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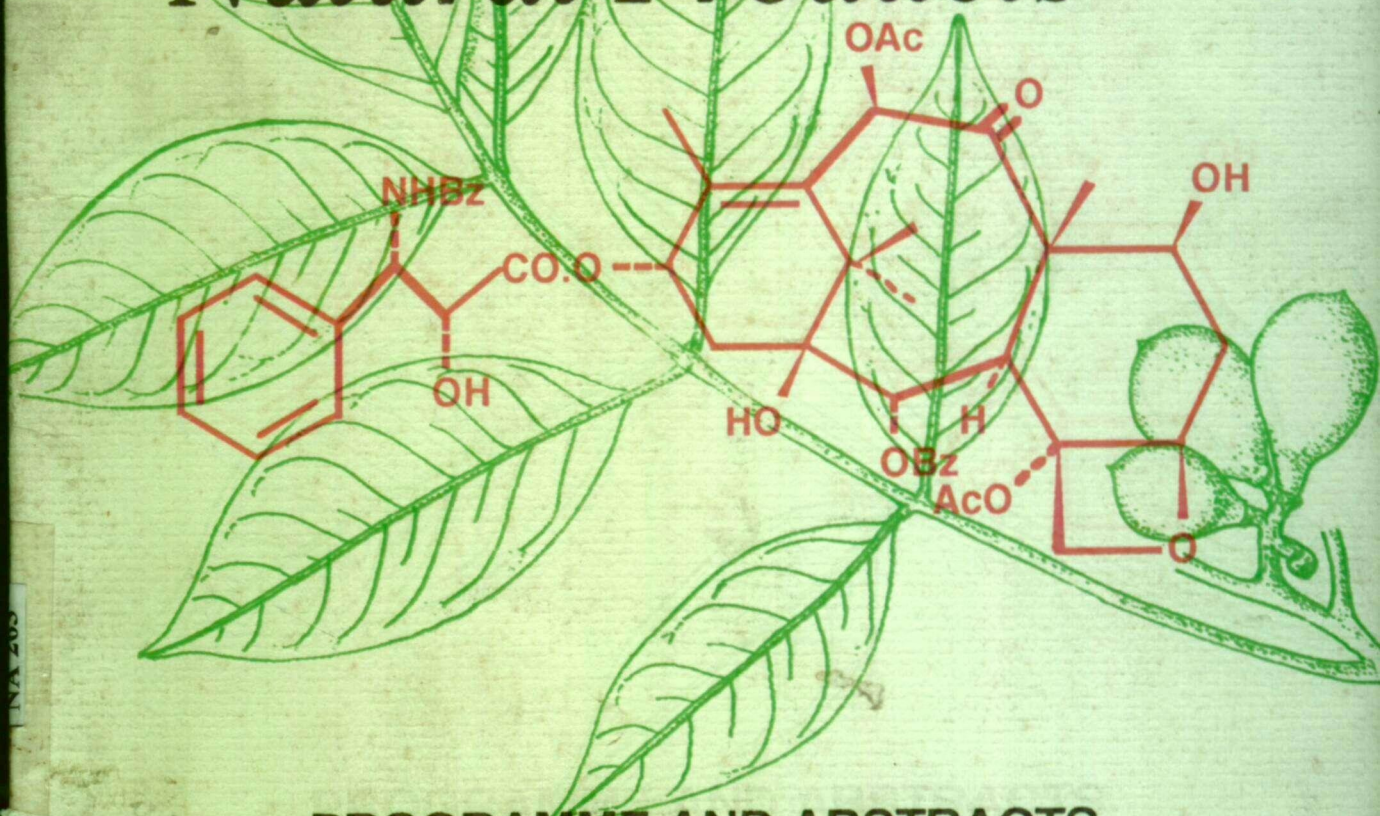
Symposium on Bioactive Natural Products



PROGRAMME AND ABSTRACTS

Organised by the Department of Chemistry,
University of Peradeniya at the Hotel Tree of Life,
Kandy, Sri Lanka. *November 11-15, 1996*

Symposium on Bioactive Natural Products

November 11-15, 1996

Hotel Tree of Life, Yahalatenne, Kandy

PROGRAMME AND ABSTRACTS

sponsored by

Department for Research Cooperation, SAREC, Sida.

International Program in Chemical Sciences,
Uppsala University, Sweden.

International Foundation for Science, Stockholm

Natural Resources, Energy and Science Authority
of Sri Lanka (NARESA)

organised by

**Department of Chemistry, University of Peradeniya,
Peradeniya, Sri Lanka.**

For Society our work means many things: pleasure, we hope, for those who follow it; instruction for those who perhaps need it; but also, and more widely, it means a common power, a power to achieve that which could not be achieved without knowledge. It means the cure of illness and the alleviation of suffering; it means the easing of labour and the widening of the readily accessible frontiers of experience, of communication, and of instruction.....

J. Robert Oppenheimer on *Research*

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ORGANISING COMMITTEE

Chairman:	Professor G. P. Wannigama
Symposium Co-ordinator:	Professor Vijaya Kumar
Members:	Professor B. M. R. Bandara
	Dr. Veranja Karunaratne
	Professor Savitri Kumar
	Dr. Anura Wickramasinghe
	Dr. Mahinda Wickramaratne
	Dr. Swarna Wimalasiri
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	Mr. Vajira Gunasekera
	Mr. A. N. Illangantilake
	Mr. A. H. M. A. Abeyratne

ACKNOWLEDGEMENTS

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International Program in Chemical Sciences,
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Internation Foundation for Science, Stockholm, Sweden.

Natural Resources, Science and Energy Authority of Sri Lanka, Colombo.

University of Peradeniya, Peradeniya.

The Committee also wishes to thank the following for their kind assistance:

Professor C. M. Madduma Bandara, Vice-Chancellor and Mrs. Madduma Bandara

Ms. Anjani M. Karunaratne, Ms. Chamalie Abeysekera and Ms. Rasika Mudalige, Department of Botany, University of Peradeniya.

Dr. D. B. Sumithraarachchi, Director, National Botanic Gardens, Peradeniya.

Dr. A. R. Ariyaratne, Dr. R. O. Thattil and Professor Kapila Gunasekera, Faculty of Agriculture, University of Peradeniya.

Programme for Inauguration of Symposium

9.30 a.m. - Traditional Lighting of Oil Lamp

9.35 a.m. - Welcome Address -

Prof. R. P. Gunawardane
Dean, Faculty of Science

9.45 a.m. - Address by Vice-Chancellor

Prof. C. M. Madduma Bandara
Vice-Chancellor, University of Peradeniya

9.55 a.m. - Inauguration Address

Hon'ble Bernard Soysa
Minister for Science, Technology and Human Resources Development

10.15 a.m. - Prof. M. U. S. Sultanbawa Felicitation Address

Prof. G. P. Wannigama

10.35 a.m. - Address

Prof. M. U. S. Sultanbawa

10.45 a.m. - Tea

11.15 a.m. - End of Inaugural Session

PROGRAMME

Monday, November 11, 1996

Chairman: Professor M. U. S. Sultanbawa

11.15 a.m. - Biologically Active Constituents of Tropical and Subtropical Plants

Wolfgang Kraus, Hohenheim University, Stuttgart, Germany

12.05 p.m. - The Bridge Over Research & Development

R. O. B. Wijesekera, Chairman, Ceylon Institute of Scientific and Industrial Research, Colombo, Sri Lanka

12.55 p.m. - Lunch

Chairman: Professor E. David Morgan

2.00 p.m. - *Pimpinella monoica* - A source of Bioactive Compounds

Asoke Banerji, Bhabha Atomic Research Centre, Mumbai, India.

2.45 p.m. - Identification and Synthesis of the Sex Pheromones of *Bongota cranaodes*

C. Rikard Unelius, The Royal Institute of Technology, Stockholm, Sweden.

3.05 p.m. - Tea

3.20 p.m. - Leave Hotel to Visit Royal Botanic Gardens, Peradeniya

4.00 p.m. - Arrive at Royal Botanic Gardens, Peradeniya

6.00 p.m. - Visit University of Peradeniya Campus

Monday, November 11, 1996

(Continued)

Social Programme

7.00 p.m. - Dinner hosted by Vice-Chancellor, University of Peradeniya
The Lodge, University of Peradeniya

10.30 p.m. - Return to Hotel

Tuesday, November 12, 1996

Chairman: Professor Wolfgang Kraus

9.00 a.m. - Synthesis of Biologically Active Natural Products with a Difference

A. V. Rama Rao, A. V. Rama Rao Research Foundation, Hyderabad, India.

9.50 a.m. - Overcoming Problems in Developing a Natural Pesticide from the Neem Tree

E. David Morgan, Keele University, Keele, England.

10.35 a.m. - Tea

Chairman: Professor G. P. Wannigama

10.50 a.m. - Pheromones, High Bioactive Natural Products as Signals in Chemical Information of Insects: Examples from Butterflies and Ants

H. J. Bestman, University of Erlangen-Nurenberg, Erlangen, Germany

Tuesday, November 12, 1996

(Continued)

11.40 a.m. - A Chemical Attractant System for the Coconut Pest, *Rhynchophorus ferrugneus*

N. E. Gunawardena, University of Kelaniya, Kelaniya, Sri Lanka.

12.10 p.m. - Napthoquinonoids from Nature: Development Towards Antiparasitic Agents

Banasri Hazra, Jadavpur University, Calcutta, India.

12.40 p.m. - Biologically Active Saponins from Sri Lankan Plants

U. L. B. Jayasinghe, Institute of Fundamental Studies, Kandy, Sri Lanka.

1.00 p.m. - Lunch

Chairman: Professor Nobuhiro Fusetani

2.00 p.m. - Biologically Active Secondary Metabolites from Marine Organisms

E. Dilip de Silva, Open University of Sri Lanka, Nugegoda, Sri Lanka.

2.30 p.m. - Studies on Use of Plant Products from Agriculture Pest Management.

M. D. Manandhar, Tribuvan University, Kathmandu, Nepal.

3.00 p.m. - An Antifungal Bis-Azasugar from *Aspergillus fumigatus*

D. T. U. Wijayaratne, University of Colombo, Colombo, Sri Lanka.

Tuesday, November 13, 1996

(Continued)

3.20 p.m. - Search for Bioactive Compounds in the *Calophyllum* species of Sri Lanka

H. R. W. Dharmaratne, Institute of Fundamental Studies, Kandy, Sri Lanka.

3.40 p.m. - Tea

Social Programme

4.00 p.m. - Visit to Kandy and the Temple of the Tooth

7.30 p.m. - Return to Hotel for Dinner

Wednesday, November 13, 1996

Chairman: Professor A. V. Rama Rao

9.00 a.m. - Search for Useful Leads from Marine Organisms

Nobuhiro Fusetani, University of Tokyo, Tokyo, Japan.

9.45 a.m. - Natural Products in Agriculture

Louis-Pierre Molleyres, Ciba-Geigy Crop Protection, Basel, Switzerland.

10.30 a.m. - Tea

Chairman: Professor H. Bestmann

10.45 a.m. - Is Natural Products Research of Relevance to Ayurveda?

Ajith Abeysekera, University of Jayawardenapura, Nugegoda, Sri Lanka.

Wednesday, November 13, 1996

(Continued)

11.30 a.m. - Hypoglycemic Effects of Some Medicinal Plants

M. Mosihuzzaman, University of Dhaka, Dhaka, Bangladesh.

12.10 p.m. - Isolation and Characterization of Terpenoids and Glycosides from *Pterospermum acerifolium* and *P. semisagittatum*

Nilufar Nahar, University of Dhaka, Dhaka, Bangladesh.

12.35 p.m. - Biological Activity of some Medicinal Plants of Sri Lanka

W. R. Wimalasiri, University of Peradeniya, Peradeniya, Sri Lanka.

12.55 p.m. - Lunch

Chairman: Professor M. Mosihuzzaman

2.00 p.m. - Alkaloids of some Bioactive Plants of Sri Lanka

L. S. R. Arambewela, Ceylon Institute of Scientific and Industrial Research, Colombo, Sri Lanka.

2.30 p.m. - Bioactive Bisbenzylisoquinoline Alkaloids from Menispermaceae

Biswapati Mukherjee, Dr. B. C. Roy Post-Graduate Institute of Basic Medical Sciences, Calcutta, India.

3.00 p.m. - Antileishmanial Activity of Jawaharene Fraction JF1 Towards *Leishmania donovani* Promastigotes

A. K. Saha, Jadavpur University, Calcutta, India.

Wednesday, November 13, 1996

(Continued)

3.30 p.m. - Effects of Azadirachtin on Specific Tissues in the Locust, *Schistocerca gregaria*

P. A. Paranagama, University of Kelaniya, Kelaniya, Sri Lanka.

4.00 p.m. - Tea

4.15 p.m. - 6.00 p.m. - **POSTER SESSION**

Posters

P 1 Screening of Plant Extracts for Nematicidal Activity

Champani Balasuriya, University of Peradeniya, Peradeniya, Sri Lanka.

P 2 Effect of Caffeine and Tannic Acid on the Growth of *Monacrosporium ambrosium*

N. K. Bandaranayake, University of Peradeniya, Peradeniya, Sri Lanka.

P 3 Extraction and Purification of Azadirachtin from Neem Seeds

D. Karunananda Bombuwela, University of Peradeniya, Peradeniya, Sri Lanka.

P 4 Neutral Sugars in Tea Stems and their Effect on *Monacrosporium ambrosium*

R. M. Thushari P. Bombuwela, University of Peradeniya, Peradeniya, Sri Lanka.

P 5 Host Attractants for the Rice Bug *Leptocorisa oratorius*

S. Dissanayake, University of Kelaniya, Kelaniya, Sri Lanka.

Wednesday, November 13, 1996

(Continued)

P 6 Mosquito Larvicidal Compounds of Stem Bark of *Pongamia pinnata* (L.) Pierre

Sarath Jayasinghe, University of Peradeniya, Peradeniya, Sri Lanka.

P 7 Photodynamic Effect of Haematoporphyrin dimethyl ester against Mosquito Larvae

Veranja Karunaratne, University of Peradeniya, Peradeniya, Sri Lanka.

P 8 A Short Asymmetric Route to Iridoids

Anoma Priyadarshanie Mudalige, University of Peradeniya, Peradeniya, Sri Lanka.

P 9 Soulattrolide an Inhibitor Of Hiv-1 Reverse Transcriptase from *Calophyllum cordato-oblongum*

W. M. A. P. Wanigasekera, Institute for Fundamental Studies, Kandy, Sri Lanka.

Social Programme

6.30 p.m. - Cultural Show at Hotel followed by Sri Lankan Speciality Dinner

Thursday, November 14, 1996

Review Sessions on SAREC Biochemical Pest Control Project

Chairman: Professor Vijaya Kumar

9.00 a.m. - Introductory remarks on Project by Chairman

9.10 a.m. - Polysaccharides - Biomolecules with Renewed Interest

Per-Erik Jansson, Karolinska Institute, Huddinge Sjukhus, Sweden.

9.40 a.m. - Making the Most of MPLC in Isolation of Natural Products, Utilizing Coarse Normal Silica Gel to its Limits

Peter Baeckstrom, Royal Institute of Technology, Stockholm, Sweden.

10.10 a.m. - Tea

Chairman: Professor Per-Erik Jansson

10.25 a.m. - Aphids - Plants and Volatiles

Jan Pettersson, Swedish University for Agricultural Sciences, Uppsala, Sweden

11.05 a.m. - Biochemical Interactions in Shot-Hole Borer Attack on Tea

Savitri Kumar, University of Peradeniya, Peradeniya, Sri Lanka..

11.35 a.m. - Caffeine And Phenolic Compounds In Shot-Hole Borer Infestation Of Tea

K. M. Swarna Wimalasiri, University of Peradeniya, Peradeniya, Sri Lanka.

Thursday, November 14, 1996

(Continued)

11.55 a.m. - Effect Of Secondary Metabolites On the Development of Shot-Hole Borer Beetle, *Xyleborus fornicatus*

W. Subodhi Karunaratne, University of Peradeniya, Peradeniya, Sri Lanka.

12.10 p.m. - Effect of *Gliricida sepium* extract on Tea Termite, *Glyptotermes dilatatus*

H. M. T. B. Herath, Institute of Fundamental Studies, Kandy, Sri Lanka.

12.35 p.m. - Lunch

Chairman: Dr. Peter Baeckstrom

1.45 p.m. - Biopesticides from Sri Lankan Plants

Vijaya Kumar, University of Peradeniya, Peradeniya, Sri Lanka.

2.15 p.m. - An Insect Ovicidal Compound from *Zingiber purpureum* (*Z. cassumunar*) (Zingiberaceae)

K. A. N. Premaratne Bandara, Horticultural Research and Development Institute, Gannoruwa, Peradeniya, Sri Lanka.

2.35 p.m. - Mosquito Larvicidal constituents of *Piper sylvestre* and *Gnidia galuca*

Kayalvili Alagesan, University of Peradeniya, Peradeniya, Sri Lanka.

2.55 p.m. - Antifungal Compounds From *Murraya gleniei* Root Timber

D. B. Mahinda Wickramaratne, University of Peradeniya, Peradeniya, Sri Lanka

Thursday, November 14, 1996

(Continued)

3.15 p.m. - Tea

3.30 p.m. - 6.00 p.m. SAREC PROJECT DISCUSSION

(A Bus will leave the Hotel at 3.40 p.m. and return from Kandy at 5.45 p.m. for those who do not wish to participate in the Discussion).

Social Programme

7.00 p.m. - Symposium Dinner

Friday, November 15, 1996

Symposium Excursion

7.00 a.m. - Symposium Excursion to Dambulla, Sigiriya and Kandalama

7.30 p.m. - Return to Hotel

PROFESSOR M. U. S. SULTANBAWA

Professor Mohamed Uvais Sideek Sultanbawa, the first Professor of Chemistry at the University of Peradeniya, celebrated his seventy fifth birthday this year. He was largely responsible for developing Peradeniya as a centre of excellence for research in Natural Products Chemistry. He believed that a University without research was no University at all and left no stone unturned in trying to convince the powers-that-be that the advancement of the frontiers of knowledge through research enriched the quality of teaching. He was indeed one of the very few who contributed to the transformation of the University system from a mere degree awarding institution into an institution of learning.

During the early years of his career, a University academic who indulged in research was considered a curiosity and even, a nuisance. At a time, when some took the easy way out and made their only contribution to academia by donning a tie, coat and cloak and delivering lectures, Professor Sultanbawa took it upon himself to develop the research element in the Department of Chemistry at the University in Colombo. With the assistance of Mr. B. J. P. Alles, who later distinguished himself in the field of educational policy, Professor Sultanbawa began working on reaction mechanisms. His work on the reactivity of vinylic halogen in unsaturated acids and the formation and stability of β-lactones was a follow up of research he had carried out at Imperial College, London.

Professor Chandrasena had initiated work on the study of the medicinal plants of Sri Lanka as far back as the 1930s. This work had been followed up by Drs. L. B. de Silva and R. O. B. Wijesekera at the Medical Research Institute. Although Professor Sultanbawa too realised the importance of reasearch on the flora of Sri Lanka and encouraged Professor G. P. Wannigama to enter the field, it took many years for Professor Sultanbawa himself to actively work in this area.

He took over the development of the Department of Chemistry at the new Science Faculty at Peradeniya in the early 1960s. The time available for research became practically zero due to his academic and administrative commitments at Peradeniya, the need to travel to Colombo every week to contribute to the Special Degree teaching programme and finally, the necessity to acquire proficiency in Sinhala.

Professor Sultanbawa spent an inspiring sabbatical year with Professor W. S. Johnson at Stanford in 1967. They used a non-enzymatic biogenetic like olefin cyclisation to generate five asymmetric centres specifically to form a steroid molecule. This biomimetic synthesis won the prestigious Roussell prize for the most significant development in Organic Chemistry for Professor Johnson in 1969. The synthesis was recognised as having the potential to revolutionise the industrial synthesis of steroids by making them available from petroleum by-products. Unfortunately, the rapid escalation of oil prices in the early 1970s made this proposition no longer practicable.

Soon after his return from the United States, Professor Sultanbawa laid the foundations for his illustrious research career in Natural Products Chemistry. The Flora of Sri Lanka consists of about 3300 species, nearly a quarter of which are endemic to the island. Medicinal uses are usually described for the non-endemic plants, not the endemic plants. It was these plants, used in India in their Ayurvedic system of medicine, which had influenced our own indigenous medicine. Professor Sultanbawa's main interest was in the study and exploitation of the endemic plants of Sri Lanka. Since the collection of these plants in the field proved difficult without a knowledge of their Sinhala and Tamil names, Prof. Sultanbawa initiated the preparation of a glossary of all Sri Lankan plants.

In this and his future research programme, he had the dedicated assistance of Professor S. Balasubramaniam, one of our finest field botanists.

The help of Mrs. Samudra Weerasekera, who was at that time in charge of the Departmental library, with her contacts with Ayurvedic practitioners and her excellent secretarial skills was contributed to the success of this project.

Professor Sultanbawa realised soon that there could be no research without external funding. He embarked on a major drive to solicit support for his research programme. He convinced the United States Dept. of Agriculture (USDA) of the usefulness of his work on the timber trees of Sri Lanka and was able to attract one of the biggest grants at that time. He was also able to inspire his research assistants like Dr. Sarath Gunasekera, Professor R. Somanathan and Dr. Gowsala Subramaniam, all of whom are now holding responsible positions in research in the United States, to work long hours at the bench under very difficult conditions. Perhaps the greatest impetus to Natural Products Chemistry at Peradeniya came from the visit of Professor, at that time, Dr. Kitagawa, even at that time a well-experienced Natural Products Chemist. Professor Kitagawa spent six months working tirelessly at the bench inculcating a sense of dedication and purpose among members of the group.

During this period, all spectroscopic analysis had to be carried out abroad, usually in Japan through Professor Kitagawa's good offices. Indeed, until the early 1970s the Department had to rely on friends abroad for all its spectral data. The many international publications regularly published by the Sultanbawa group would never have seen the light of day without the assistance of many such friends like Professor R. H. Thomson and W. D. Ollis.

Professor Sultanbawa dreamt of the day when all the spectral analysis could be carried out in Sri Lanka. He convinced the British Council that no purpose would be served in supporting exchange staff without providing the instruments they would need to contribute to the Department's programmes. Against all predictions, the British Council donated what was at that time, state of the art UV and IR Spectrophotometers, spending much more on them

than they did on the Exchange Lecturer. He then coaxed the Vice Chancellor of the University into paying for a Varian T-60 NMR spectrometer, a no mean achievement when one considers the difficulties in obtaining foreign exchange. This has proven to be the most valuable asset the Department had acquired at that time. The instrument is almost twenty years old and surprisingly still functional, considering the many stresses and strains which Sri Lanka's temperamental power supply has made it go it through.

The 1970s was the glorious period in the research career of Professor Sultanbawa. The USDA project was the backbone of his success. Over 125 tropical plant species were studied in depth. There were studies of plants of families such as *Guttiferae*, *Dipterocarpaceae*, *Ebenacea*, and the *Celastraceae*. Many novel compounds were isolated and their structures determined.

Professor Sultanbawa realised all along the importance of biological screening. He established two screening programmes, one for alkaloids and saponins and the other for antifungal and antibacterial activity. With these two programmes, research took on a multidisciplinary approach.

The four year USDA supported project resulted in twenty eight publications in the leading international Journals of the day - a remarkable result when one considers the trying conditions under which this work was carried out. Many of those who worked with Professor Sultanbawa hold high positions in research, unfortunately mostly in the United States. Professor Kitagawa retired recently after a distinguished career as a leading natural products chemist. Professor Sultanbawa and six of his colleagues at Peradeniya were nationally recognised with the Presidential award for Scientific Achievement in 1986.

Professor Sultanbawa's interest in bioactivity made him undertake a World Health Organisation project on anti-fertility compounds from plants. Peradeniya became one of six centres which worked on the project aimed at developing alternative methods of population control. This brought with it

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BIOLOGICALLY ACTIVE CONSTITUENTS OF TROPICAL AND SUBTROPICAL PLANTS

Wolfgang Kraus

Department of Chemistry, University of Hohenheim,
70593 Stuttgart, Germany.

Plant extracts are widely used as pesticides and in folk medicine in tropical and subtropical areas. In connection with our search for botanical pesticides, extracts of a number of tropical and subtropical plants collected in Cameroon, Kenya, Namibia, Zimbabwe, India, Indonesia and the Philippines have been tested for insecticidal, insect antifeedant, insect growth regulating, bactericidal, fungicidal, molluscicidal, anthelmintic, nematicidal and cytotoxic activity.

We report on bioassays used for screening of crude extracts and pure compounds and on isolation, structure determination and biological activity of some active principles.

THE BRIDGE OVER RESEARCH & DEVELOPMENT

R. O. B. Wijesekera

Ceylon Institute of Scientific & Industrial Research
363, Bauddhaloka Mawatha, Colombo 7, Sri Lanka.

In many developing countries, some scientific research is carried out most times under the most severe conditions of deprivation. Yet often observers complain that no research is done. Scientific research alone need not result in visible development. Scientific research gives rise to knowledge. Knowledge can give rise to Technology which in turn can give rise to new products or processes, and these are sometimes visible to the trained lay observer.

Because technology can give rise to processes and products which are more visible than scientific knowledge, it is often more acceptable, and hence the futile debate of basic research versus applied research. In fact knowledge, does not automatically give rise to readily usable technology, and thence development. This has to be realised by adequate investment, first in scientific research, which is costly, and then in technology development which is even more costly and finally in the development itself such as the development of new products. This last phase is the costliest of them all.

In developing countries like Sri Lanka, there is some funding (albeit at mere subsistence level) for scientific research, almost exclusively provided through the government with all its attendant bureaucratic impediments. There is, however, no funding for the development of technology and for the finalisation of technology that has been successfully developed at the pilot plant stage. Such funds are often known as "venture capital" or "risk capital", simply because they are high risk ventures and failure levels can be high. Normal sources of lending or investment require the ensurement of returns within a small period of time, sometimes even requiring guarantees of returns. Thus there has evolved venture capital sources, which are prepared to take high risks, to obtain high return which are occasional and long term.

In developing countries, mechanisms to invest in research in areas of low probability (but if successful, high possibility) are not there. Recently, there has been some thinking on a mechanism to set up a fund for 'technology incubation' which will make the resultant finished technology more visible to venture capital and other capital sources, encouraging them to invest. But however good the mechanisms, it will not work without the commitment of the government and the private sector. A partnership venture for technology development has to be forged to move research results into commercial realisation. This illusive bridge has to be built and fostered, if the country is to bring its scientists into play in its efforts at industrial development.

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Pimpinella monoica - A SOURCE OF BIOACTIVE COMPOUNDS

A. Banerji

Bio-Organic Division, Bhabha Atomic Research Centre,
Trombay, Mumbai-400 085, India

The insect deterrent properties of the extracts of the plant, *Pimpinella monoica* (Umbelliferae) prompted us to undertake a detailed phytochemical investigation which resulted in the isolation of several bioactive furochromones and furocoumarins. Though *Pimpinella* species are known to contain furocoumarins, the preponderance of furochromones in *P. monoica* was rather unusual. The antifeedant activity was attributed to the presence of khellin, visnagin and isopimpenellin. Structure-activity relations between the furochromones were studied. Biogenetic inter-relationships between the constituents have been delineated. In addition to known compounds, four compounds belonging to a new class of oxygen heterocycles were isolated and the major compound was characterised as pimolin, a dimer of visnagin. A minor component ($C_{39}H_{30}O_{12}$) has been characterised as a trimer of visnagin. The positions of the oxygenated functionalities and conformation make this molecule a potential candiate for host-recognition. Our work on the bio-organic chemistry of *P. monoica* will be summarised.

IDENTIFICATION AND SYNTHESIS OF THE SEX PHEROMONES OF *Bonagota cranaodes* (LEPIDOPTERA: TORTRICIDAE)

C. Rikard Unelius, A. Eiras,[*] P. Witzgall,[**] A. Kovaleski,[***] M. Bengtsson,[**] R. Mozuraitis, E. F. Vilela,[†] J. P. Chambon[‡] and A.-K. Borg-Karlson

Department of Organic Chemistry, Royal Institute of Technology, S-100 44 Stockholm, Sweden; [*]Setor de Semioquimicos, Laboratorio de Protecao de Plantas, CCTA, Universidade Estadual do Norte Fluminense, Campos, RJ, 28045-620, Brazil; [**]Department of Chemical Ecology, Swedish University of Agricultural Sciences, S-230 53 Alnarp, Sweden; [***]Departamento de Entomologia, EMBRAPA/CNPUV, Vacaria, RS, 25200 Brazil; [†]Departamento de Biologia Animal, Univ. Federal de Vicosa, Vicosa, MG, 36570 Brazil; [‡]6INRA, Station de Zoologie, Versailles, FRANCE

The pheromone of the apple leaf roller, *Bonagota cranaodes* (Lepidoptera: Tortricidae) was identified by GC-MS analysis of female gland extracts and field trapping of males with synthetic compounds. The solid phase micro extraction method (SPME) proved to be highly selective in detecting volatiles emitted by calling females.

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1. A. E. Eiras, A. Kovaleski, E. F. Vilela, R. S. F. Frighetto, N. Frighetto, *Resumos Cong. Brasil. Entomol.*, **14**, 86 (1983).
2. A. E. Eiras, L. R. K. Delmore, J. R. P. Parra, M. P. R. Pique, E. F. Vilela, A. Kovaleski, *An. Soc. Entomol. Brasil* , **23**, 251 (1994).
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SYNTHESIS OF BIOLOGICALLY ACTIVE NATURAL PRODUCTS WITH A DIFFERENCE

A. V. Rama Rao

A V Rama Rao Research Foundation, 54 Sai Enclave, Habshiguda, Hyderabad 500 007, India.

The author's determination to create a school of excellence in India for the synthesis of biologically active compounds met with several obstacles, which could be attributed to incidents in the long history of chemistry in a developing country. In spite of various difficulties faced by the speaker, he evolved diverse methodologies leading to the sythesis of several biologically active natural products such as anthracyclines, fredericamycin, cervinomycins, arnarosin (antitumor agents); macrolides which include FK-506 and rapamycin (immunosuppressants) and his recent work on cyclo peptides including vancomycin.

He will be highlighting some of these examples with a view to build enthusiasm and commitment among young chemists to achieve good science, which has national relevance.

OVERCOMING PROBLEMS IN DEVELOPING A NATURAL PESTICIDE FROM THE NEEM TREE (*Azadirachta indica*)

E. David Morgan, G. R. Jones and A. P. Jarvis

Department of Chemistry, Keele University, Keele, Staffordshire, England

The seeds of the Neem tree contain the most promising natural insecticide discovered in recent years. The major compound contributing to this insecticidal activity, Azadirachtin, has been known[1] and extensively studied[2] for nearly 30 years. Other biologically active compounds present in the seeds have been identified and their activity recorded. Yet, in spite of a strong popular movement to turn to natural pesticides, Neem is still not widely available for use. It is important to understand why this is so, and what can be done to overcome the problems.

Commercial introduction of Neem was first delayed by problems of supply and quality of seeds, then by problems of extraction and separation of active compounds from the seed oil, and the chemical stability of the extract, and of the stability of the product when sprayed on plants. Most recently we have the problems of regulatory requirements and licencing of products. Most of the problems have been overcome and are discussed in turn. A change of attitude of regulatory authorities is still required in many countries, to see Neem products in the same light as many other plant products that have been in use in the food and agriculture industry for a very long time. Natural Neem products may also have to face competition from industrial tissue cultured material if experimental work now in progress is successful.

In the drier tropics, Neem offers a cheap and readily available plant protectant for poor farmers, that could increase harvest yields and permit the growing of crops at present considered as too vulnerable to pests to be profitable. This presents problems of a very different, cultural and social kind, which will be touched upon, but which are largely beyond the ability of scientists to help.

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1. J. H. Butterworth and E. D. Morgan, *J. Insect Physiol.*, **17**, 969 (1972).
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Kandy, Sri Lanka, November 11-15, 1996

PHEROMONES, HIGH BIOACTIVE NATURAL PRODUCTS AS SIGNALS IN CHEMICAL INFORMATION OF INSECTS

Examples from butterflies and ants

H. J. Bestmann

Institute for Organic Chemistry, University of Erlangen-Nurenberg, Germany

A short review is given on the development of chemistry and biology of pheromones. Furthermore, the state of art of analysis and structure elucidation of pheromones is reported as well as the systematics of sex pheromones in Lepidoptera and some aspects of biogenesis. A number of structure activity relationships are discussed.

In the second part, first systematical investigations on ant trail pheromones are reported as well as the first results of investigations with respect to the biosynthesis of such pheromones.

A CHEMICAL ATTRACTANT SYSTEM FOR THE COCONUT PEST, *Rhynchophorus ferrugineus*

N. E. Gunawardena, S. Dissanayake and H.M.W.K.B. Herath, F. Kern*, O. Vostrowsky* and H.J. Bestmann*

Department of Chemistry, University of Kelaniya, Sri Lanka; *Institute of Organic Chemistry, University of Erlangen-Nurnberg, Erlangen, Germany.

A chemical attractant system has been developed to trap the major coconut pest, *Rhynchophorus ferrugineus* (Coleoptera: Curculionidae). Fermenting coconut sap (toddy), coconut bark steam distillate and 4-methyl-5-nonanol (ferrugineol), the aggregation pheromone of *R. ferrugineus*, were investigated as potential field attractants. Gas chromatography with electroantennographic and FID detectors in parallel (GC-EAD) and GC-MS was used to identify the attractants. Ferrugineol was synthesized in 61% overall yield by the coupling of butylmagnesium bromide and 2-methyl-pentanal.[1]

Of the coconut sap constituents, n-pentanol showed the highest and n-propanol slightly lesser EAG activity. Bark steam distillate contained two host attractants, 4-hydroxy-3-methoxystyrene, produced by the decarboxylation of 4-hydroxy-3-methoxycinnamic acid, and α-nonanoic acid lactone. Synthetic equivalents of these attractants showed synergism when combined in a ratio of 1:1 (v/v).[2]

In a binary choice test with olfactometer, the highest attractant properties were elicited by pentanol and propanol, while in a field assay, only ferrugineol and the host attractants lured adult weevils. When ferrugineol was combined separately with the short-range attractants, pentanol and propanol, the combined ferrugineol and pentanol (1:1, v/v) caused the highest trap catch. This combination was field attractive for over 60 days.[3]

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NAPTHOQUINONOIDS FROM NATURE: DEVELOPMENT TOWARDS ANTIPARASITIC AGENTS

B. Hazra, S. Acharya, R. Sarkar, R. Ghosh*, A. K. Saha and A. Banerjee

Department of Pharmacy, Jadavpur University, Calcutta-700 032, India;
*Department of Chemistry, Tripura University, Agartala-799 004, India.

The pronounced biological activities shown by naturally occurring quinonoid compounds have far-reaching consequences of pharmaceutical, toxicological and technical relevance. Recently, a number of naphthoquinonoids are being investigated for potential clinical applications against several parasites,[1] some of them being responsible for the opportunistic infections in AIDS patients. In our laboratory, a bisnapthoquinonoid, diospyrin, isolated from the stem bark of *Diospyros montana* Roxb., was found to possess significant antiprotozoal activities.[2] Subsequently, several derivatives of diospyrin have been prepared in order to modify the toxicity of this natural product for increasing the therapeutic index.[3] From the structure-activity relationships obtained so far, it has been observed that hydroquinonoid derivative (DIII) of diospyrin dimethyl ether is more effective than the quinonoid analogues. DIII showed the IC_{50}(μM) of 2.4 against the multidrug-resistant K1 strain of *Plasmodium falciparum*, as compared to 523 for its quinonoid precursor, and 0.2 for the standard drug chloroquine diphosphate.[4] The ED_{50} (μM) values for DIII were 0.7 and 2.17 against *Trypanosoma brucei brucei* blood-stream from trypomastigotes and *Leshmania donovani* amastigotes, respectively, *in vitro*.[5] At a concentration of 3μM, DIII caused 100% inhibition of the intracellular amastigotes of *Trypanosoma cruzi in vitro*, the ED_{50} for the standard drug nifurtimox being 2.7 μM.[5] More derivatives in this series are under development through synthesis and concurrent bioassays. Some of these results will be presented.

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BIOLOGICALLY ACTIVE SAPONINS FROM SRI LANKAN PLANTS

U. L. B. Jayasinghe and G. P. Wannigama*

Institute of Fundamental Studies, Hantana Road, Kandy, Sri Lanka;
*Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

In a continuation of our studies towards the discovery of biologically active compounds from Sri Lankan plants, we have chemically investigated *Diploclisia glaucescens* and *Anamirta cocculus* (Family Menispermaceae) and *Pometia eximia* (Family Sapindaceae). Preliminary investigations showed that these three plants contained saponins.

Chemical investigation of the methanol extracts of these plants led to the isolation of nineteen saponins. Of them, fifteen triterpenoidal saponins were found to be new natural products. Some of them showed insecticidal, anti-inflammatory and strong molluscicidal activity.

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BIOLOGICALLY ACTIVE SECONDARY METABOLITES FROM MARINE ORGANISMS

E. Dilip de Silva

Chemistry Division, Open University of Sri Lanka, P. O. Box 21, Nugegoda, Sri Lanka

Oceans represent a vast and largely unexplored resource for the discovery of novel metabolites with interesting biological activities. The vast majority of marine natural products isolated to date have come from four groups of organisms: macro algae, sponges, coelenterates and echinoderms.[1]

Our recent work has largely focused on marine invertebrates, mainly tropical sponges. From the sponge, *Cymbastela* sp. collected in Papua New Guinea we have isolated two different families of cytotoxins, the geodiamolides and the hemiasterlins/criamides. Geodiamolides are a unique group of depsi-peptides composed of three amino acids and a C_{12} polyketide fragment. Hemiasterlins are tripeptides with unusual amino acids.[2] Both geodiamolides and hemiasterlins have high potential as anti-tumor drugs. The potent protein phosphates inhibitor, motuporin was isolated from the sponge *Theonella swinohei* also from the same location.[3]

Nudibranchs are a group of brightly coloured soft bodied animals belonging to phylum Gastropoda. We have investigated nudibranch species from the coastal waters of Sri Lanka and have characterized a number of interesting metabolites including highly functionalised spongian diterpenoids, sesquiterpene isonitriles and isoquinoline quinones.[4]

The chemistry and biological activities of these metabolites will be discussed.

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STUDIES ON USE OF PLANT PRODUCTS FOR AGRICULTURE PEST MANAGEMENT

Mangala D. Manandhar, H. P. Acharya, P. Basnet, J. K. Prasain and *W. C. Taylor

Tribuvan University, Nepal ; *University of Sydney, Australia

Alcoholic extracts of *Swertia chirata* Hamilt, *S. augustifolia* Buch-Ham, *Taxus wallichiana* (zucc.) Pilger and *Boeninghaunesia albiflora* were screened for their cytotoxic, anti-feeding and insecticide activities. The extracts of *Swertia* species and *B. albiflora* have shown marked activities whereas *T. wallichiana* has shown only a mild activity. Detailed chemical investigation of *B. albiflora* has led to the isolation of a number of coumarins and triterpenes including some benzene derivatives. The essential oil components of this plant were also estimated using GC mass.

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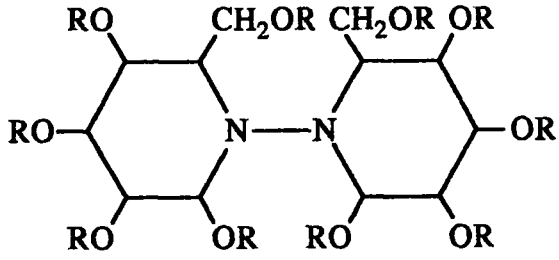
AN ANTIFUNGAL BIS-AZASUGAR FROM *Aspergillus fumigatus*

D. T. U. Wijayaratne, K. P. Abeywickrama* and P. D. F. Chandanie*

Department of Chemistry and *Department of Botany,
University of Colombo, Colombo, Sri Lanka.

Aspergillus fumigatus, isolated from mushroom compost[1] was grown in yeast-starch liquid medium for 14 days. After the filtration of mycelium, the filtrate was freeze-dried and extracted into methanol. The methanol fraction was purified using column chromatography with methanol/methylene chloride mixtures as eluent and preparative TLC on cellulose plates to isolate bis-azasugar **1**. Its mass spectral molecular ion at *m/e* 356 and its NMR spectra and those of its acetate **2** were in accordance with the structures shown. Azasugar **1** showed antifungal activity against *Cladosporium*.

Bis-azasugar are known to have other interesting biological properties.[2]



Compound 1 R = H
Compound 2 R = Ac

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SEARCH FOR BIOACTIVE COMPOUNDS IN THE *Calophyllum* SPECIES OF SRI LANKA

H. R. W. Dharmaratne

Natural Products Programme, Institute of Fundamental Studies, Kandy, Sri Lanka.

The pantropical *Calophyllum* species (Family Clusiaceae) consists of more than 150 species of which 13 are found in Sri Lanka. Of them, ten of them are claimed to be endemic to the island and only *C. inophyllum*, *C. tomentosum* and *C. walkeri* are used in the traditional systems of medicine.

Work on the Sri Lankan *Calophyllum* was initiated in the early seventies by Sultanbawa and his research group. Early work was mainly focused on chemical investigation of various parts, particularly with chemotaxonomic interest. Until the early nineties, out of hundreds of secondary metabolites isolated from *Calophyllum* species, bioactive compounds reported were the piscicidal pyranocoumarins of *C. inophyllum*. A number of pyranocoumarins isolated recently from *C. lanigerum* showed strong anti-HIV activity,[1] and one of them calanolide is undergoing clinical trial. This result prompted us to focus our attention on the bio-activity of the Sri Lankan *Calophyllum*, with special reference to antiviral/HIV activity. So far, nearly 49 xanthones, 29 coumarins, 16 chromone acids and some triterpenoids have been reported from this species. Of these, 34 xanthones and 14 coumarins have been reported to be present in local *Calophyllum* species.[2] This clearly indicates the richness of secondary metabolites in the Sri Lankan endemic species.

Soulattrolide and inophyllum D reported from *C. monii* and *C. inophyllum* were found to inhibit HIV-1 RT. Antiviral/HIV activity studies of pyranocoumarins such as cordatolide A, cordatolide B and oblonguloid from the endemic species are in progress. Apart from the coumarins, the xanthones have been reported to show antibacterial[3] and antifungal activities. The *Calophyllum* species is a very rich source of prenylated and pyranoxanthones and the bioactivity of these metabolites is a promising field for investigation.

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SEARCH FOR USEFUL LEADS FORM MARINE ORGANISMS

Nobuhiro Fusetani

Laboratory of Aquatic Natural Products Chemistry
Graduate School of Agriculture and Agricultural Life Science,
The University of Tokyo, 1-1-1 Yayoi, Bunkyo-ku, Tokyo 113, Japan.

Marine organisms, particularly marine invertebrates, have proved to be a rich source of pharmaceutically active metabolites, some of which are under clinical trials. For more than 15 years we have been trying to discover potential drugs from Japanese marine invertebrates, which resulted in the isolation of more than 150 new compounds, many of which are structurally novel and biologically interesting.[1]

In the earlier stages of our research we focussed on the discovery of antibacterial, antifungal, and cytotoxic/antitumor metabolites mainly from marine sponges. We have isolated a diverse array of active compounds ranging from simple terpenoids to complex polypeptides, including such important substances as halistanol sulfates, discondermins, kabiramides, calyculins, 13-deoxytedanolides, cinachyolide A, theopederins, ritterazines and polytheonamides.

Later, our research has expanded to the discovery of inhibitors of enzymes associated with diseases as well as of antagonists of interleukin 6 and muscarinic acetylcholine receptors and N-type Ca^{2+} channels. Again we could isolate various types of active compounds, among which cyclotheoamide A is most interesting from the viewpoints of chemical structure and biological activity. Cyclotheonamide A, which is a cyclic peptide containing two new amino acids is highly inhibitory against serine proteases including thrombin, trypsin and plasmin. It has become an important model compound for antithrombin drugs due to its unique structure and potent activity.

In this lecture I will focus on the structures and bioactivities of important compounds mentioned above, and discuss marine organisms as potential sources of drugs/leads.

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NATURAL PRODUCTS IN AGRICULTURE

Louis-Pierre Molleyres

Leadfinding Natural Products, Ciba-Geigy Crop Protection
4002 Basel, Switzerland

The constant growth of the world population and the need to produce more food can lead to undesirable consequences including the loss of uncultivated areas and deforestation: Production should increase whilst environmental damage should be minimized.

One way of discovering new pesticides capable of fighting most types of crop plagues consists of finding in nature itself new weapons with insecticidal, fungicidal, herbicidal and antiparasiticidal properties.

This approach will be presented with emphasis on the following aspects:

- Source and specific properties of natural products
- Isolation of secondary metabolites
- Synthesis of natural products
- Development of new products for agriculture
- Biological and toxicological requirements, costs, position in the crop protection market

These aspects will be illustrated by successful examples from different companies including our activities and experiences at Ciba.

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IS NATURAL PRODUCTS RESEARCH OF RELEVANCE TO AYURVEDA ?

Ajith Abeysekera

Dept. of Chemistry, University of Sri Jayawardenapura,
Nugegoda , Sri Lanka.

Research projects dealing with medicinal plants often justify their relevance by stating that the plants in question are used in ayurveda. The research methodology itself involves phytochemical analysis of the plant with the emphasis being on interesting new structures. As bioactivity is considered to be important, fractions and compounds are screened, but sometimes with no consideration of the ethnomedical usage of the plant. Occasionally, judgement is passed on the ayurveda system or on the usefulness of the plant in question for a particular ailment, based on these results.

This mode of research on medicinal plants is conditioned by the philosophy and requirements of the developed world and geared towards developing drugs for inclusion in western pharmacopoeias. Its relevance to the development of ayurveda is questionable. With increasing attention being given throughout the world to the intrinsic merits of alternative medical systems, it is worthwhile to look at the development of alternative modes of research.

Activity guided fractionations based on bioassays relevant to the ethnomedical usage, is more likely to lead to results that can be used to rationalise the ayurvedic system, develop new drugs and standardize existing drugs. An assesment must be made of the relative contribution of the different fractions to the overall activity of the plant and the existence of synergism should not be disregarded.

The final test of a drug is its clinical efficacy. The standard clinical trial used to test one drug against another, violates the principles of ayurvedic treatment, which is more patient oriented rather than disease oriented. Further, ayurveda rarely uses a single drug in isolation for a given illness. Scientific evaluation of the clinical efficacy of ayurvedic drugs awaits the development of a suitable research methodology.

Perhaps the one area in which phytochemical research on medicinal plants is of direct relevance to ayurveda is in providing baseline data for the chemical standardization of ayurvedic drugs.

HYPOGLYCEMIC EFFECTS OF SOME MEDICINAL PLANTS

M. Mosihuzzaman, Nilufar Nahar, A. K. Azad Khan,* Liaquat Ali* and Begum Rokeya*

Department of Chemistry, University of Dhaka, Dhaka-1000, Bangladesh;
*Research Division, BIRDEM, Dhaka, Bangladesh.

Studies of hypoglycemic effects of medicinal plants is a collaborative research programme between the Department of Chemistry, University of Dhaka and the Bangladesh Institute of Research and Rehabilitation of Diabetes, Endocrine and Metabolic Disorders (BIRDEM) Dhaka. The programme started a few years ago with the aim of finding out new hypoglycemic agent(s) from medicinal plants for diabetic management and to find out the mechanism of action of the active component(s). During the last few years protocols for extraction of plant materials and testing methodologies of the extracts *in vivo* and *in vitro* have been developed by our research group towards achieving the goal. A good number of plant extracts from local sources and also from the region were tested for their acute glucose lowering effects on healthy/nondiabetic and diabetic (NIDDM & IDDM) model rats at different prandial states. A good number of the plant material showed significant blood glucose lowering effects. Some of the active extracts were tested for long-term/chronic glucose lowering effects on the model rats where the parent extract showed highest activity. Lipid profile and vitamin E level of the extract-fed model rats were also studied.

Some sub-fractions of the active extract and pure compounds isolated from the active extracts were tested on pancreatic islets and single B-cells for their insulin releasing effects. A few of them showed significant insulin releasing properties.

From our studies of a large number of plant it is found that hypoglycemic agents are chemically quite different in nature and their modes of action are different. Low molecular secondary metabolites, free amino acids, small molecular peptides, some polar glycosides and polymeric carbohydrates have promising effects.

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ISOLATION AND CHARACTERIZATION OF TERPENOIDS AND GLYCOSIDES FROM *Pterospermum acerifolium* AND *P. semisagittatum*

Nilufar Nahar, Iqbal Rouf Mamun and M. Mosihuzzaman

Department of Chemistry, University of Dhaka, Dhaka-1000, Bangladesh.

Extracts of *Pterospermum acerifolium* and *P. semisagittatum* (leaves and bark) were tested for their oral hypoglycemic effects on healthy and diabetic model rats and significant hypoglycemic effects (p=0.01-0.001) were found. Chloroform and methanol extracts of *P. semisagittatum* were also tested for anticancer and anti-AIDS efficacy. Methanol extract of the bark showed positive anti-AIDS activity and positive test against colon cancer.

The chloroform and methanol extracts of *P. acerifolium* were fractionated by repeated silica gel flash column followed by centrifugal chromatography (Chromatotron) and Sephadex LH-20 column followed by HPLC. Two pure pentacyclic triterpenoids having molecular formula $C_{30}H_{50}O$ and another with molecular fomula $C_{30}H_{48}O$ and three sesquiterpenoid glucosides having molecular fomula $C_{21}H_{31}O_{10}$ and $C_{19}H_{31}O_8$ were isolated. The terpenoids and sesquiterpenoid glucosides were characterized by UV, IR, high resolution NMR and mass spectroscopy. ^1H-^1H, ^1H-^{13}C correlation spectroscopy, HMQC, HMBC and NOESY techniques were used for identification..

BIOLOGICAL ACTIVITY OF SOME MEDICINAL PLANTS OF SRI LANKA

W.R.Wimalasiri, Manel M. Goonasekara* and P. H. P. Fernando**

Division of Biochemistry, Faculty of Dental Sciences and **Department of Biochemistry, Faculty of Medicine, University of Peradeniya and *Department of Biological Sciences, Rajarata University, Mihintale.

A large number of asthma patients in Sri Lanka prefer indigenous medicines to allopathic medicine because of their effectiveness. The treatments include prepared mixtures or aqueous decoctions of the fresh and/or dried plant parts. Twenty commonly used plants were identified and twelve remedies and aqueous extractions of these plant materials were prepared for bioassay using asthma induced guinea-pigs.

The importance of plants as a source of anti-fertility drugs is well recognized. Such an agent would be useful in the developing countries and a drug given in the form of a crude extract or semi-purified isolate would be acceptable to rural people, in addition to being freely available.

Of 160 extracts of medicinal plants tested by the Peradeniya centre of a WHO multicentre progamme, the highest anti-fertility activity was found to be in*Calotropis gigantea* root bark extracts. Activity directed fractionations were carried out to obtain semi-purified or purified active constituent/s. Activity was tested using female Sprague-Dawley rats according to WHO bioassay protocol MB 30. A mixture containing 80% of a major component, probably a steroid glycoside, was found to be active at a dose of 5.16mg/Kg with 0% pregnancy rate.

Hyperlipidaemia is a predisposing factor for coronary heart disease, a major cause of mortality among the Sri Lankan population. This is due to deposition of cholesterol in the arterial walls as a result of elevated blood cholesterol. Hypolipidaemic activity was tested with aqueous extracts of *Allium sativum*, *Murraya koenigii*, *Sida acuta* and *Tinospora cordifolia* using hyperlipidaemic male Sprague-Dawley rats. Animals were dosed orally for six weeks and sera analyzed for their lipid profiles. The results showed that there was a reduction in the total:HDL cholesterol ratio in all test groups, with *S. acuta* most effective in improving lipid profile (1.45) compared with control (1.83).

This work was supported by the grants from IFS (Sweden), HRP(Sri Lanka) and University of Peradeniya

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BIOACTIVE BISBENZYLISOQUINOLINE ALKALOIDS FROM MENISPERMACEAE

Biswapathi Mukherjee and Tapan Seal

Department of Pharmacology, Dr. B. C. Roy Post-graduate Institute of Basic Medical Sciences, Calcutta - 700 020, India

So far around four hundred bisbenzylisoquinoline alkaloids have been reported from plants belonging to diverse families. These alkaloids have been categorised into twenty seven different structural types depending on their oxygen substitution pattern and mode of attachment between the two individual benzylisoquinoline moieties. Most of these types are available in the family Menispermaceae. Besides (+)-tubocurarine, an important member of the series with neuromuscular blocking effects and having clinical application as an adjunct to anesthetic reagents in abdominal surgery, many compounds have shown promising results as anti-leishmanial, anti-malarial, anti-tumour, hepatoprotective and anti-histaminic agents. This triggered up the research on these alkaloids in search of valuable pharmacophores of medicinal importance.

Chemical and pharmacological investigations on *Tiliacora racemosa* Colebr. (Menispermaceae) yielded (+)-tiliacorine, (+)- tiliacorinine, (+)-tiliamosine, (+)-tiliarine, (+)-nortiliacorinine, (+)-N-methyltiliamosine, (+)-N-methyl-tiliarine, (+)-tiliaresine, (+)-tiliacosine and (+)-tiliasine. Structures of these complicated alkaloids were elucidated by the application of sophisticated spectral techniques. (+)-Tiliacorine showed muscle relaxant activity comparable to (+)-tubocurarine. (+)-Tiliarine was found to inhibit significanly human melanoma cell (G-361) *in vitro* multiplication without affecting normal fibroblast cell line (CRL-1906) under the same experimental condition.

ALKALOIDS OF SOME BIOACTIVE PLANTS OF SRI LANKA

L.S.R. Arambewela, P. G. Madawela, S. Weerakoon and C. K. Ratnayake

Ceylon Institute of Scientific & Industrial Research
363, Bauddhaloka Mawatha, Colombo 7, Sri Lanka.

Rauvolfia canescens, R. densiflora, Taeberanaemontana divaricata and *Alstonia scholaris* are four common medicinal plants of Sri Lanka.

R. canescens and *R. desiflora* are being used as substitutes and adulterants for *R. serpentina* in India. Their crude alkaloid extracts showed sedative activities[1] evaluated by the rat hole board technique. Nine known alkaloids and the new alkaloids, lankanescine, lankafoline and sridensine were isolated from *R. canescens* .

Tabernaemontana divaricata is used in indigenous medicine for the treatment of skin disease.[2] Its crude alkaloid extract was shown[3] to be active against *Staphylococcus aureus.* It contained ten known alkaloids and the new 11-methoxy-N-methyldihydropericyclivine.

Alstonia scholaris is used as a remedy for fevers and in the treatment of malaria, diarrhoea and dysentery. In our studies,[4] the alkaloid extract of *A. scholaris* exhibited a prominent inhibition of classical activation pathway. Echitimine, picrinine, tubotaiwine and picraline deacetyl were also isolated from this plant.[5]

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ANTILEISHMANIAL ACTIVITY OF JAWAHARENE FRACTION JF_1 TOWARDS *Leishmania denovani* PROMASTIGOTES

Ahindra K. Saha and Ananta K. Das

Department of Pharmacy, Jadavpur University, Calcutta - 700 032, India.

Approximately 7 percent of the global population is estimated to be threatened by Leishmaniasis, a vector-borne parasitic disease.[1] The actual quantum of cases reported worldwide is about 12 million,[2] of which the major share belongs to the third world. The conventional chemotherapy of the disease is not radically foolproof; currently, the recognition of the extensive use of traditional herbal therapy in endemic regions has renewed the interest in evaluation of potential antileishmanials from natural sources. In our laboratory, preliminary investigation was undertaken for the antileishmanial activity of JF_1,[3] a fractional product of Jawaharene,[4] an antitumor antibiotic[5] complex obtained from the mycelia of a fungal strain. *Aspergillus niger*. JF_1 showed a cytotoxic effect on the promastigotes of *Leishmania donovani*. A considerable growth-inhibiting effect of JF_1 was observed, the minimum inhibitory concentration (MIC) of the compound being 10μg/ml (2μg/ml for pentamidine, the standard drug). JF_1 also effectively inhibited the respiration of *L. donovani* promastigotes by about 75 percent at the dose of 40μg/ml; at the same concentration, pentamidine caused 80 percent inhibition.

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EFFECTS OF AZADIRACHTIN ON SPECIFIC TISSUES IN THE LOCUST, *Schistocerca gregaria*

P. A. Paranagama, *R. H. C. Strang and **J. D. Connolly

Department of Chemistry, University of Kelaniya, Kelaniya, Sri Lanka; *Department of Biochemistry and **Department of Chemistry, University of Glasgow, Glasgow, U. K.

Azadirachtin[1] was isolated from the seeds of the neem tree (*Azadirachta indica*) by solvent extraction and flash chromatography in >95% purity and identified by reversed phased HPLC, NMR and melting point determination. Pure azadirachtin was used to obtain the reduced derivative, (22,23)-dihydroazadirachtin and (22, 23-$^3H_2$) dihydroazadirachtin.[2,3] The latter was used to follow tissue uptake, metabolism and excretion in the locust, *Schistocerca gregaria.* It was found that an injected dose of the tracer was removed at a high speed from the haemolymph into many of the locust tissue, most likely by carrier-mediated specific mechanisms. Unlabelled analogues of azadirachtin and dihydroazadirachtin injected in large excess, inhibited the clearance of the tracer to different extents and the results suggested that azadirachtin and its dihydroderivative have different affinities for the uptake mechanism.

Radio-labelled dihydroazadirachtin applied topically to the locusts was shown to penetrate into the insect only to a limited extent whereas a large fraction of the tracer was absorbed into the fat body as well as into the gut, Malpighian tubules and nervous tissues. Binding of dihydroazadirachtin was persistent and not easily replaced. Metabolism of the dihydroazadirachtin was slow and largely restricted to fat body and crop.

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REVIEW SESSION ON SAREC PROJECT ON BIOCHEMICAL PEST CONTROL

The following papers and many of those in the Poster Section represent contributions from scientists involved in the SAREC funded research project on Biochemical Pest Control. The Project was begun in 1994 and involves the Department of Chemistry, University of Peradeniya, The Natural Products Programme of the Institute of Fundamental Studies, Kandy and the Tea Research Institute of Sri Lanka as the Sri Lankan collaborating Institutions. The Swedish partners are the Department of Entomology, Swedish University of Agricultural Sciences, Ultuna, Uppsala, the Clinical Research Centre of the Karolinska Institute at Huddinge, the Department of Organic Chemistry, Royal Institute of Technology, Stockholm and the International Program in Chemical Sciences, Uppsala University,

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POLYSACCHARIDES - BIOMOLECULES WITH RENEWED INTEREST

Per-Erik Jansson

Clinical Research Centre, Karolinska Institute, Huddinge Hospital,NOVUM
S-141 86 Huddinge Sweden

Polysaccharides and carbohydrates in general are ubiquitous molecules in nature and the last twenty years have witnessed an amazing renaissance. Only little was known previously about the biological significance of carbohydrates. For example, the role of starch as reserve substance and cellulose and chitin and skeleton compounds was well understood. Development of carbohydrate chemistry led to the discovery of important molecules like glycoconjugates, especially glycolipids and glycoproteins. Analytical methods like NMR and MS have revolutionised the structure determination of polysaccharides and other carbohydrate materials. Modern chromatography has also simplified life for the chemist.

The increased knowledge of polysaccharide structures that revealed, in some cases an increased complexity but sometimes a higher degree of organisation. The complexity has in many cases consisted of new sugars that could not be analysed with older techniques. One example is C-9 sugars where for a long time only one was known, now several.

In the lecture, polysaccharides will be reviewed in terms of occurrence, structure, biological significance and methods for analysing them. Representative examples will be taken from plant, fungal and bacterial polysaccharides. NMR and MS will be emphasised as some of the most important tools of modern analytical chemistry.

MAKING THE MOST OF MPLC IN ISOLATION OF NATURAL PRODUCTS, UTILIZING COARSE NORMAL SILICA GEL TO ITS LIMITS

Peter Baeckstrom and Zandra Jiron

Department of Chemistry, Organic Chemistry, Royal Institute of Technology, S-100 44 Stockholm, Sweden

The following topics will be discussed: Group separation of compounds with similar retention factors from crude extracts or directly from plant material by means of gradient elution. Rough separation of compounds within groups of interest. Fine tuning to achieve singular compounds.

The underlying principals will be illustrated by means of computational models.

A method for simple calculations of minimum absorbent and solvent consumption for separating pairs of compounds by liquid chromatography using binary mixture based on determining the k values of the two compounds will be presented. The calculation, made in a spread sheet program, predict surprisingly short columns. Stimulations reveal that TLC data, used to selected solvent composition according to prevalent recommendations, can lead to grossly wasteful conditions.

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APHIDS - PLANTS AND VOLATILES

Jan Pettersson

Dept. of Entomology, Box 7044, Uppsala, Sweden

Volatile substances may be important messengers in aphid behavioural ecology both for social interaction and in host plant discrimination and search for good feeding sites. Aphids have high fecundity and a short developmental time. Under normal conditions, *Rhopalosiphum padi*, can make 2.5 generations in the crop[1] during one growing season. This means that a specific feeding site for an aphid colony has a short duration and the rapid development of the aphid population is a provocation for the host plant and stress reactions of different types may occur. There is experimental evidence that when aphids attack a cereal plant[3] or other crop plants,[4] volatiles are released which induce aphid antixenosis in neighbouring plants. The response of the cereal plant is limited in time but it is also correlated to a change in biomass allocation, stimulating root growth at the cost of leaf elongation.

At a certain aphid density, compounds are released that stimulate the mobility of apterous *R. padi.*[5] This could be expected to be part of a spacing mechanism which would decrease plant stress at a feeding site - a more sensitive mechanism than the initiation of alate individuals which is the best known aphid response so far to high population density.

In a joint Sri Lanka/Swedish project, studies were made on the odour based behavioural responses of the cowpea aphid, *Aphis craccivora.* Density dependent social interactions were seen and the response of the cowpea plant to aphid attack shows a temporal dynamic with regard to aphid attractivity.[6]

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BIOCHEMICAL INTERACTIONS IN SHOT-HOLE BORER ATTACK ON TEA

N. Savitri Kumar

Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

The Shot-hole borer (SHB) infestation of tea involves interactions between several organisms-the shot-hole borer beetle (*Xyleborus fornicatus*), the tea plant (*Camellia sinensis*), the symbiotic Ambrosia fungus (*Monacrosporium ambrosium*) and a bacterial symbiote. The microbial complex apparently makes nutrition from the woody substrate available to the beetle.The tea clone TRI 2023 is the most tolerant or least susceptible, while the clone TRI 2025 is the most susceptible to attack by the beetle.

The resistance of a particular tea clone may be due to the fact that the SHB has rejected a particular clone (tolerant/resistant) or because SHB has selected another clone (susceptible). The Ambrosia fungus synthesises chemicals required by the beetle. The degree of infestation by SHB may depend on the growth of the fungus lining the galleries. Another basis for clonal resitance/tolerance could be that the tolerant clone TRI 2023 provides a less favourable environment for the fungus.

Therefore the chemical composition of tea clones may play a major role in the resistance/susceptibility of certain tea clones to SHB attack. We have studied the composition of neutral sugars, and caffeine and phenolic contents of healthy and beetle infested stem samples from the two clones, and their effect *in vitro* on growth and development of the Ambrosia fungus and the SHB.

Formation of phenols and polyphenols may constitute part of the biochemical defense response of the tea plant to attack by the beetle. Formation of these compounds occurs via the conversion of phenylalanine to cinnamic acid. This reaction is catalysed by the enzyme *phenylalaninelyase* (PAL). An increase in PAL activity indicates an increase in the synthesis of phenolic compounds. We are studying PAL activity in healthy and beetle infested tea stems in order to determine whether there is any reltaionship between PAL activity and the resistance of tea clones.

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CAFFEINE AND PHENOLIC COMPOUNDS IN SHOT-HOLE BORER INFESTATION OF TEA

K. M. Swarna Wimalasiri and N. Savitri Kumar

Dept. of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

Shot-hole borer beetle (*Xyloborus fornicatus*) bores galleries in tea stems and introduces spores of the symbiotic fungus *Monacrosporium ambrosium* into th egalleries.[1] Caffeine, a major constituent of tea stems, was shown to have an inhibitory action against the symbiotic fungus *M. ambrosium*. Attack by the shot-hole borer beetle led to a higher accummulation of caffeine in the more resistant clone.[2]

Fungi are known to be capable of eliciting an active defense response from plants. the defense response may lead to the secretion of enzymes and the accummulation of secondary plant metabolites such as phytoalexins.[3] Therefore, *in vitro* experiments were carried out to determine whether attack of the tea stem by either the fungus *M. amrbosium* or the beetle led to the accummulation of caffeine.[4] Microtome sections of tea stems were treated with the fungal mycelia, spore suspension, culture filtrate, hydrolysed fungal mycelia (0.1N TFA) and the beetle. Caffeine content was determined (HPLC) after a 20 h. incubation period. No significant differences were observed in the caffeine contents fo the tea stem sections.

Plants produce a variety of secondary metabolites such as phenolics, alkaloids and terpenoids that act against herbivores and fungal pathogens. Polyphenols are important in defense mechanisms of plants. Attack by insects and pathogenic fungi are known to increase the phenolic contents and may lead to resistance of different cultivar. The phenolic contents of tea stems were analysed using HPLC. A significant difference was observed in the composition of the phenolic extracts from healthy and infected tea stems of the two clones.

Acknowledgement: Authors wish to thank Messers B. S. Mendis and A. S. T. B. Wijetunga for preparing microtome sections of tea stems.

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EFFECT OF SECONDARY METABOLITES ON THE DEVELOPMENT OF THE BEETLE, *Xyleborus fornicatus*

W. Subodhi Karunaratne, N. Savitri Kumar and Vijaya Kumar

Department of Chemistry, University of Peradeniya.

The shot-hole borer *Xyleborus fornicatus* of tea was cultured *in vitro* to study its life cycle. This was carried out by incorporating different types of secondary metabolites into the culture medium to see their effect on the developmental stages of the beetle. The beetles were introduced into the different culture media and observations made on tunnel boring, egg laying, developing stages of the larvae, pupation, number of healthy female adults emerging and the time taken to complete each stage. Five different media with six replicates each were studied for several generations.

Previous work has shown that the chemical composition of tea clones which are susceptible and resistant to beetle attack could differ with respect to the content of saponins, caffeine, tannins and other metabolites. This experiment was designed and carried out to show the effect, if any, of the secondary metabolites on the beetle. It was seen that the time taken to complete each developing stage and the whole life cycle is prolonged by addition of the secondary metabolites. The number of offspring produced was the same in both the control (which contained yeast) and the experiment where yeast was absent. There was a reduction of offspring when tannic acid was added and a greater reduction with caffeine. The number of emerging beetles however increased when caffeine and tannic acid were present together in the medium.

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EFFECT OF GLIRICIDIA SEPIUM EXTRACT ON TEA TERMITE, Glyptotermes dilatatus

H. M. T. B. Herath

Natural Products Programme, Institute of Fundamental Studies, Kandy, Sri Lanka.

Gliricidia sepium (Family Legumunosae) is grown as a shade plant in the tea plantations of Sri Lanka.Biological investigations have shown that the heartwood of this plant is toxic to the tea termite, *Glyptotermes dilatatus*, which is threat to the low country tea plantations of Sri Lanka.[1] There have been previous reports on the insecticidal activity of parts of*G. sepium* . The chemical investigation of *G. sepium* has been mainly focused on the leaves. Fifteen allelopathic compounds including a coumarin have been isolated.[2] Three flavonoids[3,4], two flavonoid glycosides[5] and related compounds have also been reported from *G. sepium.*

In our bioassays, we found that of the different extracts, the hexane extract of the heartwood of the species has the greatest attraction for the tea termites. We describe the chemical and biological investigation of the hexane extract of the heartwood of *G. sepium.* The isolation and the structural elucidation of two new isoflavans, 8,2'-dihydroxy-7,4'-dimethoxyisoflavan (1) and 7,4'-dihydroxy-3'-methoxyisoflavan (2), and three other compounds, new to the species afromosin (3), stigmasterol(4) and stigmasterol glucoside (5) are also reported. Afromosin (3) is described as a powerful anti-tumour agent.6 Tentative structures of the four other flavonoids including a dimer isolated from the same extract will also be described.

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BIOPESTICIDES FROM SRI LANKAN PLANTS

K.A.N.Premaratne Bandara, Champani A. Balasuriya, Vijaya Kumar, Kavitha Thillainathan and Nela Weerasekera.

Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

Dichloromethane and methanol extracts of different parts of selected Sri Lankan plants were screened against *Aedes aegypti*, *Plutella xylostella*, *Callosobruchus maculatus* and *Aphis craccivora*. and attempts made to isolate their active constituents using the mosquito larvicidal screen.

167 Extracts from 44 plant species were screened for larvicidal activity against the second instar of *A. aegyptii*. Extracts which showed good activity were those from *Grewia macrocos* stem bark, *Xylopia nigricans* root and leaves, *Pongamia pinnata* stem bark, *Gnidia glauca* flower and stem bark, Bahuchi seed and stem bark, *Piper* sp. leaves and twigs, *Lobelia nicotianifolia* whole plant, *Erythrospermum zeylanicum* whole plant, *Zingiber purpureum* rhizome, *Curcuma zedoaria* rhizome and *Piper argyophyllum* leaves and creeper. Eleven extracts showed moderate activities while the remainder did not show any activity.

13 extracts from 6 plant species were screened against*P. xylostella* larvae at 4000 ppm using leaf discs treated with plant extracts. Active extracts included those from *G. macrocos* stem bark, *Z. purpureum* rhizome, *P. argyrophyllum* leaves and creepers and *C. zedoaria* rhizome. Screening against *C. macculatus* was carried out using residual films of extracts or by treating cowpea seeds. Of 11 extracts screened, only *G. macrocos* stem bark, *Z. purpurium* rhizome and *C. zedoaria* rhizome were active.

A microapplicator or Potter's spray tower was used in screening for activity against *A. craccivora.*

Bioassay directed fractionation/enrichment of these extracts gave several compounds of varying activity.

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AN INSECT OVICIDAL COMPOUND FROM *Zingiber purpureum* (*Z. cassumunar*) (ZINGIBERACEAE)

K. A. N. Premaratne Bandara, Vijaya Kumar and Ramesh C. Saxena*

Department of Chemistry, Univeristy of Peradeniya, Peradeniya, Sri Lanka;
*International Centre for Insect Physiology and Ecology, Mbita Point, Kenya

The dichloromethane extract of *Zingiber purpureum* (Rh.) was found to be ovicidal to cowpea bruchid, *Callosobruchus macculatus*. Bioassay directed fractionation of the extract showed that the active principle was 4-(3',4'-dimethoxyphenyl)buta-1,3-diene. This compound has been previously isolated from the same source and its insecticidal activity on the larvae of *Spodoptera littoralis* was recently reported.[1,2]

In the present investigations, we found that both the extract and the phenyl butanoid compound have a strong ovicidal effect on bruchids. Hatchability of eggs deposited by both pre-mated or post-mated adults treated with the extract (5 mg/vial) on untreated seeds and untreated adults on treated cowpea seeds (2.5 mg/ 5 g seeds) were reduced by the treatment. The adult bruchids and cowpea seeds treated with lower concentrations of the compound (0.5mg/vial and 2.5mg/5g seeds respectively) showed similar effects. However, pre-mated adults were found more susceptible than the post-mated adults for their ovicidal activity.

Only transparent egg shells were found in affected eggs and no larval development or yolk material was found inside them. However, in some affected eggs, partially developed and dead larvae were found confined to one half of the egg white the other half of the egg was clean. These findings suggested that the compound has ovicidal and maternally applied larvicidal activity on bruchids. Borkovec (1985)[3] indicated the importance of this type of population controlling agents as insect controlling agents. The mechanism of this ovicidal activity of *Z. purpurium* is being investigated.

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MOSQUITO LARVICIDAL CONSTITUENTS OF *Piper sylvestre* and *Gnidia glauca*

Kayalvily Alagesan and Vijaya Kumar

Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

Piper sylvestre Lam. (Piperaceae), the most widespread naturalized *Piper* species in Sri Lanka and *Gnidia glauca* Fresen. (Thymelaeaceae), reputed to be an indigenous insect control agent in rice fields were investigated for mosquito larvicidal activity. The dichloromethane extracts of *P.sylvestre* creeper and leaves and *G.glauca* stem bark were found to be active. Several species of *Piper* have been investigated previously and insecticidal and growth-reducing lignans and isobutylamides have been isolated.[1]

The extracts were subjected to bioassay-guided fractionation in an attempt to isolate the active components.

Five compounds were isolated from the *P.sylvestre* extract and one of them, an amide was found to be very active, two lignans were found to be moderately active and a long chain hydrocarbon was weakly active against the second instar of *Aedes egyptii* larvae. A flavanoid from *G.glauca* was also found to show activity.

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ANTIFUNGAL COMPOUNDS FROM *Murraya gleniei* ROOT TIMBER

D. B. Mahinda Wickramaratne, Vijaya Kumar and N. K. B. Adikaram*

Dept. of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka;
*Dept. of Botany, University of Peradeniya, Peradeniya, Sri Lanka.

Bioassay directed fractionation using an antifungal TLC bioassay[1] led us to isolate several indoles from the cold dichloromethane extract of *M. gleniei* root timber. Unlike the root bark[2] and leaves which were enriched with coumarins, the root timber was found to contain four major indoles. Three of them were found to be active. Structural elucidation of these compounds using spectroscopic and chemical methods will be discussed. The compounds have shown varying but impressive activity against five fungi, *Alteraria tenuis*, *Cercospora nicotianae*, *Fusarium* sp., *Botridiplodia theobromae* and *Cladosporium cladoporioides.*

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SCREENING OF PLANT EXTRACTS FOR NEMATICIDAL ACTIVITY

Champani Balasuriya

Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

Screening of Sri Lankan plant extracts for activity against the following parasitic nematodes infesting tea, *Camellia sinensis*: the root lesion nematode, *Pratylenchus loosi* Loof syn. *P. cofeae* of Loos and the borrowing nematode, *Radophalus similes* (Cobb), Thorne forms part of the SAREC Research Project. *P. loosi* and *R. similes* are known to significantly reduce tea crop causing loss of productivity. A nematode laboratory has been established at Peradeniya and the screening for nematicidal activity is being initiated.

Dichloromethane and methanol extracts of different parts of selected plant species are screened for their nematicidal activity against 2nd, 3rd and 4th instar larvae and active adult females of *R. similis* and *P. loosi*. Larval and adult mortality 24, 48 and 72 HAT can be used to estimate their LC_{50} values. Bioassay directed fractionation can be used to isolate active constituents of nematicidal extracts.

Ten active adult female nematodes are introduced by means of a dropper into the plant extract in acetone solution contained in a cavity glass slide. Cavity blocks containing acetone solution are kept as control and five replications are carried out for each treatment. The above procedure is repeated using 2nd, 3rd and 4th instar larvae. The cavity blocks are kept in a BOD incubator at 25°C. The percentage mortality of nematodes is recorded at intervals of 24, 48 and 72 h. Observations are recorded by counting live and dead nematodes under a stereoscopic binocular microscope after keeping for 1 hr in sterile distilled water placed on a cavity glass slide. The procedure is also used to determine the hatchability of nematode eggs in the presence of plant extracts.

R. similis and *P. loosi* are cultured on carrot (*Daucas carota* L.) discs.

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EFFECT OF CAFFEINE AND TANNIC ACID ON THE GROWTH OF *Monacrosporium ambrosium*

Nivandana K. Bandaranayake, N.Savitri Kumar, Vihanga Pahalawatta and Santhushtika Udakara

Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

Caffeine was found to inhibit the growth of the fungus *Monacrosporium ambrosium* [1] and to accumulate to a greater extent in tea clone TRI 2023, which is more resistant to attack by the Shot-Hole Borer beetle, *Xyleborus fornicatus*. Caffeine, at concentrations far lower than in tea stems, was found to inhibit oviposition of beetle in laboratory cultures. Therefore the inhibitory effect of caffeine may be masked by other phytochemicals, such as phenols, found in tea stems. Phenolic compounds are widely distributed in plants and are known to be involved in plant defence mechanisms. Phenols, polyphenols and complex polyphenols (the vegetable tannins) reversibly associate with a wide range of substrates including proteins, polysaccharides and nitrogenenous metabolites like alkaloids. Thus potassium chlorogenate has been isolated as a 1:1 complex with caffeine from green coffee beans.[2]

The effect, if any, of polyphenol-caffeine interactions on the growth of *M. ambrosium*. was studied. It was found that both caffeine (2 mg) and the hydrolysable polyphenol, tannic acid (2 mg) inhibited the growth of the fungus in a TLC bio-assay, but a mixture of caffeine (2 mg) and tannic acid (4 mg) did not have an inhibitory effect on the fungus.

While caffeine and tannic acid reduced the average number of conidia in colonies of *M. ambrosium* grown on agar, the effect of caffeine was found to be much greater. The inhibitory effect of caffeine on sporulation was much less when mixtures of caffeine and tannic acid were used. Spore germination was reduced in the presence of caffeine, although mixtures of caffeine and tannic acid had no effect. Non-covalent intermolecular bonding between caffeine and tannic acid apparently reduces the inhibitory effect of caffeine.

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EXTRACTION AND PURIFICATION OF AZADIRACHTIN FROM NEEM SEED

B. D. Karunananda Bombuwela* and Peter Baeckstrom

Department of Organic Chemistry, Royal Institute of Technology, Stockholm, Sweden; *on leave from Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

The Neem tree, *Azadirachta indica* (A.Juss) (Family Meliaceae) is a common tree in South Asia. Neem products have been used as pesticides since they are safe to non-target organisms including humans. Neem is bitter in taste due to the presence of an array of compounds collectively known as limonoids.[1]

Neem seeds contain azadirachtin and many other compounds which have been shown to possess pesticidal, bactericidal and fungicidal properties.[2] Azadirachtin B, $C_{35}H_{44}O_{16}$, which has been isolated in the pure state,[1] is the principal limonoid of neem seed. It belongs to a group of ten related mostly insecticidal compounds[3] called azadirachtin A-K.

We report a method of isolating pure azadirachtin B as a colourless amorphous powder from neem seed kernel using column chromatography. Azadirachtin was obtained pure after three MPLC columns, the method being both silica gel and solvent efficient. Neem seed kernel (45 g) gave azadirachtin B (250 mg) using silica gel (87 g) and solvent (2.35 l).

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NEUTRAL SUGARS IN TEA STEMS AND THEIR EFFECT ON *Monacrosporium ambrosium*

R.M.Thushari P. Bombuwala, Nivandana K. Bandaranayake and N.Savitri Kumar

Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

The shot-hole borer beetle (*Xyleborous fornicatus*) has a symbiotic relationship with the fungus, *Monacrosporium ambrosium* . Morphological and chemical plant factors may result in the selection of a tea clone by the beetle. Lack of sufficient levels of phytochemicals, such as carbohydrates, may affect the growth of the fungus/beetle and/or the selection of tea stems by the beetle. The neutral sugar composition of the clones 2023 and 2025, and their effect on growth and development of *M.ambrosium* was investigated.

Sugar analysis was carried out on methanol extracts of healthy and infected pencil thick tea stems.The extracts were partitioned with butanol, the water layer was separated and freeze dried. The neutral sugar composition was determined as alditol acetates by GC. Glucose was the major (47-71%) reducing sugar in all samples. Galactose and mannose were found in low concentrations while an appreciable amount of inositol (7-28%) was detected. Inositol is known to be essential for the growth of certain strains of fungi.[1]

Mycelial growth sporulation and spore germination of *M.ambrosium* in culture media containing different neutral sugars were studied.

Sporulation was observed using a haemocytometer 14 days after incubation. Germination of spores was counted for eight hours at one hour intervals in a liquid medium containing glucose and inositol as the carbohydrate source. Sporulation was observed in media containing i. glucose, ii. inositol, and iii. combinations of glucose and inositol. Sporulation was reduced in mixtures of glucose and inositol. Germination of spores was 60% in glucose and in inositol it was 26%. Combinations of glucose and inositol; (1:1, 3:1, 5:1) respectively reduced the germination of spores.

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HOST ATTRACTANTS FOR THE RICE BUG, *Leptocorisa oratorius*

N. E. Gunawardena, S. Dissanayake, H. M. W. K. B. Herath and A. B. Attygalle*

Department of Chemistry, University of Kelaniya, Sri Lanka; Department of Chemistry, *Cornell University, Ithaca 14853-1301, New York, U. S. A.

Leptocorsia oratorius (Hemiptera: Coriedae) is a serious pest on rice in Asia and the Pacific region. Investigations have been conducted on a host attractant for this pest based on the observation that it is highly attractive to the milk producing rice grains. Steam distillation of the green leaves, flowering panicles, milky grains, mature grains and yellow leaves of the rice plant were subjected to electroantennogram (EAG) assay. The highly active samples were subjected to gas chromatographic-electroantennogram detection GC-EAD and GC-mass spectrometry (GC-MS) analysis.

The results revealed that steam volatiles of the milky grains were highly attractive to *L. oratorius* (EAG 1.01 mV) and this was significantly different ($P<0.05$, ANOVA, Scheffe's test) from those of the other parts of the rice plant. The flowering rice panicles also had relatively high EAG activity (0.83 mV) compared to those of the other parts (EAG range 0.58-0.68 mV). GC-EAD analysis of the rice steam distillate indicated n-octanal, n-nonanal, n-decanal, undecanal and *cis*-2-pentanol as major active constituents.

In a behavioural bioassay (binary choice test) using olfactometer, a synthetic equivalent of the milky rice grain volatiles elicited the highest response in *L. oratorius* (64% rice bugs attracted) compared to the other syntehtic combinations assayed. The above response, however, was lower than that of the natural attractant *viz.* the steam volatiles of the milky grains which attracted 80% of rice bugs under similar conditions.

MOSQUITO LARVICIDAL COMPOUNDS OF STEM BARK OF *Pongamia pinnata* (L.) Pierre.

Sarath Jayasinghe and Vijaya Kumar

Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

Pongamia pinnata (L.) Pierre (Sinhala: Magul-karanda) belongs to Family Fabaceae.[1] Parts of the plant have been extensively used as in crude drugs for the treatment of tumours, piles, skin diseases, wounds and ulcers, especially in India.[2]

The dichloromethane and methanol extracts of the stem bark of *P. pinnata* showed mosquito larvicidal activity against the second instar larvae of *Aedes aegypti*. Bioactivity guided fractionation of the dichloromethane extract gave an active compound (85% mortality at 2.5 ppm), which was deduced to be a flavonoid by spectroscopic methods. Nine inactive compounds were also isolated.

The active compound was found to be present in the methanol extract as well.

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PHOTODYNAMIC EFFECT OF HAEMATOPORPHYRIN DIMETHYL ESTER AGAINST MOSQUITO LARVAE

Yeranja Karunaratne, Vijaya Kumar, Sakuntala Jayasinghe and Nela Weerasekara

Department of Chemistry, University of Peradeniya, Peradeniya, Sri Lanka.

Photosensitizers of the porphyrin and phthalocyanine series, when exposed to visible light, generate singlet oxygen. This property of haematoporphyrin and its derivatives have been successfully used in tumour phototherapy.[1]

We have investigated the photodynamic effect of haematoporphyrin dimethyl ester against the 2nd instar larvae of *Aedes aegypti* (L.) [Diptera:Culicidae] under sunlight.

Two sets of dilution series of haematoporphyrin dimethyl ester in water ranging from 2.5-0.625 ppm were made. Mosquito larvae were introduced and one set was kept under sunlight (green-house conditions) and the other in the dark. In each experiment, an untreated control (without haematoporphyrin dimethyl ester) was maintained.

The treated larvae kept under sunlight showed 90% mortality even at 0.625 ppm after 48 h. The treated larvae kept in the dark showed no mortality even after 48 h. Both the untreated larvae kept under sunlight and in the dark showed 0% mortality after 48 h.

Reference

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A SHORT ASYMMETRIC ROUTE TO IRIDOIDS

C. Rikard Unelius and Anoma Priyadarshanie Mudalige*

Department of Organic Chemistry, Royal Institute of Technology, Stockholm, Sweden; *on leave from Natural Products Division, Institute of Fundamental Studies, Kandy.

The stereoselective synthesis of novel iridoid monoterpenes was attempted. The key step involves an efficient intramolecular [4+2]-cycloaddition of an enamine derivative of 8-oxocitral,[1] wherein the enamine moiety acts as a chiral inductor.[2] Earlier acid- or base-catalyzed ring formations of 8-oxocitral led to dial-lactol mixtures with unspecified stereochemistry at the ring junction, sometimes with double bond migration.

Although gastrolactone and *cis-cis* nepetalactone are relevant for studies on the chemical communication of chrysomelid beetles[3,4] and aphids[5] respectively, the most important achievement is the synthesis of gastrolactol, which seems to be an ideal, or at least a very useful intermediate in the synthesis of more elaborate iridoids. The double bond in the five-membered ring can be used for further addition reactions, allylic oxidations or halogenations. A synthesis of loganin seems to be within reach.

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SOULATTROLIDE AN INHIBITOR OF HIV-1 REVERSE TRANSCRIPTASE FROM *Calophyllum cordato-oblongum*

H. R. W. Dharmaratne, W. M. A. P. Wanigasekera, E. M. Greenwood* and J. M. Peszzuto*

Natural Products Programme, Institute of Fundamental Studies, Kandy, Sri Lanka; *Department of Medicinal Chemistry and Pharmacognosy, College of Pharmacy, Chicago IL 60612-7231, USA.

Screening assays have shown that *Calophyllum* species are potent inhibitors of HIV-1 RT. The pyranocoumarins, calanolide A and its derivatives,[1] inophyllum[2] and soulattrolide[3] are claimed to be active principals. We are studying the anti viral/HIV studies on the *Calophyllum* species of Sri Lanka. The hexane extract of *C.cordato-oblongum* (Cluseacea) afforded soulattrolide and cordatolide B.[4] Soulattrolide showed 98.20% inhibition at 200μg/ml and IC_{50}, r^2 0.187μM in the anti-HIV reverse transcriptase activity test. According to literature[1] calanolide A has an activity of 0.32μM. These results show that soulattrolide has a slightly higher activity than calanolide A which has been developed.[1] Its activity was previously reported using HIV-1 RT inhibitory screening assays.[3]

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4. H. R. W. Dharmaratne et al. *Phytochemistry*, **24**, 155 (1985).

LIST OF PARTICIPANTS

BANGLADESH

NAHAR, NILUFAR, PROF. Fax: +880-2-865583
Department of Chemistry,
University of Dhaka,
Dhaka-1000, Bangladesh.

MOSIHUZZAMAN, M., PROF. Fax: +880-2-865583
Department of Chemistry,
University of Dhaka,
Dhaka-1000, Bangladesh.

GERMANY

BESTMANN, H. J., PROF. Fax: +49-9131-866864
Institute for Organic Chemistry,
University of Erlangen-Nurenberg,
D-91054, Erlangen, Germany.

KRAUS, WOLFGANG, PROF. Fax: +49-711-459-2951
Department of Chemistry,
University of Hohenheim,
D-70593 Stuttgart, Germany.

INDIA

BANERJI, ASOKE, DR. Fax: +91-22-5560750
Head, Bio-Organic Division,
Bhabha Atomic Research Centre,
Trombay, Mumbai-400 085, India.

HAZRA, BANASRI, DR. Fax: +91-33-3370843
Department of Pharmacy,
Jadavpur University,
Calcutta 700 032, India.

MUKHERJEE, BISWAPATHI, DR. Fax: +91-33-2413222
Department of Pharmacology,
B. C. Roy Post-Graduate Institute of Basic Medical Sciences,
244B, Acharya J. C. Bose Road,
Calcutta 700 020, India.

RAMA RAO, A. V., DR. Fax: +91-40-675605
A. V. Rama Rao Research Foundation,
54 Sai Enclave, Habashiguda,
Hyderabad 500 007, India.

SAHA, AHINDRA K., DR. Fax: +91-33-3370752
Department of Pharmacy,
University College of Medicine,
Jadavpur University,
Calcutta-700 032, India.

JAPAN

FUSETANI, NOBUHIRO, PROF. Fax: +813-5684-0622
Laboratory of Aquatic Natural Products Chemistry,
Graduate School of Agriculture Life Sciences,
University of Tokyo,
1-1-1 Yoyoi,
Tokyo 113, Japan.

NEPAL

MANANDHAR, MANGALA, PROF. Fax: +977-1-226964
Central Department of Chemistry,
Tribuvan University,
Kathmandu, Nepal.

SRI LANKA

ABEYSINGHE, SARATH, DR. Fax: +94-052-8311
Biochemistry Division,
Tea Research Institute,
Talawakelle, Sri Lanka.

ALAGESAN, KAYALVILI, MS. Fax: +94-8-88018
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.

ARAMBEWELA, LAKSHMI, DR. Fax: +94-1-686567
Ceylon Institute for Scientific Industrial & Research,
363, Bauddhaloka Mawatha,
Colombo 7, Sri Lanka.

BALASURIYA, B. M. CHAMPANI A., MS. Fax: +94-1-88018
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.

BANDARA, K. A. N. PREMARATNE, MR. Fax: -
Division of Entomology,
Horticultural Crop Research and Development Institute,
Peradeniya, Sri Lanka.

BANDARA, B. M. R., PROF. Fax: +94-8-88018
Department of Chemsitry,
University of Peradeniya,
Peradeniya, Sri Lanka.

BANDARANAYAKE, NIVANDANA K., MR. Fax: +94-8-88018
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.

BASNAYAKE, CHANDANA S., MR. Fax: +94-1-686567
Ceylon Institute for Scientific Industrial & Research Institute,
363, Bauddhaloka Mawatha,
Colombo 7, Sri Lanka.

BOMBUWELA, B. D. KARUNANANDA, MR. Fax: +94-8-88018
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.

BOMBUWELA, R. M. THUSHARI P., MS. Fax: +94-8-88018
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.

DE SILVA, DILIP, DR. Fax: +94-1-436858
Chemistry Division,
Open University of Sri Lanka,
P. O. Box 21,
Nugegoda, Sri Lanka.

DHARMARATNE, H. R. W., DR. Fax: +94-8-232131
Institute of Fundamental Studies,
Hantane Road,
Kandy, Sri Lanka.

HERATH, H. M. T. B., DR. Fax: +94-1-232131
Institute of Fundamental Studies,
Hantane Road,
Kandy, Sri Lanka.

GUNAWARDENA, N. E., DR.
Department of Chemistry,
University of Kelaniya,
Kelaniya, Sri Lanka.
Fax: +94-1-521485

JAYASINGHE, SARATH, MR.
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-88018

JAYASINGHE, U. L. B., DR.
Institute of Fundamental Studies
Hantane Road,
Kandy, Sri Lanka.
Fax: +94-8-232131

KARUNARATNE, W. SUBODHI, MS.
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-88018

KARUNARATNE, VERANJA, DR.
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-88018

KUMAR, N. SAVITRI, PROF.
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-223294

KUMAR, VIJAYA, PROF.
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-223294

MUDALIGE, ANOMA P., MS.
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-88018

PARANAGAMA, PRIYANI A., DR.
Department of Chemistry,
University of Kelaniya,
Kelaniya, Sri Lanka.
Fax: +94-1-521485

WANIGASEKERA, W. M. A. P., MS.
Institute of Fundamental Studies,
Hantane Road,
Kandy, Sri Lanka.
Fax: +94-8-232131

SULTANBAWA, M. U. S., PROF.
89/1, Dickman's Road,
Colombo 5, Sri Lanka
Fax: -

WICKRAMARATNE, D. B. M., DR.
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka
Fax: +94-8-88018

WIJAYARATNE, D. T. U., DR.
Department of Chemistry,
University of Colombo,
Kumaratunga Munidasa Mawatha,
Colombo 3, Sri Lanka.
Fax: -

WIMALASIRI, W. R., DR.
Department of Biochemistry,
Faculty of Medicine,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-32572

WIMALASIRI, K. M. SWARNA, DR.
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-88018

WANNIGAMA, G. P., PROF.
Department of Chemistry,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-88018

SWITZERLAND

MOLLEYRES, LOUIS-PIERRE, DR.
Ciba-Geigy Ltd - Natural Products,
R-1059.605, CH-4002,
Basel, Switzerland.
Fax: +41-61 697 9342

SWEDEN

BAECKSTROM, PETER, DR.
Department of Organic Chemistry
Royal Institute of Technology,
S-100 44 Stockholm, Sweden.
Fax: +46-8-6369936

JANSSON, PER-ERIK PROF. Fax: +46-8-7465230
Clinical Research Centre, Karolinska Institute,
Huddinge Hospital, NOVUM,
S-141 86 Huddinge, Sweden.

LIMINGA, RUNE PROF. Fax: +46-18-183495
Director, International Program in Chemical Sciences,
Uppsala University,
S-752 37 Uppsala, Sweden

PETTERSSON, JAN, PROF. Fax: +46-18-672890
Department of Entomology
Swedish University for Agricultural Sciences,
Box 7044, S-75007 Uppsala, Sweden.

UNELIUS, C. RIKARD, DR. Fax: +46-8-7912333
Department of Organic Chemistry
Royal Institute of Technology,
S-100 44 Stockholm, Sweden.

UNITED KINGDOM

MORGAN, E. DAVID, PROF. Fax: +44-1782-712378
Department of Chemistry
Keele University,
Staffordhire, England

INVITEES TO SAREC REVIEW MEETING

ADIKARAM, N. K. B., PROF. Fax: +94-8-88018
Department of Botany,
University of Peradeniya,
Peradeniya, Sri Lanka.

BRECKENRIDGE, W. R., PROF. Fax: +94-8-88018
Department of Zoology,
University of Peradeniya,
Peradeniya, Sri Lanka.

EDIRISINGHE, JAYANTHI, DR. Fax: +94-8-88018
Department of Zoology,
University of Peradeniya,
Peradeniya, Sri Lanka.

GNANAPRAGASAM, NALINI, DR.
"Cropoptima",
78/3, Pansala Road,
Hatton, Sri Lanka.
Fax: +94-1-591111

KARUNARATNE, ANJANI M., MS.
Department of Botany,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-88018

KARUNARATNE, S. P. H. H., DR.
Department of Zoology,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-88018

MODDER, W. W. D., DR.
Director, Tea Research Institute,
Talawakelle, Sri Lanka.
Fax:

SANTIAPILLAI, C. V. M., PROF.
Department of Zoology,
University of Peradeniya,
Peradeniya, Sri Lanka.
Fax: +94-8-88018

SIVAPALAN, A., DR.
"Cropoptima",
78/3, Pansala Road,
Hatton, Sri Lanka.
Fax: +94-1-591111

WICKRAMASINGHE, R. L., DR.
17, Arethusa Lane,
Colombo 6, Sri Lanka.
Fax: -

OBSERVERS

Teaching Assistants and Final Year Students in Chemistry at the University of Peradeniya who has been invited to participate in some sessions.

ABEYSINGHE, Thushara

DIYABALANAGE, Himashini

FAHIR, Ziyana

FALOON, Sithy

GAMAGE, Chaminda

GANKEWELA, Rajika

HANIFFA, Faleela
JAYASOORIYA, Udeni
KARUNARATNE, Indika
KULARATNE, Sepalika
KUMARIHAMY, B. M. M.
SADIQUE, Azwana R.
RANASINGHE, Athula
RANASINGHE, Mahinda
RATNAYAKE, Mangala
SAMARANAYAKE, Anupama
SiSIRAKUMARI, Nayana

ACCOMPANYING PERSONS

Mr. L. K. Y. Abeytunga
Mrs. Anita Baeckstrom
Mrs. H. J. Bestmann
Mrs. Pradeepa de Silva
Mrs. Michiko Fusetani
Mr. Magnus Jansson
Mrs. Hymayanthi Rama Rao
Mrs. M. U. S. Sultanbawa
Mrs. Marina Wijesekera

TECHNICAL ASSISTANTS

Mr. P. H. S. S. Ariyapala
Mr. G. A. V. Gunasekera
Ms. Achala Karunaratne