella grimontii* were isolated from the fruits infested by *B. correcta*. We speculate that the microbes alter the volatile profile of the fruits and thus prevent the competing population from ovipositing in the same fruit. Further GC-EAD/GC-MS studies revealed several important cues that can be employed as ovipositional deterrents. Such niches thus help the populations co-exist without displacing each other.

Keywords : Niche Partition, Oviposition, *Bactrocera dorsalis*, *Bactrocera correcta*, Microbes, Cues

S9-O107

Salicylate alters the volatile profile of mango thereby affecting the behavior and development of the Oriental fruit fly, *Bactrocera dorsalis*

Vivek Kempraj[1,2], Ravindra Aurade[3], Kamala Jayanthi Pagadala Damodaram[4]

[1]USDS-ARS, Hilo, USA. [2]RCUH, Honolulu, USA. [3]Central Sericultural Research and Training Institute, Mysuru, India. [4]Indian Institute of Horticultural Research, Bengaluru, India

The Oriental fruit fly, *Bactrocera dorsalis*, is an economically important pest of mango. Although, present control measures are effective in controlling their population, more novel strategy of pest control is required. A pest control strategy that is promising is the induction of 'natural plant defenses' by using phytohormones prior to pest attack to keep the plants defense ready. Here, we investigated the effect of a phytohormone (Salicylic acid; SA) treatment on mango fruit (cv. Totapuri) for oviposition and larval development of the *B. dorsalis*. In oviposition choice assays, gravid females laid significantly fewer eggs in SA treated fruits compared to untreated fruit. Headspace volatiles collected from SA treated fruit were less attractive to gravid females compared to volatiles from untreated fruit. GC-MS analysis of the headspace volatiles from SA treated and untreated fruit showed noticeable changes in their chemical compositions. Cis-ocimene and 3-carene (known attractants of *B. dorsalis*) were significantly reduced in the headspace volatiles of the SA treated fruit. Further, reduced pupae formation and adult emergence was observed in treated fruit compared to control, which was associated with increased phenol and flavonoid content in the treated fruit. We also observed increased activity of anti-oxidative enzymes, such as catalase (CAT), polyphenol oxidase (PPO) and peroxidase (POD). Taken together, the results indicate that SA treatment reduced oviposition, larval development, and adult emergence in *B. dorsalis*.

Keywords: *Bactrocera dorsalis*, Behaviour, Volatiles, Headspace volatiles, Salicylic acid

S9-O87

Insect-Plant interaction: A new approach of Bactrocera fruit flies (Diptera: Tephritidae) on the Cape Jasmine flower (Gentianales: Rubiaceae) in Bangladesh

Farzana Yesmin[1], Md Nazim Uddin[1,2], Md. Hasanuzzaman[1]

[1]*Cytology and Biocontrol Research (CBR), Radiation Entomology and Acarology Division (READ), Institute of Food and Radiation Biology (IFRB), Atomic Energy Research Establishment (AERE), Bangladesh Atomic Energy Commission (BAEC), GPO Box 3787, Dhaka 1000, Bangladesh.* [2]*Department of Zoology, Darsana Govt. College, Chuadanga, Bangladesh*

Bactrocera fruit flies have been significant as model organism for basic research considering their economic context that reduce fruit yields and generating constrains on quarantine issues. They damage a wide variety of fruits and vegetables in South-East Asia, Asia-Pacific and African countries. Insect-Plant interactions being adopted not only to develop pest control mechanisms but also establishing their genetic diversity. In most cases plants contain a huge array of chemical compounds primarily to establish their natural defense against herbivores and pathogens while floral colour and fragrance to attract their pollinators. A consecutive two years study have been carried out in IFRB premises to find out interactions between Bactrocera and Cape Jasmine (*Gardenia jasminoides*) flower. Male of *Bactrocera dorsalis* and *B. zonata* were found having spell-bounded while sitting on petals of Cape Jasmine. Mass gathering on petals was noticed just after raining in April (maximum flower blooming time). Most of the flowers were enchanted by more than one fly. Fresh and full bloom flowers were cherished than those of half bloom or old flowers. *B. dorsalis* was much glamor than *B. zonata*. More than twenty flies were crowed on a single flower while in some cases seven to eight flies rushed on individual petal. They enchanted so much as they were captured even with free hand. Flies were gathered from noon to dusk only. Maximum numbers of flies were gathered just after rainfall with clear sun light. The study revealed that Cape Jasmine is a medicinal plant, it contains chemicals including eugenol which is successfully used in fruit fly surveys, quarantine detection, estimation of native fruit flies, determine interaction between fruit phenology and fruit fly population, monitoring their activities in native and exotic territory and control of fruit flies via male annihilation technique (MAT) through mass trapping.

Keywords: Bactrocera, Gardenia, Pheromone attractants

Symposium 10: Chemical defenses in vertebrates (1)

Friday, August 12th
12:05 pm - 12:25 pm

Session moderated by Adriana M. Jeckel

Sequestration timeframe and systemic distribution of alkaloids in a Dendrobatid poison frog
Adriana M. Jeckel, Kunihiro Matsumura, Keisuke Nishikawa, Yoshiki Morimoto, Demian R. Ifa, Taran Grant, Ralph A. Saporito

S10-O169

Sequestration timeframe and systemic distribution of alkaloids in a Dendrobatid poison frog

Adriana M. Jeckel[1,2], Kunihiro Matsumura[3], Keisuke Nishikawa[3], Yoshiki Morimoto[3], Demian R. Ifa[4], Taran Grant[2], Ralph A. Saporito[5]

[1]*Institute of Plant Sciences, University of Bern, Bern, Switzerland.* [2]*Institute of Biosciences, University of São Paulo, São Paulo, Brazil.* [3]*Department of Chemistry, Graduate School of Science, Osaka City University Sumiyoshi-ku, Osaka, Japan.* [4]*Center for Research in Mass Spectrometry, Department of Chemistry, York University, Toronto, Canada.* [5]*Department of Biology, John Carroll University, University Heights, USA*

Several species of animals independently evolved the ability to sequester defensive compounds from their diet. Sequestration involves the ingestion, absorption, transportation, and storage of chemicals in specialized tissues. Poison frogs are the most well studied group of vertebrates that sequester alkaloids from dietary sources. Alkaloids are largely sequestered from ants and mites and are accumulated in dermal poison glands. An important characteristic of this system is the high inter- and intraspecific variation in the types and quantity of alkaloids. Ecological factors such as geographic location, season, sex, age, life stage, and body size are known to directly influence the composition of alkaloids among individuals and populations. However, little is known about the mechanism of sequestration itself and its role in the variation of alkaloids in poison frogs. In the present study, we aimed to understand the anatomical pathway of an alkaloid after ingestion by investigating the systemic distribution of two alkaloids (decahydroquinoline and histrionicotoxin **235A**) in a poison frog over the course of a 24 h period. We used gas chromatography coupled with mass spectrometry to quantify the two alkaloids in different organs of the body, and then desorption electrospray ionization mass spectrometry imaging (DESI-MSI) to map the two alkaloids in different organs and tissues of the body. In the 1 h experiment, there were high quantities of alkaloids detected in the stomach, but a percentage was already present in the skin. However, after 24 hours, no alkaloid was detected by DESI-MSI and the amount of alkaloid in the skin remained similar to the 1 h treatment. Our results suggest that the alkaloid transport system from digestive tract to dermal skin glands following ingestion appears to be quicker than expected, but apparently not very efficient in the first 24 hours.

Keywords: Decahydroquinoline, *Dendrobates tinctorius*, Histrionicotoxin, Mass spectrometry imaging

Symposium 10: Chemical defenses in vertebrates (2)

Friday, August 12th
8:45 am - 10:25 am

Session moderated by Adriana M. Jeckel

Another evidence regarding the dietary source for skin alkaloids of poison frogs
Clement Tourbez, Philippe Gaucher, Grégory Genta-Jouve

New insights into dietary toxin metabolism: diversity in the abilityof the natricine snake *Rhabdophis tigrinus* to convert toadderived bufadienolides
Takato Inoue, Ryu Nakata, Alan Savitzky, Naoko Yoshinaga, Akira Mori, Naoki Mori

Diet and genetics influence population differences in poison frog alkaloid profiles
Lauren O'Connell

Non-lethal methods open new possibilities to perform a metabolite profiling of skin secretions from endangered and endemic amphibians
Mabel Gonzalez, Jhony Oswaldo Turizo, Alexander Aksenov, Pieter Dorrestein, Andrés E. Brunetti, Marco González-Santoro, Pablo Palacios-Rodríguez, Jack Hernández-Restrepo, Chiara Carazzone

Autoresistance to toxins in poisonous *Pitohui* birds is not rooted in sodium channel mutations
John Dumbacher, Fayal Abderemane-Ali, Megan Kobiela, Gopinathan Menon, Daniel Minor

S10-O101

Another evidence regarding the dietary source for skin alkaloids of poison frogs

Clement Tourbez[1,2], Philippe Gaucher[1], Grégory Genta-Jouve[1]

[1]UAR 3456 CNRS LEEISA - Laboratoire Ecologie, Evolution, Interactions des Systèmes amazoniens, Cayenne, French-Guiana. [2]Laboratory of Zoology, Research Institute for Biosciences, University of Mons, Mons, Belgium

Dendrobates species are known to store a wide range of poisonous alkaloids such as pumiliotoxins, histrionicotoxins or decahydroquinolines. While it is well accepted that the diet has a major role in the acquisition process of such compounds, but little is known about the underlying mechanism. Here we present the chemical analysis of skin extracts from *Dendrobates tinctorius* raised in French Guiana inside terraria either on wingless fruit flies or on leaf-litter arthropods collected every day from a site where a population of this dendrobatid frog occurs. We report the identification of 13 alkaloids using numerical methods including MS/MS spectra prediction and in silico metabolism based on high resolution mass spectrometry coupled with high performance liquid chromatography data and a statistical analysis. All alkaloids were exclusively identified in the frogs raised on leaf litter arthropods this confirming previous results obtained on *Dendrobates auratus.* A chemical analysis was also performed on the arthropods after identification. Unfortunately, none of the previously reported alkaloids were found in those extracts leading us to the conclusion that only precursors are sequestered by the frogs.

Keywords: Chemical defence, poisonous alkaloids, dendrobatid frog, diet, metabolites identification

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S10-O46

New insights into dietary toxin metabolism: diversity in the abilityof the natricine snake *Rhabdophis tigrinus* to convert toadderived bufadienolides

Takato Inoue[1], Ryu Nakata[1], Alan Savitzky[2], Naoko Yoshinaga[1], Akira Mori[3], Naoki Mori[1]

[1]*Graduate School of Agriculture, Kyoto University, Kyoto, Japan.* [2]*Department of Biology, Utah State University, Logan, UT, USA.* [3]*Graduate School of Science, Kyoto University, Kyoto, Japan*

The Japanese natricine snake *Rhabdophis tigrinus* sequesters cardiotonic steroids, bufadienolides (BDs), from ingested toads in the nuchal glands as defensive toxins. A previous study showed that R. tigrinus in captivity converts dietary BDs when it sequesters them. However, it is unknown whether the dietary BDs are actually converted and the modified products accumulated under natural conditions. It is also unknown to what extent the BD profile of ingested toads is reflected in that of the snake. We collected 123 snakes from throughout Japan, analyzed their BD profiles by liquid chromatography/mass spectrometry, and identified 15 BDs from *R. tigrinus* by nuclear magnetic resonance analyses. We also compared their BD profiles using hierarchical cluster analysis (HCA). HCA exhibited two main clusters associated with their collection locations: eastern and western regions of the Japanese main islands. These results, coupled with previous findings on the BDs of Japanese toads, suggest that 1) *R. tigrinus* converts toad-derived BDs into other compounds under natural conditions; 2) there are both universal and regionally specific conversions of dietary BDs by *R. tigrinus*; and 3) geographic variation in toad BD profiles is partially reflected in the variation of snake BD profiles.

Keywords: Biotoxin, Bufadienolide, Rhabdophis tigrinus, Sequestration, Chemical analysis

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S10-O204

Diet and genetics influence population differences in poison frog alkaloid profiles

Lauren O'Connell

Stanford University, Stanford, USA

Many forms of life obtain their chemical defenses from external sources, such as a specialized diet, which have a profound impact on physiology and fitness. Poison frogs acquire their skin alkaloids from their arthropod diet, which are bioaccumulated and repurposed to avoid predation. However, we do not how this evolutionary adaptation is shaped by genetic background of these frogs. Here, we show that populations of the Diablito poison frog (*Oophaga sylvatica*) vary in their chemical profiles. Using captive feeding experiments, we show that frog populations differ in their ability to metabolize dietary alkaloids into more potent molecules. To examine how alkaloid profiles co-vary with physiology, we used untargeted proteomics to quantify protein abundance differences in the liver, intestines, and skin. In general, we found populations vary in abundance of small molecule transporters and immune system-related proteins. Overall, our results show that poison frog toxicity is an environmental trait with strong genetic underpinnings in the physiological machinery to sequester, bioaccumulate and metabolize alkaloids.

Keywords: -

S10-O164

Non-lethal methods open new possibilities to perform a metabolite profiling of skin secretions from endangered and endemic amphibians

Mabel Gonzalez[1,2], Jhony Oswaldo Turizo[3], Alexander Aksenov[4], Pieter Dorrestein[5], Andrés E. Brunetti[6], Marco González-Santoro[7], Pablo Palacios-Rodríguez[7], Jack Hernández-Restrepo[7], Chiara Carazzone[1]

[1]Department of Chemistry, Universidad de los Andes, Colombia. [2]Department of Biology, Stanford University, USA. [3]Department of Physics, Universidad de los Andes, Colombia. [4]Department of Chemistry, University of Connecticut, USA. [5]Skaggs School of Pharmacy and Pharmaceutical Sciences, University of California San Diego, USA. [6]Laboratorio de Genética Evolutiva, Instituto de Biología Subtropical (CONICET - UNaM), Universidad Nacional de Misiones, Argentina. [7]Department of Biological Sciences, Universidad de los Andes, Colombia

The huge chemodiversity found in amphibians is comparable with that of some plants, insects, and marine organisms. Dendrobatids are recognized by the unique diversity of lipophilic alkaloids found on their skin. To analyze those compounds, total skinning following euthanization is the most widely used technique. However, total skinning is not suitable for endemic or endangered species or populations. The aim of this study was to test the extraction efficiency of two non-lethal methods of amphibian secretions, following a mild electrical stimulation of 3 min using Ag/AgCl electrodes. First, we perform a swab-based standardization process comparing three types of swabs applied to two endemic poison frogs from Colombia (*Dendrobates truncatus* and *Oophaga histrionica*) by single quadrupole GC-MS. Secondly, we present a temporal analysis of the skin secretions of Dendrobates auratus employing PDMS-patches analyzed by GC-Q-TOF. Results from skin swabs resulted in similar chemical profiles to those obtained by the gold standard lethal method, detecting a total of 94compounds. Our findings demonstrate that chemical and ecological qualitative interpretations between species, populations, and specimens were maintained with the non-lethal method. On the other hand, PDMS-patches allowed an in vivo temporal monitoring of 63 compounds. However, after 36 hours of successive sampling of the secretions, the extraction reaches a limit. These results show that mild electrical stimulation from amphibian dorsal skin and posterior swabbing using different materials has proven to be a successful method for sampling small metabolites from amphibians. From now on, this method could be applied directly in the frogs' habitat and extended for the chemical characterization of other small molecules and species. Also, it allows closer monitoring of health indicators that could be assayed from amphibian secretions over time series, which could be especially valuable now, when amphibians are facing panzootic chytridiomycosis.

Keywords: Metabolomics, Poison frogs, *In vivo*, Alkaloids, Volatiles.

S10-O172

Autoresistance to toxins in poisonous *Pitohui* birds is not rooted in sodium channel mutations

John Dumbacher[1], Fayal Abderemane-Ali[2], Megan Kobiela[3], Gopinathan Menon[1], Daniel Minor[2]

[1]*Institute for Biodiversity Science and Sustainability, California Academy of Sciences, San Francisco, CA, USA.* [2]*Cardiovascular Research Institute, University of California, San Francisco, CA, USA.* [3]*School of Biological Sciences, University of Nebraska–Lincoln, Lincoln, NE, USA*

Four avian genera from New Guinea are known to carry potent neurotoxins belonging to the batrachotoxin (BTX) family of alkaloids. The most toxic bird species, the Hooded and Variable Pitohuis (genus *Pitohui*), carry significant BTX concentrations in skin and feathers (where they can be most useful for defense), but toxins are also found in the muscle, heart, and other tissues that are normally poisoned by BTX. Batrachotoxins bind to voltage gated sodium channels (Nav) and hold them in the open conformation, and thus normally poison nerve and muscle membranes, causing numbness, tingling, burning and other irritation, and at higher concentrations can cause paralysis and heart and breathing cessation. We investigated the mechanisms of toxin resistance in pitohuis, and sequenced Pitohui sodium channel genes, expressed those proteins. We found that multiple pitohui Nav channels have no mutations that render them inherently resistant to BTX, and therefore these channels should be poisoned by natural toxin levels. Other work that we have done suggests that toxins are distributed non-randomly in the body and even within single skin cells, and this suggests active toxin transport, and possibly "sponge proteins" that can escort toxins away from tissues that they might harm and into tissues where they can perform selective functions. Although we have not been able to elucidate the exact mechanisms in vertebrates, we discuss why this explanation for BTX resistance is more parsimonious than resistance mutations, and why it is likely a more general mechanism across vertebrates, and why resistance is important to study in vertebrates.

Keywords: Pitohui, Toxin resistance, Batrachotoxin, Sponge protein

Symposium 11: Evolutionary chemical ecology: Diverging signaling mechanisms contributing to species differentiation

Friday, August 12th
8:45 am - 11:25 am

Session moderated by Jan Buellesbach

Unraveling evolutionary conserved signaling elements in chemical profiles of insects
Jan Buellesbach, Weizhao Sun, Neil Tsutsui, M. Alejandra Esparza-Mora, Dino McMahon, Jürgen Gadau

Decoding cuticular hydrocarbon-mediated female sexual attractiveness in the parasitoid wasp *Nasonia vitripennis*
Weizhao Sun, Juergen Gadau, Jan Buellesbach

Beyond cuticular hydrocarbons: Development of SPME on-fiber derivatization methods to investigate volatile organic surface compounds in parasitoid wasps
Quoc Hung Le, Jan Buellesbach

Changing scents: how pollinator-attractive odors in *Ficus racemosa* vary across distance in Asia
Lucy Nongbri, Kaveri Dey, Jean-Marie Bessiere, Yuvaraj Ranganathan, Renee Maria Borges

Molecular mechanisms underlying flower colour and volatile variation in a deceptive orchid
Darren Wong, James Perkins, Zemin Wang, Grace Marsh, Rod Peakall

Functional evolution of odorant receptors in termites
Jibin Johny Souleymane Diallo, Ondřej Lukšan, Blanka Kalinová, Robert Hanus, Ewald Große-Wilde

Mapping out olfactomes to unravel tephritid olfactory ecology and evolution
Chaymae Fennine, Sebastian Larsson Herrera, Advaith Chakravarthy, Sergio Angeli, Teun Dekker

S11-O103

Unraveling evolutionary conserved signaling elements in chemical profiles of insects

Jan Buellesbach[1], Weizhao Sun[1], Neil Tsutsui[2], M. Alejandra Esparza-Mora[3,4], Dino McMahon[3,4], Jürgen Gadau[1]

[1]Institute for Evolution & Biodiversity, WWU, Münster, Germany. [2]Department of Environmental Science, Policy, & Management, UC, Berkeley, USA. [3]Institute of Biology, FU, Berlin, Germany. [4]Department for Materials and Environment, BAM, Berlin, Germany

How exactly chemical information is encoded by the vast number of semiochemicals with signaling capabilities remains poorly understood. Cuticular hydrocarbons (CHCs) are an excellent example for this, as they have been demonstrated to encode and convey a vast array of differential chemical information, from sex pheromones to nestmate recognition. CHC profiles constitute complex mixtures of up to hundreds of different compounds, and it has been notoriously difficult to unravel the main signaling components except in a few case studies. My current research aspires to elucidate the underlying mechanisms by which chemical information such as sexual attractiveness, health status, and species- as well as nestmate affiliation can be encoded through differential chemical profile patterns. Intriguingly, closer chemical investigations of communication modalities as diverse as sexual signalling in parasitoid wasps, social immunity cues for fungal infections in termites, and nestmate recognition in invasive ants, collectively hint at the little investigated potential of methyl-branched alkanes as the main coding compounds. This intuitively makes sense, as this CHC compound class indeed carries the most potential for encoding a wide variety of differential chemical information through the multitude of possible positions and numbers of methyl branches. This suggests that basic signalling properties of methyl-branched alkanes can be evolutionary conserved across vast phylogenetic boundaries, while the actual communicated information can still vary considerably. Therefore, we hypothesize that this particular compound class contributes the basic elements of a potentially common, unified "chemical language" across different insect taxa.

Keywords: Chemical communication, Semiochemicals, Cuticular hydrocarbons, Methyl-branched alkanes

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S11-O104

Decoding cuticular hydrocarbon-mediated female sexual attractiveness in the parasitoid wasp *Nasonia vitripennis*

Weizhao Sun, Juergen Gadau, Jan Buellesbach

Institute for Evolution & Biodiversity, Münster, Germany

As the earliest and most wide-spread form of communication, chemical signaling has permeated through all known taxa of life. Insects, in particular, have exploited chemical signaling as their primary mode of communication. Cuticular hydrocarbons (CHCs), a group of unpolar waxes on the insects' epicuticle, have been demonstrated to encode and convey a vast array of differential information, commonly associated with sexual communication. In the parasitoid wasp *Nasonia vitripennis*, female CHCs serve as sexual cues, eliciting courtship and copulation behavior in conspecific males. *N. vitripennis* female CHC profiles constitute more than 50 different compounds, which makes it difficult to unravel the main components encoding sexual attractiveness. The present study addresses this knowledge gap by knocking down a fatty acid synthase gene we hypothesized to be involved in the CHC biosynthetic pathway, and further correlating the altered female CHC profiles with their sexual attractiveness. Specifically, the gene knockdown results in a consistent pattern of primarily up- and down-regulated methyl-branched alkanes with specific branching positions. We concordantly document a dramatic reduction of the sexual signaling function normally encoded in female CHC profiles, demonstrated by significantly reduced courtship and copulation behavior towards knockdown females by conspecific males. Intriguingly, this suggests a potential coding mechanism for sexual attractiveness mediated by specific branching patterns of methyl-branched CHCs in *N. vitripennis*.

Keywords: Chemical communication, Cuticular hydrocarbon, Sexual attractiveness

S11-O89

Beyond cuticular hydrocarbons: Development of SPME on-fiber derivatization methods to investigate volatile organic surface compounds in parasitoid wasps

Quoc Hung Le[1,2], Jan Buellesbach[1]

[1]*Institute for Evolution and Biodiversity (IEB), Münster, Germany.* [2]*Institute of Food Chemistry, Münster, Germany*

Nasonia (Hymenoptera: Chalcidoidea) is a species complex of parasitoid wasps that infests various flesh- and blowflies. In recent decades, *Nasonia* has become a prevalent model organism for chemical ecology and has been exceptionally well investigated for its chemical communication mechanisms. One of the major communication modalities in *Nasonia* is mediated by non-polar cuticular hydrocarbons (CHCs). However, under certain circumstances, CHC profiles are insufficient to convey complete sex and species discrimination. In this respect, polar cuticular lipids, which remain usually neglected in conventional liquid extractions of insect surfaces, have a high potential to play additional roles in chemical signaling. In order to investigate this potentially new chemical communication modality in *Nasonia*, the present study is aiming at developing sensitive and selective methods for analyzing polar volatile organic compounds. Investigations of volatile substances are undertaken using solid phase microextraction (SPME) coupled with derivatizations. A combination of these two methods, so-called SPME on-fiber derivatization, allowed us to identify many compounds generally undetectable under normal gas chromatographic conditions. A wide range of carbonyl compounds with chain lengths of up to C11 was found in both sexes of all *Nasonia* species with qualitative and quantitative differences. In addition, organic acids and alcohols with a hydrocarbon chain length of up to C14 were also discovered, with several components being identified as promising new sex- and species-specific compounds displaying clear qualitative differences between the investigated *Nasonia* species. This novel methodology opened up a completely new venue of hitherto undetected polar chemical profiles within *Nasonia*, with vast potential to close the gaps in our knowledge on chemically mediated behaviors in this chemical ecological model system.

Keywords: *Nasonia*, SPME on-fiber derivatization, Polar volatile organic compounds, Pheromones

S11-O179

Changing scents: how pollinator-attractive odors in *Ficus racemosa* vary across distance in Asia

Lucy Nongbri[1], Kaveri Dey[1], Jean-Marie Bessiere[2], Yuvaraj Ranganathan[1], Renee Maria Borges[1]

[1]*Indian Institute of Science, Bengaluru, India.* [2]*Ecole Nationale Superieure, Montpellier, France*

Floral scents play a crucial role in the highly-specific mutualistic interaction between *Ficus* species and their pollinating wasps. While floral scents are often species-specific, intrapopulation scent variation could arise from selection, differences in pollinator assemblages, genetic drift or phenotypic plasticity. Ficus is pollinated by species-specific agaonid wasps in a co-evolved brood-site pollination mutualism. Monoecious *Ficus racemosa* with an Indo-Australasian distribution is supposedly pollinated only by *Ceratosolen fusciceps* throughout. Previous studies reported that pollen-receptive floral scents did not differ between populations 800 km apart (China and Thailand) but differed over 2700 km in southern India. Across south-east Asia, pollinator populations were genetically indistinguishable (1600 km apart) indicating high dispersal ability; southern India and south-east Asia populations were genetically distinct. *Ficus racemosa* may be a species-complex with corresponding pollinator population genetics. Only south Indian scents were earlier investigated. To fill the geographical gap, we selected north-east India (NE) and compared *F. racemosa* pollen-receptive floral scents with south India (SI), 3000 km apart. Volatile organic compounds (VOCs) collected using PDMS were analysed using thermal desorption-GC-MS. We identified around 50 VOCs from each region. Sesquiterpenes dominated NE scents, while fatty acid derivatives dominated SI scents. Major VOCs included (*E*)-β-ocimene, (E)-4,8-dimethyl-1,3,7-nonatriene, zizaene, β-ylangene and epi-zizaene from NE while (*Z*)-3-hexenyl acetate and (*E*)-β-ocimene were most abundant in the SI region. Non-metric multidimensional scaling (NMDS) showed separate clusters between regions that differed significantly (PERMANOVA, $P<0.001$). (*Z*)-3-hexen-1-ol and (Z)-3-hexenyl acetate were the predicted volatiles that separated NE and SI profiles. However, NE scent was more similar to that from Thailand than that from China. The present study revealed that distance between populations affects floral scent even with a highly dispersive pollinator and supports the hypothesis that *F. racemosa* constitutes a species complex.

Keywords: *Ficus*, Fig wasps, Floral scent, Interpopulation variation, Intraspecific spatial variation

S11-O44

Molecular mechanisms underlying flower colour and volatile variation in a deceptive orchid (*Glossodia major*)

Darren Wong[1], James Perkins[1], Zemin Wang[2], Grace Marsh[1], Rod Peakall[1]

[1]*Australian National University, Canberra, Australia.* [2]*Gansu Agricultural University, Lanzhou, China*

Flowers have evolved diverse strategies to attract potential pollinators by conveying visual and olfactory signals. The highly diverse Australian orchids in the subtribe Caladeniinae are dominated by two contrasting deceptive pollination strategies. Food deceptive (FD) species are brightly-coloured (mostly pink/blue/purple) and often strongly-scented while sexual deceptive species are typically dull-coloured (green/red) flowers that are scentless to humans. The waxlip orchid (*Glossodia major*), is a widespread FD species that has bright purple-mauve flowers with a vanilla-like scent (wild-type). However, white-flowered morphs (mutant) are occasionally observed. While little is known about the pollination ecology of the rare white morphs, the purple flowers attract a variety of insects including hoverflies, beetles, and bees, with small native bees the likely pollinator. As the first step toward understanding the chemical and molecular basis of floral colour and floral volatile in this species, we leveraged targeted metabolite and transcriptome analysis of petal tissues across the common purple and rare white morph to ask the following questions: (1) What floral pigments and volatiles are present? (2) Are their pigment and volatile different between the colour morphs? (3) If so, can they be explained by differential expression and sequence variation of specific pigment and volatile pathway genes? Preliminary findings revealed that delphinidin-based anthocyanins and the differential expression of a *dihydroflavonol reductase* (*DFR*) homolog underpins the absence/presence of colour. Furthermore, frameshift mutations in the *DFR* coding sequence led to the premature termination of the functional protein in the white morph. Phenylpropanoid-derived volatiles (e.g. zingerone) were the major scent constituent in purple flowers but was generally absent in white morphs. Differential expression of a eugenol/isoeugenol synthase homolog implicated in phenylpropanoid-derived volatiles biosynthesis was observed. We conclude by discussing the ecological and evolutionary implications of these findings.

Keywords: Orchid, Flower, Anthocyanin, Volatile, Zingerone

S11-O79

Functional evolution of odorant receptors in termites

Jibin Johny[1], Souleymane Diallo[1], Ondřej Lukšan[2], Blanka Kalinová[1], Robert Hanus[2], Ewald Große-Wilde[1]

[1]EXTEMIT-K, Faculty of Forestry and Wood Sciences, Czech University of Life Sciences, Prague, Prague, Czech Republic. [2]Chemistry of Social Insects, Institute of Organic Chemistry and Biochemistry of the Czech Academy of Sciences, Prague, Prague, Czech Republic

Termites are eusocial insects that use a wide range of pheromones for communication, like trail-following pheromones (TFPs), sex-pairing pheromones, and alarm pheromones. The chemical diversity of TFPs displays a clear evolutionary trend: comparing basal lineages to more derived ones indicate a shift from branched C_{13} or C_{14} primary alcohols and aldehydes to C_{12} unsaturated primary alcohols. We hypothesize a complementary evolutionary transition in terms of olfactory perception and thus aim at exploring the functional evolution of chemosensory gene families across termites. We generated antennal transcriptomes of three termite species selected for their specific location on the tree of life -*Inquilinitermes inquilinus, Prorhinotermes simplex* and *Neotermes cubanus*, complementing genome data of other termite species. The data was used to identify our genes of interest, with a focus on olfactory receptors. This enabled transgenic expression of these receptors in *Drosophila melanogaster* in conjunction with electrophysiological measurements for functional characterization.

Keywords: Trail-following Pheromones, Termites, Functional Evolution, Pheromone Receptors

S11-O66

Mapping out olfactomes to unravel tephritid olfactory ecology and evolution

Chaymae Fennine[1], Sebastian Larsson Herrera[2], Advaith Chakravarthy[2], Sergio Angeli[1], Teun Dekker[2]

[1]*Free university of Bozen-Bolzano, Bolzano, Italy.* [2]*Swedish University of Agriculture, Alnarp, Malmö, Sweden*

Most phytophagous insect species have evolved specialized host preferences. In their search for their preferred hosts, they rely strongly on their olfactory system, following an array of odor stimuli. Tephritid fruit flies are also divergent on their oviposition choice. Interestingly, convergence is also common, i.e., a considerable overlap in host preferences that doesn't necessarily follow their genetic relatedness. This study mapped out olfactory responses of four fruit fly species, namely, *Bactrocera oleae*, *Bactrocera latifrons*, *Bactrocera dorsalis* and *Zeugodacus cucurbitae*, towards their host headspace volatiles. The species are related at genus level (*Bactrocera* genus) but differ in their ecological niches. To compare whether these divergences and convergences in host preference were reflected in olfactory sensitivities, we recorded the flies' performed comparative electrophysiological recordings. Using GC-EAD we comprehensively recorded and annotated antennal responses of all four fly species to the headspace volatiles from six different fruits: papaya, mango, cucumber, zucchini, olive and tomato. These comparative data of olfactory tuning of the species was subsequently analyzed and provides important inferences of how olfactory tuning is linked to ecology and/or phylogeny of species and how this can be used to extract candidate attractants for designing lures for pest control.

Keywords: Olfactome, Electroantennography, Host selection, Chemical ecology

Symposium 12: Chemical ecology in biosecurity and conservation (1)

Friday, August 12th
11:25 am - 12:05 pm

Session moderated by Kye-Chung Park

Dual defense system induced by pest damage in on-tree apple fruit
Yuto Ohata, Akihiro Itai, Yoichi Ishiguri, Naoki Mori, Naoko Yoshinaga

Application of chemical ecology to pre-release risk assessments for the intentional introduction of biological control agents
Kye Chung Park, Thomas E. Saunders, Lee-Anne Manning, Mark McNeill, Karina Santos, Gonzalo Avila

S12-O47

Dual defense system induced by pest damage in on-tree apple fruit

Yuto Ohata[1], Akihiro Itai[2], Yoichi Ishiguri[3], Naoki Mori[1], Naoko Yoshinaga[1]

[1]Kyoto University, Kyoto, Japan. [2]Kyoto Prefectural University, Kyoto, Japan. [3]AITC Apple Research Institute, Aomori, Japan

Apple fruits are damaged by various insects. Among them, peach fruit moth *Carposina sasakii* is an important orchard pest in Northeast Asian countries. Its larvae inflict direct damage on fruits such as peach, apple and pear, boring into fruitage. In this reason, spraying insecticides from outside the fruit has no effect on the larvae inside the fruit. In this study, we aim to elucidate the potential defense mechanisms of apple fruits and develop resistant varieties. When compared the survival rates of *C. sasakii* in on-tree and picked fruit, the survival rate in the on-tree fruits was extremely low. We speculated that this was due to chemical defenses expressed only in on-tree fruits. As a result of LCMS analysis, phenolic compounds (chlorogenic acid, *p*-coumaroylquinic acid) were induced by feeding damage in the on-tree fruits. In addition, measurement of peroxidase activity in fruit showed a marked increase localized to the feeding area. Furthermore, the enzyme product of phenolic compounds reacted with peroxidase inhibited the growth of peach fruit moth larvae. These results suggested that apple fruit might have a dual defense system inducing phenolics and peroxidase against feeding damage. Although chlorogenic acid together with peroxidase has been known to work for plant defense against herbivores, no research has been reported on the growth inhibitory by *p*-coumaroylquinic acid. In this presentation, we will discuss the mechanism of the growth-inhibitory activity and the crosstalk between tree and fruit.

Keywords: Apple fruit, Defense system, Pest damage, Phenolics, Peroxidase

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S12-O78

Application of chemical ecology to pre-release risk assessments for the intentional introduction of biological control agents

Kye Chung Park[1,2], Thomas E. Saunders[2,3], Lee-Anne Manning[1,2], Mark McNeill[2,4], Karina Santos[5,2], Gonzalo Avila[2,5]

[1]Bioprotection, New Zealand Institute for Plant and Food Research, Christchurch, New Zealand. [2]Better Border Biosecurity (B3), Wellington, New Zealand. [3]Faculty of Science Research Centres, University of Auckland, Auckland, New Zealand. [4]AgResearch, Christchurch, New Zealand. [5]Bioprotection, New Zealand Institute for Plant and Food Research, Auckland, New Zealand

When parasitoids are intentionally introduced into new environments as biological control agents, thorough pre-release host-specificity testing in containment is a common requirement to understand the potential risk they pose to non-target insects. We conducted a series of electrophysiological recordings and chemical analyses to evaluate the feasibility of applying chemical ecology techniques to pre-release host-specificity testing to predict the risk of parasitoids attacking non-target species, and therefore complement current risk assessment methods and enhance predictions around host-specificity. We used various parasitoid model systems including *Trissolcus* species (egg parasitoids of stink bugs), *Aridelus* species (nymphal parasitoid of stink bugs) and *Microctonus* species (parasitoids of *Sitona* weevils). GC-EAD recordings were conducted to identify olfactory-active compounds produced by host insects for each parasitoid species, and the chemical profiles of the EAD-active compounds were investigated through GC-MS analysis. Our results indicated that different parasitoid species share somewhat similar peripheral olfactory sensitivity for some host volatiles whereas their olfactory sensitivity to other host-related volatile compounds is highly species-specific. Our results also demonstrated that chemical profiles of the EAD-active compounds tend to be species-specific although some volatiles are common to multiple host insect species and can be produced in large quantities. Our results suggest that parasitoids have species-specific peripheral olfactory sensory systems for host and non-host volatiles, and they should be able to discriminate between hosts and non-hosts through olfaction alone. In this context, our study demonstrated that olfactory profiling with a combination of electrophysiology and chemical analyses has strong potential to complement and enhance the accuracy of risk assessments when considering biological control agents for introduction, which can further be confirmed with proper behavioral tests.

Keywords: Biocontrol agent, GC-EAD, Olfactory profiling, Parasitoid, Risk assessment

Symposium 12: Chemical ecology in biosecurity and conservation (2)

Friday, August 12th
12:25 pm - 12:45 pm and
1:30 pm - 2:30 pm

Session moderated byAndrea Clavijo McCormick

Oviposition inhibitory activity of calcium carbonate wettable powder against quarantine pest *Carposina sasakii*
Haruna Kazama, Yuto Ohata, Yoichi Ishiguri, Naoki Mori, Naoko Yoshinaga

Could metabolomic profiling of host plant biochemistry improve the effectiveness and safety of classical weed biocontrol?
D. Paul Barrett, Simon V. Fowler, Arvind K. Subbaraj, Ronny Groenteman, Andrea Clavijo-McCormick

Impact of invasive plants on native plants and herbivores: the role of volatile organic compounds
Evans Effah, Logan Svendsen, D. Paul Barrett, Andrea Clavijo McCormick

The innate floral template and its unlearning in hoverflies
Aditi Mishra, Deepa Rajan, Shannon Olsson

S12-O53

Oviposition inhibitory activity of calcium carbonate wettable powder against quarantine pest *Carposina sasakii*

Haruna Kazama[1], Yuto Ohata[1], Yoichi Ishiguri[2], Naoki Mori[1], Naoko Yoshinaga[1]

[1]*Kyoto Univeristy, Kyoto, Japan.* [2]*Aomori Prefectural Industrial Technology Research Center, Kuroishi, Japan*

The peach fruit moth, *Carposina sasakii*, is a serious pest of rosaceous fruits in Northeast Asia, and recently attracts attention as a quarantine pest. Spraying a calcium carbonate suspension called "White coat" on apple fruit significantly suppresses the *C. sasakii* moth oviposition. The main component of White coat is calcium carbonate micropowder that causes insect slipperiness. Electron microscope analysis revealed that direct contact with calcium carbonate particles clogged the moth arolium, a flexible pad of the tarsus tip to grip smooth surfaces. Interestingly, such adhesion was not observed with White coat treatment. We found that TXIB (2,2,4-trimethyl-1,3-pentanediol diisobutyrate), a rain/wind resistance adjuvant, reduced the calcium carbonate coating's slipperiness while, coincidentally, maintaining the oviposition inhibitory activity of the White coat by its deterrent odorant. Adult female antennae showed GC-EAD responses to TXIB and subsequent oviposition-choice tests revealed clear deterrent activity of TXIB. Female moths spent longer on self-grooming and searching around TXIB-treated fruits. In further investigation of the structure-activity relationship, two monoester analogs of TXIB contained in White Coat were also GC-EAD active but showed no obvious inhibitory activity in the bioassay. The stereochemistry of bioactive TXIB was identified to be 3*R*. (3*S*)-TXIB had no activity and did not negate the enantiomer's activity. This is consistent with the fact that commercially available TXIB is active in racemic mixtures. No sexual bias was observed in GC-EAD analyses. The reason why the moth sense this chemical is unclear, but these findings may provide a useful information on the chemical communication of *C. sasakii* and a sustainable pest control method.

Keywords: TXIB, peach fruit moth, oviposition, enantiomer

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S12-O77

Could metabolomic profiling of host plant biochemistry improve the effectiveness and safety of classical weed biocontrol?

D. Paul Barrett[1], Simon V. Fowler[2], Arvind K. Subbaraj[3], Ronny Groenteman[2], Andrea Clavijo-McCormick[1]

[1]Massey University, Palmerston North, New Zealand. [2]Manaaki Whenua Landcare Research, Lincoln, New Zealand. [3]AgResearch, Lincoln, New Zealand

Classical biological control of weeds reunites specialist co-adapted insect herbivores or pathogens with their original host plant that has become invasive in a new environment and is often the only viable management option, especially on conservation lands. The main challenges to biocontrol agent selection, is the ability to predict agent establishment, effectiveness, and safety. Established protocols are used to assist such assessments, however despite these efforts, many agents either fail to establish or are ineffective when they do. A literature review revealed that where failures occur, seldom is target host plant biochemistry considered a possible factor. Plant metabolomics, is the study of plant biochemistry, utilizing analytical chemistry, bioinformatics and multivariate statistics, to elucidate known and unknown metabolites which allows characterization of plant biochemical phenotypes. Abiotic and biotic factors influence and alter plant biochemical phenotypes, including nutritional value and defensive secondary compounds. These compounds have profound effects on insect performance and population dynamics, both key elements determining establishment and effectiveness of released agents. Using invasive heather (*Calluna vulgaris*) and the biocontrol agent heather beetle (*Lochmaea suturalis*) as a model system, we present evidence of significant changes to this plant's biochemical phenotype between its native range and its invaded range and discuss aspects of how understanding host plant biochemical phenotype, may aid weed biocontrol programs, agent selection, and better predict agent effectiveness.

Keywords: Metabolomics, Plant-phenotype, Biocontrol, Establishment, Effectiveness

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S12-O22

Impact of invasive plants on native plants and herbivores: the role of volatile organic compounds

Evans Effah, Logan Svendsen, D. Paul Barrett, Andrea Clavijo McCormick

Massey University, Palmerston North, New Zealand

Volatile organic compounds produced by plants often modify the behaviour of receiving organisms (including other plants, animals and microbes). Despite that, the potential of invasive plants to disrupt chemical communication in native ecosystems is less-explored. In New Zealand, several introduced weeds, including *Calluna vulgaris* (heather) and *Cytisus scoparius* (Scotch broom), are highly invasive on the North Island Central Plateau. The limited information shows that these invaders, particularly heather, contribute to restructuring this unique ecosystem and decline in arthropod abundance. Still, the exact mechanisms aiding the invasion remain largely unknown. In this study, we explored the response of a common NZ native shrub, *Leptospermun scoparium* (mānuka), towards volatile cues of heather and broom and the impact of volatile cues of heather on a native beetle *Pyronota festiva* (mānuka beetle) and a biocontrol agent *Lochmaea suturalis* (heather beetle). In a semi-field experiment, we found variations in mānuka volatile profiles when paired with conspecifics, heather or broom, with a significant reduction in green leaf volatiles, sesquiterpenes and total volatile emissions when paired with heather. The host-searching and feeding behaviours of *P. festiva* and *L. suturalis* were investigated by offering their host plant, non-host plant and a combination of the two in Y-tube olfactometer and Petri dishes. Results of the Y-tube and Petri dish trials reinforced the high host-specificity of *L. suturalis*. *P. festiva* poorly discriminated between its host and non-host plant volatile cues, although it performed relatively well in Petri dishes, where other cues were present. The results suggest that *P. festiva* could have difficulties finding its host in environments dominated by heather. However, it is likely that upon landing on an unsuitable host, it may use other cues like vision, touch or taste to make feeding decisions.

Keywords: Insect olfaction, Invasive species, Plant communication, Plant-insect-interactions, Plant volatiles

S12-O196

The innate floral template and its unlearning in hoverflies

Aditi Mishra[1], Deepa Rajan[2], Shannon Olsson[1]

[1]*National Center for Biological Sciences, Bengaluru, India.* [2]*University of California San Francisco, San Francisco, USA*

Most insects are solitary in nature. Hence, these generalists must efficiently locate and identify food within hours of emergence. Their affinity to relevant food cues must be specific enough discriminate objects, but general enough to map the diversity of food. How these tiny brains encode these cues innately is an area of intense study for ecologists and neuroscientists alike. We used the solitary generalist pollinator *Eristalinus aeneus* to understand how the innate floral search template can arise through a small number of sensory cues spanning multiple sensory modalities. Using field and laboratory behavioral assays and electrophysiology, we found that innate floral choices of the hoverfly *E. aeneus* are a product of contextual integration of broad plant based olfactory cues and the visual cues of high spectral intensity and radial symmetry. Further, we investigated if hoverflies can learn to lose attraction to innately attractive floral models in paradigms where these innately attractive floral models were paired with aversive stimuli. We found that hoverflies can learn to avoid their innately attractive floral objects after several trials, and they retain the aversion robustly, with some flies retaining this memory for the rest of their lifetime. Conversely, hoverflies learn to associate neutral objects with food within a few exposures of it, but this information is not retained for more than 48-72 hours. Hoverflies have a simple multimodal innate floral template which helps them find flowers. This coupled with their ability to quickly associate neutral objects with appetitive cues can make them important, resilient pollinators. Globally hoverflies visit 72% of all food crops and wild flowers, estimated at $300 billion annually. Coupled with their resilience to changes in land use and virtual immunity to colony collapses, hoverflies are powerful tool to mitigate the pollinator crisis.

Keywords: Innate behavior, Generalist pollinator, Hoverfly, Object recognition, Learning, Memory

Symposium 13: Insect behavior and evolution (1)

Friday, August 12th
12:05 pm - 12:45 pm

Session moderated by Koji Noge

***Drosophila melanogaster* stress odorant is an interspecific alarm cue.**
Ryley Yost, Yu Min Liang, Megan Stewart, Selwyn Chiu, Andrew Greco, Shirley Long, Ian McDonald, Tim McDowell, Jeremy McNeil, Anne Simon

The impact of environmental stressors on mating behaviour of wild bees
Samuel Boff, Taina Conrad, Josué Raizer, Marten Wehrhahn, Melis Bayer, Anna Friedel, Panagiotis Theodorou, Thomas Schmitt, Daniela Lupi

S13-O161

Drosophila melanogaster stress odorant is an interspecific alarm cue.

Ryley Yost[1], Yu Min Liang[1], Megan Stewart[1], Selwyn Chiu[1], Andrew Greco[1], Shirley Long[1], Ian McDonald[1], Tim McDowell[2], Jeremy McNeil[1], Anne Simon[1]

[1]*University of Western Ontario, London, Canada.* [2]*Agriculture and Agrifood Canada, London, Canada*

While it has been shown that *Drosophila melanogaster* adults emit an olfactory alarm signal, termed the Drosophila stress odorant (dSO) and individuals avoid areas previously occupied by stressed conspecifics, little is known about the emission of, and response to dSO. Using a binary choice assay, we have found that neither the emission of, or response to, dSO is affected by the age and sex of emitters or the time of the day the assays are conducted. However, both the sex and mating status of the receives affect the response to dSO. The profiles of dSO released by *D. melanogaster, D. simulans*, and *D. suzukii* differ but the responses observed were not species specific, so dSO should be considered a general alarm signal rather than an alarm pheromone. However, the response levels to both intra and inter-specific cues differed between species and possible reasons for these differences are discussed.

Keywords: *Drosophila*, Alarm cue, Intra and interspecific communication

S13-O203

The impact of environmental stressors on mating behaviour of wild bees

Samuel Boff[1], Taina Conrad[2], Josué Raizer[3], Marten Wehrhahn[4], Melis Bayer[5], Anna Friedel[6], Panagiotis Theodorou[6], Thomas Schmitt[4], Daniela Lupi[7]

[1]Department of Evolutionary Ecology and Conservation Genomics, Ulm, Germany. [2]University of Bayreuth, Department of Evolutionary Animal Ecology, Bayreuth, Germany. [3]Federal University of Grande Dourados, Faculty of Biology and Environmental Sciences, Dourados, Brazil. [4]University of Würzburg, Biocentre, Animal Ecology and Tropical Biology, Würzburg, Germany. [5]Ludwig Maximillians University, Department of Neurobiology, Munich, Germany. [6]Martin Luther University Halle-Wittenberg, Institute of Biology, General Zoology, Halle, Germany. [7]University of Milan, Department of Food, Environmental and Nutritional Sciences, Milan, Italy

Females of several beneficial insect species, including the solitary horned mason bee Osmia cornuta (Megachilidae), use complex pre-copulatory behaviors and odors of males as cues to assess male mating quality, which may be disrupted by sub-lethal exposure to agrochemicals. Herein, it was found that exposure to a common pesticide used to control fungi on important crop species significantly reduced mating success of male horned mason bees. Males exposed to the pesticide exhibited reduced thoracic vibrational signaling and an altered cuticular hydrocarbon profile, which are traits used by females to assess male quality. These results indicate that pesticides may negatively impact bee mating behaviors and reproductive success, with long term effects that could explain reduced pollinator populations.

Keywords: -

Symposium 13: Insect behavior and evolution (2)

Friday, August 12th
1:30 pm - 3:10 pm

Session moderated by Koji Noge

Identifying livestock dung headspace volatiles that drive the preference of the dung beetle *Bubas bison* (Coleoptera: Scarabaeidae)
Nisansala N. Perera, Paul Weston, Russell Barrow, Leslie Weston, Geoff Gurr

Social behavior and colony trajectory in bumble bees are shaped by the brood
Etya Amsalem

Multimodal signaling by queens regulates mating behavior in the bumble bee *Bombus impatiens*
Sarah K. Spence, Etya Amsalem

The defensive eversible organ in papilionids: source of volatiles, exposure of toxic hemolymph and aposematic warning
Valeria Palma-Onetto, Jan Bergmann, Marcia González-Teuber

Chemical substances underlying different host utilization between host races in the leaf-mining moth *Acrocercops transecta* (Lepidoptera: Gracillariidae)
Hajime Ono, Issei Ohshima

S13-O150

Identifying livestock dung headspace volatiles that drive the preference of the dung beetle *Bubas bison* (Coleoptera: Scarabaeidae)

Nisansala N. Perera[1,2], Paul Weston[1,2], Russell Barrow[1], Leslie Weston[1,2], Geoff Gurr[1,3]

[1]Gulbali Institute of Agriculture, Water and Environment, Charles Sturt University, Wagga Wagga, Australia. [2]School of Agriculture, Environment and Veterinary Sciences, Charles Sturt University, Wagga Wagga, Australia. [3]School of Agriculture, Environment and Veterinary Sciences, Charles Sturt University, Orange, Australia

Dung beetles (Coleoptera: Scarabaeoidae) are known to rely on volatile cues that play a significant role in the location and discrimination of food resources. They have been reported to discriminate among livestock dung types, thereby exhibiting behavioral preferences. However, the role of volatile organic compounds (VOCs) in dung localization and preference remains largely unexplored in dung beetles. The objective of this study was to determine the olfactory preference of the dung beetle *Bubas bison* to fresh livestock dung and to characterize the dung VOCs associated with dung attraction. We employed cage olfactometer bioassays to assess the behavioral responses of adult *B. bison* to VOCs from fresh horse, sheep, and cattle dung, and concurrent volatilome analysis to characterize volatilomes of these dung types. In the assays, *B. bison* exhibited greater attraction to horse dung than to cattle dung, and preferred dung from horses fed a pasture-based diet over those fed lucerne hay. Chemometric analysis revealed that headspace volatilomes of the corresponding dung samples from each livestock species had unique volatile profiles with VOCs belonging to diverse chemical groups. However, the composition and abundance of annotated VOCs varied with dung type and livestock diet. From electroantennography and supplementary olfactometry, we found strong evidence that indole, butyric acid, butanone, p-cresol, skatole, and phenol, as well as toluene, may have been associated with the attraction of *B. bison* to dung, with a mixture of these VOCs significantly more attractive than individual constituents. The activity of these identified compounds will also be assessed under field conditions, with the eventual goal of developing a robust and efficient trapping system for monitoring dung beetle activity.

Keywords: Dung volatiles, VOCs, Olfactometer, EAG, Dung beetle attraction

S13-O10

Social behavior and colony trajectory in bumble bees are shaped by the brood

Etya Amsalem

Penn State University, University Park, USA

A hallmark of sociality is reproductive division of labor, mediated by pheromones, between reproductive and sterile females. While the role of the queen in regulating worker reproduction in social insects attracted much of the attention in selected model organisms, the role of the brood and its ability to manipulate worker reproduction via pheromones remained poorly studied. Here, we used bumble bees to understand the various roles of brood in maintaining and manipulation the structure of a social colony. Findings show that the brood regulates workers' behavior and physiology in various ways. Young larvae were shown to inhibit egg laying behavior in workers whereas pupae induced the opposite effect. Both effects are dose-dependent, not influenced by relatedness or the previous experience of workers and are mediated by both unique behaviors exhibited by the brood and chemical signals produced by young larvae and pupae. We further show that the impact of the queen on worker reproduction is partial without the brood, and that the presence of brood regulates large-scale processes at the colony level such as the beginning of the competition phase and the production of sexuals. Despite the various impacts brood has on workers, a transcriptome analysis of workers' brains in response to young larvae demonstrate weak impacts on gene expression in comparison with the impact of the queen. These studies overall point to a significant impact of the brood on the social behavior of bees and demonstrate the need to examine the effects of brood and the chemical mechanisms underlying these effects more thoroughly across other species.

Keywords: Bumble bees, Larvae, Pupae, Reproduction, Pheromones, Hunger signal, Gyne production

S13-O75

Multimodal signaling by queens regulates mating behavior in the bumble bee *Bombus impatiens*

Sarah K. Spence, Etya Amsalem

Department of Entomology, Center for Chemical Ecology, Center for Pollinator Research, Huck Institutes of the Life Sciences, Pennsylvania State University, University Park, PA 16802, USA

Insect mating is often regulated by sex pheromones that convey information about mate species, quality, and receptiveness. These signals are often released from exocrine glands and elicit response in conspecifics. In bumble bees, sex pheromones in males have been investigated intensively but the queen pheromones or their glandular source remained poorly studied. Bumble bee males mark mating sites with labial gland secretions, however, the male's signaling alone has not been found to stimulate mating behavior. Our study examines the signals produced by the virgin queens and their role in short range attraction and mating behavior in Bombus impatiens. We propose that males rely on multimodal signaling, combining both visual and chemical signals produced in the queens' Dufour's and labial glands. Using a 2-choice bioassay, we show that males are attracted to both live and dead queens over workers but are not attracted to dummy queens that were stripped from their chemical odors, or to the chemical signals alone. We further show that this attraction does not occur under red light (a spectrum that the bees cannot see), suggesting males are relying on a combination of visual and chemical signals, but not on visual or chemical signals alone. Current experiments aim to identify the source of the pheromone by testing male attraction to the total secretion and selected compounds produced in the Dufour's and labial glands of the gynes. These results demonstrate the role of virgin queens in stimulating mating behavior. Furthermore, our study models the importance of multimodal signal bioassays for accurate sex pheromone identification.

Keywords: Sex pheromone, Mating, Bumble bees, Visual signals

S13-O147

The defensive eversible organ in papilionids: source of volatiles, exposure of toxic hemolymph and aposematic warning

Valeria Palma-Onetto[1], Jan Bergmann[2], Marcia González-Teuber[1]

[1]*Universidad Católica de la Santísima Concepción, Concepción, Chile.* [2]*Pontificia Universidad Católica de Valparaíso, Valparaíso, Chile*

Lepidoptera larval period largely defines the development and performance that butterflies will have in their adult state. However, the larval state is highly vulnerable and will suffer the attack of different predators. Accordingly, Lepidoptera larvae have developed diverse defensive strategies, such as the use of defensive specialized exocrine glands. Papilionidae family (Insecta: Lepidoptera) have developed an eversible organ whose defensive function stands out, the osmeterium. Nevertheless, despite many studies that have aimed to discover the chemistry and ethology of the osmeterium, the mechanisms underlying the osmeterium defensive function were still unclear. Here, we aimed to disentangle the functionality of the osmeterium, focusing on describing the biology, structure, ultrastructure, morphology, chemistry and ethology of the osmeterium in *Battus polydamas archidamas*, a species well known for using the osmeterium as a main defensive strategy. We observed that *Battus polydamas archidamas* osmeterium is composed of two differentiated sectors: the tubular arm, which contains epidermal triangular cells; and, an ellipsoid gland located at the base of the dorsal (posterior) side of each arm, which contains several organelles associated with a secretory function. The eversion and retraction of the osmeterium are mainly related to hemolymph pressure, which seems to comply an important role in its whole defensive function. In fact, chemicals associated with the osmeterium are highly toxic, but most of them are also present in the hemolymph running through the larva, except by germacrene A which would be specifically secreted by the ellipsoid gland. Bioassays confirmed a defensive function of osmeterial content but also of the larva hemolymph. Our results lead us to conclude that the osmeterium is a full defensive organ, although not a gland on its whole, and its defensive mechanism would be mainly due to its property as a source of volatiles, exposure to toxic hemolymph and aposematic warning.

Keywords: -

S13-O49

Chemical substances underlying different host utilization between host races in the leaf-mining moth *Acrocercops transecta* (Lepidoptera: Gracillariidae)

Hajime Ono[1], Issei Ohshima[2]

[1]*Kyoto University, Kyoto, Japan.* [2]*Kyoto Prefectural University, Kyoto, Japan*

The importance of plant chemistry in the host specialization of phytophagous insects has been emphasized. However, only a few chemicals associated with host shifting have been characterized. Herein, we focused on two host races in a leaf-mining micro-moth, *Acrocercops transecta* (Gracillariidae), in which the ancestral race utilizes juglandaceous species (Juglandaceae) while a derived race only associated with *Lyonia ovalifolia* (Ericaceae). To understand the process of host shifting between distantly related plant taxa, from Juglandaceae to Ericaceae, we addressed elucidation of the roles of plant chemicals underlying the different oviposition preferences and larval performance between the two host races. i) Females of the *Lyonia* race lay eggs on a cover glass treated with an extract from *L. ovalifolia* leaves. Therefore, the extract was fractionated using multiple chromatographies to isolate oviposition stimulants. We characterized two analogous *Lyonia*-specific triterpenoid glycosides as oviposition stimulants. Notably, the *Juglans* race did not respond to the oviposition stimulant. Therefore, ancestors of the *Lyonia* race probably acquired the ability to detect the newly encountered triterpenoid glycosides as oviposition stimulants. ii) Larvae of the derived *Lyonia* race can survive and grow on juglandaceous plants into adulthood, although larvae of the *Juglans* race cannot survive on the derived host, *L. ovalifolia*. Ericaceae plants including *L. ovalifolia* specifically contain toxic diterpenoids, grayanotoxins. Therefore, we forced larvae to ingest grayanotoxins by injecting solutions containing each compound into mines where larvae inhabited. We tested available grayanotoxins, grayanotoxin III (GIII), and rhodojaponin III (RIII). The *Juglans* race larvae died by injection of either GIII or RIII at ca. 0.2 mM. In contrast, almost all larvae of the *Lyonia* race survived after ingestion of GIII or RIII even at 3 mM. These results indicate that the acquisition of larval tolerance to the grayanotoxins played important roles in the host shifting to the toxic plant.

Keywords: Phytophagous insect, Host shift, Host race, Oviposition stimulants, Grayanotoxins

Symposium 14: Environmental impacts of plant-insect interaction

Friday, August 12th
1:30 pm - 3:30 pm

Session moderated by Wen-Po Chuang

Plant responses to the third trophic level: An underestimated interplay shaping multitrophic interactions
Christelle Robert, Paul Himmighofen, Xi Zhang, Pierre Mateo

Environmental impacts on the insect resistance in rice
Wen-Po Chuang, Zhi-Wei Yang, Charng-Pei Li, Shou Horng Huang

Water stress effects on arthropod communities and plant-aphid interactions: Investigating the impact of water stress intensity
Jessica Kansman, Deborah Finke

Copper elemental defense of rice against *Cnaphalocrocis medinalis*
Boon Huat Cheah, Wen-Po Chuang, Jing-Chi Lo, Yi Li, Chih-Yun Cheng, Zhi-Wei Yang, Chung-Ta Liao, Ya-Fen Lin

Low water availability enhances volatile-mediated direct defenses but disturbs indirect defenses against herbivores
Po-An Lin, Gary Felton

S14-O156

Plant responses to the third trophic level: An underestimated interplay shaping multitrophic interactions

Christelle Robert, Paul Himmighofen, Xi Zhang, Pierre Mateo

University of Bern, Bern, Switzerland

Interactions between plants, herbivores, and herbivore natural enemies, so-called tritrophic interactions, are important determinants of ecological processes and crop yields. Interestingly, plants can respond directly to natural enemies of herbivores. However, the mechanisms and ecological relevance of these responses are not well understood. We characterized how maize plants respond to soil-dwelling entomopathogenic nematodes, natural enemies of root herbivores, and assessed the ecological relevance of this phenomenon under controlled and field conditions. Under exposure to entomopathogenic nematodes, no change in the root metabolism could be observed. Yet, the presence of the third trophic level modulated the root exudation of sugars, amino acids, and benzoxazinoids. Entomopathogenic nematodes further induced systemic changes in the leaf primary metabolism. However, some of the plant responses were not consistent over different experiments. We thus repeated the assay under field conditions to identify (agro)ecologically relevant metabolomic changes and their impact on plant-environment interactions. Maize plants were able to perceive and respond to the presence of entomopathogenic nematodes in two independent field experiments. While nematode-induced metabolic changes are still under investigation, the presence of entomopathogenic nematodes in soil reduced natural infestation rates by the stem borer aboveground. We argue that the time is ripe to acknowledge the relevance of plant direct responses to the enemies of herbivores and discuss potential adaptive values of this phenomenon.

Keywords: Multitrophic Interactions, Entomopathogenic Nematodes, Belowground Interactions, Exudation, Metabolomic

S14-O14

Environmental impacts on the insect resistance in rice

Wen-Po Chuang[1], Zhi-Wei Yang[2], Charng-Pei Li[3], Shou Horng Huang[4]

[1]National Taiwan University, Taipei, Taiwan. [2]Taoyuan District Agricultural Research and Extension Station, Taoyuan City, Taiwan. [3]Taiwan Agricultural Research Institute, Taichung City, Taiwan. [4]Chiayi Agricultural Experiment Station, Chiayi, Taiwan

The impact of climate change on insect resistance genes is elusive. Hence, we investigated the responses of rice near-isogenic lines (NILs) that carry resistance genes against brown planthopper (BPH) under different environmental conditions. We tested these NILs under three environmental settings (the atmospheric temperature with corresponding carbon dioxide at the ambient, year 2050 and year 2100) based on the Intergovernmental Panel on Climate Change prediction. Comparing between different environments, two of nine NILs that carried a single BPH-resistant gene maintained their resistance under the environmental changes, whereas two of three NILs showed gene pyramiding with two maintained BPH resistance genes despite the environmental changes. In addition, two NILs (NIL-*BPH17* and NIL-*BPH20*) were examined in their antibiosis and antixenosis effects under these environmental changes. BPH showed different responses to these two NILs, where the inhibitory effect of NIL-*BPH17* on the BPH growth and development was unaffected, while NIL-*BPH20* may have lost its resistance during the environmental changes. In addition, nitrogen is an essential macronutrient for plant growth and development. Crops with a high nitrogen input usually have high yields. However, outbreaks of BPH frequently occur on rice farms with excessive nitrogen inputs. The amount of nitrogen applied had an impact on the resistance of some lines with BPH resistance genes. In addition, three NILs (NIL-*BPH9*, NIL-*BPH17*, and NIL-*BPH32*) were further examined for antibiosis and antixenosis under varying nitrogen regimes. Our results indicated that the resistance of three tested NILs did not respond to different nitrogen regimes and that NIL-*BPH17* exerted the most substantial inhibitory effect on BPH growth and development.

Keywords: *Nilaparvata lugens*, Near-isogenic lines, Climate change, Insect resistance gene, Nitrogen

S14-O37

Water stress effects on arthropod communities and plant-aphid interactions: Investigating the impact of water stress intensity

Jessica Kansman[1], Deborah Finke[2]

[1]*Penn State University, State College, USA.* [2]*University of Missouri, Columbia, USA*

Climate change is causing an increase in drought events around the world. Water limitation affects the quality and quantity of plant resources for herbivorous insects, leading to cascading effects for higher trophic levels. Additionally, water stress occurs along gradients of intensity, and the impact of drought intensity on plant-insect interactions is understudied. Our research examined the response of arthropod communities to water limitation in wheat (*Triticum aestivum* L.), and further assessed the plant-mediated mechanisms driving these responses. In the field, we found that any level of water limitation reduced aphid abundance, and aphid population response was not simply driven by reduced plant biomass. On the contrary, predatory insect abundance appears to be strongly mediated by changes in plant biomass as a result of water limitation instead or other stress-induced changes within the plant. We further investigated which plant-mediated mechanisms mediate the aphid response to drought by assessing plant nitrogen, amino acids, sugars, and phytochemistry. Water limitation did not affect whole-plant nitrogen; however, water limitation did reduce amino acid concentration and increase sugars, but only under high stress intensity. The phytohormones abscisic acid (ABA), jasmonic acid (JA), and salicylic acid (SA), and the expression of their associated gene transcripts were also differentially affected by water stress intensity. In well-watered conditions, aphid feeding increased concentrations of the defense-related hormones SA and JA over time; however, any amount of water limitation prevented aphid induction of JA. Although aphids may experience a reprieve from JA-related defenses in stressed conditions, SA levels remain high in response to aphid feeding, indicating aphids are still vulnerable to SA-related defenses. Understanding the mechanisms driving aphid and plant performance under water stress conditions can improve our ability to predict how aphid populations and arthropod communities will respond to climate change.

Keywords: Abiotic stress, Plant-Insect Interactions, Aphids, Community Ecology

S14-O84

Copper elemental defense of rice against *Cnaphalocrocis medinalis*

Boon Huat Cheah[1], Wen-Po Chuang[1], Jing-Chi Lo[2], Yi Li[1], Chih-Yun Cheng[3], Zhi-Wei Yang[3], Chung-Ta Liao[4], Ya-Fen Lin[1]

[1]*Department of Agronomy, National Taiwan University, Taipei, Taiwan.* [2]*Department of Horticulture and Biotechnology, Chinese Culture University, Taipei, Taiwan.* [3]*Crop Improvement Division, Taoyuan District Agricultural Research and Extension Station, Taoyuan City, Taiwan.* [4]*Crop Environment Division, Taichung District Agricultural Research and Extension Station, Changhua County, Taiwan*

Farmers lose an average of 37% of their rice (*Oryza sativa* L.) yield to pests and diseases every year. According to the elemental defense hypothesis, accumulated metals in plant tissues may enhance plant defense against herbivorous insects and pathogens. However, it is unclear whether the exogenous elemental application can induce rice defense to these biotic stressors. Here, we examined the potential of copper (Cu) and iron (Fe) micronutrient supplements for the protection of rice plants against a major rice insect pest, the rice leaffolder (*Cnaphalocrocis medinalis*). *C. medinalis*-susceptible rice seedlings were grown under different concentrations of $CuSO_4$ or Fe-citrate, and the effects of elemental treatments on larval growth, as well as on rice growth and yield components were evaluated. Our results showed that intermediate levels of Cu (20 µM $CuSO_4$) and high Fe (265 mg/L Fe) did not inhibit the growth of *C. medinalis* larvae, but significantly inhibited rice growth and reduced grain yield. A high level of Cu (80 µM $CuSO_4$) was found to inhibit *C. medinalis* larval growth and pupal development but at the same time adversely affected the growth of rice plants. Prolonged treatment with 10 µM $CuSO_4$ had no adverse effects on rice growth or yield components. These results indicate that pest control based on the application of Cu may be possible through the intervention of nanotechnology to achieve effective larval sublethal and lethal doses at less than 10 µM $CuSO_4$.

Keywords: Micronutrient, Trace elements, Inorganic chemical defense, Pest management

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S14-O42

Low water availability enhances volatile-mediated direct defenses but disturbs indirect defenses against herbivores

Po-An Lin[1], Gary Felton[2]

[1]*University of Bern, Bern, Switzerland.* [2]*Pennsylvania State University, University Park, USA*

Interactions between plants and natural enemies of insect herbivores influence plant productivity and survival by reducing herbivory. Plants attract natural enemies via herbivore-induced plant volatiles (HIPVs), but how water availability (WA) influences HIPV-mediated defenses is unclear. We use tomato (*Solanum lycopersicum*), tomato fruitworm (*Helicoverpa zea*), and two natural enemies, the parasitoid wasp (*Microplitis croceipes*) and the predator spined soldier bug (*Podisus maculiventris*), to investigate the effect of WA on HIPV emission dynamics and associated plant defense. We show that low WA initially increases total HIPV emission by tomato on the first day of herbivore exposure and, in contrast, reduces HIPV emission on the second day. Low WA enhances HIPVs that are mostly found in tomato trichomes. Notably, some volatiles inhibited by low WA are known attractants of natural enemies. Evidence from Y-tube and in-cage behavioral assays indicates that changes in HIPV emissions by low WA compromise the ability of tomato plants to attract natural enemies. Based on our results, we propose a hypothesis where plants respond to low WA by enhancing repellent HIPV emissions and reducing the emission of HIPVs that attract natural enemies, which disrupts natural enemy-mediated plant indirect defenses, but enhances plant direct defense against herbivores.

Keywords: -

Posters (In-Person & Virtual)

MONITOR 1

Adriana	**Moriguchi Jeckel**	Benzoxazinoid detoxification by the Western Corn Rootworm	TU12:30(1)
Gabriele	**Bumbulyte**	Behavioural effects of essential oils and single VOCs on mealworm (*Tenebrio molitor* L.) larvae	TU13:00(1)
Mayuri	**Shewale**	Deciphering the semiochemistry underlying host selection in *Ips duplicatus* Sahlberg. Antennal morphological and physiological study.	WE12:30(1)
Stefan	**Schulz**	MACE - Mass Spectra for Chemical Ecology	WE13:00(1)

MONITOR 2

Andres	**Gonzalez**	Chemical communication studies in Cyrtogenius luteus, a bark beetle recently introduced in southern South America	TU12:30(2)
Jessil	**Pajar**	Metabolic profile shifts in *Brassica nigra* leaves after belowground nematode infection affect aphid performance aboveground	TU13:00(2)
Mohammed	**Sajidha**	Laboratory glass chamber to conduct dynamic headspace analysis of insect volatiles	WE12:30(2)
Sungjun	**Choung**	JA-dependent pith lignification in *Nicotiana attenuata* performs a defensive function against the stem-boring herbivore	WE13:00(2)

MONITOR 3

Anjana	**Unni**	The behavior of *Locusta migratoria* to conspecific odors	TU12:30(3)
Junheon	**Kim**	Identification of a sex pheromone of hibiscus-leaf caterpillar moth	TU13:00(3)
Nils	**Schöfer**	Sublethal effects of insecticides on partner and host finding of Lariophagus distinguendus	WE12:30(3)
Tammy	**Shim**	Odorant receptors' structure – function relationships are decoded by in silico / in vitro joint approaches. Mammal receptors as a test case.	WE13:00(3)

MONITOR 4

Barbora	**Stribrska**	Measurable physiological and biochemical changes in Norway spruce freshly attacked by *I. typographus*, as a potential base of alternative early detection methodology.	TU12:30(4)
Kevin	**Bartl**	Pollution in beeswax	TU13:00(4)
Rasa	**Čepulytė**	On chemical ecology of plant-parasitic nematodes	WE12:30(4)
Tsuyoshi	**Maruoka**	Search for active role of FACs hydrolysis with CRISPR-Cas9	WE13:00(4)

MONITOR 5

Dominykas	**Aleknavičius**	Feed associated bacteria effect on behavioural responses of edible cricket cultivars	TU12:30(5)
Laima	**Blazyte-Cereskiene**	VOCs of sea buckthorn fruits attractive for fruit fly *Rhagoletis batava*: search for kairomone compounds	TU13:00(5)
Shinnosuke	**Mori**	Floral color strategy in *Camellia* attracting different pollinators	WE12:30(5)
Yuki	**Miyake**	Pheromone synthesis of Coleopteran pests, *P. californicus* and *O. rhinoceros*, and their application in field	WE13:00(5)

MONITOR 6

Eraldo	**Lima**	Method to access the mating status of males collected in pheromone traps. A study with *Tuta absoluta* (Lepidoptera: Gelechiidae) in wind tunnel and field	TU12:30(6)
Natália	**Ribas**	Bt-protein expression impairs early *Spodoptera frugiperda* oviposition induced volatiles and egg parasitoid attraction in maize plants	TU13:00(6)

S6-P168

Benzoxazinoid detoxification by the Western Corn Rootworm

Adriana M. Jeckel[1], Meng Ye[2], Paul Himmighofen[1], Franziska Beran[3], Christelle A. M. Robert[1]

[1]*Institute of Plant Sciences, University of Bern, Bern, Switzerland.* [2]*Tea Research Institute, Chinese Academy of Agricultural Sciences, Hangzhou, China.* [3]*Max Planck Institute for Chemical Ecology, Jena, Germany*

Highly adapted herbivores can highjack plant secondary metabolites for their own benefit. For instance, the Western Corn Rootworm, *Diabrotica virgifera virgifera*, can detoxify and sequester maize benzoxazinoids for its own nutrition and defense against entomopathogenic nematodes. However, the detoxification mechanisms remain unclear. We investigated how the insect larvae detoxify MBOA (6-methoxy-2-benzoxazolinone), a benzoxazinoid breakdown product formed upon maize injury. We observed that MBOA is stabilized by the insect through *N*-glucosylation. By combining transcriptomic analyses and genome annotation available in NCBI, we identified 50 genes encoding for *N*-glucosyltransferases in the insect. We quantified the gene expression of these genes in insects reared in diet containing different levels of benzoxazinoids and evaluated possible candidate genes for further analysis. RNAi-mediated silencing of 11 of these candidates was tested to evaluate a difference in levels of MBOA-Glc in the insect. We observed a decrease in MBOA-Glc formation by silencing two of these candidates. However, when these genes were expressed in Sf9 cells and screened for *N*-glucosylation activity, there was no formation of MBOA-Glc. Our next step is to screen for other possible candidate genes responsible for stabilizing MBOA. Discovering how a pest herbivore detoxifies benzoxazinoids and their derivatives may contribute to develop targeted pest management strategies.

Keywords: Sequestration, Maize, *Diabrotica virgifera virgifera*, Glucosyltransferase

S2-P12

Behavioural effects of essential oils and single VOCs on mealworm (*Tenebrio molitor* L.) larvae

Gabrielė Bumbulytė, Vincas Būda

Nature Research Centre, Vilnius, Lithuania

Mealworms (*Tenebrio molitor* L.) are easily cultivated, have a relatively large biomass increase, and are great source of proteins, thus larvae could replace animal meat (partially). In insect farms it is very important effectively harvest insect biomass from substrate. Application of repellents could be effective tool to manipulate behaviour of the insect larvae to give them away from substrate. We analysed if essential oils, could be used as natural repellents for this purpose. Behavioural tests were performed using essential oils of 6 plants under laboratory conditions. Behaviour of larvae was recorded and monitored by a computer program – EthoVision XT 12 (Noldus, the Netherlands) and analysed as two-choice assay. Behaviour of each larva was recorded for 5 minutes, and 10 replicates were performed with each essential oil or compound tested. The results revealed that the most repellent for the mealworm larvae were the essential oils of thyme (*Thymus vulgaris* L.) and spearmint (*Mentha spicata* L.). GC-MS analysis revealed composition of the essential oils, and synthetic analogs of the most abundant components were tested. Data on the most repellent compounds will be presented.

Keywords: Behaviour, Entomophagy, Food-source, Mentha, Thymus

S7-P64

Deciphering the semiochemistry underlying host selection in *Ips duplicatus* Sahlberg. Antennal morphological and physiological study

Mayuri Shewale, Jaromír Blaha, Jan Prchal, Jaromír Hradecký, Blanka Kalinová

Faculty of Forestry and Wood Sciences, Czech University of Life Sciences Prague, Czech Republic, Prague, Czech Republic

The double-spined spruce bark beetle *Ips duplicatus* has become infamous secondary pest of spruce over past few decades in many Central European countries. *I. duplicatus* has a quite similar biology to the notoriously known European spruce bark beetle Ips typographus. Both species share common primary host, Norway spruce (Picea abies (L.), but elicits differences with respect to the host preference and selection. Olfaction plays a crucial role for host selection in bark beetles and different structures on the antenna define distinct function in detection of odors from the chemosphere. While extensive research has been done on *I. typographus*, the olfactory aspects of *I. duplicatus* remain unexplored. In the current study, we investigated the diversity of sensillar types on the antennal surface of *I. duplicatus* using scanning electron microscopy. Additionally, we collected volatiles using headspace method from spruce varying in their age and height to analyze secondary metabolites. Microscopic studies revealed presence of five types of sensilla with no sexual dimorphism was observed between the sexes. Comprehensive two-dimensional gas chromatography coupled with time-of-flight mass spectrometer (GCxGC-TOF-MS) analyses revealed differences in the chemical profile of spruce sample volatiles with respect to age and height. Gas chromatography-electroantennographic detection (GC-EAD) analysis with *I. duplicatus* antennae depicted many antennally active volatiles. The volatile spectrum detected by *I. duplicatus* antenna largely overlapped with those reported for *I. typographus*. The morphological studies and identification of active volatiles will provide a basis for future investigation of *I. duplicatus* olfactory receptors tuning and specificity using single sensillum recording.

Keywords: *Ips duplicatus*, Antennal sensilla, Host volatiles, GCxGC-TOF-MS, GC-EAD

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S3-P91

MACE - Mass Spectra for Chemical Ecology

Stefan Schulz, Anton Möllerke

Technische Universität Braunschweig, Braunschweig, Germany

Mass spectral libraries of EI-mass spectra are an important tool in the identification and structure elucidation of natural products by GC/MS, a typical task in Chemical Ecology. Unfortunately, spectra of new compounds are usually only available as figures even in recent publications, making it tedious to keep track of them and difficult to integrate them into user libraries. We therefore implemented an open access data repository for EI mass spectra, called MACE. In this database high quality spectra not found in common databases can be downloaded as a collection in a simple format, readily integrable into local spectral databases. The spectra are from original publications that have been synthesized or isolated. Compounds found widespread commercial databases such as NIST 17 or Wiley 7 are not included. MACE is therefore an add-on database of high-quality EI-mass spectra. MACE is designed as a community effort with open access, needing input from research groups worldwide. The list of compounds will be continuously extended, hopefully with spectra submitted by other groups. We have started with compounds from our own group, but further additions are very welcome. Submitted spectra will be checked for quality and added to MACE. The complete MACE library text file can be downloaded from the research data repository of TU Braunschweig, thus ensuring long-term data storage. The data can be freely distributed for non-commercial use according to the creative commons license CC-BY-SA.

Keywords: GC/MS, Open access, Database, Mass spectra, Community effort

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S2-P163

Chemical communication studies in *Cyrtogenius luteus*, a bark beetle recently introduced in southern South America

Gianna Zinola[1], Mariela Suárez[2], Gissel Cantero[2], Analía García Feijó[2], Carmen Rossini[1], Gonzalo Martínez[2], Andrés González[1]

[1]*Laboratory of Chemical Ecology, Faculty of Chemistry, Universidad de la República, Montevideo, Uruguay.* [2]*Laboratory of Entomology, National Forestry Research Program, Instituto Nacional de Investigación Agropecuaria, Tacuarembó, Uruguay*

International trade expansion has caused a steady increase in the introduction of non-native species into new habitats. Bark beetles are commonly transported in solid wood packaging materials, sometimes causing profound alterations in native forests as well as economic damage. About a decade ago the bark beetle *Cyrtogenius luteus* (Curculionidae: Scolytinae), native to Asia, was first reported in South America. Population outbreaks have been reported in pine plantations in Uruguay, based on observed damage and trap records using pine volatile attractants. No pheromones have been reported for *C. luteus*, so we performed chemical and behavioral studies in search for aggregation pheromones that may increase trapping efficiency. Male and female *C. luteus* came from cut *Pinus taeda* trunks stored outdoors in plastic tanks. The insects were shipped to the laboratory and either stored for bioassays or allowed to infest small cylindrical *Pinus taeda* disks. Male- or female-infested disks were used as behavioral stimuli or as volatile sources for chemical studies. Attraction was tested in a Y-tube olfactometer with various dual combinations of odorant stimuli, with individual males or females as focal insects. Both males and females are cleared attracted to non-attacked pine volatiles. When contrasting attraction to male-infested pine disks against non-attacked disks, both males and females showed significant preference for male-attacked disks. When comparting volatiles from attacked and non-attacked disks collected at the entrance of the insect gallery, a single differential compound was consistently observed in the volatiles from male galleries, absent in female or artificial galleries. This compound is an unusual oxidation product of pine monoterpenes and differs from reported *Dryocoetini* male aggregation pheromones. Structure confirmation and synthesis are underway for conducting field tests in the 2022-23 summer.

Keywords: Coleoptera: Curculionidae: Scolytinae, Forest pests, Pheromones, *Pinus* sp.

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S6-P72

Metabolic profile shifts in *Brassica nigra* leaves after belowground nematode infection affect aphid performance aboveground

Jessil Ann Pajar[1,2], Pius Otto[1], April Lyn Leonar[1,3], Nicole M. van Dam[1,4]

[1]*Molecular Interaction Ecology, German Centre for Integrative Biodiversity Research (iDiv), Jena-Halle-Leipzig, Germany.* [2]*International Max Planck Research School, MPI-Chemical Ecology, Jena, Germany.* [3]*Department of Biology, Nematology Research Unit, Ghent University, Ghent, Belgium.* [4]*Institute of Biodiversity, Friedrich-Schiller-University, Jena, Germany*

Previous studies showed below-aboveground interactions in naturally-occurring herbivores of the black mustard plant, *Brassica nigra*. This plant species is simultaneously attacked by several genera of plant-parasitic nematodes belowground and the aphid *Brevicoryne brassicae* aboveground. Experimental studies showed that aphid population growth was positively influenced by prior root-knot nematode (*Meloidogyne* spp.) infection, yet was reduced on plants infested by root-lesion nematodes (*Pratylenchus penetrans*, *Pp*) or cyst nematodes (*Heterodera schachtii*)[1,2]. While this was attributed to nematode-induced phytohormonal signalling interfering with aphid-induced defence responses, many possible mechanisms are still yet unexplored. In addition, these experiments were on single nematode infections, whereas in nature plants interact with many nematode species simultaneously. Here we addressed the question how single and dual nematode infections belowground may affect aphid performance aboveground. Using targeted- and non-targeted metabolomics, we show that shifts in metabolic profiles of *B. nigra* leaves after nematode infection are linked to aphid infestation success. This was measured by changes in aphid fecundity and population growth. Glucosinolates are the main defence compound in Brassicaceae. Pp-infection caused systemic increase of 4-hydroxyglucobrassicin –a glucosinolate previously reported to have aphid-deterring properties[3]. Dual nematode infection resulted in increase of rosmarinic acid in the leaves –a compound known for its insecticidal property[4]. Moreover, dual nematode infection also caused significant decrease of aromatic (phenylalanine, tyrosine) and branched-chain (valine, isoleucine) amino acids. Overall, our study shows plant-mediated interaction between spatially separated herbivores. Analysis of plant metabolic profiles in response to herbivory shows ecological influence of herbivores beyond direct damage to host plants.

Keywords: Metabolomics, Simultaneous herbivory, Plant defense

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S8-P158

Laboratory glass chamber to conduct dynamic headspace analysis of insect volatiles

Sajidha Mohammed[1], Thomas Hesselberg[2], Rafeeq KUMA[1], Shameer K.S[3]

[1]*Department of Zoology, MES Mampad College (Autonomous), University of Calicut, Malappuram, India.*
[2]*Department for Continuing Education & Department of Zoology, University of Oxford, United Kingdom.*
[3]*Insect Ecology and Ethology Lab, University of Calicut, Calicut University, India*

Headspace analysis has always been a crucial part of volatile studies. The complication of the same increases within the case of insects, along with their decreasing size and active nature. An insect body secretes a cocktail of volatile organic compounds (VOCs), the study of which can provide valuable insight into their chemical communication and its impact in various contexts. *Luprops tristis* (Mupli beetle), is one nuisance pest that aggregates in millions at indoor spaces and also releases certain phenolic secretions when disturbed that cause skin and eye irritations [1]. This study aimed to facilitate profiling the whole body volatilome of *Luprops tristis* using an economically feasible sampling chamber to collect the headspace volatiles from the entire insect body, particularly when in aggregation in laboratory conditions. A glass chamber with an approximate volume of 1000 L was designed to collect the volatiles of beetles. A glass disc with holes of 2mm will rest upon the constriction ¼ way of the chamber which was used to prevent the insects from blocking the inlets and outlets. The holes on the disc will allow the volatiles released to move towards the headspace region. Whole-body VOCs were collected from sets of 500 and 1000 beetles at different time intervals (12hr, 24hr, 48hr and 72hr periods) and analyzed by solvent-assisted desorption followed by gas chromatography-mass spectrometry (GC-MS). The glass chamber was successful in the studies of headspace semiochemical analysis, where a range of compounds was detected and identified a few of which also included carboxylic acids, aliphatic and aromatic hydrocarbons. This method facilitates standardized and quantitative analytical profiling of the insect's body volatilome.

Keywords: Dynamic headspace sampling, Insect volatiles, Glass chamber

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S6-P55

JA-dependent pith lignification in *Nicotiana attenuata* performs a defensive function against the stem-boring herbivore

Sungjun Choung[1], Moonyoung Kang[1], Gisuk Lee[1], Youngsung Joo[2], Sang-Gyu Kim[1]

[1]Korea Advanced Institute of Science and Technology, Daejeon, Korea, Republic of. [2]Chungbuk National University, Cheongju, Republic of Korea

The interaction between *Nicotiana attenuata* and *Trichobaris mucorea* is a rising model system for studying the plant defense against stem herbivory. When the larva of *T. mucorea* feed on the pith of *N. attenuata*, the pith is ectopically lignified. However, the function and molecular mechanism of pith lignification are not known. To elucidate the function of induced lignification, we inoculated eggs of *T. mucorea* to lignin-deficient lines (ir*NaCAD*) and compared larval performances with WT. Interestingly, silencing the lignin biosynthetic gene significantly enhanced larval growth. Moreover, in JA-biosynthetic mutant lines (*Naaoc*), larval performance was greater than in WT, and no lignification was induced upon the *T. mucorea* attack. These results showed that pith lignification performs a defensive function in a JA-dependent manner at insect-damaged *N. attenuata* stem. To further investigate the molecular mechanism of JA-dependently induced pith lignification, we performed a comparative transcriptomic analysis between damaged WT and *Naaoc* stem. Among the induced genes in damaged WT pith, we discovered five MYC transcription factors including previously reported two MYC genes. Single mutants of each transcription factor were generated using the virus-induced gene-editing system based on the CRISPR-Cas system. The roles of these MYC transcription factors in pith lignification will be analyzed. This study will uncover the molecular mechanism of tissue lignification against the stem-boring insect herbivore.

Keywords: Nicotiana attenuata, Stem-borer, Jasmonic acid (JA), MYC, Lignification

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S2-P57

The behavior of *Locusta migratoria* to conspecific odors

Anjana Unni, Markus Knaden, Bill Hansson

Max Planck Institute for Chemical Ecology, Jena, Germany

The swarming locust bands have caused huge plagues across the globe since biblical times. Locusts have been and remain a major pest and a threat to food security. These pests exhibit a dramatic phase change from gregarious to solitary phase. The cause of this phase change is a complicated interplay of cues from the environment and conspecifics, and still not well understood. Ethology is pivotal in understanding this polyphenism and developing an efficient pest management technique. The behavior in Locusta migratoria, the migratory locusts is a particularly overlooked area. In this study, we look at the behavior of both solitary and gregarious migratory locusts towards the headspace odors of conspecifics. We asked if the gregarious phase is innately attracted to the odors of conspecifics and whether solitary animals are repelled by the odors of gregarious. We also investigate, whether gregarious males or females emit odors to attract the other sex, and, if so, whether these odors are also attractive to potential mating partners of the solitary phase. The study aims to fill a decisive gap in understanding the aggregation and mating behaviors of the migratory locusts.

Keywords: Locusts, Odors, Behavior, Aggregation, Mating

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S4-P54

Identification of a sex pheromone of hibiscus-leaf caterpillar moth

Junheon Kim, Sujin Lee

National Institute of Forest Science, Seoul, Republic of Korea

Anomis privata (Lepidoptera: Noctuidae) (Syn. *Rusicada private* (Lepidoptera: Erebidae) is one of the primary defoliator insects on *Hibiscus* spp. in Korea. The moths of this species are difficult to detect without pheromone traps due to being active at night. To develop a suitable tool for monitoring and mate disrupt method for control this pest in *Hibiscus syriacus*, we investigated the attractiveness of sex pheromone to *A. privata* adults. We performed gas chromatography-mass spectrometry (GC-MS) analysis of female abdominal tip extracts and identified 7-methylheptadecane (7-MeC17) as a major component of sex pheromone candidate. 7-MeC17 was chemically synthesized and were tested for field attractancy. Male *A. privata* were captured in 7-MeC17 baited traps from April to October in Seoul and Suwon, Korea in 2021, and from April in Suwon, Korea in 2022. In 2021 field tests, traps baited with 7-MeC17 attracted significantly more males than those with hexane as control in both sites. Adult A. private occurred through May and August. Based on these results, it was revealed that 7-MeC17 is one of pheromone components of *A. privata*. It suggests that 7-MeC17 could be useful in a monitoring strategy, which can determine an accurate assessment of the threshold for insecticide applications of target insects.

Keywords: Monitoring, Mate disrupt, Control

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S2-P18

Sublethal effects of insecticides on partner and host finding of *Lariophagus distinguendus*

Nils Schöfer, Joachim Ruther

Institute Of Zoology, University of Regensburg, Regensburg, Germany

As modern agriculture works to produce crops more effectively, new insecticides are being developed to reduce economic damage by herbivorous insects. As these substances are not selective, beneficial non-target species can also be affected. We studied *Lariophagus distinguendus*, a parasitoid of various stored product infesting beetle species, and the sublethal effects of four insecticides (ACEtamiprid, DIMethoate, FLUpyradifurone and SULfoxaflor) on mate finding, courtship and host finding. Sublethal doses of ACE (0.21ng), DIM (≥0.21ng), FLU (≥0.105ng) and SUL (≥0.021ng) decreased the recognition of females by males. Copulation rates were measured separately depending on whether the male, female or both partners were applied with the insecticides. When only males were applied, copulation rates were decreased by ACE (0.105ng), FLU (0.42ng) and SUL (0.021ng & 0.105ng) while DIM (≥0.21ng), FLU (≥0.105ng) and SUL (0.021ng, 0.063ng) showed effects when exclusively females were treated. When both partners were treated all substances decreased copulation rates (ACE: 0.21ng, DIM: 0.105ng, FLU: ≥0.105ng, SUL: 0.021ng, & 0.105ng) with higher doses of FLU (≥0.21ng) reducing the copulation rate to 35%. When tested on their ability to locate hosts, females no longer showed a preference for host odours when treated with any of the four substances. ACE affected the females from the lowest tested dose (≥0.105ng), while only the highest doses of DIM (0.63ng) and SUL (0.105ng) had an effect. When treated with FLU the lowest dose (0.105ng) affected the females, while higher doses did not. Parasitic wasps may come into contact with insecticides at sublethal dosages via nectar, honey dew or guttation water as well as by direct contact with treated crops. The effects at sub-nanogram levels demonstrated here for *L. distinguendus* suggest that sublethal insecticide doses may reduce the reproductive success of parasitic wasps, thus compromising their function as a biological control agents.

Keywords: Sublethal effects, Insecticides, *Lariophagus*, Olfaction, Courtship

S4-P97

Odorant receptors' structure – function relationships are decoded by *in silico / in vitro* joint approaches. Mammal receptors as a test case

Tammy Shim[1,2], Jody Pacalon[3], WonCheol Kim[1], Xiaojing Cong[3], Jérémie Topin[3], Jérôme Golebiowski[1,3], Cheil Moon[1,2]

[1]Department of Brain Science, DGIST, Daegu, Korea, Republic of. [2]Convergence Research Advanced Centre for Olfaction, DGIST, Daegu, Korea, Republic of. [3]Université Cote d'Azur, CNRS, Institut de Chimie de Nice UMR7272, Nice, France

Living organisms use odorant receptors (ORs) to detect airborne chemicals. Although mammal and insect ORs are structurally very different, they play similar roles in chemical detection. In insects, understanding the mechanistic events associated with odorant binding will be extremely helpful to design chemicals that could be of interest to pest control for example. Although the structures of insect ORs were recently better known, the parts of the receptors required for ligand binding are still unknown. This study has focused on a mammalian OR considered as a model system, but a similar study on insect ORs is currently ongoing. We have benchmarked a joint approach combining molecular modeling and in vitro functional assays to understand how a mammalian odorant receptor detects odorant. Molecular dynamics simulation of the modeled human OR1A1 with its representative ligand, (-)-Carvone demonstrated the role of the third extracellular loop in the ligand-binding process. Functional cellular assay confirmed that this third extracellular loop differentially impacts ligands' binding. These results highlighted the role of this extracellular loop as a vestibule site during the ligand-binding process [3,4]. Using this model system, we showed how in vitro - in silico joint research is a powerful tool to decode how ORs detect their ligands.

Keywords: Odorant_Receptors, molecular modeling, functional assays

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S7-P33

Measurable physiological and biochemical changes in Norway spruce freshly attacked by *Ips typographus*, as a potential base of alternative early detection methodology

Barbora Stříbrská[1], Jaromír Hradecký[2], Jaroslav Čepl[2], Ivana Tomášková[2], Roman Modlinger[1], Rastislav Jakuš[1, 3], Anna Jirošová[1]

[1]*Faculty of Forestry and Wood Sciences, Czech University of Life Sciences Prague, Prague, Czech Republic.*
[2]*Faculty of Forestry and Wood Sciences, Czech University of Life Sciences Prague, Prague, Czech Republic.*
[3]*2 Institute of Forest Ecology, Slovak Academy of Sciences, Zvolen, Slovakia*

In recent years, spruce stands in middle Europe experienced intensive bark beetle (*Ips typographus*) calamities. The bark beetle calamity management is based on the early detection of freshly infested trees, which must be cut down and decontaminated before the new beetle generation emerges. Besides the classical infestation checking provided by foresters are developed alternative methodology as an aerial scanning. Our study mainly reports the measurable physiological and biochemical changes in freshly infested trees as a new option for alternative early detection. The changes were compared with long-distance attraction and host acceptance of such trees by conspecific beetles. The experiment was conducted in the naturally *I. typographus* attacked Norway spruces in central Czechia. In trees, bark surface temperature, sap flow, tree ring increment, VOC emission, and resin compounds in the phloem were monitored. In non-choice bioassay, beetles' host acceptance of these trees and the long-range attraction using passive traps were checked. These features were compared with soil water potential showing the water availability for trees and meteorological conditions. The infested trees were located in the dryer locality then monitored control trees were. In infested trees, the bark surface temperature did not change significantly compared to the control but sap flow and ring increment decreased. Phloem VOC emitted by freshly infested trees increased significantly, and the decisive compounds for clustering in PCA analysis were mainly compounds such as eucalyptol, α-terpinol, and trans-4-thujanol. In bioassays, within a week of the infestation, more conspecific beetles were attracted to the infested trees and more beetles accepted these trees as hosts. According to bark beetle management, it would be highly desirable to scan for the infestation in larger forest areas than in individual trees, we suggest developing a detection of specific VOC emitted after infestation.

Keywords: Bark beetles attack, *Picea abies*, VOCs profiling, Defense, Host acceptance

S8-P88

Pollution in beeswax

Kevin Bartl, Erhard Strohm, Joachim Ruther

Regensburg University, Regensburg, Germany

The decline in insects, especially bees, which has been observed for several years, is a severe ecological problem. Among other factors, anthropogenic environmental chemicals are considered a possible cause of population declines. Little attention, however, has been paid on the role of the beeswax which adult honeybees are in constant contact with and bee larvae develop in. The pollution of the landscape, but especially beekeeping measures lead to the accumulation of pollutants in the non-polar beeswax. This might affect the development, health, performance and thus the productivity of honeybees. To get an overview of the current status of contaminations of beeswax in Bavaria, we performed a state-wide monitoring. Wax samples from conventional and organic beekeepers were chemically analysed for possible residues of environmental chemicals. There was generally a higher contamination of wax from conventional compared to organic beekeepers. A total of 80 anthropogenic compounds were detected in the samples at levels ³1 µg/kg. These included 29 fungicides, 28 insecticides/acaricides, 7 herbicides, 1 disinfectant, 9 currently or previously used varroacides and 6 multiple source compounds (MSP). Furthermore, we investigated the accumulation of contaminations in clean beeswax at conventionally and organically farmed areas. A total of 37 anthropogenic contaminations were detected in concentrations ³1 µg/kg. These included 13 fungicides, 9 insecticides/acaricides, 4 herbicides, 1 disinfectant, 6 currently or previously used varroacides and 4 MSP. The quantities of anthropogenic contaminants newly introduced into the beeswax within one season were between 0 and 281 µg/kg. Moreover, we investigated the performance of bee colonies and worker bees that developed in heavily and weakly contaminated wax, respectively. Hitherto, we neither detected any significant differences between treatments concerning colony size and the honey yield nor concerning homing ability, learning, flight performance and immune competence.

Keywords: Honeybee, Apis mellifera, Wax contamination, Pesticides

S6-P59

On chemical ecology of plant-parasitic nematodes

Rasa Čepulytė, Vincas Būda

Laboratory of Chemical and Behavioral Ecology, Institute of Ecology, Nature Research Centre, Vilnius, Lithuania

Knowledge on plant-parasitic nematode (PPN) chemical ecology facilitates environmentally friendly pest management integration. This could be achieved by disrupting biointeractions between nematodes and their host plants and/or between nematodes. Over 500 natural chemical compounds, both organic and inorganic, involved in PPN behavior were classified and reviewed following a system accepted by chemoecologists. Kairomonal egg-hatching stimulants, as well as attractants for juveniles, are presented. PPN pheromones: sex, aggregation, egg-hatching, and putative diapause are analyzed and grouped into clusters of primers and releasers. The most widely analyzed and least studied fields of PPN chemical ecology is indicated. Data on biologically active chemicals reveals targets for resistant plant selection, including through application of gene silencing techniques.

Keywords: Aggregation, Attractant, Diapause, Egg-hatching, Review

S6-P68

Search for active role of FACs hydrolysis with CRISPR-Cas9

Tsuyoshi Maruoka[1], Yu Shirai[2], Takaaki Daimon[2], Naoki Mori[1], Naoko Yoshinaga[1]

[1]Lab of Chemical Ecology, Department of Applied Life Sciences, Kyoto University, Kyoto, Japan. [2]Lab of Insect Physiology, Department of Applied Biosciences, Kyoto University, Kyoto, Japan

Volicitin [N-(17-hydroxylinolenoyl)-L-glutamine] and N-linolenoyl-L-glutamine were originally identified in the regurgitant of Spodoptera exigua larvae. These fatty acid amino acid conjugates (FACs) are known as elicitors that induce plants to release volatile compounds which in turn attract natural enemies of the larvae such as parasitic wasps. FACs concentrations are regulated by enzymatic biosynthesis and hydrolysis in the intestine of Lepidoptera larvae (1.3-3 nmol/µl). We proposed that FACs metabolism activates glutamine synthetase and plays an important role in nitrogen metabolism in larvae. In this study, we identified a candidate gene of FACs hydrolase in Spodoptera litura using genomic information of various related Lepidoptera and knocked it out using CRISPR-Cas9 to analyze the importance of FACs hydrolysis on caterpillar performance. LC/MS analysis revealed that larvae of strains with an inactive FACs hydrolase excreted FACs in their feces whereas WT insects excrete only trace amounts of FACs. Furthermore, analysis using element analyzer has shown that they absorbed 30% less nitrogen from the diet compared to WT caterpillars, which resulted in the slower growth rate of the larvae. These results suggested that the hydrolysis of FACs is an important metabolism for insects and that FACs metabolism play an active role in nitrogen assimilation in the larval body.

Keywords: Tritrophic interactions, Fatty acid amino acid conjugates (FACs), FACs hydrolysis, Nitrogen metabolism, CRISPR-Cas9

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S13-P94

Feed associated bacteria effect on behavioural responses of edible cricket cultivars

Dominykas Aleknavičius[1], Eglė Markaitytė[1], Ramunė Stanevičienė[2], Elena Servienė[2]

[1]*Laboratory of Chemical and Behavioural Ecology, Nature Research Centre, Vilnius, Lithuania.* [2]*Laboratory of Genetics, Nature Research Centre, Vilnius, Lithuania*

Crickets *Acheta domesticus* and *Gryllus assimilis* are easily grown and bred insects that can be a sustainable source of protein used in food or feed production. Currently, this is an increasingly popular practice due to high nutritional values, environmentally friendly cultivation, and effective feed conversion ratio during mass rearing. Regarding that, edible crickets must comply food safety requirement. The aim of this study was to estimate the ability of crickets to recognize bacterially contaminated feed. A two-choice test was performed between bacterially contaminated feed and blank feed using a video tracking software EthoVision XT, which tracks and analyses animal behaviour, movement, and activity. Three bacterial species were tested as potential contaminants: *Escherichia coli*, *Pseudomonas aeruginosa* and *Bacillus subtilis*. The cultivars of crickets differ in their ability to identify contaminated feed. House cricket (*A. domesticus*) did not recognize any of the feeds contaminated with any of the bacterial species tested. While Jamaican field cricket (*G. assimilis*) avoided feed associated to *E. coli*, but the other two bacteria did not cause differences in behaviour. This shows that Jamaican field crickets can recognize some bacteria contaminants and avoid spoiled feed. Further, gas chromatographic - electroantennographic detection will be performed to determine biologically active volatile organic compounds of *E. coli* perceived by Jamaican field cricket. This research is funded by the European Social Fund under the No 09.3.3-LMT-K-712-19-0021 "Development of Competences of Scientists, other Researchers and Students through Practical Research Activities" measure.

Keywords: *Escherichia coli*, Feed safety, House cricket, Jamaican field cricket

S9-P58

VOCs of sea buckthorn fruits attractive for fruit fly *Rhagoletis batava*: search for kairomone compounds

Laima Blažytė-Čereškienė, Vincas Būda, Violeta Apšegaitė, Sandra Radžiutė, Jurga Būdienė, Raimondas Mozūraitis

Laboratory of Chemical and Behavioural Ecology, Institute of Ecology, Nature Research Centre, Vilnius, Lithuania

Rhagoletis batava (Diptera: Tephritidae), is the most important pest of *Hippophae rhamnoides* fruits. For detection and monitoring of *R. batava,* traps supplied with nonspecific attractants are used. New and more specific attractants for environment-friendly pest control are needed. Such attractants could be fruit-related semiochemicals that are involved in the host location by flies. Behavioural Y-olfactometer tests revealed that *R. batava* males were attracted to ripe fruit odour, while females preferred unripe and semi-ripe fruits. Thermal desorption and GC-MS analyses revealed substantial quantitative and qualitative changes in VOCs of unripe/ripe fruits. In the unripe fruit emission, 41 volatile compounds were isolated whereas 64 compounds were sampled from the ripe fruits. The total amount of volatiles increased five times during the fruit ripening. GC-EAD analysis of the fruit headspace volatiles revealed at least 26 compounds in unripe and 27 compounds in ripe fruits eliciting antennal *R. batava* responses of both sexes. The fruits of these two ripening stages differed qualitatively in a single compound only: 3-methylbutyl 2-methylpropionate. Esters were the most abundant volatiles composing 84% and 93% of EAG-active compounds in the emissions of unripe and ripe fruits, respectively. Based on the persistent EAG responses 17 compounds were selected as the most promising candidates for kairomone attractants of the sea buckthorn pest *R. batava*.

Keywords: *Hippophae rhamnoides* fruits, volatiles, *Rhagoletis batava,* electroantennography, kairomone.

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S13-P28

Floral color strategy in *Camellia* attracting different pollinators

Shinnosuke Mori[1], Yoichi Hasegawa[2], Yoshinari Moriguchi[3]

[1]*Keio University, Yokohama, Japan.* [2]*Forestry and Forest Products Research Institute, Tsukuba, Japan.* [3]*Niigata University, Niigata, Japan*

Flower color is a major trait to allure foraging pollinators. Red coloration predominates among ornithophilous flowers, which has been associated with 'the bee avoidance hypothesis'. In the sect. *Camellia*, the snow camellia (*C. rusticana*) and the Japanese camellia (*C. japonica*) bear red flowers and recruit different pollinators; the former is entomophilous (bees, flies, beetles), while the latter is ornithophilous (passerine). *C. japonica* is considered to have been speciated from a common ancestor later than *C. rusticana*, accompanying a pollinator shift from insects to birds. Nevertheless, factors explaining the pollinator difference in camellias remain rudimentary. In this study, the color traits of the two camellias were examined to determine their strategy to attract different pollinators. The two-choice assay between the two species unveiled the exclusive visitation of bees towards *C. rusticana*, indicating that bees chose *C. rusticana* by visual and/or olfactory cues before accessing rewards (nectar, pollen). We analyzed flower color characteristics of the two camellias with diffuse reflectance spectra, testing bee avoidance hypothesis. *C. rusticana* petals were more conspicuous to bees and less to birds due to UV-reflection secondary peak; especially, *C. japonica* petals displayed crucially low chromatic contrast against leaf background for bees, suggesting its petal is almost indistinguishable from the background. On the other hand, *C. japonica* flowers appeared conspicuous to birds. Furthermore, another difference was found between the two species; the anther of *C. rusticana* exhibited blue fluorescence, likely serving as attractive cue for bees[1], while *C. japonica* did not. The fluorescent compounds were identified as two anthranilates. The two camellias offered different color strategies to be conspicuous against their respective pollinators. *C. japonica* appeared to have evolved to avoid bees through the alterations in these color traits.

Keywords: Pollinator shift, Bee avoidance hypothesis, Flower color

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S2-P32

Pheromone synthesis of Coleopteran pests, *Prionus californicus* and *Oryctes rhinoceros*, and their application in field

Yuki Miyake, Erina Ohno, Naoki Ishibashi, Tatsuya Hojo, Takeshi Kinsho, Takeru Watanabe

Specialty Chemicals Research Center, Shin-Etsu Chemical Co., Ltd., Niigata, Japan

The California prionus (CP), *Prionus californicus* (Motschulsky) (Coleoptera: Cerambycidae), is the most destructive pest of hops in the USA. The Coconut rhinoceros beetle (CRB), *Oryctes rhinoceros* (Linnaeus) (Coleoptera: Scarabaeidae), is one of the most important pests of oil palm in Southeast Asia. Insecticide application against both CP and CRB has been used, however, it is sometimes ineffective because pesticide does not reach the inside of the stem or soil where the pests exist. To control these serious pests, pheromone-based mating disruption (MD) and attract and kill (A & K) have been strongly desired. The female produced sex pheromone of CP is (3*R*, 5*S*)-3,5-dimethyldodecanoic acid and the male produced aggregation pheromone of CRB is (4*S*)-ethyl 4-methyloctanoate. We have established an industrially scalable production method of both CP and CRB pheromones. 3,5-dimethyldodecanoic acid (CP pheromone) and ethyl 4-methyloctanoate (CRB pheromone) were both synthesized from corresponding malonic acid derivatives via dealkoxycarbonylation reaction, resulting in a short and efficient synthesis of these pheromones. Using tube-type dispensers, field MD trials for CP have been conducted in USA and A & K trials for CRB in Southeast Asia. MD efficacy was demonstrated through suppression of the male trap capture and reduction of larvae populations. This MD trial became a rare example of successful field MD control of coleopteran pests.

Keywords: Prionus californicus, Oryctes rhinoceros, Mating disruption, Attract and kill

S2-P171

Method to access the mating status of males collected in pheromone traps. A study with *Tuta absoluta* (Lepidoptera: Gelechiidae) in wind tunnel and field

Eraldo Lima, Suelen Speridião, Giovana Huayhua, Natália Ribas, Manuel X-Balam

Laboratory of Semiochemicals and Insect Behavior/Department of Entomology/Federal University of Viçosa, Viçosa, Brazil

Mass trapping has great potential to control *Tuta absoluta*. This technique uses traps with synthetic attractants to capture as many males as possible and keep the population below the economic injury level. Results are not satisfactory, mainly because of the competition of virgin females with traps. The success depends directedly on the unmated males trapped. We aimed to test the hypothesis that the male quality affects the competition between feral females and artificial lures with pheromones. The work was done in a wind tunnel and field. We developed a method to distinguish the mating status of males by using the curve produced by decreasing spermatophore size in successive matings. The size of the spermatophore was affected by the number of matings of the males on consecutive days, and the weight of the males. In a wind tunnel, old males reach the gland extract of females faster than young males, while young males reach the source of synthetic attractants faster than the old males. The method was based on the mean spermatophore size of the first and second copula (0.1082 ± 0.0158 mm2). The male collected in the field was offered to a virgin female for mating and the collected spermatophore sized. Males that produced a spermatophore below the s.d. from the mean were considered mated males, while males with a spermatophore above the s. d. from the mean were considered virgin males. In the field, traps containing virgin females attracted proportionately more mated males than virgins. The number of virgin males collected in traps with the main component was significantly higher than mated males. Is viable to determine the mating status of males in the field and depending on the type of attractant, the proportion of unmated and mated males differs significantly.

Keywords: Male Quality, Pheromone Trapping, Mating Status, Mass Trapping

S6-P121

Bt-protein expression impairs early *Spodoptera frugiperda* oviposition induced volatiles and egg parasitoid attraction in maize plants

Natália Ribas[1], Eraldo Lima[1], Wellington Firmino[1], Carla Arce[2]

[1]*Universidade Federal de Viçosa, Viçosa, Brazil.* [2]*University of Neuchâtel, Neuchâtel, Switzerland*

Plants often induce volatile organic compounds in response to herbivory. However, feeding is not necessarily the first contact between plants and herbivores, as females mainly lay their eggs on the host plants, which can induce volatiles. The use of transgenic crops expressing insecticidal proteins from *Bacillus thuringiensis* (Bt) is the most successful strategy for controlling the fall armyworm (FAW) *Spodoptera frugiperda*, the primary pest of maize in South American countries. FAW has shown resistance to most Bt proteins; therefore, their management seeks more sustainable tools as biological control with parasitoids. Therefore, in this study, we evaluated the effect of resistant and susceptible FAW oviposition on Bt and isogenic maize varieties, the defense oviposition-induced volatiles, and how the volatiles induced affects the searching behavior of the egg parasitoid *Trichogramma pretiosum*. Our results demonstrated that *T. pretiosum* females prefer volatiles emitted by isogenic maize varieties (var. 7280 and var. 709) with eggs of *S. frugiperda* either susceptible and resistant rather than the odors from control plants without eggs. The females did not discriminate between control and plants with FAW eggs on the Bt maize varieties. This pattern could be explained due to the induction of green leaf volatiles (GLVs: (E)-2-hexenal and (Z)-3-hexenal) emitted in higher amounts by the isogenic maize varieties but not by the Bt varieties. Our study unravels the effects of Bt mediated resistance on crucial biological control agents. Thus, these results may help optimize the biological control with *T. pretiosum* in maize fields with Bt and isogenic refuge areas to control FAW promoting an increase in parasitism rates and the viability of biological control programs with *T. pretiosum*.

Keywords: FAW, Natural enemies, Oviposition, Maize, Volatiles

Posters (In the App only)

Brown planthopper honeydew-associated microbes and their role in rice defense; a laboratory and field approach
David Wari, Yuko Hojo, Akio Tani, Tomonori Shinya, Ivan Galis

Dynamics of silicification and the role of silicon in rice defense against chewing and sucking herbivores
Dandy Ahamefula Osibe, Yuko Hojo, Tomonori Shinya, Ivan Galis

E-β-farnesene mediated adaptive evolution between wheat aphid and its parasitoid *Aphidifus gifuensis* based on olfactory plasticity
Xin jiang, Miaomiao Yu, Jun Jiang, Siyu Zhang, Frédéric Francis, Yun Xu, Jia Fan, Julian Chen

Evolution of sex-pheromone receptors in *Drosophila*
Melissa Díaz-Morales, Mohammed A. Khallaf, Rongfeng Cui, Jerrit Weißflog, Markus Knaden, Bill S. Hansson

Functional differentiation of three pheromone binding proteins in *Orthaga achatina* using mixed-type sex pheromones
Qi Yan, Yu-Xiao Si, Jin-Meng Guo, Yu Ma, Shuang-Lin Dong

Influence of semio-chemicals (Methyl eugenol and Zingerone) aromatherapy on the response and mating success of the sterile peach fruit fly *Bactrocera zonata* (Saunders) (Diptera:Tephritidae)
Mahfuza Khan, Md. Azharul Islam, Md. Abdul Bari

Knocking out a simple extracellular leucine-rich repeat protein gene confers host plant resistance to a piercing-sucking herbivore
Peng Kuai, Na Lin, Miaofen Ye, Meng Ye, Shuting Chen, Yonggen Lou

Methyl benzoate is a new natural fumigant with the potential to control post-harvest pest species
Md Munir Mostafiz, Kyeong-Yeoll Lee

Occurrence of *Drosophila suzukii* population monitored by sugar-vinegar liquid trap and the trap with liquid volatile compounds
Jin-Ping Zhang, Rui Tang, Muhammad Yasir Ali, Xin-Yue Tian, Zheng-Yu Luo, Feng Zhang

Odorant receptors mediate beetle's aggregation behavior
Tianliang Ji, Zhi Xu, Youming Hou, Guirong Wang

Regulation of orientation behavioral responses in *Bemisia tabaci* by CCYV-induced dynamic variations of plant terpenoids
Fengming Yan, Zelong Zhang, Jingjing Li

Systemic defense against herbivores in rice: Not really the case
Ivan Galis, Taiga Kuwabara, Yuko Hojo, David Wari, Tomonori Shinya

The "hidden players": Nectar-inhabiting microbes mediate interactions between flowering plants and parasitoids in conservation biological control
Jay Darryl Ermio, Antonino Cusumano, Patrizia Bella, Ezio Peri, Michael Rostás, Salvatore Guarino, Bart Lievens, Stefano Colazza

Brown planthopper honeydew-associated microbes and their role in rice defense; a laboratory and field approach

David Wari, Yuko Hojo, Akio Tani, Tomonori Shinya, Ivan Galis

Institute of Plant Science and Resources, Kurashiki, Japan

Plants evolved the ability to sense insect damage, for example tissue wounds caused by feeding of caterpillars, to timely activate their defense. However, feeding of sucking insects, such as the rice brown planthoppers (*Nilaparvata lugens*; BPH), causes only limited mechanical damage on plants. We hypothesize that plants could be using other signals to detect BPH attacks. Previously, we established a model where rice plants selectively recognize symbiotic microbes in the honeydew of BPHs and use them as important functional signals to activate their innate defense systems. In search for more insect-associated microbes, we have sampled BPHs from different rice cultivars grown in different geographical locations in Japan. Insect-associated microbes from the BPH honeydew were isolated on a growth media, screened and identified using 16S rRNA gene sequencing. The insect associated microbes from the BPH honeydew were further characterized through Next Generating Sequencing (NGS). Representative isolates from 10 orders were then subjected to further screening and characterization of induced rice innate defense systems in the laboratory conditions. Representative isolates from 6 orders that induced rice innate defense were then further characterized in field trials. Results showed that application of isolates from the orders Enterobacteriales, Flavobacteriales, Bacillales, Lysobacteriales, Pseudomonadales and Sphingobacteriales induced high levels of rice secondary metabolites in laboratory and field conditions. In this poster, we discuss about the potentially useful roles of insect-associated microbes in pest management and their interaction with the host plants (rice), the host-insect (BPH) and other pests/herbivores.

Keywords: Honeydew-associated microorganisms, Plant defense, Rice (*Oryza sativa*), Rice brown planthopper (*Nilaparvata lugens*)

Chain length of exocrine gland components ubiquitously decreases in bumble bee queens throughout their life cycle.

Margarita Orlova, Etya Amsalem
[1]Pennsylvania State University, University Park, USA

Queen pheromones regulating worker reproduction have been extensively studied in social insects, but most studies have focused on one exocrine source and searched for the one, all-important compound that would perform the entirety of the queen's functions. However, these pheromones often convey information about the queen's physiological state, and the physiological state can be reflected simultaneously in multiple compounds produced in various exocrine glands. To address this gap, we examined whether changes in bumble bee queen physiology led to a pervasive shift in the chemical composition of multiple exocrine secretions. Bumble bees are especially interesting models to study this question because queens change their chemical output and strategy to control worker reproduction during their life cycle. We examined the chemical composition of cuticular hydrocarbons, the Dufour's, labial and the mandibular glands in bumble bee queens that were either unmated (gynes), had recently founded the colony and produced workers (young queens) or headed a mature colony and produced sexual offspring (old queens). Our findings indicate that the chain length of hydrocarbons and the alcohol moiety of wax esters decreases across exocrine sources as the queens age. Older queens produced a higher proportion of short-chained and lower proportion of long-chained compounds in all glands compared to younger queens. Gynes also produced larger proportions of terpenoid compounds in the labial and the Dufour's glands. These findings suggest that changes in queen physiology pervasively affect exocrine gland composition and are likely to be regulated via shared mechanisms of biosynthesis and transportation of chemical signals.

Keywords: queen pheromone, hydrocarbons, aging, biosynthesis, worker reproduction, esters

Dynamics of silicification and the role of silicon in rice defense against chewing and sucking herbivores

Dandy Ahamefula Osibe[1, 2], Yuko Hojo[1], Tomonori Shinya[1], Ivan Galis[1]

[1]Institute of Plant Science and Resources, Okayama University, Kurashiki, Japan. [2]Department of Plant Science and Biotechnology, University of Nigeria, Nsukka, Nigeria

Rice accumulates and deposits silicon (Si) as amorphous silica in specialized cells in the leaves which helps the plant to overcome various stresses [1]. Here, using scanning electron microscopy and X-ray microanalysis (SEM/EDX) we showed the pattern of distribution of the dumb-bell shaped silica cells in rice leaves treated with or without Si. Our EDX, provided evidence for the localization of these silica cells in the leaf epidermis and their higher accumulation thereof in Si-treated plants compared to untreated. Furthermore, the effect of Si on trichome numbers in rice was also investigated using the acquired SEM images. However, no decisive variation was found in the number of macrohairs (non-glandular trichomes) on Si-treated and untreated plants. We also examined the effects of Si treatments on the growth performance and survival of chewing (*Mythimna loreyi*) and sucking (*Nilaparvata lugens*) rice herbivores. Si-treated plants deterred feeding by *M. loreyi* and as such reduced their weight gain and survival rates. More so, SEM analysis of frass from the chewing rice herbivore fed on Si-treated rice revealed the presence of trichomes and silica cells suggesting a possible reduction in digestion efficiency. However, the effect of Si treatment appeared to be more complex in the case of sucking insects. We then examined the expression of genes involved in Si uptake and distribution after herbivory. Intriguingly, transcripts of genes involved in Si transport were not largely affected by herbivory in the presence of Si. Overall, our current study provides additional insights to the accumulation and defense roles of Si in rice.

Keywords: Insect herbivores, Silica, SEM, Trichomes, Rice defense

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E-β-farnesene mediated adaptive evolution between wheat aphid and its parasitoid *Aphidifus gifuensis* based on olfactory plasticity

Xin jiang[1], Miaomiao Yu[1], Jun Jiang[1], Siyu Zhang[1], Frédéric Francis[2], Yun Xu[3], Jia Fan[1], Julian Chen[1]

[1]*State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, Beijing, China.* [2]*Functional and Evolutionary Entomology, Gembloux Agro-Bio Tech, Gembloux, Belgium.* [3]*Agricultural Environment &Resources Institute of YAAS, Kunming, China*

(E)-β-farnesene is the major component of aphids that elicits defense or escape from nearby conspecific aphids, but whether the natural enemy, such as parasitoid, utilizes that cue is still debated. Odorant-binding proteins (OBPs) are a gene family which integral to the peripheral detection of odoriferous chemicals. Here, we focused on the molecular basis of perceiving EBF within *Aphidifus gifuensis* and also the foraging behavior responses between *A. gifuensis* and *Sitobion miscanthi* to the nanogram level of EBF. Firstly, we discovered a molecular basement that perceives EBF in *A. gifuensis*. Multi-AgifOBPs genes bound to EBF and elevated to induction with EBF, we also revealed that hydrophobic interactions are the primary driving forces between AgifOBP26 and EBF and AgifOBP26 was detected in the main olfactory organs of antennae. For the sources of EBF, we confirmed that a trace level of EBF could be detected from the aphid's body. After spreading a reciprocal amount of EBF throughout the surface of S. miscanthi, the behavior parameters between *S. miscanthi* and *A. gifuensis* were considerably elevated in the short ranges. Finally, we demonstrated that artificially releasing *A. gifuensis* appears to be a viable technique, as *A. gifuensis* could dramatically suppress the total number of surviving aphids while also increasing the relative mummified ratio of alate, which relates to *A. gifuensis*' preference for different phenotypes of *S. miscanthi*. Our results suggest that *A. gifuensis* acquired a more sensitive olfactory mechanism to detect the trace level of EBF that remained on aphid surfaces, resulting in reciprocal adaptation between the two species. Our findings may provide light on the parasite wasp's olfactory sensitivity to host cues, as well as aid improve the use of *A. gifuensis* as a natural biocontrol agent.

Keywords: *Aphidifus gifuensis*, Odorant-binding protein, (E)-β-farnesene, Foraging behavior, Evolutionary adaptation

Evolution of sex-pheromone receptors in *Drosophila*

Melissa Díaz-Morales[1,2], Mohammed A. Khallaf[3], Rongfeng Cui[4], Jerrit Weißflog[1], Markus Knaden[1], Bill S. Hansson[1]

[1]*Max Planck Institute for Chemical Ecology, Jena, Germany.* [2]*University of Costa Rica, San Jose, Costa Rica.* [3]*Max Delbrück Center for Molecular Medicine, Berlin, Germany.* [4]*Sun Yat-sen University, Guangdong, China*

Insects use olfaction for a diversity of inter- and intra-specific communications. In that way, sex pheromone perception plays an important role in speciation, allowing the attraction of conspecific individuals and repelling individuals from different species [1]. Recent studies on the evolution of sex pheromones within the genus *Drosophila* identify a high diversity of sex-specific compounds that are mainly detected via olfaction [2]. These studies also demonstrate that to keep the efficiency of inter- and intra-specific reproductive communication systems, rapid and independent evolution of the sex pheromones and their associated olfactory channels is necessary. Using the transgenic expression of olfactory receptors with the empty neuron system in D. melanogaster, and the posterior measurement of electrophysiological responses through single sensillum recordings to previously identified sex pheromones, we are investigating how pheromone receptors coevolve to match the high diversity of sexual pheromones. We would also like to identify potential hotspots in the receptors undergoing rapid evolution.

Keywords: Pheromone receptors, Single sensillum recording, Drosophlid flies, Empty neuron system

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Functional differentiation of three pheromone binding proteins in *Orthaga achatina* using mixed-type sex pheromones

Qi Yan, Yu-Xiao Si, Jin-Meng Guo, Yu Ma, Shuang-Lin Dong

Nanjing Agricultural University, Nanjing, China

Pheromone-binding proteins (PBPs) play important roles in perception of insect sex pheromones, functioning to recognize and transport pheromone components onto the olfactory receptors of the odorant sensing neurons. *Orthaga achatina*, a serious pest of camphor trees, uses a mixture of three Type I (Z11-16:OAc, Z11-16:OH and Z11-16:Ald) and one Type II (Z3,Z6,Z9,Z12,Z15-23:H) sex pheromone components in its sex communication, in which Z11-16:OAc is the major component and others are minor components. In this study, we for the first time demonstrated that the three PBPs differentiated in recognition among pheromone components in a moth using mixed-type sex pheromones. First, tissue expression study showed that all three PBPs of *O. achatina* were expressed only in antennae and highly male-biased, suggesting their involvement in perception of the sex pheromones. Second, the three PBPs were expressed in *Escherichia coli* and the binding affinities of PBPs to four sex pheromone components and some pheromone analogs were determined by the fluorescence competition binding assays. The results showed that OachPBP1 bound all four sex pheromone components with high binding affinity, while OachPBP2 had high or moderate binding affinity only to three Type I components, and OachPBP3 had high binding affinity only to three minor pheromone components. Furthermore, key amino acid residues that bind to sex pheromone components were identified in three PBPs by 3-D structure modeling and ligand molecular docking, predicting the interactions between PBPs and pheromone components. The study provides a fundamental insight into the olfactory mechanism in moths that use mixed-type sex pheromones.

Keywords: *Orthaga achatina*, Pheromone binding protein, Mixed-type sex pheromone, Ligand binding assay

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Influence of semio-chemicals (Methyl eugenol and Zingerone) aromatherapy on the response and mating success of the sterile peach fruit fly *Bactrocera zonata* (Saunders) (Diptera:Tephritidae)

Mahfuza Khan[1], Md. Azharul Islam[2], Md. Abdul Bari[3]

[1]*Institute of Food and Radiation Biology (IFRB), Atomic Energy Research Establishment (AERE), Bangladesh Atomic Energy Commission (BAEC), Ganak bari, Savar, Dhaka -1349, Bangladesh.* [2]*Department of Zoology, Faculty of Life and Earth Science, Jagannath University, Dhaka, Bangladesh.* [3]*Insect Biotechnology Division (IBD), Institute of Food and Radiation Biology (IFRB), Atomic Energy Research Establishment (AERE), Bangladesh Atomic Energy Commission (BAEC), Ganak bari, Savar, Dhaka-1349, Bangladesh*

The peach fruit fly, *Bactrocera zonata* (Saunders) is a severe invasive pest of horticultural crops. Males of many *Bactrocera* species under the family Tephritidae are strongly attracted to lures, methyl eugenol (ME) and Zingerone (Zn). Control of fruit flies using Sterile Insect Technique (SIT) relies heavily on mating competitiveness and the survival of the released sterilized male flies. The mating performance of male flies is influenced by the pre-release nutritional supplements/semio-chemicals, and the age of individuals. Accordingly, we investigated to determine the response of male *B. zonata* to two different semio-chemicals viz: ME and Zn, separately using aromatherapy. The mating success in terms of percentage mating pairs, mating latency and copula duration was studied using ME and Zn aroma treated sterile male: control male and control female *B. zonata*, followed by modified method of Haq et al. [1] under laboratory condition. The experimental results showed significantly higher response of male *B. zonata* to ME than Zn aromatherapy at the age of day 7. The percentage of mating pairs (71.42%) of sterile male *B. zonata* on ME aromatherapy was significantly higher (P>0.05) than the percentage (28.57%) pairs formation of untreated *B. zonata*. Similarly, a significantly higher percentage of mating pairs (53.842%) of sterile Zn aroma treated *B. zonata* was recorded than untreated (28.57%) *B. zonata.* The mating latency (mins) and mating duration (mins) did not differ significantly between treated and untreated flies. The experimental results suggested that immature virgin male *B. zonata* strongly responded to ME aromatherapy compared to Zn aromatherapy under controlled laboratory conditions. Both ME and Zn aromatherapy enhanced the mating success of sterile male *B. zonata*, but the effect was much stronger to ME aromatherapy. The present findings indicated semio-chemicals aromatherapy as prerelease supplement have significant influence on the implementation of SIT against *B. zonata.*

Keywords: -

Knocking out a simple extracellular leucine-rich repeat protein gene confers host plant resistance to a piercing-sucking herbivore

Peng Kuai, Na Lin, Miaofen Ye, Meng Ye, Shuting Chen, Yonggen Lou

Institute of Insect Sciences, Zhejiang Univeisity, Hangzhou, China

Simple extracellular leucine-rich repeat (eLRR) proteins, which structurally resembles the ectodomain but without the transmembrane and kinase domains of eLRR receptors-like kinases (LRR-RLKs), have been revealed that also participate in plant defense responses. However, the molecular mechanisms of these simple eLRRs in regulating plant defense responses to herbivores are still poorly understood. Here, we discovered a novel plasma membrane-localized eLRR protein in rice (*Oryza sativa*), OsLRR6, whose transcript levels could be strongly up-regulated by infestation of gravid females of the piercing-sucking herbivore, brown planthopper (BPH, *Nilapavata luguns*), as well as treatments with methyl jasmonate (MeJA) or abscisic acid (ABA). Knocking out *OsLRR6* increased the activity of constitutive mitogen-activated kinase protein 6 (OsMPK6) and BPH-induced OsMPK3 and changed the mRNA level of several defense-related WRKY transcript factors. Additionally, the knockout of *OsLRR6* enhanced levels of BPH-induced jasmonic acid (JA), JA-isoleucine (JA-Ile), ethylene and ABA accumulations, but decreased the level of hydrogen peroxide (H_2O_2). Moreover, contents of defensive compounds, like some flavonoids and rice volatiles, were increased in *OsLRR6*-knockout rice lines compared with those in wild-type (WT) plants. Bioassays in the lab showed that the knockout of *OsLRR6* significantly decreased the hatching rate of BPH eggs, the feeding and oviposition preference of gravid BPH females but prolonged the development duration of BPH eggs. Field experiments showed that the population density of BPH and the white-backed planthopper (*Sogatella furcifera*) in *OsLRR6*-knockout lines was significantly lower than that in WT plants. Also, the higher attractiveness to the common egg parasitoid, *Anagrus nilaparvatae*, were displayed *OsLRR6*-knockout rice lines compared with WT plants in both the greenhouse and field experiments. Collectively, OsLRR6 acts as a negative modulator of rice defense responses to BPH and that BPH might exploit this modulator for its own benefit.

Keywords: Rice, Defense responses, *Nilaparvata lugens*, Defense-related signaling pathways, *Anagrus nilaparvatae*

Methyl benzoate is a new natural fumigant with the potential to control post-harvest pest species

Md Munir Mostafiz, Kyeong-Yeoll Lee

Kyungpook National University, Daegu, Republic of Korea

Methyl benzoate (MBe), a volatile organic compound that occurs naturally as a metabolite in many plants, it is also used as an insect semiochemical. The Indian meal moth, *Plodia interpunctella* (Hübner) (Lepidoptera: Pyralidae), is an insect pest that commonly affects stored and postharvest agricultural products. For the control of insect pests and mites, MBe is lethal as a fumigant and also causes contact toxicity. However, the fumigation toxicity of MBe has yet to be demonstrated in *P. interpunctella*. Herein, we evaluated MBe as a potential fumigant for controlling adults of *P. interpunctella* in two bioassays. Compared to the monoterpenes examined under laboratory conditions, MBe demonstrated high fumigant toxicity using a 1-L glass bottle at 1 µL/L air within 4 h of exposure. The median lethal concentration (LC_{50}) of MBe was 0.1 µL/L air; the median lethal time (LT_{50}) of MBe at 0.1, 0.3, 0.5, and 1 µL/L air was 3.8, 3.3, 2.8, and 2.0 h, respectively. Compared with commercially available monoterpene compounds used in pest control, MBe showed the highest fumigant toxicity (toxicity order as follows): MBe > citronellal > linalool > 1,8 cineole > limonene. Moreover, in a larger space assay, MBe caused 100% mortality of *P. interpunctella* at 0.01 µL/cm^3 of air after 24 h of exposure. Therefore, MBe can be recommended for use in food security programs as an ecofriendly alternative fumigant. Specifically, it provides another management tool for curtailing the loss of stored food commodities due to *P. interpunctella* infestation.

Keywords: Stored-product insect, Fumigation toxicity, Naturally available compound, Methyl benzoate, Monoterpenes

Occurrence of *Drosophila suzukii* population monitored by sugar-vinegar liquid trap and the trap with liquid volatile compounds

Jin-Ping Zhang[1,2], Rui Tang[3], Muhammad Yasir Ali[1,2], Xin-Yue Tian[1,4], Zheng-Yu Luo[1,5], Feng Zhang[1,2]

[1]MARA-CABI Joint Laboratory for Bio-safety, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China. [2]CABI East & South-East Asia, Beijing, China. [3]Guangdong Key Laboratory of Animal Conservation and Resource Utilization, Guangdong Public Laboratory of Wild Animal Conservation and Utilization, Institute of Zoology, Guangdong Academy of Sciences, Guangzhou, China. [4]College of Plant Protection, Jilin Agricultural University, Changchun, China. [5]Hubei Engineering Technology Center for Forewarning and Management of Agricultural and Forestry Pests, Institute of Entomology, College of Agriculture, Jingzhou, China

The vinegar fly *Drosophila suzukii* (Matsumura) (Diptera Drosophilidae), it was thought native to east and southeast Asia, recently invaded North American, European, South American, African, and Oceanian countries, and causing considerable damage on soft-skinned and stone fruits. Sugar-vinegar liquid (SVL) trap was confirmed as an effective tool to detect and monitor D. suzukii, and provide information on predicated time for management of this pest. The objectives of this study were to investigate the population occurrence and dynamics of *D. suzukii* at different elevations in mountain and cherry orchard. It was also expected to identify the activity compounds of SVL by chemical ecology technology. Three SVL traps were set up at cherry orchard and five elevations of nature mountain in Beijing, respectively. Traps were checked once per week, and the number of D. suzukii trapped were sexed and counted. The traps were replaced with the new solution weekly and continued with monitoring for two years. The results showed the pest population dynamic was consistent at cherry orchard and mountain. Furthermore, the outcome from all tested sites revealed that there was one sharp population peak in summer among all seasons, whereas none flies were caught from December to April. Moreover, elevation gradient did not provide a clear pattern. Ethanol, 1-propanol, isopentyl alcohol, ethyl (S)- (-)-lactate and acetic acid from SVL were identified as electrophysiological active compounds by GC-MS and GC-EAD. These five compounds mixed with water did not show any significant attractiveness compared to SVL in the field.

Keywords: Spotted wing drosophila, Sugar-vinegar liquid, Monitor, Population dynamic, Chemical ecology

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Odorant receptors mediate beetle's aggregation behavior

Tianliang Ji[1,2], Zhi Xu[1], Youming Hou[2], Guirong Wang[1]

[1]*State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection, Chinese Academy of Agricultural Sciences, Beijing, China.* [2]*State Key Laboratory of Ecological Pest Control for Fujian and Taiwan Crops, Key Lab of Biopesticide and Chemical Biology, Ministry of Education & Fujian Province Key Laboratory of Insect Ecology, College of Plant Protection, Fujian Agriculture and Forestry University, Fuzhou, China*

Odorant receptors (ORs) are expressed on the dendritic membrane of olfactory receptor neurons (ORN) and play an important role in odor detecting and signal transduction in the peripheral olfactory system. The red palm weevil (RPW), *Rhynchophorus ferrugineus* (Olivier) (Coleoptera: Curculionidae), is considered as one of the most serious pests for palm trees worldwide. RPW mass gathering regulated by male-produced pheromones. Behavior experiments indicated RPW was significantly attracted to aggregation pheromone from 0.1 to 10 µg (low dosage), while was obviously repelled from 100 to 1000 µg (high dosage). Therefore, we speculate may exist different pheromone recognized pathways that mediate aggregation behavior of RPW. Single sensillum recordings (SSR) results showed type A sensilla trichoid housing two neurons, and neuron-B was responsive to the two pheromone components, 4-methyl-5-nonol and 4-methyl-5-nonone even at dose of 10 ng, while type B sensilla trichoid, and only tuned to 4-methyl-5-nonanol from 10 µg. Using RNAi and Xenopus oocyte expression system, we demonstrate RferOr1 was sensitive to low dosage of 4-methyl-5-nonol and 4-methyl-5-nonone to mediate aggregation behavior of RPW. While the repellent effect of aggregation pheromone to RPW disappeared at high concentration when RferOr33 was knock-down. Our results reveal RferOr1 and RferOr33 may play a completely role in mediating aggregation behavior of PRW.

Keywords: *Rhynchophorus ferrugineus*, Aggregation pheromone, Antennal sensilla, Odorant receptor, RNAi

Regulation of orientation behavioral responses in *Bemisia tabaci* by CCYV-induced dynamic variations of plant terpenoids

Fengming Yan, Zelong Zhang, Jingjing Li

Henan Agricultural University, Zhengzhou, China

The cucurbit chlorotic yellows virus (CCYV) has caused serious damage to melon crops in many countries in recent years. This plant virus is exclusively transmitted by the whitefly *Bemisia tabaci* (Gennadius) in a semi-persistent mode. Previous studies have shown that both persistent and non-persistent viruses can affect the orientation and performance of insect vectors, through changing host phenotype or interacting with insect vectors directly to facilitate the spread of viruses. However, how CCYV affects host-plant selection by *B. tabaci* has not been reported. In this study, we investigated the visual and olfactory preferences of *B. tabaci* between healthy and CCYV-infected host plants *Cucumis sativus* (Cucurbitaceae). Volatile profiles of healthy and CCYV-infected *C. sativus* plants were analyzed using gas chromatography-mass spectrometry (GC-MS). In the choice assay, whiteflies preferred to settle on CCYV-infected *C. sativus* seedlings. However, the concentrations of total volatiles and terpenes in *C. sativus* plants decreased after CCYV infection. Interestingly, in the Y-tube assay and vision preference test, whitefly *B. tabaci* adults showed significant visual preference to CCYV-infected host but showed olfactory preference to healthy plants. These results indicated that CCYV infection in plants differently affected the visual and olfactory-mediated orientation behaviors of vector whiteflies and implied that visual cues could play a more important role than olfactory cues in whiteflies in locating CCYV-infected host plants.

Keywords: Cucurbit chlorotic yellows virus, *Bemisia tabaci*, Orientation, Terpenes.

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Systemic defense against herbivores in rice: Not really the case

Ivan Galis, Taiga Kuwabara, Yuko Hojo, David Wari, Tomonori Shinya

Institute of Plant Science and Resources, Okayama University, Kurashiki, Japan

It is commonly accepted idea that plants use systemic signaling to induce their defense responses in distal parts, apart from directly damaged areas by herbivores. Such responses seem very logical for a stationary plant that is facing a highly mobile attacker. However, our previously reported results focusing on large scale metabolomics changes in the local and systemic leaves of rice after attack by an adapted strain of fall armyworm, *Spodoptera frugiperda*, suggested that rice plants may not be as strongly relying on the activation of their systemic defenses. In the current experiment, we further investigated a specific set of defense metabolites, and hormonal responses in the hydroponically-grown rice that was exposed to an attack by herbivores from two distinct feeding guilds: chewing larvae (*Mythimna loreyi*) and sucking insects (*Nilaparvata lugens*). Five rice tissue parts comprised of younger (+1), local infested (0), and older (-1) leaves, leaf sheaths and roots were collected over a three-day time period and subjected to chemical analysis by LC-MS/MS. A clear accumulation of jasmonic acid was detected in the locally attacked leaves by both *M. loreyi* and *N. lugens*, and in stems by *M. loreyi*. However, bioactive JA-Ile only accumulated in the locally infested leaves. Salicylic acid levels remained unchanged in all tissues while abscisic acid was elevated only in local tissues. i.e. those directly damaged by chewing insects. Analysis of defense secondary metabolites showed a strictly local inducible character of defensive compounds, isopentylamine and phenolamides in herbivore-damaged leaves. Furthermore, high constitutive levels of phenolamides were found in the roots of hydroponic rice. In conclusion, which contradicts our general expectations and systemic defense in plants, rice plants did not show any strong signs of induced defense responses in their younger and/or other systemic parts.

Keywords: Herbivory, Hormones, Metabolites, Rice (*Oryza sativa*), Systemic defense

The "hidden players": Nectar-inhabiting microbes mediate interactions between flowering plants and parasitoids in conservation biological control

Jay Darryl Ermio[1], Antonino Cusumano[1], Patrizia Bella[1], Ezio Peri[1], Michael Rostás[2], Salvatore Guarino[1], Bart Lievens[3], Stefano Colazza[1]

[1]*Department of Agricultural, Food and Forest Science, Università degli Studi di Palermo, Palermo, Italy.*
[2]*Department of Agricultural Entomology, Georg-August-Universitaet Goettingen, Goettingen, Germany.*
[3]*Laboratory for Process Microbial Ecology and Bioinspirational Management, Department of Microbial and Molecular Systems, KU Leuven, Leuven, Belgium*

Floral nectar emits sweet alluring scents that serve as cues for insects to locate flowers. There are different factors why nectars create a distinct trail of signals which affects foraging behavior of insects. Studies have shown that floral nectar is inhabited by a multitude of microbes that influence volatile emission and suspected as the "hidden players" in the highly understudied plant-parasitoid communications. Here, we isolated nectar microbes from buckwheat (*Fagopyrum esculentum*) to test the hypothesis that these microbes affect the foraging behavior of the stink bug egg parasitoid *Trissolcus basalis* via changes in nectar traits. Culturable microbes were used to ferment synthetic nectars, and their impact on parasitoid olfaction was studied. Parasitoid life history parameters, such as longevity and survival were also investigated as affected by the microbe-fermented synthetic nectars to evaluate if these microbes affect parasitoid performance. Among the microbes associated with buckwheat nectar, we found 14 bacterial strains (and 6 fungal strains). The nectar yeasts *Metschnikowia gruessii* and *M. reukaufii*, though not found in buckwheat nectar, were also included in the list of the test microbes since these yeasts are known to be specialist of flower nectar. Out of the 14 bacterial strains evaluated, four have elicited significant olfaction response in parasitoids (*Staphylococcus epidermidis*, *Terrabacillus saccharophilus*, *Pantoea* sp. and *Curtobacterium* sp.). The synthetic nectar fermented by *M. gruessii*, on the other hand, recorded significant parasitoid attraction than the *M. reukaufii*-fermented nectar. Chemical analyses showed that changes in the nectar traits and impacts on parasitoid performance varied among the nectar microbes used. We discuss our results in an applied perspective as nectar-rich flowering plants such as buckwheat are widely integrated in agricultural landscapes to promote conservation biological control of pests.

Keywords: *Trissolcus basalis*, Nectar, Conservation Biological control, Plant-parasitoid interaction

Participants

Full Name	Institute/Company	Email Address	Country
Adams, Bashiru	International Centre of Insect Physiology and Ecology (icipe), Kenya	badams@icipe.org	Kenya
Afify, Ali	Johns Hopkins University School of Medicine	aafify2@jh.edu	USA
Agnihotri, Aniruddha	Murdoch University	AR.Agnihotri@murdoch.edu.au	Australia
Aleknavičius, Dominykas	Nature Research Centre	dominykas.aleknavicius@gamtc.lt	Lithuania
Ali, Jamin	Keele University	j.alirana@yahoo.com	United Kingdom
Alinc, Tugcan	University of Palermo	tugcan.alinc@unipa.it	Italy
Amsalem, Etya	Penn State University	eua6@psu.edu	USA
Arce, Carla	University of Neuchatel	carla.marques@unine.ch	Switzerland
Barrett, Paul	Massey University	d.p.barrett@massey.ac.nz	New Zealand
Bartl, Kevin	University of Regensburg	kevin_bartl@yahoo.de	Germany
Benrey, Betty	University of Neuchatel, Switzerland	betty.benrey@unine.ch	Switzerland
Biwer, Peter	Institute for Organic Chemistry, Technische Universität Braunschweig	p.biwer@tu-bs.de	Germany
Blazyte-Cereskiene, Laima	Nature Research Centre	laima.blazyte@gamtc.lt	Lithuania
Boff, Samuel	Ulm University	samboff@gmail.com	Germany
Bohman, Björn	the Swedish University of Agricultural Sciences	bjorn.bohman@slu.se	Sweden
Borges, Renee M.	Indian Institute of Science	renee@iisc.ac.in	India
Buda, Vincas	Nature Research Centre	vincas.buda@gamtc.lt	Lithuania
Buellesbach, Jan	Institute for Evolution & Biodiversity, WWU Münster	buellesb@uni-muenster.de	Germany
Bumbulytė, Gabrielė	Nature Research Centre	gabriele.bumbulyte@gamtc.lt	Lithuania
Bustos, Carlos	University of Neuchatel	bustossc@gmail.com	Switzerland
Butterwick, Joel	Yale University	joel.butterwick@yale.edu	USA
Cao, Li	CEFE,CNRS	li.cao@cefe.cnrs.fr	France
Cepulyte, Rasa	Nature Research Centre	rasacepulyte@gmail.com	Lithuania
Cha, Dong	USDA-ARS	dong.cha@usda.gov	USA
Chan, Jeremy	University of Washington	jeremy_runescape@hotmail.com	USA
Cheah, Boon Huat	Department of Agronomy, National Taiwan University	boonhuatcheah@ntu.edu.tw	Taiwan

Chen, Jian	USDA	jian.chen@usda.gov	USA
Chen, Li	China	chenli1@hbu.edu.cn	China
Chen, Ruixu	Jiangsu Vocational College of Agriculture and Forestry	ruixuchen2017@icloud.com	China
Chen, Shuting	Zhejiang University	shtchen@zju.edu.cn	China
Chen, Xiaofeng	Southwest University	18883375009@163.com	China
Chiba, Yuki	Graduate School of Agricultural Science, Tohoku University	yuki.chiba.r6@dc.tohoku.ac.jp	Japan
Choung, Sungjun	Korea Advanced Institute of Science and Technology	sungjun123@kaist.ac.kr	Republic of Korea
Chuang, Wen-Po	National Taiwan University	wenpo@ntu.edu.tw	Taiwan
Clancy, Mary	University of Neuchatel, FARCE Laboratory	clancymv@tcd.ie	Switzerland
Clavijo McCormick, Andrea	Ministry of Primary Industries New Zealand/Massey University	a.c.mccormick@massey.ac.nz	New Zealand
Colazza, Stefano	University of Palermo	stefano.colazza@unipa.it	Italy
Costa, Arnaud	CABI (Centre for Agriculture and BioSciences International)	a.costa@cabi.org	Vietnam
Crowley-Gall, Amber	University of California Davis	acrowleygall@ucdavis.edu	USA
Cui, Mo	Pherobio Technology Co., Ltd	cui@pherobio.com	China
Cusumano, Antonino	University of Palermo	antonino.cusumano@unipa.it	Italy
Daenen, Lucien	Guest	lucien.daenen@idiv.de	
De Faveri, Stefano	Department of Agriculture and Fisheries	Stefano.defaveri@daf.qld.gov.au	Australia
Diaz-Morales, Melissa	Max Planck Institute for chemical Ecology	mdiaz-morales@ice.mpg.de	Germany
Ding, baojian	Lund University	baojian.ding@biol.lu.se	Sweden
Divakaran, Keerthi	Institute of Plant Sciences, University of Bern	keerthi.divakaran@ips.unibe.ch	Switzerland
Dumbacher, John	California Academy of Sciences, Department of Ornithology and Mammalogy	jdumbacher@calacademy.org	USA
Duplais, Christophe	Cornell University - Cornell AgriTech	c.duplais@cornell.edu	USA
Effah, Evans	Massey University	effahevans5@gmail.com	New Zealand
Ermio, Jay Darryl	Università degli Studi di Palermo, Italy	ermiojaydarryl@gmail.com	Italy
Fan, Jia	State Key Laboratory for Biology of Plant Diseases and Insect Pests, IPPCAAS	jfan@ippcaas.cn	China
Fan, Yongliang	Northwest A&F University	yfan@nwafu.edu.cn	China
Fandino, Richard	Cornell University	raf272@cornell.edu	USA
Fang, Jiaxing	Chinese academy of forestry	fjxinsect@163.com	China

Fang, Jiaxing	Ecology and Nature Conservation Institute,Chinese Academy of Forestry	jxfang@caf.ac.cn	China
Faraone, Nicoletta	Acadia University	nicoletta.faraone@acadiau.ca	Canada
Fennine, Chaymae	Free university of Bozen-Bolzano	chaymae.fennine@natec.unibz.it	Italy
Galis, Ivan	Okayama University, Institute of Plant Science and Resources	igalis@okayama-u.ac.jp	Japan
Geng, Yishu	Nanjing Forestry University	angel520-007@163.com	China
Genta-Jouve, Grégory	CNRS	Gregory.genta-jouve@cnrs.fr	French Guiana
González, Andrés	Facultad de Química, Universidad de la República	agonzal@fq.edu.uy	Uruguay
Gonzalez, Mabel	Stanford University	mabelg@stanford.edu	USA
Gu, Tianzi	Anhui Agricultural University	tianzigu@ahau.edu.cn	China
Guo, Hao	Institute of Zoology, Chinese Academy of Sciences	guohaoinsect@126.com	China
Guo, Mengbo	Institute of Plant Protection, Chinese Academy of Agricultural Sciences	guo_mb@163.com	China
Hayes, R. Andrew	University of the Sunshine Coast	rhayes@usc.edu.au	Australia
He, Peng	Guizhou Univeristy	phe1@gzu.edu.cn	China
Heckel, David	Max Planck Institute for Chemical Ecology	heckel@ice.mpg.de	Germany
Hee, Alvin Kah-Wei	Universiti Putra Malaysia	alvinhee@upm.edu.my	Malaysia
Hewa Ranaweera, Pradeepa	Universiti Putra Malaysia	ranaweerapra@gmail.com	Malaysia
Huang, Yongping	Shanghai Institute of Plant Physiology and Ecology, Chinese Academy of Sciences	yphuang@sibs.ac.cn	China
Il'ichev, Alex	Agriculture Victoria Research-retired	ilichev@bigpond.com	Australia
Inoue, Takato	Kyoto university	chem-riku.it@outlook.jp	Japan
Islam, Tarikul	Western Sydney University - Hawkesbury Institute for the Environment	tarikul.islam@westernsydney.edu.au	Australia
Ji, Tianliang	State Key Laboratory for Biology of Plant Diseases and Insect Pests, Institute of Plant Protection,	15506908618@163.com	China
Jiang, Dun	Northeast Forestry University	821880041@qq.com	China
Jiang, Hong bo	Southwest University	jhb8342@swu.edu.cn	China
Jiang, Xin	Institute of Plant Protection, CAAS	jiangxinn61@gmail.com	China
Jirosova, Anna	Extemit, CULS Prague, FFWS	jirosovaa@fld.czu.cz	Czech Republic
Johny, Jibin	Czech University of Life Sciences, Prague	jibinmjo@gmail.com	Czech Republic
Kang, Le	Institute of Zoology, Chinese Academy of Sciences	lkang@ioz.ac.cn	China

Kansman, Jessica	Penn State University	kansmanj@psu.edu	USA
Kaplan, Fatma	Pheronym, Inc.	fkaplan@pheronym.com	USA
Katsumata, Ayako	North Carolina State University	akatsum@ncsu.edu	USA
Kazama, Haruna	Agilent	cabbagec_a_t@yahoo.co.jp	Japan
Kempraj, Vivek	University of Hawai'i	vivek.kempraj@gmail.com	USA
Khan, Mahfuza	Bangladesh Atomic Energy Commission	mahfuza79@gmail.com	Bangladesh
Kieu Hanh, Duong	Can Tho University	dkhanh@ctu.edu.vn	Vietnam
Kim, Junheon	National Institute of Forest Science	junheon@gmail.com	Republic of Korea
Kim, Taein	Laboratory for Ecology, Department of Biological Sciences, KAIST	tedori725@kaist.ac.kr	Republic of Korea
Kong, Xiangbo	Ecology and Nature Conservation Institute, Chinese Academy of Forestry	xbkong@sina.com	China
Kuai, Peng	Zhejiang University	kpchen7493@163.com	China
Kumar, Dharmesh	Individual	atdharmesh@gmail.com	India
Kumar, Manish	IISER PUNE	manishkumar.sah@students.iiserpune.ac.in	India
Kurenshchikov, Dmitry	Institute of Water and Ecology Problems of the Far Eastern Branch of the Russian Academy of Sciences	dkurenshchikov@gmail.com	Russian Federation
Lahondere, Chloe	Virginia Tecj	lahonder@vt.edu	USA
Lambaere, Kevin	Guest	kevin.lambaere@outlook.com	
Le, Quoc Hung	Institute for Evolution and Biodiversity (IEB) - University of Münster (WWU)	hle@uni-muenster.de	Germany
Leal, Walter	UC Davis	wsleal@ucdavis.edu	USA
Levi Zada, Anat	Agricultural Research Organization	anatzada@volcani.agri.gov.il	Israel
Levit, Debora Mirtha	Guest	deboralevit65@gmail.com	
Li-Byarlay, Hongmei	Central State University	hli-byarlay@centralstate.edu	USA
Li, Bin	Guest	jimmy.tobbesson@gmail.com	
Li, Shouyin	Nanjing Forestry University	Leo15077855180@njfu.edu.cn	China
Li, Yaning	Northeast Forestry University	964838394@qq.com	China
Liang, Dangsheng	Apex Bait Technologies, Inc.	dliang@apexbait.com	USA
Lima, Eraldo	Universidade Federal de Viçosa	Eraldo.lima@ufv.br	Brazil
Lima, Nerilda	Guest	nerilda@gmail.com	
Lin, Na	Zhejiang University	lin_na@zju.edu.cn	China
Lin, Po-An	National Taiwan University	poanlin1@hotmail.com	Taiwan

Liu, Fu	Ecology and Nature Conservation Institute,Chinese Academy of Forestry	liufu2006@163.com	China
Liu, Xiaolan	Henan Agricultural University	xiaolanberry@126.com	China
Liu, Yang	Institute of Plant Protection, Chinese Academy of Agricultural Sciences	liuyang02@caas.cn	China
Loeb, Greg	Cornell University	gme1@cornell.edu	USA
Löfstedt, Christer	Lunds universitet	christer.lofstedt@biol.lu.se	Sweden
Lorenzo, Marcelo Gustavo	FIOCRUZ-MINAS	marcelo.lorenzo@fiocruz.br	Brazil
Lou, Yonggen	Zhejiang University	yglou@zju.edu.cn	China
Lu, Yujie	Jiangsu University of Science and Technology	luyjlyj71@just.edu.cn	China
Luan, Jun-Bo	Shenyang Agricultural University	jbluan@syau.edu.cn	China
Ma, Baiwei	Institute of Plant Protection, Chinese Academy of Agricultural Sciences	mabaiweilmr@163.com	China
Machado, Ricardo	University of Neuchatel	ricardo.machado@unine.ch	Switzerland
Machtinger, Erika	Pennsylvania State University	etm10@psu.edu	USA
Mamin, Marine	University of Neuchâtel, FARCE laboratory	marine.mamin@unine.ch	Switzerland
Maruoka, Tsuyoshi	Lab of Chemical Ecology, Department of Applied Life Sciences, Kyoto University	maruoka.tsuyoshi.82n@st.kyoto-u.ac.jp	Japan
Mateus, Eduardo	NOVA School of Science and Technology / NOVA University Lisbon	epm@fct.unl.pt	Portugal
Matsuyama, Shigeru	University of Tsukuba, Graduate School of Life and Environmental Sciences	matsuyama.shigeru.fu@u.tsukuba.ac.jp	Japan
Mauck, Kerry	UC Riverside	kerry.mauck@ucr.edu	USA
McClure, Melanie	CNRS	melanie.mcclure@cnrs.fr	French Guiana
McNeil, Jeremy	University of Western Ontario	jmcneil2@uwo.ca	Canada
Menuz, Karen	University of Connecticut	karen.menuz@uconn.edu	USA
Meredith, LorMona	Pacific Rim Concepts	lormona@gmail.com	USA
Miao, Xuexia	CAS Center for Excellence in Molecular Plant Sciences, Chinese Academy of Sciences.	xxm@cemps.ac.cn	China
Mishra, Aditi	National Centre for Biological Sciences	aditi.niser@gmail.com	India
Miyake, Yuki	Shin-Etsu Chemical Co., Ltd.	miyake_y@shinetsu.jp	Japan
Mohammed, Sajidha	MES Mampad College (Autonomous), University of Calicut	sajidha.mohammed@gmail.com	India

Mori, Naoki	Kyoto University	mori.naoki.8a@kyoto-u.ac.jp	Japan
Mori, Shinnosuke	Keio University	mori.shinnosuke.72x@kyoto-u.jp	Japan
Moriguchi Jeckel, Adriana	Universität Bern	adriana.jeckel@ips.unibe.ch	Switzerland
Mostafiz, Md Munir	Kyungpook National University	munirmostafiz12@gmail.com	Republic of Korea
Najar-Rodriguez, Adriana	Plant and Food Research	adriana.najar-rodriguez@plantandfood.co.nz	New Zealand
Niogret, Jerome	Mars Wrigley	jerome.niogret@effem.com	Australia
Nishida, Ritsuo	Kyoto University	nishida.ritsuo.o33@kyoto-u.jp	Japan
Noge, Koji	Japan	noge@akita-pu.ac.jp	Japan
O'Connell, Lauren	Stanford University	loconnel@stanford.edu	USA
Ohata, Yuto	Kyoto University	oohata.yuuto.87e@st.kyoto-u.ac.jp	Japan
Omura, Hisashi	Hiroshima University	homura@hiroshima-u.ac.jp	Japan
Ono, Hajime	Kyoto University	ono.hajime.5a@kyoto-u.ac.jp	Japan
Osibe, Dandy	Institute of Plant Science and Resources, Okayama University	pnz1088u@s.okayama-u.ac.jp	Japan
P D, Kamala Jayanthi	Indian Institute of Horticultural Research	jaiinsect@gmail.com	India
Pajar, Jessil Ann	German Centre for Integrative Biodiversity Research	jessil_ann.pajar@idiv.de	Germany
Pal, Elisa	University of Pretoria, FABI	elisa.pal@fabi.up.ac.za	South Africa
Palma Onetto, Valeria	Universidad Católica Santísima Concepción	vpalma@ucsc.cl	Chile
Parepally, Saravanan Kumar	Indian Institute of Horticultural Research	sravanchikki@gmail.com	India
Park, Kye Chung	New Zealand Institute for Plant and Food Research	kpark@plantandfood.co.nz	New Zealand
Park, Soo Jean	Macquarie University	soojean.park@mq.edu.au	Australia
Pask, Greg	Middlebury College	gpask@middlebury.edu	USA
Peach, Daniel	The University of Georgia	peach@zoology.ubc.ca	USA
Peñaflor, Maria Fernanda	LAVRAS FEDERAL UNIVERSITY (UFLA)	fernanda.penaflor@ufla.br	Brazil
Perera, Nisansala	Charles Sturt University	nperera@csu.edu.au	Australia
Peri, Ezio	University of Palermo	ezio.peri@unipa.it	Italy
Perkins, James	ANU	james.perkins@anu.edu.au	Australia
Petrén, Hampus	Philipps-University Marburg	hampus.petren@biologie.uni-marburg.de	Germany
Pickett, John	Cardiff University	pickettj4@cardiff.ac.uk	United Kingdom
Potter, Chris	Johns Hopkins University School of Medicine	cpotter@jhmi.edu	USA
Proffit, Magali	CNRS-CEFE	magali.proffit@cefe.cnrs.fr	France

Ramakrishnan, Rajarajan	Faculty of Forestry and Wood sciences, Czech university of Life sciences, Prague, Czech Republic.	ramakrishnan@fld.czu.cz	Czech Republic
Ramiaranjatovo, Gaëlle	CIRAD - UMR PVBMT	ramiaranjatovo@gmail.com	France
Ramos, Sergio	University of Zurich	sergio.ramoscastro@uzh.ch	Switzerland
Rering, Caitlin	USDA-ARS	caitlin.rering@usda.gov	USA
Ribas, Natália	Universidade Federal de Viçosa	natalia.sribas@gmail.com	Brazil
Robert, Christelle Aurélie Maude	University Bern	christelle.robert@ips.unibe.ch	Switzerland
Rossini, Carmen	Facultad de Química	crossinister@gmail.com	Uruguay
S, Aswathi	Indian Institute of Science Education and Research (IISER), Kolkata	as20rs137@iiserkol.ac.in	India
Sakamoto, Shunta	Kyoto University	sakamoto.shunta.44m@st.kyoto-u.ac.jp	Japan
Sarkar, Sumitro	KW Conferences	sumitro@kwconferences.com	India
Saul-Gershenz, Leslie	University of California, Davis	lsaulgershenz@ucdavis.edu	USA
Schöfer, Nils Hauke	University of Regensburg	n.h.schoefer@gmail.com	Germany
Schulz, Stefan	TU Braunschweig	stefan.schulz@tu-bs.de	Germany
Schuman, Meredith Christine	University of Zurich	meredithchristine.schuman@uzh.ch	Switzerland
Shen, Jianmei	Zhongkai University of Agriculture and Engineering	shenjianmei@zhku.edu.cn	China
Shewale, Mayuri	Czech University of Life Sciences	shewale@fld.czu.cz	Czech Republic
Shim, Tammy	DGIST	tammyshim22@gmail.com	Republic of Korea
Siderhurst, Matthew	Eastern Mennonite University	matthew.siderhurst@emu.edu	USA
Silva, Rodrigo	Bedoukian Research	rdasilva@bedoukian.com	USA
Song, Junyong	Korea Advanced Institute of Science and Technology	sjyplay333@kaist.ac.kr	Republic of Korea
Sorrells, Trevor	Rockefeller University	trevorsorrells@gmail.com	USA
Spence, Sarah	Pennsylvania State University	sks6958@psu.edu	USA
Stříbrská, Barbora	Czech university of Life sciences in Prague, Faculty of Forestry and Wood Sciences	stribrska@fld.czu.cz	Czech Republic
Sun, Longlong	Henan Agricultural University	sunlonglong0313@163.com	China
Sun, Weizhao	Institute for Evolution and Biodiversity	wsun1@uni-muenster.de	Germany
Sun, Xiaoling	Tea Research Institute Chinese Academy of Agricultural Sciences	xlsun@tricaas.com	China
Syed, Zainulabeuddin	University of Kentucky - Lexington	zsy224@uky.edu	USA
Takahashi, Teruki	Sumitomo Chemical, Co., Ltd.	takahashit34@sc.sumitomo-chem.co.jp	Japan

Thiyagarajan, Nalini	Department of Entomology, Faculty of Agriculture, Annamalai University	nalinisenthilvelan@gmail.com	India
Torto, Baldwyn	International Centre of Insect Physiology and Ecology (ICIPE)	btorto@icipe.org	Kenya
Touhara, Kazushige	The University of Tokyo	ktouhara@g.ecc.u-tokyo.ac.jp	Japan
Túler, Amanda	ESALQ/USP NCSU	amandatuler@usp.br	USA
Turlings, Ted	University of Neuchâtel	ted.turlings@unine.ch	Switzerland
Umezaki, Yujiro	UC Davis	yumezaki@ucdavis.edu	USA
Unni, Anjana	Max Planck Institute for Chemical Ecology	aunni@ice.mpg.de	Germany
Usmani, Mohd Tariq	Russell IPM India Pvt Ltd	synthesis@russellipm.com	India
Valbon, Wilson	Duke University	wilson.valbon@duke.edu	USA
Valterová, Irena	Institute of Organic Chemistry and Biochemistry, Czech Academy of Sciences	irena@uochb.cas.cz	Czech Republic
van Dam, Nicole	German Center for integrative Biodiversity Research (iDiv)	nicole.vandam@idiv.de	Germany
Van Vang, Le	Can Tho University, Can Tho city, Vietnam	lvvang@ctu.edu.vn	Vietnam
Vander Meer, Robert	USDA/ARS	bob.vandermeer@usda.gov	USA
Vanderplanck, Maryse	CNRS, CEFE	maryse.vanderplanck@cefe.cnrs.fr	France
Venkatesan, Radhika	Indian Institute of Science Education and Research Kolkata	rv@iiserkol.ac.in	India
Venkateswaran, Vignesh	Maxplank institute for chemical ecology	vignesh.venk.90@gmail.com	Germany
Venugopal, Vibina	ICAR-CPCRI	vibinavenugopal@gmail.com	India
Verdecia-Mogena, Arletys	Institute of Plant Sciences (University of Bern)	arletys5.verdecia@gmail.com	Switzerland
Wang, Bing	Institute of Plant Protection, Chinese Academy of Agricultural Sciences	wangbing02@caas.cn	China
Wang, Chen-Zhu	Institute of Zoology, Chinese Academy of Sciences	czwang@ioz.ac.cn	China
Wang, Guangyu	Hebei University	17863800784@163.com	China
Wang, Guirong	Institute of Plant Protection, Chinese Academy of Agricultural Sciences	wangguirong@caas.cn	China
Wari, David	Institute of Plant Science and Resources, Okayama University	pmi89t6q@okayama-u.ac.jp	Japan
Warr, Coral	La Trobe University	c.warr@latrobe.edu.au	Australia
Watanabe, Takeru	Shin-Etsu Chemical Co., Ltd.	takeru.watanabe@shinetsu.jp	Japan
Wee, Suk-Ling	Faculty of Science and Technology, Universiti Kebangsaan Malaysia	slwee@ukm.edu.my	Malaysia
Wickham, Jacob D	Institute of Ecology and Evolution, Russian Academy of Sciences	jacobwickham@gmail.com	Russian Federation

Wong, Darren	Australian National University	darren.wong@anu.edu.au	Australia
Xiao, Wei	Southwest University, China	xiaowei@swu.edu.cn	China
Xiao, Wenhan	Zhejiang University	2223171532@qq.com	China
Xiao, Yong	Institute of Plant Protection, Guangdong Academy of Agricultural Sciences	xiaoyong@gdaas.cn	China
Xu, Guangxin	Hebei University	jainxuguang@163.com	China
Xu, Li	Southwest University	xll718287@swu.edu.cn	China
Xu, Shuqing	University of Mainz	shuqing.xu@uni-mainz.de	Germany
Xu, Wei	Murdoch University	W.Xu@Murdoch.edu.au	Australia
Yan, Fengming	Henan Agricultrual University	fmyan@henau.edu.cn	China
Yan, QI	Nanjing Agricultural University	yanqi@njau.edu.cn	China
Yang, Bin	IPPCAAS	byang@ippcaas.cn	China
Yao, Chengcheng	Institute of Plant Protection, Chinese Academy of Agricultural Sciences	yaochengcheng128@163.com	China
Yeo, Shelley	Guest	shelleyeo@hotmail.com	
Yesmin, Farzana	Bangladesh Atomic Energy Commission	farzana.baec@gmail.com	Bangladesh
Yoshinaga, Naoko	Kyoto University	yoshinaga.naoko.5v@kyoto-u.ac.jp	Japan
Zhang, Aijun	USDA-ARS	aijun.zhang@usda.gov	USA
Zhang, Jin	Tea Research Institute Chinese Academy of Agricultural Sciences	zhangjin1369@tricaas.com	China
Zhang, Jin	Nanjing Agricultural University	jinzhang001@njau.edu.cn	China
Zhang, Jinping	CABI	j.zhang@cabi.org	China
Zhang, Longwa	Anhui Agricultural University	zhanglw@ahau.edu.cn	China
Zhang, Qing He	Sterling International Inc	qing-he@rescue.com	USA
Zhang, Sufang	Nature Conservation Institute, Chinese Academy of Forestry	zhangsf@caf.ac.cn	China
Zhang, Xiaxuan	Agricultural Genomics Institute at Shenzhen, Chinese Academy of Agricultural Sciences	zhxiaxuan@163.com	China
Zhang, Xun	-	zhx3355@126.com	China
Zhao, Lilin	Institute of Zoology, Chinese Academy of Science	zhaoll@ioz.ac.cn	China
Zhou, Fengwen	Nanjing Agricultural University	2020102085@stu.njau.edu.cn	China
Zhu, Dan	Guest	dan.zhu@gmail.com	
Zhu, Junwei Jerry	USDA-ARS, AMRU	jerry.zhu@usda.gov	USA
Zuest, Tobias	University of Zürich	tobias.zuest@uzh.ch	Switzerland

叶, 馨儿	梅州市华师昆虫发育生物学与应用技术重点实验室广梅园研发中心	914134805@qq.com	China
叶, 馨儿	梅州市华师昆虫发育生物学与应用技术重点实验室广梅园研发中心	624060707@qq.com	China
庆, 高	浙江大学	11916008@zju.edu.cn	China
李, 娜	梅州市华师昆虫发育生物学与应用技术重点实验室广梅园研发中心	lina5sxnd@sina.com	China

Thank you for your participation!

We hope you enjoy(ed) the conference and are looking forward to meeting you again next year!

2022

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Abstract Book

Managing sustainability in challenging times

Asia-Pacific Association of Chemical Ecologists