# International Society of Chemical Ecology

## 15th Annual Meeting
## Ithaca, New York

## June 20-24, 1998

## Saturday, June 20

1900-2100 **Welcome reception and registration,** Mary Donlon Hall

## SCIENTIFIC PROGRAM

(All activities in Alumni Auditorium, Kennedy Hall)

## Sunday, June 21

0830-1200 **Registration,** Kennedy Hall
0900-0915 **Official Welcome**
Alan Renwick, Meeting Chairman
Theodore Hullar, Director, Cornell Center for the Environment
Jim Tumlinson, President, ISCE

**Opening Session**
0915-0925 **John Simeone**, Chemical Ecology: in the beginning
0925-0950 **Thomas Eisner**, Chemical Ecology: in retrospect and prospect
0950-1035 **Daniel Janzen**, Biodiversity development of a Costa Rica conservation area as a garden: chemicals and a whole lot more
1035-1100 Coffee break
1100-1145 **Jo Handelsman**, Discovering the chemical repertoire of microbial diversity in soil: a metagenome approach
1145-1230 **Baldomero Olivera**, Cone Snail Venoms: fifty million years of drug development
1230-1330 Lunch break

**Intraspecific Interactions**
1330-1400 **Walter Leal**, Chemical diversity in the sex attraction of scarab beetles
1400-1515 Contributed papers
1515-1545 Coffee break
1545-1715 Contributed papers

## Monday, June 22

**Intraspecific Interactions, continued**
0900-0945 Contributed papers

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| | **Interspecific Interactions I** |
| 0945-1015 | **Jim Tumlinson**, Tritrophic interactions and plant signaling |
| 1015-1045 | Coffee break |
| 1045-1230 | Contributed papers |
| 1230-1330 | Lunch break |
| 1330-1430 | Contributed papers |
| | **Interspecific Interactions II** |
| 1430-1500 | **Ritsuo Nishida**, Chemistry of plant-insect associations: phytochemical niches in butterflies |
| 1500-1530 | Coffee break |
| 1530-1615 | Contributed papers |
| 1630-1730 | Annual Business Meeting |
| | **Special Lecture (Silverstein-Simeone Award)** |
| 1900-2000 | **Ian Baldwin**, The chemical ecology of *Nicotiana attenuata*: an example of adaptive plasticity |
| | **Poster Session** |
| 2000-2200 | Kennedy Hall and Corson-Mudd Atrium (refreshments) |

## Tuesday, June 23

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| | **Function and use of Bioactive Molecules** |
| 0900-0930 | **Jon Clardy**, Natural products and their macromolecular receptors |
| 0930-1030 | **David Cane**, Making sense of nature's scents: how nature makes terpenes |
| 1030-1100 | Coffee break |
| 1100-1130 | **Ashit Ganguly**, Drugs from natural sources |
| 1130-1300 | Contributed papers |
| 1300 | Afternoon free for recreation and informal discussion groups |

## Wednesday, June 24

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| | **Chemistry of bioactive molecules** |
| 0900-0930 | **Koji Nakanishi**, Natural products chemistry: past, present and future |
| | **Interspecific Interactions II, continued** |
| 0930-1030 | Contributed papers |
| 1030-1100 | Coffee break |
| 1100-1215 | Contributed papers |
| 1215-1330 | Lunch break |
| 1330-1500 | Contributed papers |
| 1500-1530 | Coffee break |
| 1530-1630 | Contributed papers |
| | **Human Health Applications** |
| 1630-1700 | **Charles Arntzen**, Creating transgenic plants for pharmaceutical production |

**Banquet**

1930 Statler ballroom

**Meeting Chairman:** Alan Renwick,
Boyce Thompson Institute, Ithaca

**Program Committee:** Alan Renwick (Chairman), Boyce Thompson Institute
Athula Attygalle, Dept. of Chemistry, Cornell University
Thomas Eisner, Section of Neurobiology and Behavior, Cornell University
Jerrold Meinwald, Dept. of Chemistry, Cornell University
Wendell Roelofs, N.Y. State Agr. Exp. Sta., Cornell Univ., Geneva, NY.
Eloy Rodriguez, L.H. Bailey Hortorium, Cornell University

**Local Arrangements:** Donna Coye, Cornell Conference Services

**Formatting of Abstracts:** Holly Beermann,
Elaine VanEtten, Boyce Thompson Institute

**Conference logo and T-shirt design:** Kimberly Lopez, Boyce Thompson Institute

The organizers acknowledge a generous grant from an anonymous donor and financial as well as logistic support from the Boyce Thompson Institute.

# Sunday, June 21, A.M., Opening Session

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| **O-1** | 0915-0925 | **John Simeone**<br>Chemical Ecology: in the beginning |
| **O-2** | 0925-0950 | **Thomas Eisner**<br>Chemical Ecology: in retrospect and prospect |
| **O-3** | 0950-1035 | **Daniel Janzen**<br>Biodiversity development of a Costa Rica conservation area as a garden: chemicals and a whole lot more |
| | 1035-1100 | **Coffee break** |
| **O-4** | 1100-1145 | **Jo Handelsman**<br>Discovering the chemical repertoire of microbial diversity in soil: a metagenome approach |
| **O-5** | 1145-1230 | **Baldomero Olivera**<br>Cone Snail Venoms: fifty million years of drug development |
| | 1230-1330 | **Lunch break** |

**Chemical ecology: in the beginning.** **O-1**

John Simeone

College of Environmental Science and Forestry, SUNY, Syracuse, NY 13210

**Chemical ecology: in retrospect and prospect.** **O-2**

Tom Eisner

Section of Neurobiology & Behavior, Cornell University, Ithaca, NY 14853

**Biodiversity development of a Costa Rica conservation area as a garden : chemicals and a whole lot more**. **O-3**

Daniel Janzen

Department of Biology, University of Pennsylvania, Philadelphia, PA 19104

**Discovering the Chemical Repertoire of Microbial Biodiversity in Soil: A Metagenome Approach.** **O-4**

Jo Handelsman, Michelle R. Rondon, Jon Clardy, and Robert M. Goodman

Department of Plant Pathology, University of Wisconsin, Madison, WI 53706, USA.

Microorganisms' lives are governed by small molecules. In soil, microorganisms communicate with plants, animals, and other microorganisms through signaling compounds that have a vast array of biological activities. These activities include modification of plant development and physiology and initiation of endosymbiotic relationships with soil fauna. Small molecules contribute to defensive tactics of microorganisms living in communities, and are central to the assembly of microbial consortia, influencing the growth of other microorganisms and sensing the density of like organisms. What we know about the chemical virtuosity of microbes in soil is entirely based on those organisms we can grow in defined culture media in the laboratory. But recent evidence shows that less than 0.1% of the microorganisms in soil can be cultured readily. Molecular phylogenetic analysis reveals that the other 99.9% of the community contains rich diversity, with entirely new groups of microorganisms and new members of known groups. Indeed, the soil is perhaps the greatest frontier of unknown biology with thousands of new species and their physiology and chemistry waiting to be understood. Our work focuses on a functional analysis of the noncultured community of microbes using a genomics approach that

we call “metagenomics” to characterize new chemistry in the biologically diverse and unknown microflora of the soil.

**Cone snail venoms; fifty million years of drug development.** **O-5**

Baldomero M. Olivera

Department of Biology, University of Utah, Salt Lake City, UT 84112, USA.

Venomous cone snails have evolved a neuropharmacological strategy to capture prey and defend against predators. Most pharmacologically active components of the venoms are small, highly constrained peptides that have multiple disulfide linkages; these are the direct translation products of genes. Every cone snail species has its own distinct complement of ca.100 peptides; since there are about 500 *Conus* species, these venoms comprise a neuropharmacological resource of 50,000 peptides. The majority are probably generated from only 6-8 peptide superfamilies with extremely rapid diversification as speciation occurs. Each peptide targets a specific molecular form of receptor or ion channel on the cell surface. *Conus* peptides have a remarkable ability to discriminate between closely related members of receptor and ion channel families. Recent structure-activity work indicates that some *Conus* peptides have two distinct interaction faces, a design permitting both high affinity and high discrimination. Thus many *Conus* peptides represent a new type of pharmacologically active agent, for which we coin the term “Janus ligand.”

# Sunday, June 21, P.M., Intraspecific Interactions

Chairpersons: Wendell Roelofs and Charles Linn, Cornell University

**O-6** 1330-1400 **Walter Leal**
Chemical diversity in the sex attraction of scarab beetles

**O-7** 1400-1415 **Jong-yoon Kim**
Pheromone Glands in Some Scarabs

**O-8** 1415-1430 **Jocelyn G. Millar**
Sex attractant pheromones for moths in the genus *Saturnia* (Lepidoptera:Saturniidae).

**O-9** 1430-1445 **Allan Cameron Oehlschlager**
Synthesis & field testing of the chiral isomers of the aggregation pheromone of the banana weevil, *Cosmopolites sordidus*.

**O-10** 1445-1500 **Ring T. CardŽ**
Effects of Light Levels and Plume Structure on the Orientation Maneuvers of Male Gypsy Moths Flying Along Pheromone Plumes of Differing Structure.

**O-11** 1500-1515 **Darryl K. Jewett**
Evidence for chemical communication between males and females of two pupal parasitoids (Ichneumonidae: Hymenoptera).

1515-1545 **Coffee break**

**O-12** 1545-1600 **Christoph Messer**
Phenolic compounds in *Neanura muscorum* (Templeton): Collembola, Neanuridae, and their role as Deterrents and alarming Substances.

**O-13** 1600-1615 **Michael J. Greene**
Pheromonal inhibition of male courtship behavior in the brown tree snake, *Boiga irregularis*.

**O-14** 1615-1630 **Jeremy N. McNeil**
Evidence of a male pheromone in the fall armyworm, *Spodoptera frugiperda,* and its role in mate choice.

**O-15** 1630-1645 **Jeffrey R. Aldrich**
Pheromone-mediated augmentation of the predatory spined soldier bug, *Podisus maculiventris* (Heteroptera: Pentatomidae).

**O-16** 1645-1700 **Shigeru Matsuyama**

Chemical ecology in the Japanese honeybee, *Apis cerana japonica* rad. *(acj)*: relation between free fatty acids in royal jelly and worker mandibular gland components.

**O-17** 1700-1715 **Abraham Hefetz**
Caste specificity of dufour's gland secretion and its regulation in the honey bee *Apis mellifera*.

**Diversity in insect pheromones.** **O-6**

Walter Soares Leal

Laboratory of Chemical Prospecting, National Institute of Sericultural and Entomological Science, 1-2 Ohwashi, Tsukuba 305-8634, Japan.

Since the milestone identification of bombykol more than three decades ago, about 950 insect sex pheromones and sex attractants have been reported. The most thoroughly studied female-produced sex pheromones of Lepidoptera show very little diversity in chemical structures, with monounsaturated straight chain aldehydes, alcohols and acetates being especially common. By contrast, the pheromone chemistry of Coleoptera is remarkably diverse. This will be highlighted by the case of scarab beetles, in which chemical communication is achieved by sex pheromones with a wide variety of structures, ranging from the ubiquitous phenol and anisole to amino acid derivatives and a unique anti-inflammatory and analgesic alkaloid. This diversity in scarab beetle pheromones and a pivotal role of chirality in pheromonal activity have been explored in my laboratory in order to get a better understanding of the molecular basis of insect olfaction.

**Pheromone Glands in Some Scarabs** **O-7**

Jong-yoon Kim and Walter Soares Leal

Laboratory of Chemical Prospecting, National Institute of Sericultural and Entomological Science, 1-2 Ohwashi, Tsukuba 305-8634, Japan.

Careful analyses using GC-MS showed that pheromone glands in genus *Anomala* (Scarabaeidae: Rutelinae) were integumental epithelial cells on a few last abdominal segments. Histological studies suggested that these epithelial cells differentiated from ordinary integumental epithelia, after the latter having secreted the adult cuticle. Subsequent investigations showed that these epithelial pheromone glands were a common feature among ruteline beetles but were not necessarily shared by Melolonthinae, a closely-related subfamily. In a melolonthinae, *Holotrichia parallela*, calling females everted from their abdominal tip a ball-like structure, which contained high titers of the major pheromone component, L-isoleucine methyl ester (LIME). GC-mass spectrometry analyses identified the posterior part of the structure as the center for pheromone production which was confirmed by histological analyses of this part revealing a layer of secretory columnar cells. Paired accessory glands known as pheromone glands in some other melolonthine species were found not involved in the pheromone production in *H. parallela*.

**Sex attractant pheromones for moths in the genus *Saturnia* (Lepidoptera: Saturniidae).** **O-8**

Jocelyn G. Millar[1], J. Steven McElfresh[1], and Dan Rubinoff[2]

[1]Dept. of Entomology, University of California, Riverside CA 92521, USA.
[2]Div. of Environmental Science, Policy, and Management, 201 Wellman Hall, University of California, Berkeley CA 94720, USA.

Relatively few sex attractant pheromones have been identified for moths in the family Saturniidae, in part because few species in this family are injurious to agricultural crops. In conjunction with studies of the sex attractant pheromones of other Saturniid moths in the western US, we had occasion to examine the pheromone chemistry of species in the genus Saturnia. This genus is represented by only three species in the New World, *S. mendocino, S. walterorum, and S. albofasciata*. All three species are univoltine, and are found in the chaparral plant

communities of Oregon and California. The brown and orange, day-flying *S. mendocino* and *S. walterorum* are similar in color, host range, and general biology, and adults of both species fly in spring. In contrast, *S. albofasciata* has a different host range than the other two species, and the sexually dimorphic adults emerge in mid to late fall. We will describe the identification of the sex pheromones of these three species by combined analytical chemistry and electroantennogram techniques, and the synthesis and field testing of the active components.

**Synthesis & field testing of the chiral isomers of the aggregation pheromone of the banana weevil, *Cosmopolites sordidus*.** **O-9**

Seetharaman Jayaraman[1], Allan Cameron Oehlschlager[2], Lilliana Gonzalez[2], Dennis Alpizar[3], and Mario Fallas[3]

[1]Dept. Chem., Simon Fraser Univ., Burnaby, B.C., Canada.
[2]ChemTica Int., S.A., Apdo. 159-2150 San Jose, Costa Rica.
[3]Min. Agric., Guapiles, Costa Rica.

Male *Cosmopolites sordidus* produce four structural isomers (A,B,C,D) of their aggregation pheromone, sordidin. Chiral isomers of mixtures of sordidin A & B and sordidin C & D have been prepared & field tested. Although only one chiral isomer of each sordidin structural isomer is produced by the males both chiral isomers of sordidin structural isomers attract weevils to pitfall traps in the field. Both chiral isomers of sordidin A & B are biologically attractive but do not exhibit synergy when presented as racemic mixtures. Both chiral isomers of sordidin C & D are biologically active & exhibit synergy when presented as racemic mixtures.

**Effects of Light Levels and Plume Structure on the Orientation Maneuvers of Male Gypsy Moths Flying Along Pheromone Plumes of Differing Structure.** **O-10**

Ring T. CardŽ[1], Bart G.J. Knols[2] and L.P.S. Kuenen[3]

[1]Department of Entomology, University of California, Riverside, California 92521 USA.
[2]International Centre for Insect Physiology and Ecology, Nairobi, Kenya.
[3]Department of Entomology, Cornell University, Ithaca, New York 14853, USA.

Gypsy moth (*Lymantria dispar*) males headed more toward upwind and increased their velocities flying along wide turbulent plumes of pheromone, compared to flight along narrow, ribbon-like plumes. The mean rates of counterturning were concentrated within a band of 3.5 to 4 turns $s^{-1}$.

These differences are consistent with a response template in which moment-to-moment contact with pheromone dictates heading and velocity. Flights along both ribbon plumes and turbulent plumes in 450 lux and a near dusk level of 4 lux differed in the amplitudes of the zigzags and in velocities. These differences may be related to strategies to enhance a male's ability to detect wind-induced drift when light levels are reduced.

**Evidence for chemical communication between males and females of two pupal parasitoids (Ichneumonidae: Hymenoptera).** **O-11**

Darryl K. Jewett and James E. Carpenter

USDA, ARS, Insect Biology Lab, P.O. Box 748, 2747 Davis Road, Tifton, GA 31793, USA.

Data supporting chemically-mediated communication between males and females of two pupal parasitoids *Diapetimorpha introita* (Cresson) and *Ichneumon promissorius* (Erichson) were gathered with field trapping, a behavioral bioassay, and electroantennography. Responses of male antennae to conspecific females or their extracts were greater than to males, their extracts or a control. In behavioral bioassays, more males were recovered from traps baited with females than with males or a control. Extracts from females of neither species attracted males in the bioassay. In the field, more *D. introita* males were caught in traps baited with live females than with live males or a control. *D. introita* males were caught in traps baited with live females than with live males or a control. *D. introita* and *I. promissorius* are potentially important to the biological control of armyworms and bollworms in the southeastern United States, and methods of monitoring their relative seasonal abundance are being considered including use of their sex pheromones.

**Phenolic compounds in *Neanura muscorum* (Templeton): Collembola, Neanuridae, and their role as Deterrents and alarming Substances.** **O-12**

Christoph Messer[1], Konrad Dettner[1], Stefan Schulz[2] and Wittko Franke[2]

[1]Lehrstuhl Tierškologie II, UniversitŠt Bayreuth, UniversitŠtsstr. 30, 95440 Bayreuth, Germany.
[2]Lehrstuhl fŸr Organische Chemie, UniversitŠt Hamburg, Martin-Luther-King Platz 6, 20146 Hamburg, Germany.

Among other volatiles, four phenolic compounds were identified in whole body samples of the collembolan *Neanura muscorum* (Templeton): 2,4-dimethoxyaniline, 1,3-dimethoxybenzene, phenol and 2-aminophenol. The major compounds, 1,3-dimethoxybenzene and 2,4-dimethoxyaniline, were also found in the hemolymph and the eggs of this species. The phenolics were quantified for individual animals by gas chromatographic methods. Synthetic substances as well as crushed conspecifics were used to examine the behavior of *N. muscorum* in bioassays. 1,3-dimethoxybenzene apparently serves as an intraspecific alarming substance. Behavioral tests with the predatory mite *Pergamasus norvegicus* (Berlese) showed deterring properties of 2,4-dimethoxyaniline.

**Pheromonal inhibition of male courtship behavior in the brown tree snake, *Boiga irregularis*.** **O-13**

Michael J. Greene and Robert T. Mason.

Department of Zoology, Oregon State University, 3029 Cordley Hall, Corvallis, OR 97331, USA.

The brown tree snake is a nocturnal, rear-fanged colubrid native to Australia, Papua New Guinea and the Solomon Islands that has caused significant economic and ecological damage on the island of Guam since its accidental introduction during W.W.II. This species displays robust courtship and male-male combat behaviors in the laboratory. In 6 of 25 courtship trials, female brown tree snakes released a bolus of liquid from their cloacae in response to male courtship. In each case, male courtship rapidly ceased after the release of the cloacal secretion (CS). To test the hypothesis that female CS inhibits male courtship and is courtship specific, we performed four experiments: 1) the effect of female CS on male courtship, 2) the effect of male CS on male courtship, 3) the effect of female CS on male-male combat and 4) the effect of male CS on male-male combat. The results of these experiments demonstrate that only female CS inhibits male courtship and that this is a courtship specific response. As male snakes cannot force copulation, this may represent a mechanism for females to reject males deemed unsuitable mates.

**Evidence of a male pheromone in the fall armyworm, *Spodoptera frugiperda,* and its role in mate choice.** **O-14**

Jeremy N. McNeil and Jean Philippe Gravel

Department of Biology, Laval University, Ste-Foy, P. Q., Canada, G1K 7P4.

While the two host races of the fall armyworm are sympatric there appears to be a low level of hybridization under field conditions. While the calling windows are temporally separated and there are small differences in pheromone production in the two strains, males of one strain will fly upwind in a wind tunnel to calling females of the opposite strain. We, therefore, undertook a study to determine if the mating success of males varied depending on the host strain of the female. Initial experiments found that mating of within-strain pairs was generally about 80% and while a similar level was seen when corn males were held for one scotophase with rice females, there was less than 30% mating when rice males were held with corn females. This suggested the possibility of asymmetry in mate choice. However, it was clear, following detailed behavioral observations throughout the scotophase, that females of both strains generally vigorously

rejected males of the other stain. The results strongly suggest that male sex pheromones play a role in mate choice in the fall armyworm and the inter-strain mating observed are an artifact of laboratory conditions.

### Pheromone-mediated augmentation of the predatory spined soldier bug, *Podisus maculiventris* (Heteroptera: Pentatomidae). O-15

Jeffrey R. Aldrich

USDA-ARS Insect Chemical Ecology Laboratory, B-007, rm301, Agricultural Research Center-West, Beltsville, Maryland 20705 USA.

As part of a 3-year USDA Pilot Test, we demonstrated that enough wild *P. maculiventris* adults can be captured in 30 pheromone-baited traps during 2 weeks in early spring to produce over 400,000 immature predators. We have also shown that spined soldier bug nymphs will walk toward Soldier Bug Attractors™, indicating that the synthetic pheromone can also be used to disperse nymphs from points of augmentation. In the final year of the pilot test we combined pheromone-mediated harvest of adult predators with pheromone manipulation of their offspring. Cages were made using a mesh size allowing young predators to escape while retaining the adults, and these cages were placed in the center of treatment plots to serve as predator nurseries. Wild adult predators caught in pheromone-baited traps were transferred to nursery cages, and synthetic pheromone dispensers were positioned at the perimeter of plots to attract immature predators from the nursery cages into the surrounding rows of potato plants. Colorado potato beetle infestations were significantly suppressed using this approach.

### Chemical ecology in the Japanese honeybee, *Apis cerana japonica* rad. relation between free fatty acids in royal jelly and worker mandibular gland components. O-16 *(acj)*:

Shigeru Matsuyama[1], Takahisa Suzuki[1], and Hiromi Sasagawa[2]

[1]Institute of Applied Biochem., Univ. of Tsukuba, Tsukuba, Ibaraki, 305 Japan.
[2]PRESTO "Intelligence and Synthesis", JST c/o National Institute of Sericultural and Entomological Science, 1-2 Oowashi, Tsukuba, Ibaraki, 305-8634 Japan.

(E)-10-hydroxy-2-decenoic acid (10-HDA) and 10-hydroxydecanoic acid (10-HDAA) are found as major components in both mandibular glands (MG) of forager and royal jelly (RJ), product of young worker honeybees, in the European honeybee, *Apis mellifera* L. (*Am*). On the other hand, forager of *Acj* produces mainly (R)-(-)-3-hydroxyoctanoic acid (3-HOA) in MG. These facts

prompted us to examine free fatty acids in RJ of *Acj*. Interestingly, RJ from *Acj* contained 10-HDA and 10-HDAA as major components, implying that worker MG components may change along with aging. By comparing the MG components between different ages, we found that *Acj* workers change their MG components according to their ages. Younger bees produce 10-HDA and 10-HDAA in their mandibular glands. Biological significance of 3-HOA for *Acj* foragers and why they change the MG components during their lifetime are unknown.

**Caste specificity of dufour's gland secretion and its regulation in the honey bee *Apis mellifera*.** **O-17**

Abraham Hefetz, Tamar Katzav and Victoria Soroker

Department of Zoology, Tel Aviv University, Ramat Aviv 69978 Israel.

Caste specificity in pheromonal composition seems to be the rule in honey bees. The morphological and physiological differences between the castes are already evident upon emergence, suggesting that their regulation occurs during the preimaginal state. Nevertheless, recent findings indicate that some plasticity in the biosynthetic capability of exocrine glands is retained in the imago implying the existence of additional regulating mechanisms. We have focused our study on Dufour's gland, a modified accessory gland associated with the sting apparatus characteristic to the Hymenoptera. The chemical composition of glandular secretion, revealed by GC/MS analyses, is caste specific. Qualitatively, the exudate of workers is composed of a homologous series of odd *n*-alkanes, whereas the glandular exudates of the queens also possess long chain esters. However this caste specificity is not rigid, since under queenless conditions egg laying workers, but not foragers produce queen like esters. To evaluate whether Dufour's gland biosynthetic potential is modulated by social and physiological factors, we have initiated a study of the *de novo* biosynthesis of esters and hydrocarbons in queens' gland *in vivo* and *in vitro,* using [1-$^{14}$C] sodium acetate as a precursor. In *in vivo* incubations, esters and hydrocarbons comprised the major *de novo* products in ratios comparable to those naturally occurring. Whereas *in vitro* incubated glands synthesized significant levels of esters and fatty acids but no hydrocarbons. Extrinsic and intrinsic factors that may be responsible for these differences are currently investigated.

# Monday, June 22, A.M., Intraspecific Interactions, continued

Chairpersons: Wendell Roelofs and Charles Linn, Cornell University

**O-18** 0900-0915 **Anne-Genevieve Bagn res**
Biological and biochemical processes which regulate cuticular hydrocarbons in insects.

**O-19** 0915-0930 **Joachim Ruther**
The European hornet, *Vespa crabro* L., perceives artificial modifications of cuticular hydrocarbon profiles.

**O-20** 0930-0945 **Allan Cameron Oehlschlager**
Pheromone-based mass trapping of the banana weevil, *Cosmopolites sordidus* & the West Indian sugarcane weevil, *Metamasius hemipterus* in plantain & banana.

# Interspecific Interactions I

Chairperson: Ted Turlings, University of Neuch‰tel

**O-21** 0945-1015 **Jim Tumlinson**
Tritrophic interactions and plant signaling.

1015-1045 **Coffee Break**

**O-22** 1045-1100 **Manuel Aregullin**
The chemical defenses of tortoise beetles (Cassidinae).

**O-23** 1100-1115 **Fredric V. Vencl**
Shield-bearing leaf beetles: chemical, ecological, and phylogenetic aspects.

**O-24** 1115-1130 **Johannes Steidle**
Grain and faeces: host finding and host recognition cues for the granary weevil parasitoid *Lariophagus distinguendus* (fšrst.)

**O-25** 1130-1145 **Gert Petersen**
Odor-mediated influence of the hyperparasitoid *Alloxysta victrix* on the defense behavior of *Sitobion avenae*.

**O-26** 1145-1200 **Fran ois Lorenzetti**
Plant stress, plant resistance, herbivores, and predators: bottom-up effects.

**O-27** 1200-1215 **Rupert Keller**
Plant stress, plant resistance, herbivores, and predators: bottom-up effects.

**O-28** 1215-1230 **Torsten Meiners**
What is the basis of pederin polymorphism in *Paederus riparius* rove beetles?

1230-1330 **Lunch break**

**Biological and biochemical processes which regulate cuticular hydrocarbons in insects.** **O-18**

A.-G. Bagn res[1], S. Ziani[1], G. J. Blomquist[2], M. Kuenzli[2], S. Mpuru[2], C. Schal[3], V. L. Sevala[3], and J.-L. ClŽment[1]

[1]LNB-Communication Chimique-CNRS UPR 9024. 31 Chemin J. Aiguier, 13402 Marseille Cedex 20. France.
[2]Dept of Biochemistry, MS 330, University of Nevada, Reno, NV 89557. USA.
[3]Dept of Entomology, Box 7613, North Carolina State University, Raleigh, NC 27695 USA.

Cuticular hydrocarbons (HCs) serve as signals in highly regulated chemical signatures allowing species and sex recognition in many insect species and nest and caste recognition in social insects. Control of the chemical signatures in different species is finely tuned. Under artificial experimental conditions, control is less strict. We have studied two different types of regulation in two kind of species; the regulation of biosynthesis and transport under hormonal cues in houseflies and internalization and degradation/detoxification in termites. Our findings in houseflies confirm strong hormonal control of HC production. We were able to gain insight into the internalization processes by applying non-labeled and radiolabeled (tritiated) components to the cuticle of termites. We observed quick internalization of the added component as shown by a rapid decrease of the compound on the cuticle followed by an increase inside the insects. In *Reticulitermes* spp. the process depends on whether the compound is of exogenous or endogenous origin and on the nature of the cuticle. In *Zootermospis* we have noted the same type of internalization depending upon the exogenous compound, and have followed internalization into the hemolymph.

**The European hornet, *Vespa crabro* L., perceives artificial modifications of cuticular hydrocarbon profiles.** **O-19**

Joachim Ruther[1] and Stefan Sieben[2]

Free University of Berlin, Institute of Zoology:
[1]Department of Applied Zoology/Animal Ecology, Haderslebener Str.9, 12163 Berlin, Germany.
[2]Department of Bee Research, Kšnigin-Luise-Stra§e 1-3, 14195 Berlin, Germany.

The behavioral response of workers of the European hornet *V.* crabro L. (Hymenoptera, Vespidae) towards differently treated dead conspecifics (dummies) was observed. Seven types of dummies were compared: Untreated nestmates (1) untreated non-nestmates (2) extracted nestmates whose cuticular hydrocarbons had been reapplied (control) (3), extracted nestmates whose cuticular hydrocarbon profiles had been modified before reapplication by adding 10 g of heneicosane (4), tricosane (5), (*Z*)-9-tricosene (6), or a mixture of the three compounds (7). All tested compounds are common components of cuticular hydrocarbons in *V. crabro*. Treatment of the dummies with either heneicosane or the mixture caused a significant increase of agonistic behavior towards the dummies in leaving workers, when compared with the control treatment. Incoming foragers reacted much less aggressive than leaving ones. This is the first behavioral proof that modification of cuticular hydrocarbon profiles can be perceived by a social wasp species and can induce agonistic behavior. The results support the hypothesis that colony specificity of cuticular hydrocarbon profiles are responsible for nestmate recognition in social wasps.

**Pheromone-based mass trapping of the banana weevil, *Cosmopolites sordidus* & the West Indian sugarcane weevil, *Metamasius hemipterus* in plantain & banana.** **O-20**

Allan Cameron Oehlschlager[1,] Dennis Alpizar[2], Mario Fallas[3], Lilliana Gonzalez[1], & Seetharaman Jayaraman[3]

[1] ChemTica Int., S.A., Apdo. 159-2150 San Jose, Costa Rica.
[2] Min. Agric., Guapiles, Costa Rica.
[3] Dept. Chem., Simon Fraser University, Burnaby, B.C., Canada.

*Cosmopolites sordidus* and *Metamasius hemipterus* are economically important world-wide pests of plantain and banana. Mass trapping *C. sordidus* using a pheromone-baited pitfall trap and *M. hemipterus* using a pheromone-sugarcane-baited open gallon trap over 13-17 months at

four traps for each insect per ha gave 75-85% reduction in corm damage and 25-32% increase in yields in both banana and plantain.

**Tritrophic interactions and plant signaling.** **O-21**

Jim Tumlinson

CMAVE, USDA, ARS, 1700 SW 23d Drive, Gainesville, FL 32608

**The chemical defenses of tortoise beetles (Cassidinae).** **O-22**

Manuel Aregullin[1], Karen Olmstead[2], and Eloy Rodriguez[1]

[1]Phytochemistry Laboratory, L.H. Bailey Hortorium, Division of Biological Sciences, Cornell University, Ithaca, NY 14853 USA.
[2]Department of Biology, University of South Dakota, Vermillion, SD 57069 USA.

Sequestration of plant natural products, for purposes of predator deterrence, is a well documented trait among many species of insects. Larvae of the tortoise beetle *Physonota* sp. (Cassidinae), exhibit peculiar behavior of sequestration by retaining and covering exuvia with feces forming shields (parasols) containing active chemicals from plants they specialize on. The beetle uses these shields in overt defensive behavior when threatened by a potential enemy. We have studied populations of *P. arizonae* that feed on *Ambrosia ambrosioides* in the southwest, and *P. helianthi* that feed on *Helianthus grosseserratus* in the midwest, for the chemistry of their shields and the comparative chemistry of the plant species. Toxic sesquiterpene lactones and diterpenes have been isolated from beetle shields and plants and their structures elucidated. The chemical evolution of defensive shields will also be presented.

**Shield-bearing leaf beetles: chemical, ecological, and phylogenetic aspects.** **O-23**

Fredric V. Vencl and Timothy C. Morton[1]

Department of Neurobiology and Behavior, State University of New York at Stony Brook, Stony Brook, NY 11794-5230, USA.
[1]Department of Ecology and Evolution, University of Chicago, Chicago IL, 60637, USA.

The remarkable diversification of phytophagous insects, particularly leaf beetles (*Chrysomelidae* ), is thought to be due primarily to overcoming two major hurdles: host-plant chemical barriers and natural enemies. Mechanisms by which these factors might interact to promote cladogenesis are not well understood but dietary specialization, enforced by generalist predation pressure, may be requisite conditions. Larval members of several leaf beetle subfamilies have convergently evolved an unusual defensive strategy: fecal retention to form 'shields' against predators. Bioassay-guided chemical analyses of host plants and shield feces revealed that larvae expropriate and transform particular host secondary, and primary metabolites, and then eliminate them in feces that become strategically positioned above larvae to form chemically deterrent shield defenses. We compared unrelated species occupying the same trophic niche to determine whether particular chemicals occur in feces as selective responses to predation, or appear as fortuitous by-products of digestion. Phylogenetic analysis indicated that clades colonizing chemically more 'complex' hosts are derived and are twice as diverse as sister taxa that retained the ancestral, chemically 'simpler' hosts. Shield (and host) chemical complexity positively relates to the shield's effective spectrum. The relationships between defensive spectrum, dietary specialization, and enhanced beetle diversification is examined with respect to the 'predator-free space' hypothesis and chemical 'bridges'.

**Grain and faeces: host finding and host recognition cues for the granary weevil parasitoid *Lariophagus distinguendus* (fšrst.)** **O-24**

Johannes Steidle and Joachim Ruther

FU Berlin, Angewandte Zoologie, Haderslebenerstr. 9, 12163 Berlin, Germany.

The parasitoid *Lariophagus distinguendus* parasitizes the larvae of the granary weevil *Sitophilus granarius* that develop in kernels of grain. The parasitoid female has to find and identify grain kernels infested by the host within a bulk of healthy grains. Former experiments revealed that the females of *L. distinguendus* use chemical cues from the damaged kernels and mainly from the faeces of the weevil larvae for host finding and host recognition. To identify the active compounds extracts were tested in an olfactometer (volatiles) and in Petri dishes (contact chemicals). This demonstrated that arrestment during host finding and host recognition behavior (antennae drumming, tapping, drilling) could be stimulated by hexane extracts of the faeces. For fractionation the hexane extract of the faeces was applied on columns of silica gel and eluted with hexane, hexane/methylenchloride (1:1), methylenchloride, methylenchloride/methanol (1:1) and methanol. Host recognition experiments revealed a synergism between the fractions, with the methylenchloride fraction being most important. Several fatty acids, a-tocopherol and related compounds, and sterols, have been identified so far. Their activity in inducing host recognition behaviour has been investigated.

**Odor-mediated influence of the hyperparasitoid *Alloxysta victrix* O-25 on the defense behavior of *Sitobion avenae*.**

Gert Petersen, Wittko Francke, Urs Wyss

Institut fuer Phytopathologie, Universitaet Kiel, Hermann-Rodewald-Strasse 9 24118 Kiel, Germany Institut fŸr Organische Chemie, UniversitŠt Hamburg, Martin-Luther-King-Platz 6 20146 Hamburg, Germany.

When attacked by females of the primary parasitoid *Aphidius uzbekistanicus*, most of the aphids defend themselves by typical reactions such as jerking, kicking, lifting of the abdomen, turning, dropping and releasing cornicle secretions. In contrast to *A. uzbekistanicus*, *A. victrix* females have to mount the aphids in order to probe the presence of host larvae of the aphid primary parasitoid. Mounting is initially inhibited by similar defense reactions, except that aphid dropping and cornicle secretions do then usually not occur. Later, however, the aphids reduce their defense activities and within about 50 seconds after the first contact with an *A. victrix* female, they have calmed down completely so that mounting can now easily be achieved. We found that the calming of the aphids by *A. victrix* females is obviously mediated by a mixture of hyperparasitoid volatiles, most likely in combination with antennal drumming on the aphid antennae. Until now 6-methyl-5-heptene-2-on and some iridoid substances have been identified to be involved in the calming down process.

**Plant stress, plant resistance, herbivores, and predators: bottom-up effects.** **O-26**

Lorenzetti, Fran ois and John Thor Arnason

Department of Biology, University of Ottawa, 30 Marie Curie, Ottawa, Ontario, Canada, K1N 6N5.

Laboratory, greenhouse, and field experiments were conducted to investigate the interaction between plant stress and host-plant resistance on an insect herbivore, and the effects of this plant-insect interaction on a predator in the maize-aphid-ladybird tritrophic system. Maize genotypes used in these investigations included the DIMBOA-deficient *bxbx*, high-DIMBOA genotypes, and a cross between one of the latter and the *bxbx* material, yielding an intermediate-DIMBOA genotype. Plant stress was imposed by either high planting density or low nitrogen fertilization. HPLC analyses on tissue of greenhouse grown plants indicated that DIMBOA increased significantly in stressed high-DIMBOA genotype plants, but not in stressed intermediate-DIMBOA genotype plants. Aphids (*R. maidis* ) on similar greenhouse grown plants did not develop on either the stressed or the non-stressed plants of the high-DIMBOA genotype, but developed significantly slower on stressed *bxbx* plants, indicating a strong interaction between plant stress and resistance level. A similar pattern was observed in field trials. In the lab, the duration of the larval stages of the ladybird *P. quatuordecimpunctata* was not affected when larvae were fed aphids from either non-stressed or stressed plants of either the high-DIMBOA or the DIMBOA-deficient genotypes. However, the duration of the pupal stage was slightly but significantly increased in the treatment where the larvae were fed aphids from stressed plants of the high-DIMBOA genotype. These results are discussed in the light of the different risks involved for ladybird beetles in a field situation in an effort to link lab results and population dynamics. For example, the scarcity of aphids on resistant plants in the field is likely to be a bigger threat to ladybird larvae than plant-mediated effects on their development. Survivorship of ladybird larvae in the field on differentially resistant maize genotypes will be presented in support of this conclusion.

**What is the basis of pederin polymorphism in *Paederus riparius* rove beetles?** **O-27**

Rupert Kellner and Lehrstuhl Tieroekologie II

Universitaet Bayreuth, D-95440, Bayreuth, Germany.

Pederin, a toxic amide found in the hemolymph of *Paederus* rove beetles, is accumulated by most females of *Paederus riparius*. These females lay eggs containing the toxin and thus are called (+)-females. Some females, however, do not transfer pederin into the eggs as they cannot accumulate the amide. Those are termed (-)-females. Larvae and males only store pederin acquired maternally or by ingestion. The polymorphism of females, which is also observed in other *Paederus* species, might have a genetic basis. As (+)-females produce (+)-females and (-)-females whereas the offspring of (-)-females comprises (-)-females again, the ability to accumulate pederin is clearly a matrilineal trait. However, (+)-females descended from (-)-females can be obtained experimentally if the specimens are fed with eggs from (+)-females during larval development. This indicates existence of microorganisms needed for pederin biosynthesis which are transferred from (+)-females to their offspring.

**Induction of plant synomones by oviposition of a phytophagous insect.** **O-28**

Torsten Meiners and Monika Hilker

Freie UniversitŠt Berlin, Institut fŸr Angewandte Zoologie, Haderslebenerstr. 9, D-12163 Berlin, Germany.

Oviposition of the elm leaf beetle *Xanthogaleruca luteola* (Coleoptera, Chrysomelidae) induced elm leaves (*Ulmus campestris*) to emit volatiles which attracted the egg parasitoid *Oomyzus gallerucae* (Hymenoptera, Eulophidae). On the other hand, the egg parasitoid showed no positive response towards volatiles from elm leaves artificially damaged or damaged only by feeding elm leaf beetles. Also host eggs themselves did not attract the parasitoids. The attractiveness of elm leaves onto which eggs had been deposited was not caused by a synergism of volatiles from eggs and leaves, since an airstream combining volatiles from eggs and volatiles from leaves without egg depositions did not attract the parasitoid. Instead, attractive volatiles from elm leaves were experimentally induced by slightly scraping the leaf surface like the elm leaf beetle usually does prior to oviposition and smearing a secretion squeezed from the beetle's oviduct onto this wound. In order to examine whether the induction of synomones was locally restricted to a single elm leaf or systemically active, elm leaves without any egg depositions were

removed from a twig with leaves onto which eggs were deposited. Scent from these egg-free leaves neighbored to egg-laden leaves was also attractive to the parasitoid. The role of jasmonic acid for induction of plant synomones by oviposition is currently studied.

# Monday, June 22, P.M., Interspecific Interactions I

Chairperson: Monika Hilker, Freie UniversitŠt, Berlin

**O-29** 1330-1345 **NŽlida E. G—mez**
Use of plant-derived secondary metabolites: evidence of chemical defense in tortoise-beetle larvae (Chrysomelidae).

**O-30** 1345-1400 **John P. Berry**
Chemical ecology of the mountain gorilla (*Gorilla gorilla beringei*), with special reference to antimicrobial constituents.

**O-31** 1400-1415 **M. Lawrence Henneman**
Fruit damage cues and selection of fruits by a parasitoid of the walnut-feeding tephritid *Rhagoletis juglandis* .

**O-32** 1415-1430 **Ted Turlings**
Factors that determine variability in herbivore-induced odor emissions in corn plants.

# Interspecific Interactions II

Chairperson: Meena Haribal, Boyce Thompson Institute

**O-33** 1430-1500 **Ritsuo Nishida**
Chemistry of plant-insect associations: phytochemical niches in butterflies.

1500-1530 **Coffee break**

**O-34** 1530-1545 **Inoue A. Takashi**
Oviposition responses of *Papilio polytes* butterflies to extracts of 8 species of Rutaceae plants.

**O-35** 1400-1415 **Drew Harvell**
Mechanisms of sea fan (*Gorgonia spp*) chemical resistance to a fungal pathogen (*Aspergillus sp*).

**O-36** 1600-1615 **Hiromi Sasagawa**
Co-evolution of plant and insect: the interaction between the Oriental Orchid (*Cymbidium floribundum* Lindl.) and the Japanese Honeybee.

# Annual Business Meeting

1630-1730

# Monday, June 22, evening

## Special Lecture (Silverstein-Simeone Award)

Chairperson: Thomas Eisner, Cornell University

**O-37** 1900-2000 **Ian Baldwin**
The chemical ecology of *Nicotiana attenuata*: an example of adaptive plasticity.

## Poster Session

2000-2200 Kennedy Hall and Corson-Mudd Atrium (refreshments)

**Use of plant-derived secondary metabolites: evidence of chemical defense in tortoise-beetle larvae (Chrysomelidae).** **O-29**

NŽlida E. G—mez[1], Ludger Witte[2] and Thomas Hartmann[2]

[1]Smithsonian Tropical Research Institute, Panama.
[2]InstitutfŸr Pharmazeutische Biologie, Technische UniversitŠt Braunschweig, Germany.

Tortoise beetle larvae construct a maneuverable dorsal structure using fecula and cast skins. These structures, also called fecal shields, functions as physical defense against enemies. However, their role as chemical barrier has received little attention. We chemically examined fecal shields of *Eurypedus nigrosignata* and the foliage of its host plant, *Cordia curassavica* (Boraginaceae). *Cordia curassavica* produced volatile monoterpenes and sesquiterpenes as major secondary compounds, which were grouped into the §-terpinene, a-pinene and sabinene chemotypes. Fecal shields contained the same terpenoid profile as its hostplant. The concentration of plant-derived compounds in the fecal shield appeared to depend on larval age and plant chemical type. The fecal shields increased the larval survival in the field. In the laboratory, *E. nigrosignata* fecula had feeding deterrent activity. Fecal structures seemed to retain volatile terpenes much better than loose fecula. We also discuss implications of carrying a defensive structure containing volatile compounds.

**Chemical ecology of the mountain gorilla (*Gorilla gorilla beringei*), special reference to antimicrobial constituents.** **O-30** **with**

John P. Berry and Eloy Rodriguez

L. H. Bailey Hortorium, Cornell University, Ithaca, NY 14853, USA.

Understanding the chemical ecology of endangered species has potentially important applications for protection of these species and their habitats. We have investigated the phytochemical aspects of herbivory in the highly endangered mountain gorilla (*Gorilla gorilla beringei*). This work was conducted in Bwindi Impenetrable and Mgahinga National Parks, Uganda, home to approximately 300, or half of the world's population of, mountain gorillas. Analysis of nutritional and non-nutritional chemistry of the plant-based diet, in conjunction with behavioral and microbiological data, reveal important and "interactive" contributions of these chemical parameters to the feeding ecology of this anthropoid primate. Both behavioral and physiological adaptations allow mountain gorillas to cope with presence of plant toxins, and obtain adequate nutrition, in these marginal habitats. In particular, we have focused on the diverse antimicrobial, and otherwise biologically-active, chemical constituents of plant foods, and the susceptibility of the enteric microflora with respect to these constituents. Possible roles, or consequences, of the antimicrobial chemistry of food plants with respect to both pathogenic and beneficial endosymbionts will be discussed.

**Fruit damage cues and selection of fruits by a parasitoid of the walnut-feeding tephritid *Rhagoletis juglandis* .** **O-31**

M. Lawrence Henneman[1], Robert A. Raguso[1], Junji Takabayashi [2], and Eric G. Dyreson[1]

[1]Dept. of Ecology and Evolutionary Biology, University of Arizona, Tucson AZ 85721 USA.
[2]Pesticide Research Institute, Faculty of Agriculture, Kyoto University, Kyoto 606, Japan.

*Biosteres juglandis* (Braconidae) attacks larvae of *Rhagoletis juglandis* (Tephritidate) as they feed within the husks of walnuts. Behavioral and chemical assays were conducted to study the relationship between odor profiles and wasp foraging decisions. Volatiles of infested, uninfested, and artificially damaged fruits at various phenological stages were analyzed using a gas chromatograph/mass spectrometer. Profiles from fruits from the same tree were most similar, with high variation among trees, but overall the profiles of damaged fruits tended to resemble those of uninfested fruits more than they did those of infested fruits. In behavioral assays, wasps did not distinguish between infested and damaged fruits when there was visual damage, but were

able to distinguish infested from uninfested fruits in the wind tunnel when no visual cues were available. Wasps probably rely on visual information when it is present, but are able to use volatiles to distinguish fruits when visual cues are not available.

**Factors that determine variability in herbivore-induced odor emissions in corn plants.** **O-32**

Ted Turlings, Sandrine GouinguenŽ, and Thomas Degen.

Laboratory of Animal Ecology and Entomology, Institute of Zoology, University of Neuch‰tel CH-2007, Switzerland.

Plants under herbivore attack emit specific odors that are used by parasitoids and predators as cues to locate the herbivores. Using corn as our model plant, we have studied several aspects of the phenomenon of herbivore-induced odor emission. Different induction methods and a range of corn varieties were used to study the variability in odor emissions. Among different plant genotypes, quantitative and qualitative variation can be considerable. Some compounds (e.g. linalool, (*E* )-4,8-dimethyl-1,3,7-nonatriene, indole, (*E* )-b-farnesene) are released by all genotypes, while others (e.g. (*Z* )- b-ocimene, phenethyl acetate, (*E* )- b-caryophyllene) appear to be only released by some corn varieties. The composition of the odor blend does not seem to be dependent on the type of herbivore that does the damage, but the intensity of the emission can vary tremendously depending on which herbivore feeds on the plants. Our results do not give much support for the notion that plants may emit specific chemical signals that indicate which herbivore feeds on them, but they do suggest that it will be possible to select and breed plants that emit particularly large quantities of odors that attract beneficial insects.

**Chemistry of plant-insect associations: phytochemical niches in butterflies.** **O-33**

Ritsuo Nishida

Chemical Ecology Lab, Faculty of Agriculture, Kyoto University, Kyoto 606, Japan.

Plants produce a vast array of secondary metabolites as a result, in part, of coevolutionary interactions with phytophagous organisms. A number of insects have not only overcome these chemical barriers but positively utilize them as host-finding cues during both oviposition and larval feeding. In recent years, suites of contact oviposition stimulants have been characterized for several classes of butterfly species (Papilionidae, Pieridae and Nymphalidae). Some of them, instead of detoxifying plant poisons, sequester these compounds, thereby obtaining immediate defensive advantage, enabling them to establish new adaptive zones. Papilionids and

danaines are exemplified here to illustrate the use of plant toxins both as host finding cues and subsequently for defense by sequestering the compounds, in that an assessment of host plant quality with the target toxins would most efficiently guarantee consequent protection.

## Oviposition responses of *Papilio polytes* butterflies to extracts of 8 species of Rutaceae plants. O-34

Inoue A. Takashi[1], Seta Kazuaki[2], Niki Sawako[2], Hashimoto Kanako[2] and Asaoka Kiyoshi[1]

[1]National Institute of Sericultural and Entomological Science: Japan 305-8634 Ibaraki, Tsukuba, Ohwashi 1-2, NISES, Neurophysiology.
[2]Biopark of Tokyo, Adachi: JAPAN 121-0064 Tokyo, Adachi, Hogima 2-17-1.

*Papilio polytes* is one of the most common butterflies in South-East Asia and it sometimes becomes a pest of orange trees. We have begun to study this butterfly's host plant recognition system. We researched female butterflies' responses to methanol extracts of 8 species of Rutaceae plants (*Citrus* spp., *Toddalia asiatica, Skimmia japonica, Phellodendron amurense, Zanthoxylum piperitum, Z. ailanthoides, Evodia glauca* and *Orixa japonica* ) using the method described by Ichinose. These plants could be divided into three groups according to the butterflies' responses. In group-1 are plants to which female butterflies always showed an ovipositional response (*Citrus* spp. and *T. asiatica* ). In group-2 are plants (*Z. piperitum*, *Z. ailanthoides* and *Evodia glauca* ). to which are sometimes rejected for oviposition by females that will oviposit on the group-1 plants. In group-3 are plants that are almost always rejected (*P. amurense*, *S. japonica* and *O. japonica* ). The results of the responses to the extracts of the plants in group-1 or group-3 almost corresponded to the observations in the field or green house, but responses to the plants in group-2 sometimes differed from these observations. We have observed that most female butterflies in the field or green house do lay eggs on these plants.

**Mechanisms of sea fan (*Gorgonia spp*) chemical resistance to a pathogen (*Aspergillus sp*).** **O-35**

C.D. Harvell[1], K. Kim, R. Taylor[2], and E. Rodriguez[2].

[1]Section of Ecology and Systematics, Corson Hall, Cornell University, Ithaca, NY 14853, USA.
[2]Bailey Hortorium, Cornell University, Ithaca, NY 14853, USA.

Although diseases of corals appear to be on the increase, very few of the causative agents have been identified, and additionally nothing is known about mechanisms of coral resistance to disease. The fungal pathogen *Aspergillus* syndowii is currently affecting sea fans (*Gorgonia ventalina* and *G. flabellum* ) throughout the Caribbean. The disease causes variable-sized lesions and even colony death at some sites. In the Bahamas and Florida Keys, many monitored lesions enter long-term stasis (> 12 months) suggesting the possibility of effective resistance against the fungus within colonies. The chemical extracts of the two species of sea fan have significant and similar anti-fungal activity against *Aspergillus* in laboratory assays (MIC 6-9 mg/ml). Anti-fungal activity is higher in extracts of diseased sea fans (*Gorgonia ventalina* ) proximal to fungal lesions relative to non-diseased controls, suggesting the possibility of localized induction of resistance. Chemical elucidation of the partially purified compound (MIC= 0.9 mg/ml) is on-going.

## Co-evolution of plant and insect: the interaction between the Oriental Orchid (*Cymbidium floribundum* Lindl.) and the Japanese Honeybee. O-36

Hiromi Sasagawa[1] and Shigeru Matsuyama[2]

[1]PRESTO "Intelligence and Synthesis", Japan Science and Technology Corporation (JST): c/o National Institute of Sericultural and Entomological Science, Tsukuba, Ibaraki, 305, Japan.
[2]Inst. of Applied Biochemistry, Univ. of Tsukuba, Tsukuba, Ibaraki, 305, Japan.

The Japanese honeybee (*Apis cerana japonica* Rad.:*Acj*) and the European honeybee (*Apis mellifera* L.:*Am*) share the same habitat in Japan. The red and white flower varieties of the oriental orchid (*Cymbidium floribundum* Lindl.:*Cf*) attract workers, drones, queens as well as the entire swarming colonies of *Acj*, but not of *Am*. The Nasonov gland and the mandibular glands extracts induced aggregation behavior in both species, but the GC profiles of the extracts from *Am* and *Acj* were significantly different. This is due to the fact that the flower scent mimics the Nasonov and mandibular glands pheromone of *Acj*. We report here: (1) the identification of more than 15 semiochemical compounds ((S)-linalool, 4 types of linalool oxide, fatty acids) of *Cf* and Nasonov gland of *Acj*, (2) 3-hydroxyoctanoic acid was found in *Cf* scent and in the mandibular glands of *Acj*, (3) the identification of *Cf* original compounds (alkanals, gamma-lactones, Alkanols) attract only *Acj*, (4) 2-heptanone was also found in *Cf* scent and in the mandibular glands of both *Acj* and *Am*., (5) differences between *Acj* and *Am* might be due to differences in their semiochemical components as well as their sensitivity to and recognition of these semiochemicals.

## The chemical ecology of *Nicotiana attenuata*: an example of adaptive plasticity. O-37

Ian Baldwin

Max-Planck-Institut of Chemical Ecology, Tatzendpromenade 1a, 07745 Jena, Germany.

# Tuesday, June 23, Function and use of Bioactive Molecules

Chairpersons: Jerrold Meinwald and Frank Schršder, Cornell University

**O-38** 0900-0930 **Jon Clardy**
Natural products and their macromolecular receptors

**O-39** 0930-1030 **David Cane**
Making sense of nature's scents: how nature makes terpenes

1030-1100 **Coffee break**

**O-40** 1100-1130 **Ashit Ganguly**
Drugs from natural sources

**O-41** 1130-1145 **Jacques Einhorn**
Structural characterization of the oleander scale *Aspidiotus nerii* sex pheromone : a long-term international adventure.

**O-42** 1145-1200 **Carsten Muller**
Strategies for the identification of waterborne chemical cues: sex pheromones in *Nereis succinea*.

**O-43** 1200-1215 **Frank C. Schršder**
Combinatorial chemistry in insects: a library of defensive

**O-44** 1215 1230 **JosŽ-L. Giner**
Biometric Synthesis of Insecticidal Steriods from Petunias.

**O-45** 1230 1245 **Eric A. Schmelz**
Induced 20-hydroxyecdysone accumulation in spinach: insect feeding and jasmonates increase levels of *de novo* biosynthesis.

**O-46** 1245-1300 **Shigefumi Kuwahara**
Synthesis of Some Pheromone Components of *Nezara viridula* and *Piezodorus hybneri*.

## Afternoon free for recreation and informal discussion groups

**Natural products and their macromolecular receptors.** **O-38**

Jon Clardy

Department of Chemistry, Cornell University, Ithaca, NY 14853

**Making sense of nature's scents: how nature makes terpenes.** **O-39**

David Cane

Department of Chemistry, Brown University, Providence, RI 02912

**Drugs from natural sources.** **O-40**

Ashit Ganguly

Schering-Plough Research Institute, Kenilworth, NJ 07033

**Structural characterization of the oleander scale *Aspidiotus nerii* sex pheromone : a long-term international adventure.** **O-41**

Jacques Einhorn[1], Angel Guerrero[2], Paul-Henri Ducrot[1], Fran ois-Didier Boyer[1], Mary Gieselmann[3] and Wendell Roelofs[3]

[1]UnitŽ de Phytopharmacie et MŽdiateurs Chimiques, INRA, route de St Cyr, 78026 Versailles Cedex, France.
[2]Department of Biological Organic Chemistry, CID (CSIC), Jordi Girona 18-26, 08034-Barcelona, Spain.
[3]New York State Agricultural Experiment Station, Cornell University, Geneva, NY 14456-0462, USA.

The sex pheromone emitted by the female oleander scale *Aspidiotus nerii* (Homoptera : Diaspididae) has been isolated and characterized as (1*R, 2S*)-*cis*-2-isopropenyl-1-(4'-methyl-4'-penten-1'-yl)-cyclobutaneethanol acetate using advanced MS and NMR spectroscopic methods as well as a variety of microderivatization sequences. The structure including the relative stereochemistry has been confirmed by stereo- and enantioselective synthesis of the four possible stereoisomers. The absolute configuration has been determined by comparison of the activity of the *cis* (1*S,*2*R*) and (1*R,*2*S*) enantiomers towards *A. nerii* males with that displayed by the natural material in greenhouse bioassays and field tests. The structure of this sesquiterpenoid pheromone is not only new in the coccids but appears as completely original in the pheromone field. Initiated independently in France and in

USA more than 15 years ago, the success of this project was made possible by an intense and diverse international cooperation that started in the early 90's.

**Strategies for the identification of waterborne chemical cues: pheromones in *Nereis succinea*.** **O-42 sex**

C. T. Muller[1], M. Beckmann[2] and J. D. Hardege[1]

[1]School of Pure and Applied Biology, Cardiff University of Wales, PO Box 915, Cardiff CF1 3TL, U.K.
[2]Dept. of Biology, Hongkong University of Science and Technology, Clear Water Bay, Kow Loon, Hongkong.

The isolation of waterborne chemical cues still represents a challenge in chemical ecology. The recent evaluation of the structure of a waterborne pheromone of *Nereis succinea* can be described as a major step forward The work started with the observation that female *Nereis succinea* excrete remarkable amounts of pheromone prior to any release of gametes. Like in insects where the transfer medium (air) is sampled in this case sea water which had been exposed to one or several females was used. The following processing of the sample allowed the bioactive compound to be detected along the whole purification to ensure no bias to the work and ended with a single substance sample large enough to undertake NMR studies. Any derivatisation and/or addition of most buffers and other chemicals was excluded in order to keep track of the bioactivity throughout the whole separation. Even under such constraints it was possible to achieve the isolation and identification of this substance in a relatively short time. The sample was filtered (sterile 0.45um, ultra <1000D) to remove any particles and larger molecules and freeze dried to reduce the volume. After desalting (BioRad P2) the subsequent separation via HPLC (size exclusion, reversed phase) yielded only one pure compound which was submitted to the NMR. The analysis revealed the substance to be Cysteinyl-glutathion which was confirmed by comparison to the synthetic product. Cysteinyl-glutathion or "Nereithione" is, therefore, the second truly waterborne cue identified in *Nereis succinea* and the purification strategy developed could be generally suitable for "waterborne" cues in the marine environment.

**Combinatorial chemistry in insects: a library of defensive macrocyclic polyamines produced by a ladybird beetle.** **O-43**

Frank C. Schršder[1]*, Jay J. Farmer[1], Athula B. Attygalle[1], Scott R. Smedley[2], Thomas Eisner[3], Jerrold Meinwald[1]

[1]Dept. of Chemistry, Cornell University, Ithaca, NY 14853.

[2]Dept. of Biology, Trinity College, Hartford, CT 06106.

[3]Section of Neurobiology and Behavior, Seeley G. Mudd Hall, Cornell University, Ithaca, NY 14853.

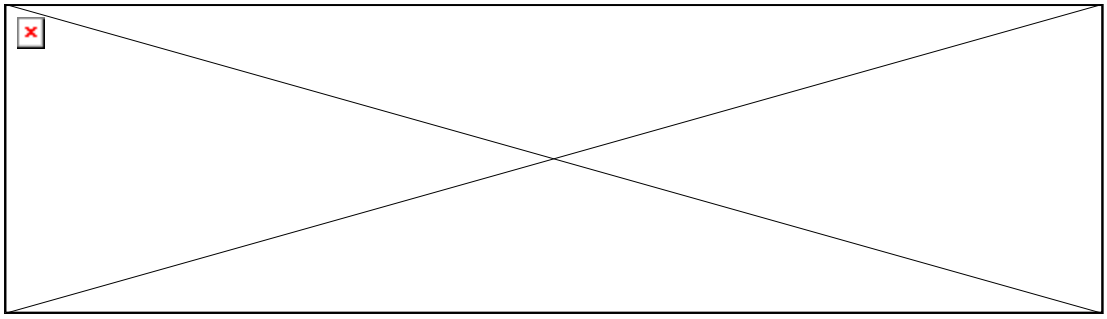
The pupal defensive secretion of the squash beetle (*Epilachna borealis*) is composed principally of a combinatorial library of macrocyclic polyamines. These compounds constitute a new family of natural products, characterized by lactonic structures with extremely large ring-sizes (26 to several hundred members), which are based on the apparently non-selective oligomerization of a small set of homologous building blocks (**1**-**3**). Slow, spontaneous intramolecular rearrangement of the macrocycles further increases the structural diversity.

**Biometric Synthesis of Insecticidal Steriods from Petunias.** **O-44**

JosŽ-L. Giner and Juan Faraldos

Dept. of Chemistry, State University of New York - ESF, Syracuse, NY, 13210, USA.

The chemical structures of the insecticidal natural products present in *Petunia* species were determined in the laboratory of Carl Elliger at the USDA Center at Albany, California. They are steroidal compounds bearing orthoester groups in the side chain. The most potent petuniasteroids exhibit $ED_{50}$'s against lepidopteran larvae such as *Heliopthis zea* of about 1 ppm. The bicyclic orthoester is essential for bioactivity. We have developed an efficient synthesis of petuniasteroids. This synthesis features a reaction that produces the bicyclic orthoester in a fashion mimicking the natural biosynthetic process. This biomimetic reaction is exceptionally facile and proceeds in near quantitative yield.



**Induced 20-hydroxyecdysone accumulation in spinach: insect feeding and jasmonates increase levels of *de novo* biosynthesis.** **O-45**

Eric A. Schmelz[1], Robert J. Grebenok[2], David W. Galbraith[3], and William S. Bowers[1]

[1]Dept. of Entomology, University of Arizona, Tucson, AZ 85721, USA.
[2]Dept. of Biology, Canisius College, Buffalo, NY 14208, USA.
[3]Dept. of Plant Sci., University of Arizona, Tucson, AZ 85721, USA.

Phytoecdysteroids are believed to serve a defensive function in plants, however this paradigm has not been conclusively demonstrated. Closer inspection of plant responses to insect herbivory may suggest better tests of this hypothesis. The roots of spinach (*Spinacia oleracea* ) rapidly induce 20-hydroxyecdysone (20E) concentrations, 2-6 fold above controls, following treatments including mechanical damage, insect herbivory, or additions of the plant wound-hormone analog methyl jasmonate (MJ). Induction inhibition studies with methyl salicylate support the involvement of the jasmonic acid pathway in signaling this response. Labeling studies, using intact plants and $^{14}$C-mevalonic acid, demonstrate that root 20E induction is largely caused by increases in root *de novo* biosynthesis. Root PE induction is not associated with concurrent increases in membrane sterols, thus indicating a level of pathway specificity in this response . The numerous similarities between PE induction and known inducible plant defenses, lend new support to the hypothesis that PE's may aid in plant defense against insects.

**Synthesis of Some Pheromone Components of *Nezara viridula* and *Piezodorus hybneri*.** **O-46**

Shigefumi Kuwahara,* Daisuke Itoh, and Walter Soares Leal[#]

Laboratory of Agricultural Chemicals, Faculty of Agriculture, Ibaraki University, 3-21-1 Ami-machi, Inashiki-gun, Ibaraki 300-0393, Japan.
[#] Laboratory of Chemical Prospecting, National Institute of Sericultural and Entomological Science, Tsukuba 305-8634, Japan.

A convenient stereoselective synthesis of (2*S*,3*R*,6*S*,7*Z*)- and (2*R*,3*S*,6*S*,7*Z*)-2,3-epoxy-7,10-bisaboladienes, sex pheromone components of *Nezara viridula*, will be reported. The former was synthesized from (*S*)-4-methyl-3-cyclohexene-1-carboxylic acid in only four steps and 19% overall yield. Its transformation into the latter was accomplished through 3 steps in 64 % yield by inverting the configuration of the epoxide ring. On the other hand, a simple synthesis of both enantiomers of 15-hexadecanolide, a sex

pheromone component of *Piezodorus hybneri*, will also be reported. They were synthesized in 5 steps by using ethyl (*R*)-3-hydroxybutanoate as the only chiral source.

# Wednesday, June 24, A.M.

## Chemistry of Bioactive Molecules

Chairperson: Athula Attygalle, Cornell University

**O-47** 0900-0930 **Koji Nakanishi**
Natural products chemistry: past, present and future.

## Interspecific Interactions II, continued

Chairperson: Athula Attygalle, Cornell University

**O-48** 0930-0945 **Brian K. Penney**
Chemical and physical defenses of *Cadlina marginata* (Opisthobranchia: Nudibranchia).

**O-49** 0945-1000 **Valerie J. Paul**
Chemical diversity in seaweeds: implications for herbivores.

**O-50** 1000-1015 **Dietland MŸller-Schwarze**
Do beaver (*Castor canadensis*) process food by leaching? Behavioral and chemical experiments.

**O-51** 1015-1030 **Chris Mullin**
Modeling of pollen chemoreception in *Diabrotica* beetles.

1030-1100 **Coffee break**

**O-52** 1100-1115 **Brice A. McPherson**
Feeding stimulants for subterranean termites isolated from associated fungi in laboratory colonies.

**O-53** 1115-1130 **Eduardo Mateus**
Relationship between the attack level by the processionary moth *Thaumetopoea pityocampa* and volatile monoterpene composition for twelve pine species.

**O-54** 1130-1145 **Cheryl A. Krasowski**
Compounds of alfalfa (*Medicago sativa*) root nodule extract which are phagostimulatory to clover root curculio (*Sitona hispidulus*) neonates.

**O-55** 1145-1200 **Kotaro Konno**
Gaba, glycine and b-alanine: amino acids secreted in digestive juice of herbivorous insects that protect against plant phenolics.

**O-56** 1200-1215 **Sanford D. Eigenbrode**
Principles and ecological implications of differential adhesion by an insect predator to plant surface waxes.

1215-1330 **Lunch break**

**Natural products chemistry: past, present and future.** **O-47**

Koji Nakanishi

Department of Chemistry, Columbia University, New York, NY 10027

**Chemical and physical defenses of *Cadlina marginata* (Opisthobranchia: Nudibranchia).** **O-48**

Brian K. Penney

Department of Biological Sciences, University of Alberta, Edmonton, AB, CANADA T6G 2E9; and Bamfield Marine Station, Bamfield, BC, CANADA V0R 1B0

Dorid nudibranchs have been extensively investigated by chemists for their putative defenses. However, these investigations have generally not used ecologically relevant predators, and have ignored the physical defenses (spicules) of the dorids. I investigated the defenses of *Cadlina marginata* (Chromodorididae) against common Pacific Northwest invertebrate generalist predators. Spicule investment in *C. marginata* is heavier in the mantle than in the foot, rhinophores, and gills, suggesting a defensive role. Extracts from *C. marginata* deter feeding by *Cancer productus*, while spicules have no effect. Neither spicules nor extracts alone deterred *Anthopleura elegantissima*, but extracts and spicules combined significantly deterred feeding. Evidently, predators respond differently to nudibranch chemical defenses, and spicules may supplement the effectiveness of compounds in these cases.

**Chemical diversity in seaweeds: implications for herbivores.** **O-49**

Valerie J. Paul

University of Guam Marine Laboratory, UOG Station, Mangilao, Guam 96923

Seaweeds in coral reef habitats experience intense grazing by generalist herbivores; therefore it is not surprising that tropical seaweeds produce a diverse array of chemical defenses. Many seaweeds occupy a variety of reef habitats, and the types and concentrations of compounds they produce will vary among collection sites. This chemical variation among collections of different seaweeds can influence the feeding and behavior of both generalist and specialist herbivores. The red alga *Portieria hornemannii* varies in the types and concentrations of monoterpenes (apakaochtodenes A and B) it produces in different reef sites on Guam. These metabolites deter grazing by reef fishes and the sea hare *Aplysia parvula*; however, *A. parvula* still prefers *P. hornemannii* over most other seaweeds. Chemical variation in seaweeds can be very pronounced for the filamentous cyanobacteria, which form large mats in many coral reef habitats. Most generalist reef herbivores avoid these chemically rich seaweeds. However, one specialist, the sea hare *Stylocheilus longicauda*, feeds on *Lyngbya majuscula*, a common cyanobacterium in reef habitats. The larvae of *S. longicauda* also preferentially settle on *L. majuscula*. Chemical variation in *L. majuscula* can influence the feeding behavior of adults and the settlement behavior of the larvae.

**Do beaver (*Castor canadensis*) process food by leaching?** **O-50**

**Behavioral and chemical experiments.**

Dietland MŸller-Schwarze[1], Heather Brashear[1], Robin Kinnel[2], Katharine A. Hintz[2] and Jason S. Kingsbury[2]

[1]SUNY College of Environmental Science & Forestry, Syracuse, NY 13210, USA.
[2]Dept. of Chemistry, Hamilton College, Clinton, NY 13323, USA.

Beaver store branches and feed in the water. We present an idea and preliminary experimental results. Do beaver condition less palatable food by leaching out in water digestion-inhibiting secondary plant metabolites? In two experiments, we soaked unpalatable red maple (*Acer rubrum* ) sticks in a pond and in tubs with ground water, respectively, for periods of 1 to 36 days and presented the sticks to free-living beaver at 10 colonies. Soaking for 1 to 2 days increased acceptance by beaver slightly. During their natural feeding behavior, beaver tend to leave branches of certain tree species in the water for 1 to 2 days before consuming the bark. Chemically, red maple is very rich in total phenolics. The dried bark contains 1.4% phenolics, the most abundant of which is acertannin. In laboratory experiments, large amounts of phenolics leached into the water, peaking after about 4 days of soaking. Processing unpalatable food by leaching may be an important feeding strategy by beaver and may force wildlife managers to reconsider what vegetation to classify as suitable for beaver.

**Modeling of pollen chemoreception in *Diabrotica* beetles.** **O-51**

Chris Mullin, Ben Hollister, Jae Hak Kim and Sisi Lin

Pesticide Research Lab, Department of Entomology, Pennsylvania State University, University Park, PA, 16802 USA.

Computer molecular modeling is a powerful technique to determine quantitative structure-function relationships for three-dimensional spatial interactions at protein chemoreceptors. Organ-specific herbivory is primarily dictated by taste, which in the insect occurs at sensory neurons that are a direct connection between the central nervous system and the outside. We are investigating the phagostimulatory and antifeedant ingredients in pollen that jointly limit its acceptability as a food. In pollens are high contents of short-chain neutral amino acids and some lipids, polyamides and flavonoids that provide stimulatory inputs for pollen feeding and specialization in adult corn rootworms. L-alanine was the most stimulatory amino acid followed by L-serine and §-alanine among the 52 amino acids studied. The presence of inhibitory amino acids, sugars, and other components indicates that a net integration of positive and negative chemosensory inputs is driving pollen consumption. Structure/dose-response correlations were determined for the phagoactive factors, and evaluated based on concentrations representative of pollen. Co-docking of structures to equal or homologous atomic sites of energy minimized model taste conformers was performed, guided by behavioral potency relative to 3-D goodness of fit. Molecular modeling techniques have allowed development of representative chemical templates to predict structural features for potent amino acid phagostimulation and potent alkaloid and terpenoid antifeedant taste in adult beetles.

**Feeding stimulants for subterranean termites isolated from associated fungi in laboratory colonies.** **O-52**

Brice A. McPherson and David L. Wood

Department of Environmental Science, Policy, and Management, Division of Insect Biology, University of California, Berkeley, CA 94720, USA.

The influences of substrate chemistry on subterranean termite (*Isoptera: Rhinotermitidae* ) feeding behavior are largely unknown. In addition to constitutive wood chemistry, the presence of fungi in dead wood may alter its chemical profile and affect behavioral responses of termites. Fungus-colonized filter paper taken from laboratory colonies of *Reticulitermes hesperus* , the western subterranean termite, yielded polar extracts that were stimulatory in cellulose TLC feeding assays against three North American *Reticulitermes* species. Open column chromatography and HPLC of polar extracts showed that stimulatory activity was not limited to one fraction. Fungi were isolated from the colonized filter paper, grown in cultures, and extracts were assayed. Extracts from a zygomycete soil fungus isolated from the termite colonies that produced the filter paper extracts were stimulatory for the three termite species in feeding assays. Although nonpolar extracts of the fungus-colonized filter paper were deterrent, those from the zygomycete elicited feeding. In contrast to the fungus-cultivating Termitidae, obligate termite-fungus associations have never been shown for the Rhinotermitidae. Although the nature of the relationship between the zygomycete and *Reticulitermes* species is not clear, this appears to be the first evidence that fungal metabolites can serve as feeding stimulants for lower termites.

**Relationship between the attack level by the processionary moth *Thaumetopoea pityocampa* and volatile monoterpene composition for twelve pine species.** **O-53**

Eduardo Mateus, Helena Farral, Qing-He Zhang and Maria Rosa Paiva

New University of Lisbon, Faculty of Sciences and Technology, Department of Environmental Sciences and Engineering, 2825 Monte Caparica, Portugal.

For several phytophagous forest insects, primary attraction, that is the process of host selection, has been correlated with variations of the monoterpene composition in the volatiles emitted by the trees. The winter pine processionary moth, *Thaumetopoea pityocampa*, is a pest on *Pinus* and *Cedrus*, attaining serious economic importance in the Mediterranean region. In the present work, the volatile monoterpene composition of 12 pine species, 3 of which were native - *P. pinea*, *P. pinaster* and *P. halepensis* - and 9 introduced - *P. nigra*, *P. brutia, P. patula, P. radiata, P. taeda, P. elliotti, P. kesyia, P. sylvestris* and *P. eldarica* - was analyzed and related to the level of attack by the processionary moth. Needles from all pine species were sampled between June and September 1996. Except for *P. eldarica*, all trees originated from a plot planted in 1991 in central Portugal, Abrantes region (N 39¼ 26«; W 8¼ 04«). *P. eldarica* twigs were collected 50 Kms further North, from 25 years old trees. For all samples, head space of the needles was collected, using Solid Phase Micro Extraction (SPME) and analyzed by High-Resolution Gas Chromatography (HRGC) on a DB-5 (J&W Scientific) fused-silica capillary column. Monoterpene identification was made by mass spectrometry (MS) and standard co-injection. A total of 16 - were identified from the sample headspaces of all species: thujene, a-pinene, camphene, sabinene b-pinene, myrcene, a-phellandrene, d-3-carene, a-terpinene, *p* -cymene, limonene, b-phellandrene, ocimene, g -terpinene, terpinolene and linalool. A statistical analysis was performed and a significant correlation coefficient was found between the relative amounts of the monoterpenes emitted, by the different species, and the attack level by the processionary moth.

**Compounds of alfalfa (*Medicago sativa*) root nodule extract which are phagostimulatory to clover root curculio (*Sitona hispidulus*) neonates.** **O-54**

Cheryl A. Krasowski[1], Christopher A. Mullin[2], and Arthur A. Hower[2]

[1]Intercollege Program in Plant Physiology, Penn State University, Univ. Park, PA 16802, USA.
[2]Dept. of Entomology, Penn State University, Univ. Park, PA 16802, USA.

Alfalfa (*Medicago sativa*), a leading forage legume in the United States, forms nitrogen-fixing nodules when inoculated with the appropriate symbiont Rhizobium meliloti. Unfortunately, alfalfa is then susceptible to nodule herbivory by early instars of the clover root curculio (*Sitona hispidulus*). This results in diminished productivity, forage quality, and stand persistence if the plant cannot readily or adequately compensate for nodule and root damage. To more effectively reduce *M. sativa* herbivory by *S. hispidulus*, we must better understand the root nodule-insect larvae interaction. A bioassay employing agar cylinders (3 mm x 3 mm diam.) which allows first instars of *S. hispidulus* to mimic burrowing behavior common to their nodule feeding habit has been developed. It has been used to drive fractionation of cold water-hot ethanol extractions of wildtype *M. sativa* nodules. This 2 hr bioassay has demonstrated that upon centrifugation (1800X G) of nodule extract, the supernatant has significantly greater activity ($p < 0.05$) than the pelleted material and that methanol and water soluble components of the supernatant have significantly greater activity than nonpolar components of the supernatant. Double column ion exchange chromatography has allowed investigation of phagostimulatory abilities of classes of compounds including amino acids. organic acids, and neutral components. This on-going work suggests that a suite of chemicals of various ionic classes may interact to provide the neonate with sufficient cues to stimulate feeding This work was supported by The Root Biology Training Program, a unit of the DOE/NSF/USDA Collaborative Research in Plant Biology Program, funded by NSF grant BIR-9220330.

## Gaba, glycine and b-alanine: amino acids secreted in digestive juice of herbivorous insects that protect against plant phenolics. O-55

Kotaro Konno, Hiroe Yasui, Chikara Hirayama, Sachiko Okada, and Masatoshi Nakamura

National Institute of Sericultural and Entomological Science, 1-2 Ohwashi, Tsukuba, Ibaraki 305-8634, Japan.

Leaves of the privet tree *Ligustrum obtusifolium* contain oleuropein, a phenolic compound, which causes a strong protein-denaturing activity that makes protein non-nutritive by decreasing lysine content [1]. High concentration of glycine exists in the digestive juice of several Lepidoptera and Hymenoptera species specialized in the privet tree [2,3]. As glycine inhibits the denaturing activity, these insects seem to secrete glycine to counter_adverse effects of the denaturation (e.g. loss of lysine, inactivation of digestive enzymes) [1,2]. However, not all the privet specialists have glycine in there digestive juice. Here, we report that high concentration of GABA, a compound known as a neurotransmitter, exists in the digestive juice of larvae of a privet-specialist butterfly. In another specialist, b-alanine was found. GABA and b-alanine, as well as glycine, inhibited the denaturing activity in physiological concentration. Several amino acids, amine, and ammonium ion also inhibited the denaturing activity, indicating that the amino residue of GABA, glycine and b-alanine are responsible for the inhibition. Inhibitory activities were much higher in GABA, glycine and b-alanine, which were found in digestive juice, than in alanine and ammonium ion, which were not. These data suggest that GABA has an ecological role, and that herbivores have independently developed different but effective chemical strategies to counter a plant chemical during evolution.

**Literature**

1. Konno K., Yasui H., Hirayama C., Shinbo H.: J. Chem. Ecol. 24, 735 (1998)
2. Konno K., Hirayama C., Shinbo H.: J. Insect Physiol. 43, 217 (1997)
3. Konno K., Hirayama C., Shinbo H.: Comp. Biochem. Physiol 115**A**, 229 (1996)

**Principles and ecological implications of differential adhesion by an predator to plant surface waxes.** **O-56 insect**

Sanford D. Eigenbrode and Nelson Kabalo

University of Idaho, Department of Plant, Soil and Entomological Sciences, Moscow, ID, USA.

Active insect predators must adhere well to plant surfaces to forage effectively, and this adhesion depends upon the physico-chemical properties of the leaf surface. This is illustrated by experiments with 9 genetic variants of *Brassica oleracea* with different surface wax composition and structure. Adhesive force produced by first instar *Chrysoperla plorabunda* varies by 2 orders of magnitude among these genotypes. In turn, adhesive force is positively correlated with the proportion of time the larvae spend walking on the plant surfaces. Lastly, the proportion of time larvae spend walking is related to the effectiveness of the predators attacking *P. xylostella* larvae on caged plants and in small arenas. The results indicate that predator-prey dynamics involving *C. plorabunda* in the field will be influenced by surface waxes of the plants. To understand the principles involved *C. plorabunda* tarsal adhesion to pure wax compounds and plant wax extracts deposited on glass was measured. Pure primary alcohols, fatty acids, and an alkane were deposited as amorphous films or as crystals densely covering a glass surface and assessed for effects on adhesion by *C. plorabunda* larvae. Differences in adhesion among these artificial substrates permit partial isolation of the physical and chemical characteristics of waxes that can affect adhesion and effectiveness of a generalist predator.

# Wednesday, June 24, P.M.

# Interspecific Interactions II, continued

Chairpersons: Chris Mullin, Pennsylvania State University
Sanford Eigenbrode, University of Idaho

**O-57** 1330-1345 **Jihong Jiang**
Antifungal activity of quinolizidine alkaloids extracted from *Ophora alopecuroides.*

**O-58** 1345-1400 **Benedict Hollister**
Sensory coding of phagostimulatory amino acid chemistry from host pollens for adult western corn rootworm.

**O-59** 1545-1600 **Justin Schmidt**
Chemical composition of the allomones of adult and immature vinegaroons *Mastigoproctus giganteus*.

**O-60** 1415-1430 **Meena Haribal**
Oviposition responses by experienced zebra swallowtail butterflies, *Eurytides marcellus* to the volatiles present in hexane extract of their host *Asimina triloba*.

**O-61** 1430-1445 **Bong-Seop Kil**
Phytotoxic and anti-microbial effects of chemical substances from *Artemisia lavandulaefolia*.

**O-62** 1445-1500 **Gus E. Dria**
Trichome constituents of *Calamintha ashei*.

1500-1530 **Coffee break**

**O-63** 1530 1545 **Maureen Carter**
Host plant chemistry influences oviposition choice of the spicebush swallowtail butterfly.

**O-64** 1545-1600 **Boguang Zhao**
Oviposition Responses of the Spruce Budworm, *Choristoneura fumiferana* (LEP.: Tortricidae) to Aliphatic and Aromatic Carboxylic Acids, and Some Methyl Esters.

**O-65** 1600-1615 **Kyung Saeng Boo and Jeon Hong**
Composition and circadian rhythm in release of sex pheromone in *Aphis spiraecola* (Homoptera: Aphididae)

**O-66** 1615-1630 **Manfred Ayasse**
Flower-specific variation of pollinator-attracting odor signals and reproductive strategies in the early spider orchid *Ophrys sphegodes*.

# Human Health Applications

Chairperson: Eloy Rodriguez, Cornell University

**O-67** 1630-1700 **Charles Arntzen**
Creating transgenic plants for pharmaceutical production

# Banquet

1930 **Statler Ballroom**

**Antifungal activity of quinolizidine alkaloids extracted from *Ophora alopecuroides.*** **O-57**

Jihong Jiang[1] and Boguang Zhao[2]

[1]Department of Biology, Xuzhou Normal University, Xuzhou 221009, P.R. China
[2]Forestry University, Nanjing 210037, P.R. China

*Sophora alopecuroides* is a common Chinese plant, well known as an abundant source of quinolizidine alkaloids. Although these alkaloids have a wide range of useful biological activities, their antifungal properties are largely unexplored. We tested alkaloid extracts of this plant and seven quinolizidine alkaloids isolated from the extracts for their antifungal activity on five species of fungi, including *Glomerella cingulata* (Chinese fir isolate), a serious pathogen of valuable Chinese fir. Quinolizidine alkaloid extracts had only moderate effect on the mycelial growth of fungi. They were more effective in inhibiting germination of conidia, and the effects on all five fungal species tested was significant, with the greatest effect on *G. cingulata* (effective concentration of extract causing inhibition, EC50 = 60 mg/ml). All seven quinolizidine alkaloids isolated from the extract showed significant inhibitory activity against the conidia of *G. cingulata*. Activity of these alkaloids was highly variable, with sophocarpine having the strongest effect on conidia (EC50 = 24 mg/ml). The structure-activity relationship between the quinolizidines and their antifungal activity is discussed.

**Sensory coding of phagostimulatory amino acid chemistry from pollens for adult western corn rootworm.** **O-58 host**

Benedict Hollister and Christopher A. Mullin

Pesticide Research Laboratory, Department of Entomology, The Pennsylvania State University, University park, PA, 16802, USA.

Sequential solvent extraction and bioassay, chromatographic fractionation, and HPLC analysis have identified amino acids as dominant phagostimulants from host pollen species for adult western corn rootworm. A strong correlation has been shown between taste cell inputs and phagostimulatory outputs for individual dominant amino acids with strongest responses elicited by L-alanine and L-serine. These compounds result in largely single cellular chemosensory responses. Simple ternary mixtures of dominant amino acids, approximating levels found in sweet corn and squash pollen, give equivalent phagostimulation compared to crude extracts. There is a clear dose-dependent response exhibiting greater maximal response, less variation, and steeper response slope relative to individual amino acids. Taste cell responses for ternary mixtures result in similar dose-response characteristics compared to individual amino acids, but with a distinctly multicellular firing pattern. These results suggest an across-fiber pattern as the sensory code driving pollen feeding behavior for adult western corn rootworm.

**Chemical composition of the allomones of adult and immature vinegaroons *Mastigoproctus giganteus*.** **O-59**

J. O. Schmidt[1], F. R. Dani[2], G. R. Jones[2], and E. D. Morgan[2].

[1]Southwestern Biological Institute, 1961 W. Brichta Dr., Tucson, Arizona 85745, USA.
[2]Chemical Ecology Group, Department of Chemistry, Keele University, Staffordshire ST5 5BG, UK.

Vinegaroons are so named for their ability to spray concentrated acetic acid at potential predators. In an early pioneering chemical investigation, Meinwald used elegant classical techniques to demonstrate that pooled samples from many individuals of unknown sex and instar consisted of aqueous solutions of concentrated acetic acid plus octanoic acid (Eisner et al. 1961. J. Insect Physiol. 6:272-98). Several questions have remained since that study: 1) does the secretion contain minor components that might enhance its activity; 2) does the composition of the secretion vary among individuals, or depend upon the sex and instar of the individual; 3) do the relatively tiny first instar free-living immatures produce and spray the secretion; and 4) could the secretion to serving as a pheromonal or communicative function in addition to being an allomone. Our investigation revealed: 1) in addition to acetic acid, water, and octanoic acid, vinegaroon spray contains small quantities of 2-ethyl-hexanoic acid, (Z)-5-octenoic acid, heptanoic acid, decanoic acid, hexanoic acid, and (E)-5- octenoic acid; 2) the chemical composition of the spray is relatively constant between males, females, and all four free-living instars of immatures; 3) the small first instars contain relatively large quantities of secretion which differs in no noticeable way from older individuals; 4) there is no evidence for a pheromonal or other non allomonal function of the secretion.

**Oviposition responses by experienced zebra swallowtail butterflies, *Eurytides marcellus* to the volatiles present in hexane extract of their host *Asimina triloba*.** **O-60**

Meena Haribal and Paul Feeny

Ecology and Systematics, Corson Hall, Cornell University, Ithaca, NY 14853, USA.

Experienced females of *Eurytides marcellus* laid significantly more eggs on a model leaf treated with contact stimulant in the presence of volatiles, extracted by hexane from the host plant, *Asimina triloba*. Higher number of eggs on leaves with volatile were related to more frequent approaches. This suggested that the volatiles act primarily by modifying pre-alighting behavior rather than by enhancing the probability of oviposition after each approach or landing. However, in the absence of contact stimulants significantly more eggs were laid on the volatiles treatment than on the hexane control. When hexane extract was further fractionated by column chromatography on silica gel into four fractions and bioassayed, there was no single fraction that accounted for all the activity. This could have been due to loss of synergism between the fractions. When each fraction of hexane extract was bioassayed against the total hexane extract in a choice test, there was no significant difference between the fractions and the total hexane extract. This suggests that the individual females learn to recognize a different set of chemical cues. This may be due to variations in populations or genetics.

**Phytotoxic and anti-microbial effects of chemical substances from *Artemisia lavandulaefolia*.** **O-61**

Bong-Seop Kil and Dong Min Han

Division of Life Science, Wonkwang University, Iksan, 570-749, Republic of Korea.

To investigate phytotoxic substances in *Artemisia lavandulaefolia*, the donor plant, and their biological activities, seed germination and seedling growth of receptor plants were examined at different concentrations of aqueous extracts of the donor plant. Germination of four receptor species was inhibited by the extracts, while seedling growth was decreased to a lesser degree than in the germination test. Essential oil of the plant extracted by Karlsruker's apparatus inhibited growth of several microorganisms and callus growth of *Pinellia ternata* and *Oryza sativa*.. The GC/MS method was employed for analysis and identification of allelochemicals from *A. lavandulaefolia* leaves. Sixty-one chemical substances were identified from essential oil of *A. lavandulaefolia*. The results of this experiment on seed germination, seedling growth,

anti-microbial test and tissue culture indicated that naturally occurring chemical substances from *A. lavandulaefolia* would be responsible for the phytotoxic and anti-microbial effects.

**Trichome constituents of *Calamintha ashei*.** **O-62**

Gus E. Dria and Jeffrey D. Weidenhamer

Department of Chemistry, Ashland University, Ashland, Ohio 44805, USA.

The perennial shrub *Calamintha ashei* occurs in the Florida scrub along Florida's Lake Wales ridge. *Calamintha* was previously shown to contain novel, water-soluble menthofuran monoterpenes which inhibit the germination and growth of grasses from the neighboring Florida sandhills. This study was undertaken to characterize the trichomes which dot the surface of *Calamintha* leaves. It was presumed that the trichomes would be found to contain the previously identified menthofurans. However, GC-MS analyses of trichomes individually removed from leaves show no evidence of the menthofurans, but do show three major constituents and several minor constituents that also can be detected in headspace by solid phase microextraction. The mass spectra of the three principal compounds all show a molecular ion at 204, indicative of sesquiterpenes. An extract of surface compounds was prepared by briefly dipping leaves in dichloromethane, followed by vacuum liquid chromatography and preparative HPLC to isolate these compounds. The mixture of the three principal constituents has proven extremely difficult to separate, suggesting that the compounds are very closely related isomers. A proton NMR of the mixture is consistent with sesquiterpenes, and proton-proton and carbon-proton correlation experiments are being undertaken to establish the structures of the compounds.

**Host plant chemistry influences oviposition choice of the spicebush swallowtail butterfly.** **O-63**

Maureen Carter and Paul Feeny

Department of Ecology and Systematics, Cornell University, Ithaca, NY 14853, USA.

The spicebush swallowtail, *Papilio troilus*, lays its eggs on Sassafras (*Sassafras albidum*), spicebush (*Lindera benzoin*), redbay (*Persea borbonia*) and camphortree (*Cinnamomum camphora*), plants in the family Lauraceae. No oviposition preference was recorded between sassafras and spicebush leaves. In one-choice tests females laid eggs on chemical extracts of the leaves of each of the four host plants. In two-choice experiments, females always preferred to oviposit on an extract of sassafras compared to an extract of one of the other three hosts. Previously we (Carter, Feeny and Haribal, in preparation) had identified one of the host plant chemicals acting as an oviposition stimulant in sassafras extract as 3-caffeoyl-muco-quinic acid (3-CmQA). Extracts of the other three hosts did not contain this compound. The addition of 3-CmQA alone to spicebush extract did not increase oviposition activity. It did, however, increase discrimination between hosts and non-hosts. When a fraction of sassafras extract containing 3-CmQA and other synergistic stimulants was added to spicebush extract, preference for sassafras extract was no longer recorded. Oviposition choice in the spicebush swallowtail is mediated by responses to different combinations of contact stimulants, probably interacting also with visual and volatile cues.

**Oviposition Responses of the Spruce Budworm, *Choristoneura Fumiferana* (LEP.: Tortricidae) to Aliphatic and Aromatic Carboxylic Acids, and Some Methyl Esters.** **O-64**

Boguang Zhao[1], Dorothy Langevin[2] and Gary G. Grant[2]

[1] Nanjing Forestry, University, Nanjing, P.R. China
[2] Canadian Forest Service, Sault Ste. Marie, Ontario, P6A 5M7 Canada

Organic acids are common constituents of insect host plants and are implicated increasingly as insect semiochemicals involved in oviposition and other behaviors. Using a dual-choice laboratory bioassay, we investigated the structure-activity relationship of carboxylic acids on the oviposition behavior of the spruce budworm, a major defoliator of coniferous forests in North America. Test compounds (10-100 mM solution) were applied to a filter paper substrate and compared to a solvent-treated control. At dosages of 78 - 780 nmol/cm$^2$, the stimulating activity of aliphatic monocarboxylic acids peaked at C8-C9 chain length. Strong activity was also observed with some di-carboxylic acids and longer chain unsaturated acids. Methyl esters of long chain acids tended to be deterrent. Some aromatic acids were also strongly stimulating although phenolic acids were not. At higher dosages, stimulating acids became repellent; the repellent dosage was dependent on chain length. Consistent with an olfactory mode of action, EAG responses elicited by C6-C16 aliphatic acids correlate with their behavior activity.

**Composition and circadian rhythm in release of sex pheromone in *Aphis spiraecola* (Homoptera: Aphididae)** **O-65**

Kyung Saeng Boo and Jeon Hong

Division of Applied Biology and Chemistry, College of Agriculture and Life Sciences, Seoul National University, Suwon 441-744, Korea.

*Aphis spiraecola* (Patch) was reared on the dropwort, for artificial production of sexual forms, for 3 generations under low temperature (12~18¡C ) and short day (12L:12D to 10L:14D) conditions. Sexual males were produced in the second generation and sexual females (oviparae) in the third generation. The percentage of males induced was 21%, at most, among the total progeny produced. These slender males were observed to mate with oviparae with distinctly swollen hind tibiae, which were raised together with the abdomen and waved from time to time during the photophase, as an obvious behavior of releasing sex pheromone. The air around these oviparae was collected and analyzed by a gas chromatograph for pheromone components to reveal (+)-(4αS, 7S, 7αR)-nepetalactone and (-)-(1R, 4αS, 7S, 7αR)-nepetalactol at the ratio of 6:1 ~ 8:1. Oviparae showed a circadian rhythm in releasing sex pheromone. They began to release nepetalactone with the start of lights-on and abruptly stopped with lights-off. Their sex pheromones were always detected during the photoperiod, even when the photophase was altered to a different period from 10L:14D to 16L:8D.

**Flower-specific variation of pollinator-attracting odor signals and reproductive strategies in the early spider orchid *Ophrys sphegodes*.** **O-66**

Manfred Ayasse[1], Florian P. Schiestl[1], Hannes F. Paulus[1], Christer Lšfstedt[2], Bill Hansson[2], Dirk Erdmann[3]and Wittko Francke[3]

[1]Institute of Zoology, University of Vienna, Althanstrasse 14, A-1090 Vienna, Austria
[2]Department of Ecology, Lund University, S-22362 Lund, Sweden.
[3]Institute of Organic Chemistry, D-20146 Hamburg, Germany.

The orchid *Ophrys sphegodes* is pollinated by sexually excited males of *Andrena nigroaenea*, which are lured to the flowers by visuaI cues and volatile semiochemicals. In *O. sphegodes*, visits of pollinators are rare: in 1996 we recorded that only 4.85% of 887 plants were visited by a pollinator. Therefore, *Ophrys* plants should have evolved strategies in order to optimize pollination events. Our data from the behavioral tests indicate that male bees learn the odor bouquets of individual flowers during mating attempts and recognize them in further encounters. They thus avoid trying to mate with flowers they have visited previously, but not with other flowers either on the same or on a different plant. We used GC-MS analyses to identify more than 100 compounds in the odor bouquets of *O. sphegodes*; 27 compounds were found to be ”biologically active” (GC-EAD analyses). GC analyses of individual flower odors showed that the floral volatile bouquets differed between plants as well as between flowers within an inflorescence. Biologically active compounds showed a smaller intraspecific variation of the odor bouquets as compared to non-active compounds. This can be explained by a higher selective pressure on the pollinator-attracting communication signal. We assume that variation in the odor bouquets of individual flowers within an inflorescence raises the chance of more than one flower being visited by the same male and thereby influences the plant’s pollination success and individual fitness.

**Creating transgenic plants for pharmaceutical production.** **O-67**

Charles Arntzen

Boyce Thompson Institute, Ithaca, NY 14853

# Monday, June 22

# 20:00-22:00

# Poster Session

# Kennedy Hall

# Posters P1 to P21

**Sex pheromones and fatty acid desaturases in *Drosophila.*** **P-1**

Claude Wicker-Thomas, Renaud Dallerac, CŽline Henriet, Carole Labeur and Jean-Marc Jallon

UniversitŽ Paris-Sud, URA C.N.R.S. 1491, B‰t. 446, 91405, Orsay CŽdex, France.

In *Drosophila*, pheromones are long-chain unsaturated cuticular hydrocarbons and there is a marked qualitative polymorphism for pheromones among species. A fatty acid desaturase gene has been recently characterized in *D. melanogaster*. The polypeptide encoded by the gene has a high homology with the D9 desaturases from rat and yeast desaturases and could be involved in the biosynthesis of pheromones. In other *Drosophila* species desaturase genes have also been evidenced. The possible co-evolution of contact pheromones and desaturases in the genus *Drosophila* is discussed.

**Pheromonal enhancement of artificial diet for the predator *Podisus nigrispinus* .** **P-2**

Miryan D. A. Coracini[1], Evaldo F. Vilela[1], JosŽ C. Zanuncio[1] and Jeffrey R. Aldrich[2]

[1] Lab. of Biological Control and Pheromones, Federal University of Vi osa, MG, 36571-000 Brazil.
[2] USDA-ARS Insect Chemical Ecology Laboratory, Beltsville, Maryland 20705, USA.

An artificial diet for rearing *Podisus nigrispinus* (Heteroptera:Pentatomidae), a caterpillar predator of *Eucalyptus* pests in Brazil, was developed by Saavedra and co-workers, in our laboratory. The 'artificial caterpillar' needs improvements because the developmental time of the insects reared on the diet is prolonged, and the acceptance of the diet by the predator is still unsatisfactory, probably due to the lack of kairomonal cues. Extracts of the sex pheromone gland of *P. nigrispinus* brushed on artificial caterpillars made them as attractive as larvae of *Musca domestica*; therefore, we evaluated the synthetic sex pheromone components as stimulants for *P. nigrispinus* feeding on artificial diet. The male-produced pheromones of pentatomids act as aggregation pheromones, as males attract not only females, but also other males and nymphs. We treated artificial caterpillars with the following synthetic pheromone formulations: 1) the complete blend of *P. nigrispinus* pheromone [*(E)*-2-hexenal + a-terpineol + benzyl alcohol + *trans*- piperitol + linalool], 2) the blend of the three major components and, 3) each component singly. Artificial caterpillars were treated with 0.05 mg of pheromone fomulation/ml dichlormethane. Diets containing either the blend of the three major components or *trans*-piperitol, produced insects with the same weights and the same nymph to adult developmental time, as insects fed on larvae of *M. domestica* (Tukey test, p=0.05). Despite the improvement of attraction to the diet, the complete pheromone blend failed to improve nymphal development, while artificial diet containing single or major pheromone components proved to be useful in this respect. Nevertheless, further work needs to be carried out in order to test the development of subsequent generations of the predator and to improve the quality of the artificial diet itself.

**Attractants for honey bee swarms: does nasonov "pheromone" act as a pheromone or simply as a detectable odor?** **P-3**

J. O. Schmidt

Carl Hayden Bee Research Center, USDA-ARS, 2000 E. Allen Road, Tucson, AZ 85719, USA

Nasonov "pheromone" is used by honey bees to orient conspecifics seeking locational or directional information. During reproductive swarming, it is released by scouts both on the swarm cluster and at entrances of potential nest sites. However, evidence suggests that nectar or water foragers use Nasonov simply to mark low odor sources of forage and does not act as a pheromone. I tested the hypothesis that Nasonov odor is a true pheromone for nest seeking bees, versus an alternative hypothesis that Nasonov is merely a bee generated odor no better than other odors to aid other searching scouts to locate low-odor nest sites. To test these hypotheses, crossover design experiments using artificial nest cavities in which Nasonov pheromone and a test odor were altered over a season were conducted. Test odors included a floral odor (linalool), a strong odorous plant natural product (clove oil), an animal odor expected to be released by cavity-inhabiting animals (skatole), and the pheromone of wax moths (scavengers that consume the remnants of dead honey bee colonies). In these comparison tests Nasonov pheromone was 4.75 times more attractive than no odor, 3.0 times more attractive than linalool, 6.5 times more attractive than skatole, 8.5 times more attractive than clove oil, and at least twice as attractive as wax moth pheromone. The results support the hypothesis that Nasonov pheromone acts as a true pheromone rather than simply as a detectable odor.

**Synthesis of 9*E, 11Z* -Hexadecadienal: Sex attractant pheromone of the pecan nut casebearer, *Acrobasis nuxvorella* Neunzig.** **P-4**

Linda C. Passaro and Francis X. Webster

Department of Chemistry, SUNY College of Environmental Science and Forestry, 1 Forestry Drive, Syracuse, NY 13210, USA.

The pecan nut casebearer, *Acrobasis nuxvorella* Nuenzig, is one of the most significant pests to the pecan nut industry in the southwestern United States and Mexico. Extensive research has been aimed at finding more environmentally sound ways to manage and control the pecan nut casebearer. An alternative syntheses of 9*E,* 11Z -hexadecadienal, the major sex attractant of the pecan nut casebearer has been carried out. Starting with 1-heptyn-3-ol, 9*E*, 11Z

-hexadecadienal was obtained in seven steps and characterized using NMR. Key transformations include an orthoester Claisen rearrangement, rearrangement of the product to a 2*E*, 4Z-diene ester using basic $Al_20_3$, and Cu (I) I coupling of 1-(tetrahydropyran-2-yloxy)-7-heptylmagnesium bromide to a diene acetate.

**Heading upwind - Pheromone-induced behaviours in pine sawflies.** **P-5**

Fredrik …strand and Olle Anderbrant

Dept. of Ecology, Animal Ecology, Lund University, S-223 62 Lund, Sweden.

This study was set out to investigate the pheromone-induced behaviour of the European pine sawfly, *Neodiprion sertifer* (Hym: Diprionidae). The possible effect of different climatic factors on males' performance was studied with the aid of a weather station. Males were marked with an instant marker on the thorax, and released in the field downwind pheromone traps, baited with 100 mg of the sex pheromone. Males were either released 5m from one trap, or 50 or 200 m away from five traps, in line approximately perpendicular to the wind. As a control males were released identically but without any pheromone source present. A total of 1824 males were released, and 80% of them took off. Males entered a platform where their behaviour prior to take off was studied. The direction of grooming, wing fanning and take off were scored and found to be significantly directed to the upwind direction (one of eight possible directions) in all experiments including the control At 5m, 50m and 200m experiments the males showed a significantly stronger preference for displaying the behaviours in the upwind direction than in the control. The frequency of wing fanning was negatively correlated with wind speed, whereas the frequency of grooming did not show any relation to wind speed. Recapture figures after 24h were 26% and 4% for the 50m and 200m experiments, respectively. Males took off significantly faster in the 200m experiments compared with the control, and even faster in the 5m experiments. Low radiation lowered the take off frequency significantly in both 50m and 200m experiments. Pheromone-induced behaviour in diprionids seem to be less distinct than *e.g.* in Lepidoptera. The micro climate can have significant effect on the male's performance close to a pheromone trap.

**Sex pheromone of the rosy russian gypsy moth, (*Lymantria mathura* Moore).** **P-6**

James E. Oliver[1,*] Joseph C. Dickens[2], Marina Zlotina[3], and Victor C. Mastro[4]

[1]Insect Chemical Ecology Laboratory, Plant Sciences Institute, ARS, Beltsville Agricultural Research Center, USDA, Beltsville, MD 20705-2350, USA.
[2]Vegetable Laboratory, Plant Sciences Institute, ARS, Beltsville Agricultural Research Center, Beltsville, MD 20705-2350.
[3]Department of Entomology, University of Massachusetts, Amherst MA 0100, USA.
[4]Otis Plant Protection Center USDA, APHIS, PPQ Building # 1398, Otis ANGB, MA 02542-500, USA.

Two electrophysiologically active compounds have been isolated and identified from abdominal tip extracts of female *Lymantria mathura* Moore: Z,Z,Z-3,6,9-nonadecatriene **1** and its monoepoxide Z,Z-(9*S*, 10*R*)-9,10-epoxy-3,6-nonadecadiene **4a.** This type of pheromone system, unusual for a Lymantriidae, is more typical of those found in the families Arctiidae, Noctuidae and Geometridae.

**Chemical communication in the silver-washed fritillary, *Argynnis paphia* (Lepidoptera, Nymphalidae).** **P-7**

P. Ockenfels, M. BopprŽ, O. Fischer and S. Schulz*

Institute of Forest Zoology, University of Freiburg, Bertoldstrasse 17, 79085 Freiburg i. Br., Germany.
* Institute for Organic Chemistry, University of Braunschweig, 38106 Braunschweig, Germany.

In the course of studies on pheromone communication in the Silver-washed Fritillary, we have found that the male valves are glandular and that the female ‘dorsal sac’, described by URBAHN (1913) and TREUSCH (1971), contains a glutinous liquid after mating. Morphological and GLC/MS studies have revealed that during copulation males transfer a-farnesene and 1-dodecyl-acetate from the valves into the female’s dorsal sac where it coagulates and apparently serves as a repellent for conspecific males.

**Identification of the scent-marking sex pheromone of the North American decorator wasps *Eucerceris rubripes, conata* and *tricolor.*** **P-8**

S. R. Clarke[2], F. R. Dani[2], G. R. Jones[2], E. D. Morgan[2] and J. O. Schmidt[1]
[1]Agricultural Research Science, 2000 E. Allen Road, Tucson, Arizona, 85719-1596, USA.
[2]Department of Chemistry, Keele University, Staffordshire, ST5 5BG, UK.

The males of the decorator wasps *Eucerceris* have been observed to display abdomen-dragging behaviour on plants surrounding their nest in order to secrete a scent-marking sex pheromone. This serves to alert females to their territory for courtship and mating. Three species, *E. rubripes*, *E. conata* and *E. tricolor* were analysed by GC/MS. The gas chromatograms of all three species revealed the presence in large quantities of one common volatile compound in the male body extracts. Utilising micro-reactions and with the aid of FT-IR, $^{1}H$ and $^{13}C$ NMR spectroscopy (*R*), (*Z*)-3-hexenyl 3-hydroxybutanoate (**1**) was proposed and confirmed by synthesis. The absolute configuration of the chiral centre was determined by preparing the Mosher's ester. 2- and 3-hexenoic acid and the aromatic compounds (**2**), (**3**) and (**4**) were found in varying quantities in both males and females along with hydrocarbons and fatty acids.

**Chemical ecology of honeybees in Asia: chemical analyses of worker pheromone components** **P-9**

Shigeru Matsuyama[1], Takahisa Suzuki[1] and Hiromi Sasagawa[2]

[1]Institute of Applied Biochemistry, University of Tsukuba, 1-1-1, Ten-noudai, Tsukuba, Ibaraki 305, Japan.
[2]PRESTO "Intelligence and Synthesis", Japan Science and Technology Corporation (JST), c/o: National Institute of Sericultural and Entomological Sciences (NISES). 1-2 Oowashi, Tsukuba, Ibaraki 305-8634, Japan.

We report the pheromone gland chemistry of several honeybee species in Asia, *Apis cerana japonica (Acj), A. c. cerana (Acc), A. c. indica (Aci), A. c. himalaya (Ach), A. dorsata (Ad), A. laboriosa (Al)*and *A. andreniformis (Aa).* In the Nasonov gland extracts of *Acj, Acc , Aci*, and *Ach*, none of the compounds known as Nasonov pheromone in *A. mellifera (Am)* was detected. Instead, linalool oxide was identified as common component in *Acj* and *Acc.* 3-Hydroxyoctanoic acid was identified as a major mandibular gland component in *Acj, Aci* and *Acc.* Isopentyl acetate was identified in the sting gland among the species tested. These results suggest that

Asian honeybees had developed different sets of chemicals as their Nasonov and mandibular gland pheromone from those of *Am*.

**Multi-methylene interrupted alkadienes in insects.** **P-10**

Larry L. Jackson, David Cummings, Julie A. Brown, Jamie Smith and Joe Evans

Chemistry and Biochemistry Department, Montana State University, Bozeman, MT 59717, USA.

Alkadienes of the type $CH_3$-($CH_2$)x-CH=CH-($CH_2$)y-CH=CH-($CH_2$)z-$CH_3$, where y is 14-24, are rather rare; but have been recently characterized in the cuticular lipids of a number of insects. *Drosophila willistoni* have four tritriacontadienes: 8,24-33:2 [C8=C16=C9]; 7,25-33:2 [C7=C18=C8]; 5,25-33:2 [C5=C20=C8]; 5,27-33:2 [C5=C22=C6]; and two pentatricontadienes: 9,25-35:2 [C9=C16=C10]; and 8,26-35:2 [C8=C18=C9] as major components of their cuticular hydrocarbons. The tritriacontadienes each increase nearly five fold during the first three days following eclosion in flies reared at $25^0$C. The hydrocarbon profiles of males and females are very similar and only close examination reveals the differences that occur as the flies reach sexual maturity. *D. malerkotliana* have 5,27-33:2 [C5=C22=C6] as their major tritricontadiene and lesser quantities of 6,26-33:2 [C6=C20=C7]. However the alkadienes of *D. nebulosa* are the pentatricosadienes 7,27-35:2 [C7=C20=C8]; 5,25:2 [C5=C20=C10] and 5,27-35:2 [C5=C22=C8]. The Caribbean fruit fly (*Anastrepha suspensa*) also makes large quantities of C33 and C35 multi-methylene interrupted alkadienes during their first few days after eclosion. The three major alkadienes are: 9,23-33:2 [C9=C14=C10]; 8,24-33:2 [C8=C16=C9] and 9,25-35:2 [C9=C16=C10], where in each case one of the double bonds is nine carbons from the end. In *A. suspensa*, 9,25-35:2 is produced in great quantities immediately after eclosion; however as the flies mature, production of 35:2 decreases while production of the 33:2's increases and peaks at sexual maturity.

**Cuticular hydrocarbons suggest three lineages in *Reticulitermes* North America.** **P-11**

Marion Page, Lori J. Nelson, Michael I. Haverty and Brian T. Forschler[1]

Pacific Southwest Research Station, USDA Forest Service, P. O. Box 245, Berkeley, California 94701, USA.
[1]Department of Entomology, 413 Biological Sciences Bldg. University of Georgia, Athens, GA 30603-2603, USA.

Species specific cuticular hydrocarbon mixtures have utility for determining phylogenetic relationships. We report inferred degrees of relatedness among the chemical phenotypes of *Reticulitermes* from PAUP (Phylogenetic Analysis Using Parsimony) analyses of cuticular hydrocarbon characters. One hundred eighteen *Reticulitermes* colonies collected from Georgia, Arizona, Nevada and California were used in these analyses. Collections of *Reticulitermes* contain representatives of the five recognized species: *R. hesperus* Banks, *R. tibialis* Banks, *R. virginicus* Banks, *R. hageni* Banks, and *R. flavipes* (Kollar). Initial maxium parsimony analyses sorted the 118 colonies into 25 chemical phenotypes. Subsequent analyses, using the ancestral species *Coptotermes formosanus* and *Heterotermes aureus* as outgroups, sorted *Reticulitermes* taxa into three major lineages, I, II and III, each characterized by a different set of dominant hydrocarbon components. *Reticulitermes* in Lineage I have chemical phenotypes with a preponderance of 5,17-dimethylalkanes. Hydrocarbon profiles of Lineage II have a preponderance of internally and terminally branched monomethylalkanes and 11,15-dimethylalkanes. Lineage III phenotypes make an abundance of alkenes, dienes, trienes, tetraenes and one pentaene. Within Lineage III two major clades are evident. The phenotypes with major quantities of the diene C27:2 are separate from those phenotypes with the major component being the alkene C27:1. Bootstrap analyses of 1000 replications and decay indices provided statistical support and robustness for this putative chemical based phylogeny.

**Host- and non-host-plant discrimination in *Zygaena trifolii* (esper, 1783) and *Z. transalpina* (esper, 1780) (Lepidoptera, Zygaenidae).** **P-12**

Tilmann MŸller[1] Peter Ockenfels[2], and Clas. M. Naumann[1]

[1]Zoologisches Forschungsinsitut und Museum Alexander Koenig, Adenauerallee 160, 53113 Bonn, Germany.
[2]Forstzoologisches Institut der UniversitŠt Freiburg, Bertoldstr. 17, 79098 Freiburg i.Br., Germany.

"Zygaenidae are highly cyanogenic, day-flying moths with a world-wide distribution. In the Palaearctic region the genus *Zygaena* Fabricius, 1775 with nearly 100 species represents one of the most striking examples of geographic variation and highly aposematic wing patterns ("the old world Heliconius"). The partial adaptation to cyanogenic larval foodplants is considered a fairly recent phenomenon linked with tremendous evolutionary success. The chemical protection of the moths has also contributed to complex reproductive strategies."
In the course of studies on the oviposition behaviour of two related *Zygaena* species, volatiles of the host-plants elicit the strongest EAG responses in comparison to non-host-plants, except for the cyanogenic *Trifolium repens*. The results are considered as an exemplification of a basic condition for behavioural discrimination between host- and non-host-plants by the female, prior to oviposition. The role of cyanogenic glucosides in host-plant recognition is discussed.

**Cuticular hydrocarbons and soldier defense secretions for chemotaxonomy P-13 of reticulitermes in North America.**

Michael I. Haverty[1], Lori J. Nelson[1], Brian T. Forschler[2], Laurence G. Cool[3], and Marion Page[1]

[1]Chemical Ecology of Forest Insects, Pacific Southwest Research Station, USDA Forest Service, P.O. Box 245, Berkeley, CA 94701, USA.
[2]Department of Entomology, University of Georgia, Athens, GA 30602, USA.
[3]Forest Products Laboratory, University of California, Richmond, CA 94804, USA.

Cuticular hydrocarbon mixtures and soldier defense secretions from samples of Reticulitermes collected from disparate locations in Georgia, New Mexico, Arizona, Nevada, and California were characterized and correlated with species determinations. Twenty-five hydrocarbon phenotypes, and at least 7 soldier defense secretion phenotypes have been identified. Hydrocarbons from the following classes have been found: normal alkanes, alkenes (1-5 double bonds), terminally branched and internally branched monomethylalkanes, dimethyl- and trimethylalkanes. Soldier defense secretions are comprised of 38 terpenoid compounds, including monoterpenes, sesquiterpenes, and a diterpene alcohol, geranyl linalool. The sesquiterpenes include γ-cadinene, γ-cadinene aldehyde, germacrene A, α-, §-himachalene, δ-amorphene, §-farnesene, and numerous other minor and unknown compounds. Some soldier defense secretion phenotypes correlate with more than one cuticular hydrocarbon phenotype, however each hydrocarbon phenotype is correlated with only one soldier defense secretion phenotype. On the basis of these chemical characterizations, we suggest that there are numerous undescribed taxa of Reticulitermes and conclude that the taxonomy of this genus in North America is in need of revision.

**Induction of isoprenoid volatile synthesis during oak bark beetle (*Scolytus intricatus*) maturation feeding.** **P-14**

Pavlina Vrkocova, Jiri Kindl, Jan Vrkoc and Bohumir Koutek

Institute of Organic Chemistry and Biochemistry, Academy of Sciences of the Czech Republic, Flemingovo nam. 2, CZ - 166 10 Praha, Czech Republic.

The European oak bark beetle (*Scolytus intricatus*) breeds in cut or weakened oak branches. After emerging, the young adults fly up to the crown of healthy trees and feed to maturation on 1-2 year old twigs. *S. intricatus* prefers the species of *Quercus* to the other broad leaves tree species [1]. Analyzing the volatiles that are emitted from fresh oak twigs during the maturation feeding, several terpenic compounds were found, (*E* )-b-ocimene being the major component of the volatiles. (*E* )-b-ocimene was released from herbivory damaged twigs in amount 15 - 20 % of the total volatiles found (measured as GC total peak area). Both undamaged and artificially damaged twigs release only minute amounts of this compound. It is known, that (*E* )-b-ocimene and others terpenoids, elicited from herbivore damaged plants by leaf-feeding caterpillars and a grasshopper (or applying a regurgitate of these species on mechanically damaged plants) can serve as a chemical signal that attracts natural enemies of the herbivore to the damaged plant [2]. Even maize plants injured by an herbivore that feeds inside the stem (stem borer, *Chillo partellus*) can trigger the emission of signals attracting parasitoids [3]. Searching behavior of predatores or parasites during the *S. intricatus* maturation feeding is not known so far. The similarity of induction of (*E*)-b-ocimene released by oak bark beetle maturation feeding on fresh oak twigs with de novo biosynthesis following a herbivory attack on green leaves of plants leads to a hypothesis that this plant signal could be also used as a cue for *S. intricatus* parasites.

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**Comparison of ovipositional responses of the 2 subspecies of *protenor* butterflies.** **P-15** ***papilio***

Seta Kazuaki[2], Inoue A. Takashi[1], Niki Sawako[2], Hashimoto Kanako[2] and Asaoka Kiyoshi[1]

[1]National Institute of Sericultural and Entomological Science, JAPAN 305-8634 Ibaraki, Tsukuba, Ohwashi 1-2, NISES, Neurophysiology
[2]Biopark of Tokyo, Adachi, JAPAN 121-0064 Tokyo, Adachi, Hogima 2-17-1

The Japanese *Papilio protenor* is divided into 2 subspecies. One is ssp. *demetrius* occurring between Honshu and Amani-Shotou (Northern area of Japan), and the other is ssp. *liukiuensis* occurring between Okinawa-Hontou and Yaeyama-Shotou (South-west end of Japan). These subspecies are easily distinguished from each other by the color patterns on the 2nd-4th instar larvae and on the adult female wing. The first subspecies' responses to some Rutaseae plants' extracts were already reported by Ichinose and Honda. In contrast, the later subspecies was not researched. We researched both subspecies oviposition responses to methanol extracts of 8 species of Rutaseae plants (*Citrus* spp., *Toddalia asiatica, Skimmia japonica, Phellodendron amurense, Zanthoxylum piperitum, Z. ailanthoides, Evodia glauca* and *Orixa japonica* ) using the method described by Ichinose. The subspecies did not significantly differ in their responses to those plants' extracts.

**GC-EAD analysis of semiochemicals of a pamphiliide sawfly (*Acantholyda erythrocephala* ).** **P-16**

Joseph K. Staples and Stephen A. Teale

State University of New York College of Environmental Science and Forestry Department of Environmental Forest Biology Syracuse, New York 13210, USA.

The Pine False Webworm (PFW) (*Acantholyda erythrocephla* ), a Pamphiliide sawfly that defoliates white pine, and to a lesser extent, red and Scotch pines, is currently in outbreak levels in northern New York and southern Ontario. The purpose of our research has been to develop a pheromone lure for this insect. Using GC-MS and GC-EAD, we have identified three female-produced compounds that elicit male antennal responses. The identity of the compounds was confirmed with GC-EAD analysis of synthetic samples. Field trapping experiments showed that male PFW are selectively attracted to specific combinations of the three compounds. Additionally, GC-EAD analysis of non host volatiles was conducted to identify those compounds that elicit antennal responses in male and female PFW. The results from these experiments will facilitate the development of future management strategies.

**Phytochemical basis for stalk borer resistance in maize.** **P-17**

A. Ramputh[1], D. Bergvinson[2], J.T. Arnason[1] and J. Simmonds[3]

[1]Department of Biology, U. of Ottawa, Ottawa, ON, K1N 6N5, Canada.
[2]CIMMYT, Mexico.
[3]ECORC, Agriculture Canada, Ottawa, ON K1A 0C6, Canada.

Lepidopteran stalk boring larvae cause economically significant losses to maize production throughout the world. Host plant resistance (HPR) is an effective and environmentally safe means of control for these pests. The basic challenge is identifying sources of resistance for insect-resistance breeding programs. Resistance mechanisms involve both toxic and mechanical defenses against insect pests. The hydroxamate based toxic defenses in maize are well studied. This present study focuses on newly discovered mechanical defenses of HPR involving phenolic dimers. Recent research has shown that cell wall bound phenolic acids can strengthen the cell wall through a peroxidase mediated dimerization that cross-links adjacent arabinoxylan molecules with diferulic acid (DFA). This is evident in the positive correlation between DFA content and leaf toughness ($r=0.68$). Genetic transformation of maize is now being used to generate germplasm for ECORC breeding programs. Genes are delivered into embryogenic cell cultures by a biolistic procedure and transgenic plants are recovered. The wheat gene, (germin), coding for oxalate oxidase has been transferred to maize. This enzyme generates $H_2O_2$ from Ca oxalate. It has been proposed that $H_2O_2$ dependent peroxidative formation of phenolic cross-links between cell wall polymers serves as a mechanism for cell wall stiffening and subsequent increased insect resistance. These transgenic lines are being analyzed for altered cell wall characteristics.

**Potential for insecticide discovery in temperate and tropical tree extracts.** **P-18**

Semir Omar[1], Krishan Goel[2], Bernard J.R. Philog ne[1] and John T. Arnason[1]

[1]Dept. Of Biology, University of Ottawa, Ottawa, Canada K1N 6N5.
[2]Domtar innovation Centre, Senneville, Canada.

Thirty extracts of wood and bark of hardwood trees from Eastern North America were compared with extracts from Tropical American trees for insect growth reducing activity in bioassay with European corn borer, *Ostrinia nubilalis*. The growth reducing activity of both wood and bark extracts of temperate trees was comparable to tropical extracts. Statistical analyses of relative growth rate (RGR) values showed that there were no significant difference between the bark extracts. However, the mean RGR values of wood extracts are significantly different from each other indicating that tropical wood extracts were more active.

**Biological activity of fifteen crude extracts from native plants against P-19 larvae of *Tecia solanivora* (Lepidoptera: Gelichiidae)**

B‡rbara Moreno[1], G. Castillo[2], M. V. Bejarano[2] Emilio Luque[2], V. Fajardo M.[3] and Margoth Su‡rez M. [1]

[1]Facultad de Ciencias -Universidad Nacional de Colombia, A.A. 27586 SantafŽ de Bogot‡ , Colombia
[2]Facultad de Agronom'a Universidad Nacional de Colombia A.A. 14490, SantafŽ de Bogot‡ , Colombia.
[3]Facultad de Ciencias- Universidad de Magallanes, casilla 113-D Punta Arenas, Chile.

Colombia, as other countries produces and consumes high amounts of different kind of potatoes (*Solanum tuberosum* and related) and many families depend economically on their cultures ( >100.000 Ha). During last decade great areas have been affected by the potatoes moth *Tecia solanivora* (Lepidoptera, Gelichiidae ) known as " moth from Guatemala", which destroys the tubers and have generated serious losses to producers incomes of about US $ 70 millions in 1997, in fields and storage[1,2]. In our continuous search for new natural bioactive compounds through bioassay- guided fractionation insecticidal and growth reducing properties of crude extracts of 15 native or growing species have been evaluate with the brine shrimp test (BST, with *Artemia salina* larvae[3]) and against 1st and 2nd instar larvae of the moth *Tecia solanivora* using a non choice assay with "creole potatoes" *( S. pureja* ) as natural diet. Alcoholic extracts of fifteen species belonging to Solanaceae (*Salpichroa diffusa, S. tristis, Nicandra physaloides, Jaborosa magellanica* ), Berberidaceae (*Berberis saboyana, B. monguiensis, B. tabiensis, B. cusianensis, B. carupensis. B. horrida* and *B. darwinii* ), Euphorbiaceae (*Hieronyma moritziana , H. macrocarpa* ), Valerianaceae (*Valeriana carnosa* ) and Myrtaceae (*Eucaliptus globulus* ) were assessed at least for three concentrations (5%, 3% and 1% ) with five replications. The Effective Concentration for 50% of individuals ($EC_{50}$ ppm) were estimated and observed effects such as physical and behavioral changes were analyzed. Active extracts were partitioned by organic solvents and separated and purified by chromatographic techniques in order to identify the responsible constituents of the produced affections. We thank Colciencias, CINDEC-UN and Universidad de Magallanes for financial support.

References:

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**Chemistry of wilted red maple (*Acer rubrum*).** **P-20**

Elizabeth Miller and Jeffrey D. Weidenhamer

Department of Chemistry, Ashland University, Ashland, Ohio 44805 USA.

Wilted leaves of the red maple tree (*Acer rubrum*) are toxic to horses. The most obvious result of this poisoning is a type of methemoglobinemia known as Heinz-Ehrlich body anemia. Currently, the only treatment for red maple poisoning is supportive care; a horse that has ingested 3g leaves/kg body weight has little chance of survival. Fresh leaves do not cause this poisoning, nor do leaves of other varieties of maples, whether they are wilted or not. This study was undertaken to investigate the chemistry of wilted red maple leaves for possible toxins. Extracts of both fresh and wilted leaves were prepared by methanol extraction, followed by removal of chlorophylls and other pigments with hexane. Analysis of the crude extracts by thin layer chromatography on silica revealed a compound unique to the wilted leaves which fluoresces blue under longwave UV light. A purified extract of the compound has been isolated by vacuum liquid chromatography and recrystallization. Proton NMR data suggest a complex structure containing many aliphatic protons. Further characterization of this compound is in progress.

**Cuticular hydrocarbons and their roles in the courtship behavior of three parasitoids of stored product insects.** **P-21**

Ralph W. Howard

USDA-ARS, USGMRL, 1515 College Ave., Manhattan, Kansas 66502 USA.

Cuticular hydrocarbons from *Cephalonomia tarsalis*, *C. waterstoni* (Hymenoptera: Bethylidae) and *Pteromalus cerealellae* (Hymenoptera: Pteromalidae) have been analyzed and are compared. Although all three species share a few common components, distinct qualitative and quantitative species and gender differences are found. Behavioral studies on the courtship behavior of these parasitoids have been conducted and the role of the hydrocarbons as possible pheromonal components examined. The cuticular hydrocarbons appear to be playing a close-range, gender identity role rather than serving as long range pheromonal cues.

# Monday, June 22

# 20:00-22:00

# Poster Session

# Corson-Mudd Hall

# Posters P22 to P54

**Molecules and Movement: Female *M. sexta* Moths and Hostplant Volatiles**. **P-22**

Wendy L. Mechaber and John G. Hildebrand

ARL Division of Neurobiology, University of Arizona, Tucson, AZ 85721, USA.

This report presents our recent findings on female M. sexta behavior stimulated by moth exposure to hostplant volatiles alone (no plant present) in a flight tunnel bioassay. These new data continue our work with the M. sexta-tomato hostplant model system. In our previous experiments with female moths and potted tomato plants, we documented the range of behaviors that individual female moths display in the presence of potted hostplants, e.g., odor-mediated upwind flight and pre-oviposition postures following continued exposure to plant volatiles. For these current experiments with volatiles, intact, potted hostplants were used as the odor source. Plant volatiles were trapped by dynamic headspace collection, and the classes of compounds contained within the samples were identified via GC-MS. The initial volatile samples were serially diluted and tested for their stimulatory effects on female moth behavior in the same flight tunnel bioassay. The most
concentrated volatile samples stimulated upwind orientation, while the increasingly more dilute volatile samples stimulated pre-oviposition postures.

**Phagostimulants from sunflower pollen for adult western corn rootworm.** **P-23**

Sisi Lin and Christopher A. Mullin

120 Pesticide Research Laboratory, The Pennsylvania State University, University park, PA 16802, USA.

Adult Diabroticites including western corn rootworm (WCR), *Diabrotica virgifera virgifera* LeConte, readily consume pollen both in nature and as a lab diet. Pollen, as a highly preferred *Diabrotica spp*. food, provides a unique opportunity for determining the phytochemosensory basis for an insect-plant interaction. The interacting phagostimulants for adult WCR in Giant Gray Stripe sunflower, *Helianthus annuus* L., pollen were isolated and identified to understand the chemical basis of pollinivory. Short-chain amino acids in the methanol-water extract of pollen have been identified in our lab as strong phagostimulants for *Diabrotica* both behaviorally and electrophysiologically. With bioassay driven fractionation and purification, we characterized WCR phagostimulants from an ethyl acetate fraction of sunflower pollen. Some lipids and mid-polarity compounds were found with significant phagostimulatory activity. Lipids including triglycerides, free fatty acids, phosphatidylethanolamine, phosphatidyl acid,

phosphatidylinositol, and phosphatidylcholine were active. Other phagostimulatory components included hydroxycinnamic acid-polyamine amides and two flavonoids. The structural characteristics of these phagoactive compounds and their additive or synergistic interactions on WCR feeding will be described.

**Behavioral and Physiological Responses of Two Insect Herbivores to Variation in Glucosinolates and Myrosinase Activity in *Brassica juncea*.** **P-24**

Qun Li and Sanford Eigenbrode

Division of Entomology, Department of Plant, Soil and Entomological Sciences, University of Idaho, Moscow, ID 83844-2339, USA.

Glucosinolates and their hydrolytic products have different effects on herbivore behavior and physiology. Glucosinolates are nonvolatile, stimulate feeding in crucifer specialists and deter some general feeders. Their hydrolysis products, especially the isothiocyanates, are volatile, attractive to some specialists, and broadly biocidal. Myrosinase is the enzyme that catalyzes hydrolysis of glucosinolates. We developed a model to examine the effect of varying both glucosinolate concentration and myrosinase activity on insect herbivores. Newly hatched larvae of the crucifer specialist, *Plutella xylostella*, and the general feeder, *Spodoptera eridania*, were observed on cotyledons of *Brassica juncea* plants with different levels of myrosinase activity and glucosinolate concentrations for 30 minutes. Neither myrosinase activity nor the dominant glucosinolate, sinigrin concentration showed significant effects on biting frequency and feeding damage by *P. xylostella*, but high sinigrin concentration significantly reduced biting frequency and feeding damage by *S. eridania*. High myrosinase activity depressed biting frequency and feeding damage by *S. eridania* irrespective of glucosinolate concentration, but the significance of the effect was marginal. Physiological effect was measured by comparing the larval weight change after feeding on the plants with different levels of myrosinase activity, glucosinolate concentrations, and different contents of glucosinolates. Behavioral and physiological responses of the two insect herbivores will be discussed in context of evolution of the glucosinolate/myrosinase binary defense system.

**Antagonistic effects of some carbohydrates on amino acid feeding stimulation in adult western corn rootworm, *Diabrotica virgifera virgifera* Leconte.** **P-25**

Jae Hak Kim and Christopher A. Mullin

Pesticide Research Laboratory, Department of Entomology, Pennsylvania State University, University Park, PA 16802, USA.

The effects of binary mixtures of phagostimulatory amino acids and carbohydrates on feeding responses of the adult western corn rootworm (WCR) were investigated. Among 12 carbohydrates tested in a choice bioassay, sucrose, fructose and glucose appeared to have antagonistic effects on consumption of phagostimulatory amino acids (L-alanine, L-serine and b-alanine) by nondiapause adult WCR. Neutral interactions were found between amino acids and other carbohydrates. On the contrary, synergistic interactions occurred between amino acids and carbohydrates in field-collected WCR. Different interactions may in part reflect history of food exposure by the adult stage of these populations or genetic difference.

**Allelopathic effects of *Artemisia capillaris* on selected species.** **P-26**

Jae Hong Lee, Bong-Seop Kil, Byung Sam Kim and Ji Hyun Kil

[1]Division of Life Science, Wonkwang University, Iksan, 570-749, Republic of Korea.
[2]National Institute of Environmental Research, Seoul, 122-706, Republic of Korea.

Allelopathic effects on germination and seedling growth of selected species were investigated with aqueous extracts and volatile substances from *A. capillaris*. The extracts inhibited the germination and seedling growth of several species including *Leonurus sibiricus*, but those of *Cosmos bipinatus* was not severely reduced. The inhibition effect was dependent on the concentration of volatiles. Various chemicals from *A. capillaris* essential oil were identified such as *n*-hexanal, terpinen-4-*ol*, etc. by GC/MS. These chemical substances seemed to be responsible for allelopathic effects.

**Attraction of *Epitragus salaei* Champion (Coleoptera: Tenebrionidae) to odor of mango inflorescence *Manguifera indica* L.** **P-27**

Jose Alfredo JimŽnez and Leopoldo Cruz L—pez

El Colegio de la Frontera Sur, Carretera Antiguo Aeropuerto Km 2.5, Tapachula, Chiapas, Mexico, CP 30700 AP 36.

Olfactometry using adults of the tenebrionide beetle *Epitragus salaei* Champion, a potential pest in the mango crop, showed an attraction effect to hexane extract from mango inflorescence *Manguifera indica* L. Volatiles from mango inflorescence were also sampled by air entrainment in the field, fourteen compounds were identified by GC-MS from extract and entrainment sample, 3-carene was found as the main component in both samples. Many of the identified compounds attracted more *Epitragus salaei* than the control but 3-carene showed the highest response.

**New non-polar oviposition stimulants for the diamondback moth.** **P-28**

Patrick R. Hughes, J. Alan A. Renwick and Kimberly D. Lopez

Boyce Thompson Institute for Plant Research at Cornell University, Tower Road, Ithaca, NY 14853, USA.

The diamondback moth (DBM), *Plutella xylostella* L. (Lepidoptera: Plutellidae) is an important pest of cruciferous crop plants throughout the world, and an understanding of the chemical stimuli mediating host selection by adult moths might provide new alternatives for controlling the insect. Glucosinolates have been thought to be the primary oviposition stimulants for the DBM on its various host plants. However, we recently found non-polar constituents of cabbage foliage that are very potent stimulants for DBM. Two groups of active compounds have been isolated by silica gel chromatography, and normal phase HPLC is being employed to further purify the stimulants. Extraction of foliage from other hosts and related plant species has shown that these or similar compounds are widely distributed.

**Influence of acylsugars on the predominance of *Solanum sisymbrifolium* lamark in prairies.** **P-29**

M.V. Cesio, C. Barra, H. Heinzen and P. Moyna

C‡tedra de Farmacognosia . Facultad de Qu'mica, Universidad de la Repœblica . Avenida General Flores 2124 Montevideo UR- 11800, Uruguay.

*Solanum sisymbrifolium* (Revientacaballos) is an around the world spread weed whose biology is not fully understood. How periods of seed dormancy and germination proceed, has not been explained, but once the plant is established it acts as an aggressive weed which quickly develops covering soil rich fields. Toxic and aggressive to cattle, is a major concern for Uruguayan cattle breeders. Field infestation of *S.sisymbrifolium* forms dense and healthy shrub arrays in relatively short periods. Grass population is diminished, insects and fungi are scarce on and around the plants. Searching for the reasons of this behaviour, leaves Leachates were investigated for allelopathic activity. They showed inhibition of the germination (up to 30%) and diminished development up to 50% of wheat seeds in a concentration dependent effect. Chemical analysis and bio-assay guided fractionation allowed the identification and characterisation the bioactive fraction of acyl sugars. This fraction was composed mainly of esters of glucose and methyl butyric and short chain fatty acids as revealed after GC and GC-MS analysis. On the other hand, glucose esters from *S. Sisymbrifolium* showed antifungal properties against common fungi as *A.niger, A. flavus* and *P.crysogenum.* These findings are discussed in terms of the field observations gathered and their relevance on the general defensive system of the plant.

**Acknowledgments**
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**The potential of Thiarubrine C as a nematicidal agent against plant parasitic nematodes.** **P-30**

Susanna Sˆnchez de Viala[1], Bill B. Brodie[2], Eloy Rodriguez[3], and Donna M. Gibson[2]

[1]Plant Pathology Dept., Cornell University, Ithaca, NY 14853, USA.
[2]ARS, USDA, U.S. Plant, Soil, and Nutrition Lab, Cornell University, Ithaca, NY 14853, USA.
[3]L. H. Hortorium and Plant Biology Dept., Cornell University, Ithaca, NY 14853, USA.

Thiarubrine C, a polyacetylenic 1,2-dithiin isolated from the roots of *Rudbeckia hirta* (Asteraceae), exhibited strong nematicidal activity in *in vitro* and growth chamber assays. Thiarubrine C was toxic, in the absence of light, to the plant parasitic nematodes *Meloidogyne incognita* and *Pratylenchus penetrans* at $LC_{50}$s of 12.4 ppm and 23.5 ppm, respectively. A minimum exposure time between 12 and 24 hours was the critical period for nematode mortality due to thiarubrine C. Although thiarubrine C was not totally dependent on light for toxicity, activity was enhanced in the presence of light, especially with *Teratorhabditis dentifera*, a microbivorous nematode. Upon exposure of *M. incognita* juveniles to 20 ppm thiarubrine C for 1 hour, infectivity of tomato plants was greatly reduced compared to untreated checks. Thiarubrine C was also effective in reducing plant infection when mixed with soil 24 hours prior to or at planting, unlike other related compounds such as ¶-terthienyl.

**Bird repellents: how agents interact in mixtures.** **P-31**

Larry Clark

U.S. Department of Agriculture, National Wildlife Research Center, 1716 Heath Parkway, Fort Collins, CO 80524

Numerous studies characterize the concentration-behavioral response for odorants, tastants, and irritants. However, to achieve ecological validity, interaction of agents in mixture must be considered. Equiresponse and equimolar molar models of interactions have been proposed and methods for testing whether agents in mixture interact independently have been evaluated. Yet these averaging models cannot a priori predict whether agents will interact antagonistically, independently or synergistically. I studied the bird repellent properties of several structurally similar and well described trigeminally mediated avian irritants, singly and in mixture. Compounds within a chemical class, where the electron withdrawing groups were similar, interacted independently to produce their repellent effects, e.g. 2-amino methyl benzoate v methyl-2-methoxy benzoate, and o-amino acetophenone v 2-methoxy acetophenone. The response to mixtures drawn from compounds of dissimilar chemical class, e.g., 2-amino methyl benzoate v o-amino acetophenone, interacted antagonistically, suggesting mediation by a different mechanism within the trigeminally mediated sensory modality. These observations underscore the our previous findings for the importance of the molecular properties of the carbonyl group for aromatic bird repellents, and suggests the possible existence of multiple receptor mechanisms for avian trigeminal repellents. These data also underscore the importance of attending to interactions of agents in mixtures when designing repellents as tools for the management of wildlife and resolution of conflicts between humans and wildlife.

## *Hypsipyla*/Meliaceae interactions: a chemical perspective on the attack of *Hypsipyla grandella* on *Cedrela odorata.* P-32

Ian Castro-Gamboa*, Jo‹o Oiano Neto, JosŽ Realino de Paula, Maria F‡tima das Gra as Fernandes da Silva*, Jo‹o Batista Fernandes, and Paulo Cezar Vieira

Departamento de Qu'mica, Universidade Federal de S‹o Carlos (UFSCar), Caixa Postal 676, S‹o Carlos - S‹o Paulo, Brazil.

Efforts to establish large scale homogenous plantations of valuable members from Latin American Meliaceae have almost invariably failed due to larval attacks by the shoot borer *Hypsipyla* (Lepidoptera: *Pyralidae*). Main damage is caused by the larvae, which destroys the succulent terminal shoots by boring into the tip and tunneling inside the juvenile stems of saplings and seedlings. Re-sprouting, followed by repeated attacks of the insect, generally results in the development of numerous side branches, making the tree unsuitable for timber production.

However, *Toona ciliata* the Australian red cedar, introduced in Latin America, shows excellent growth and absence of attack by *H. grandella*, in contrast to the native *Cedrela odorata*. The latter has been grafted to stems of *T. ciliata* and the resistance has been translocated from *Toona*'s stock to *Cedrela*'s graft. The chemistry of these grafts have not been previously studied. Thus, we examined all the organs of *T. ciliata*, seedlings and the stem of *C. odorata* graft, in order to determine if secondary metabolites present in the former could be translocated to the latter. The stem of the grafted tree produces a considerable number of limonoids which are common in *C. odorata*. On the other hand, the occurrence of cedrelone, limonoid typical of *Toona* in the seedlings from the grafted tree, is of good value to clarify the basis of the induced resistance, since aside from the C-*seco* limonoids, the most active compounds appear to be the intact *apo*-euphol limonoids with a 14,15-epoxide and either a 3-oxo-1-ene A ring as in the former. The cycloartanes and catechin could also been translocated from *Toona*'s stock to *Cedrela*'s graft.

*Authors to whom correspondence should be addressed

**Flavonoids of *Calamintha ashei.*** **P-33**

Brian Burtch and Jeffrey Weidenhamer

Department of Chemistry, Ashland University, Ashland, Ohio 44805, USA.

*Calamintha ashei* is a perennial shrub which occurs in the Florida sand pine scrub community along Florida's Lake Wales ridge. *Calamintha* and other scrub perennials have been of interest because several scrub species appear to produce allelopathic chemicals that inhibit the germination and growth of grasses from the neighboring Florida sandhills. As part of an investigation of the chemistry of trichomes on the *Calamintha* leaves, an extract of surface compounds was prepared by briefly dipping leaves in dichloromethane, followed by vacuum liquid chromatography. Several grams of flavonoid-rich fractions were obtained from the extraction of approximately 1kg of Calamintha leaves. One compound has been identified by NMR as 5-hydroxy-6,7,8,3',4'-pentamethoxyflavone, which has previously been reported from this plant. Isolation and characterization of several other flavonoids is proceeding. Once the compounds are characterized, our objective is to develop an HPLC method to investigate the distribution of these compounds on and within the leaves.

**Phytochemistry and biological activity of *Echinacea* spp.** **P-34**

Shannon E. Binns. B. Purgina, B.R. Baum, J.T. Arnason, D.V.C. Awang and J. Livesay

University of Ottawa, Department of Biology, 30 Marie Curie Street, P.O. Box 450 stn A, Ottawa, ON, Canada K1N 6N5.

Native populations of *Echinacea* spp. (Compositae) have been assessed for alkylkamide, polyacetylene and phenolic content by HPLC as part of a taxonomic study of the genus. Hexane fractions of ethanolic extracts from roots and flower heads contained polyacetylenes which were found to have phototoxic activity against clinical fungal isolates. Antifungal activity in the absence of photo-activation was attributed to isobutylamide content. Chemical variation was observed between populations and ages classes within populations. Degradation within ethanolic extracts of *Echinacea* spp. was observed over time. Also, the lipophilic and hydrophilic constituents found in ethanolic extracts of young seedlings were markedly different from those in older plants of the same population.

**Greenhouse study of the effect of nitrogen availability on dillapiol production by dill, *Anethum graveolens*: a confirmation of the C/N Balance hypothesis.** **P-35**

Anne-Sophie Belzile and John Thor Arnason

Ottawa-Carleton Institute of Biology, 30 Marie Curie, Ottawa, Ontario, Canada, K1N 6N5.

The lignan dillapiol is an efficient naturally occurring insecticide synergist and a potential anti-cancer agent produced by plants from different families such as the Umbellifereae and the Piperaceae. Eventual commercialization of this botanical would require optimization of the production. The present study, set in a chemical ecology perspective, proposed to look at factors that could affect this production. A greenhouse study was conducted in order to assess the effect of Nitrogen treatments on the dillapiol content of fresh tissues of dill, *Anethum graveolens* (Umbellifereae). It was found that dillapiol concentration, which is higher in the aerial parts of the plant, increases significantly per gram of fresh weight when Nitrogen input in the medium is lower. These findings agree with the Carbon/Nutrient balance hypothesis.

**Why are the larvae of the Gall Midge *Schizomyia impatientis* O.S. (Cecidiomyiidae) a bright orange color?** **P-36**

Constance Nozzolillo

Department of Biology, University of Ottawa, Ottawa, Canada K1H 5G6.

Flower buds of touch-me-not (*Impatiens capensis* Meerb., Balsaminaceae) in which the gall midge *Schizomyia impatientis* deposits its eggs develop into globular galls instead of the characteristic spotted orange flower. The larvae that emerge from the eggs are initially colorless but become a bright orange color as they mature. No one has ever identified the chemical nature of the pigmentation which is exactly the same color as that of the normally developed flower petals. Is it possible that the insect coopts the carotenoids that the plant would deposit in the petals for its own unknown purpose? Galls were picked off touch-me-not plants growing in the author's garden. They were immediately opened up and the larvae were removed and stored in the freezer. When two to three dozen larvae had been collected, attempts were made to remove the pigmentation into solution following maceration in both polar and non-polar solvents. In contrast to the ready solution of the carotenoids of the flower into organic solvents, the orange color remained firmly associated with the insect debris. Covering the insects with

mineral oil and heating in a microwave oven finally resulted in an orange-colored solution that had a characteristic carotenoid spectrum. Comparison of these carotenoids to those of the flower is in progress. The question of why the insect deposits this orange color in its body remains unanswered.

**Is methyl gallate a natural constituent of maple (Genus *Acer*) leaves?** **P-37**

Mamdouh M. Abou-Zaid, Constance Nozzolillo[1] and Domenic A. Lombardo.

Natural Resources Canada, Canadian Forest Service, Great Lakes Forestry Centre, 1219 Queen St. East, Sault Ste. Marie, Ontario, Canada P6A 5M7.
[1]Ottawa-Carleton Institute of Biology, University of Ottawa, Ottawa, Ontario, Canada K1N 6N5.

Methyl gallate has been reported to be present in extracts of silver maple leaves (*Acer saccharinum L.*) (Bailey et al., J. Nat. Prod. 49:1149, 1986). Given the current interest in the biological activity of methyl gallate (e.g. Mendez and Mato, Phytochemistry 44: 41, 1997), it is important to establish whether methyl gallate is a natural constituent of the leaves and not an artifact of extraction in methanol. In the present study, leaves of three *Acer* species (family Aceraceae) indigenous to North America, *A. rubrum* L. (red maple), *A. saccharum* Marsh. (sugar maple) and silver maple, were extracted either with methanol or ethanol. The extracts were then analyzed by HPLC. Methyl gallate was present in methanolic extracts but was also found in ethanolic extracts. In addition, methyl gallate was present in extracts of red maple leaves made with diethyl ether, methylene chloride, acetonitrile, or ethyl acetate. The implication that this compound is found in the leaves of red maple in particular is under investigation for its possible role in the innate resistance of this species to feeding by larvae of forest tent caterpillar.

**Sequestered aristolochic acids as deterrents to parasitism.** **P-38**

Karen R. Sime

Ecology & Systematics and Department of Entomology Corson Hall Cornell University, Ithaca, NY 14853, USA.

The parasitoid *Trogus pennator* (Hymenoptera: Ichneumonidae) attacks the larvae of swallowtail butterflies (Lepidoptera: Papilionidae) in the tribes Papilionini and Graphiini. It does not, however, attack those in the tribe Troidini, despite geographical and ecological overlap.

Troidine larvae sequester aristolochic acids; in this research I investigated possible roles of these compounds in protecting the larvae from attack by *T. pennator*. I found that 1) rejection of troidine larvae occurs only after antennal contact with the larval cuticle; 2) on otherwise acceptable host larvae, ethanolic washes of troidine larvae deter parasitism; 3) aristolochic acids are at least partly responsible for this effect.

**Hygienic Grooming Behavior Induced by Parasitic *Varroa* Mites in the Japanese Honey, *Apic cerana japonica* RAD.** **P-39**

Hiromi Sasagawa[1], Shigeru Matsuyama[2], Walter S. Leal[3] and Christine Y. S. Peng[4]

[1]PRESTO "Intelligence and Synthesis", Japan Science and Technology Corporation (JST): c/o National Institute of Sericultural and Entomological Sciences (NISES), Tsukuba, Ibaraki, 305, Japan.
[2]Inst.of Applied Biochem., Univ. of Tsukuba, Tsukuba, Ibaraki, 305, Japan.
[3]National Institute of Sericultural and Entomological Sciences (NISES), Tsukuba, Ibaraki, 305, Japan.
[4]Department of Entomology, University of California, Davis, CA 95616, USA.

The Japanese honeybee (*Apis cerana japonica*: *Acj*) and the European honeybee (*Apis mellifera*:*Am*) have been sharing the same habitat in Japan since the introduction of the latter bee species at the turn of the century. *Varroa jacobsoni* Oud, originally an ectoparasitic mite of *A. cerana* species, has since infested *Am* colonies in the world. Heavy infestations of *Varroa* are commonly found in the *Am* colonies, hence the mite has become a major pest of the new host bee species. In contrast, the colonies of *Acj* have very few mites. We conducted experiments to compare the hygienic grooming behavior of *Acj* and *Am* in response to *Varroa* parasitism. We report here the identification a semiochemical compound, ethyl (*Z*)-9-octadecenoate (ethyl oleate:EO), from the cuticular extract of *Varroa*, using GC-MS and further established its biological activity in *Acj* and *Am* by GC-EAD and behavioral bioassays. We demonstrate that EO is the principle chemical cue used by the *Acj* worker bees to olfactorily detect the ectoparasite mites. It is highly potent in eliciting the entire behavioral sequence of a typical hygienic allo-grooming, which consists of searching, detecting, and removing the target object with the mandibles. *Am* worker bees showed no response to mite and mite extract. The biological function of EO in the bee-mite association is strictly serving as a chemical cue that triggers hygienic self- and allo-grooming behavior in the *Acj* against their ectoparasitic *Varroa* mites. This semiochemical role is most pronounced in the long evolutionary relationship between the mite and Japanese honey bee.

**Defensive egg endowment in *Utetheisa ornatrix*: mother provides a fixed fraction of her alkaloid load.** **P-40**

Carmen Rossini and Thomas Eisner

Cornell Institute for Research in Chemical Ecology, W 347 Mudd Hall, Cornell University, Ithaca, NY 14853, USA.

*Utetheisa ornatrix* sequester pyrrolizidine alkaloid (PA) from their diet (legumes of the genus *Crotolaria* ). Males transfer PA to the female at mating, thereby supplementing the female's intrinsic PA load. Females protect their eggs by endowing them with PA, using for the purpose both their intrinsic PA and that received from the male. We showed that the PA content of the eggs is variable, but a precise reflection of the female's PA load. The more PA the females have, the more they bestow upon the eggs. The relationship holds, irrespective of the relative magnitude of the female's intrinsic PA load and the quantity of PA she receives from the male. In provisioning the eggs, the female never entirely depletes her PA load.

**The Biochemical responses of cotton plants to insect herbivory and methyl jasmonate application.** **P-41**

Ursula S.R. Ršse, Jšrg Bohlmann, James H. Tumlinson and Jonathan Gershenzon

Max-Planck-Institut fŸr chemische …kologie, Tatzendpromenade 1A, 07745 Jena, Germany.

When attacked by herbivores, several plant species release volatile compounds that are attractive to predators and parasitic wasps. In cotton these herbivore-inducible compounds include acyclic terpenoids [e.g. (*E* )-b-ocimene, (*E* )-b-farnesene, (*E,E* )-a-farnesene, linalool, (*E* )-4,8-dimethyl-1,3,7-nonatriene, (*E,E* )-4,8,12-trimethyl-1,3,7,11-tridecatetraene], (*Z* )-3-hexenyl acetate, indole, isomeric hexenyl butyrates, and 2-methylbutyrates. Many of these compounds are not only released from the damaged parts of the plants, but also systemically from undamaged parts of caterpillar damaged plants including (*E* )-b-ocimene, (*E* )-b-farnesene, (*E,E* )-a-farnesene, linalool, (*E* )-4,8-dimethyl-1,3,7-nonatriene, (*E,E* )-4,8,12-trimethyl-1,3,7,11-tridecatetraene, and(*Z* )-3-hexenyl acetate. Several of these compounds are not induced by artificial damage alone, but require the presence of caterpillar regurgitate to be released from the cotton plants. These herbivore inducible volatiles are known to be synthesized de novo. Cotton plants that were fed upon by caterpillars or sprayed with 0.025% methyl jasmonate showed elevated levels of monoterpene-and sesquiterpene synthase

activities compared to untreated control plants. In assays with tritium-labeled geranyl diphosphate and tritium-labeled farnesyl diphosphate, a significant increase of enzyme activity in the cotton leaves could be demonstrated with herbivore-damage leading to higher levels of activity than methyl jasmonate.

**How chemical signals mediate foraging and recruitment in subterranean termites.** **P-42**

Judith Reinhard[1] and Manfred Kaib[2]

[1]FU Berlin, Angewandte Zoologie, Haderslebenerstr. 9, 12163 Berlin, Germany.
[2]UniversitŠt Bayreuth, Lehrstuhl Tierphysiologie, 95440 Bayreuth, Germany.

Foraging and recruitment in subterranean termites is investigated in the French species *Reticulitermes santonensis* De Feytaud (Rhinotermitidae). Few pioneer workers explore a new territory in every direction. For orientation they lay pheromone trails with their sternal gland by putting the abdomen in intervals briefly on the ground. These trails are followed slowly by further foraging workers. Following the course of the trails, termites build a regular net of runways. Wood is detected over distance by volatiles emanating from it. Then foragers appear more frequently and their runs are directed towards the odour source. Following contact with the wood, successful workers lay a recruitment trail when returning to the nest by constantly dragging the abdomen on the ground. This recruitment trail must differ from the foraging trails in quantity and/or quality of its pheromonal composition, for it is highly attractive: it is followed immediately and in high speed by many workers. Even soldiers, which were not seen before, appear on the recruitment trail. Their weaker reaction is probably due to their higher perception threshold for the trail pheromone, which makes them follow only strong or very attractive trails. Finally, following the course of the recruitment trail, termites build a direct runway leading straight to the wood.

**Behavioral & fitness consequences of pyrrolizidine alkaloids in *utetheisa* caterpillars to their *polistes* wasp predators.** **P-43**

Linda S. Rayor

Department of Entomology, Cornell University, Ithaca, NY 14853, USA.

Tritrophic interaction theory suggests that herbivores capable of feeding on host-plants with unpalatable or toxic allelochemicals should reduce their predation risk. My research has focused on whether generalist, invertebrate predators, such as *Polistes dominulus* and *P.fuscatus* wasps (Hymenoptera: Vespidae), are deterred by *Utetheisa ornatrix* (Lepidoptera:Arctiidae) caterpillars which have fed on *Crotolaria* host plants rich in pyrrolizidinealkaloids (PA), and the consequences to the predator's fitness by consuming chemically defended prey. In behavioral choice tests, live *Utetheisa* that had been reared on artificial diets with or without 10% *Crotolaria* seeds, were presented to captive free-flying wasps. Predation by two wasp species was significantly reduced on caterpillars with PA. Response to PA changed with wasp experience. Additionally, wasp colonies reared on *Utetheisa* caterpillars with PA suffered reduced fecundity, produced fewer reproductive wasps, and had a 10% greater probability of chitinous deformities, than wasps reared on caterpillars without PA. Generalist predators, such as predatory wasps, experience serious consequences of consuming caterpillars which have fed on host-plants containing significant quantities of PA.

**Interactions by allelochemicals between tentiform leafminers (Lepidoptera, Gracillariidae) and other moth species.** **P-44**

Raimondas Mozuraitis[1,2], Vincas Buda[1] and Anna-Karin Borg-Karlson[2]

[1]Institute of Ecology, Akademijos 2, Vilnius 2600, Lithuania.
[2]Royal Institute of Technology, Dept. Chemistry, Organic Chemistry, SE-100 44 Stockholm, Sweden.

Interaction by allelochemicals can occur (i) when individuals of one species perceive and react to compounds emitted by organisms of another species, (ii) when the species are dispersed in distances allowing communication, and (iii) when the periods of communication activity of the species overlap. Almost all of the attraction inhibitors affecting the species *Pyllonorycter ulmifoliella*, *P. sorbi*, *P. mespilella* and *P. sylvella*, which we have found, have been known earlier as components of sex pheromones or attractants in lepidopteran species belonging to other families. On the basis of available data on moth sex pheromones, sex attractants and their inhibitors, we present schemes of probable interactions by means of allelochemicals acting between *P. ulmifoliella* and 363 other moth species, *P. sorbi* and 206, *P. mespilella* and 4, and *P. sylvella* and 5 other moth species . As the host-plants of these tentiform leafminer species are common and widespread in the temperate climatic zone, the phyllonoryctids are found at the distances short enough to allow interactions by allelochemicals with other moth species. It is known, that females of most moth species, for which pheromone emission periods were determined, signal during the dark hours of the day. Our investigation of the daily signalling activity of tentiform leafminers showed that females of the species investigated emitted sex pheromones during the light period of the day, early in the morning. We hypothesize, that the uncommon timing of the pheromone communication in phyllonoryctids may have been caused by the sensitivity of the males to inhibitors, which females of many other species emit into the environment at other times of the day. The communication during the period free of inhibitors should be adaptive for the species.

**Chemical comparison of waste-based defenses of two *Solanum dulcamara* feeders: *Lema trilinea* (criocerinae) fecal shield vs. *Plagiometriona clavata* (hispinae) parasol.** **P-45**

Timothy C. Morton[1] and Fredric V. Vencl[2]

[1]Department of Ecology and Evolution, University of Chicago, Chicago IL, 60637, USA.
[2]Department of Neurobiology and Behavior, SUNY at Stony Brook, Stony Brook, NY 11794, USA.

This study compares the defensive chemistry of *Lema trilinea* (Chrysomelidae subfamily Criocerinae) fecal shields with *Plagiometriona clavata* (subfamily Hispinae) parasols. Our previous work with fecal shields of other species showed them to be dietary-based chemical defenses and, though parasol defenses differ morphologically and are found in a different beetle subfamily, we wondered if Hispine beetle larvae might use host-plant chemistry similarly. To facilitate the comparison, both insects were reared at the same time on cuttings of the same host-plant. Extracts of *L. trilinea* and *P. clavata* feces were compared with each other and to plant extracts using TLC, GC-MS, and FAB-MS. Bioassays using the ant, *Formica subserecea*, showed that both *L. trilinea* and *P. clavata* defenses deterred this generalist predator. Active components of defense were then identified using bioassay-guided fractionation. Both beetles incorporate fatty acids and derivatives of host-plant saponins and steroidal glycosides into their feces. Both beetles performed similar metabolic transformations to saponins and steroidal glycosides before incorporating them into their defenses. It would appear that the transformations are not inadvertent and might serve to 1) prevent intoxication by enhancing compartmentalization, 2) supply energy, and 3) enhance and/or activate anti-predator activity. This is the first demonstration that the parasol-type defense of Hispinae larvae can be a chemical defense. Feeding on noxious host-plants, when coupled with retention of feces, is likely to represent the first and simplest route to the evolutionary sequestration of host-plant chemistry.

**Specifity of kairomones for two egg parasitoids of chrysomelidae.** **P-46**

Torsten Meiners and Monika Hilker

Freie UniversitŠt Berlin, Institut fŸr Zoologie, Haderslebenerstr. 9, D-12163 Berlin, Germany.

Host finding behaviour of eulophid egg parasitoids of the polyphagous tansy leaf beetle *Galeruca tanaceti* and the elm leaf beetle *Xanthogaleruca luteola* was investigated. Within their host habitats, both the wasp *Oomyzus galerucivorus*, which parasitizes *Galeruca* spp., and *O. gallerucae*, which attacks only *X. luteola*, use kairomones from faeces of their hosts for host finding. The specifity of these kairomones were studied in dependence of the feeding host stage, the host plant, and the insect species feeding on a host plant. (A) Kairomones were not specific for a developmental stage of the feeding host: faeces from larvae and adults elicited host finding behaviour. (B) Kairomones from faeces were not specific for the plant which had been consumed by the host: faeces from *G. tanaceti* which fed upon yarrow or cabbage were recognized by *O. galerucivorus*. (C) However, only faeces from host species contained kairomones: faeces from a lepidopteran larva which fed upon elm did not elicit host finding behaviour in *O. gallerucae*; faeces from the mustard leaf beetle which fed upon cabbage had no kairomonal effect on *O. galerucivorus*. Thus, the two parasitoid species studied showed no significant differences in specifity of kairomones from host faeces. The similarity of host finding behaviour in *O. galerucivorus* and *O. gallerucae* contrasts with the differences in habitat location of both wasps (1,2).

**Literature:**
1 Meiners et al. (1997): J. Insect Behav. 10, 523;
2 Meiners & Hilker (1997) Oecologia 112, 87

**Piscicidal properties of piperovatine from *Piper piscatorum* (piperaceae).** **P-47**

Marcus A. McFerren and Eloy Rodriguez

Cornell University, LH Bailey Hortorium, Ithaca, NY 14853, USA.

Extraction of the roots of Amazonian medicinal plant, *Piper piscatorum* Trelease and Yuncker with MeOH and subsequent bioassay guided fractionation for piscicidal activity using the guppy, Girardina guppii, yielded 2 isobutyl amides. Spectroscopic methods elucidated N-isobutyl-6-(p-methoxyphenyl) 2E, 4E-hexadieneamide (piperovatine) as the active component and N-isobutyl-(E)-7-(3,4-methylenedioxyphenyl)hept-2-enamide (pipercallosidine) as an

inactive component. The former displayed an LC50 of 115 ng/ml in toxicity tests and proved to be the constituent responsible for the dual ethnobotanical uses of this plant: that of fish stupefacient (barbasco) and oral local anesthetic.

**New ophiobolins from cochliobolus heterostrophus.** **P-48**

Stuart B. Krasnoff, Xiaoyu Shen, Shunwen Lu, B. Gillian Turgeon, Olen C. Yoder, Donna M. Gibson and Mark T. Hamann

USDA, ARS, PPRU, Tower Road, Ithaca, NY 14853

*Cochliobolus heterostrophus* (the Southern corn leaf blight fungus), produces ophiobolins, sesterterpenes that exhibit a range of antibiotic activities. We analyzed ophiobolin production by a strain of *C. heterostrophus* race 0. Preparative HPLC of methylene chloride extracts of liquid cultures yielded 6 previously identified ophiobolins, ophiobolin A, 3-anyhydro-ophiobolin A, 6-epi-ophiobolin A, 6-epianhydroophiobolin A, ophiobolin B, and ophiobolin I as well as two previously unreported analogues, 6-epiophiobolin B, and 3-anhydroophiobolin B. Tentative structures for the new analogues were proposed based on data from NMR experiments. In cytotoxicity assays $IC_{50}$'s for ophiobolin A were estimated at 50 ng/ml for cancer cell-lines A-549, HT-29, and MEL-20, and 25 ng/ml for cell-line P-388. This potent anti-tumor activity suggests that ophiobolin chemistry could be optimized by mutant strains, bioconversions, or semi-synthesis to yield compounds that may eventually prove clinically useful.

**Some alkaloids common to ants and frogs: decahydroquinolines and a novel quinolizidine**. **P-49**

Tappey H. Jones[1], Jeffrey S.T. Gorman[1], Roy R. Snelling[2], H. Martin Garraffo[3], T.F. Spande[3], John W. Daly[3] and Jacques H.C. Delabie[4]

[1]Department of Chemistry, Virginia Military Institute, Lexington, VA 24450, USA.
[2]Los Angeles County Museum of Natural History, 900 Exposition Blvd., Los Angeles, CA 90007, USA.
[3]Laboratory of Bioorganic Chemistry, NIDDK/NIH, Bethesda, MD 20892, USA.
[4]Divisao de Zoologia, Centro de Pesquesa do Cacau, CEPLAC, Caixa Postal 7, 15600 Itabuna, Bahia, Brazil.

The major component of the extract from a thief ant, *Solenopsis (Diplorhoptrum)* species, collected in Brazil was shown to be (6Z, 10E)-4-methyl-6-propylquinolizidine, a new class of ant venom alkaloids. The structure of this alkaloid was established by comparison of the FTIR and mass spectra of the natural compound with the products of stereoselective syntheses. Two minor decahydroquinolines (DHQs) were also detected in this extract. One of these was identical to a frog-skin alkaloid, cis-**195A** (cis-5-methyl-2-propyldecahydroquinoline), and the other, designated **195J**, is tentatively cis-2-methyl-5-propyldecahydroquinoline. This same triad of alkaloids, with the quinolizidine predominating has been found in a number of frog-skin extracts. The common occurrence of these compounds in ants and frogs supports ants being a likely dietary source for sequestered frog-skin alkaloids and brings to six the alkaloid classes common to ants and frogs.

**Induction of odor emissions in maize plants by different larval instars of *spodoptera littoralis* lepidoptera: noctuidae).** **P-50**

Sandrine GouinguenŽ, Thomas Degen and Ted Turlings.

Laboratory of Animal Ecology and Entomology, Institute of Zoology, University of Neuch‰tel CH-2007, Switzerland

Maize plants emit a specific blend of volatiles after they are attacked by caterpillars. The resulting odor is highly attractive to parasitic wasps. A factor in the caterpillar oral secretions is responsible for the induction of the odor emissions. We tested the effect of larval instar of *Spodoptera littoralis* on the intensity and quality of odor emissions. Maize plants were treated with regurgitant from second, third and fifth instar caterpillars. The emission of volatiles was not different for the different instars, neither qualitatively nor quantitatively. However, because of the manner in which they feed, differences may occur when the different instars actually feed on the leaves. Young instar larvae damage the leaves in a very different manner than do older larvae. We tested our hypothesis that differences in induction by different instars is merely due to this physical difference in how leaves are damaged. We used live plants damaged with caterpillars of respective instars feeding directly on them. The volatile emission appears to be dependent on the number of larvae feeding on the plant, with differences according to the instar. We mimicked the manner of feeding of each larval instar to test the importance of the physical damages of the leaves on the odor emission. It is discussed how larval instar may affect variability in odor emissions and if this would allow natural enemies to distinguish among instars.

**Sexually transmitted chemical defense in a moth.** **P-51**

AndrŽs Gonz‡lez, Carmen Rossini, and Thomas Eisner

Cornell Institute for Research in Chemical Ecology, W347 Mudd Hall, Cornell University, Ithaca, NY 14853, USA.

The arctiid moth *Utetheisa ornatrix* is protected against predation by pyrrolizidine alkaloids (PA) that it sequesters as a larva from its food plant. In courtship, the female *Utetheisa* favors males of high PA content. Males transmit PA to the female with the sperm package, in quantity proportional to the male's PA load. Earlier work had shown that the female bestows part of the PA gift upon the eggs, which are protected against predation as a result. We have now demonstrated that the female herself derives protection from the gift. Females deficient in PA are vulnerable to spider predation (*Nephila clavipes*; *Lycosa ceratiola*). If mated with a PA rich male, they are invulnerable. The effect takes hold immediately: females are unacceptable to spiders as soon as they uncouple from the male. Chemical data showed that the female allocates the received PA promptly to all body parts, and that even as she ages and loses PA to the eggs she retains sufficient quantity of the chemical for her own protection. To our knowledge this is the first demonstration of a female insect being rendered invulnerable by receipt of a sexually transmitted chemical.

**Cellulysin is a high molecular elicitor of plant volatile biosynthesis.** **P-52**

Andreas Jux, Thomas Koch, Jšrn Piel, and W. Boland

Max-Planck-Institut of Chemical Ecology, Tatzendpromenade 1a, 07745 Jena, Germany.

Treatment of higher plants (*Nicotiana plumbaginifolia*, *Phaseolus lunatus*, and *Zea mays*) with cellulysin, a crude cellulase of the plant parasitic fungus *Trichoderma viride*, induces the biosynthesis and the emission of volatile compounds from the plant leaves. The pattern of emitted substances closely resembles the pattern of volatiles released after treatment of the same plants with jasmonic acid. This observation matches with the observation that ca. 30 minutes after treatment of a freshly detached plantlet of the Lima bean (*P. lunatus*) with cellulysin through the petiole (aq. soln. at 50 µg $ml^{-1}$), an 8-fold increase of the endogenous JA-level is observed. The peak maximum decreases within two hours to the resting level of an undamaged plant. The JA-increase, together with inhibitor experiments proved the octadecanoid cascade as the only relevant signalling pathway for the cellulysin-induced volatile production. Administration experiments of $d_5$-mevalolactone and $d_2$-deoxy-D-xylulose clearly showed that the biosynthesis of the induced (cellulysin- or jasmonate-treatment) volatiles proceeds via the novel glycerinaldehyde-3-phosphat/pyruvat pathway (*Rohmer-Sahm*-pathway) and not via the classic acetate/mevalonate route. The same pathway is also operative for the biosynthesis of constitutively present volatiles. In the case of sesquiterpenoids both the *Rohmer-Sahm* as well as the mevalonate pathway contribute to the biosynthesis of this class of terpenoids. This finding may be of importance for the plant defense by adding a greater plasticity to the defense response in case of injection of inhibitors from the herbivore salivary secretion or from enzyme cocktails of attacking pathogens into the leaves.

**Soilborne bacteria and phenolic acid bioassays.** **P-53**

Udo Blum[1] and Steven R. Shafer[2]

[1]Department of Botany, Box 7612, North Carolina State University, Raleigh, NC 27695, USA.
[2]USDA-APHIS, Center for Plant Health Science & Technology, Box 7559, North Carolina State University, Raleigh, NC 27695, USA.

The role of soilborne bacteria in allelopathy involving phenolic acids was studied. Model soil or plant-soil systems in the laboratory were treated with vanillic, p-hydroxybenzoic, ferulic, or p-coumaric acid. Changes in numbers of bacteria in the soil and rhizosphere were determined by cultural methods. Phenolic acids remaining in soil after periods of utilization by bacteria were extracted with EDTA or citrate and quantified by HPLC. Phytotoxicity of phenolic acids was quantified by inhibition of cucumber seedling growth. Numbers of bacteria that could utilize phenolic acids as a carbon source were stimulated by phenolic acids in the presence of adequate mineral nutrition. Chronic (0.025 or 0.1 µmol/g soil applied 4 times between days 7 and 13) phenolic acid treatments had no effect on numbers of bacteria in the rhizosphere on day 17 and did not alter rhizosphere bacterial responses to a subsequent acute (0.125 to 1 µmol/g soil, applied once on day 15) treatment. Acute phenolic acid treatments increased the numbers of bacteria in the rhizosphere (day 17). Utilization of ferulic and p-coumaric acid (cinnamic acids) by bacteria resulted in the production of vanillic and p-hydroxybenzoic acids (benzoic acids), respectively. Cinnamic acids were more phytotoxic than their corresponding benzoic acids. Proper interpretation of phenolic acid dose - plant response bioassays requires characterization of both phytoxicity and soil microbial populations, because responses of microbes to phenolic acids differ in the bulk soil and the rhizosphere, and differential utilization of phenolic acids and their breakdown products by microbes can substantially influence the phytotoxicity of phenolic acids.

**Aromatic Polyketide Pigment Biosynthesis in the Chestnut Blight Fungus *Cryphonectria parasitica*** **P-54**

Alice C. L. Churchill[1], Gregory J. Podgorski[2], and Neal K. Van Alfen[3]

[1]Molecular Mycology Center, Boyce Thompson Institute for Plant Research, Ithaca, NY, 14853-1801
[2]Molecular Biology Program, Department of Biology, Utah State University, Logan, UT, 84322-1503
[3]Department of Plant Pathology and Microbiology, Texas A & M University, College Station, TX, 77843-2132.

The orange and yellow pigments of *C. parasitica* are monomeric and dimeric anthraquinones, which are synthesized by a polyketide pathway, and have been identified as emodin, chrysophanol, skyrin, oxyskyrin, and rugulosin. *C. parasitica* is a model system for studying the biosynthesis of aromatic polyketides because it is genetically tractable, i.e., DNA-mediated transformation, molecular genetic manipulations, and *in vitro* sexual crosses are routine. Chemical and structural analyses of the pigments suggest they are synthesized by a polyketide synthase, which is hypothesized to be part of a gene cluster required for pigment biosynthesis. We have isolated a cosmid clone with a 32 kb insert that complements a pigmentation defect in an albino mutant of the fungus. Genetic characterization of the cosmid clone is in progress. Conditions for characterization of pigments by TLC and reverse-phase HPLC with PDA have been determined, and analyses of wild-type, mutant, and complemented transformants will be reported. It is notable that polyketide pigment production is suppressed in strains of the fungus that are infected with transmissible fungal viruses of the genus *Hypovirus*. In particular, the pigment profile of *Hypovirus*-infected strains appears to be dependent on the genetic makeup of the viral strain present in the fungus. The mechanism by which Hypoviruses regulate secondary metabolite production in *C. parasitica* is unknown but hypothesized to be through perturbation of signal transduction pathways.

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