10th ANNUAL MEETING

International Society of Chemical Ecology

Tampa/Clearwater Florida

July 31-August 4, 1993

I S C E

Promoting The Understanding Of Interactions Between Organisms and Their Environment That Are Mediated by Naturally Occurring Chemicals.

Symposia Organizers

William Fenical
Jean Langenheim
Wittko Francke
Lincoln Brower
David Carlson
John Romeo

Local Arrangements

John Romeo

Acknowledgements

The ISCE is grateful to the following institutions and foundations for support of this meeting.

University of South Florida College of Arts and Sciences
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PROGRAM SCHEDULE

Saturday, July 31

3:00 - 5:00 pm	Executive Meeting - Conference Room 254
4:00 - 5:00 pm	Registration - Lobby
6:00 - 8:00 pm	Welcome Reception - Poolside

Sunday, August 1

8:30 am	Registration (late) - Lobby
8:45 - 8:50 am	Welcome - Beach Gulf Room John Romeo, Secretary ISCE
8:50 - 9:00 am	Rollin Richmond, Dean College of Arts and Sciences University of South Florida
9:00 - 12:20 pm	Symposium I Marine Chemical Ecology Beach Gulf Room
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12:20 - 2:00 pm	Break For Lunch
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2:00 - 3:30 pm	Session I Contributed Papers - Marine Subjects Palm Room
2:00 - 4:45 pm	Session II Contributed Papers - General Subjects Beach Gulf Room
7:30 - 10:00 pm	Posters Bay Room

Monday, August 2

9:00 - 12:10 Noon Symposium II Chemical Ecology of Terpenoids
Beach Gulf Room

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12:10 - 2:00 pm Break For Lunch

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2:00 - 4:45 pm Session III Contributed Papers - Terpenoids
Island No. I Room

2:00 - 5:30 pm Session IV Contributed Papers - Pheromones
Beach Gulf Room

5:30 - 6:30 pm ISCE Business Meeting
Beach Gulf Room

Tuesday, August 3

9:00 - 12:00 Noon Symposium III Physical Stress & Chemical Ecology
Beach Gulf Room

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12:00 - 2:00 pm Break For Lunch

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2:00 - 5:30 pm Session V Mini-Symposium - Insect Pheromones & Hormones
Beach Gulf Room

7:00 - 8:00 pm Social - Beach Gulf Room

8:00 - 10:00 pm Banquet - Beach Gulf Room

Wednesday, August 4

8:30 - 12:30 pm Symposium IV Tropical Chemical Ecology
Beach Gulf Room

SYMPOSIA

10th ANNUAL MEETING
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Symposium I (Sunday - August 1)

Marine Chemical Ecology

Beach Gulf Room - William Fenical Presiding

9:00 - 9:40 am	***Symposium 1*** Joseph Pawlik, University of North Carolina-Wilmington, *Chemical Defenses of Caribbean Sponges.*
9:40 - 10:20 am	***Symposium 2*** Mark Hay, University of North Carolina-Chapel Hill *Why Small Herbivores Eat Toxic Plants: Marine-Terrestrial Contrasts.*
10:20 - 10:40 am	**COFFEE BREAK**
10:40 - 11:00 am	***Symposium 3*** Steve Pennings, University of Guam, *Mediation of Algal-Sea Hare-Predator Interactions by Algal Secondary Metabolites.*
11:00 - 11:20 am	***Symposium 4*** Jim McClintock, University of Alabama-Birmingham *Aspects of the Chemical Ecology of Shallow-Water Marine Invertebrates in McMurdo Sound, Antarctica*
11:20 - 11:40 am	***Symposium 5*** Nancy Targett, University of Delaware, *The Effect of Phlorotannins on Digestion in Marine Herbivores: The Role of the Gut Environment.*
11:40 - 12:00 Noon	***Symposium 6*** Peter Steinberg, University of New South Wales, Australia, *Quantitative Variation in Temperate Seaweed Secondary Metabolites.*
12:00 - 12:20 pm	***Symposium 7*** Niels Lindquist, University of North Carolina-Chapel Hill, *Chemical Defense of Invertebrate Larvae.*

Symposium 1

CHEMICAL DEFENSES OF CARIBBEAN SPONGES. Joseph R. Pawlik. Center for Marine Science Research, University of North Carolina at Wilmington, NC 28403-3297 USA.

Marine sponges are sessile, lack obvious physical defenses, and are rich in unusual secondary metabolites, much like many terrestrial plants and marine algae. Preliminary surveys of crude extracts of over 50 species of reef and mangrove sponges from Belize, the Bahamas and Florida for their capacity to deter feeding by generalist predatory reef fishes have revealed considerable variability between some families, but also within some genera. Sponges in the orders Verongida and Axinellida were uniformly deterrent, while several species in the Haplosclerida yielded non-deterrent extracts, including very common species such as *Callyspongia vaginalis* and *Xestospongia muta*. Pronounced intraspecific variability in deterrence was noted for some species. There is no apparent relationship between the feeding deterrence of crude extracts and previous assessments of their toxicity to fish. Glass spicules, which perfuse the tissues of many marine sponges, did not deter feeding of generalist predators. Two common species of mangrove sponges, *Chondrilla nucula* and *Tedania ignis*, were rapidly eaten by angelfishes when they were transplanted to reef habitats, suggesting that these specialist predators may limit the distributions of some Caribbean sponges.

Symposium 2

WHY SMALL HERBIVORES EAT TOXIC PLANTS: MARINE-TERRESTRIAL CONTRASTS. Mark E. Hay. University of North Carolina at Chapel Hill's Institute of Marine Sciences, Morehead City, NC 28557 USA.

Attempts to understand the evolution of chemically-mediated host-plant specialization in terrestrial communities have been complicated by the huge numbers of specialist species, the fact that almost all of these are insects, and the problem that these specialized relationships evolved over millions of years when now extinct megaherbivores were common and could have play a large role in affecting host-plant specialization by insects. In contrast, marine systems have very few specialist species, these are distributed broadly among different taxonomic groups, and the tremendous terrestrial changes that occurred during the Pleistocene extinctions did not occur in marine communities. Marine communities, thus, offer advantages for investigating the evolution of chemically-mediated specialization. Investigations of Caribbean, tropical Pacific, and temperate Atlantic seaweed-herbivore interactions showed that specialists are invariably small, relatively sedentary, subject to high potential rates of predation, and selectively use plants that are chemically defended from fishes. The specific compounds that deterred fishes were often used as host-plant cues by the specialists. In all cases, specialists used their chemically-defended host to avoid or deter predators. Host-plant specialization appears to be driven be the need for safe habitats rather than the need for specific foods. Similar constraints also may be driving host-plant specialization in terrestrial communities.

Symposium 3

MEDIATION OF ALGAL-SEA HARE-PREDATOR INTERACTIONS BY ALGAL SECONDARY METABOLITES. Steven C. Pennings and Valerie J. Paul. Marine Laboratory, University of Guam, Mangilao, Guam 96923 USA.

Sea hares are large, herbivorous, opisthobranch gastropods. They commonly consume chemically-rich algae and sequester secondary metabolites. However, we suggest that sea hares may not be highly adapted to either 1) preferentially consume algae containing secondary metabolites or 2) effectively utilize algal secondary metabolites for defense. 1) Consumption. Many sea hares eagerly consume palatable algae such as *Ulva* spp. that lack known secondary metabolites. Although the generalist sea hare *Dolabella auricularia* is indifferent to crude extracts of many algae, the specialist sea hare *Stylocheilus longicauda* is deterred from feeding by the secondary metabolites malyngamide A and malyngamide B from its host plant, *Microcoleus lyngbyaceus*. 2) Defense. Mortality rates of small sea hares are much higher than is generally realized. Sequestered algal compounds are primarily located internally, rather than in the skin where they would be readily detected by predators. Consequently, predators are often unable to distinguish between sea hares that contain and lack secondary metabolites, although the post-ingestive consequences of feeding on the two groups are different. Furthermore, the in vivo transformation of some compounds by sea hares appears to produce less effective feeding deterrents. Sea hare eggs and ink (released when animals are disturbed) appear to contain defensive compounds that are not derived from the algal diet.

Symposium 4

ASPECTS OF THE CHEMICAL ECOLOGY OF SHALLOW-WATER MARINE INVERTEBRATES IN McMURDO SOUND, ANTARCTICA. James B. McClintock and *Bill J. Baker. Department of Biology, University of Alabama at Birmingham, AL 35294-1170 USA and *Department of Chemistry, Florida Institute of Technology, Melbourne, FL 32901 USA.

Little information is available on the presence of bioactive compounds in polar marine invertebrates. In contrast to earlier predictions, our ongoing studies of the chemical ecology of antarctic marine invertebrates reveal a moderate incidence of bioactivity. Tube-foot chemotactic responses of the spongivorous antarctic sea star *Perknaster fuscus* to organic extracts of 18 species of sponges suggest bioactive secondary metabolites occur in a number of species. Two of the sponges which show very little or no biological activity (*Mycale acerata* and *Homaxinella balfournesis*) are rapidly growing, "weedy" species, both of which are the primary sponges in the natural diet of this spongivorous sea star. A number of the antarctic sponges which show biological activity have recently been found to possess secondary metabolites. We also report data which suggests that chemical defense occurs in representatives of antarctic soft corals, gastropods, pteropods, brachiopods, nemerteans and echinoderms. These studies indicate that, similar to temperate and tropical marine invertebrates, bioactive chemicals are present in a wide diversity of antarctic marine invertebrates. Statistically robust biogeographical comparisons will require further chemical and ecological analyses of larger numbers of species. Supported by NSF grants # DPP-9118864 and DPP-9117216.

Symposium 5

THE EFFECT OF PHLOROTANNINS ON DIGESTION IN MARINE HERBIVORES: THE ROLE OF THE GUT ENVIRONMENT. Nancy M. Targett. University of Delaware, Graduate College of Marine Studies, Lewes, DE 19958 USA.

Polyphenolic compounds are known to act as both feeding deterrents, digestibility reducing substances, and toxins in terrestrial and marine systems. Their action is dependent upon the chemical nature of the polyphenolics and the chemical environment in which they are acting. Here I report on *in vivo* assimilation efficiency studies of phenolics from several brown algal species with four marine herbivores having different gut conditions. Marine polyphenolic extracts negatively affected assimilation efficiency in the temperate herbivorous fishes, *Xiphister mucosus* and *Girella nigricans* (acidic guts) at naturally occurring concentrations (2% dry weight). The effect was dose-dependent and transitory. It was also dependent upon phenolic molecular size with high molecular weight phlorotannins, >10 kDa, decreasing assimilation efficiency to zero at naturally occurring phenolic concentrations. Phlorotannin structure type (e.g. fuhalols, fucophlorethols, eckols) was not as important as phlorotannin size in their effects on assimilation efficiency. Two tropical herbivores, the fish *Sparisoma radians* (basic gut) and the crab *Mithrax sculptus* (acid gut) were unaffected by brown algal phlorotannins at naturally occurring concentrations(2%-15% dry weight). Factors intrinsic and extrinsic to the guts of marine herbivores and to brown algal polyphenolics affect polyphenolic-macromolecule complexations and thus alter their effect on *in vivo* herbivore assimilation efficiencies.

Symposium 6

QUANTITATIVE VARIATION IN TEMPERATE SEAWEED SECONDARY METABOLITES. Peter D. Steinberg, Rocky de Nys and John Runcie. University of New South Wales, Kensington, NSW, 2033 Australia

Persistent broad scale variation of algal secondary metabolites is reasonably well described for a number of systems, and is generally linked to parallel variation in the intensity of herbivory. However, with a few notable exceptions, we know little about factors determining small scale, quantitative variation in seaweed secondary metabolites. We have quantitatively described temporal and small scale spatial variation in metabolites for three seaweeds in temperate NSW, Australia: *Ecklonia radiata*, which produces phlorotannins (polyphenolics); *Delisea pulchra*, which produces non-polar halogenated lactones, and; *Zonaria diesingiana*, (phlorotannins and prenylated quinones). Small scale variation in the intensity of herbivory (either spatially, or for *E. radiata*, temporally) does not explain variation in metabolites, and we are now focussing on the effects of light and nutrients. In contrast to most models, and some data, variation in metabolites is not inversely correlated with variation in nitrogen levels among plants or across the habitat. Moreover, in contrast to the predictions of most models, *E. radiata* appears to emphasize maintenance of high phlorotannin levels over increased growth, even when nitrogen levels are adequate for rapid growth.

CHEMICAL DEFENSE OF INVERTEBRATE LARVAE. Lindquist, N. Institute of Marine Sciences, University of North Carolina - Chapel Hill, Morehead City, NC 28557 USA.

It is now apparent that adult stages of common sessile invertebrates are often defended against consumers by the production of unusual secondary metabolites, however, little is known about chemical defenses in larval and juvenile stages. Using feeding preference assays, I show that the physically-defenseless larvae of many tropical sponges, ascidians, and octocorals are distasteful to co-occurring reef fishes and benthic cnidarians. Similarly, larvae of several temperate bryozoans and hydroids are also rejected as prey by co-occurring consumers. Surprisingly, the survivorship of larvae tasted and rejected by fishes did not differ significantly from unattacked control larvae; however, benthic cnidarians were more likely than fish to consume larvae or kill larvae that they ultimately rejected as food. The rejection by consumers of artificial food pellets treated with larval crude extracts indicates that the unpalatability of many larvae is chemically based. For many species studied, both adults and larvae are commonly chemically defended, however, for some invertebrates, crude extracts of adult stages were readily eaten by consumers that rejected foods treated with larval extracts. Results of these investigations suggest that predation may have minimal impact on the numbers of chemically-defended larvae available for recruitment.

Symposium II (Monday - August 2)

Chemical Ecology of Terpenoids

Beach Gulf Room - Jean Langenheim Presiding

9:00 - 9:50 am	***Symposium 8*** Jean Langenheim, University of California - Santa Cruz *Higher Plant Terpenoids: A Phytocentric Overview of Their Ecological Roles.*
9:50 - 10:20 am	***Symposium 9*** Jonathan Gershenzon, Washington State University *The Metabolic Costs of Terpenoid Accumulation in Higher Plants.*
10:20 - 10:40 am	**COFFEE BREAK**
10:40 - 11:10 am	***Symposium 10*** Nikolaus Fischer, Louisiana State University - Baton Rouge, *Recent Studies on Allelopathic Mechanisms Involving Terpenoids.*
11:10 - 11:40 am	***Symposium 11*** Carleton S. White, University of New Mexico - Albuquerque, *Monoterpenes: Their Effects on Ecosystem Nutrient Cycling.*
11:40 - 12:10 am	***Symposium 12*** Junji Takabayashi, Kyoto University - Japan *Terpenoids as Mediators of Induced Indirect Defense of Plants Against Herbivores.*

Symposium 8

HIGHER PLANT TERPENOIDS: A PHYTOCENTRIC OVERVIEW OF THEIR ECOLOGICAL ROLES. Jean H. Langenheim. Department of Biology, University of California, Santa Cruz, CA 95060 U.S.A.

Characteristics of higher plant terpenoids that result in mediation of numerous kinds of ecological interactions will be discussed as a framework for the Symposium. However, the role that mixtures of terpenoids, either constitutive or induced, their qualitative and quantitative compositional variation and dosage dependent effects will be emphasized in subsequent discussions. Selected phytocentric examples will be presented of terpenoid interactions: 1) defense against generalist and specialist insect and mammalian herbivores, 2) defense against insect-vectored fungi and potentially pathogenic endophytic fungi, 3) attraction of entomophages and pollinators, 4) allelopathic effects that inhibit seed germination and soil bacteria, and 5) interaction with reactive troposphere gases. The results will be integrated by discussing how these terpenoids are contributing factors in determining some properties of terrestrial plant communities and ecosystems. A terrestrial phytocentric approach is necessitated due to the magnitude and scope of terpenoid interactions. This presentation will have a more broadly based ecological perspective than the several excellent recent reviews of the ecological chemistry of terpenoids.

Symposium 9

THE METABOLIC COSTS OF TERPENOID ACCUMULATION IN HIGHER PLANTS. Jonathan Gershenzon. Washington State University, Pullman, WA 99164-6340 USA.

The ability of plants to accumulate terpenoid compounds for defense, allelopathy or other functions is generally thought to be constrained by the metabolic costs of these substances. I will discuss several components of terpenoid accumulation costs in the light of recent progress in the biochemistry and physiology of plant terpenoid metabolism. 1) *Costs of synthesis*. Terpenoid formation requires a supply of raw materials (substrates and cofactors) and various types of macromolecular machinery, including enzymes and nucleic acids. Raw materials costs calculated from biosynthetic pathways indicate that the most expensive terpenes to produce per gram are those with the highest degree of chemical reduction. Biosynthetic machinery costs cannot yet be estimated with any degree of certainty, but the fact that terpenoid formation is frequently confined to brief periods in plant development would appear to minimize the possible costs of enzyme turnover. 2) *Costs of storage*. Terpenoid substances are usually stored in complex secretory structures whose construction entails a significant investment of resources. 3) *Costs of maintenance*. The cost of compensating for terpenoids lost during storage is thought to be quite substantial due to the high incidence of turnover. However, recent studies have found no evidence for rapid rates of terpene turnover suggesting that maintenance costs may actually be very low.

RECENT STUDIES ON ALLELOPATHIC MECHANISMS INVOLVING TERPENOIDS. Nikolaus H. Fischer, G. Bruce Williamson and Jeffrey D. Weidenhamer ‡. Louisiana State University, Baton Rouge, LA 70803 USA; ‡ Ashland University, Ashland, OH 44805 USA.

The hypothesis that allelopathic agents released from members of the Florida scrub community deter the invasion of fire-prone sandhill grasses was investigated. Constituents of the endemic scrub members *Ceratiola ericiodes*, *Conradina canescens*, *Calamintha ashei* and *Chrysoma pauciflosculosa* were examined for phytotoxic activity. Effects of the plant natural products and their derivatives on the germination and radicle growth of little bluestem (*Schizachyrium scoparium*) and green sprangletop (*Leptochloa dubia*), two native grasses of the Florida sandhill community, were tested.This will include studies related to the release mechanism of allelopathic agents from a source plant and their transport to the target species. Also, allelopathic activations of nontoxic agents, after release from a source plant into the environment, will be discussed.

Symposium 11

MONOTERPENES: THEIR EFFECTS ON ECOSYSTEM NUTRIENT CYCLING. Carleton S. White. Department of Biology, University of New Mexico, Albuquerque, NM, 87131 USA.

This article explores the evidence for monoterpenes to alter rates of nutrient cycling, with particular emphasis on the nitrogen cycle, from an ecosystem perspective. First, the article reviews the nitrogen cycle and notes particular processes where monoterpenes could exert control. Next, the theoretical and conceptual basis for the proposed mode of action by monoterpenes on particular processes within the nitrogen cycle is presented along with recent developments. Experimental support for these roles is presented that includes effects at the cellular level and progresses through populations and communities (microbial and invertebrate) involved in nitrogen mineralization and immobilization processes. Particular processes within the carbon cycle, which has similarities with the nitrogen cycle, that may be altered by monoterpenes also are noted. Finally, areas for future research that appear most promising are suggested.

TERPENOIDS AS MEDIATORS OF INDUCED INDIRECT DEFENSE OF PLANTS AGAINST HERBIVORES.

Junji Takabayashi[1] and Marcel Dicke[2]. [1]Pesticide Research Institute, Faculty of Agriculture, Kyoto University, Kyoto 606, Japan. [2]Department of Entomology, Wageningen Agricultural University, P.O.Box 8031, 6700 EH, Wageningen, The Netherlands.

It has recently been shown that herbivores may induce in plants the production of volatile infochemicals that attract predators of the herbivores. This response is considered to be an induced indirect defense mechanism of plants against herbivores. Most of the predator attractants produced by plants upon herbivore damage have been found to be terpenoids. Different plant species and cultivar produce their own characteristic combination of chemicals according to the attacking herbivore species.

In this presentation, I will report on the specificity of attractant production in the system of plants, two-spotted spider mites Tetranychus urticae and the predatory mites Phytoseiulus persimilis, and also the tritrophic interaction of plants, armyworms pseudaletia separata and the parasitic wasps Cotesia kariyai.

Finally, I will compare the effectiveness of the two systems.

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Symposium III (Tuesday - August 3)

Physical Stress and Chemical Ecology

Beach Gulf Room **John Romeo Presiding**

9:00 - 9:40 am ***Symposium 13*** Jerry McClure, Miami University - Ohio, *Epidermal Flavonoids and UV-B Attenuation: Plant Responses To Ozone Depletion.*

9:40 - 10:20 am ***Symposium 14*** Barbara Bentley, State University of New York *The Effects of Global Change on The Interactions Between An Insect Herbivore and Its N-Fixing Host Plant.*

10:20 - 10:40 am **COFFEE BREAK**

10:40 - 11:20 am ***Symposium 15*** John Pickett, AFRC Rothamsted Experimental Station - UK,*Wound-induced Biosynthetic Pathways and Phytopheromones in Herbivore, and Pathogen Interactions With Plants.*

11:20 - 12:00 Noon ***Symposium 16*** Rainer Jaenicke, University of Regensburg - Germany *Protein Stability and Molecular Adaptation To Extremes of Physical Conditions.*

EPIDERMAL FLAVONOIDS AND UV-B ATTENUATION: PLANT RESPONSES TO OZONE DEPLETION. Jerry W. McClure, Miami University, Oxford OH 45056 USA.

Although flavonoids and hydroxycinnamic acids absorb UV-B and could theoretically protect underlying photosynthetic tissues from damage, they are not restricted to the epidermis of all species, their level of accumulation varies greatly between species and cultivars apparently well adapted to current UV-B levels (which vary markedly with latitude), and they are induced or quantitatively increased by many stimuli in addition to UV light. We find that when grown with supplemental UV-B simulating 25% ozone depletion at 40°N, Williams soybean adapts by reduced leaf area, thicker leaves, and more epidermal kaempferol while Essex soybeans lack these adaptive responses and their photosynthetic system is damaged. Atlas barley responds by decreased rates of leaf elongation, prolonged PAL activity, and increased C-Glycosylflavone accumulation in both epidermal and mesophyll tissues. Implications of such studies to projected responses of major crop plants to increased UV-B will be considered.

Symposium 14

THE EFFECTS OF GLOBAL CHANGE ON THE INTERACTIONS BETWEEN AN INSECT HERBIVORE AND ITS N-FIXING HOST PLANT. Barbara L. Bentley, State University of New York at Stony Brook, Stony Brook, NY 11794 USA.

Atmospheric CO_2 concentration is increasing at an unprecedented rate. Projections indicate that CO_2 levels will rise from the current concentration of 348 ppm to about 600 ppm in the next 75 years. The well-known "greenhouse" effect is likely to be one consequence of increased CO_2 concentrations. But, perhaps even more importantly, biological systems will also respond to the direct effects of CO_2: Carbon dioxide is a starting material for photosynthesis. In most cases, plants have increased photosynthetic rates, increased growth, and increased biomass. However, because N availability is not changed, the percent N in the tissues decreases. The response of N-fixing plants is quite different, however. N-fixers can use the "extra" photosynthate to fuel the fixation system, leading to an increase in tissue N. These differences in N can be very important for the insect herbivores feeding on the foliage.

To test these differences, we grew lupine (Lupinus arboreus and L. nanus; Leguminosae) at three levels of CO_2 (350, 500, and 650 ppm), and either with the N-fixing symbiont (Bradyrhizobium), or without Bradyrhizobium but given high or low levels of inorganic N. Biomass, carbon and N content, and alkaloid concentrations of the foliage produced by the plants were measured, then foliage was fed to an insect herbivore (Spodoptera eridania) to determine changes in the quality of the lupin foliage as food for the herbivore.

The results of our feeding experiments suggest a complex relationship between CO_2 enrichment and quality of lupin foliage. Feeding rates on foliage from the low (350 ppm) CO_2 plants were higher than the other treatments — suggesting that the food was palatable but relatively low in protein. In contrast, consumption was lower but digestibility was higher on the higher (500 and 650 ppm) CO_2 plants, suggesting that palatability was low, but protein content high in these samples. Overall growth rates, however, remained highest in those fed foliage from the 350 CO_2 plants, suggesting that the change in alkaloids is more important than the change in protein. Confirmation of this conclusion awaits the completion of the chemical analyses of the foliage from these experiments.

Symposium 15

WOUND-INDUCED BIOSYNTHETIC PATHWAYS AND PHYTOPHEROMONES IN HERBIVORE AND PATHOGEN INTERACTIONS WITH PLANTS. John A. Pickett. AFRC Rothamsted Experimental Station, Harpenden, Hertfordshire, AL5 2JQ, United Kingdom.

Plant defence mechanisms against herbivore and pathogen development, for example protease inhibitors and toxic secondary metabolites such as alkaloids, can be induced during initial damage. The investigation of induction mechanisms has recently benefited by identification of damage-related signals, including translocatable plant components. In addition, damaged plants can release signals (phytopheromones) that induce defence pathways in other individuals of the same species. The subject will be reviewed. New work on phytopheromone-mediated interactions will be discussed, including components directly influencing the behaviour of herbivores and their predators and parasitoids.

Symposium 16

PROTEIN STABILITY AND MOLECULAR ADAPTATION TO EXTREMES OF PHYSICAL CONDITIONS Rainer Jaenicke. University of Regensburg, W-8400 Regensburg FRG.

Proteins exhibit marginal stabilities, equivalent to only a few weak intermolecular interactions. Extreme environments require either molecular adaptation in terms of local structural changes or stabilization by "extrinsic factors" not encoded in the amino acid sequence. Such factors are, among others, specific metabolites, cofactors, ions and chaperones. No general strategies of stabilization have yet been established. However, certain incremental contributions to stability have been elucidated, analyzing extremely stable proteins, e.g., from "extremophiles", on one hand, and point mutants obtained by site-directed mutagenesis, on the other.
Stabilization may occur at three levels: 1. local packing of the polypeptide chain, 2. subdomains or domains and their mutual interactions, and 3. association of subunits. Experimental approaches (and representative examples) are, ad 1: fragmentation of single-chain proteins (e.g., thermolysin from *Bacillus thermoproteolyticus*), and comparison of homologous proteins (such as NAD-dependent dehydrogenases) from extremophilic and mesophilic organisms; ad 2: "nicking" of domain proteins and "transplanting" their linker peptides (e.g., crystallins from eye lens); ad 3: denaturation-renaturation experiments using oligomeric or multimeric proteins (e.g., proteolytic enzymes from *Bacillus stearothermophilus* or *Thermotoga maritima*).

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Symposium IV (Wednesday - August 4)

Tropical Chemical Ecology

Beach Gulf Room **Lincoln Brower Presiding**

8:30 - 9:10 am *Symposium 17* Jeffrey Harborne, Plenary Lecture. *Flavonoids and Insects.*

9:10 - 9:40 am *Symposium 18* Kelsey Downum, Florida International University - Miami, *Solar-powered Toxins from Tropical Plants.*

9:40 - 10:10 am *Symposium 19* Roberto Trigo, Departamento de Zoologia - Brazil *Stereochemical Inversion of Pyrrolizidine Alkaloids (PA's) by Ithomiine Butterflies: Origins of PA Use by Lepidoptera.*

10:10 - 10:30 am **COFFEE BREAK**

10:30 - 11:00 am *Symposium 20* Steve Malcolm, Western Michigan University - Kalamazoo, *Steroids versus Alkaloids In Plant-butterfly Interactions.*

11:00 - 11:30 am *Symposium 21* Doug Levey, University of Florida - Gainesville *The Chemical Ecology of Bird-fruit Interactions.*

11:30 - 12:00 Noon *Symposium 22* Lincoln Brower, University of Florida - Gainesville *Coevolution Revisited: Asclepias humistrata Latex versus First Instar Monarch Butterfly Larvae.*

12:00 - 12:30 pm *Symposium 23* Mark Hay, The University of North Carolina - Chapel Hill, *Multiple Functions of Seaweed Secondary Metabolites: Constraints on Chemically Mediated Coevolution.*

PLENARY LECTURE

Silver Medal Award

Flavonoids And Insects

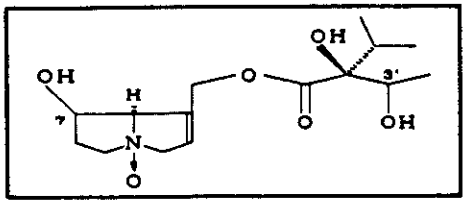
Professor Jeffrey Harborne

SOLAR-POWERED TOXINS FROM TROPICAL PLANTS. Kelsey R. Downum, Florida International University, Miami, FL 33199 USA.

Sunlight provides the energy required for all biochemical, physiological and developmental processes necessary for plant growth, reproduction and survival. The role of light in photosynthetic and photomorphogenetic processes has been appreciated for some time; however, its participation in plant defense is a comparatively recent realization. Approximately 100 photosensitizers or phototoxins, molecules that become toxic in the presence of light, have been extracted from higher plant tissues. These biologically-active compounds have diverse biosynthetic origins and belong to at least 15 different phytochemical classes. Phototoxins and/or phototoxic activity have been reported in some 40 families of the Magnoliophyta. Most of the structurally-identified phototoxins are acute toxins with little organism-specificity. As such, they are effective biocides capable of killing a wide-range of potentially harmful organisms including: viruses, pathogenic bacteria/fungi, nematodes and herbivorous insects, as well as competing plant species. The available information suggests that phototoxic phytochemicals represent defensive components in both primitive and advanced plant taxa, and may be effective under a variety of photoenvironmental conditions. The phytochemistry, taxonomic occurrence and toxicological consequences of phototoxic metabolites in flowering plants will be discussed.

Symposium 19

STEREOCHEMICAL INVERSION OF PYRROLIZIDINE ALKALOIDS (PAs) BY ITHOMIINE BUTTERFLIES: ORIGINS OF PA USE BY LEPIDOPTERA. J.R. Trigo[1,2], K.S. Brown Jr.[1] & L.E.S. Barata[2]. Departamento de Zoologia, Instituto de Biologia[1] & Instituto de Química[2], UNICAMP, Campinas, SP, 13081 Brazil.



Pyrrolizidine alkaloids (PAs) protect Danainae and Ithomiinae butterflies and Arctiidae moths against predators, and are biosynthetic precursors of male sex pheromones in these Lepidoptera. The investigation of PAs in several species of wild caught adults of Ithomiinae showed lycopsamine (7R-OH,3'S-OH) as the main alkaloid, always present in the N-oxide form. In an incorporation experiment, PA-free (freshly emerged) adults of the ithomiine Mechanitis polymnia were fed lycopsamine and three of its known natural stereoisomers - intermedine (7R-OH, 3'R-OH), echinatine (7S-OH, 3'S-OH) and rinderine (7S-OH, 3'R-OH). Males butterflies isomerized all PAs, mainly to lycopsamine. Females fed PAs changed only part of these to lycopsamine; they may have little capacity to invert PAs, since they acquire already transformed PAs from males by mating and rarely visit adult sources of PAs. The inversion from 7S to 7R (probably via oxi-reduction) may be closely related to the evolution of acquisition of PAs by butterflies and moths: the ancestral butterflies probably adapted to tolerate and then assimilate 7R-PAs (all of the widespread macrocyclic PA diesters including in the tribe Senecioneae, more primitive than Eupatorieae, are 7R); the later appearance of 7S-PAs in plants could have selected the inversion mechanism. The inversion is reflected in the 7R configuration of all known PA derived pheromones. The inversion of the 3'-asymmetric center, when the butterfly was fed rinderine remains to be explained: it could be due the oxi-reduction enzime structure or the alkaloid conformation.

Symposium 20

STEROIDS VERSUS ALKALOIDS IN PLANT-BUTTERFLY INTERACTIONS.

Stephen B. Malcolm. Western Michigan University, Kalamazoo, MI 49008 USA.

There is little doubt that some steroids and alkaloids are effective chemical defenses in some butterfly species and that these defenses can be sequestered from the larval or adult host plants of the butterflies. Thus butterfly sequestration is a reasonably well documented phenomenon. In contrast, we know very little about the same chemicals in the host plants of sequestering butterflies, or about the effects of these plant-derived chemicals on real natural enemies of butterflies. Cardenolides and pyrrolizidine alkaloids are discussed as well known examples of defensive steroids and alkaloids and comparisons are made between anecdotal claims and new evidence that these two groups of plant metabolites are plant defenses. This evidence is used to address the issue of whether cardenolides and pyrrolizidine alkaloids have blinded us to other equally significant interactions or whether there really is something special about these two very different kinds of chemical defense.

Symposium 21

The chemical ecology of bird-fruit interactions. Douglas J. Levey, Carla Restrepo, Department of Zoology, University of Florida, Gainesville, FL 32611 and Carlos Martínez del Rio, Department of Ecology and Evolutionary Biology, Princeton University, Princeton, NJ 08544.

We examine how the nutritional composition of fleshy fruits can be explained by the digestive physiology of fruit-eating birds. Because all fruits contain seeds that are indigestible to fruit-eating birds, these birds must deal with a high bulk diet. They do so by passing seeds and pulp quickly through their digestive system. We present experimental evidence that rapid passage leads to low digestive efficiencies and suggest that low digestive efficiency may have selected for easily assimilated nutrients in fruit pulp. Indeed, most fleshy fruits are rich in monosaccharides and free amino acids. We then examine how lack of a single intestinal enzyme, sucrase, can affect the types of fruits taken by some birds and how patterns of nutrient composition of two families of mistletoes and fruit choice among their dispersers suggest a complex evolutionary history of chemical ecology.

Symposium 22

Coevolution revisited: Asclepias humistrata latex *versus* first instar monarch butterfly larvae. Lincoln P. Brower, Department of Zoology, University of Florida, Gainesville, FL 32608 and Myron P. Zalucki, Entomology Department, The University of Queensland, Brisbane, Australia 4072.

An experimental study in a natural habitat of Asclepias humistrata and monarch butterflies (Danaus plexippus) in North Florida measured the effect of the latex contained in the leaves of this important spring foodplant on first instar larvae. Larvae were allowed to hatch (A) on leaves that were deprived of their latex and (B) on control leaves that were left intact. Rates of development, gluing, disappearance, and feeding behavior were all detrimentally affected by the latex. The behavioral responses of the larvae on the control and experimental leaves will be compared in a video tape.

MULTIPLE FUNCTIONS OF SEAWEED SECONDARY METABOLITES: CONSTRAINTS ON CHEMICALLY MEDIATED COEVOLUTION. Mark E. Hay, Tim Schmitt, and Niels Lindquist. University of North Carolina at Chapel Hill's Institute of Marine Sciences, Morehead City, NC 28557 USA.

Although chemically-mediated coevolution between plants and herbivores is thought to be common, such coevolution will depend on selection altering the chemical defenses of the plant following the evolution of resistance by herbivores. Such a reciprocal response will be constrained if compounds play multiple roles that are ecologically important. We show that the brown seaweed *Dictyota* produces secondary metabolites that are broadly defensive against a wide variety of both consumers and competitors. Although certain consumers may evolve resistance to these metabolites, it is unclear that feeding by these consumers will result in reciprocal responses from the plant. We suggest that coevolved interactions may be much less common than is generally thought, and that many interactions which appear to be coevolved may result from fortuitous and opportunistic preadaptations.

CONTRIBUTED PAPERS
ORAL

10th ANNUAL MEETING
International Society of Chemical Ecology
Tampa/Clearwater Florida
July 31-August 4, 1993

Contributed Papers

Session I (Sunday - August 1)

Marine Subjects

Palm Room **Nancy Targett Presiding**

2:00 - 2:15 pm ***Oral Paper 1*** CHEMICAL ASPECTS OF SHALLOW-WATER MARINE INVERTEBRATE ECOLOGY IN MCMURDO SOUND, ANTARCTICA. Bill J. Baker, James B. McClintock, Mark Hammon, Bob Kopitzke and Paul J. Scheuer.

2:15 - 2:30 pm ***Oral Paper 2*** BIOGENIC ORGANOHALOGENS FROM *SACCOGLOSSUS KOWALEVSKI* (PHYLUM HEMICHORDATA, CLASS ENTEROPNEUSTA): PATTERNS, POTENTIAL SOURCES AND SIGNIFICANCE OF CHEMICAL VARIATION. Kevin T. Fielman and Nancy M. Targett.

2:30 - 2:45 pm ***Oral Paper 3*** A MICRO-FOLIN-DENIS METHOD FOR THE RAPID QUANTIFICATION OF PHENOLIC COMPOUNDS IN MARINE PLANTS AND ANIMALS. Walter I. Hatch, Christopher E. Tanner, Nadine M. Butler and Egan P. O'Brien.

2:45 - 3:00 pm ***Oral Paper 4*** VARIATION IN THE SECONDARY METABOLITES OF THE SPONGE *DYSIDEA* CF. *AVARA*. Valerie J. Paul and Bridgette S. Davidson.

3:00 - 3:15 pm ***Oral Paper 5*** GEOGRAPHIC, TEMPORAL, AND ONTOGENETIC VARIATION IN POLYPHENOLIC ANTIHERBIVORE DEFENSES OF THE KELPS *ALARIA MARGINATA AND A. NANA*. Kathryn L. Van Alstyne, David O. Duggins, and James McCarthy.

3:15 - 3:30 pm ***Oral Paper 6*** EVIDENCE FOR BIOTRANSFORMATION OF DIETARY ALLELOCHEMICALS IN THE TROPICAL MARINE TELEOST *CHAETODON CAPISTRATUS*. Nicholas H. Vrolijk, Nancy M. Targett, Bruce R. Woodin, and John J. Stegeman.

Oral Paper 1

CHEMICAL ASPECTS OF SHALLOW-WATER MARINE INVERTEBRATE ECOLOGY IN McMURDO SOUND, ANTARCTICA. Bill J. Baker, James B. McClintock, Mark Hammon, Bob Kopitzke and Paul J. Scheuer. Department of Chemistry, Florida Institute of Technology, Melbourne, FL 32901 USA, Department of Biology, University of Alabama at Birmingham, AL 35294-1170 USA, and Department of Chemistry, University of Hawaii, 2545 The Mall, Honolulu, HI 96822 USA.

Recent chemical ecological investigation of antarctic marine invertebrates has revealed a moderate incidence of bioactivity. Using ecologically relevant assays, extracts containing bioactive substances have been identified and are currently being evaluated for novel secondary metabolites. A picture is beginning to emerge of the nature of the bioactivity in antarctic marine invertebrates and this picture suggests that these polar organisms can share metabolic pathways with their temperate relatives and that coloration plays a significant role as an indicator of secondary metabolism. A discussion will be presented to support these observations and structures will be presented of some of the new secondary metabolites we have found.

Oral Paper 2

BIOGENIC ORGANOHALOGENS FROM *SACCOGLOSSUS KOWALEVSKII* (PHYLUM HEMICHORDATA, CLASS ENTEROPNEUSTA): PATTERNS, POTENTIAL SOURCES AND SIGNIFICANCE OF CHEMICAL VARIATION. Kevin T. Fielman and Nancy M. Targett. Graduate College of Marine Studies, University of Delaware, Lewes DE 19958 USA

The occurrence of halogenated metabolites is especially consistent among the marine hemichordate worms. To aid in the interpretation of their ecological role and to contribute to our knowledge of chemical variation among marine invertebrates, we quantified spatial and temporal patterns of halocarbon variation in *S. kowalevskii*, a common species which contains Br-phenols or pyrroles. By FABMS we found that the novel Na sulfamate salt ($C_4HBr_3NO_3SNa$) (TBP A) of 2,3,4-tribromopyrrole (TBP B) predominated (11% vs. 0.6% ash-free dry weight). There was no relationship between the total amount of each form. TBP A was concentrated in the hepatic region (50%); a possible site of TBP synthesis or storage. TBP B was highest (1.5%) in the proboscis and tail, which are exposed to predation. No temporal pattern was apparent for TBP A. TBP B increased during spawning months. TBP A was not detectable by traditional GC/MS techniques, thus raising the possibility that sulfonation may be more common than realized. The sulfamate salt may serve as the non-autotoxic stable precursor to the more volatile TBP B; possibly a predator deterrent or an allelochemical agent. By reducing loss of head and tail tissue during gametogenesis, *Saccoglossus* may increase its fitness by maximizing reproductive output. Whether any of the observed interorganismal variation in TBP content (3-5x) could be attributable to differences in resource acquisition or genetic factors is unknown. With a defined and variable chemistry and its potential for laboratory culture, *Saccoglossus* constitutes an excellent marine model system in which to study the significance of variation in secondary metabolism.

Oral Paper 3

A MICRO-FOLIN-DENIS METHOD FOR THE RAPID QUANTIFICATION OF PHENOLIC COMPOUNDS IN MARINE PLANTS AND ANIMALS. Walter I. Hatch, Christopher E. Tanner, Nadine M. Butler and Egan P. O'Brien. St. Mary's College of Maryland, St. Mary's City, MD 20686 USA.

A micro-Folin-Denis method was developed for the rapid assay of phenolic compounds in small amounts of tissue (approximately 1 to 200 mg fresh weight). A primary difference between this and previous methods is the use of trichloroacetic acid in the extraction solution to prevent under estimation of phenolic compounds as a result of co-precipitation with proteins. Additional micro-methods were developed to eliminate interference from non-aromatic reducing compounds, and for determining the astringency of phenolic compounds. These methods allow for the rapid processing of large numbers of samples and the analysis of small individual organisms or portions of organisms. Using the micro-Folin-Denis method, localization of phenolic compounds were studied in the brown alga *Padina gymnospora*, the seagrass *Thalassia testudinum*, and the burrowing sponge *Cliona laticavicola*. Significant differences in concentrations of phenolics compounds from different regions within each of these organisms were observed.

Oral Paper 4

VARIATION IN THE SECONDARY METABOLITES OF THE SPONGE *DYSIDEA CF. AVARA*. Bridgette S. Davidson and Valerie J. Paul, University of Guam Marine Laboratory, UOG Station, Mangilao, GU 96923 USA.

Dysidea cf. *avara*, a common sponge on reefs in Apra Harbor on Guam, exhibits significant spatial variation in secondary metabolite concentrations. The major secondary metabolite in the sponge, avarol, deterred feeding by reef fish at natural concentrations. We hypothesized that secondary metabolite concentrations in sponges are environmentally variable and tested this by transplantation of individual sponges between sites. Transplantation of sponges caused their secondary metabolite concentrations to become similar to control sponges, even though differences were significant between the two transplant sites. To examine factors influencing site-to-site variation in secondary metabolites in *Dysidea* cf. *avara*, we tested the effects of 1) artificial grazing using scissors, and 2) light and UV radiation. Variation in secondary metabolites was evaluated in two ways: 1) chemical analysis by quantitative HPLC and then 2) extracts of experimental sponges were compared with appropriate controls in field feeding assays designed to quantify the effects on potential predators. Artificial grazing and light experiments did not significantly alter secondary metabolite concentrations or reef fishes' preference for sponge extracts in feeding assays. We are currently testing the hypothesis that observed variation in secondary metabolite concentrations is due to variation in food availability at the different sites.

Oral Paper 5

GEOGRAPHIC, TEMPORAL, AND ONTOGENETIC VARIATION IN POLYPHENOLIC ANTIHERBIVORE DEFENSES OF THE KELPS ALARIA MARGINATA AND A. NANA. Kathryn L. Van Alstyne[1], David O. Duggins[2], and James McCarthy[1].[1]Department of Biology, Kenyon College, Gambier, OH 43022 USA, [2] Friday Harbor Labs, Friday Harbor, WA 98250 USA.

Polyphenolic compounds are feeding deterrents towards many herbivorous snails, urchins, and fishes. In this study, we examined variation in concentrations of these compounds over several spatial and temporal scales. Concentrations of phenolic compounds varied within individuals. In the San Juan Islands kelps, phenolic levels were highest in meristematic tissues, and lowest in the distal ends of the plants. There were no significant differences in phenolic levels between non-meristematic vegetative tissues and sporophylls. We also examined differences in polyphenolic concentrations of kelps along the west coast of North America, ranging from San Simeon, CA to Port Renfrew, BC. Concentrations of phenolics from A. marginata were 5 to 10 times higher in the San Juan Island population than concentrations reported from central California. Ontogenetic variation in phenolic levels was also measured by quantifying phenolic concentrations of kelps of different sizes collected from the San Juan Island site in April, 1993. Little seasonal variation was seen in phenolic concentrations of San Juan Island A. marginata in the summer of 1992.

Oral Paper 6

EVIDENCE FOR BIOTRANSFORMATION OF DIETARY ALLELOCHEMICALS IN THE TROPICAL MARINE TELEOST *Chaetodon capistratus*. Nicholas H. Vrolijk[1], Nancy M. Targett[2], Bruce R. Woodin[3], and John J. Stegeman[3]. [1]Center of Marine Biotechnology, Univ. of Maryland, Baltimore, MD 21202 USA, [2]Univ. of Delaware Graduate College of Marine Studies, Lewes, DE 19958 USA, [3]Woods Hole Oceanographic Institution, Woods Hole, MA 02543 USA

High levels of cytochrome P450 and glutathione transferase (GST) were detected in hepatic tissue of the butterflyfish, *Chaetodon capistratus*, a species that regularly feeds on terpenoid-rich gorgonian corals, as compared to two sympatric congeners that do not feed on gorgonians. Two P450 isozymes that have partially evolved in response to allelochemicals, CYP2B and CYP3A, were 2 to 10-fold and 2 to 20-fold higher, respectively, in *C. capistratus* than in *C. ocellatus* and *C. striatus*. The content of total P450 (0.588-0.794 nmol mg^{-1}), the activity of NADPH-cytochrome c (P450) reductase (288-311 nmol min^{-1} mg^{-1}) and the activity of GST (2.98-3.06 μmol min^{-1} mg^{-1}) were significantly greater in *C. capistratus* and are among the highest reported in teleosts from unpolluted waters. *Chaetodon capistratus* also had significantly larger livers per unit fish weight and more microsomal protein per g liver, factors that translate into 4 to 5-fold more total P450 per g fish. These results suggest that biotransformation enzymes have a potential detoxification role in a teleost that feeds on allelochemically rich gorgonian corals.

Oral Paper 7

UPTAKE AND SEQUESTRATION OF PYRROLIZIDINE ALKALOIDS BY LARVAE OF CHRYSOMELID BEETLES. Adelheid Ehmke*,Martine Rowell-Rahier§, Jacques M. Pasteels+, Thomas Hartmann*, *TU Braunschweig, D-38106 FRG, § Zool. Inst. Basel, CH-4051 Switzerland, +ULB, Brussels, B-1050 Belgium.

Leaf beetles from the genus *Oreina* are chemically defended. *Oreina cacaliae* and *O. speciosissima* sequester pyrrolizidine alkaloid N-oxides (Pa-Nox) from their host plants in their bodies and in their glands. Additionally, *O. speciosissima* but not *O. cacaliae* synthesizes autogenous cardenolides. We tested the ability of larvae of the two species for uptake and sequestration of Pa-Nox. Larvae of both species were able to accumulate [^{14}C]senecionine N-oxide (sen-Nox), larvae of *O. speciosissima* even more effectively, although in the field they feed on Pa free *Petasites paradoxus* plants. The stored sen-Nox seems to be lost during molting. In contrast to the adults, larvae are able to N-oxidize senecionine. Monocrotaline Nox was accumulated to the same extent as seneciphylline Nox. Thus, the storage of Pas in larvae is comparable to the storage in adults as far as the body is concerned, whereas the transfer into the beetles glands is highly specific for the Pas of the host plant *Adenostyles alliariae*.

Oral Paper 8

TLC-BIOAUTOGRAPHY-GUIDED ISOLATION OF RYANOID ANTIFEEDANTS FROM *PERSEA INDICA* AGAINST *SPODOPTERA LITURA*. Azucena González-Coloma[1,3], Pierre Escoubas[1], David Terrero[2], Braulio M. Fraga[2] and Matías Reina[2]. [1]JRDC, Eniwa RBP, Megumino Kita 3-1-1, Eniwa-Shi, Hokkaido, JAPAN. [2]IPNAC, CSIC, Avda. Astrofísico F. Sánchez 2, 38206 La Laguna, Tenerife, SPAIN. [3]Current address: CIB, CSIC, Velázquez 144, 28006 Madrid, SPAIN.

A new TLC-bioautography method, previously described for the isolation of insect antifeedants (Escoubas et al., 1992), was applied to guide the isolation of the active compounds responsible for the antifeedant activity of a *Persea indica* (Lauraceae) crude extract against *Spodoptera litura*. Silica gel vacuum-liquid and column chromatography of the active TLC fractions afforded six pure ryanoids previously found in this plant (González-Coloma et al., 1990; Fraga et al., in press). The feeding indexes (FR) of the compounds, tested at 1000 ppm, ranged between 0.51 to 54.1 for the highest and lowest antifeedant activity respectively. A dose-response experiment performed for those compounds giving FR values lower than 25, showed that the ryanoid cinnceylanone had the highest activity, followed by cinnceylanol, its isomer and ryanodol. A structure-activity relationship will be discussed.

Escoubas P., Fukushi Y., Lajide L. and Mizutani J. (1992). J. Chem. Ecol. 18: 1819.
González-Coloma A., Hernández M.G., Perales A. and Fraga B.M. (1990). J. Chem. Ecol 16: 2723

Oral Paper 9

HERBICIDAL AND ALLELOPATHIC EFFECTS OF TREE-OF-HEAVEN (AILANTHUS ALTISSIMA).
Rod M. Heisey, Pennsylvania State University, Schuylkill Haven, PA 17972 USA.

Ailanthus altissima is an introduced tree that is common in much of eastern United States. It is an extremely invasive species and is a very successful competitor that often colonizes abandoned fields, vacant city lots, roadsides, cracks in pavement, and other recently disturbed areas. I hypothesized allelochemicals may contribute to the invasive and competitive ability of A. altissima. Preliminary bioassays demonstrated that aqueous extracts of A. altissima leaves and bark strongly inhibited germination and growth of other plant species in vitro. Within the mature A. altissima tree, concentrations of the toxin(s) were highest in the bark of roots, trunk, and branches, but were low in the wood. Stemflow from intact A. altissima trees, however, was not phytotoxic. Pieces of A. altissima bark mixed with soil strongly inhibited the growth of indicator plants. Methanol extracts of A. altissima bark sprayed onto a range of weed and crop plants growing in soil in the greenhouse exhibited striking herbicidal effects. The phytotoxicity of the active compound(s) persisted for only a few days in the soil, however. The major herbicidal compound in A. altissima bark has now been isolated and is being characterized. Its identity, herbicidal properties, and potential for causing allelopathy will be discussed.

Oral Paper 10

BIOCHEMICAL ASPECTS OF SEQUESTRATION AND METABOLISM OF PLANT DERIVED PYRROLIZIDINE ALKALOIDS IN ARCTIIDS.
Thomas Hartmann, Andreas Biller and Michael Boppré. Institut für Pharmazeutische Biologie der Technischen Universität, D-38106 Braunschweig und Forstzoologisches Institut der Universität, D-79098 Freiburg, Germany

Arctiids such as *Tyria jacobaeae* and *Creatonotos transiens* are long known to store pyrrolizidine alkaloids (PAs) from their larval food plants. Tertiary PAs taken up from the guts are immediately N-oxidized by a soluble mixed functional NAD(P)H-dependent oxygenase localized in the hemolymph. This enzyme guarantees that PAs are safely stored in the body as polar untoxic N-oxides. The PA N-oxides are present in the hemolymph at relatively high levels. PA storing insects obviously lost the ability to excrete PAs. Depending on their chemical structure certain PAs are preferably hydrolyzed in the guts. The resulting necine base (e.g. retronecine) is taken up, N-oxidized, stored, and transformed into "insect PAs" such as creatonotine or callimorphine. This transformation is restricted to the very early stage of pupation. Wild-caught imagines of *C. transiens* were frequently found to store "insect PAs" as exclusive or dominating type of PAs. It is an open question whether the formation of the "insect PAs" is the consequence of a physiological need (safe storage of retronecine in pupae) or of chemoecological importance (salvage of a necine base into a biological active ester alkaloid).

Oral Paper 11

A PHAGOSTIMULANT FUNGAL METABOLITE FOR THE WESTERN SUBTERRANEAN TERMITE, RETICULITERMES HESPERUS BANKS. Brice A. McPherson, David L. Wood, and Isao Kubo, University of California, Berkeley, CA 94720 USA.

Methanolic extracts of fungi associated with colonies of R. hesperus in northern California were found to elicit feeding in laboratory assays. Feeding behavior was quantified by a novel assay which uses cellulose thin layer chromatography plates as the feeding substrate. Bioassay-directed fractionation using primarily size exclusion chromatography yielded a potent phagostimulatory component. The bioassays demonstrate a clear dose-dependent effect for the phagostimulant, which elicits feeding behavior at less than 10 micrograms per cm^2. Development of effective subterranean termite baits requires that the foragers exhibit preference for baits over other cellulosic materials. A phagostimulant active in the microgram range may provide the key to the creation of selective baits.

Oral Paper 12

Localization and Specificity of the Cyanoglucoside-producing System in the Larvae of *Zygaena trifolii* (Insecta:Lepidoptera).

Adolf Nahrstedt and Guido Holzkamp.
Institute of Pharmaceutical Biology and Phytochemistry
Westf. Wilhelms-University, D-48149 Muenster, Germany.

The larvae of the moth *Zygaena trifolii* accumulate the two cyanogenic glucosides linamarin and lotaustralin which they produce from the amino acids valine and isoleucine resp. Incubation experiments with various larval tissues have shown that the fat body is the site of biosynthesis. However, the isolated fat body does not form the intact glucosides but rather their direct precursors, the non-glucosylated cyanohydrins. N-Hydroxyvaline and N-hydroxyisoleucine and their corresponding aldoximes and nitriles (known as intermediates from plant biosynthesis) were tested as precursors for cyanohydrin production by the fat body. The N-hydroxyamino acids were converted to a very low extent (ca. 3-10 nmol/fat body/24h); the aldoximes were intermediate (ca.30-70 nmol/fat body/5h) and the nitriles were the best substrates (ca. 150-200 nmol/fat body/5h) reflecting their biogenetic sequence. Conversion of the *in vivo* precursors isoleucine and valine has not yet been detected in the isolated fat body. Some aliphatic linear and branched and even aromatic metabolites derived from different amino acids were also converted to cyanohydrins; thus the active enzymes are not specific for the intermediates derived from the endogenous precursors valine and isoleucine. Since only linamarin and lotaustralin are found in the intact larvae the question of the substrate specific step in the biosynthetic pathway is still open.

Oral Paper 13

RELATIVE UNPALATABILITY OF LEAF-BEETLES WITH EITHER BIOSYNTHESIZED OR SEQUESTERED CHEMICAL DEFENSE. M. Rowell-Rahier*, J.M. Pasteels§, A. Alonso-Mejia+ and L.P. Brower+. *Zoological Inst., Basel, CH-4051 Switzerland; §ULB, Brussels, B-1050 Belgium, +U. Florida, Gainesville, FL- 32611USA.

Defence of the leaf beetles from the genus *Oreina* is chemically diverse. Some (e.g. *O. gloriosa*) rely on the secretion in small quantity of highly concentrated mixture of cardenolides biosynthesised de novo and stored only in glands. Others (e.g. *O. cacaliae*), sequester, both in their body and in their glands, pyrrolizidine alkaloid N-oxides (PAs) from their host-plants.. First, we test the impact of the defensive secretion on the palatability of the beetles to birds (wild caught red-winged black birds, *Agelaius phoeniceus*). Secondly, we compare the effect of these two different types of chemicals to examine whether they grant equal protection to the beetles.

O. gloriosa ,with their intact secretion of cardenolides, were eaten in 68% of trials When the secretion was removed, 95% of the now undefended *O. gloriosa* were eaten. The handling time by a bird decreased significantly from 41 seconds to 26 seconds. This shows that the small quantity of highly concentrated secretion produced on the surface of the beetles body affords a protection against birds. *O. cacaliae* with PAs in their secretion and body were eaten in only 39% of the cases and, when the secretion was removed leaving only the PA content of the body for defence, in 46% of trials. The handling times for *O. cacaliae* with or without secretion are similar to those for *O. gloriosa* with or without secretions.

We conclude that beetle protection is better when it is defended by PAs rather than cardenolides and that the presence of secretion of either chemical class greatly increase the handling time by the birds, thus making the prey less "valuable".

Oral Paper 14

OVIPOSITION AND FEEDING DETERRENTS RESPONSIBLE FOR REJECTION OF *ERYSIMUM CHEIRANTHOIDES* (CRUCIFERAE) BY *PIERIS RAPAE*.

J.A.A.Renwick, K. Sachdev-Gupta, Xinpei Huang and C.D. Radke. Boyce Thompson Institute, Ithaca, NY 14853 USA

The wild crucifer, *Erysimum cheiranthoides* is rejected by ovipositing *Pieris rapae* butterflies, despite the presence of a stimulant. The compounds responsible for stimulant activity were identified as glucosinolates, glucoiberin and glucocheirolin, and two major deterrent compounds were identified as the cardenolides, erysimoside and erychroside. Feeding deterrents have also been detected in *E. cheiranthoides*, and several cardenolides were found to be responsible. However, the most active feeding deterrents are digitoxin-based cardenolides, whereas the oviposition deterrents are strophanthidin-based. The results suggest that *E. cheiranthoides* has developed two separate lines of defense against the different life stages of *P. rapae*.

Oral Paper 15

CATERPILLAR FEEDING PATTERN LINKED TO DETOXIFICATION ENZYME ACTIVITY. John I. Glendinning and Frank Slansky. Univ. of Florida, Gainesville, FL 32611 USA.

We tested the hypothesis that allelochemical processing by detoxification enzymes affects feeding behavior; for insects consuming food with a potentially toxic allelochemical, uninduced individuals with baseline enzyme activity should have reduced feeding compared with induced individuals having higher enzyme activity. We observed feeding by polyphagous fall armyworms (Spodoptera frugiperda) when fed diet with indole 3-carbinol (I3C), a toxic allelochemical in certain crucifers and a well-known inducer. Larvae were either pre-exposed to control diet (uninduced) or an I3C diet (induced). The I3C concentrations used (0.2 and 0.4% fm) were non-deterrent, eliminating the influence of peripheral chemoreception. Uninduced larvae fed I3C diet had significantly fewer feeding bouts and longer intervals between bouts, but feeding bout duration was unaffected. In contrast, induced larvae given I3C diet fed similarly to control larvae on diet lacking I3C. These results support the initial hypothesis. If the altered feeding pattern is an adaptive response in which larvae adjust feeding to match detoxification activity (rather than a passive consequence of post-ingestive toxicity), then these results suggest that feeding patterns may be set by the rate of allelochemical detoxification rather than, as generally assumed, by the rates of nutrient digestion and absorption.

Oral Paper 16

FALL ARMYWORM SENSITIVITY TO FLAVONE: LIMITED ROLE OF CONSTITUTIVE AND INDUCED DETOXIFYING ENZYME ACTIVITY . G. S. Wheeler, F. Slansky, JR., AND S. J. Yu. Department of Entomology and Nematology, University of Florida, PO Box 110620, Gainesville, FL USA 32611-0620

We used inhibition and induction of detoxifying enzymes to determine whether these enzymes allow a generalist species (Spodoptera frugiperda; fall armyworms) to cope with ingestion of the flavonoid, flavone. Flavone induces polysubstrate monooxygenases (PSMO), general esterases (GE) and glutathione S-transferases (GST) in S. frugiperda, yet this species is affected deleteriously by low dietary concentrations of this allelochemical. First, in a series of experiments, larvae were fed artificial diets containing increasing concentrations of flavone, either alone or with known inhibitors of either PSMO, GE or GST enzymes. In an additional treatment, flavone and inhibitors of all three enzyme systems were administered in diets simultaneously. Significant synergism of flavone's growth-reducing activity occurred at the highest concentration tested (0.125% fresh mass, fm) when the PSMO inhibitor, piperonyl butoxide, or the GST inhibitor, diethyl maleate, was added to the diet, and at 0.08% fm flavone, when combined with the GE inhibitor, tri-tolyl phosphate. In many cases, however, the additive effect (i.e., reduction in growth owing to flavone alone + inhibitor alone) was greater than the synergistic effect. In the second approach, caterpillars were pre-exposed to a concentration of flavone (0.02% fm) that induced these enzymes ca. 1.5 to 2.5-fold, prior to switching larvae to a diet containing a higher (growth-reducing) flavone concentration (0.125% fm). The relative growth rates (RGR) of induced larvae were significantly greater (14%) than those of the uninduced larvae on the 0.125% fm flavone diet. The variable responses to inhibitor treatment and the relatively small benefit of enzyme induction suggest that these enzyme systems have minimal impact on the detoxification of flavone in S. frugiperda, even though this allelochemical induces enzyme activity and has been reported to be metabolized in vitro.

10th ANNUAL MEETING
International Society of Chemical Ecology
Tampa/Clearwater Florida
July 31-August 4, 1993

Contributed Papers

Session III (Monday - August 2)

Terpenoids

Island No. 1 Room E. David Morgan Presiding

2:00 - 2:15 pm ***Oral Paper* 17** THE CHEMICAL AND NUTRITIONAL ECOLOGY OF FOUR SIMPLE-STOMACHED PRIMATE SPECIES IN KIBALE FOREST, UGANDA: Nancy Lou Conklin and Richard W. Wrangham.

2:15 - 2:30 pm ***Oral Paper 18*** HIGH SPEED MULTILAYER COIL COUNTER CURRENT CHROMATOGRAPHY (MLCCC): A MILD ISOLATION METHOD FOR SENSITIVE, BIOLOGICALLY ACTIVE LIMONOIDS FROM NEEM SEEDS. Hans E. Hummel, Y. Ito, and H. Oka.

2:30 - 2:45 pm ***Oral Paper 19*** ALLELOPATHIC EFFECTS OF BRASSICA ROOT EXUDATES AND ISOTHIOCYANATES ON THE TRIPARTITE LEGUME SYMBIOSIS. Robert A. Kluson.

2:45 - 3:00 pm ***Oral Paper 20*** TOXIFICATION OF SECONDARY PRODUCTS BY THE DETOXIFICATION SYSTEM: CHROMENE-METABOLISM IN SPODOPTERA LITTORALIS. Arno Kunze, Luise Narang, Claudia Grimm, Ludger Witte, and Peter Proksch.

3:00 - 3:15 pm ***Oral Paper 21*** R-(-)-LINALOOL, A MINOR SEX PHEROMONE COMPONENT OF THE SCARAB BEETLE *HOLOTRICHIA PARALLELA*. Walter Soares Leal, Shigeru Matsuyama, Masaaki Sawada, Yasumasa Kuwahara, Makoto Hasegawa.

3:15 - 3:30 pm ***Oral Paper 22*** NEW BIOACTIVE HELIANNUOLS FROM CULTIVAR SUNFLOWER VARIETIES, SH-222® AND VYP®. Francisco A. Macías, Rosa M. Varela, José M. G. Molinillo and Ascensión Torres.

3:30 - 3:45 pm **COFFEE BREAK**

3:45 - 4:00 pm ***Oral Paper 23*** PHOTO-OXIDATION PRODUCTION OF INSECT-ANTIFEEDANT TRITERPENOIDS FROM NEEM SEEDS. E. David Morgan and Shaun Johnson.

4:00 - 4:15 pm ***Oral Paper 24*** SEQUESTRATION OF HIGHLY TOXIC TERPENOIDS BY ERICACEAE-FEEDING GEOMETRID MOTHS. Ritsuo Nishida.

4:15 - 4:30 pm ***Oral Paper 25*** BIOASSAY DEVELOPMENT FOR THE ATTRACTION OF A PARASITOID, *APROSTOCETUS HAGENOWII*, TO VOLATILES EMANATING FROM AMERICAN COCKROACH OOTHECAE. Daniel R. Suiter, Dave A. Carlson, Philip G. Koehler and Richard S. Patterson.

4:30 - 4:45 pm ***Oral Paper 26*** FLORAL FRAGRANCES OF ORCHIDS POLLINATED BY MALE EUGLOSSINE BEES. W. Mark Whitten and Norris H. Williams.

5:30 - 6:30 pm **Business Meeting - Beach Gulf Room**

Oral Paper 17

THE CHEMICAL AND NUTRITIONAL ECOLOGY OF FOUR SIMPLE-STOMACHED PRIMATE SPECIES IN KIBALE FOREST, UGANDA. Nancy Lou Conklin and Richard W. Wrangham. Harvard University, Cambridge, MA 02138 USA.

The potential nutritional value, based on chemical analysis, and the tannin, alkaloid and terpenoid content of fruits consumed by three sympatric monkey species are presented and compared to those of the fruits consumed by the sympatric chimpanzee community. A field project performing simultaneous feeding observations on the four primate species is currently underway at Makerere University Biological Field Station, Kibale Forest, Uganda. Preliminary results are that monkeys eat smaller fruits, containing higher levels of terpenoids but which are also higher in protein, and/or lipid compared to the higher fiber foods of chimpanzees. All four animal species consume some fruits containing alkaloids but prefer different alkaloid-containing plant species. The chimps select for higher simple sugar content than do the monkeys. All four animal species select against condensed tannins, but select less strongly against hydrolyzable tannins. The chemical results are considered along with the plant phenology to draw conclusions regarding diet choice.

Oral Paper 18

HIGH SPEED MULTILAYER COIL COUNTER CURRENT CHROMATOGRAPHY (MLCCC) : A MILD ISOLATION METHOD FOR SENSITIVE, BIOLOGICALLY ACTIVE LIMONOIDS FROM NEEM SEEDS. Hans E. Hummel, Y.Ito,and H.Oka; Justus-Liebig-University of Giessen,Germany; National Institute of Health,Bethesda,Md.,USA; Aichi Prefectural Institute of Public Health,Nagoya,Japan.

Neem seeds (Azadirachta indica :Meliaceae) contain a large number of tetranortriterpenoids with well documented insect antifeedant (salannin) and metamorphosis inhibiting properties (azadirachtin,$C_{35}H_{44}O_{16}$). Isolation of quantities large and inexpensive enough for at least small scale field trials against pest insects remains problematic. We here report on experiments with various MLCCC configurations capable of purifying crude (10-15%) neem extracts and obtaining,in one step, within 2.5 hours and without losses, solid amorphous azadirachtin samples with a minimum of 96% purity and with full biological activity (EC_{50} at 0.76 ug/g) in the sensitive Epilachna varivestis bioassay system. The (not yet optimized) two-phase solvent system hexan/ethylacetate/methanol/water = 3:5:3:5 (v/v) was employed and yielded 300-400 mg azadirachtin per run.

Some recent progress towards reducing physiological fitness including diminished sensitivity to react to host plant kairomones in the economically highly important Diabrotica virgifera virgifera beetles (Chrysomelidae) is being presented.
Prospects of the method toward characterization and isolation of other natural products interesting to chemical ecologists are most inviting.

Oral Paper 19

ALLELOPATHIC EFFECTS OF BRASSICA ROOT EXUDATES AND ISOTHIOCYANATES ON THE TRIPARTITE LEGUME SYMBIOSIS. Robert A. Kluson. University of South Florida, Tampa, Florida 33620 USA.

The allelopathic potential of root exudates of cabbage (Brassica oleraceae) was tested on nodulation, nitrogen fixation activity, and infection of vesicular-arbuscular mycorrhizae (VAM) for the tripartite legume symbiosis of Phaseolus-Rhizobium-Glomus. In studies of beans and cabbage grown together in Leonard jars, cabbage root exudates reduced all symbiotic responses, as well as bean biomass. However, in studies using the root exudate recirculating system no effects on these symbiotic responses were observed. Collections and HPLC analysis of cabbage root exudates demonstrated the presence of 2-phenyl-ethyl isothiocyanate. Bioassays with this allelochemical demonstrated that it is biologically active to the individual symbionts of the tripartite. Dosage responses elucidated relevant mechanisms involved in the observed allelopathy.

Oral Paper 20

TOXIFICATION OF SECONDARY PRODUCTS BY THE DETOXIFICATION SYSTEM: CHROMENE-METABOLISM IN *SPODOPTERA LITTORALIS*. Arno Kunze[1], Luise Narang[1], Claudia Grimm[1], Ludger Witte[2] and Peter Proksch[1]. [1]= Julius-von-Sachs-Institut für Biowissenschaften, D-W-8700 Würzburg, FRG; [2]= TU Braunschweig, D-W-3300 Braunschweig, FRG.

Chromenes occur frequently in species of the sunflower family *Asteraceae* (1). Precocene II, first isolated from *Ageratum houstonianum*, is known for its antijuvenile hormone activity towards several insects. Other chromene derivatives like encecalin, demethoxyencecalin and eupatoriochromene show no antijuvenile hormone activity, but are toxic to insects following topical contact as well as oral uptake (2). We investigated the toxicity of these four chromene derivatives towards the polyphagous pest insect *Spodoptera littoralis*. Precocene II was the most toxic compound, followed by encecalin. The other two acetylchromenes showed only marginal insecticidial activity. Metabolism of chromenes in *Spodoptera littoralis* occurs mainly in the midgut, the major source of MFO's. Isolated midguts were used as enzyme source for our *in-vitro* experiments on the metabolism of chromenes. All chromene derivatives investigated gave rise to the 3,4-diol as major metabolite. The diol is the hydrolysis product of the 3,4-epoxid, which is responsible for the cytotoxic effects of chromenes. The abundance of the diol, generated during *in-vitro* studies, was found to correlate well with the toxicity of the following topical application to larvae of *Spodoptera littoralis*. The toxic effects of precocene II or encecalin could be eliminated in the presence of tetcyclasis, a powerful MFO-inhibitor. The metabolism of precocene II or encecalin in the pest *Spodoptera littoralis* is another example for toxification of secondary plant products by the detoxification system of animals.

Acknowledgements: Financial support by the DFG is gratefully acknowledged.

References: (1) P. Proksch and E. Rodriguez (1983) *Phytochemistry* **22**, 2335-2348.
(2) M. Randriamihaly et al. (1992) *Biochem. System. Ecol.* **20**, 711-722.

R-(-)-LINALOOL, A MINOR SEX PHEROMONE COMPONENT OF THE SCARAB BEETLE *Holotrichia parallela*. Walter Soares Leal (National Institute of Sericultural and Entomological Science, Tsukuba, Ibaraki 305 Japan), Shigeru Matsuyama (Tsukuba University), Masaaki Sawada (Chiba Prefectural Agricultural Experiment Station), Yasumasa Kuwahara (Kyoto University), and Makoto Hasegawa (Chiba Pref. Agric. Exp. Stn.)

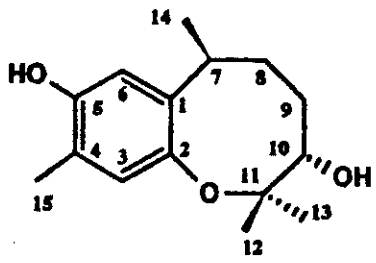
R-(-)-Linalool was identified as a minor component in the sex pheromone of the scarab beetle *Holotrichia parallela*. Enantiomeric resolution of racemic linalool was achieved on a GC chiral capillary column and the absolute configuration of the natural product determined. Field assays revealed that, although not attractive per se, R-(-)-linalool enhanced the attractiveness of the major sex pheromone, L-isoleucine methyl ester (LIME). Analyses of the pheromone titers in the glands of field-collected females demonstrated the occurrence of peak levels of LIME with a 48-hr ("circabidian") periodicity. The levels of LIME in the glands of 45-day-old virgin females increased over three times, from the scoto- to the photophase of a calling day, but the amounts of (R)-linalool did not significantly change.

Oral Paper 22

NEW BIOACTIVE HELIANNUOLS FROM CULTIVAR SUNFLOWER VARIETIES, SH-222® AND VYP®.

Francisco A. Macías, Rosa M. Varela, José M. G. Molinillo and Ascensión Torres.
Departamento de Química Orgánica, Facultad de Ciencias, Universidad de Cádiz, Apdo. 40, 11510 Puerto Real, Cádiz, Spain.

The study of bioactive fractions of cultivar sunflowers (*Helianthus annuus* L.) varieties, SH-222® and VYP® afforded two new sesquiterpenes containing a novel sesquiterpene skeleton named heliannuol, heliannuol **A** and **B**. In continuation of our systematic allelopathic activity studies on cultivar varieties of *H. annuus*, we present the isolation and structural elucidation of two new related sesquiterpenes, heliannuol **C** and **D**.



Heliannuol D

Heliannuol **A** and **D** are epimers at C-10, but the conformational equilibrium observed in heliannuol **A** at room temperature is not shown by heliannuol **D**. Computational chemical methods (molecular orbital calculation, PM3) could provide the quantitative evaluation of the conformational and electronic properties. A biogenetic sequence, that involved a phenonium ion is proposed on the biogenesis of heliannuol **C**. A structure-activity correlation is discussed based on the allelopathic bioassay results.

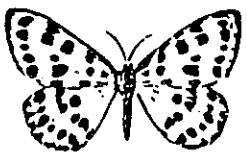
Oral Paper 23

PHOTO-OXIDATION PRODUCTION OF INSECT-ANTIFEEDANT TRITERPENOIDS FROM NEEM SEEDS. E. David Morgan and Shaun Johnson, Chemistry Department, Keele University, Staffordshire, England ST5 5BG

Photooxidation with oxygen in UV light of three major triterpenoids from the seeds of the Neem tree *(Azadirachta indica)* has been studied to learn about the degradation of insect-antifeedant properties of Neem in storage and in agricultural use. Nimbin gives a mixture of products not yet fully identified. Salannin gives three unstable oxides, all of which on standing or chromatography isomerize to a single hydroxylactone. Azadirachtin gives one photo-isomerized product but largely decomposes to polar products. The use of HPLC-NMR is being investigated for the study of these triterpenoids.

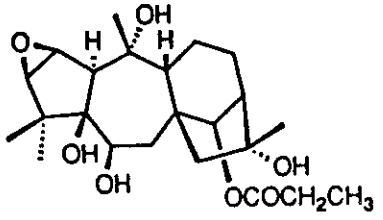
Oral Paper 24

SEQUESTRATION OF HIGHLY TOXIC TERPENOIDS BY ERICACEAE-FEEDING GEOMETRID MOTHS



Ritsuo Nishida Pesticide Research Institute, Kyoto University, Kyoto, 606-01 Japan

Larvae of the geometrid moth, *Arichanna gaschkevitchii*, (Lepidoptera) feed selectively on young leaves of *Pieris japonica* (Ericaceae), which is known to contain a group of highly toxic diterpenes called grayanoids. The adult moths were found to sequester a series of the host-originated toxins in the body tissues. The compounds were characterized as asebotoxins I and IV, grayanotoxin III, rhodojaponin III, kalmitoxin I and two new analogs, arichannatoxins I and II (total content: 300 μg/moth). The most predominant component, asebotoxin I (200 μg/moth), strongly deterred feeding of house lizards at low doses, which suggested a defensive role against predatory animals. The grayanoid diterpenes were also detected from bodies of a closely related species, *Arichanna melanaria*. Since these two diurnal species exhibit very similar aposematic wing patterns with bright orange-colored hind-wings and appear sympatrically during summer in central Japan, they are regarded as Müllerian mimics.



Arichannatoxin I

Oral Paper 25

BIOASSAY DEVELOPMENT FOR THE ATTRACTION OF A PARASITOID, *Aprostocetus hagenowii*, TO VOLATILES EMANATING FROM AMERICAN COCKROACH OOTHECAE. Daniel R. Suiter, Dave A. Carlson, Philip G. Koehler and Richard S. Patterson. Department of Entomology & Nematology, University of Florida, Gainesville, FL 32611-0620 USA.

The blattid oothecal parasitoid, *Aprostocetus hagenowii*, is attracted to oothecae of the American cockroach, *Periplaneta americana*, by volatile kairomones emanating from the host itself. A Y-tube bioassay has been developed to aid in the isolation of the kairomone(s) responsible for this attraction. The percentage of wasps responding increased with flow rate; greater than 95% of wasps chose the ootheca host.

Oral Paper 26

FLORAL FRAGRANCES OF ORCHIDS POLLINATED BY MALE EUGLOSSINE BEES. W. Mark Whitten and Norris H. Williams. Florida Museum of Natural History, Univ. of Florida, Gainesville, FL 32611 USA.

Floral fragrances of 374 species or chemotypes of orchids were analyzed by GC/MS, with emphasis on species pollinated by male euglossine bees (Apidae: Euglossini). These orchid species were compared with those belonging to other pollination syndromes (non-euglossine bees; hawkmoths). Orchid fragrances are chemically diverse, containing over 350 compounds (>0.1%) in the 374 taxa examined. Fragrances of male euglossine-pollinated plants are: 1. usually produced in large quantities over short periods of time; 2. range from simple to complex (related to pollinator specificity and number of sympatric congeners); 3. rich in oxygen-containing monoterpenes, sesquiterpenes, and simple aromatics (esters, alcohols, ketones, or epoxides). Flowers pollinated by other types of bees often produce fragrances dominated by nerol, geraniol, ocimene, and alpha-farnesene. Moth-pollinated taxa also have diverse fragrances, but many hawkmoth-pollinated species contain large amounts of trans-nerolidol and linalool.

10th ANNUAL MEETING
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Tampa/Clearwater Florida
July 31-August 4, 1993

Contributed Papers

Session IV (Monday - August 2)

Pheromones

Beach Gulf Room **Abraham Hefetz Presiding**

2:00 - 2:15 pm ***Oral Paper 27*** SAMPLING OF HONEYBEE POPULATIONS FOR CHEMOSYSTEMATIC ANALYSIS OF PHEROMONES. Robin M. Crewe.

2:15 - 2:30 pm ***Oral Paper 28*** THE ROLE OF THE POSTPHARYNGEAL GLAND SECRETION IN NESTMATE RECOGNITION IN ANTS. Abraham Hefetz, Elise Nowbahari, and Christine Errard.

2:30 - 2:45 pm ***Oral Paper 29*** ENVIRONMENTAL AND PHEROMONAL CONTROL OF SPAWNING AND REPRODUCTIVE BEHAVIOUR OF MARINE POLYCHAETES. J. D. Hardege.

2:45 - 3:00 pm ***Oral Paper 30*** EFFICIENT AND STEREOSPECIFIC SYNTHESIS OF SUSPENSOLIDE --THE SEX PHEROMONE OF THE CARIBBEAN FRUIT FLY AND THE MEXICAN FRUIT FLY. A. C. Oehlschlager and Ginna Hidlago.

3:00 - 3:15 pm ***Oral Paper 31*** THE LARVAL SECRETION OF CHRYSOMELA LAPPONICA (COLEOPTERA, CHRYSOMELIDAE). Monika Hilker and Stefan Schulz.

3:15 - 3:30 pm ***Oral Paper 32*** CHEMICAL ECOLOGY OF THE PALM WEEVIL RYNCHOPHORUS PALMARUM (L) (CLEOPTERA: CURCULIONIDAE): ATTRACTION TO HOST PLANTS AND TO A MALE PRODUCED AGGREGATION PHEROMONE. K. Jaffee., P. Sanchez, H. Cerda, H.V. Hernandez, R. Jaffee, N. Urdaneta, G. Guerra, R. Martinez and B. Miras.

3:30 - 3:45 pm **COFFEE BREAK**

3:45 - 4:00 pm ***Oral Paper 33*** SIZE, SHAPE, AND A MILIEU OF PHEROMONES -- WHAT IS GOVERNING NEST LOCATION IN HONEY BEES? Justin O. Schmidt.

4:00 - 4:15 pm ***Oral Paper 34*** VOLATILE COMPOUNDS FROM SCENT ORGANS OF TROPICAL BUTTERFLIES. Stefan Schulz.

4:15 - 4:30 pm ***Oral Paper 35*** QUANTIFICATION OF ELECTROANTENNOGRAM RESPONSES OF THE PRIMARY RHINARIA OF ALATE *ACYRTHOSIPHON PISUM* (HARRIS) (HOMOPTERA: APHIDIDAE). W. A. van Giessen, H. W. Fescemyer and J. K. Peterson.

4:30 - 4:45 pm ***Oral Paper 36*** NOVEL HOST HOST PLANT MARKERS IN EGG-LAYING BUTTERFLIES. Joop J. A. van Loon, Louis M. Schoonhoven, Anton Blaakmeer, Teris A. van Beek, AEede de Groot.

4:45 - 5:00 pm ***Oral Paper 37*** FIELD TRAPPING OF THE TOMATO WORM, SCROBIPALPULOIDES ABSOLUTA (LEPIDOPTERA, GELECHIIDAE), USING VIRGEN FEMALES. Evaldo F. Vilela and Manoel A. U. Fernandes.

5:00 - 5:15 pm ***Oral Paper 38*** BIOSYNTHETIC STUDIES OF CONTACT PHEROMONES OF DROSOPHILA MELANOGASTER. Maryse Pennanec'h and Jean-Marc Jallon.

5:30 - 6:30 pm **Business Meeting - Beach Gulf Room**

Oral Paper 27

SAMPLING OF HONEYBEE POPULATIONS FOR CHEMOSYSTEMATIC ANALYSIS OF PHEROMONES Robin M. Crewe. University of the Witwatersrand, Johannesburg, 2050 South Africa.

Sampling of honeybee populations for chemosystematic analysis of their pheromones, particularly their mandibular gland pheromones, has traditionally been carried out by collection of the bee material in the field, followed by immediate processing into an appropriate solvent for extraction. However, with the advent of solvent free injection techniques, it is necessary either to keep the experimental bees alive until they can be analysed or to use collection technique that preserves the pheromone signal until the analysis can be done. Where bees of the same age have to analyzed, keeping them alive until analysis is not feasible.

A sample technique will be described that resolves these difficulties. The bees are anacsthetising with N_2 gas, followed by immersion in liquid nitrogen. The stored material can be analysed after a period of storage of at least two weeks. Comparison of the GC analyses of stored material with that which was freshly prepared indicate that there is no significant loss of material during storage.

Oral Paper 28

THE ROLE OF THE POSTPHARYNGEAL GLAND SECRETION IN NESTMATE RECOGNITION IN ANTS. Abraham Hefetz, Elise Nowbahari, & Christine Errard. Department of Zoology, Tel Aviv University, Israel, and Laboratory of Comparative ethology, University of Paris, France.

Recent chemical and behavioral data suggest that the postpharyngeal glands are the source of the hydrocarbons that are responsible for nestmate recognition in *Cataglyphis niger*. The glandular secretion constitutes a complex mixture of hydrocarbons, that is congruent with the cuticular hydrocarbon profile, but with little quantitative difference.
Alien ants that are encountered in a neutral arena exhibit strong mutual aggression. However, ants that encounter alien ants that are smeared with the postpharyngeal secretion of their nestmates behave non-aggressively towards them. Moreover if we encounter two nestmates, one of which is smeared with alien postpharyngeal glands, the non treated ant exhibits aggression towards her nestmate.
The ability to create mixed species groups of *Manica rubida* and *Formica selysi* is apparently related to the acquisition of the heterospecific compounds, thus changing the label by which they are recognized. Analyses of the postpharyngeal glands of members of such groups revealed the presence of the heterospecific compounds in their secretion. This was true even if the group was composed of a single *F. selysi* and 20 workers of *M. rubida*. Whether these heterospecific compounds are transferred between the ants by trophalaxis or biosynthesized *de novo* remains to be clarified

Oral Paper 29

ENVIRONMENTAL AND PHEROMONAL CONTROL OF SPAWNING AND REPRODUCTIVE BEHAVIOUR OF MARINE POLYCHAETES

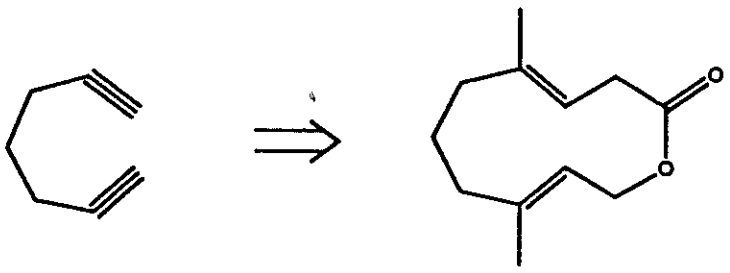
J.D. HARDEGE, University of St. Andrews, Gatty Marine Laboratory, KY16, 8LB, Scotland

A number of marine polychaetes, e.g. the *Nereidae*, undergo a somatic metamorphosis to become mature individuals before reproduction and die afterwards. Synchronization of spawning at the populationary level is achieved by endocrine factors , temperature regimes or lunar periodicity. These factors may also act to synchronize gamete release between individuals, hormones and pheromones are the main cues in the transduction of the environmental information. In marine nereid polychaete worms, e.g. *Platynereis dumerilii*, volatile sex pheromones such as 5-methyl-3-heptanone acts as aphrodisiacs, arrestant semiochemicals or provide sex determination via the use of sex specific enantiomers of the ketone in long range chemical communication during the reproductive process known as the "nuptial dance". Water soluble, non-volatiles functions in short range communication, e.g. the induction of the release of the gametes of the sexual partners. Sex pheromones are also involved in species recognition and reproductive isolation in closely related nereid species *P.dumerilii* and *Nereis succinea* which are reproducing in the same area during summer nights. Nevertheless interspecific activity of the gamete release inducing pheromones also occur in a number of nereid species. Recent investigations have focused on annual reproducing specimens such as *Arenicola marina* and *Nephtys hombergii*.

Oral Paper 30

EFFICIENT AND STEREOSPECIFIC SYNTHESIS OF SUSPENSOLIDE - THE SEX PHEROMONE OF THE CARFIBBEAN FRUIT FLY AND THE MEXICAN FRUIT FLY Ginna Hidlago and A. C. Oehlschlager, Department of Chemistry, Simon Fraser University, Burnaby, B. C. V5A 1S6 CANADA.

The Caribbean fruit fly, *Anastrepha suspensa*, Loew. and the Mexican fruit fly, *A. ludens*, Loew. infest many tropical and subtropical fruits in Central and South America. Suspensolide (E,E-4,8-dimethyl-3,8-decadien-10-olide, **1**) is a major component fo the sex pheromone released by the males of both species. Previous syntheses of **1** have involved low yield preparation of mixtures of the 3Z,8E and 3E,8E hydroxy acid precursors, their separation and cyclization. We report an 8 step high (7% overall) yield preparation of **1** based on carbometallation of **2**.



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Oral Paper 31

THE LARVAL SECRETION OF CHRYSOMELA LAPPONICA (COLEOPTERA, CHRYSOMELIDAE). Hilker, Monika[1] & Schulz, Stefan[2]. [1]Lehrstuhl für Tierökologie II, Universität Bayreuth, Germany; [2]Institut für Organische Chemie, Universität Hamburg, Germany.

The defensive secretion of *Chrysomela lapponica* larvae, which is produced by nine pairs of exocrine dorsal glands, has been chemically analyzed. *C. lapponica* larvae were kept in the laboratory either on leaves of birch (*Betula pendula*), or alder (*Alnus glutinosa*), or willow (*Salix fragilis*). Larvae on birch and willow developed normally, whereas those on alder died within a few days. GC-MS analyses of the secretion of larvae on birch and willow revealed that the composition of this secretion differs distinctly from the known ones of several other *Chrysomela* species. In the exocrine secretion of larvae on birch numerous compounds were identified, which included the main components isobutyric acid, 2-methylbutyric acid, and esters of the two. Alcoholic components of the main esters are 8-hydroxylinalool, 1,3-hexanediol, benzylalcohol and 2-phenylethylalcohol. These linalool- and diolesters are new natural components. Glycosides of 8-hydroxylinalool are known as constituents of birch leaves. Of the secretion components of the larvae feeding on birch, several were also found in the secretion of larvae feeding on willow. A striking difference between both secretions is the presence of saligenin as a main component in the secretion of the larvae feeding on willow, whereas saligenin is absent in the secretion of the larvae on birch. *C. lapponica* obviously acquires saligenin by hydrolysis of salicin from willow leaves. However, in contrast to other *Chrysomela* species, *C. lapponica* larvae oxidize only traces of saligenin to salicylaldehyde. The repellent activity of single authentic compounds of the secretion of larvae feeding on birch and willow, respectively, was tested in laboratory bioassays with ants (*Myrmica sabuleti*). Pathways of biosynthesis of the secretion compounds are suggested and discussed under evolutionary and functional aspects.

Oral Paper 32

CHEMICAL ECOLOGY OF THE PALM WEEVIL Rynchophorus palmarum (L) (CLEOPTERA: CURCULIONIDAE): ATTRACTION TO HOST PLANTS AND TO A MALE PRODUCED AGGREGATION PHEROMONE Jaffee, K., P. Sanchez, Cerda, H., H.V. Hernandez, R. Jaffee, N. Urdaneta, G. Guerra, R. Martinez and B. Miras. Universidad Simon Bolivar, Apartado Postal 89000, Caracas 1080A, and Fondo Nacional de Investigaciones Agropecuarias, Venezuela.

Attraction to host plants by adult R.palmarum was studied in the field and in the laboratory. Stems of coco palms and pineapple fruits revealed the presence of ethanol and ethyl-acetate and of pentane, hexanal and isopentanol in coco stems. In the olfactometer, the first two compounds and isoamyl-acetate were attractive to the insects and the last three compounds, although not attractive by itself, increased the attractiveness of a mixture of the first two compounds. Mixtures of these compounds in proportions similar to the one occurring in attractive plant tissue, were as attractive as natural coconut tissue. In the field the chemical compounds either presented alone or as a mixture attracted weevils at a short distance. Males produce an aggregation pheromone when smelling ethyl-acetate. Rhynchophoral, the known active component of the pheromone, attracts weevils from long distances. In the field only mixtures of Rhynchophorol and plant odours attract large numbers of weevils.

Oral Paper 33

SIZE, SHAPE, AND A MILIEU OF PHEROMONES -- WHAT IS GOVERNING NEST LOCATION IN HONEY BEES? Justin O. Schmidt. Carl Hayden Bee Research Center, USDA, ARS, 2000 East Allen Road, Tucson, AZ 85719 USA.

For long-term survival and reproduction, honey bees must locate a safe and reliable nest cavity. Microenvironment, size, and location are important physical factors affecting suitability of cavities, and pheromones coordinate the processes of selection, communication, and movement of reproductive swarms to cavities. Honey bees operate according to two hierarchies during nest seeking activities: one relates to physical condition and properties of the potential nest cavity; the other relates to pheromonal and olfactory cues that mediate the behavioral processes. Experimental designs were devised to simultaneously determine which physical and pheromonal factors were governing honey bee choice and behavior. Size was shown to be important in temperate honey bees, but not in tropical honey bees. Nasonov pheromone plays the dominant role in communication, and, in specific, is the single long-range attractant pheromone of honey bees. The confusion about the possible role of queen pheromone was clarified: queen pheromone acts only as a short-range pheromonal cue that simply conveys information about the location of the queen. The confusion about its role in swarming arose from the mistaken assumption that queen pheromone attracts swarms to cavities; when, in actuality, queen pheromone in a cavity causes swarm clustering by bees in response to "discovery" of their lost queen, rather than to nest discovery.

Oral Paper 34

VOLATILE COMPOUNDS FROM SCENT ORGANS OF TROPICAL BUTTERFLIES. Stefan Schulz, Institut für Organische Chemie, Universität Hamburg, 20146 Hamburg, Germany.

The behavior of satyrid butterflies have only rarely been studied. Males of *Elymnias thryallis*, a satyrid from Papua New-Guinea, possess an abdominal scent gland which secretes a fresh smelling liquid. The main component could be identified by GC/MS and chemical derivatization to be a new, linalool derived, furanoid monoterpene, elymniafuran.

The tropical, new world *Heliconius* butterflies are known for their elaborate chemical communication systems. Males of *H. melpomene* mark their females chemically during copulation which makes the females unattractive for other males. (*E*)-β-Ocimene is transferred from the males to the females and acts as a repellent for other males. Feeding of the males with 2-^{13}C-acetate yields ocimene enriched in ^{13}C (10 %), thus indicating that the butterflies are able to synthesize the pheromone *de novo*, despite the widespread occurrence of ocimene in plants.

A different strategy is used by some ithomiine butterflies. The absolute configuration of the male pheromone 2-hydroxy-2-(1-hydroxyethyl)-3-methyl-γ-butyrolactone (Ithomiolide A), has been determined by synthesis of enantiomers, and comparison with natural samples by GC/MS on a chiral phase. The natural product has (2*S*,3*R*,1'*S*)-configuration. The lactone is formed by the butterflies from the natural pyrrolizidine alkaloid lycopsamine which exhibits identical configurations on the stereocenters common to both compounds. The butterflies obtain lycopsamine together with other pyrrolizidine alkaloids from their foodplants.

Oral Paper 35

QUANTIFICATION OF ELECTROANTENNOGRAM RESPONSES OF THE PRIMARY RHINARIA OF ALATE *ACYRTHOSIPHON PISUM* (HARRIS) (HOMOPTERA: APHIDIDAE).

W. A. van Giessen[1,2], H. W. Fescemyer[3], and J. K. Peterson[1]. [1]USDA-ARS, U.S. Vegetable Laboratory, Charleston, SC 29414-5334. [2]Clemson University, Dept. of Plant Pathology and Physiology, Clemson, SC 29634-0377. [3]Clemson University, Dept. of Entomology, Clemson, SC 29634-0377.

Electroantennograms (EAGs) of the proximal and distal primary rhinaria (respectively DPR and PPR) were recorded from excised antennae of alate virginoparous pea aphids, *Acyrthosiphon pisum* (Harris)(Homoptera: Aphididae). Primary unsaturated alcohols and aldehydes with varying carbon length (C3 to C8) were used as volatile test stimuli. EAGs were recorded from the DPR and PPR separately through the use of sectional electroantennography. Stimulus-response curves were fitted to a (sigmoid shaped) logistic equation. Differences in the EAG response curves of the DPR and PPR to volatile stimuli were characterized by estimating five parameters that were derived from this logistic equation and relate to particular characteristics of sigmoid curves, like saturation (maximum) EAG response (R_s), EAG response at 50% of R_s (CR_{50}), tolerance ($T_{0.9}$), threshold EAG response (CR_1), and EAG response area (A_R). The EAG response area, A_R, showed the largest separation between EAG responses of the DPR and PPR to the two homologous groups and between compounds with varying carbon chain length. The DPR was significantly more responsive to alcohols than to aldehydes, while the reverse was true for the PPR, showing a basic difference between DPR and PPR. The highest overall responses were elicited by 1-hexanol, hexanal and heptanal.

Oral Paper 36

NOVEL HOST HOST PLANT MARKERS IN EGG-LAYING BUTTERFLIES.

Joop J.A. van Loon, Louis M. Schoonhoven, Anton Blaakmeer[&], Teris A. van Beek[&], Æede de Groot[&]. Department of Entomology, Agricultural University, P.O. Box 8031,6700 EH Wageningen, The Netherlands. [&]Department of Organic Chemistry, Phytochemical Section, Agricultural University, Dreijenplein 8, 6703 HB Wageningen, The Netherlands

The large white butterfly, *Pieris brassicae* L., a herbivorous pest of crucifers, produces egg-associated chemical markers that inhibit its oviposition. Here we report on the identification of the marker compounds. Separation by means of reversed-phase HPLC demonstrated the presence of 3 active substances. These were identified using mass- and NMR-spectroscopy and chemical synthesis as trans-2-[3-(3,4,5-trihydroxy-phenyl-propenoyl)-amino]-3,5-dihydroxy-benzoic acid and two derivatives. These compounds form a novel group of chemicals that have not been reported before. Two related *Pieris* species produce the same chemicals, while these compounds could not be detected from six other lepidopterous species, including two other species of Pieridae. This is the first report of genus-specific chemicals affecting butterfly oviposition behavior. Site of production is the female accessory gland that contains substantial amounts of a glycoside form which is secreted onto the egg surface during oviposition. The availability, stability and inhibitory action on colonisation of cabbage plants by butterflies make application of these compounds in the protection of cabbage crops feasible and compatible with other environmentally sound crop protection strategies.

Oral Paper 37

FIELD TRAPPING OF THE TOMATO WORM, SCROBIPALPULOIDES ABSOLUTA (LEPIDOPTERA, GELECHIIDAE), USING VIRGEN FEMALES. Manoel A. U. Fernandes and Evaldo F. Vilela. Federal University of Viçosa,Viçosa, MG 36570-000 Brazil.

During the last ten years, **S. absoluta** became one of the most serious tomato pest in Latino America. Field studies using virgen females baited traps were carried out to evaluate males catches, as the sex pheromone is yet to be identified. Six different trap heights, six distinct colors and seven trap designs were evaluated. It was found that the height of trap placement had an influence on catches and that the optimum height varied according to the growth stage of the plant. The trap design also influenced the efficiency of capture. The models PVC 200 (homemade) and Pherocon 1C were the most efficient for the capture of males **S. absoluta** . The trap PVC 200 is, therefore, an alternative to commercial models. The data about trap colors indicated no difference among them.

Oral Paper 38

BIOSYNTHETIC STUDIES OF CONTACT PHEROMONES OF *DROSOPHILA MELANOGASTER*. Maryse Pennanec'h and Jean-Marc Jallon. Mécanismes de Communication, NAM4-CNRS URA 1491, Bât 446, Université Paris Sud, 91405 Orsay Cedex, FRANCE

The pheromone blends of mature D. melanogaster flies consist of sex-specific mixtures of cuticular hydrocarbons. Only mature females (which possess 150% more cuticular hydrocarbons than males) show w7,11 dienes. Monoenes are present in both sexes, with a common double bond in position w7. All these unsaturated long-chain hydrocarbons play a major role in sexual communication. De novo biosynthesis of these pheromones was followed using radio labels (myristic, palmitic, stearic and *cis*-vaccenic acids). The incorporation of C14 or H3 labeling was higher for C14:0 > C16:0 > C18:0 > C18:1, and slightly higher in males than in females. The proportion of labeled unsaturated components is higher for 18:1 > 14:0 > 16:0 > 18.0 in both sexes (60% and 77% with C18:1 for females and males respectively). Analysis of these components with radio-CG showed that for males 74% of the recovered labeling was in 7-tricosene. Internal hydrocarbons were also found which had a similar composition to cuticular hydrocarbons; higher levels were found in males than in females.

10th ANNUAL MEETING
International Society of Chemical Ecology
Tampa/Clearwater Florida
July 31-August 4, 1993

Contributed Papers

Session V (Tuesday August 3)

Mini-Symposium - Insect Pheromones & Hormones

Beach Gulf Room **David Carlson Presiding**

2:00 - 2:30 pm ***Oral Paper 39*** COMMERCIAL ADVANCEMENT IN PHEROMONE RELATED TECHNOLOGY. Philipp Kirsch.

2:30 - 3:00 pm ***Oral Paper 40*** ENDOGENOUS REGULATION AND BIOSYNTHESIS OF LEPIDOPTERA SEX PHEROMONES: CURRENT RESEARCH AND FUTURE DIRECTIONS. Peter E. A. Teal, James H. Tumlinson, Nianbia Fang, Ronald J. Nachman and Rella A. Abernathy.

3:00 - 3:15 pm ***Oral Paper 41*** DETERMINATION OF DOUBLE BOND POSITIONS IN POLYUNSATURATED PHEROMONES AND RELATED COMPOUNDS. Athula B. Attygalle, G.N. Jham and J. Meinwald.

3:15 - 3:30 pm ***Oral Paper 42*** PBAN ACTIVATION AND ITS MODE OF ACTION; SECOND MESSENGER STUDIES IN PHEROMONE GLANDS OF *HELICOVERPA ARMIGERA*. Victoria Soroker and Ada Rafaeli.

3:30 - 3:45 pm **COFFEE BREAK**

3:45 - 4:00 pm ***Oral Paper 43*** MALE-SPECIFIC VOLATILES FROM NEARCTIC AND AUSTRALASIAN TRUE BUGS (HETEROPTERA: COREIDAE AND ALYDIDAE). Jeffrey R. Aldrich.

4:00 - 4:15 pm ***Oral Paper 44*** ANAL EXUDATE OF THE QUEEN HONEY BEE: CHEMISTRY, FUNCTION, AND DISCOVERY OF A PHEROMONAL MIMIC. Murray S. Blum, Henry M. Fales, and Robert M. Page.

4:15 - 4:30 pm ***Oral Paper 45*** THE SCARAB BEETLES *ANOMALA OCTIESCOSTATA* AND *A. CUPREA* UTILIZE THE SAME SEX PHEROMONE. Walter Soares Leal, Masaaki Sawada, and Makoto Hasegawa.

4:30 - 4:45 pm ***Oral Paper 46*** SEX PHEROMONE OF THE ORIENTAL BEETLE, *EXOMALA ORIENTALIS*. Walter Soares Leal, Wendell L. Roelofs, Aijun Zhang, Michael Villani, Masaaki Sawada, Makoto Hasegawa.

4:45 - 5:00 pm ***Oral Paper 47*** INDENTIFICATION OF RELEASER PHEROMONES MEDIATING AGGREGATION IN GREGARIOUS DESERT LOCUST, SHISTOCERCA GREGARIA (FORSKAL) (ORTHOPTERA: ACRIDIDAE). Baldwyn Torto, Daniel Obengofori, Peter G. N. Njagi, Ahmed Hassanali and Habert Amiani.

5:00 - 5:15 pm ***Oral Paper 48*** PHEROMONES OF RHYNCHOPHORUS PALM WEEVILS. A. C. Oehlschlager, A. L. Perez, R. Gries, G. Gries, R. Hallett, H. D. Pierce, Jr., L. M. Gonzalez, J. H. Borden, and R. M. Giblin-Davis.

5:15 - 5:30 pm ***Oral Paper 49*** MANAGEMENT OF RED RING DISEASE IN COMMERCIAL OIL PALM PLANTATIONS THROUGH PHEROMONE - BASED TRAPPING OF THE AMERICAN PALM WEEVIL. A. C. Oehlschlager and L. M. Gonzalez.

Oral Paper 39

Commercial Advancement in Pheromone Related Technology. Philipp Kirsch. Trece Inc. 1143 Madison Lane, Salinas, CA 93907

Despite two decades of research, use of arthropod pheromones for enhancing pest management strategies is still a small industry. Although monitoring programs in horticulture and fruit production are well-established worldwide, they are restricted on broadacre and vegetable crops. Management of the pink bollworm on cotton in the desert southwest and of tomato pinworm in Mexico are the two most extensive uses of pheromones in agriculture. Mating disruption has recently been adopted in three fruit production systems - peach, pome, and grape. Indirect pheromone-based management is focussed on the development of monitoring tools, traps and lures, which when coupled with temperature driven predictive phenology models, permit considerable precision in the timing of control strategies. Direct pheromone-based management has focussed first on mating disruption and then on attracticide strategies. This paper will review current commercial application of pheromone technology and discuss factors that affect both commercial and scientific progress. The authors will suggest priorities for ongoing research programs in both monitoring and control applications of pheromone technology.

Oral Paper 40

ENDOGENOUS REGULATION AND BIOSYNTHESIS OF LEPIDOPTERA SEX PHEROMONES: CURRENT RESEARCH AND FUTURE DIRECTIONS. Peter E. A. Teal, James H. Tumlinson, Nianbia Fang, Ronald J. Nachman and Rella A. Abernathy. Insect Attractants, Behavior and Basic Biology Research Laboratory, USDA, ARS, P. O. Box 14565, Gainesville, FL 32604 USA.

Sex pheromones are crucial for mating success in most species of Lepidoptera. Consequently, the endogenous factors that regulate sex pheromone production and the biochemical mechanisms involved in synthesis of these compounds are key elements of the pheromone communication system. Studies conducted on a number of species have been instrumental in elucidating the mechanisms responsible for biosynthesis of the fatty acyl precursors of pheromones as well as the enzymatic steps employed during the terminal steps in pheromone biosynthesis. Additionally, recent studies have resulted in the identification of hormonal and neurohormonal regulators responsible for stimulation and termination of pheromone production in moth species. Information gained from this research has been instrumental for the identification of new sex pheromone components produced by Lepidoptera species and in the design of model mimetics which may allow for development of novel compounds for use in alternative strategies for pest control.

Oral Paper 41

DETERMINATION OF DOUBLE BOND POSITIONS IN POLYUNSATURATED PHEROMONES AND RELATED COMPOUNDS. Athula .B. Attygalle, **G.N. Jham and J. Meinwald**. Cornell Institute for Research in Chemical Ecology (CIRCE), Cornell University, Ithaca, NY 14853 USA.

Determination of the positions and configurations of carbon-carbon double bonds in naturally occurring compounds is a classical problem. Such structural elucidations are particularly challenging when only nanogram amounts of unknown material is available. We find that partially hydrogenating a polyene substrate, using N_2H_4 or $N_2{}^2H_4$ and H_2O_2, and identifying the monoenes in the resulting mixture by GC, GC-MS and GC-IR is a promising method. The monoenes are further characterized by derivatizing with dimethyl disulfide. The method is directly applicable to mixtures of polyenes and only about 20-50 nanograms of each unsaturated compound is required. Several applications of the procedure will be reported.

Oral Paper 42

PBAN ACTIVATION AND ITS MODE OF ACTION; SECOND MESSENGER STUDIES IN PHEROMONE GLANDS OF HELICOVERPA ARMIGERA.
Victoria Soroker and Ada Rafaeli* Department of Zoology, Tel Aviv University, Ramat Aviv, 69978, Israel. * Department of stored products , the Volcany Center, P.O. Box 6, Bet Dagan ,Israel.

In *H. armigera* a direct PBAN activation of the pheromone production in the pheromone gland was previously demonstrated by us. This activation could be mimicked by cAMP analogs *in vitro*. Subsequent studies presented the importance of extracellular Ca^{2+} for PBAN stimulation, thus suggesting the involvement of more then one transducing pathway.
This possibility was tested in the present study, using synthetic agonists for the phosphatidylinositol as well as for the cAMP pathways.
The interaction between cAMP production and extracellular Ca^{2+} levels was directly measured using RIA.
Results of the present study provide some evidence that the mode of PBAN action on the pheromone gland involves the activation of two pathways: cAMP and phosphatidylinositol. The possible interactions between the two mechanisms is discussed.

Oral Paper 43

MALE-SPECIFIC VOLATILES FROM NEARCTIC AND AUSTRALASIAN TRUE BUGS (HETEROPTERA: COREIDAE AND ALYDIDAE). Jeffrey R. Aldrich. Insect Chemical Ecology Laboratory, USDA-ARS, Beltsville, MD 20705 USA.

Aeration and exocrine gland extracts were analyzed for three Coreidae and two Alydidae. Males of all the species studied emit volatile blends that are probably pheromones, but sexual communication in these insects evolved differently. In the alydids, *Riptortus serripes* and *Mirperus scutellaris*, the metathoracic scent glands are sexually dimorphic, and the dimorphisms are expressed chemically. Secretions from the male alydids contain high concentrations of esters or alcohols (e.g. *(E)*-2-hexenyl *(Z)*-3-hexenoate, *(E)*-2-hexenyl butyrate, and *(E)*-2-octenol), while females produce mainly acids and aldehydes (e.g. butyric and hexanoic acids, and *(E)*-2-hexenal). In the coreids, *Amblypelta lutescens lutescens*, *A. nitida* and *Leptoglossus phyllopus*, the metathoracic scent glands are not sexually dimorphic, but male- and species-specific volatiles are released, apparently from cells in the cuticular epidermis. The coreid male-specific volatiles are primarily mono- and sesquiterpenes, including (-)-(3R)-*(E)*-nerolidol as the major component from *A. l. lutescens* (an Australasian species) and *L. phyllopus* (a Nearctic species). Only (+)-(3S)-*(E)*-nerolidol is commonly found in plants, so *(E)*-nerolidol from these coreids is environmentally unique because of its chirality.

Oral Paper 44

ANAL EXUDATE OF THE QUEEN HONEY BEE: CHEMISTRY, FUNCTION, AND DISCOVERY OF A PHEROMONAL MIMIC. Murray S. Blum, Henry M. Fales, and Robert M. Page. University of Georgia, Athens, GA 30602 USA.

The chemistry and function of the anal exudate of virgin honey bee (Apis mellifera) queens change dramatically after about 48 hrs of age. At this time the anal exudate functions as a gentle repellent (= tranquillizer) for worker bees that manifests aggressive behavior toward resident young virgins. The exudate of each queen appears to be quantitatively distinctive. Analyses of the anal secretions demonstrated that they were of great structural electicism containing hydrocarbons, acids, alcohols, esters, and aromatic ketones. The activity of the pheromonal tranquillizer, identified as o-aminoacetophenone, was completely matched by an unnatural compound with some similar properties to those of the pheromone.

Oral Paper 45

THE SCARAB BEETLES *Anomala octiescostata* AND *A. cuprea* UTILIZE THE SAME SEX PHEROMONE. Walter Soares Leal (National Institute of Sericultural and Entomological Science, Tsukuba, Ibaraki 305 Japan), Masaaki Sawada, and Makoto Hasegawa (Chiba Prefectural Agricultural Experiment Station, Chiba 266 Japan).

The scarab beetle *Anomala cuprea* utilizes, in addition to the previously identified major sex pheromone (R,Z)-5-(-)-(oct-1-enyl)oxacyclopentan-2-one (1), a minor component (R,Z)-5-(-)-(dec-1-enyl)oxacyclopentan-2-one, which has been previously identified as the sex pheromone of the Japanese beetle. Although present in slightly different proportions, both chemicals were also identified as the sex pheromone components of *A. octiescostata*, which appears in the field earlier than *A. cuprea*. Over 9000 *A. octiescostata* males/trap per day were captured this year in the peak of the season (late April), with 1 mg of synthetic sex pheromone. Besides, it was found that *A. daimiana* utilizes the sex pheromone of *A. schonfeldti* (2-E-nonenol) as a major and the major component of *A. cuprea* (1) as a minor component of its own sex pheromone blend. All these species are economically important pests in Japan.

Oral Paper 46

SEX PHEROMONE OF THE ORIENTAL BEETLE, *Exomala orientalis*. Walter Soares Leal (National Institute of Sericultural and Entomological Science, Tsukuba, Ibaraki 305 Japan), Wendell L. Roelofs, Aijun Zhang, Michael Villani (Cornell University, Department of Entomology, Geneva, NY 14456-0462 USA), Masaaki Sawada, and Makoto Hasegawa (Chiba Prefectural Agricultural Experiment Station, Chiba 266 Japan)

By means of a GC-behavior bioassay method, a sex pheromone of *Blitopertha orientalis*, a turf pest in Japan (called semadarakogane in Japanese), was identified as (Z)- and (E)-tetradec-7-en-2-one. A synthetic mixture in the same ratio as the natural product (Z/E, 7/1) was more attractive in the field than caged females. Pure Z-isomer as well as an Z/E mixture (12% E) were very attractive in a flight tunnel to *Anomala orientalis* (an exotic species to the US). In addition, the sex pheromone isolated from the American population had the same capillary GLC retention times and MS as the synthetic (Z) and (E)-tetradec-7-en-2-one. Taxonomic examinations suggested that the Japanese and American species are the same, which accord to a recent revision (Baraud, J., 1991) are to be named *Exomala orientalis*.

Oral Paper 47

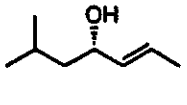
IDENTIFICATION OF RELEASER PHEROMONES MEDIATING AGGREGATION IN GREGARIOUS DESERT LOCUST, SHISTOCERCA GREGARIA (FORSKAL) (ORTHOPTERA : ACRIDIDAE). Baldwyn Torto, Daniel Obeng-Ofori, Peter G.N. Njagi, Ahmed Hassanali and Habert Amiani. ICIPE, P.O. Box 30772, Nairobi, Kenya

Olfactometric bioassays with gregarious phase desert locust showed that nymphs and adults responded only to their own respective volatiles. Analysis by GC-EAD, GC-MS of charcoal trapped air borne volatiles from nymphs, adults and faeces derived from the different locust stages revealed the presence of six electrophysiologically active compounds, identified as anisole, benzaldehyde, veratrole, guaiacol, phenylacetonitrile and phenol. These included three male-specific volatiles from older adults, identified as benzaldehyde, veratrole and phenylacetonitrile and two faecal-specific volatiles from young adults and nymphs identified as guaiacol and phenol. Behavioural tests with different synthetic pheromone blends showed that aggregation of adults was strongly synergised by phenylacetonitrile to which nymphs responded indifferently. On the other hand, nymphs were more responsive to a blend of guaiacol and phenol associated with nymphal and young adult faeces. The ecological significance of these results and the role of diet on pheromone production will be discussed.

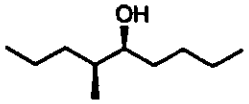
Oral Paper 48

PHEROMONES OF RHYNCHOPHORUS PALM WEEVILS A. L. Perez,[1] R. Gries,[2] G. Gries,[2] R. Hallett,[2] A. C. Oehlschlager,[1] H. D. Pierce, Jr.,[1] L. M. Gonzalez,[1] J. H. Borden[2] and R. M. Giblin-Davis[3] Department of Chemistry[1] and Department of Biological Sciences[2] Simon Fraser University, Burnaby, B. C. V5A 1S6 CANADA and [3]Fort Lauderdale Research and Education Center, University of Florida, Institute of Food and Agricultural Sciences, Fort Lauderdale, FL 33314

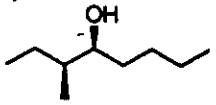
Weevils in the Rhynchophorinae (Coleoptera) are destructive pests of commercial and ornamental palms in the tropics. Male *R. palmarum* (Central and South America), *R. phoenicis* (Africa), *R. ferrugineus* and *R. vulneratus* (Middle and South East Asia), *R. bilineatus* (Papua New Guinea) and *R. cruentatus* (Florida) produce aggregation pheromones which, in combination with plant volatiles are highly effective in attracting conspecifics. This paper discusses the laboratory and field work that led to the identification of the pheromones shown below.



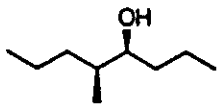
(4S)-2-methyl-4-hydroxy hept-5-ene
R. palmarum



(4S,5S)-4-methyl nonan-5-ol
R. ferrugineus
R. vulneratus
R. bilineatus



(3S,4S)-3-methyl octan-4-ol
R. phoenicis



(4S,5S)-4-methyl-octan-5-ol
R. cruentatus

MANAGEMENT OF RED RING DISEASE IN COMMERCIAL OIL PALM PLANTATIONS THROUGH PHEROMONE - BASED TRAPPING OF THE AMERICAN PALM WEEVIL. A. C. Oehlschlager and L. M. Gonzalez, Department of Chemistry, Simon Fraser University, Burnaby, B. C. V5A 1S6 CANADA.
Carlos. M. Chinchilla, Palm Research Program, Palma Tica, Coto, Costa Rica

The palm weevil, *Rhynchophorus palmarum*, causes significant damage to coconut and oil palm in the Americas as vector of red ring nematode. Adults of this weevil can be captured and killed in traps containing insecticide treated sugarcane or palm and from which is released the male-produced aggregation pheromone. Mass trapping of *R. palmarum* in a commercial oil palm plantation in Costa Rica significantly lowered red ring infection rates in the trapping areas and even in surrounding lots. This is attributed to the strong dispersal of this weevil which was established in mark-release-recapture experiments.

Trapping densities of 4 traps / Ha, 1 trap / Ha or 1 trap / 3.5 Ha resulted in reduced red ring infection rates. Trapping strategies are discussed in terms of normal plantation sanitation and replanting practises.

CONTRIBUTED PAPERS
POSTERS

Posters (Sunday, August 1)
Time: 7:30 -10:00 pm

Bay Room

Poster 1
AN ELICITOR IN CATERPILLAR ORAL SECRETIONS THAT INDUCES CORN SEEDLINGS TO RELEASE PARASITOID ATTRACTANTS. Hans T. Alborn, Ted C.J. Turlings, John H. Loughrin and J.H. Tumlinson.

Poster 2
SYNTHESIS OF DEUTERIUM LABELED POLYUNSATURATED FATTY ACIDS. Aleš Svatoš, Athula B. Attygalle and Jerrold Meinwald.

Poster 3
A NEW INFORMATIVE BIOASSAY FOR THE SEARCH OF ANTI-INSECT ACTIVITY IN PLANT SCREENING PROGRAMS. Miguel E. Alonso-Amelot, Jorge Avila, Daniel Otero, and Berenice Wolff.

Poster 4
PHENOLIC CONTENT OF THE TROPICAL BROWN ALGA *LOBOPHORA VARIEGATA* AS A FUNCTION OF NITROGEN AVAILABILITY. Thomas M. Arnold, Christopher E. Tanner and Walter I. Hatch.

Poster 5
SOME NEW AND NOVEL COMPOUNDS FROM CERTAIN SOUTH AFRICAN ANTS. John M. Brand and E. David Morgan.

Poster 6
FLOWER VOLATILES: ARE THEY ATTRACTIVE TO BITING INSECTS IN FLORIDA? David A. Carlson, M. Whitten, D. Kline and R. Stewart.

Poster 7
DUFOUR GLAND SECRETIONS OF STINGLESS BEES (MELIPONINAE). L. Cruz-López and E.D. Morgan.

Poster 8
ANTIMICROBIAL SESQUITERPENES FROM *PROSTANTHERA* (LABIATAE). Joanne E. Dellar, Michael D. Cole, Alexander I. Gray, Simon Gibbons, Peter G. Waterman.

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Poster 1

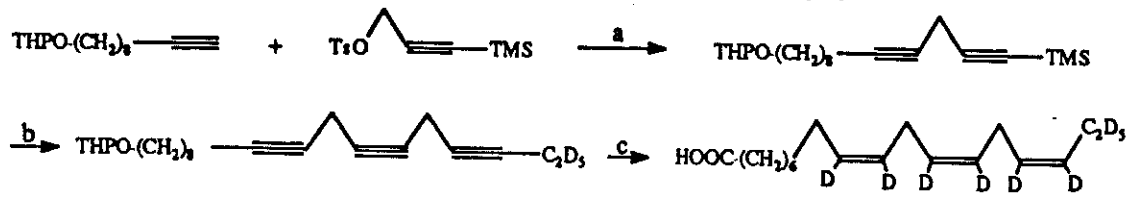
AN ELICITOR IN CATERPILLAR ORAL SECRETIONS THAT INDUCES CORN SEEDLINGS TO RELEASE PARASITOID ATTRACTANTS. Hans T. Alborn[1], Ted C.J. Turlings[2], John H. Loughrin and J.H. Tumlinson[3]. [1]Dept. of Chemical Ecology, University of Göteborg, Reutersgatan 2C S-413 20 Göteborg, Sweden. [2]Dept. of Phytomedicine/Applied Entomology, ETH Zentrum/CLS, Clausiusstrasse 21 CH-8092, Zürich, Switzerland. [3]USDA-ARS, Insects Attractants, Behavior, and Basic Biology Res. Lab., P.O. Box 15465, Gainesville, FL 32604

Several hours after caterpillars start feeding on a corn seedling (*Zea mays*) the whole plant responds with the release of relatively large amounts of terpenoids. Parasitic wasps that lay their eggs in caterpillars exploit this plant response to guide them into the vicinity of the hosts. Regurgitant of corn-fed beet armyworm (BAW) caterpillars (*Spodoptera exigua*), applied to damaged sites of seedlings as well as merely placing cut off seedlings in a solution of regurgitant in water for 12 hours causes the same release of volatiles. The diet of the insects had no effect on the activity and also regurgitant of other species of insects showed a very similar effect when tested on corn seedlings. The response therefore seems to be a general response to attack by phytophagous insects. BAW regurgitant was subjected to a series of purification steps. Each time, one distinctive and highly active fraction could be collected, although occasionally some other fractions showed minor activity. In the final purification procedure two distinctive peaks with very similar characteristics could be collected, however only one turned out to be active. Identification of this component is now in progress.

Poster 2

SYNTHESIS OF DEUTERIUM LABELED POLYUNSATURATED FATTY ACIDS. Aleš Svatoš, Athula B.Attygalle and Jerrold Meinwald. Cornell Institute for Research in Chemical Ecology (CIRCE), Cornell University, Ithaca, NY 14853 USA.

Carbon-carbon double bonds bearing two *cis* deuterium atoms can be prepared by treating acetylenes with bis(2-deuteriocyclohexyl)borane-B-d_1 [$DB(Cy)_2$] followed by CH_3COOD. As an example, the preparation of labeled linolenic acid is outlined below:



[a] CuI/NaI/K_2CO_3, DMF; [b] (i) KF, DMF; (ii) $TsOCH_2$-C≡C-C_2D_5, cond. [a]; [c] (i) $DB(Cy)_2$/CH_3COOD; (ii) Amberlyst, MeOH; (iii) PDC, DMF; [THP = tetrahydropyranyl; TMS = trimethylsilyl; Ts = p-toluenesulfonyl; DMF = dimethylformamide]

Several others unsaturated acids were also prepared by modifying the procedure appropriately. Purities of all products were high; no traces of *trans* isomers were detected by GC-MS. These acids are particularly useful for the study of insect pheromone biosynthesis.

Poster 3

A NEW INFORMATIVE BIOASSAY FOR THE SEARCH OF ANTI-INSECT ACTIVITY IN PLANT SCREENING PROGRAMS. Miguel E. Alonso-Amelot, Jorge Avila, Daniel Otero, and Berenice Wolff. Grupo de Química Ecológica, Departamento de Química, Facultad de Ciencias, Universidad de Los Andes, Mérida 5101, VENEZUELA.

Most bioassays for anti-insect activity of plant extracts are based on repellency toward treated substrates and leaf disk phago-depression of few species of lepidopteran larvae such as *Manduca, Heliothis, and Spodoptera sp.* Many of these assays frequently ignore other deleterious effects on total fitness of the insect, *e.g.* weight loss and modification of the efficiency by which ingested food is converted to insect biomass. This is probably due to the laborious methodology involved that is not applicable to a large number of samples. This may result in inadequate detection of compounds with ecological significance. Therefore, a method was developed to extract part of the missing information. The test insect, *Tribolium castaneum* (Coleoptera, Tenebrionidae) was exposed 24 h after emergence from its pupa to thin disks of artificial diet. The disks had been previously dosified with solutions of the test extract/compound at various concentrations. The change in insect weight, ingested food, mortality and distribution in one and two way bioassays were measured after 60 h. From this data phago-depression/stimulation, repellency/attractancy, LD_{50}, variation in conversion efficiency of ingested food, and their dose dependency was obtained. Results using pure phytochemical and active plant extracts will be presented.

Poster 4

PHENOLIC CONTENT OF THE TROPICAL BROWN ALGA *LOBOPHORA VARIEGATA* AS A FUNCTION OF NITROGEN AVAILABILITY. Thomas M. Arnold, Christopher E. Tanner and Walter I. Hatch. St. Mary's College of Maryland, St. Mary's City, MD 20686 USA.

Although phlorotannins in brown algae have been shown to deter herbivores and may accumulate in response to herbivory, recent evidence has suggested that concentrations of these polyphenolic compounds are inversely related to nitrogen availability. Previous studies that have examined the relationship between polyphenolics and nutrient availability were conducted in the field, and as a result other variables may have contributed to observed differences in polyphenolics. In order to more rigorously test the phenolic content:nitrogen availability hypothesis, a clone of the tropical brown alga *Lobophora variegata* was cultured for four weeks in seawater media of three different nitrate concentrations. Total phenolic content was determined weekly by a micro-Folin-Denis assay which uses as little as 1 mg of tissue. Percent carbon and nitrogen (per dry weight algal tissue) were also determined weekly for each nutrient treatment. Significant differences ($P<.001$) in phenolic concentrations were observed between nutrient treatments after two weeks. Phenolic concentrations were directly correlated with tissue C:N ratios and inversely correlated with percent tissue nitrogen. Our results support the hypothesis that nitrogen availability plays a significant role in determining polyphenolic concentrations in brown algae.

Poster 5

SOME NEW AND NOVEL COMPOUNDS FROM CERTAIN SOUTH AFRICAN ANTS. John M. Brand and E. David Morgan. University of Fort Hare, Alice 5700, South Africa, and University of Keele, Keele, UK.

Several new compounds have been identified in the poison gland and Dufours gland of certain South African ant species. A re-investigation of the Dufours gland contents of *Anoplolepis custodiens* has shown that, in addition to the previously identified *n*-alkanes and *n*-alkenes in the range C_{10}-C_{21}, primary alcohols (C_{19}-C_{22}), secondary alcohols (C_{20}-C_{23}) and 2-ketones (C_{20}-C_{23}) have been identified. It is possible that these high-boiling compounds are used by the workers in laying trails on the hot sandy surfaces of their characteristic habitat. The Dufours gland of workers of *Camponotus arminius* contains two major components that appear to be geranyllinalool and geranylnerolidol in addition to many of the usual hydrocarbons. The poison gland of these two formicine species, as well as that of some other formicine species, contains hexadecyl formate and 1-hexadecanol, in addition to formic acid. The poison gland of the myrmicine ant, *Messor capensis*, (Cape harvester ant) contains anabaseine as the major component, and minor amounts of anabasine and an uncharacterised ester of octadecenoic acid. Many of the usual hydrocarbons found in Dufours glands of ants are present in this gland.

Poster 6

FLOWER VOLATILES: ARE THEY ATTRACTIVE TO BITING INSECTS IN FLORIDA? David A. Carlson[1], M. Whitten[2], D. Kline[1] and R. Stewart[1]. [1]USDA, ARS, Medical and Veterinary Entomology Research Laboratory, Gainesville, FL 32604 USA. [2]Florida Natural History Museum, Gainesville, FL 32611. USA.

Biting diptera in Florida are often described as obtaining sugar meals from flowering plants before blood-seeking behavior begins. However, observations and evidence for this activity is rare, and the odorants responsible unknown. Mosquitoes are presumed to exhibit the same behavior. Recent observations suggest that biting midges (sand gnats, *Culicoides mississippiensis*) may obtain sugar meals from flowering plants in salt marsh, then bite. We describe terpenoid volatiles from inflorescences from candidate host plants, Holly (*Ilex* species) and Saw palmetto determined by GC-MS.

Poster 7

DUFOUR GLAND SECRETIONS OF STINGLESS BEES (MELIPONINAE)

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The Dufour gland of aculeate Hymenoptera have been widely studied but those of the eusocial bees of subfamily Apidae, which includes the tribe Meliponini and Apini have received little attention. We have recently examined the secretion of the Dufour gland of four genus of stingless bees workers (Meliponinae) from South America and find that they differ considerable between genus. The principal substances found in the Dufour gland secretion of Frieseomelitta languida were saturated and unsaturated hydrocarbons with nonadecane, heneicosane and pentacosene the major components. The Dufour gland secretion of Mannotrigona testaceicornis contained geranylgeranyl acetate as predominant component together with other esters. In Tetragonisca angustula carboxylic acids, amides and hydrocarbons were identified and in Friesella schrottkyi were found chiefly terpenes.

Poster 8

ANTIMICROBIAL SESQUITERPENES FROM *PROSTANTHERA* (LABIATAE).

Joanne E. Dellar, Michael D. Cole, Alexander L Gray, Simon Gibbons, Peter G. Waterman. University of Strathclyde, Glasgow, G31 1XW, Scotland, U.K.

The Labiatae are a rich source of terpenoid compounds, some of which have antimicrobial properties. In a continuing study of this family, members of the genus *Prostanthera* were investigated for antimicrobial compounds. Using bioassay guided isolation procedures, three sesquiterpenes were isolated from *P.rotundifolia* and *P.melissifolia*. These included the novel compound prostanthol and the known compounds aristolen-2-one and caryophyllene epoxide. All three compounds were able to inhibit germination of spores obtained from *Cladosporium cucumerinum* in the 1-50µg range. In addition, the growth of three bacterial phytopathogens was inhibited by prostanthol with MIC's ranging from 0.1-10µg per ml. It is possible that these sesquiterpenes are produced within the plant as an effective deterrent to microbial pathogen invasion.

Poster 9

THE ROLE OF HOST ODOUR PLUMES IN HOST LOCATION BY FEMALE *Aedes aegypti* (DIPTERA: CULICIDAE). Alvaro E. Eiras [1,2] John W. S. Bradshaw[2] & Paul C. Jepson[2]. [1]Universidade Federal de Viçosa, Brazil - [2] University of Southampton, UK

The response of female *Aedes aegypti* (L.) to human subjects was studied using the odour from their hands in an olfactometer and extracts of their sweat in a wind tunnel bioassay. Both assays demonstrated that there is great variability in the host preference of mosquitoes. Most of the sweat extracts did not activate flight by the mosquitoes. However, some of the extracts did elicit landing behaviour, indicating that individual human odours are likely to determine the host preference of mosquitoes. We have also discriminated the odour plumes to which female *A. aegypti* orientates at long- and short-range to locate human hosts. Mosquitoes were able to locate the source of sweat extract and carbon dioxide when presented either individually or in combination from a point source. Take-off and flight activity were enhanced by the combination, while the probability of landing decreased. When sweat extract alone was presented as a diffuse signal (ie: not from a point source), the mosquitoes became very active and were unable to locate a point source of carbon dioxide. However, when carbon dioxide was presented in diffuse form, flight activity was reduced and the probability of landings on a point source of human odour extract was significantly enhanced. These results indicate that, at long-range, mosquitoes are activated to fly upwind by a plume of carbon dioxide in combination with human odour, and at close-range, they switch to landing behaviour primarily guided by the plume of human odour.

Poster 10

OXIDATIVE STRESS AND INSECT HERBIVORY. G.W. Felton, J. L. Bi and C.B. Summers. University of Arkansas, Fayetteville, AR 72701.

Several host plants of noctuid caterpillars respond to herbivory by producing a flux of reactive oxidants that can impair the growth of larvae. Feeding by Helicoverpa zea on soybean, tomato, or cotton induces increases in the activities of foliar oxidative enzymes such as lipoxygenases and polyphenol oxidases. These enzymes produce toxic lipid peroxides and quinones. Furthermore, foliar levels of key antioxidants (ascorbic acid, glutathione) are decreased as a result of herbivory. Consequently, larvae are exposed to increased oxidative stress when feeding upon previously damaged leaf tissues.

Poster 11

FOLIAR MONOTERPENOID CONTENT OF THE CALIFORNIA BAY TREE, *UMBELLULARIA CALIFORNICA*, DURING LEAF STAGE DEVELOPMENT: RELATION TO HERBIVORY BY BLACKTAILED DEER. Raymond J. Goralka and Jean H. Langenheim. Biology Dept., University of California at Santa Cruz, CA 95064 U.S.A.

As part of a larger investigation regarding the role of monoterpenoids in deterring mammalian herbivory, browse intensity on Bay tree foliage by Blacktailed deer was monitored during a growing season. We observed a strong seasonal effect, with young leaf developmental stages in the spring and early summer being heavily browsed, while in mid-summer and fall the more mature leaf stages were left untouched. Analyses of the average total yields of foliar monoterpenoids showed very low yields in early growth leaf stages I and II (5.9 and 6.0 mg/g leaf dry wt.) and much higher yields in stages III, IV and V (30.4, 49.5 and 43.9 mg/g leaf dry wt.). Leaf stages also showed quantitative compositional variation during leaf development, especially in concentrations of Pinocarvone and Umbellulone which were inversely related. The field data suggest that monoterpenoid total yield and/or concentrations of oxygenated monoterpenoid compounds function as herbivory deterrents. We discuss these results in relation to plant chemical defense theories.

Poster 12

TERPENOIDS FROM *PINUS MARITIMA* AND THEIR TERMITE PREDATORS OF THE GENUS *RETICULITERMES* : ROLE FOR PROTECTION OF PINUS AGAINST TERMITES AND TERMITES AGAINST ANTS. Jean-Luc Clément, Laboratoire de Neurobiologie, CNRS, 31, ch. J. Aiguier 13402 Marseille, France. Patricia Nagnan, INRA, route de St. Cyr, 78026 Versailles, France, Michèle Lemaire, Musée d'histoire naturelle, parc Saint Paul, 18000 Bourges, France

Trees of the genus *Pinus* produce secondary compounds, especially terpenes, which constitute a very efficient defense against xylophagous insects. These compounds were toxins for termites of the genus *Reticulitermes* (LD50 between 3 and 20 µg/mg of termites). Differences appear between various termites species: terpenes were generally twice as toxic to *R.(lucifugus) grassei* as to *R. santonensis*. The non specific host of *Pinus* species (*R. (lucifugus) banyulensis*) was less susceptible than the sympatric species. When trees are dead or sick the quantity of terpenes decreases or they are biotransformed and they begin to act as attractants for termites workers which build galleries in the soil to invade new tree truncks. Termite species possess a particular caste: the soldier, to protect the society. Termite soldier defensive secretions are composed of the same terpenes as the wood they eat but in different proportions. Lethal doses were calculated for predators of termites, mainly ants. Ant resistance varies among species according to their trophic strategy and attack mode. Geranyl linalool, the major compound, acts as a natural insecticide against some ant predators (LD50=6 ppm). These species use raid or chemical crypsis to invade termite colonies. In contrast, some predators using a sting to kill termites were very resistant (10 000 ppm). The geographic variation in the amount of geranyl linalool in the soldiers' secretion is coincident with the average resistance of ants species. The use of the same compound by wood and termites suggests a coevolutionary process of synthesis and detoxification .

Poster 13

ANTIFEEDING EFFECT OF SELECTED ALKALOIDS FROM PLANTS ON THREE SPECIES OF INSECT STORAGE PESTS. Juraj Harmatha (1), Jan Nawrot (2), Bohumil Proksa (3). (1) Institute of Organic Chemistry and Biochemistry, 166 10 Prague, Czech Republic
(2) Institute of Plant Protection, 60-318 Poznań, Poland
(3) Faculty of Chemical Technology, STU, 812 37 Bratislava, Slovak Republic.

Twenty one alkaloids of isoquinoline, quinoline, indolizidine, indole, pyridine, piperidine and steroid types isoleted from plant material were tested for feeding deterrent activity against three species of storage pests: Sitophilus granarius, Tribolium confusum and Trogoderma granarium in a standard test. The relation between their structure and activity is discussed. The strongest feeding deterrent activity has been observed with the quinine, yohimbine and with the fraction containing 3-veratroyl- and 3-angeloylzygadenin from Veratrum album. The activity of these compounds is compared with the strongest antifeedants of terpene, rotenone, lignan, coumarin, and other types coming from our screening of secondary metabolites of plant origin.

Poster 14

ALDOXIMES AND N-HYDROXYAMINO ACIDS ARE INTERMEDIATES IN THE BIOSYNTHESIS OF THE CYANOGENIC GLUCOSIDES IN INTACT LARVAE OF THE MOTH *ZYGAENA TRIFOLII* (INSECTA : LEPIDOPTERA).

Guido Holzkamp and Adolf Nahrstedt. Institute of Pharmaceutical Biology and Phytochemistry, Westf. Wilhelms-Universität, D-48149 Muenster, Germany.

The larvae of the cyanogenic moth *Zygaena trifolii* produce the two cyanogenic glucosides linamarin (1-cyano-1-methylethyl-D-glucopyranoside) and lotaustralin (R-1-cyano-1-methylpropyl-D-glucopyranoside) using the amino acids valine and isoleucine as precursors and the nitriles 2-methylpropanenitrile and 2-methylbutanenitrile as intermediates. The aim of the present study was to determine the role of aldoximes and N-hydroxyamino acids (intermediates known from plant biosynthesis for cyanogenic glycosides) as intermediates in the lepidopteran pathway. [U-^{14}C]-2-methyl-propanealdoxime, 2S-[U-^{14}C]-methylbutanealdoxime and [U-^{14}C]-D,L-N-hydroxyisoleucine were synthesized and each compound was injected into the haemocoele of the larvae. Radioactive 2-methylpropanealdoxime was incorporated into linamarin, and radioactive D,L-N-hydroxyisoleucine and 2S-methylbutanealdoxime were incorporated into lotaustralin. Incorporation rates of the aldoximes were 18-22% and of the N-hydroxyamino acid 4-5%. These results clearly indicate that both, aldoximes and N-hydroxyamino acids, are intermediates in the biosynthesis of linamarin and lotaustralin by the larvae of *Zygaena trifolii* similar to the pathway observed for plants. The high incorporation rates, however, do not support a channeled system such as has been described for the microsomal enzyme systems that produce cyanogenic glycosides in higher plants.

Poster 15

OVIPOSITION STIMULANTS AND DETERRENTS REGULATING DIFFERENTIAL ACCEPTANCE OF *IBERIS AMARA* BY TWO *PIERIS* SPECIES.
Xinpei Huang, J.A.A. Renwick and K. Sachdev-Gupta. Boyce Thompson Institute, Ithaca, NY 14853 USA

Iberis amara contains both stimulants and deterrents that are involved in regulating oviposition by *Pieris rapae* and *P. napi oleracea*. The deterrents and stimulants were identified as 2-O-β-D-glucosyl cucurbitacins (E and I) and glucosinolates (sinigrin and glucoiberin), respectively. The contrasting responses of the two *Pieris* species to the semiochemicals can explain the differential acceptance of the plant by these butterflies.

Poster 16

ELECTROANTENNOGRAM AND OLFACTOMETER RESPONSES OF APHID PARASITOIDS TO NEPETALACTONE, A COMPONENT OF APHID SEX PHEROMONES.
Rufus Isaacs[1], Jim Hardie[1], Francesco Nazzi[2], Wilf Powell[3], Lester J. Wadhams[3] and Christine M. Woodcock[3]. [1]AFRC Linked Group in Aphid Biology, Imperial College at Silwood Park, Ascot, Berkshire. SL5 7PY, U.K. [2]Dipartmento di Biologia Applicata alla Difensa delle Piante, Universita di Udine, via delle Scienze 208, 33100 Udine, Italy. [3]AFRC Institute of Arable Crops Research, Rothamsted Experimental Station, Harpenden, Herts. AL5 2JQ, U.K.

Synthetic aphid sex pheromone, (+)-4a*S*,7*S*,7a*R*-nepetalactone, attracts female aphid parasitoids of several *Praon* species in the field. Electroantennogram studies were done on five parasitoid species: *Praon volucre, Aphidius ervi, A. matricariae, A. rhopalosiphi* and *A. urticae*. Females of all these species gave strong responses to the nepetalactone. Smaller, but significant, responses were also recorded from male parasitoids.

Behavioural responses of aphid parasitoids to the nepetalactone were investigated using a four-armed olfactometer. The chemical was released from a glass microcapillary into one of the arms, at a rate of 1.5ng/h. Single parasitoids were placed in the centre of the chamber and the number of entries and time spent in each arm were recorded over a twelve minute period. The apparatus was rotated through 45° every minute. Positive responses were recorded for both male and female *A. ervi* and *A. matricariae*. However, whilst female *A. matricariae* spent more time in the treated arm than the control arms, *A.ervi* made more entries into the treated arm than into the other arms.

Poster 17

RESPONSE OF GROWTH AND DEFENSE TO TWO LEVELS OF DEFOLIATION BY *HETEROTHECA SUBAXILLARIS*. Sandra L. Johnson and David Lincoln. University of South Carolina, Columbia, SC 29208 USA.

Heterotheca subaxillaris contains several mono- and sesquiterpenoid compounds which are thought to defend it against herbivory. We investigated the plant's response to defoliation and allocation to meet the conflicting requirements for continued growth, defense and reproduction. Defoliation on *H. subaxillaris* was simulated to resemble herbivory by its specialist folivore, *Zygograma heterothecae*, at levels comparable to those sustained in the field. We tested the hypothesis that defoliated plants would increase allocation of resources to growth, resulting in reduced defense. Analysis of leaf, shoot and root mass showed no difference in plant biomass between treatments, indicating compensatory responses to the 10-25% leaf area removals. Gas chromatographic analysis of volatile mono- and sesquiterpenes shows a tendency for lower quanitities of defense compounds in defoliated plants. This suggests a tradeoff relationship between growth and defense, rather than an induced response. Differences in flower production between treatments were not significant.

Poster 18

PHYTOTOXIC EFFECTS OF VOLATILE SUBSTANCES IN LYCOPERSICON ESCULENTUM. Young Sik Kim, Dept. of Physical therapy, Wonkwang Health Junior College, Iri, 570-750, Bong-Seop Kil, Dept. of Biology Educ., Wonkwang Univ., Iri, 570-749 and Kyeong Won Yun Dept. of Oriental Medicine Resourses, Sunchon National Univ., Sunchon, 540-742, Republic of Korea.

The phytotoxic effects of volatile substances emitted from the tomato plant(lycopersicon esculentum) on receptor plants was investigated. Vapours from the leaves inhibited seed germination and seedling growth of receptor plants in the laboratory tests. The inhibition response was dependent upon the volatile substances concentration. Laboratory bioassays showed the harvest mass reduction of grape vine planted near the tomato plants. In field bioassays, growth of lettuce and grape vine grown near the tomato plants in vinyl house were inhibited. To find out the phytotoxic compounds from tomato plant, GC/MS method was employed for analysis and identification of volatile substances. And forty-one chemical compounds such as trans-2-Hexenal, Linaleol, Phenylacetaldehyde, Methylsalicylic acid, Tetradecanaic acid, etc. were identified from tomato essential oil. These results suggest tomato plant may have allelopathic potential.

Poster 19

ALLELOPATHIC ACTIVITY OF VOLATILE SUBSTANCES BY ARTEMISIA PRINCEPS VAR. ORIENTALIS. Kyeong Won Yun, Dept. of Oriental Medicine Resources, Sunchon National Univ., Sunchon, 540-742, Bong-Seop Kil, Dept. of Biology Educ., Wonkwang Univ., Iri, 570-749 and Young Sik Kim, Dept. of Physical Therapy, Wonkwang Health Junior College, Iri, 570-750, Republic of Korea.

Artemisia princeps var. orientalis is a perennial plant often found in traditional agroecosystem and field in Korea. In a number of laboratory and field work, germination and growth of selected species were inhibitory by volatile substances emitted from A. princeps var. orientalis plant. The growth of several bacteria and fungi species supplemented with essential oils of the plant were also suppressed but species-dependent. The induction and growth of callus of selected species were inhibited severely. It was, therefore,confirmed that the volatile substances from the plant had biologically toxic activity. To detect phytotoxic substances of the plant, GC/MS method was used and identified as mainly terpenoids such as campor and (-)-thujone, etc. These data obtained from germination, seedling growth, tissue culture and microorganism test suggested that the volatile substances of A. princeps var. orientalis would be responsible for the allelopathic activity.

Poster 20

CHEMICAL AND SEASONAL VARIATIONS IN TWO POPULATIONS OF SATUREJA GILLIESII (GRAH.) BRIG. Cecilia Labbé and Francesca Faini. Universidad de Chile, Casilla 653, Santiago, CHILE.

Satureja Gilliesii, is a small shrub endemic to the Chilean Central Valley, with an unusual natural resistance to phytofagus insects. A comparative chemical study of three wild populations, growing in different ecological environments, revealed the existence of two chemical strains in this species, based upon the presence of 16 secondary metabolites isolated from leaf exudates. Eleven new compounds were characterized during this research and their structures (di-,sesqui-,and monoterpenoids) bear some unusual features disclosed by spectroscopical evidence.
A seasonal study showed that the differences are present in all periods of the year. Feeding deterrancy bioassays with Spodoptera littoralis larvae were done on the extracts and pure compounds and the results will be discussed.

This research was supported by DTI (U. de Chile) and CSIC (Spain) grants.

Poster 21

THE IMPLICATION OF PHENOLIC COMPOUNDS IN THE CHEMICAL DEFENSE OF MARINE SPONGES. Tyler R. Lindstrom, Hans D. Lemke, Walter I. Hatch, and Christopher E. Tanner, St. Mary's College of Maryland, St. Mary's City, MD 20686 USA.

Phenolic compounds are nearly ubiquitous among terrestrial plant families and are common in marine brown algae. Because of their unique properties, they have been implicated in many aspects of plant defense. They prevent infection and inhibit epiphytes by their antibacterial and antifungal activity; they deter feeding by many terrestrial and marine grazers; and they precipitate proteins, including digestive enzymes, rendering the plant material indigestible. Although phenolics are produced by animals, they have not been suggested as a defense against predation. In the present study, phenolic compounds, as meaured by a modified Folin-Denis assay, were detectable in most sponge species examined. Extremely high levels, 100 mg/g ash free dry weight, approaching those found in phenol rich brown algae, were recorded for one sponge species. Astringency, as measured by albumin precipitation, was also detectable in all of the species examined. Protein precipitating potential exceeding 200 mg tannic acid quivalents/g ash free dry weight of sponge were recorded in one species. Protein exposed to extracts of this sponge was rendered indigestible. In addition the digestive enzymes trypsin and pepsin were completely inhibited when exposed to phenols extracted from species with high phenolic content and astrigency. This marks the first time that phenolics have been hypothesized to serve an antifeedant role in animals.

Poster 22

VOLATILE COMPOUNDS FROM COTTON LEAVES ELICITED BY CATERPILLAR FEEDING DAMAGE. John H. Loughrin and James.H. Tumlinson. Insect Attractants, Behavior and Basic Biology Research Laboratory, Gainesville, FL 32604 USA.

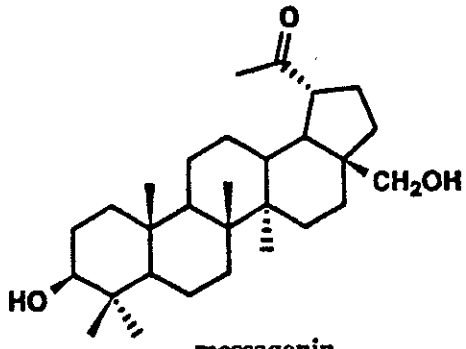
The volatile compounds emitted by leaves of five cotton cultivars and one Florida naturalized cotton were investigated *in situ*. Relatively low levels of compounds were emitted by undamaged plants (avetage less than 100 ng/hour). Plants which had been fed on overnight by beet armyworm larvae, however, emitted high levels of compounds; the principal components consisting of terpenes and lipoxygenase-hydroperoxide lyase products. Average production by commercial cultivars was approximately 1300 ng/hour while that from the naturalized cotton was approximately 9000 ng/hour. Feeding damage followed over the course of several days revealed qualitative and quantitative changes in the terpenoids emitted by the cotton plants.

Poster 23

ALLELOCHEMICALS FROM *Melilotus messanensis*

Francisco A. Macías, Ana M. Simonet and M. Dolores Esteban.
Departamento de Química Orgánica, Facultad de Ciencias, Universidad de Cádiz, Apdo. 40, 11510 Puerto Real, Cádiz, Spain.

The aerial parts of *Melilotus messanensis* (sweet clover) afforded, from the medium polar bioactive fractions, in addition to the known lupane triterpenes lupeol, betulin, betulin aldehyde and betulinic acid, the new nor-lupane messagenin (30-norlupane-3β,28diol-20-one) which have been tested as allelochemicals. Structures and their stereochemistry were elucidated by spectral methods and chemical transformations. Messagenin has been synthesized from betulinic acid.



messagenin

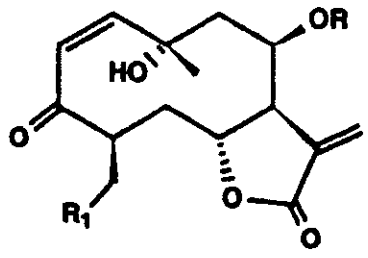
The effect of a series of aqueous solutions at 10^{-4}-10^{-9}M of eight natural and synthethic lupane derivatives were tested for their effects on the germination and growth of the dicotyledone species *Lactuca sativa* and *Lepidium sativum* and the monocotyledone species *Hordeum vulgare* and Triticum aestivum. All eight lupane triterpenes posses potential allelopathic activity in particular over dicotyledone species and their are likely to be significantly involved in the allelopathic action of *Melilotus messanensis*

Poster 24

STUDY OF THE ALLELOPATHIC POTENTIAL OF NEW GERMACRANOLIDES FROM CULTIVAR SUNFLOWERS, VAR. VYP®.

Francisco A. Macías, Ascensión Torres, Rosa M. Varela and José M. G. Molinillo.
Departamento de Química Orgánica, Facultad de Ciencias, Universidad de Cádiz, Apdo. 40, 11510 Puerto Real, Cádiz, Spain.

Chemical analysis of the medium polar fractions of *Helianthus annuus*, var. VYP®, has led to the isolation of four new germacranolide-type sesquiterpene lactones, along with other known compounds. The structures of these compounds were established by spectroscopic analysis of their ^{1}H and ^{13}C NMR, ^{1}H-^{1}H COSY, ^{1}H-^{13}C HETCOR and NOE diff spectra.



They have shown bioactivity on germination and root and shoot length on the dicotyledone *Lactuca sativa* and the monocotyledone *Hordeum vulgare* species. The study of the allelopathic potential of the natural products involved in the above mentioned effects will be discussed.

Poster 25

STUDIES ON THE MATURATION OF THE DESERT LOCUST *SCHISTOCERCA GREGARIA* FORKSAL. Hassane Mahamat, Ahmed Hassanali, Hezekiel Odongo and El-Sayeed El-Bashir. The International Centre of Insect Physiology and Ecology, P.O. Box 30772, Nairobi, Kenya

The accelerating effect of crowded mature *Schistocerca gregaria* males on the maturation of immature adult locusts has been confirmed. Immature locusts of either sex exposed to mature females and immature locusts took 24.8 ± 3.9 days to mature whereas these exposed to mature males took 12.1 ± 1.9 days (x^2 = 118.4, $P<0.01$). Immature locust exposed to hexane extracts of mature males were also induced to accelerated maturation (13.1 ± 2.0 days), thus confirming a previous suggestion that the phenomenon is due to a chemical signal. We have now demonstrated that this signal is present in the volatiles emanating from mature males (maturation time for immature locusts exposed to such volatiles was 12 ± 2.3 days). Candidate compounds in the volatiles of mature males have been isolated and identified, and these are now being tested for their role in accelerating maturation.

Poster 26

HEAVY METAL ACCUMULATION IN ROOTS, STEMS, AND LEAVES IN TYPHA LATIFOLIA IN A MAN-MADE WASTEWATER WETLANDS SYSTEM. William D. Mahon and John T. Romeo. University of South Florida, Tampa, Florida 33620, USA.

Abstract

Small, immature Typha latifolia (cat-tails) plants were transplanted from a near-pristine pond in central Florida to three separate plots in a man-made wastewater wetlands system. Heavy metal concentrations of Pb, Cd, and Al were measured in the roots, stems, and leaves of the plants. The study coincided with the cat-tail plant's rapid growing period. Metal concentrations in the roots stayed level or decreased when growth was most rapid, then increased as growth decreased. Uptake of Al in a plot with high Al concentration rapidly increased. Metal concentrations in stems and leaves initially decreased, then increased during the study period. Uptake rates correlated with metal concentrations in the soil, and not with metal concentrations in the water.

Poster 27

THE CHEMICAL ECOLOGY OF REPTILES: A SELF-GUIDED SLIDE PRESENTATION. Paul J. Weldon and Robert T. Mason. Texas A&M University, College Station, TX 77843 USA, and Oregon State University, Corvallis, OR 97331 USA.

Educational slide presentations are inexpensive, accessible to most secondary and undergraduate institutions, and easily updated as new information is acquired. Thus, they provide an ideal medium to foster appreciation for the field of chemical ecology. A sample self-guided slide presentation on the chemical ecology of reptiles will be shown, describing responses to pheromones, the discharge of predator repellents, and the structure, function and natural products of skin glands.

Poster 28

SYNOMONES IN INSECT-MITE PHORESY. Sergio Merino and Douglas Whitman. Biology, Illinois State University, Normal, IL 61761 USA.

Mesostigmata mites inhabit vertebrate carrion, an extremely ephemeral and dispersed habitat. The tiny mites cannot fly, but get from one carcass to another by "phoresy" -- riding on another animal. The mites attach themselves to Nicrophorus burying beetles, and in this way are carried to the next carcass. Mites recognize burying beetles by polar compounds on the beetles body. The interaction is beneficial for both the mite and the burying beetle, because the mites feed on competitors of the burying beetles: nematodes, fly eggs, and maggots. Thus, the mite-attractive compound acts as a synomone.

Poster 29

DETERMINING HOST PLANT UTILIZATION OF THE QUEEN BUTTERFLY USING CARDENOLIDE FINGERPRINTING. Raymond A. Moranz and Lincoln P. Brower. Department of Zoology, University of Florida, Gainesville, FL 32611 USA.

The larvae of the queen butterfly (Danaus gilippus berenice) feed upon plants in the Apocynaceae (the milkweeds and dogbanes). Using traditional methods (field observations), lepidopterists in north central Florida have recorded queen larvae on only a few species of plants, even though over twenty species of apocynaceous plants occur in the area. By taking advantage of the queen's ability to sequester cardenolides from their host plants, we hope to demonstrate the relative importance of various host plant species to the queen. Queens sequester a suite of cardenolides from each plant species that they feed upon; this set of cardenolides manifests itself on a thin-layer chromatogram as a distinctive pattern of blue spots (a "fingerprint"). By matching cardenolide fingerprints of queens collected in the field with fingerprints of butterflies reared on known host plants in the laboratory, we can determine the larval host plant used by each individual. We have reared queens on a variety of apocynaceous host plants in order to obtain standard fingerprints. This summer, we will capture adult queens in coastal and inland habitats of north Florida, and obtain their cardenolide fingerprints so that we can ascertain their larval host plants. Our poster will demonstrate the methodology used for our experiments, and will also show the results that we have obtained thus far.

Poster 30

BEHAVIOURAL TEST AND CHEMICAL IDENTIFICATION OF THE TRAIL PHEROMONE OF *ACROMYRMEX SUBTERRANEUS SUBTERRANEUS* FOREL. Ruth do Nascimento and Professor E.D. Morgan. Department of Chemistry, Keele University, Keele, Staffordshire, ST5 5BG, England.

The poison gland reservoir is the source of the trail pheromone in many species of myrmicine ants, including members of the tribe Attine which are commonly known as leaf-cutting ants. Previous work on the trail pheromone of Attine ants, using large numbers of ants, has identified methyl 4-methylpyrrole-2-carboxylate (M4MPC) as the trail pheromone of two species of *Atta*, namely *A. cephalotes* and *A. texana*, and one species of *Acromyrmex;* A. *octospinosus* In addition, ethyl-2,5-dimethylpyrazine (EDMP) and a mixture of EDMP and M4MPC have been found to be the trail pheromones of *A. sexdens rubropilosa* and *A. sexdens sexdens* respectively.
Behavioural tests on the poison gland apparatus of workers of *A. subterraneus subterraneus* located the poison gland of this species as the source of the trail pheromone. Solution of pure M4MPC elicited strong trail-following response in the ants while EDMP was inactive. Chemical analysis of single poison glands performed by gas chromatography using the solventless method of injection and a nitrogen selective detector, showed M4MPC as the sole component present in the secretion. From these results we conclude that M4MPC is indeed the trail pheromone of *A.s. subterraneus.*

Poster 31

CONTENTS OF THE MANDIBULAR GLANDS AND THE FUNCTION OF WORKERS IN THE QUEENLESS PONERINE ANT DINOPONERA AUSTRALIS. Neil J. Oldham and E. David Morgan. Keele University, Staffs. England. R. V. S. Paiva and C. R. F. Brandao. Universidade de Sao Paulo, SP, Brazil. B. Gobin, E. Schouters and J. Billen. Universiteit Leuven, Leuven, Belgium.

The mandibular gland contents of two entire colonies of the queenless ponerine ant Dinoponera australis have been analysed by gc-ms. The secretion has been found to contain a mixture of trisubstituted alkylpyrazines and tetrasubstituted hydroxyalkyl and alkenylpyrazines. Behavioural observations identified the function of each individual within the two colonies. Egg laying is confined to the most dominant worker, known as the gamergate, which remains inside the nest with workers of intermediate dominance which serve as nurses. Foraging is carried out by a few individuals which are low in the colony hierarchy. There is a tendency for foragers to contain more mandibular gland secretion than the nurses or gamergate. The function of the secretion is discussed in the light of these results.

Poster 32

THE INFLUNCE OF SIX MONOTERPENES ON EGG HATCH, LARVAE SURVIVAL AND GROWTH OF HELIOTHIS ARMIGERA HUBNER. Cao Ri Qiang and Yuan Kai Lai. Dept. of Bio., Nanjing Univ., Nanjing, 210008, P.R.China

Compared with control, the hatch rate of eggs in tubes containing α-pinene, β-pinene, ocimene, myrene, bicyclic isoprene and limonene is low. The hatching rate, control: 85%; limonene:60%; myrene:20%; β-pinene:10%; bicyclic isoprene:5%; α-pinene:3%; ocimene:0. The meridic diet is improved. The improved diet contains no nonporlar components except cholesterol. The improved diet has no bad influence on growth, development and reproduction. Mix each monoterpenes in the improved diet and feed larvaes. Larvaes fed diet containing each terpene have low survival rate and growth rate. The survival rate, control:93%; β-pinene:72%; myrene:42%; limonene:22% ; bicyclic isoprene: 20%; α-pinene: 14%; ocimene:0. The fresh weight of 7-day larvae, control:131mg; β-pinene: 7.3mg; limonene:5.2mg; bicyclic ieoprene:2.5mg; myrene: 0.9mg; α-pinene: 0.5mg; ocimene: 0.

Six monoterpenes occur in glanded conton. So, they contribute to the chemical resistence of conton to conton bollworm Heliothis armigera Hubner.

Poster 33

TOXIC EFFECT OF TERPENOIDS UPON SOME INSECTS FROM COLEOPTERA , DIPTERA AND LEPIDOPTERA ORDERS

Catherine REGNAULT -ROGER & Abdelaziz HAMRAOUI
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The efficiency of mediterranean aromatic plants as a practice to protect *Leguminosae* from the attacks of their beetles had been tested and it appeared that the most active plants belonged to *Labiatae* family and were rich in essentials oils. The lethal toxicity of these extracts was observed and the main compounds, benzenoids and isopropanoids,determined by gas chromatography. Following these first observations, the toxicity of some terpenoids were presently examined in this work. Carvacrol, thymol, linalool, eugenol, cynamaldehyde,1,8 cineole, p-cymene, a- pinene , borneol,camphor, terpineol, anethol, cuminaldehyde and estragol were tested upon insects belonged to *Coleoptera*, *Diptera* and *Lepidoptera* orders and their toxic effect analyzed by statistical analysis . A toxic effect by inhalation was observed upon *Acanthoscelides obtectus* SAY, bruchid of kidney bean (*Phaseolus vulgaris* L.), *Ceratitis capitata*, mediterranean fly and *Sitotroga cerealella* Ol.. CL50 for 24 hours and 48 hours in regard to adults mortality were determined.

This direct toxic effect on insects adults might be enhanced by an indirect toxic effect involving an inhibition of the reproduction : oviposition , larval growing and adult emergence of *A.obtectus* were inhibited with variable intensity by tested terpenoids. A larvicide effect was also noticed upon *Sesamia nonagroides* and *Ostrinia nubilalis* Hb, pests of maize.

This toxicity might be considered to use terpenoids as alternative substances within insecticidal compositions for a best management of the insect pests.

Poster 34

TOXIC EFFECT OF TERPENOIDS UPON *ACANTHOSCELIDES OBTECTUS* SAY.

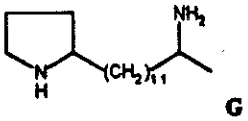
Catherine Regnault-Roger et Abdelaziz Hamraoui, Université de Pau et des Pays de l'Adour, 64000 Pau, France.

Aromatic and medicinal mediterranean plants showed some efficiency to protect Leguminosae from beetles attacks: the most active tested plants belonged to Labiatae, well-known for essential oils production. The lethal toxicity of these extracts was observed and the main compounds, benzenoids and isoproprenoids determined. Following these first observations, the toxic effect of some terpenoids (carvacrol, thymol, linalool, eugenol, cynamaldehyde, p-cymene, α-pinene, borneol, camphor, terpineol, anethol, cuminaldehyde and estragol) was tested upon *Acanthoscelides obtectus* Say (Coleoptera), a bruchid of kidney bean (*Phaseolus vulgaris*): a direct toxicity by inhalation was noticed and CL_{50} (24^H and 48^H) determined. This effect was enhanced by an indirect toxic activity involving the inhibition of reproduction: oviposition, larval growth and adult emergence decreased with variable intensity. So, besides their recognized attractive or repellent effects, appropriate concentrations of terpenoids appear to be defensive allelochemicals in plant-insect relationships. This toxicity might be considered to use them as alternative substances within insecticidal composition for a better management of the insect pest.

Poster 35

DEFENSIVE ALKALOIDS OF THE MEXICAN BEAN BEETLE, *Epilachna varivestis* (COCCINELLIDAE). Shang-Cheng Xu, Athula B. Attygalle, Kevin D. McCormick, Jerrold Meinwald, Curtis L. Blankespoor and Thomas Eisner. Cornell Institute for Research in Chemical Ecology (CIRCE), Cornell University, Ithaca, New York

Abstract: Two novel alkaloids, 2-(12-aminotridecyl)-pyrrolidine [G] and its 1-(2-hydroxyethyl) derivative [I], were characterized from extracts of adult Mexican bean beetles. These pyrrolidines, together with a previously identified homotropane alkaloid, euphococcinine, account for 90% of the alkaloids present in this beetle. A number of piperidine derivatives were also identified as minor components. The total mixture represents the most complex bouquet of alkaloids reported hitherto from any coccinellid beetle.



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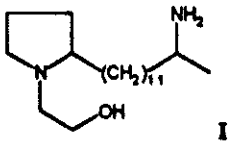

I

Poster 36

NESTMATE DISCRIMINATORS BIOSYNTHESIS IN *CATAGLYPHIS NIGER*. Victoria Soroker and Abraham Hefetz. Department of Zoology, Tel Aviv University, Ramat Aviv, 69978, Israel.

The mixture of cuticular hydrocarbons was shown to act as a fingerprint for nestmate recognition. The similarities between the hydrocarbon profile of the post pharyngeal glands and the cuticle implies that the glands may be the source of these hydrocarbons.
In the present study the hydrocarbon biosynthesis in *Cataglyphis niger* was examined.
In vivo and *in vitro* biosynthesis of hydrocarbon was followed by the use of ^{14}C sodium acetate as a precursor. The radioactive lipid products were separated by TLC and the fraction corresponding to the hydrocarbons was monitored by liquid scintillation counter. The identity of the hydrocarbons was further ascertained by GC.
In vivo experiments showed that labeled hydrocarbons were present both on the cuticle and in the postpharyngeal gland.
The involvement of postpharyngeal gland in hydrocarbon biosynthesis is discussed.

Poster 37

POLYPHENOLS IN EXTRACTS OF FOUR FORESTRY SPECIES. RELATION WITH THEIR ALLELOPATHIC POTENTIAL. J. Carlos Souto, Luis González and Manuel J. Reigosa. Dpto. Recursos Naturales y Medio Ambiente. Universidad de Vigo, Apdo. 874. 36200 Vigo. SPAIN.

The effects of extracts of four forestry species (*Quercus robur, Eucalyptus globulus, Acacia melanoxylon* and *Pinus radiata*) and of some polyphenols identified in these extracts on the germination of seeds and growth of radicles of *Lactuca sativa* and several undergrowth species are reported here.
In Galicia (NW of Spain) *Q. robur* is an autochthonous species and it is dominant in climax forests. *P. radiata, E. globulus* and *A. melanoxylon* are foreign species, introduced with commercial purposes.
Various studies show important differences regarding the structure and diversity in the undergrowth of the oakwood and rest arboreal communities which are studied in this work. The ecophysiological mechanisms which control the understory of woodlands structure and diversity are numerous and it is difficult to separate and define individual effects. Normally a group of related factors showing synergistic effects are responsible.
The possible allelopathic activity of the aloctonous species monocultures was taken as a starting hypothesis, considering it as one of the probable causes that justify the loss of species richness and low cover in the understory of woodlands.
Allelopathic capacity of the species in study and the role of polyphenols (mainly flavonoids) in this process is discussed.

Poster 38

INDUCED OXIDATIVE STRESS AS A POTENTIAL MODE OF ACTION FOR TANNIN TOXICITY IN THE LEPIDOPTERAN LARVAE HELICOVERPA ZEA. Clinton B. Summers and Gary W. Felton. University of Arkansas, Fayetteville, AR 72701 USA.

The midgut tissues of H. zea larvae ingesting dietary tannins were found to contain higher levels of lipid peroxidation products and oxidized proteins. The observation, in vitro, that both hydrolysable and condensed tannins can generate reactive oxygen species in alkaline conditions with catalytic amounts of ferrous iron, may indicate that the induction of oxidative stress is an important component of tannin toxicity to lepidopteran larvae.

Poster 39

HOST PLANT TROPANE ALKALOIDS IN THE ITHOMIINE *PLACIDULA EURYANASSA* (LEPIDOPTERA: NYMPHALIDAE). A.V.L. Freitas[1], J.R. Trigo[1], K.S. Brown Jr.[1], L. Witte[2] & T. Hartmann[2]. [1] Depto. Zoologia, I.B., UNICAMP, Campinas, SP, 13081, Brazil, [2] Institut für Pharmazeutische Biologie, Braunschweig, D3300 Germany.

Most Ithomiine butterflies feed as larvae on Solanaceous plants, rich in alkaloids (tropanes, steroid glycosides and others), bitter steroids (withanolides), saponins, phenolics, many types of terpenes, and strong-smelling oils. Even though the adults of Solanaceae-feeding Ithomiinae are unpalatable to a variety of vertebrate and invertebrate predators, no host plant-derived defensive chemicals have been detected in these butterflies; they sequester their primary defensive compounds (dehydropyrrolizidine alkaloids, PAs) from flower nectar (Compositae) or decaying Boraginaceae (*Heliotropium*). We have found that gregarious aposematic larvae of the primitive Mechanitine *Placidula euryanassa* (a monotypic genus restricted to southeastern Brazil and adjacent countries) store some of the tropane alkaloids (0.32% of their dry weight) from the Solanaceous host plant *Brugmansia suaveolens*. Part of these are also passed on to the pupae (0.07%) and imagos (0.0027%). Field-caught adults often have no PAs or very small amounts, go rarely to PA baits, and are frequently accepted by the spider predator *Nephila clavipes*. The retained tropanes may be adequate protection against some important predators of this species in its environment (larvae are rejected by birds and monkeys), but these substances did not confer protection for the adults against *Nephila*.

Poster 40

REPELLENT ACTIVITY OF INTERSPECIFIC ANT ALARM PHEROMONES AGAINST *SOLENOPSIS INVICTA*, THE RED IMPORTED FIRE ANT. Robert K. Vander Meer and Jack S. Seawright. USDA/ARS, P.O. Box 14565, Gainesville, FL 32604 USA.

The alarm pheromones of many ant species have been isolated and identified. They are normally small very volatile molecules containing 3-11 carbons. The alarm pheromones often consist of blends of several compounds and several different behaviors have been associated with these pheromones. The most obvious elicited behavior is a dramatic increase in the rate of locomotion, characterized by a looping search pattern. The movement is directed toward the source by the action of an attractive component of the alarm pheromone. The alarm pheromone of the fire ant is unknown; however, it was surprising that Y-tube olfactometer evaluation of several interspecific alarm pheromone components demonstrated strong repellent activity. These repellents have the potential to provide non-insecticidal exclusion of fire ants from areas where they are unwanted. One of these compounds impregnated in soil prevented newly mated fire ant queens from founding colonies in the treated soil. Also, when applied to peat pots it prevented fire ant colonies from moving into the soil filled pots.

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MATING BEHAVIOUR OF MARUCA TESTULALIS

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The podborer Maruca testulalis (Lepidoptera: Pyralidae) is a pest of many tropical legumes. Observations showed that three days after emerging a calling female was attractive to males when its ovipositor was protruded and curved upwards. The successful male suitor had to thump the females long waving antennae with his protracted abdominal tip hair-pencils. By doing so he deposited bead-like hair-pencil material which immobilized the antennae by holding them together in an upright position and then the female raised her abdomen (acceptance response). The male still in flight, immediately turned around, made genital contact and the pair remained in copula for 1-2h. Soon after pairing the female disentangled her antennae, with her fore legs. Electroantennogram (EAG) bioassays showed that the hair-pencil extracts contained stimuli which were more effective on the female antennae than the male antennae but the reverse was true when the ovipositor extracts were tested.

In this pyralidae, semiochemicals from both sexes appear essential for mating to succeed. Since the biologically active compounds from the ovipositor and hair-pencil material are unknown, combined flame ionization and electroannographic detector (FID-EAD) methods are currently employed to facilitate the identification of active components.

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ALLELOPATHIC POTENTIAL OF MENTHOFURAN MONOTERPENES FROM CALAMINTHA ASHEI.

Jeffrey D. Weidenhamer, Marios Menelaou, Francisco A. Macias, Nikolaus H. Fischer and G. Bruce Williamson.

A reversed-phase HPLC analysis was used to separate and quantify five menthofuran monoterpenes in the leaf soaks and washes of Calamintha ashei, a perennial shrub of the Florida scrub community. Epi-evodone and desacetylcalaminthone were the major constituents of both soaks and washes. Concentrations of epi-evodone and desacetylcalaminthone were as high as 0.66 and 0.74 mM, respectively, in leaf soaks. Total concentrations of monoterpenes in leaf washes by misting were in no case higher than 0.021 mM. Aqueous solubilities of the menthofuran monoterpenes were determined to exceed concentrations required for growth inhibition. Bioassays of Calamintha monoterpenes have demonstrated effects on germination as low as 0.05 mM for epi-evodone. The analytical results are evaluated against the bioassay data with respect to the allelopathic potential of Calamintha.

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INFLUENCE OF FOLIAR MONOTERPENOIDS FROM *UMBELLULARIA CALIFORNICA* ON NITRIFICATION POTENTIAL IN SOIL BENEATH *UMBELLULARIA* Swarup E. Wood and Jean H. Langenheim. University of California at Santa Cruz, Santa Cruz, California 95064 USA.

Nitrification potential soil bioassays were performed on monoterpenoid amended soil, taken from under *Umbellularia* trees, to determine the influence of *Umbellularia* foliar monoterpenoids on nitrification. Nitrification potentials of all monoterpenoid (monoterpene hydrocarbons and oxygenated monoterpenes) amended soils were significantly ($p<0.05$) less than controls at additions of 0.59mg monoterpenoid/g dry soil. Monoterpene hydrocarbons, sabinene, p-cymene, and a-pinene, were significantly more inhibitory than oxygenated monoterpenes, cineole, terpinen-4-ol, and umbellulone. Significant ($p<0.05$) inhibition of nitrification was observed with monoterpene hydrocarbon additions of 0.26mg monoteprene/g dry soil. However, similarly significant inhibition by oxygenated monoterpenes required additions of 0.59 mg monoterpene/g dry soil. The possible inhibition by monoterpenoids of nitrification in forest soils and its implication regarding nutrient cycling in forest ecosystems will be discussed.

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CHEMICAL COMMUNICATION IN THE MALADERA MATRIDA ARGAMAN BEETLE. Gal Yarden and Arnon Shani. Ben-Gurion University of the Negev, Beer-Sheva 84105 ISRAEL.

The *Maladera matrida argaman* beetle (Coleoptera, Scarabaeidae, Melolonthinae), a relative newcomer to Israel, was first identified there by B. Argaman in 1983 and has recently appeared in Saudi Arabia. This beetle is considered a serious polyphagic pest, affecting many agricultural crops and is also nuisance to man. Preliminary experiments were carried out to study the biological activity of the species and to characterize the volatiles produced by live virgin beetles. Field experiments involving trapping with attractants and in olfactometer experiments, were designed as follows: live virgin females, with or without food, live virgin males, with or without food, and food alone were exposed to beetles in the field or at the lab in olfactometer experiments. Live virgin females in the presence of food (cut peanuts leaves) proved more attractive to both males and females than other treatments tried ($\alpha < 0.001$). Live virgin females with food attracted more males than females in a 3:1.5-2 ratio. In another set of field trapping experiments, volatiles produced by live virgin females plus food had the same attracting ability as live virgin females with food. Volatiles produced by virgin females with food attracted males and females more strongly than treatments with volatiles produced by live virgin male beetles with or without food or volatiles produced by virgin females without food ($\alpha < 0.001$). The results of the behavioral experiments as well as preliminary GC/MS and EAD measuurments will be presented.

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OVIPOSITION - AGGREGATING PHEROMONE OF THE DESERT LOCUST, SCHISTOCERCA GREGARIA. Manoj M. Rai and Rajinder K. Saini, International Centre of Insect Physiology and Ecology; Box 30772, Nairobi, Kenya.

Experiments undertaken confirm that gravid females prefer to oviposit in sand previously used for egg lying. The preference was shown to be due to a chemical factor originating from the froth and not from the eggs. Hexane and methanol extracts of froth and volatiles collected from it were shown to have the attractive compounds. Electroantenograms indicated that the receptors for the pheromone are present on the antennae.

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TRANSFER BETWEEN WORKERS OF AN EXOGENOUS HYDROCARBON TOPICALLY DEPOSITED ON THE CUTICLE IN THE ANT *CAMPONOTUS VAGUS* SCOP.: ROLE OF THE POST-PHARYNGEAL GLANDS IN INTRACOLONY CHEMICAL HOMOGENEITY. Mohamed Meskali, Annie Bonavita-Cougourdan, Eric Provost and Jean-Luc Clément. C.N.R.S. LNB8. 31, Ch. Joseph Aiguier, 13009 Marseille, France.

In order to study variations in the cuticular hydrocarbon profiles as a function of the social environment in ants, we analysed the mechanisms involved in hydrocarbon transfer between members of an ant colony. We performed experiments in which one worker of the ant *Camponotus vagus* was treated topically with (Z)-9-Tricosene (an unsaturated hydrocarbon not normally synthesized by this species), and placed together with 5 non-treated workers.
The cuticular extracts of all six workers in each group were analysed after 30, 90 minutes and 9, 24, 48, 96, 168, 264 and 336 hours of cohabitation. The GC-MS analyses show that (Z)-9-Tricosene was present in the cuticles of some non-treated workers which had spent 96 and 168 hours in contact with a treated worker and no (Z)-9-Tricosene was detected either before 96 hours or after 168 hours of cohabitation. The post-pharyngeal glands were dissected out and a GC-MS analysis was performed on groups after the same periods of cohabitation as above: the results show that from 30 min to 168 hrs of cohabitation, the post-pharyngeal glands contained (Z)-9-tricosene. The highest levels were recorded in all the individuals in each group at about 24 hours of cohabitation.
These results suggest that this substance may have been actively transferred between individuals in the same group during licking or grooming activities. After being absorbed by the non-treated workers via the post-pharyngeal glands, (Z)-9-tricosene was probably transported through the cuticle after 96 and 168 hours of contact with a treated worker. These data may show some of the ways in which chemical homogeneity is achieved in social insect colonies, especially as regards the chemical substances involved in nestmate recognition.

EFFECTS OF ISOLATED NATURAL PRODUCTS FROM CORAL REEF SPONGES ON FEEDING BY PREDATORY FISHES AND CRABS. Sonia R. Pablo, Steven C. Pennings and Valerie J. Paul. Marine Laboratory, University of Guam, Mangilao, Guam 96923 USA.

Although sponges contain large numbers of secondary metabolites, often present in high concentrations, little is known about the natural function(s) of these metabolites. We offered secondary metabolites from tropical marine sponges at natural concentrations to 3 groups of consumers. A mixture of avarol and isoavarol usually deterred feeding, as did a brominated diphenyl ether. Luffariellolide and heteronemin usually had no effect or stimulated feeding, while chondrillin had variable effects. Pufferfish, *Canthigaster solandri*, were most sensitive to sponge secondary metabolites, followed by crabs, *Leptodius* spp., followed by reef fishes, which were deterred from feeding by none of the metabolites. These results indicate that sponge secondary metabolites vary in their effectiveness as feeding deterrents, and that potential consumers vary in their sensitivity to sponge secondary metabolites.

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