# Phytochemical Society of North America Sociedad Fitoquímica de América del Norte Société Phytochimique de L'Amerique du Nord

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# PRESIDENT'S LETTER Phytochemistry and Some Thoughts on the Future of the PSNA

I would like to use the opportunity of my second contribution to the PSNA Newsletter to think aloud, as it were, on what the science of phytochemistry really encompasses at the start of the 21<sup>st</sup> century. My objective, which I will state openly, is to encourage the membership of the society to think more broadly about their field, and to embrace change at a time when technological advances in analytical chemistry and genomics are pushing phytochemistry into the spotlight.

The PSNA is a relatively small society compared to related societies such as the American Society of Plant Physiologists (ASPP), although it does have a stable and significant "core membership". At the last meeting of the Executive Committee of the PSNA in Beltsville, MD, it was decided that a new recruiting drive would be very much in order, and this will be initiated shortly. However, it was also apparent that the society has a problem in "mobilizing" its core membership to attend its annual meetings, in spite of the fact that these feature excellent presentations that form the basis of articles in the highly respected Recent Advances in Phytochemistry series. Why is this? I believe the problem reflects the fact that there is a perception among the membership that recent meetings have only catered to a sub-set of the membership, or, put another way, have appealed to either the "molecular" or "chemical" factions of the membership, but not to both. I would argue that these two branches of our science should be viewed as inseparable.

To start thinking about where phytochemistry is going in the future, let us first consider the past achievements of some scientists who can clearly be classified as phytochemists based on the dedication to them of special issues of the journal Phytochemistry. These include some of the founding members of the society and the journal. Eric Conn, Tom Mabry, Neil Towers, Jeffery Harborne and Clarence Ryan have all made major contributions to phytochemistry, and have trained many of the new generation of phytochemists. The important point is that, together, their work encompasses a variety of sub-disciplines, including chemotaxonomy, ethnobotany, pharmacognosy, ecological biochemistry, enzymology, metabolic biochemistry, and molecular biology. Each has taken a unique approach to understanding the chemical processes that make plants such a rich source of molecular diversity. Understanding this chemical diversity at the level of simply cataloging molecules was never the be all and end all of phytochemistry, but a first step towards utilizing plant natural products to benefit humankind by exploitation of their activities as, for example, medicines or plant protectants.

The enormous power of molecular biology, particularly genomics, now makes it possible, for the first time, to truly exploit our knowledge of plant chemical diversity. This can be done by isolating the genes

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2001 PSNA Annual Meeting

Phytochemistry in the Genomics and Post-Genomics Eras

August 5 - 9

Oklahoma City, Oklahoma

The Phytochemical Society of North America (PSNA) is a nonprofit scientific organization whose membership is open to anyone with an interest in phytochemistry and the role of plant substances in related fields. Annual membership dues are U.S. \$40 for regular members and \$20 for student members. Annual meetings featuring symposium topics of current interest and contributed papers by conference participants are held throughout the United States, Canada, and Mexico. PSNA meetings provide participants with exposure to the cutting-edge research of prominent international scientists, but are still small enough to offer informality and intimacy that are conducive to the exchange of ideas. This newsletter is circulated to members to keep them informed of upcoming meetings and developments within the society, and to provide a forum for the exchange of information and ideas. If you would like additional information about the PSNA, or if you have material that you would like included in the newsletter, please contact the PSNA Secretary and Newsletter Editor. Annual dues and changes of address should be sent to the PSNA Treasurer. Also check the PSNA website at www.psnaonline.org for regular updates.

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#### **PSNA ADVISORS**

Kelsey Downum (2000) John T. Arneson (2001) C. Peter Constabel (2003) Nikolaus H. Fischer (2002) Jonathon Poulton (2003)

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that encode the enzymes and regulatory transcription factors that together constitute the genetic machinery underlying plant natural product biosynthesis. With these materials in hand, it will be possible to generate genetically modified organisms to provide unlimited supplies of complex bioactive natural products, and to design in vitro synthetic methods using immobilized recombinant enzymes (a form of "combinatorial biochemistry"). Elucidation of the three-dimensional structures of natural product biosynthetic enzymes will lead to a better understanding of the exact protein structures that help determine substrate specificity, regiospecificity and kinetics of the enzymatic reactions of natural product biosynthesis. In turn, this knowledge will facilitate the design of novel enzymes for the production of totally new natural products.

In my opinion, gene discovery through molecular biology, genetics or genomics is, if the question being addressed relates to plant products or biochemical processes, a branch of phytochemistry. So too is structural biology when targeted at plant proteins, and genomics when it involves metabolic profiling, or other screens based upon a knowledge of plant metabolism, to identify genes important for any plant process. These molecular approaches now join classical natural product isolation and structural elucidation as the tools of our trade. At the exploitation end, pharmacognosy and ethnobotany may make strange bed-fellows with the genetic manipulation of organisms using natural or "designer" enzymes and regulatory transcription factors, but these new approaches represent an important future direction for the application of phytochemistry to improving the human condition. This is why I find it disappointing that the PSNA has had limited success in bringing together scientists from the different sub-disciplines of phytochemistry as I have defined them above. Someone about to start a functional genomics project aimed at natural product diversity could probably learn more from an hour's discussion with a good ethnobotanist than from any molecular biologist. At the same time, the molecular biologist could probably help a plant chemist think of new ways of utilizing his or her knowledge of natural product chemistry to discover new facets of plant biology.

Collaborations between the members of our society who represent the different ends of the phytochemical spectrum are essential if we are to fully exploit the revolution in molecular science to further our understanding of, and interest in, plant chemistry. To this end, the 2001 meeting of the PSNA will be on the subject "Phytochemistry in the Genomics and Post-Genomics Eras". The meeting will be held in Oklahoma City between August 5 and 9. It will review recent advances in chemical separation and analytical technologies, along with the latest approaches in genetics, genomics and structural biology as they relate to natural product pathway discovery and manipulation. The format will hopefully allow for a better understanding of how these (at first sight) disparate disciplines rely on one another. I hope we can expect a strong attendance from all sectors of the membership. In his accompanying article in this volume of the Newsletter, John Romeo rightly points to the important role that the Recent Advances in Phytochemistry series has had in defining the direction of phytochemistry at certain points in its development. I hope that volume 36, based on the symposium talks at the Oklahoma City meeting, will likewise be a record of a new definition of our science that will keep it at the forefront of advances in the broader field of plant biology.

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Further details of the program for the August 2001 meeting will appear in the next volume of the Newsletter, and in a letter to be sent to existing and potential new members of the society.

I realize that the above views are not unbiased. I speak as a biochemist who has taken molecular approaches to understanding plant natural product pathway regulation, function, and exploitation. Others in the membership may disagree as to whether molecular and genetic techniques can help them address the questions they are interested in. I would like to encourage the membership to air their views on these issues here in the Newsletter, by sending their comments to the editor. Peter Facchini, preferably by e-mail. Comments might also contain suggestions for topics for future meetings of the society. Peter will incorporate a selection of these comments in subsequent editions of the Newsletter.

Richard A. Dixon Samuel Roberts Noble Foundation Ardmore, Oklahoma radixon@noble.org

# FROM THE EDITOR

This issue of the PSNA Newsletter is my first as its new Editor. I accepted the challenge of producing a professional quality newsletter for our society at the request of Vincenzo De Luca. Actually, considering that Vince is my former postdoctoral supervisor, it was somewhat of an offer that I couldn't refuse! My instructions were simple - make the newsletter into something that members of the society will find valuable and want to read. I should say that this first issue has arrived in your hands more by

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shear force of will rather than as the result of cleaver planning. The difficulties faced by past newsletter editors has become evident over the last few months. It has been very difficult to obtain contributions from society members. Thus, the theme of this brief editorial is to emphasize the importance of contributing articles, advertisements, announcements, and other materials to the newsletter. The vitality of this publication is directly proportional to the quality and quantity of the contributions made to it from PSNA members. I would like to extend the comments of Rick Dixon in the President's Letter and appeal to the membership to consider making regular, or even one-time, contributions on any relevant topic to the PSNA Newsletter. There are many ways that interested individuals can contribute. For example, a good way to promote the international flavor of the PSNA would be to have a regular feature article by a 'correspondent'

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# SOUTH OF THE BORDER

#### Mexican Tradition, Phytochemistry and Biochemistry

The richness of Mexican phytogenetic resources is a matter of frequent praise.With a flora of more than 20,000 species, Mexico ranks third in plant diversity. Such abundance in phytoresources, together with the cultural advances of ancient inhabitants. led to the domestication of important crops and the development of an impressive herbolaria or collection of medicinal herbs. Today, traditional healers in rural areas of Mexico outnumber physicians 4 to 1, and still depend on this millenary knowledge to treat all kinds of diseases. Therefore, the social importance of medicinal plants for Mexico should not be overlooked.

The systematic study of Mexican medicinal plants started in 1552 with the elaboration of the Libellus de Medicinalibus Indorum Herbis, also known as the Badiano Codex. This book, which describes the properties of more than 200 plants, was followed by others written through the colonial period. Once Mexico became an independent country, the government founded the Instituto Mèdico Nacional (IMN), where the information available on medicinal plants was analysed and concentrated during the period between 1888 and 1915. Such information was used later in the formulation of the Farmacopea Mexicana which, along with other colonial manuscripts, led to the publication of Plantas Medicinales de Mèxico by Maximino MartÌnez in 1933. Studies on the distribution and

uses of medicinal plants have continued mainly at the Instituto de Ecologia of the National University and at the Instituto Mexicano del Seguro Social (IMSS), and also in many state universities. The contributions on this subject by researchers, such as Robert Bye and Xavier Lozoya, have been particularly important in making Mexican traditional medicine one of the best documented in the Western Hemisphere.

It can be assumed that Dr. Leopoldo Rìo de la Loza established the basis for the development of phytochemistry in Mexico at the turn of the century. His pioneering work revealed the chemical composition of some medicinal plants. Many other scientists have continued the identification of chemical products in plant extracts, while training younger scientists. This approach has yielded an important number of researchers actively working on the identification and characterisation of natural products from Mexican plants. The contributions of Mexican chemists to the knowledge on medicinal plants have been very important and, sometimes, without proper recognition.

So where are the biochemists? Undoubtedly, there is a strong tradition on the study of Mexican medicinal plants. Botanists, chemists, medical doctors, anthropologists and even economists have contributed to it. Why not biochemists? There is not an easy answer to this question since several biochemically-oriented

groups have been operating in Mexico for many years. More recently, groups studying plant biochemistry and molecular biology have appeared in different parts of the country. However, such groups have not shown any interest in studying the metabolic pathways leading to the biosynthesis of the active principles contained in our medicinal plants. The importance of this information is obvious, since a frequent problem related with the use of medicinal plants, or products made from them, is the variability of the pharmacological effects. The understanding of these processes could allow us to control the accumulation of active metabolites. Indeed, there are a few isolated efforts. Fewer than five groups are currently working on the subject at the molecular and biochemical level. However, given the volume of botanical and medical information already available, the number of biochemists involved should be much higher. The use of that information by biochemists, combining their expertise with those of botanists and chemists could lead to the formation of strong, multidisciplinary groups on medicinal plants. Apart from the academic value of such research, the social impact would be considerable. Who will take the first step? Please, send your comments, they will be much appreciated.

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## 2000 ANNUAL MEETING

#### Arthur Neish Young Investigators' Minisymposium

The Arthur Neish Young Investigators' Minisymposium was held for the second consecutive year as part of the 2000 PSNA Annual Meeting held in Beltsville, MD, from August 6 to 10. The theme of this year's minisymposium, "Molecular Manipulation of Alkaloids, Flavonoids, and Terpenoids" fit in well with the overall focus of the meeting, "Regulation of Phytochemicals by Molecular Techniques".

Four young scientists were invited to present their research as part of the minisymposium. Frédérique Hilliou, from the Institute of Molecular Plant Sciences at Leiden University in The Netherlands, discussed her work on the isolation of regulators of genes involved in terpenoid indole alkaloid metabolism in Catharanthus roseus using a T-DNA activation tagging approach. Dr. Hilliou showed that the DNA flanking the T-DNA tag in one positive cultured cell line encoded a jasmonate-responsive AP-2 domain transcription factor called ORCA3 (an acronym for Octadeca-noid-Responsive Catharanthus AP2-domain). Overexpression of ORCA3 in stably transformed C. roseus cell lines increased the expression of genes involved in terpenoid indole alkaloid biosynthesis resulting in increased secondary metabolite accumulation.

Edward Braun from the Department of Plant Biology at Ohio State University described his work on the regulation of flavonoid metabolism by *myb* genes. Several plant *myb* genes have been implicated in the regulation of phenolic compounds including flavonoids and phenylpropanoids. Dr. Braun is interested in the use of evolutionary analyses to make functional inferences about members of the *myb* 

gene family. He has used various molecular techniques to explore the implications of these functional inferences in terms of genome dynamics and metabolic diversity. His analyses suggest that the *myb* gene family underwent a remarkable expansion near the origin of land plants, coincident with the early evolution of some phenolic secondary compounds. More recent expansions in the *myb* gene family are also suggested within the grasses.

The third speaker in the minisymposium was Lukas Mueller from the Carnegie Institute of Washington in Stanford, California. Dr. Mueller discussed recent developments in the establishment of a model for anthocyanin sequestration in plants. He reported that in maize and petunia, glutathione S-trans-ferases (GSTs) are required for the vacuolar sequestration of antho-cyanins. He has recently demonstrated that AN9, the petunia GST required for pigment sequestration, is a flavonoid binding protein, and proposed that AN9 serves as a cytoplasmic flavonoid carrier protein.

Mark Schoenbeck, from the Department of Agronomy at the University of Kentucky, was the final speaker in the minisymposium. Dr. Schoenbeck described his work on the sesquiterpene cyclase gene family in Nicotiana tabacum. He showed that members of the tobacco cyclase gene family have a conserved gene structure within their coding region, but diverge significantly in their flanking genomic sequences, including the promoter regions. N. tabacum is the product of hybridization between ancestors of N. sylvestris and N. tomento-siformis, followed by chromosome doubling that allowed meiotic pairing. Dr. Shoenbeck's

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work has shown that each ancestor likely contributed unique members to the cylase gene family in *N. tabacum*.

In an effort to promote the excellent research and highlight the promising careers of these scientists, a brief biography on each speaker at this year's Young Investigators' Minisymposium is provided below.

#### Frédérique Hilliou

Frédérique Hilliou is currently a postdoctoral fellow at the Institute for Molecular Plant Science, Clusius Laboratory, at Leiden University in The Netherlands. She is studying the isolation of regulators of terpenoid indole alkaloid biosynthesis. Frédérique's supervisors are Drs. Johan Memelink and Jan Kijne and she is supported by a Marie Curie Postdoctoral Fellowship. Frédérique did her Ph.D. on the bioengineering of terpenoid indole alkaloid biosynthesis in Catharanthus roseus from October 1996 to 1999 under the supervision of Drs. Mark Leech and Paul Christou in the Molecular Biotechnology Unit, John Innes Centre, Norwich, UK. This work was sponsored by a John Innes Foundation Fellowship. From 1994 to 1995 Frédérique completed a Masters degree in plant breeding and genetic resources (DEA de Resources Gènètiques et Amèlioration des Plantes). The topics covered by this research were quantitative genetics, genetic resources, biostatistics, population genetics, and biotechnology. Frédérique did her Masters work at the University of Orsay (Paris XI), France and was supported by a grant from the French Ministry of Research.

#### **Edward Braun**

Edward Braun's research program has focused on the use of evolutionary genomics and bioinformatics to elucidate gene function. He is also very interested in understanding patterns of molecular and organismal evolution at a variety of levels. Edward received his Ph.D. in biology from the University of New Mexico in 1996 for his work with Dr. Margaret Werner-Washburne concerning the regulation of gene expression in response to changes in nutrient availability in the budding yeast Saccharomyces cerevisiae. He then accepted a postdoctoral position that was split between the National Center for Genome Resources and the University of New Mexico where he worked on the Neurospora crassa genome project (NGP) with Dr. Donald Natvig. The NGP project provided the first evidence for genes encoding enzymes from secondary metabolism in a filamentous fungus and allowed Edward to document patterns of gene loss in S. cerevisiae. After completing his postdoctoral work with the NGP, Edward moved to the Department of Plant Biology at Ohio State University to work with Dr. Erich Grotewold. He has worked at OSU since 1998 and has been supported by a postdoctoral fellowship from the USDA since 1999. While at OSU he has used a combination of computational and laboratory-based approaches to establish the timing of gene duplications in the plant myb gene family and the types of changes that have occurred during its evolution. Using these data, he has worked on projects to examine the biological role of specific Myb homologs. Edward also has an interest in the use of molecular data to examine the relationships between organisms as well as the application of statistical and computational methods to the analysis of sequence data. Since many myb genes act as regulators of specific phytochemicals he believes that these analyses also provide insights into the evolution of secondary metabolism.

#### Lukas Mueller

Lukas Mueller is a native of Zurich, Switzerland, where he studied biochemistry and biotechnology at the Swiss Federal Institute of Technology (ETH). During that time he became interested in plant secondary metabolites and later did his doctoral dissertation at the University of Lausanne on the biosynthesis of betalain pigments in the mushroom Amanita muscaria and the Caryophyllales. Lukas has just finished a postdoctoral position in Virginia Walbot's lab at Stanford University where he worked on the import of anthocyanins into the vacuole, and recently joined the Arabidopsis Information Resource (TAIR) at the Carnegie Institution of Washington at Stanford. He mainatins a keen interest in plant secondary metabolism and ways in which the pathways could be integrated into the TAIR database.

#### Mark Schoenbeck

Mark Schoenbeck received his BS in agronomy at the University of Missouri and his Ph.D. from the Plant Biological Sciences program at the University of Minnesota. His interest in plant biology research began during his college days, working as a technical assistant in the laboratory and field plots of Dr. Dale Blevins, examining the relationship between mineral nutrition and carbohydrate metabolism. After two years of lab work as an undergraduate, Mark moved to Minnesota to study the developmental physiology and molecular biology of symbiotic nitrogen fixation in the laboratories of Drs. Carroll Vance and Deborah Samac. After defending his Ph.D. thesis in 1997, Mark joined the lab of Dr. Joe Chappell at the University of Kentucky where he is currently working as a research associate. Initially charged with identifying specific terpene synthase genes activated in plant-pathogen interactions, his interests have broadened to include the chemical ecology of plant defense and the conservation of defensive mechanisms among the Solanales.

#### Student Award Winners at the 2000 PSNA Annual Meeting

**Best Oral Presentation** 

Allan Phipps Florida International University,

Japanese Use of Beni-tengu-dake (Amanita muscaria) and the Efficacy of Traditional Detoxification Methods **Best Poster Presentation** 

Alexander Walz Technical University of Dresden,

Molecular Cloning and Characterization of the IAA-Protein Gene from *Phaseolus vulgaris*.

#### **Travel Awards**

Leon Adler, UMBC, An *Arabidopsis* Mutant with Sucrose Dependent Meristem Growth

Monira Ahsan (with S.K.N. Islam), University of Strathclyde, Novel Diterpenes from *Scoparia Dulcis* and Their Cytotoxic Activities on Human Cancer Cell Lines

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Jacqueline Bede, University of Toronto, Biosynthetic Pathway of Insect Juvenile Hormone III in the Sedge, *Cyperus iria* L.

Jamie Bretz, University of Maryland, Identification of *Pseudomonas syringae* pv. Tomato DC3000 Host Range Determinants

Maryam Farzad, Georgetown University, Floral Color Change in *Viola cornuta*: a Model System to Study Regulation of Anthocyanin Production

Mi Kwon, Washington State University, In situ Hybridization and Immunolocalization of Dirigent Protein and Lignan Reductases in *Forsythia intermedia* and *Pinus taeda* 

Kothandaraan Narasimhan, National University of Singapore, An LC/MS-Based Metabolic Profiling of the Flavonoid Pathway Intermediates in the Tissues and Root Exudates of *Arabidopsis* Mutants and Transgenics





Daniel Owens, East Tennessee State University, Development of a Sensitive Quantitative Assay for Flavanone-3-Hydroxylase using Capillary Electrophoresis

Isagani Padolina, Universty of Texas at Austin, Deoxyflavonoids in Elicited Liquid Cell Cultures of *Cephalocerus* senilis (Old Man Cactus): Metabolic Profiles and Molecular Mechanisms

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Jennifer Pelt, East Tennessee State University, A Comparison of Flavanone-3-O-Hydroxylase (F3H) mRNA Levels in Petunia Hybrida and *Citrus Paradisi* 

Allan Phipps, Florida International University, Japanese Use of Benitengu-dake (*Amanita muscaria*) and the Efficacy of Traditional Detoxification Methods



Jennifer Smith, East Tennessee State University, Bryophyte Phylogeny: Evidence Based on the Chloroplast *psbA* Gene Sequence

Reynold Tan, University of Maryland, Stimulation of Spore Germination in a Plant-Pathogenic Fungus in Response to Host Flavonoids

Daniel Tanner, East Tennessee State University, Structural Elucidation of Flavanone-7-*O*-Glucosyltransferase from *Citrus paradisi* 

Alexander Walz, Technical University of Dresden, Molecular Cloning and Characterization of the IAA-Protein Gene from *Phaseolus vulgaris* 

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## Faces at the 2000 PSNA Annual Meeting



Ed Braun discusses the lighter side of science.





Mark Schoenbeck. Lukas Mueller considers a question.





Daniel Tanner at his poster.

Mi Kwon.



Eric Conn and friends hope to see you again in Oklahoma!



Jim Saunders thanks the Organizing Committee for their help in putting on a very successful meeting.



Hector Flores, Kelsey Downum, and John Romeo enjoy the banquet.



Kelsey Downum, Allan Phipps, John Romeo, and Connie Nozzolillo.

# MESSAGE FROM THE EDITOR-IN-CHIEF The New World of Publishing

Symposium volumes are not as lucrative for publishers as they once were. Due to increasing costs, sales are off because many libraries have ceased buying them. Individuals who in the past have routinely purchased proceedings now reconsider whether or not they are affordable. Publishing houses have instituted a number of cost-cutting changes to protect their interests. In general, most of these are unfavorable to groups such as the PSNA. Our previous contract with KluwerAcademic/Plenum Press expired in 1999. Their conditions for renewal for another five year period were so unfavorable that, after much deliberation, the PSNA Executive Committee voted to sign a 5-year contract with Elsevier for Volumes 34 to 39. Their terms were far better for us.

The changes can be summarized briefly. In-house formatting has reverted to 'camera-ready' copy (more work for authors and the editor(s) and more preparation expenses); royalties and page budgets have been maintained at current levels (we receive no royalties on discounted copies, but they have been increased slightly on others and we have retained page preparation charges); the Society purchases 75 books as a bulk order prior to publication - PSNA members can purchase from this stock from the Treasurer at a 50% discount prior to publication, or at a 40% discount after publication.

We hope, among PSNA members and other meeting attendees, that selling this number of books will not be difficult. Early returns indicate this may not be the case. The Executive Committee is concerned. Why are meeting attendees and other members not buying our books? Why are we publishing these volumes? Are the topics inappropriate for our members? (Certainly molecular biology has invaded the traditional phytochemical terrain!) Does the Society need to more aggressively market the books? Do we need to make the cost of the annual volume a mandatory part of the registration fee at our meetings? (We made this an option at the Beltsville meeting). Do we wish to abandon entirely the practice of publishing annual volumes? Both the membership and the Executive Committee need to wrestle with these questions. The future of the series may be in jeopardy.

We are making a special appeal to members to: 1) Purchase symposium volumes. Volume 34 can be obtained from the Treasurer at the 40% discounted price. Please enquire with Cecilia McIntosh for the current price; 2) Make a pre-publication commitment to purchase Volume 35 at the 50% discounted price. This too can be ordered from the Treasurer; 3) Encourage your institutional library to purchase our volumes and commit to the series by notifying Elsevier. They need to be on an automatic renewal list so that volumes are sent each year. These are the volumes on which the Society receives royalties; 4) give the Executive Committee your ideas for dealing with this issue. There certainly are no magic fixes, but there may be things we have not considered.

The rich history of Recent Advances in Phytochemistry provides a perspective obtainable nowhere else in the phytochemical literature. Several volumes have played pivotal roles in shaping the direction of future research. Others have solidified ideas and summarized topics in extremely useful ways. The sheer variety of topics addressed and the depth of their treatment is unsurpassed in the literature. The respect for and prestige of this series is internationally recog-

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nized. It would be a tragedy should the series not be able to continue.

John T. Romeo University of South Florida Tampa, Florida romeo@chuma.cas.usf.edu

#### FROM THE EDITOR

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from Canada, the United States, and Mexico (please see the excellent article by Felipe Vàzquez-Flota, our Mexican correspondent, in this issue of the Newsletter) who will write, or solicite from a colleague, an article relevant to a topic that is important in each country. Others might want to summarize a few recently published papers for each newsletter, or contribute an article that would be of special interest to certain groups. For example, is there anyone out there who would be willing to bring a chemical ecology or pharmacognosy perspective to the newsletter? Please do not hesitate to contact me if you would like to contribute, or if you have any ideas or comments. I especially encourage the participation of students and postdoctoral fellows. The PSNA and its Newsletter will only be as good as what we make it. Please think about getting involved.

> Peter Facchini University of Calgary Calgary, Canada pfacchin@ucalgary.ca

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# PSNA EXECUTIVE COMMITTEE Minutes of the Last Meeting

Present: Susan McCormick, Celia McIntosh, Rick Dixon, John Romeo Absent: Vince De Luca, Dennis Clark

The following is a summary of the issues discussed at the PSNA Executive Committee meeting held on Sunday, August 6 at the 2000 PSNA Annual Meeting in Beltsville, MD. **Newsletter and Website** - There was a general discussion on the state of the Newsletter and website, and the need to improve both immediately. It was discussed to eventually have the Newsletter online as an option to reduce annual membership fees.

Future Meetings - Rick Dixon originally offered to host 2001 meeting in Santa Fe, NM, although this was dependent on the availability of Pedro Mendez as a coorganizer in New Mexico. However, Dr. Mendez has moved to Virginia Tech, so the meeting will probably be in Oklahoma City, or in Dallas, at a hotel. The general theme of Analytical Phytochemistry and Genomics was suggested. It was mentioned that funding in support of the meeting should be available from the Samuel Roberts Noble Foundation. It was suggested that the Young Investigators' Minisymposium topic be different from those selected previously. There is an offer from Rachel Mata and colleagues to host the meeting in Mexico in 2002. Hector Flores has also offered to help with contacting potential organizers for this meeting. Rick Dixon offered to contact Charlie Arntzen if the topic were medicinal in orientation. Cecelia McIntosh talked about possible meeting facilities at East Tennessee State University.

**Dues** - Discussed raising dues to defer costs of an improved Newsletter and website. the Executive Committee decided to raise dues to \$40 for full members, and \$20 for student members. Society bylaws do not require a vote of the membership to change dues. **Student Awards** - The amount of money available for student travel awards was decided. Jim Saunders was asked to arrange for judges for the student paper competition. The Executive Committee authorized judges to select up to 2 posters and 2 oral presentations for awards

Editor-in-Chief's Report - A new publisher and contract for the conference proceedings book are now in place. Important features of the new deal include the requirement for camera ready copy and better marketing of the books. The book from last year's meeting is not available yet, although a nice brochure was produced and will be available at the annual meeting. Our contract requires sales through the PSNA of 75 books per volume. The book editor will push book sales in the newsletter and at the annual meetings. Book orders can be placed in conjuction with dues notices and meeting registration. The treasurer will receive what remains unsold from the bulk order of books.

**Treasurer's Report** - The student travel award and membership lists will be updated, and members in arrears will be dropped from the list. It was reported that the membership directory costs about \$1000 to produce each year. Unfortunately, the Arthur Neish fund is still not fully endowed. Interest on this account is supposed to support the Young Investigators' Minisymposium.

Jim Saunders came to the meeting to report on the 2000 meeting with regard to arrangements, registration, and other matters. It was estimated that there would be about 150 registrants. He reported that he had about \$10,000 in outside funding between the USDA and MAPBS, the group that is co-hosting the meeting. He also suggested that attendance from MAPBS membership may be low because of the registration fees.

Susan McCormick

# FUTURE MEETINGS OF INTEREST

#### Third IUPAC International Conference on Biodiversity (ICOB-3)

Antalya, Turkey

November 3 - 8, 2001

Contact Prof. B. Sener blgsener@tr-net.net.tr

#### 42nd Annual Meeting of the American Society of Pharmacognosy

Oaxaca, Mexico

July 14 - 18, 2001.

Contact Dr. Luis Manuel Rodrìguez Imanuel@cicy.mx http://www.phcog.org

4th International Congress on Chemistry, and 13th Caribbean Conference on Chemistry and Chemical Engineering

Havana, Cuba

April 16 - 20, 2001.

Contact Alicia García aliciag@palco.get.cma.net

## PERSPECTIVE

#### History and Future of the Young Investigators' Minisymposium: A Good Idea Whose Time Has Come

The PSNA Young Investigators' Minisymposium was born in early 1994 when Kelsey Downum, who was the PSNA President in 1993, explored this idea with me during a telephone conversation. I was very much interested in organizing such a minisymposium, as it could greatly facilitate the rejuvination and modernization of the PSNA. The concept that evolved during these discussions was that senior graduate students and/or postdoctoral fellows whose research had created an impact in a particular area of phytochemistry would be invited to participate in a yearly minisymposium. The scientific theme would vary each year in order to address each of the major phytochemical areas (biochemistry, chemistry, molecular biology and ecology). In this way young scientists from each discipline could discover, and participate in the revival of, the PSNA. The very first Young Investigators' Minisymposium took place at the PSNA Annual Meeting held in 1995 in Sault Sainte-Marie, Canada. The theme of this meeting was "Phytochemical Redundancy in Ecological Interactions". We decided that the young investigators' minisymposium would deal with "The biosynthesis and Molecular Biology of Secondary Metabolites". Invitations were extended to Nicholas Bate, Clint Chapple, Peter Constabel, Peter Facchini, and Nancy Paiva who all eagerly accepted the opportunity to participate, although several required financial support for travel to the meeting. Although the PSNA was unable to fund the minisymposium, we succeeded in raising \$3,000 (from Agriculture Canada, the National Research Council of Canada, Pfizer and

Novartis) to help finance the participation of speakers. The young investigator presentations were very well received and complemented the original theme of the meeting on phytochemical redundancy.

The excellent reception of this new minisymposium convinced several senior members of the PSNA that it should become a regular feature of the annual meetings. However, this would not be possible without a permanent source of financial support as highlighted by the fact that there was no minisymposium at the annual meetings between 1996 and 1998. Therefore, we decided to initiate a campaign in 1998 to raise a \$50,000 Young Investigators' Minisymposium fund, which could be used to support this event at our annual meetings. The PSNA initiated the permanent fund with a \$10,000 contribution and we succeeded in raising \$23,000 from donations by the National Research Council of Canada, Novartis Agribusiness Biotechnology Inc., the Samual Roberts Noble Foundation, Pfizer Canada, as well as donations from PSNA members and a special contribution from Mrs. Dorothy Neish. It was unanimously agreed to name this fund the "Arthur Neish Young Investigators' Fund", in honor of this well known phytochemist.

The 1st Arthur Neish Young Investigators' Minisymposium was held in 1999 at the PSNA Annual Meeting in Montreal, Canada. The meeting theme was "The Evolution of Metabolic Pathways" and the minisymposium covered the "Biochemistry and Molecular Biology of Brassinosteroid Hormones". The young investigators, Jurgen Schmidt, Man-Ho Oh, Z-Y Wang, and Frederique Marsolais, were invited by Luc Varin to present a fascinating overview of their contributions to the chemistry, biochemistry and regulation of this rapidly evolving area of hormone biology. The minisymposium was held again at the 2000 PSNA Annual Meeting in Maryland when Mark Schoenbeck, Frédérique Hilliou, Edward Braun and Lukas Mueller were invited by Peter Facchini to speak about advances in the "Regulation of Terpenoid, Monoterpenoid Indole Alkaloid, Flavonoid, and Anthocyanin Pathways". respectively. In 2001 the PSNA will meet in Oklahoma City and the minisymposium should focus on phytochemical analytical methods.

It is clear that the financial support of this fund has been useful to encourage the annual holding of this minisymposium and I believe we are well on our way toward the rejuvination of our society. In order to maintain this momentum I am soliciting a tax deductible donation from our membership in order to bring the Arthur Neish fund up to the level of \$50,000. In addition, we need volunteers from the membership who are willing to organize the minisymposium or who can suggest an area they wish to have highlighted. The diversity of phytochemistry should ensure many different and interesting subject areas that can be explored. However, it is essential that more of us get excited and become involved in these activities. The future of the PSNA rests on the membership.

Vincenzo De Luca Novartis, Inc. Research Triangle Park, NC vince.deluca@nabri.novartis.com

#### PHYTOCHEMICAL SOCIETY OF NORTH AMERICA

#### FINANCIAL REPORT (01 January 1999 - 31 December 1999)

| RECEIPTS  |             |
|---|-------------|
| Membership dues                                     | \$5100.00   |
| Plenum Publishing : royalties & page charges on RAP | 6991.91     |
| Interest on Fortis Money Market                     | 3.49        |
| Interest on TN FAIR account                         | 806.74      |
| Dividends Fortis Advantage Account                  | 133.59      |
| 1998 meeting refund                                 | 4220.21     |
| Art Neish Young Investigator Symposium Fund         | 6623.11*    |
| Symposium Fund Interest                             | 965.50      |
| Mailing list rental                                 | 200.00      |
| 1999 meeting refund                                 | 5421.00     |
| TOTAL RECEIPTS                                      | \$30,465.55 |

\*From generous contributions by NSRC, Pfizer, Mrs. Art Neish, Ann Oaks, and Ragai Ibrahim

#### **EXPENDITURES**

| Executive Committee expenses                     |             |
|--|-------------|
| Treasurer (dues notices, supplies for directory) | \$331.92    |
| Editor, RAP                                      | 1500.00     |
| Secretary  | 2000.00     |
| Travel   | 380.97      |
| Montreal meeting advance                         | 5000.00     |
| 1999 Paper/Poster Awards                         | 500.00      |
| 2000 meeting advance                             | 2000.00     |
| Phytochem. Soc. Eur. (1st year share royalties)  | 1369.92     |
| TOTAL EXPENDITURES                               | \$13,082.81 |
| ASSETS   |             |
| Checking account                                 | \$2,703.73  |
| FAIR account (investment reserve)                | 32,375.93   |
| Young Investigator Symposium Account             | 24,960.33   |
| Fortis Money Market (opened 2/99 \$100)          | 103.49      |
| Fortis Advantage Account** (opened 2/99 \$9900)  | 11,796.32   |
| TOTAL ASSETS                                     | \$71,939.80 |
| **stock/bond investment account                  |             |

C. McIntosh

# **PSNA 2001**

# Phytochemistry in the Genomics and Post-Genomics Eras

August 5 - 9, 2001

Oklahoma City Oklahoma, USA

The 2001 Annual Meeting of the Phytochemical Society of North America will be held August 5 - 9 in Oklahoma City. The theme of the meeting will be Phytochemistry in the Genomics and Post-Genomics Eras. The aim is to provide something for everyone as the Symposium outlines the state-of-the-art in phytochemical analysis and molecular approaches aimed at phytochemical pathway discovery.

Topics to be covered will include:

- ✓ LC/MS for metabolic profiling in Medicago truncatula.
- ✔ Fourier Transform Ion Cyclotron MS for metabolic profiling
- ✓ Functional genomics by metabolic profiling using transient gene expression
- ✓ Profiling isoprenoid pathway metabolites
- ✓ Profiling alkaloids
- ✓ Isotopic analysis and modeling of pathway flux.
- ✓ Linking metabolic pathway and gene expression databases.
- ✓ A genomics approach to one carbon metabolism.
- ✓ A mutational approach to dissection of flavonoid biosynthesis in Arabidopsis.
- ✓ Biopanning of natural product pathways using T-DNA activation tagging.
- ✓ Functional genomics of plant cytochrome P450s.
- ✓ Heterologous expression systems for plant natural product biosynthetic enzymes.
- ✔ Chemical synthesis of flavonoids, isoflavonoids and condensed tannins.
- ✓ Combinatorial chemistry approaches
- ✓ Structural biology of the plant polyketide synthase superfamily.
- ✔ Genetic approaches to brassinosteroid biosynthesis and function.
- ✓ Genetic approaches to triterpene saponin biosynthesis
- ✓ Metabolic engineering of lignin.
- ✓ Metabolic engineering of lignans.
- ✓ Vitamins and nutritional genomics.
- ✓ Metabolic engineering of cyanogenic glycosides and glucosinolates.

Full details of the program and how to register will appear in the next edition of the Newsletter, and on the PSNA website (www.psna-online.org).

# Elsevier Publishes Volume 34 of Recent Advances in Phytochemistry

Volume 34 of Recent Advances in Phytochemistry - *Evolution of Metabolic Pathways*, the symposium volume resulting from the excellent 1999 PSNA Annual Meeting in Montréal, Canada organized by Vincenzo De Luca, Luc Varin, and Ragai Ibrahim, is the first to be published by our new publisher Elsevier. It contains 14 chapters, has 467 pages, and is available for purchase by PSNA members at a significant discount.

The past decade has seen major advances in the cloning of genes encoding enzymes of plant secondary metabolism. This has been further enhanced by the recent project on the sequencing of the Arabidopis genome. These developments provide the molecular genetic basis to address the question of the Evolution of Metabolic Pathways. This volume provides indepth reviews of our current knowledge on the evolutionary origin of plant secondary metabolites and the enzymes involved in their biosynthesis.



Contributors to this volume explore a wide range of topics that include:

- If the role of secondary metabolites in evolution
- $\ensuremath{\mathscr{D}}$  the evolutionary origin of polyketides and terpenes
- ${\mathscr O}$  oxidative reactions and evolution of secondary metabolites
- $\ensuremath{\mathscr{C}}$  the evolutionary origin of substitution reactions
- $\mathscr{A}$  structure-function relationships in the evolution of steroid sulfonation

The PSNA, under terms of our new contract, can sell you this volume at the **40% discounted price** of \$112 US. Send your check or a credit card number to the Treasurer, Cecilia McIntosh.

# **Recent Advances in Phytochemistry Series** PSNA members receive a 40% discount on the following titles

| Volume 35 (2000)** | Regulation of Phytochemicals by Molecular Techniques                |
|--------------------|---|
|                    | (List \$210.00, PSNA \$112.00)                                      |
| Volume 34 (1999)** | Evolution of Metabolic Pathways                                     |
|                    | (List \$222.50, PSNA \$133.50)                                      |
| Volume 33 (1998)   | Phytochemicals in Human Health Protection, Nutrition, Plant Defense |
|                    | (List \$165.00, PSNA \$99.00)                                       |
| Volume 32 (1997)   | Phytochemical Signals and Plant-Microbe Interactions                |
|                    | (List \$95.00, PSNA \$57.00)  |
| Volume 31 (1996)   | Functionality of Food Phytochemicals                                |
|                    | (List \$114.00, PSNA \$68.40)                                       |
| Volume 30 (1995)   | Phytochemical Diversity and Redundancy in Ecological Interactions   |
|                    | (List \$89.50, PSNA \$53.70)  |
| Volume 29 (1994)   | Phytochemistry of Medicinal Plants                                  |
|                    | (List \$71.00, PSNA \$42.60)  |
| Volume 28 (1993)   | Genetic Engineering of Plant Secondary Metabolism                   |
|                    | (List \$89.50, PSNA \$53.70)  |
| Volume 27 (1992)   | Phytochemical Potential of Tropical Plants                          |
|                    | (List \$79.50, PSNA \$47.70)  |
| Volume 26 (1991)   | Phenolic Metabolism in Plants                                       |
|                    | (List \$89.50, PSNA \$53.70)  |
| Volume 25 (1990)   | Modern Phytochemical Methods  |
|                    | (List \$85.00, PSNA \$51.00)  |
| Volume 24 (1989)   | Biochemistry of the Mevalonic Acid Pathway to Terpenoids            |
|                    | (List \$85.00, PSNA \$51.00)  |
| Volume 23 (1988)   | Plant Nitrogen Metabolism   |
|                    | (List \$89.50, PSNA \$53.70)  |
| Volume 22 (1987)   | Opportunities for Phytochemistry in Plant Biotechnology             |
|                    | (List \$59.50, PSNA \$35.70)  |
| Volume 21 (1986)   | Phytochemical Effects of Environmental Compounds                    |
| . ,                | (List \$75.00, PSNA \$45.00)  |
| Volume 20 (1985)   | The Shikimic Acid Pathway   |
|                    | (List \$55.00, PSNA \$33.00)  |

\*\*Volumes 34 and 35 are available from our new publisher, Elsevier. Please contact the PSNA treasurer, Cecilia McIntosh, for information on ordering these volumes.

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## **Phytochemical Society of North America**

#### **Membership Application**

| Please fill in the following application and return to the Treasurer with your dues payment. We are also in the process of updating the PSNA website, so please respond to the question regarding posting information on the website and give information on your personal web page if you wish it to be included. Once your application has been processed, you will receive newsletters and special mailings. You are also eligible for PSNA member discounts on the Recent Advances in Phytochemistry series. |   |  |  |
|--|---|--|--|
| Please make check or money order payable to the <b>Phytochemical Society of North America.</b> Payment must be made in U.S. dollars, drawn on a U.S. bank. Traveler's Checks or Canadian Postal Money Orders, payable in U.S. dollars, are also acceptable. We are unable to accept payment via credit card.   |   |  |  |
| Dues schedule:   | Life (or emeritus) member - no charge<br>Regular member - \$40.00 per year<br>Student member - \$20.00 per year |  |  |

Return this statement along with your payment to:

#### Dr. Cecilia A. McIntosh, PSNA Treasurer Department of Biology, Box 70703 East Tennessee State University Johnson City, TN 37614-0703 USA

| Please take a mome                      | nt to provide/up                    | odate the following inform                          | ation:        |                              |
|---|-------------------------------------|---|---------------|------------------------------|
| Name (Dr., Mr., Mrs                     | ., Ms.):                            |   |               |                              |
| Mailing Address:                        |                                     |   |               |                              |
| City:                                   | Sta                                 | ate/Province:                                       |               | Zip/Postal Code:             |
| Phone:                                  | Fa:                                 | x:  | E-I           | Mail:                        |
| Homepage URL:                           |                                     |   | _             |                              |
| The PSNA homepag<br>May we include/link | e is now availa<br>/our directory/h | ble at www.psna-online.o<br>nomepage information on | rg<br>the PSI | NA website? Yes/No           |
| Research Interests (circle              | up to 4 items):                     |   |               |                              |
| A. Acetylenes                           | S.                                  | Terpenoids  |               | stress                       |
| B. Alkaloids                            | Т.                                  | Vitamins  | nn.           | Industrial applications      |
| C. Amino acids/proteins                 | aa.                                 | Biochemistry/physiology of                          | 00.           | Structure identification     |
| D. Coumarins                            |                                     | herbicides  | pp.           | Marine natural compounds     |
| E. Cyanogenics                          | bb.                                 | Enzymology  | qq.           | Medicinal chemistry          |
| F. Flavonoids                           | CC.                                 | Cell wall chemistry                                 | rr.           | Membrane structure/function  |
| G. Glucosinolates                       | dd.                                 | Chemotaxonomy                                       | SS.           | Molecular/immunological      |
| H. Lignans                              | ee.                                 | Biotechnology                                       |               | techniques                   |
| I. Lipids                               | ff.                                 | Plant-insect interactions                           | tt.           | Nitrogen fixation/metabolism |
| J Nitrogen compounds                    | gg.                                 | Plant-microbe interactions                          | uu.           | Pharmacology/pharmacognosy   |
| K. Nucleic acids                        | hh.                                 | Plant-plant interactions                            | VV.           | Plant pathology              |
| L. Organic acids                        | И.                                  | Chemical reactions/organic                          | WW.           | Plant genetics               |
| M. Phenolics                            |                                     | synthesis   | XX.           | Recognition-cell surface     |
|   | JJ.                                 | Biochemistry of secondary                           |               |                              |
| O. Quinones                             | L.I.                                | metabolism  | уу.           | I ISSUE/CEII CUITURE         |
| P. Stildenes                            | KK.                                 | Fungal metabolism                                   | ZZ.           | TOXICOLOGY OF NATURAL        |
| Q. Sugars/polysaccharl                  | ues II.                             | Growin regulators                                   |               |                              |
| R. Sullui compounds                     | THTT                                | . Diochemistry/physiology of                        | 011           | TER                          |

R. Sulfur compounds

# Phytochemical Society of North America Sociedad Fitoquímica de América del Norte Société Phytochimique de L'Amerique du Nord

Volume 41, Number 2

March 2001

# PRESIDENT'S LETTER PSNA 2001: A Preview of the Annual Meeting

Phytochemists have a distinct view of the world, in which plant chemistry is central. Nevertheless, this centrality is fed on one side by the processes of evolution, genetics, and gene expression that lead to the diverse array of natural products found in nature, and, on the other side, itself feeds disciplines such as pharmacognosy, ethnobotany, and biotechnology that make use, in one way or another, of the biological activities of plant natural products. Plant genetics and biotechnology are research areas that other societies also claim as central to their members' interests. However, PSNA is unique in its strong chemical component. Although I want to see a broadening of the society's scope to reflect the relevance of phytochemistry to genomics, plant chemistry must nevertheless remain at its heart if the society is to retain its individual identity.

Because of the broadening scope of the discipline of phytochemistry, meetings have tended to become

"factionalized" in recent years, and have been seen as catering to either the "chemical" or "molecular" groups within the society. I think this is unfortunate, as many members may have missed important opportunities to share ideas with colleagues in different areas of the discipline to the benefit of their programs. I would therefore like to draw the attention of the membership to the August 2001 Annual Meeting of the PSNA to be held in Oklahoma City which I have organized. The topic of this meeting is "Phytochemistry in the Genomics and Post-Genomics Eras", and the meeting has received significant funding from the Samuel Roberts Noble Foundation. It is our intention to make this meeting a showcase of modern phytochemistry, linking phytochemical analysis with the latest developments in understanding gene function. As a significant number of plant genes encode enzymes involved in the biosynthesis or catabolism of natural products, functional genomics

is the science that will ultimately tell us how all the molecules we are so interested in are made in the plant. Conversely, doing this is impossible without strong expertise in phytochemical analysis.

The program for the meeting is included in this edition of the Newsletter. It features internationally recognized scientists working at the interface of phytochemistry and genomics. The two keynote speakers will address the importance of phytochemical bioprospecting for medicine and the power of Arabidopsis genetics for revealing the secrets of complex biosynthetic pathways. The Arthur Neish Young Investigator Symposium has been an attractive feature of recent meetings, giving outstanding young phytochemists an opportunity to present their work in the setting of an international conference. The topic of this year's symposium is phytochemical synthesis and analysis. I hope you will agree that we have some extremely high profile speakers,

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- ✔ Chemical Ecology: Spotlight on St. John's Wort
- ✓ Northern Exposure: A Case of Mistaken Identity?
- ✓ Perspective: A Phytochemical Renaissance?
- ✓ PSNA 2001: Registration Forms and Program



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2001 PSNA Annual Meeting

Phytochemistry in the Genomics and Post-Genomics Eras

August 4 - 8

Oklahoma City, Oklahoma

The Phytochemical Society of North America (PSNA) is a nonprofit scientific organization whose membership is open to anyone with an interest in phytochemistry and the role of plant substances in related fields. Annual membership dues are U.S. \$40 for regular members and \$20 for student members. Annual meetings featuring symposium topics of current interest and contributed papers by conference participants are held throughout the United States, Canada, and Mexico. PSNA meetings provide participants with exposure to the cutting-edge research of prominent international scientists, but are still small enough to offer informality and intimacy that are conducive to the exchange of ideas. This newsletter is circulated to members to keep them informed of upcoming meetings and developments within the society, and to provide a forum for the exchange of information and ideas. If you would like additional information about the PSNA, or if you have material that you would like included in the newsletter, please contact the PSNA Secretary and Newsletter Editor. Annual dues and changes of address should be sent to the PSNA Treasurer. Also check the PSNA website at www.psnaonline.org for regular updates.

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#### **PSNA ADVISORS**

Kelsey Downum (2000) John T. Arneson (2001) C. Peter Constabel (2003) Nikolaus H. Fischer (2002) Jonathon Poulton (2003)

#### continued from page 1

and that there is something for everyone at this meeting.

I hope to see you in Oklahoma City, where we can re-affirm the central role of phytochemistry in modern plant biology.

Richard A. Dixon Samuel Roberts Noble Foundation Ardmore, Oklahoma radixon@noble.org



Oklahoma City skyline

Brick Town Canal

# PHYTOCHEMICAL PIONEERS

#### **Helen Stafford**

My life and scientific career are described in a recent ASPP web page (www.aspp.org) article concerning "pioneer" women in Plant Physiology by the "Committee on the Status of Women in Plant Physiology". I published my first paper in 1948 (American Journal of Botany 35: 706) following my MA degree with Richard Goodwin at Connecticut College for Women. I followed this with excerpts from my Ph.D. research with David Goddard on cytochrome oxidase in peas, and a series of papers with Birgit Vennesland, such as DPN-dependent D-glyceric dehydrogenase, during my post-doctoral career. Publications concerning laboratory research with plant phenolics continued through 1985 at Reed College, followed by reviews in the area of phenolics, for a total of 70 publications through 2000. My book on flavonoid metabolism was published in 1990, and I edited volumes 24 to 28 of Recent Advances in Phytochemistry from 1990 to 1994.

Beginning in the 1940s and 1950's, paper chromatography of crude methanol- $H_2O$  extracts was used. Visual and UV derived visible

colors were noted, before and after  $NH_3$  fuming and with various sprays. TLC was later substituted for paper chromatography and finally HPLC analyses. Early Beckman spectrophotometers were used to determine UV and visible spectra. I studied the enzymology of various enzymes in phenolic metabolism and their products, resulting in original publications through 1989. Clearly, techniques for studying plant enzymology are much more complex today.

I attempted for years without success to isolate 'synthases' producing lignin and two flavonoids, proanthocyanidins and anthocyanidins. The enzymes were too unstable for isolation in purified form. Future research in this area required knowledge of the genes involved. In order to explore this area, I would have had to work with a molecular biology laboratory for future progress. After 'retiring' from laboratory research in the late 1980's at age 65, I continued to write reviews and review papers for various journals. I am now 78. A 'model' of the unstable anthocyanidin synthase protein was finally produced via modern cDNA tech-

3

niques in 1999 (Plant Journal 172: 181), and hopefully similar techniques can be used for the final steps in lignin and proanthocyanidin pathways in the near future to remove my frustrations with my own research in this area.

> Helen Stafford Reed College Portland, Oregon helen.stafford@reed.edu

The PSNA has a rich and fascinating history created by members who have made many important contributions to the field of phytochemistry. Beginning with this issue of PSNA News, our pioneering members will be graciously requested to provide us with an autobiographical perspective of their research careers. We've all heard that "to know where we're going, we must know where we've been". This is especially true for younger members of our society. If anyone would like to see a pioneering PSNA member recognized in this feature, please contact the editor.

#### PSNA NEWS

March 2001

# SOUTH OF THE BORDER

#### Mexican National Council for Science and Technology Appoints New Director

On January 16, forty-six days after taking Office, Mexican president Vicente Fox appointed Jaime Parada-Avila as the new Director of CONACyT (the Mexican National Council for Science and Technology). In some respects, Parada's appointment was a surprise despite his entrepreneurial profile which fits in well with the new government. His name was never openly mentioned among the top contenders for this position. The new head of CONACyT is a 52 year-old mechanical engineer who claims to have experience converting technology into viable business ventures. A former university professor since the early 1980's, Parada-Avila has been involved in the technological development of some of the most important private industries of Mexico, including Sydermex (the iron industry), and steel the electrodomestic division of Vitro Group and Cydsa (a powerful fiber plastic manufacturing and consortium). With Parada-Avila as its new Director, CONACyT now appears closer to achieving the longsought-after link with the private sector, 30 years after its inception. CONACyT's previously meager participation in the funding of science and technology has been frequently

pointed out by international organizations. The new Director is now attempting to persuade the Congress to approve tax exemptions for those industries willing to invest in science and technology in Mexico. This strategy could play a major role in his plan to increase the level of resources dedicated to science and technology from 0.4 to 1.0% of the Gross Internal Product by the end of the current administration's term in 2006. Even though this figure is still short from the 1.5% recommended by the UN, it certainly may represent a major breakthrough. However, the Mexican scientific community has expressed its concern about the rules for the distribution of those resources. Recently, Parada-Avila has declared that CONACyT will identify priority areas for support and, due to his industrial connections, some researchers fear that projects with extensive social relevance, but lacking in immediate economic potential, might be overlooked. Furthermore, many scientists dedicated to basic research have expressed concern that they will be at a disadvantage in terms of the allocation of research funds. compared to their colleagues working in fields of applied research and technological development. As a

result of this debate. Parada-Avila has ratified the commitment of CONACyT to support all areas of scientific research. However, funding will be conditional on the ability of scientists to produce original research papers and train highly qualified Ultimately, personnel. the appointment of Parada-Avila as the new Director of CONACyT represents an important change in the science and technology policies in Mexico. Despite some reservations, his appointment has been well received. It is now crucial for Mexican scientists to attract the attention of both the government and the private sector to find real solutions to critical problems in Mexico. The scientific community must clearly define these goals. The delicate balance necessary to satisfy the demands of the diverse groups with interests in the emerging science and technology sector in Mexico will be the major challenge of the new Director.

Felipe Vázquez-Flota Unidad de Biología Experimental Centro de Investigación Científica de Yucatán Merida, Mexico felipe@cicy.mx



# CHEMICAL ECOLOGY

#### Shedding Some Light on St. John's Wort

In the last PSNA newsletter, Rick Dixon emphasized the importance of the interdisciplinary nature of our society. I have focused the "spotlight" of my article on the chemical ecology of a popular plant to illustrate this point. In "light" of the recent popularity of St. John's wort as an antidepressant, I thought it would make an interesting topic.

Hypericum perforatum L. (family Guttiferae (Clusiaceae)) or St. John's wort is a small perennial plant between 1 to 3 feet tall. Its distinctive oblong leaves are scattered with transparent dots, known as schizogenous secretory cavities, making identification easy. Around the edges of the bright yellow flowers, one observes minute black dots, which are composed of cell clusters filled with the phototoxins hypericin and pseudohypericin. These glands are also found along the leaf margin and the stem. The use of St. John's wort in herbal medicine has been documented since the Middle ages (Wills et al. 2000; Mazza and Oomah, 2000; Griesen et al., 2001). The medicinal parts of the plant consist of the aerial tissues including the buds and flowers. Traditional uses of this herb include the treatment of anxiety and depression. Moreover, the oil of St. John's wort has been used externally for the treatment of minor burns, wounds and muscle aches.

Recently, St. John's wort has been the focus of considerable media attention due to its popular use as an alternative treatment for minor depression. Research suggests that the antidepressant properties of *H. perforatum* stem from the influence of catecholamine neurotransmission by compounds in the extract which were believed to be the naphthodianthrones, hypericin and pseudohypericin (Wills et al. 2000; Mazza and Oomah, 2000; Griesen et al., 2001). However, recent evidence points to hyperforin as the primary active constituent of St. John's wort extract since this compound has been shown to have serotonergic, noradrenergic, dopaminergic and cholinergic activity (Griesen et al., 2001). The biological compound or compounds responsible and the mechanism of action are still topics of much research. Adding an interesting twist to this debate is the fact that St. John's wort extracts are standard-



ized by their hypericin content which, in light of recent data, may not be an appropriate marker (Greeson et al., 2001). Further studies are being conducted to characterize the compounds and mechanisms responsible for the pharmacological activity of St. John's wort and to identify possible side effects or herb-drug interactions.

Hypericin and pseudohypericin, are eight-ringed structures comprised of two anthrone units joined through three bonds. These highly

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conjugated compounds are bright red and localized in specialized glands along the floral edges and leaf margins where they may account for 0.09 % (hypericin) and 0.23 % (pseudohypericin) of the plant's fresh weight (Kitanov, 2001). In general, the highest levels of these naphthodianthrones were detected in the floral tissues, followed by the leaves and stem (Sirvent et al., in press). However, in two H. perforatum populations, higher levels of these compounds were found in the leaf tissue. The authors speculate that this may be the result of the plant's defense response against insect herbivory. Hypericin and pseudohypericin are phototoxins. Upon activation by light, these quinones enter an excited state that interacts with molecular oxygen to produce highly reactive singlet oxygen or superoxide radicals (Arneson et al., 1983). This activity is believed to be, at least partially, responsible for the observed antiviral and antitumor properties of these compounds (Lavie et al., 1989). Hyperforin and adhyperforin are phloroglucinols and comprise up to 2.0-4.5 % and 0.2-1.8 % of the plant's fresh weight, respectively (Kitanov, 2001). Other plant constituents that might have pharmacological activity or could modify, either synergistically or antagonistically, the activity of these compounds include flavonoids, procyanidins, tannins and a variety of essential oils.

St. John's wort is indigenous to Europe, western Asia and northern Africa and was introduced in the early twentieth century into North America (California, Oregon, British Columbia) and Australia. By 1944, it is estimated that this invasive weed occupied over 1,000,000 ha in California (Driesche and Bellows, 1996). Not only was this weed taking over natural habitats, but livestock ingesting the

the plant suffered dermatitis and, in extreme cases, central nervous system toxicity, blindness and death (Bourke, 2000). In a well known and successful example of biological control, three insect species, Chrysolina hyperici, C. quadrigemina, C. varians, Zeuxidiplosis giard and Agrilus hyperici, were introduced into the United States and Canada. The defoliating beetles, C. hyperici and C. quadrigemina, were the most effective control agents and weed infestation levels were reduced by 99% (Driesche and Bellows, 1996). Larval and adult Chrysolina beetle feed on St. John's wort's leaves and reduce the plant's ability to overwinter. However, defoliation is minimal (25-50%) and plant mortality results from the fungal pathogen Colletotrichum gloeosporioides which uses these insects as a vector (McCoy and Gorsuch, 1999).

In the mid-1970s, reports of the spread of C. quadrigemina onto a related Hypericum species, H. concinnum (a native Californian species), appeared (Andres, 1985). This posed a potential concern because of the possible interference with native plant ecology by the introduced insect species. The extent of damage of H. concinnum by C. quadrigemina appears to be minimal and was attributed either to the ability of the plant to regenerate daughter plants through a trailing root system, or to the avoidance of direct sunlight by C. quadrigemina which probably relates to its ability to evade hypericin phototoxicity. This insect prefers to "hide" under the rosette foliage of H. perforatum rather than H. concinnum, which has restricted basal foliage.

Insects use a number of adaptive strategies to avoid the phototoxicity of hypericin in St. John's wort. A number of insects in the order Lepidoptera exhibit light avoidance behaviour while feeding, such as through leaf rolling or tying, boring into the plant or case-making (Sandberg and Berenbaum, 1989). Some generalist grasshoppers avoid the leaf edge that contains the hypericin-filled glands while feeding (Guillet et al., 2000). This behaviour is reminiscent of the early instars of the caterpillar, Anaitis plagiata, which also selectively feeds on leaf tissue (Fields et al., 1991). Later instars of this insect consume the entire leaf. It is thought that the cuticle of these older larvae is less permeable to the critical photosensitizing wavelengths. A similar story is observed in Chrysolina larvae where early instars feed at night and seek shelter from the light in the day by burying under the topsoil (Fields et al., 1991). Adults avoid phototoxicity with opaque cuticles that shield them from the light. Guillet et al. (2000) have also observed that generalist and specialist insect species possess different biochemical adaptations to St. John's wort. Specialist insect species showed lower constitutive activities of the antioxidant enzymes glutathione-Stransferase (GST) and glutathione reductase in their fat bodies and midgut. However, hypericin induced GST activity in specialist insect species.

I have tried to present a brief and, perhaps, Daliesque view of H. perforatum. One purpose was to represent one plant species from a botanical, plant physiological, pharmacological, biochemical and chemical ecological perspective. As Rick suggested in his previous article, we must all strive to find "a unique approach to (understand) the chemical processes that make plants such a rich source of molecular diversity".

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# NORTHERN EXPOSURE

#### Authentication of Medicinal Plant Identity

The rapid growth in the popularity of herbal medicines has given rise to a number of pressing issues and concerns, not only for the scientific community but also for consumers, regulators and healthcare professionals. In all of these arenas, authentication of the botanical identity of medicinal plants is the most critically important concern. Accurate plant identification is not only a safety and quality issue, it is a fundamental factor in evaluating the efficacy of botanical products and an obvious essential for phytochemical profiling. Failure to adequately confirm the identity of the material used renders any subsequent scientific work meaningless. The most pressing medical concerns center on botanical identity as the majority of adverse and toxic effects, as well as treatment failures, are due to incorrect identifications or failure to adequately confirm identity. Identification is the most critical factor for assurance of public safety, for effectively documenting both positive and adverse events associated with botanical products, for developing and maintaining quality assurance standards in manufacturing, and, ultimately, for meaningful efficacy testing of these products. In a larger sense, authentication of botanical identity has a gateway function as the most fundamental prerequisite for any type of scientific research on herbal medicines.

For the past four years, natural health (primarily botanical) products have consistently formed the fastest growing segment of the retail market. This steady growth in the popularity of herbal medicines provides a strong indication that the importance and impact of these products on society also continues to grow. There is increasing recognition in North America, that herbal medicines have the potential to substantially reduce

both social and economic burdens on health care systems. The most fundamental obstacle to the realization of this potential is the critical need for authentication of botanical identity. According to the World Health Organization (WHO), ensuring the correct identity of herbal medicines would provide the most significant decrease in the risks associated with these products, far greater than any other factor. The United Nation's "Quality Guidelines for Herbal Remedies" states that the greatest improvement in quality assurance of herbal remedies would be attained through the establishment of an international system for 'certificates of botanical identity'.

The popularity of herbal medicines has negative impact on other important public and scientific concerns, those of conservation and maintenance of biodiversity. Rapid increases in demand, in tandem with non-sustainable harvesting, have already driven some medicinal species to the brink of extinction and many more are threatened. The placement of endangered species such as goldenseal (*Hydrastis canadensis*) and pygeum



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(*Pygeum africanum*) on the CITES Appendix 2 list has introduced the need for an effective means of monitoring of international trade in these species. The growing importance of accurate identification of plant products to protect consumer health and to regulate the burgeoning international trade in wild and domesticated plant species at risk has placed burdens on regulatory agencies that currently cannot be met.

Public health and safety, as well as conservation issues, have created a demand for accurate and efficient plant identification methods that classical taxonomic approaches based on morphology, cytology, and chemistry can not necessarily fulfill. It is estimated that there are over 1200 species of medicinal plants commonly sold in North America for which identification methods are needed. In addition to this basic requirement to identify diverse plant species, appropriate identification tools for plant materials in various states of biological integrity, ranging from raw herbs, powders, to liquid and solid extracts, are needed.

Many processed formats are not amenable to any current classical morphological or chemotaxonomic methods and require taxonomic data that is not readily available. The limitations of classic morphological methods, primarily based on flower structure, in identifying herbal medicines composed of roots, stems or leaves, are obvious. Chemical analyses can provide highly sophisticated chemical fingerprinting or quantification of marker compound(s); however, method development is costly and there is no consensus in methodology. Phytochemical profiles can vary dramatically with genetic and environmental factors, developmental stage and tissue type, as well as post-har-

vest physiology and processing. Additionally, it is important to recognize that the most common adulterants/ contaminants are often related taxa, which can not always be reliably differentiated chemically in a timely and cost-effective manner. These factors must be considered in order to develop accurate and reliable chemical methods of authentication of identity. In some cases, even when sufficient resources in terms of both time and money are expended, chemical methods can not reliably discriminate between species due to infra- and interspecific variation. When the necessary morphological features are lacking and chemical methods are inadequate or as yet undeveloped, expensive and time-consuming research or extensive pharmacognosy expertise may be required. In many cases, equivocal identification is accepted. Consequently, unsubstantiated identifications are common in the natural health products industry and are the leading cause of misinformation regarding herbal medicines.

We suggest that another level of biological information, identification using genetic markers, may help to resolve many of these identification issues. It is widely accepted that DNA based approaches can be ideal for a range of identification applications. The use of DNA markers in plant breeding and in phylogenetic studies, as well as for forensic identification, is well established. In contrast to morphological or some chemical analyses, one of the strengths of DNA analysis is that small amounts of processed biological material, (e.g. dried, ground), are often still amenable to DNA marker identification and, depending on the strategies employed, can be semi-quantitative (e.g. QC-PCR). Compared to the range of tests and expertise required for current methods of plant identification, DNA based strategies present the best candidate for development of a single standardized approach that would be

applicable to widespread plant species in various states of biological integrity. A hierarchy of DNA markers could provide a comprehensive tool kit for identification at the appropriate taxonomic level. Additionally, genetic markers at the genus or family level could provide a much more efficient and accurate means of detecting toxic botanicals compared to the onerous task of individually checking for the presence of a broad spectrum of chemical markers. Research into DNA-based medicinal plant identification has been initiated at the National Center for Natural Products Research at the University of Mississippi, the Hong Kong University of Science and Technology, and Agriculture Canada, Ottawa.

At present, there are no internationally recognized, validated scientific methods for medicinal plant species identification. Several European pharmacopoeias describe traditional identification criteria for selected herbal medicines, although they do not all specify the same organoleptic, microscopic and/or chemical methods. The Kew Garden's program for Traditional Chinese Medicine collection is an important initiative since voucher specimens play an essential role in identity authentication: however, in isolation these are often not sufficient. In North America, there are no institutions or government bodies with adequate tools and expertise to conduct comprehensive plant identification, let alone develop industrial standards. The United States Pharmacopeia (USP) and the nonprofit American Herbal Pharmacopoeia (AHP) have started to publish herb monographs that outline authenticating methods for medicinal plant material; these valuable projects are based on traditional taxonomic criteria using morphology and chemistry. There is international industry participation and sponsorship of the Methods Validation Program of the American Institute of Nutraceutical Advan-

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cement (INA) for chemical analytical methods specifically for raw botanicals. The validated chemical methods made accessible by the INA are primarily for quality assessments using marker compounds and are not necessarily appropriate for routine authentication of identity in raw or finished products. Although these programs are important, the research investment to develop the expertise and techniques needed for accurate biological identification of the diverse array of medicinal plant products has yet to be made.

We submit to the membership of the Phytochemical Society of North America that the production of DNA markers for a range of medicinal plant species of commercial or conservation significance and for which an accessible, reliable means of efficient identification is not currently available, is an undertaking that could provide the strategic keystone to resolve some of these plant identification issues and facilitate phytochemical research of these species. The development of a medicinal plant pharmacopoeia, which we envision as an integration of molecular tools for identification with classical morphological and chemical tools for plant identification, may be the most significant scientific contribution that natural product researchers can provide for global public health and the medicinal plant industry worldwide.

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March 2001

## PERSPECTIVE

#### The Renaissance of Phytochemistry: A Reality or Just a Pipe Dream?

During the last century, scientists have been intensively exploiting the plant world in their search for secondary metabolites, which provide for a large range of human uses. Developments in the last 20 years have clearly demonstrated that secondary metabolites fulfill many key biological roles in plants (Plant Physiology 125: 58-60) and that they are not simply produced as products without function. For example, many interesting reports have been published which illustrate how molecules such as the ubiquitous flavonoids may play key roles in regulating legume-rhizobium symbiosis, plant-micchorriza associations, plant fertility and plant defense interactions. An important conclusion of these studies has been that the specificity of the interaction is guided by the particular substitution pattern of the flavonoid. In the human arena, epidemiological and in vitro studies have continued to suggest that flavonoids induce apoptosis in prostate cancer cell lines (Cancer Letters 160: 219-228 [2000]), exhibit anti-inflammatory activities by binding to chemokines (Immunopharmacology 49: 295-306 [2000]), promote antibacterial activity in methicillinresistant Staphylococcus aureus (Journal of Ethnopharmacology, 72: 483-488 [2000] & Journal of Pharmacy and Pharmacology 52: 361-366 [2000]), show anti-hepatitis B activity (Planta Medica 66: 694-698 [2000]) and inhibit HIV-1 infection at the level of viral entry (Biochemical and Biophysical Research Communications 276: 534-538 [2000]).

A sampling of the literature for other secondary metabolites shows that the structural diversity of terpenoids and alkaloids also provides access to remarkable variation leading to biological activity. Extensive studies on the biochemistry and mol-

ecular biology of a few key secondary metabolism pathways have revealed that the variation occurring in different species is derived from their ability to produce a restricted number of versatile chemical backbones and to evolve diverse substitution patterns. The emerging picture is that living organisms produce biochemistry through an unexpectedly small number of protein folds, which "can be combined, adapted and fine-tuned to achieve the diverse and quite specific functions mediated by the very large number of proteins that operate at the cellular level (Plant Physiology 125: 54-57 [2001])". This feature has produced a restricted set of large recognizable gene families (oxygenases, methyltransferases, glucosyltransferases etc.), that are responsible for the chemical diversity of plants, and from which further evolutionary diversification is possible (Trends in Plant Sciences 5: 439-445 [2000]).

The large efforts to sequence the genomes of several organisms, including those of Arabidopsis, rice, corn, soybean and tomato have resulted in the identification of many thousands of genes belonging to various gene families. The recognition that these genes catalyze a particular class of biochemical reaction has not been followed with extensive functional characterization of these genes. The industrial community has begun to develop proteomic and metabolic profiling technologies in the hope that genes required for desired biochemical reactions can be rapidly characterized and protected. However, this is an important new niche for the academic community to occupy, since industry will only exploit a fraction of the potential of these databases.

Functional genomic approaches, combined with computational and expression-based analyses are only beginning to be used to accelerate the comprehensive understanding of specialized cellular metabolism. For example, a recent study with a peppermint oil gland secretory cell cDNA library demonstrated that 25% of randomly selected cDNA clones were involved in essential oil synthesis (Proc Nat Acad Sci USA 97: 2934-2939 [2000]). This study confirms that cellular specialization results in greatly enhanced expression of genes involved in that specialization. Another interesting recent study helps to confirm the value of sequencing cell specific biochemical factories for isolating pathways responsible for the assembly of secondary metabolites (Plant Physiology 125: 539-555 [2001]). The glands of sweet basil accumulate phenyl-propene defense compounds and random sequencing of a cDNA library produced from the peltate glands of Basil confirmed that the phenylpropanoid pathway was expressed at high levels in these cells, since 13 % of the sequences obtained were for phenylpropanoid biosynthetic genes.

Although this has yet to be tried, similar random sequencing of cell-specific cDNA libraries from alkaloid producing plants could be used to isolate whole alkaloid pathways. For example, the data presented for the cell-specific compartmentation of alkaloid biosynthesis in *Catharanthus roseus* does suggest that epidermal, idioblast and laticifer cells are specialized sites of alkaloid biosynthesis and may be enriched for these pathways (Trends Plant Sci 5: 168-173 [2000] and Plant Cell 11: 887-900 [1999]).

A proteomic approach to analyze the proteins in opium poppy latex, which is thought to be the major site of morphine biosynthesis, has recently been published (Electrophoresis 21:3500-3516[2000]).The study focused on the analysis of latex-specific proteins by 2-dimensional SDS-PAGE after separating the latex into cytosolic serum and alkaloid containing vesicles. Internal peptide sequences were obtained for 75 protein spots and a putative function could be assigned to 69 of them. This type of analysis together with analysis of expressed sequence tags, promises to help define which genes are required for the creation of the specialized cell factories that are responsible for the biosynthesis of alkaloids and more generally all secondary metabolites.

These examples illustrate that there are many more secondary metabolism genes available than human resources to characterize them. Functional characterization of sequenced 'secondary metabolism' genes will require extensive collaboration between phytochemists and molecular biologists. It is clear that one block to progress in this area has been the loss of skilled phytochemists that has occurred over the past 20 years. Another problem relates to the possible apathy that may be present between phytochemists and molecular biologists. This is palpable in the lack of attendance by members of either discipline when PSNA meetings are dedicated to a phytochemical or a molecular topic. Can this situation be remedied and what can the PSNA do to bring about these essential collaborations? A major challenge for our society should be to bring phytochemists and molecular biologists together through generalized participation in the PSNA annual meeting,

through participation in the PSNA newsletter and by providing a forum for contact between the two groups. What should be done to promote these collaborations? Feedback providing possible answers to these questions would be much appreciated and may help to activate greater discovery efforts through the synergies created by collaborations between phytochemists and molecular biologists. Please think about this and respond!

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# MEETINGS OF INTEREST

#### IVth International Legume Conference

The Australian National University and the Royal Botanic Gardens, Melbourne are co-sponsoring the IVth International Legume Conference in Canberra, July 1 to 6, 2001. One of the sessions at the Conference is entitled "Chemosystematics and Natural Products Chemistry of the Leguminosae' co-organized by Ben-Erik van Wyk, Professor of Botany at Rand Afrikaans University in Johannesberg, and Dr. Barbara Meurer-Grimes, Leader, Natural Products Chemistry at Exgenix pharmaceuticals in Melbourne.

The session will include invited and contributed papers. The three invited speakers are: Prof. Peter Waterman, Lismore, Australia — "Phytochemical/ chemosystematic patterns in the Leguminosae" Dr. Geoff Kite, Jodrell Laboratory, Royal Botanic Gardens, Kew — 'New methodologies applied to chemosystematics" Prof. Michael Wink, University of Heidelberg — "The evolution of phytochemical traits in the Leguminosae"

#### VTT Biotechnology Conference

The VTT Biotechnology Conference will be held in Helsinki, Finland from June 10 to 13, 2001. The theme of the conference will be Plant Biotechnology - Better Products from Better Plants. The development of plant molecular biology has been very rapid during the past years. Today, the emphasis of genetic engineering of plants has shifted from improvement of agronomic traits to improvement of quality traits. This applies to medicinal as well as to food or feed plants. The production of desired plant compounds can be enhanced by metabolic engineering, or novel products such as therapeutic proteins, enzymes and bioploymers can be produced through heterologous systems. This congress will cover a wide spectrum of the most up-dated scientific outcome in the field of plant biotechnology and the topics will include: plant secondary metabolites; therapeutics from plants; food quality improvement; industrial applications; regulatory affairs and consumers attitudes; past and future of plant biotechnology

For further information, please contact: Dr. Kirsi-Marja Oksman-Caldentey (fax: ++358 9 455 2103; e-mail: kirsimarja.oksman@vtt.fi ) or Dr. Anna Maria Nuutila (fax: ++358 9 455 2103; e-mail: anna-maria.nuutila@vtt.fi). Conference web-site: http://www.vtt.fi/bel/new/plant/.

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**PSNA NEWS** 

August 4-8, 2001

To be held at The Westin Hotel, One North Broadway, Oklahoma City, OK, USA

#### "Phytochemistry in the Genomics and Post-Genomics Eras"

#### INDIVIDUAL ROOM RESERVATION FORM (Please type or print clearly)

All rooms are charged an additional 9.875% for state & occupancy tax.

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| Send reservation form and payment in   | formation to:   | Phytochemical Society of North America<br>C/o Ms. Marilyn Nance<br>The Noble Foundation (580)-221-7311 - c<br>Plant Biology Division (580) 221-7380 – fa<br>2510 Sam Noble Parkway<br>Ardmore, Oklahoma 73401 USA<br>e-mail: manance@noble.org | office<br>ax   |

## August 4-8, 2001

To be held at The Westin Hotel, One North Broadway, Oklahoma City, OK, USA

#### "Phytochemistry in the Genomics and Post-Genomics Eras"

#### OKLAHOMA CITY TOUR FORM (Please type or print celarly)

To sign up for the tour of the Oklahoma City National Memorial, stockyards, State Capitol and the National Cowboy Hall of Fame, please fill out the form below and send it by mail, fax or return e-mail with registration.

| Name:              |          |               |              |  |
|--------------------|----------|---------------|--------------|--|
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| City:              | _ State: | Zip:          | Country:     |  |
| Number of Persons: |          | Cost (\$32.00 | per person): |  |

Send to: Marilyn Nance, Plant Biology Division, The Noble Foundation, P. O. Box 2180, Ardmore, OK 73402, phone: (580)-221-7311, FAX: (580) 221-7380 or e-mail: manance@noble.org.

#### Tuesday, August 7, 2001 – Let's See OKC! Tour 1:00-5:00 pm

Price includes transportation, guided tour, admission to the National Cowboy and Western Heritage Museum.

Driving tour includes world's largest working stockyards, downtown, Brick Town, Church Row, Myriad Gardens, historic homes of Heritage Hills, and Remington Land.

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Stops include:Oklahoma National Memorial (walking tour).Inside the State Capitol to view murals.Self-guided tour of National Cowboy and Western Heritage Museum.

Tour will begin and conclude at the Westin Hotel in Oklahoma.

PSNA NEWS

#### August 4-8, 2001

To be held at The Westin Hotel, One North Broadway, Oklahoma City, OK, USA

#### "Phytochemistry in the Genomics and Post-Genomics Eras"

#### **ABSTRACT SUBMISSION FORM (Please Type or Print Clearly)**

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| Gradu  | ate Students and Re  | cent Ph.D.s   |   |
| To encourage participation of young investudents and recent Ph.D.s to offset a portion you would be a set of the set of t | estigators in this meeting, f<br>on of the travel and registr<br>like to be considered for fin | inancial assistance will be<br>ation costs. Please check a<br>nancial assistance. | provided to graduate<br>any of the following if |
| Best Short Talk/Poste  | rTravel/Reg  | istration Assistance  | Date of Ph.D.                                   |

#### Abstracts must be received by June 15, 2001.

Send two copies of the Abstract and a copy on diskette (MS-Word, Work Perfect or ASCII) by Air Mail or e-mail to:

Phytochemical Society of North America 2001 Meeting c/o Richard A. Dixon Director Plant Biology Division The Noble Foundation 2510 Sam Noble Parkway Ardmore, Oklahoma 73401 USA (580)-221-7322 – office (580) 221-7380 – fax e-mail: radixon@noble.org

ABSTRACT FORMAT: Typed on plain paper in 12 point font and not exceeding 16.5 cm x 7.5 cm (6.5 in x 3.0 in).

#### Please submit abstracts by June 15, 2001

#### August 4-8, 2001

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#### "Phytochemistry in the Genomics and Post-Genomics Eras"

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| E-mail:           |          |      |          |  |

REGISTRATION FEES (Includes admission to all scientific sessions, reception, and banquet.) Due by May 30, 2001.

| Regular Registration <sup>a</sup> (with Symposium Vol. 36)    | US \$300.00 |
|---|-------------|
| Economy Registration (without Symposium Vol. 36)              | US \$200.00 |
| Non-member Registration <sup>d</sup> (with Symposium Vol. 36) | US \$350.00 |
| Non-member Registration <sup>d</sup> (without Symposium Vol.) | US \$250.00 |
| Student Registration <sup>b</sup> (without Symposium Vol.)    | US \$85.00  |
| Late Registration Fee <sup>c</sup>                            | US \$50.00  |
| Additional Banquet Tickets                                    | US \$30.00  |
| Additional Symposium Volume                                   | US \$100.00 |
| Tour of OKC Memorial, etc.                                    | US \$32.00  |
| 2001 Meeting T-Shirt (size: M, L, XL, XXL)                    | US \$10.00  |
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<sup>a</sup>Symposium Vol. 36 "Recent Advances in Phytochemistry", if purchased at the conference 50% off. <sup>b</sup>Requires Supervisor's Signature in Part A – below. °If Registration is received after June 15, 2001. <sup>d</sup>Includes membership fees.

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## August 4-8, 2001

To be held at The Westin Hotel, One North Broadway, Oklahoma City, OK, USA

#### "Phytochemistry in the Genomics and Post-Genomics Eras"

#### **MEETING PROGRAM**

#### Day I. Saturday, August 4.

6.00pm. Dinner

7.30pm. Opening remarks.

7.45pm. Keynote lecture: **Charles Arntzen** (Arizona State University, Tempe, AZ). Bioprospecting of saponins in desert legumes; a link between chemical ecology and human medicine.

#### Day II. Sunday, August 5. am.

#### Session I. Metabolic Profiling in Functional Genomics

8.30am: Lloyd Sumner (Noble Foundation, Ardmore, OK). LC/MS for metabolic profiling in Medicago truncatula.

- 9.05am: **Dayan Goodenowe** (Yol Bolsum Inc, Rycroft, Alberta, Canada). Fourier Transform Ion Cyclotron MS for metabolic profiling.
- 9.40am: **Richard Trethewey** (Metanomics Gmbh and Co KG, Berlin, Germany). High throughput metabolic profiling in potato and Arabidopsis.
- 10.15am: Coffee break
- 10.45 am: **Guy della-Cioppa** (Large Scale Biology, Vacaville, CA). Functional genomics by metabolic profiling using high throughput transient gene expression from a viral vector.
- 11.15am: Non-symposium talk.
- 11. 40am: Non-symposium talk.

12.00 noon. Lunch

#### Day II. pm

#### Session II. Bioinformatics and Phytochemical Pathway Modeling

2.00 pm: Douglas Gage (Michigan State University, East Lansing, MI). Isotopic analysis and modeling of pathway flux.

- 2.35 pm: **Pedro Mendes** (Virginia Bioinformatics Institute, Blacksburg, VA). Linking metabolic pathway and gene expression databases.
- 3.05 pm: Andrew Hansen (University of Florida, Gainesville, FL). A genomics approach to one carbon metabolism.
- 3.40 pm Poster Session I (with coffee, and cash bar at 5.00pm)

6.00 pm Dinner

- 7.30 pm Evening Session. Neish Young Investigator Symposium. "Phytochemical Analysis and Synthesis" Theunis G van Aardt (University of Mississippi, Oxford, MI). Chemical Synthesis of Pterocarpans.
  - **David Gang** (University of Michigan, Ann Arbor, MI). Analytical tools to investigate phenylpropanoid biosyn thesis in basil peltate glandular trichomes.
  - Ikhlas A. Khan (University of Mississipi, Oxford, MI). The role of analytical tools in the standardization and quantification of phytomedicines.

#### Day III. Monday, August 6. am

#### Session III. Loss- and Gain-of Function Genetic Approaches for Metabolic Pathway Gene Discovery

- 8.30 am: **Anne Osbourn** (Sainsbury Laboratory, John Innes Centre, Norwich, UK). Genetic approaches to understand ing the biosynthesis of triterpene saponins.
- 9.05 am: **Brenda Winkel-Shirley** (Virginia Polytechnic Institute, Blaksburg, VA). A mutational approach to dissection of flavonoid biosynthesis in Arabidopsis.

#### Day III. Monday, August 6. am (continued)

9.40 am: Yiji Xia (Akkadix Inc, La Jolla, CA). Biopanning by activation tagging.
10.15 am Coffee break
10.45 am: Ken Feldman (Ceres Inc, Malibu, CA). Functional genomics of cytochrome P450s.
11.20 am: Non-symposium talk

#### Day III pm

#### Session IV. Sequence-Based Approaches to Natural Product Pathway Gene Discovery

2.00 pm: Greg May (Noble Foundation, Ardmore, OK). Medicago truncatula functional genomics.

- 2.35 pm: **Mark Lange** (Novartis Agricultural Discovery Institute, Inc., San Diego, CA). Terpenoid biosynthesis as revealed by mass sequencing of mint oil gland cDNAs.
- 3.05 pm: **Toni Kutchan** (Leibniz Institute for Plant Biochemistry, Halle) Heterologous expression systems for plant natural product biosynthetic enzymes.

Poster Session II.

Free evening

#### Day IV. Tuesday, August 7. am

#### Session V. Chemical Synthesis, Structural Biology and Enzyme Specificity

- 8.30 am: **Daneel Ferreira** (National Center for Natural Products, University of Mississippi, Oxford, MS). Chemical synthesis of flavonoids, isoflavonoids and condensed tannins.
- 9.05 am: Seiichi Matsuda (Rice University, Houston, TX). Tinkering with terpene biosynthesis
- 9.40 am: **Joe Noel** (Structural Biology Laboratory, Salk Institute, La Jolla, CA). Structurally guided alteration of substrate and product selectivity in plant type III polyketide synthases.
- 10.15 am: Coffee break
- 10. 45 am: **Chloe Zubieta** (Structural Biology Laboratory, Salk Institute, La Jolla, CA) Structural biology of phenolic O-methyltransferases.

11.20 am: Non-symposium talk

#### Day IV. pm

Free afternoon. Organized tour of Oklahoma National Memorial and National Cowboy Hall of Fame, or opportunity to visit the Wichita Mountains National Wildlife Refuge.

6.30pm. Conference Banquet

After dinner speaker: **Joanne Chory** (Plant Biology Laboratory, Salk Institute, La Jolla, CA). Genetic approaches to brassinosteroid biosynthesis and function.

#### Day V. Wednesday, August 8. am

#### Session VI. Metabolic Engineering

8.30 am: Clint Chapple (Purdue University, West Lafayette, IN). Lignin.

**PSNA NEWS** 

9.05 am: Norman Lewis (Institute for Biological Chemistry, Washington State University, Pullman, WA) Lignans.

9.40 am: Dean DellaPenna (Michigan State University, East Lansing, MI) Vitamins and nutritional genomics.

- 10. 15am: Coffee break
- 10. 45 am: **Barbara Halkier** (Royal Veterinary and Agricultural University, Copenhagen) Cyanogenic glycosides and glucosinolates.

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March 2001

11.20 am: Non-symposium talk

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# Elsevier Publishes Volume 34 of Recent Advances in Phytochemistry

Volume 34 of Recent Advances in Phytochemistry - *Evolution of Metabolic Pathways*, the symposium volume resulting from the excellent 1999 PSNA Annual Meeting in Montréal, Canada organized by Vincenzo De Luca, Luc Varin, and Ragai Ibrahim, is the first to be published by our new publisher Elsevier. It contains 14 chapters, has 467 pages, and is available for purchase by PSNA members at a significant discount.

The past decade has seen major advances in the cloning of genes encoding enzymes of plant secondary metabolism. This has been further enhanced by the recent project on the sequencing of the Arabidopis genome. These developments provide the molecular genetic basis to address the question of the Evolution of Metabolic Pathways. This volume provides indepth reviews of our current knowledge on the evolutionary origin of plant secondary metabolites and the enzymes involved in their biosynthesis.



Contributors to this volume explore a wide range of topics that include:

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- $\ensuremath{\mathscr{V}}$  the evolutionary origin of polyketides and terpenes
- ${\mathscr O}$  oxidative reactions and evolution of secondary metabolites
- $\ensuremath{\mathscr{C}}$  the evolutionary origin of substitution reactions
- ${\mathscr S}$  structure-function relationships in the evolution of steroid sulfonation

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# Phytochemical Society of North America Sociedad Fitoquímica de América del Norte Société Phytochimique de L'Amerique du Nord

Volume 41, Number 3

October 2001

# PRESIDENT'S LETTER An Exciting Time for Phytochemistry

It is indeed an honor to address all of you as Society President for 2001-2002. It is a humbling privilege to be able to serve the PSNA membership and the community of phytochemists and plant biologists at large during this coming year. I must say that it is also a personal challenge to engage in phytochemical "business as usual" in light of the unspeakable tragedies of September 11. For me, it is a strange coincidence that 18 years ago, on that same date, I learned of the coup d'etat in Chile, the neighboring country to the south of Peru, which resulted in the death of President Salvador Allende. As a college biology major in Peru at that time, I remember the feelings of distress and helplessness at this tragedy, which shook me and my friends. As some of you might know, that event started a reign of terror from which Chileans have not yet fully recovered. It is sad that humanity keeps failing to learn from horrors of the past and continues to breed the new terrors for the future. My heart goes to all of you who might have

been directly affected by recent tragedies, and it is my sincere hope that those who hold the power to destroy will allow sanity, rather than madness, to prevail.

Let me share with you a few thoughts and proposals about the task at hand. First, I would like to say that I am here to promote the ambitions of the whole Society rather than impose my own opinions. My task is made much easier by the earlier addresses in this newsletter from our Past-President, Richard Dixon, as he has laid the ground work for what I believe should be the future direction of the PSNA. Like Rick, I believe that the coming of age of genomics and the start of post-genomics will change the nature of biological research for many years to come. While all of the life sciences are affected by these developments, I am convinced that this is an especially exciting time to be engaged in phytochemical research. We can now revisit fascinating problems, such as the origin and evolution of biochemical diversity, with amazingly powerful tools. As amazing as these tools are, however, it is actually our ability to reformulate old questions with a new perspective that will lead to novel and exciting science. As Rick has already alluded, we will be successful in these new endeavors only to the extent that we can make connections and establish fruitful partnerships with all the branches of knowledge that impact on phytochemistry. The recent PSNA meeting in Oklahoma City is a great example of what we should strive for in future PSNA endeavors. I was very pleased to see the start of productive dialogue between the "classic" phytochemists and our molecular and genomic colleagues. I was particularly delighted at the large attendance of bright young scientists and the high quality of their presentations and posters. This is, in my view, the most hopeful indication that PSNA is poised to take on future challenges. At the same time, I was a bit saddened by the lack of participants from Mexico and Latin America. This

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- ✓ Stars and Stripes: The Emancipation Oak
- ✔ Great Expectations: 2001 Annual Meeting Review



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Phytochemistry as Integrative Biology: from Ethnobotany to Molecular Ecology

August 3 - 7

Mérida, Yucatán, México

The Phytochemical Society of North America (PSNA) is a nonprofit scientific organization whose membership is open to anyone with an interest in phytochemistry and the role of plant substances in related fields. Annual membership dues are U.S. \$40 for regular members and \$20 for student members. Annual meetings featuring symposium topics of current interest and contributed papers by conference participants are held throughout the United States, Canada, and Mexico. PSNA meetings provide participants with exposure to the cutting-edge research of prominent international scientists, but are still small enough to offer informality and intimacy that are conducive to the exchange of ideas. This newsletter is circulated to members to keep them informed of upcoming meetings and developments within the society, and to provide a forum for the exchange of information and ideas. If you would like additional information about the PSNA, or if you have material that you would like included in the newsletter, please contact the PSNA Secretary and Newsletter Editor. Annual dues and changes of address should be sent to the PSNA Treasurer. Also check the PSNA website at www.psnaonline.org for regular updates.

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# PHYTOCHEMICAL PIONEERS

#### **Neil Towers**

#### Some Memories of a Budding Scientist in North America (1946-1965)

I grew up in Myanmar (formerly Burma) when it was a British colony. My parents sent me to boarding schools around the country run by Christian brothers who sadly lacked an interest in the natural sciences, particularly natural history. Living and travelling as a schoolboy in perhaps one of the most beautiful tropical countries on this planet, I developed a craze for natural history. I collected snakes, beetles, butterflies, dissected animals for parasites and tried to identify plants from books. It was a happy boyhood. On reflection I think I was lucky not to have lived in our computer and television age. I did not see a television program until I was about twenty two! I spent all of my holiday time escaping prayers and wandering through the enchanting countryside exploring nature. I was spellbound by the travels, adventures and ideas of Darwin, Wallace, Bates and many other famous explorers. That is exactly what I wanted to be. World War II intervened.

I came to Canada on a scholarship for ex-naval officers at the end of the war. I had many adventures during the war, quite a number of which would have been called unforced errors of life were they to be compared to a game of tennis! Having escaped from the Japanese and winding up in England and then Canada, my life changed and I was suddenly plunged into the cloisters of academia. My sunny days of adventure were over – perhaps forever.

I was saddened to find that there were very few enthusiastic natural historians in this new life in a university. My fellow undergraduates in fact never seemed to have had time to talk about the excitement of biology they were so busy cramming for exams. I found out also that the world appeared to have been already explored by my arrogant zoology instructors and there was little new to discover other than to climb very tall mountain peaks or dive deep under the sea. I was an Honours Zoology student at McGill University in Montréal at the time and was advised by zoologists that the secrets of the animal world really lay in the realm of statistics! Even genetics was all statistics according to them.



Botanists, in contrast, were fascinated by apparent trivia: they were excited by the shapes of leaves, the hairiness of plant structures (for which there are many unpronounceable names) the geometry of flowers and a phenomenon called 2N versus N. However these botanists seemed to love what they were doing and I was encouraged to join their ranks. They actually worked with their microscopes in the evenings. They suggested to me that the inner workings of plants e.g. how sugars are manufactured from a gas in light was irrelevant and for Heaven's sake don't spoil things by dragging chemistry into the picture in order to understand how a plant lives. Of course, electron

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October 2001

**PSNA NEWS** 

microscopy, the role of nucleic acids, the nature of enzymes etc. were not even dreamed of at that time. Professor R. D. Gibbs, a feisty botanist at McGill, kindled my interest in plant chemistry. He was considered a crank by other botanists as I found out later because he was fascinated by the chemical relationships between plants. In fact he was a chemotaxonomist at a time when chemists did not know the meaning of the word taxonomy and a botanist might have been embarrassed if accused of understanding anything about chemistry.

Here was a botanist who actually knew some phytochemistry and, Good Lord, this chap could actually draw chemical structures! We became good friends and I obtained an M.Sc. under his supervision. The research involved the chemotaxonomy of plant lignins and was published in Nature. Perhaps I was the first person at McGill to use the new technique of paper chromatography. Certainly, the chemists and biochemists at McGill seemed to be as yet unfamiliar with the use of this technology. Later on during a sabbatical leave with the enzymologist D.D. Davis at the University of East Anglia, I used to drink beer every afternoon with Dick Synge, one of the discoverers of paper chromatography, a Nobel Laureate, a seasoned beer drinker, and a very dangerous cyclist (after drinking of course). Curiously Gibbs suggested that paper chromatography would never work and that I should use fractional sublimation instead to separate my products of alkaline nitrobenzene oxidation, namely p-hydroxybenzaldehyde, vanillin and syringaldehyde. Clearly this was not good advice because his total NRC budget for the year was

#### continued from page 3

about \$115! Gibbs was old fashioned by current standards. It was good for me. It made me more of an independent scientist. When I asked him if he would be kind enough to read a draft of my thesis he was astounded. "I am here to *examine* you Towers, not to *help* you. It is your thesis - not mine!" Nowadays, of course, there are many rules in Canadian universities to chaperone graduates in thesis writing so that in the end I feel we sometimes produce the well-known camel instead of the desired racehorse.

I went to Cornell for my Ph.D. studies with Professor F. C. Steward, a distinguished English plant physiologist who boasted that he had never taken a course in botany in his life. Needless to say he thought that he was the founder of botany. He had achieved fame for his work on ion accumulation in plants. His students called him the Golden Bantam because he was small, a fearless fighter, and rather arrogant. He once told me that he regretted the fact that dueling was no longer encouraged as a means of resolving departmental quarrels among faculty. He meant it!

Steward had become interested in the use of paper chromatography for separating and identifying the many unidentified non-protein amino acids in plants. My Ph.D thesis was concerned with designing new methods for the identification of  $\alpha$ keto acids in plants. Quite boring actually. It was a wonderful period of study, however, because Steward had attracted extremely knowledgeable postdocs, such as John F. Thompson, and clever graduate students to his lab. He was also a research supervisor who was so busy chasing research dollars that we had complete freedom in our own programs. I think that this is still the case in many universities.

At Cornell I took Botany and Biochemistry and a course on enzymes by J. B. Sumner. After many years of tedious research Sumner had discovered that the fewer steps involved, the better were his yields of the hydrolytic enzyme urease which he was studying in Canavalia ensiformis (Jack bean). In fact, he discovered one day that a 32% acetone extract heated to about 60°C, filtered overnight through Whatman paper into a graduate cylinder, and placed in a refrigerator yielded a precipitate which, when examined under a microscope, was found to consist of "octahedral" crystals. The crystals had tremendous urease activity. Repeated analyses showed that it was a protein. This was in 1926. Sumner wrote in his lab notebook about this momentous day: "That night I slept but little". At that time of course the true nature of enzymes was unknown and the leaders in the field, among them the very famous German biochemists, Willstatter and Waldschmidt-Leitz, refused to acknowledge that a 26year-old American had actually isolated an enzyme and proven that it was nothing more than a protein. His discovery was treated with some ridicule which unfortunately made him rather bitter. Four years later when Northrup crystallized the proteolytic enzymes pepsin and trypsin from animal sources at the Rockefeller Institute and showed that they are also proteins, Sumner's achievement was acknowledged - they shared a Nobel prize. We had the privilege of repeating Sumner's work in our laboratory course and even of recrystallizing urease. Of course, like Sumner, we made the entry "That night I slept but little" in our laboratory notebooks.

I was offered a job as Assistant professor in the Botany Department at McGill and assigned to teach plant anatomy, plant physiology, plant biochemistry and help run introductory botany labs. I inherited an old physics lab which could only be accessed through a men's urinal, a bit of an annoyance to my women graduate students. After four years of working in this "lab" we discovered an open pool of about 40 kg of mercury

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under the wooden floor. It must have been "lost" by the physicists during their war research years. Also contributing to the poor working conditions were the feral cats that had taken up residence in the dark corners of this medieval set of rooms, occasionally emerging to produce a litter of young kittens on our chromatograms which had to be stored on the floor. The lab was cold and we often had to use gloves and overcoats to stay warm during the winter months. At that time, university startup money for research was unthinkable. Besides, there were tons of microscopes and herbarium sheets lying around. What more could a botanist want? Roy Waygood, the plant physiologist at McGill was most encouraging, allowing me access to his lab equipment and his knowledge of plant physiology and biochemistry.

Among the many wonderful graduate students in my lab was our illustrious PSNA stalwart Ragai Ibrahim. Seichi Yoshida of Tokyo Metropolitan University also joined me as a postdoc. Later his student, Minamikawa, joined my lab and much later on Minamikawa's student, Etsuo Yamamoto, came to my lab as a postdoc. That's three generations of great Japanese phytochemists! We spent a lot of time making 2D chromatograms of plant extracts, cutting out spots, eluting them, and so on. I remember my eight year old son spending an afternoon in my lab. After watching me for half an hour he asked "What are you going to be when you grow up Dad?" It seems that in his eyes I have never really grown up.

I was delighted to learn about the Birch and Donovan hypothesis in relation to flavonoids. Instead of being neatly derived (on paper) from two hexoses and a triose according to Geissman and Hinreiner, they now appeared to be derived from a hydroxycinnamate and acetate! We resolved to test this with the dihydro-

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#### **Neil Towers**

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chalcone glucoside, phloridzin. Alas! We were beaten by Neish's group at the National Research Council (NRC) of Canada in Saskatoon who proved this hypothesis with quercetin, and Grisebach's group in Germany who proved the hypothesis with an anthocyanin. It seems silly now but we were dreadfully disappointed not to have been the first to have proven that Birch and Donovan were right.

I spent a summer at the NRC laboratories in Ottawa with D.C. Mortimer and Paul Gorham, learning radiotracer techniques and carrying out <sup>14</sup>C photosynthesis studies. The following summer I spent with Stewart "Coumarin" Brown and Arthur Neish at the NRC laboratory (then called the Prairie Regional Laboratory) in Saskatoon studying coumarin biosynthesis. When I returned to McGill at the end of the summer Ibrahim and I prepared twodirectional chromatograms of the phenolic acids from a range of plants. Sprayed with diazotized nitroaniline or diatized sulfanilic acid they gave a range of beautiful colors. We had a special room set up with these large chromatograms adorning the walls for participants of the IX<sup>th</sup> Botanical Congress which was held at McGill, the Université de Montréal, and Sir

George Williams College (now Concordia University) that year. These chromatograms were works of art and admired by all who visited us.

After enjoying more than nine years at McGill I was invited by Art Neish to head up the Plant Biochemistry section of the NRC's Atlantic Regional Laboratory in Halifax, Nova Scotia to which he had been appointed Director. Neish was considered to be one of the outstanding phytochemists in Canada and I was delighted to join his institute as I had a great admiration for him as a scientist and also because I was jointly appointed as an Associate Professor to Dalhousie University in Halifax where Neish and I taught a course in comparative biochemistry. My graduate students at McGill accompanied me there and had the advantage of working in the well-equipped NRC labs and alongside distinguished Canadian scientists in chemistry (Gavin McInnis) and biochemistry (Leo Vining). We published many papers especially on the biosynthesis of interesting lichen compounds as well as on comparative phenylpropanoid metabolism in lycopods and fungi. We showed clearly that Ltyrosine is metabolized quite differently from L-phenylalanine, especially in vascular plants. Tyrosine is metabolized to acetate and its derivatives when introduced into plant tissues and phenylalanine is the gateway to phenylpropanoid metabolism. We also identified a new cyanogen from *Taxus* and studied its biosynthesis showing that both the nitrogen and carbon are derived from L-phenylalanine. We discovered psilotin, a glucoside derived from ahydroxycinnamate and one equivalent of acetate, in the primitive ferns *Psilotum* and *Tmesipteris*.

I next moved to the University of British Columbia in Vancouver as Head of Biology and Botany, an administrative position which tore me away from thinking time and plunged me into the petty life of administration in a then impoverished Canadian university. After five years, a sabbatical leave in England where I studied enzymes with D.D. Davies at the University of East Anglia, convinced me to resign as Head and settle down again to research and teaching. As most biology students at our universities do not enjoy chemistry my classes were small and were therefore a great pleasure to teach. Many more graduate students and postdocs passed through my research program and it would take many more pages to describe our further achievements in phytochemistry.

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#### **President's Letter**

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clearly shows that we must work harder to forge appropriate links with our colleagues to the south. As I am sure you would agree, there is a wealth of high quality phytochemical research being done in countries such as Mexico, Argentina, and Brazil. We must make a greater effort to welcome these colleagues to our PSNA family so that all of us can benefit from their enormous diversity of views and resources. I will be in touch with the Executive Committee soon to discuss these and other issues, and I welcome your ideas to recruit and retain the greatest possible diversity of membership in PSNA. In line with the initiatives of Rick Dixon last year, I proposed at the business meeting in Oklahoma City to host the 2002

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PSNA annual meeting in a location which addresses both the challenges and opportunities outlined above. I was pleased that my proposal met with the approval of the membership and am delighted to confirm that next year's meeting will be held on August 3-7, 2002 in Merida, Mexico. Merida

# **GREAT EXPECTATIONS**

#### Review of the 2001 PSNA Annual Meeting in Oklahoma City

With the development of bioinformatics, fueled by the ever-expanding accumulation of sequence information and an ever-increasing array of molecular techniques, phytochemistry is moving into a new era. These powerful technologies are allowing high-throughput analyses of chemical and biological information producing huge volumes of data. How these new tools can be used to solve phytochemical problems was the theme of the 2001 PSNA meeting held in Oklahoma City, August 4-8. The conference proceedings were officially opened by Michael A. Cawley, the president of the Noble Foundation. Mr. Cawley welcomed conference delegates to Oklahoma City and gave a short pep-talk to us about research. Charles Arntzen (Arizona State University) gave the keynote address describing the studies resulting from a collaborative research program between the M.D. Anderson Cancer Center and the Arizona Biomedical Institute. The identification of target sites for saponins in mammalian cells resulted in the identification of a novel class of saponins from Acacia victoriae, a desert legume. This novel class of saponins appears to have significant anti-cancer properties and Dr. Arntzen presented a possible model for its mode of action in vivo involving the inhibition of nf-KB from binding DNA.

Starting the first session, Lloyd Sumner (Samual Roberts Noble Foundation) described his group's work on metabolic profiling of *Medicago trunculata*. Sumner presented a strategy in which the metabolome is divided into subclasses, based on solubility. Sequential extraction followed by multidimensional analysis techniques, including GC-MS and HPLC-MS, allowed for the separation and specific identification of individual metabolites. Sumner reported that as many as 300-500 primary metabolites can be identified and quantified using this technique, allowing for the comparison of the metabolic phenotypes of mutants. The use of tandem mass spectrometry (MS/MS, MS<sup>n</sup>) for chemical structure identification was described as well as an application for demonstrating the profiling of saponins from Medicago. Dayan Godenowe (Phenomenome Discoveries, Inc., Saskatoon, Canada) presented the latest developments in metabolome analysis at his company. Using strawberry as a model system, the proprietary technology developed at Phenomenome Inc was able to identify over 6000 metabolites in the fruit. Fourier Transform Ion Cyclotron MS allowed for the identification of thousands of individual metabolites. Coupling this staggering amount of data with micro-array analysis and proprietary software to analyze the data, the company was able to identify over 20 pathway changes during fruit ripening as well as a number of other new observations. Richard Trethewey (Metanomics GmbH, Berlin, Germany) discussed the importance of analyzing phenotypes by metabolic profiling, allowing for the categorization of the corresponding genotypes. Trethewey described how using GC-MS and tandem mass spectrophotometry (MS<sup>n</sup>) allowed for a faster and more comprehensive analysis of the various metabolic networks involved in starch production of a number of mutant and transgenic lines of potato. Mycorrhyzal symbiosis between roots and fungi is a poorly understood, yet ubiquitous condition in nature. Dieter Strack (Institut für Pflanzenbiochemie, Halle, Germany) offered evidence to show that the de novo synthesis of isoprenoids is involved in the interaction between the root and the fungi. Root leucoplastids were

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transformed to chromoplastids following colonization by the fungus, which were closely associated with arbuscules. Moreover, root cortex cells containing arbuscules also accumulated carotenoids and apocarotenoids, suggesting that isoprenoid biosynthesis, via the nonmevalonate pathway, is an intrinsic part of this fascinating interaction. Asaph Aharoni presented the results of a collaboration between Plant Research International (Wageningen, Netherlands) and Phenomenome Inc using the proprietary analysis systems of Phenomenome to categorize the gene expression and metabolite changes during strawberry fruit development. This analysis lead to the identification of SAAT, a non-specific ester-forming enzyme that is expressed during fruit development and may be important for the flavours and aromas of the strawberry fruit. Guy della-Cioppa (Large Scale Biology, California) described to us the utility of "geneware," a virus-based gene delivery system that allows for the quick temporary overexpression of a gene in growing plants. Entire fields can be infected with the viral vector allowing for large scale analysis of multiple genes. Using the viral vector to transform plants with an entire cDNA library may provide a powerful method of gene-function analysis by generating a large volume of metabolic data that could be further analyzed to determine specific metabolic changes to actual pathways. Such analyses would certainly require automated high throughput systems to be feasible. Douglas Gage (Michigan State University), described the value of computer modeling of pathway flux and demonstrated the efficacy of modeling using glycine betaine metabolism. Gage explained the importance of models to allow for the calculation of flux and maximum

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yields, and for the formulation of testable hypotheses. Metabolic engineering of glycine betaine, so far has been largely unsuccessful, because flux in this pathway is not well understood, claimed Gage. (For more information visit the Web site http:// www.purdue.edu/cfpesp/models/ models.htm). C1 metabolism is crucial to secondary metabolism and in fact, as Andrew Hanson (University of Florida) described, secondary metabolism creates a massive demand for C1 far in excess of the synthesis of primary metabolites. The C1 network is difficult to study because many of the cofactors are highly unstable and in very low abundance. Using available genomic data, EST libraries and reverse genetic approaches, Hanson described how he and his collaborators have been able to find exciting new information about this pathway including the identification of a formerly unknown gene, an ATP-dependant 5-formyltetrahydrofolate cycloligase. Pedro Mendes (Virginia Tech) described the importance of using bioinformatics systems to better understand the vast amount of data that can be created from genomic, proteomic, and metabolic profiling. Mendes defines the identification of gene function as the "identification and quantification of the molecular interactions involving the products of a gene". Using this broader understanding of gene function, the importance and power of combining the data from genomic, proteomic, and metabolomic profiling was described. This "cellular profiling" as described by Mendes could be used to generate a robust model based on the rich data sets that are becoming accessible.

The 2001 Arthur Neish Young Investigator Minisymposium included four speakers. T.G. Van Aardt (University of Mississippi) described his group's success in the chemical synthesis of enantiomer-specific pterocarpins including the first transpterocarpin. Using purified glandular trichomes from basil (Ocimum basilicum), David Gang (University of Michigan) and collaborators were able to identify new methyltransferases involved in phenylpropanoid metabolism by combining an EST database with metabolic analysis. Interestingly, these two methyltransferases differ only in one amino acid yet have different substrate specificities. Most phytochemists have been encouraged by the increasing demand for herbal remedies in North American markets, but with this increase in demand comes a need for the assurance of the authenticity, quality, safety, and efficacy of the wide range of products available to the consumer. Ikhlas Khan (University of Mississippi) is developing new approaches to address these concerns based on HPLC, GC, and CE technologies. Confirming the validity of these products is not a simple matter, as often the active compounds have not yet been identified and reliable surrogate markers need to be developed. M.H. Cho described new findings in the biosynthesis of allylphenol lignans in the creosote bush (Larrea tridentata). Specifically of interest is the antioxidant nordihydroguaiaretic acid.

Anne Osbourn (Sainsbury Laboratory, Norwich, UK) described her group's work on triterpenoid saponins. Saponins are common secondary metabolites with anti-fungal properties. Using a variety of genetic approaches, Osbourn's group has been able to identify a key step in this pathway, cycloartenol synthase, as well as a new distinct enzyme, amryn synthase. Brenda Winkel-Shirley (Virginia Tech) presented work from her lab that demonstrated that proteinprotein interaction occurs among enzymes involved in flavonoid biosynthesis. Chalcone synthase was shown to associate with both DFR and chalcone isomerase, both in vitro and in vivo. Moreover, Winkel-Shirely presented evidence that CHI, which is normally a soluble cytosolic enzyme,

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interacts with DFR - an ER-membrane-bound enzyme - using immunogold localization. Activation tagging is a technique that allows the functional identification of genes by forcing an overexpression of the gene product. Yiji Xia (Noble Foundation, Oklahoma) presented the discovery of *pap1-D*, a MYB-like gene involved in phenylpropanoid biosynthesis. PAP1 acts as a transcription factor to up-regulate the phenylpropanoid pathway resulting in an increase in phenylpropanoids throughout development. Kenneth Feldmann (Ceres, Inc) described the efficacy of using a reverse genetics approach in Arabidopsis (T-DNA knock-out) to identify the function of the difficultto-isolate cytochrome P-450 monoxygenases. Kazuki Saito (Chiba University, Japan) used differential display to compare the expression of genes between two forms of Perilla frutescens. This led to the identification of a range of genes, including a MYC-transcription factor and a novel anthocyanin 5-glucosyltransferase. John Jelesko (Virginia Tech) and collaborators to examine alkaloid biosynthesis in tobacco used a similar new tool, fluorescent differential display. This method uses random oligos coupled with a poly-T primer to reversetranscribe the mRNA pool followed by high-throughput analysis on an automated sequencing machine. This method allows for the analysis of genes without using an EST or cDNA library. Using this approach they were able to identify a gene, an AAA-AT-Pase, whose role in alkaloid biosynthesis isn't yet clear, but has eluded identification by more traditional means. Gregory May (Noble Foundation) described ongoing research involving many groups at the Noble Foundation to develop an integrated functional genomics approach to Medicago trunculata. Using a variety of expression profiling tools, including microarray, differential display,

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#### **President's Letter**

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is the capital of the province of Yucatan, a beautiful colonial city with marvelous surroundings, from gorgeous beaches to fascinating archaeological ruins, not to mention a superb (and very spicy) regional cuisine. Merida is also the home of the Centro de Investigacion Científica de Yucatan (CICY), a 20-year-old research institute which stresses plant biochemistry and biotechnology as major areas of focus. I am currently working with two colleagues (and friends), Felipe Vazquez-Flota and Victor Loyola-Vargas, on the logistics of the meeting and will report in the next newsletter on various plans as they develop. I can also assure the PSNA membership that you can fully expect to experience Latin American "joie de vivre" at its very best, as Mexican people are among the most hard-working and funloving folks you'll ever meet, an unbeatable combination. This means, among other things, that you can expect to (and will be expected to) imbibe a large number of margaritas

#### **2001 Annual Meeting Review**

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and EST analysis, combined with high-throughout metabolite profiling, it is hoped that this integrated approach will provide scientists with a new powerful way to answer key problems in legume biology. Bernd Lange (Syngenta Research & Technology, California) reported the results of using combined computational and expression-based analysis on peppermint oil biosynthesis. Coupling expression-based data with metabolic profiling allowed an integrated approach to describe the dynamic flux through the isoprenoid pathway. Toni Kutchan (Institut für Pflanzenbiochemie, Halle (Saale), Germany) and participate in several merengue and salsa dances, in addition to our planned scientific activities. If only for these reasons, it is my ardent hope that many of you will be able to join us next year in Merida. But wait, there's more. Consistent with the location of the meeting and future direction of PSNA, I have proposed the following tentative theme for next year's meeting: Phytochemistry as Integrative Biology: From Ethnobotany to Molecular Ecology. The rationale is to continue encouraging the building of bridges between disciplines that normally do not talk to each other a whole lot. My colleagues in Mexico and I are currently working on speaker selection and we are open to any ideas for topics, special sessions, or other activities. Please feel free to contact me or any Executive Commitee member at any time (preferably very soon) with your suggestions, and feel free to be creative. Cautionary note: I am not known to shy away from wacky ideas. It is my hope that we will be able to span the phytochemical disciplines from ethnobotany to ecology throughout the meeting. For example, a topic such as glucosinolates is now amenable to molecular and genetic analysis in model systems such a Arabidopsis. At the same time, there is substantial classical phytochemical literature on these fascinating compounds as well as impressive ethnobotanical studies. More recently, groups at the Max Planck Institute for Chemical Ecology at Jena (Germany) are applying molecular and genomic approaches to approach the evolution of glucosinolate diversity as well as their ecological significance. I am hopeful that we can bring these people together in the same session and apply this model to the rest of the meeting. The program will be included in the next newsletter. I thank you again for the honor of serving as your President and look forward to working with you over the next year. Please accept my warmest wishes for a successful, productive, and peaceful year. I hope to see you all next summer in Merida!

> Hector E. Flores Pennsylvania State University University Park, Pennsylvania hef1@psu.edu

described some successes in cloning Papaver somniferum alkaloid biosynthetic genes using a sequencebased approach. For example salutaridinol-O-acetyltransferase was cloned using a consensus sequence developed from an acetyl-transferase in Cantharanthus roseus. A proteomics-based approach was used to analyze proteins in poppy latex and was able to identify a number of the proteins involved in morphine biosynthesis. Daneel Ferreira (University of Mississippi) described recent advances in the stereospecific synthesis of monomeric flavanoids, using poly-(L)-lysine or poly-(D)-lysine, they

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able to synthesize stereo-specific monomers. Altering enzyme specificity was a goal of Seiichi Matsuda (Rice University, Texas) using cycloartenol synthase. Matsuda reported that a single amino acid conversion (isoleucine to valine) within the active site reduced the "steric bulk" of the site allowing for alternative substrates to fit within the pocket. The mutant cycloartenol synthase also acted as a lanesterol synthase. Altering enzyme specificity was also the focus of Joe Noel's (Salk Institute, California) work. Working with

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# NEISH MINISYMPOSIUM FOR YOUNG INVESTIGATORS

#### **Profiles of This Year's Speakers**

#### Man-Ho Cho

Man-Ho Cho began his work in natural products biochemistry as an undergraduate in the Department of Genetic Engineering, Kyung Hee University, Korea, and completed both his M.Sc. and Ph.D. at the same institution under the guidance of Prof. Tae-Ryong Hahn. His M.Sc. studies involved the purification, expression and characterization of a pea chloroplastic fructose 1,6-biophosphatase and an Escherichia coli thioredoxin. For his Ph.D., he focused more on various important natural products and their biosynthesis. This included biosynthesis of the chalcone pigments in safflower (Carthamus tinctorius), during which time he purified and characterized precarthamin decarboxylase, an important enzyme involved in the color transition of its floral tissues from yellow to red. Precarthamin decarboxylase catalyzes the oxidative decarboxylation of precarthamin (yellow pigment) to carthamin (red pigment). Additionally, Dr. Cho isolated and identified anthocyanins from pigmented rice, shikonins from Lithospermum erythrorhizon, and carthamin and safflower yellow pigments from safflower. Dr. Cho is currently studying lignan biosynthesis using the creosote bush (Larrea tridentata) in Dr. Norman G. Lewis' laboratory in the Institute of Biological Chemistry at Washington State University. The creosote bush has a vast array of allylphenol lignans, of which nordihydroguaiaretic acid (NDGA), a potent antioxidant, is the major metabolite. Some of these lignans also have striking anti-viral activities, including against HIV. To elucidate the biosynthetic pathway to NDGA and its congeners, he administered  $[U^{-14}C]$ -Phe to L. tridentata seedlings, which established that it was efficiently converted into NDGA and related allylphenol congeners. Dr. Cho is currently completing his studies on the enzymology and protein biochemistry of allylphenol coupling, as well as on the downstream hydroxylation stages involved. He is also finalizing papers on the work completed to date, especially on the synthesis, biosynthesis and protein biochemistry of the optically active allylphenol lignans present in *Larrea* species.

#### **David Gang**

David Gang is currently a post-doc in the lab of Eran Pichersky in the Department of Molecular, Cellular and Developmental Biology at the University of Michigan, Ann Arbor. His current research focuses on the biosynthesis specialized of metabolites in aromatic culinary herbs, such as sweet basil and perilla. David's research seeks to elucidate the biosynthetic pathways that produce novel and important plant specialized metabolites, on uncovering the mechanisms responsible for the evolution of these pathways in the plant kingdom, and on understanding the function of a given specialized metabolite in the biology and physiology of a given plant species. David believes that the most productive approach in this area has been a multidisciplinary one that utilizes tools from the fields of chemistry, biochemistry, molecular biology, plant physiology, whole organism biology and ecology. Understanding the role of specialized metabolites in plants requires an understanding of the whole complexity surrounding its formation and utilization.

David obtained a B.S. in Botany-Molecular Biology from Brigham Young University, Provo,

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Utah. During his undergraduate years he worked with Darrell Weber on several projects, incorporating biochemical, molecular and ecological approaches. He then moved to Washington State University, Pullman, where he worked with Norman Lewis as he obtained his Ph.D. in Plant Physiology. While at WSU, David cloned the first genes to be identified in the lignan biosynthetic pathway, which leads to important specialized metabolites such as podophyllotoxin (an anticancer drug) and SDG (a phytoestrogen). Included among these genes were those for the dirigent proteins, which are the first proteins identified that are able to control phenoxyradical coupling, yielding enantiomerically pure products. In addition to their role in producing optically active lignans, these dirigent proteins may also provide a template for lignin formation in plants.

While at Michigan, David was able to demonstrate that the branch of the phenylpropanoid pathway leading to important flavor and aroma molecules, such as eugenol and methylchavicol (estragole), is localized to the specialized peltate glandular trichomes on the surface of the leaves. This has made characterization of this pathway much simpler since these structures can be isolated from the rest of the plant tissue for biochemical and molecular analysis. In addition, using a biochemical genomics approach, he was able to isolate cDNAs encoding the final enzymes in the pathway to methylchavicol and methyleugenol, namely chavicol O-methyltransferase (CVOMT) and eugenol O-methyltransferase (EOMT), respectively. He found that these enzymes evolved within the plant small molecule Omethyltransferase gene family, but

#### **Neish Symposium Speakers**

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not from (iso)eugenol *O*-methyltransferase from *Clarkia breweri* (IEMT), another enzyme in this gene family that is able to carry out the same reactions. David worked in collaboration with Chloe Zubieta in Joe Noel's group at the Salk Institute, LaJolla, CA, to show a rational structural basis for the substrate discrimination between CVOMT, EOMT and IEMT. The difference in substrate specificity between CVOMT and EOMT was caused by a single amino acid substitution resulting from a single point mutation in the corresponding genes.

David has recently accepted a position as an Assistant Professor in the Department of Plant Science at the University of Arizona. He will be moving to Tucson early in 2002 and will continue with the work he began while at Michigan.

#### Ikhlas Khan

Ikhlas Khan finished his Master's degree in chemistry from the Aligarh Muslim University Aligarh in India and joined Dr. H. Wagner's group at the Institute for Pharmaceutical Biology, University of Munich, Germany for his Ph.D. After completing his Ph.D., Ikhlas travelled to the United States for a postdoc position for two years, but returned to Europe and worked for three years with O. Sticher at the Swiss Federal Institute of Technology (ETH) Zurich, Switzerland on the isolation and structure elucidation of natural products from Papua-New Guinea plants. Finally, he returned to the U.S. and has since been working at the National Center for Natural Products Research at the University of Mississippi. The main focus of his research is the isolation and structure elucidation of natural products from herbal medicines and fingerprinting the chemical profile of

such plants for quality control purposes. His group has developed several analytical methods for the quantification of marker compounds. His main research program is focused on developing new tools for analysis, isolation of standards and identifying active components in herbs with collaborative work. In summary, his research is aimed at providing consumers, as well as industry, with scientific evidence of the efficacy of these traditional medicines, and also developing the tools to analyze them.

#### **Theunis van Aardt**

Theunis van Aardt started his training in 1992 at the University of the Orange Free State, South Africa and completed his Bachelors' degree majoring in chemistry and zoology. After completing his Honors degree he continued with an M.Sc in organic chemistry, but upgraded that study in November 1997 to a Ph.D. in synthetic organic chemistry. During the course of his studies he also applied himself to both practical and theoretical lecture responsibilities. Having completed his studies in May 2000 under the supervision of Dr. D. Ferreira and Dr. H. van Rensburg, he is currently in the middle of postdoctoral studies in the National Center for Natural Products Research, School of Pharmacy, University of Mississippi. As a synthetic organic chemist, his current project supervised by Dr. D. Ferreira is the stereoselective synthesis of radiolabeled flavonoid precursors to condensed tannins.

The work discussed in his seminar was taken from his thesis: Direct synthesis of pterocarpans *via* aldol condensation. Although pterocarpans have interesting medicinal properties, lengthy multistep routes and a lack of phenolic hydroxylation pattern diversity, limit synthetic protocols to these phytoalexins. He thus developed a synthetic approach towards pterocarpans based on the

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aldol condensation between benzaldehydes and phenylacetates to afford 2,3-diaryl-3-hydroxypropanoates. Reduction and cyclization would afford 3-benzylsulfanylisoflavans which could then be cyclized to yield the racemic pterocarpans. The 3-benzylsulfanylisoflavans also served as precursor to enantiopure cis- and trans-6αhydroxy-pterocarpans via oxidation and thermal elimination, yielding isoflav-3-enes, followed bv asymmetric dihydroxylation and cyclization. The 2,3-diaryl-3hydroxypropanoate aldol products allowed him to reverse the order of cyclization, *i.e.* initial C-ring formation followed by B-ring closure, thus providing the first synthetic access to the hitherto unknown 6α,11α-*trans*-pterocarpans. Thus, aldol condensation and cyclization afforded the transfused 2,3-disubstituted dihydrobenzofuran. Subsequent reduction and Mitsunobu cyclization yielded the  $6\alpha$ ,  $11\alpha$ -*trans*-pterocarpans.

He sees himself applying my training to the development of synthetic routes to natural products. Thus, allowing the structure elucidation of these and also, where applicable, access to naturally rare metabolites. This would necessitate not only the development of new synthetic reactions and/or conditions, but also new protective groups. Within an academic environment he sees himself as both teacher and researcher, developing the theoretical and practical abilities of students at all levels, thus giving them the foundation to address the field of natural products chemistry. In conclusion, he dares to make the statement that synthetic chemistry must not be neglected, since despite it's limitations, it is still the most powerful tool to understanding the physico-chemical properties of molecules as well as a tool to unambiguous structure elucidation.

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## STUDENT AWARD WINNERS

#### Profiles: 2001 Winners of the Best Paper Awards: Ora Carter and Michael Austin

#### **Ora Carter**

I am currently working towards finishing my Ph.D. in Plant Physiology in Rod Croteau's laboratory at the Institute of Biological Science, Washington State University. I work on two projects, the first being pathway reconstruction in microbial hosts. Carvone, a secondary metabolite from spearmint (Mentha spicata), is used as a model system for metabolic engineering and pathway flux of a simple, four step, biosynthetic pathway. This monoterpene is biosynthesized in glandular trichomes and the biochemistry and cellular organization has been well defined. Carvone is an important insecticidal deterrent and has pharmaceutical value. The carvone pathway provides an opportunity to establish the feasibility of and requirements for the transplantation of an entire metabolic pathway, first to E. *coli* and then to a non-producing plant. I also work on the bifunctional diterpene cyclase, abietadiene synthase. This work investigates the interaction of two distinct reactions in separate active sites. Its triterpenoid products: abietadiene, levopimaradiene and neo-abietadiene, are the major constitutive and wound-inducible components of rosin acids in conifers. By mutational analyses and enzymatic assays of the recombinant protein, we can dissect the A and B domain activities and investigate the bases involved. Abietadiene synthase's bipartite proton-initiated and ionization-initiated cyclizations of geranyl geranyl diphosphate (GPPS) to form the abietane skeleton are targets for protein engineering. My future goals are to continue working in the field of metabolic engineering and perhaps to contribute to our understanding of the complexities of plant mechanisms which are involved in

pathway regulation and flux. Ultimately, I hope to be involved in a project to improve crops for human health and nutrition. I would like to comment on how I came to work in the field of natural products. A career in the biological sciences came a little later for me than most of my colleagues, as one of my children was already in Jr. High School. But my interest in natural products actually began some years ago. I was looking for preventative health care for my children, especially immunity enhancers. This led me to attend several herbal conferences, where I was immediately struck by the health and vigor of the elderly women who were lifelong practitioners. After researching endogenous medicinals, I began to experiment with crude extractions from plants collected from my New England property and the surrounding woodland. Back then I considered a career in therapeutic herbalism, only later to discover I had a surprising and overwhelming fascination with the biological sciences. I knew by experience that certain plants had beneficial effects but I wanted to understand why these plant extracts had very specific physiological activity and what compounds were responsible for the effects. And I wanted to ascertain for myself that the overall benefits of crude plant extracts were not actually diminished in the further processing to achieve pharmaceutically pure compounds. My observation had been that the best of the herbals were hardy weeds and that the roots were often the plant part chosen by native healers. Further, I began to wonder about what we could achieve, insofar as human health and nutrition, if crops were selected with some of the inherent qualities of our hardy herbals. In this manner, already a mother of three wonderful children, my scientific

career began to take root. Since then, developing as a plant scientist has provided a platform of inquiry for this innocuous fascination with plants to continue to grow.

#### **Michael Austin**

I grew up in Springfield, Missouri, and spent a few years after high school studying philosophy and sociology at the local state college (Southwest Missouri State University). I eventually rejected this career path, but was unsure of what alternative I should pursue. I postponed completion of my degree for a number of years while I explored various positions related to the care and education of people with developmental disabilities.

Reflecting upon which aspects of philosophy I either loved or disliked eventually led me to consider science, a career that I had not previously given much thought. A fascination for life pulled me towards biology, but I quickly realized that a chemical understanding afforded a better perspective from which to examine life. Although I have taken a rather circuitous and non-traditional path to the study of biochemistry, I regret none of these 'distractions'. "Nothing is a waste of time if we use the experience wisely", as Auguste Rodin once put it. I feel that each of these experiences has contributed to my development both as a person and a scientist.

I graduated from SMSU in 1998 with a B.S. in chemistry (emphasis in biochemistry), with minors in philosophy and biology. I then entered the Chemistry and Biochemistry graduate program at the University of California at San Diego, where I joined Joe Noel's group in the Structural Biology Laboratory (SBL) at the

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**PSNA NEWS** 

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# STUDENT TRAVEL AWARD WINNERS

Aharoni Asaph. Profiling Fruit Maturation:From Gene Expression to Metabolic Pathways in Strawberry. Plant Research International, The Netherlands.

**Michael B Austin.** Structural and Engineering Studies of Stilbene Synthases. The Salk Institute for Biological Studies, USA.



Hector Flores and Michael Austin

Antoine Bily. Phenylpropanoïds and Corn Resistance to *Fusarium* graminearum:: Role of Cell-Wall Bound Dehydrodimers of Ferulic Acid. University of Ottawa, Canada and Université de Pau et des Pays de l'Adour, France.

**David A. Bird.** Identification of a Vacuolar Sorting Determinant in the Berberine Bridge Enzyme has Implications for Sanguinarine Biosynthesis. University of Calgary, Canada.

**Jennifer Boatright.** Identification and Characterization of a Petal-Specific ALDH Gene from Snapdragon. Purdue University, USA.

**Rico A. Caldo.** Dissecting Wheat-Leaf Rust Interaction with Gene Expression Profiling. Oklahoma State University, USA.

**Ora Carter..** Engineering Carvone for Production in Microbial Hosts. Washington State University, USA.

**Christopher D. Dana.** Homology Modeling of Mutations in the *Arabidopsis* Chalcone Synthase Gene. Virginia Tech, USA.

**Ying-Shan Han.** Analysis of Anthraquinones in the Elicited Cell Cultures of *Cinchona* 'Robusta' by HPLC with Photodiode Array and Mass Spectrometry Detection. Gorlaeus Laboratories, The Netherlands.

Jeremy Kapteyn. Genetic Diversity of *Echinacea* Species and the Regulatory Role of PAL in the Accumulation of Caffeic Acid Derivatives. Rutgers University, USA.

**Soo-Hee Kim.** Hydrogen Migration in Cyclization of Farnesyl Diphosphate by Amorpha-4,11-diene Synthase As Probed by Mass Spectrometry. Seoul National University, Korea.



David Bird



Ora Carter

**Sean G. Koenig.** Structural Comparison of Plant Sesquiterpene Cyclases. The Salk Institute for Biological Studies, USA.

Andres Lopez. Phytochemistry and Biological Activity of *Piper lanceaefolium* University of British Columbia, Canada.

**Susan Marles.** Reduced Expression of Dihydroflavonol Reductase is Associated with the Yellow-Seed Phenotype in Certain Brassicaceae. University of Saskatchewan, Canada.

**Fatima Mouhajir.** Antimicrobial Thymohydroquinones of Moroccan Nigella Sativa Seeds Detected by Electron Spin Resonance. University of British Columbia, Canada.

**Hosana M.D. Navickiene.** Molluscicide, Insecticide and Fungicidal Activity of Extracts and Amides from *Piper tuberculatum*. NuBBE-Instituto de Quimica, UNESP, Brazil.

**Isagani D. Padolina.** Metabolic Channeling in Elicited Liquid Cell Cultures of *Cephalocereus senilis* 



(Old Man Cactus): Metabolic Profiles and Pulse-Chase Experiments. University of Texas, USA.

Sang-Un Park. Antisense RNA-Mediated Suppression of Benzophenanthridine Alkaloid Biosynthesis in Transformed Cell Cultures of California Poppy. University of Calgary, Canada.

**Darren Peters.** Induction of Dihydroflavonol Reductase and Condensed Tannin Synthesis in Response to Wounding and Herbivory in Trembling Aspen (*Populus tremuloides*). University of Alberta, Canada.

Jeannine Ross. The Biological Roles of Methylsalicylate and Salicylic Acid in the Flowering Plant, *Clarkia breweri*. University of Michigan, USA.

Sittiruk Roytrakul. Proteomics Biosynthesis of Alkaloids in *Catharanthus roseus*- Effect of Elicitors on Indole Alkaloid Formation. Leiden University, The Netherlands.

**Timmy D. Samuels.** Differential Gene Expression in Wheat Roots in Response to Infection by the "take-all" Fungus *Gaeumannomyces* 



Hector Flores and Sang-Un Park



Ying-Shan Han

*graminis* var.*tritici*. Oklahoma State University, USA.

**Tara M. Sirvent.** Increased levels of the Anthraquinone Hypericins in *Hypericum perforatum* L. in Response to Exogenous Application of Methyl Jasmonate and Salicylic Acid. Cornell University, USA.

**Desmond Slade.** Olfactory Communication: Chemical Characterization of the Interdigital Secretion of the Black Wildebeest, *Connochaetes gnou*. University of Stellenbosch, South Africa.



Christopher Dana

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Soo-Hee Kim

**Andrew Swanson.** Functional Genomics of the *Stevia rebaudiana* Diterpene Glycoside Metabolic Pathway. Agriculture Agri-Food Canada.

Xinkun Wang. Variation of Taxol Content in Needles of *Taxus x media* Cultivars with Different Growth Characteristics. Oklahoma State University, USA.

**Yan Zhang.** Detection of Fungal-induced cDNAs from Bermudagrass using Subtractive Hybridization Coupled to cDNA Microarray. Oklahoma State University, USA.



Jennifer Boatright

# IMAGES OF PSNA 2001 in OKLAHOMA CITY



Antoine Bily explains a point to Neil Towers



Darren Peters discusses his work



Mark Bernards, Thomas Vogt and... you see, this is what happens when you don't wear your name tag



The PSNA banquet

Norman Lewis introduces Neil Towers

PSNA NEWS 14 October 2001



Neil Towers accepts his Pergamon Prize

Enjoying the banquet

"Did I ever tell you about the time ... "

Seeing old friends and meeting new ones

Sang-Un Park discusses his research at the Poster Session

Noble Foundation President, Michael Cawley

# PSNA FINANCIAL SUMMARY

2000 PSNA FINANCIAL SUMMARY

| RECEIPTS:                          |            | RECEIPTS:                |            |
|------------------------------------|------------|--------------------------|------------|
| Dues                               | \$3760.00  | Advance from PSNA        | \$2000.00  |
| Plenum/Kluwer (Royalties)          | 4524.77    | Advance from MAPMBS      | 3000.00    |
| Investment Reserve Acct (Interest) | 965.91     | Registration             | 16615.00   |
| Neish Symposium Acct (Interest)    | 1110.96    | Book Orders              | 459.60     |
| Fortis Money Market (Dividend)     | 6.11       | Sponsors                 | 6250.00    |
| Fortis Advantage Acct (Dividend)   | 172.79     | Membership dues          | 90.00      |
| Book sales (RAP orders)            | 2321.00    | Misc (T-shirt sales)     | 310.52     |
| Anonymous Donation (Travel awards) | 100.00     | TOTAL                    | \$28725.12 |
| Elsevier (Page charges)            | 3500.00    |                          |            |
| TOTAL                              | \$16461.54 | EXPENDITURES:            |            |
|                                    |            | Conf Acct Adm Charge     | \$1909.32  |
| EXPENDITURES:                      |            | Bang/meals/refresh       | 6409.39    |
| Executive Committee                |            | Guest Activities         | 175.50     |
| RAP Editor                         | \$4000.00  | Promotions               | 2175.83    |
| Secretary (Newsletter)             | 4000.00    | Travel (Speakers)        | 7437.89    |
| Treasurer (Mailings)               | 1959.23    | Van shuttle              | 689.95     |
| Neish Symposium Speakers           | 1500.00    | Office Supplis/Misc      | 1147.81    |
| PSNA 2000 Travel Awards            | 2050.00    | Dues to PSNA             | 90.00      |
| PSNA 2000 Presentation Awards      | 500.00     | Kluwer                   | 99.60      |
| Elsevier Bulk order, vol 34        | 8343.75    | Bulk book orders         | 3222.00    |
| PSE (share 1st yr royalties)       | 1369.92    | Reg. Refunds             | 155.00     |
| TOTAL                              | \$23722.90 | Refund to PSNA           | 2000.00    |
|                                    |            | Refund to MAPMBS         | 3000.00    |
| ASSETS:                            |            | TOTAL                    | \$28512.29 |
| Checking Acct                      | \$2620.09  |                          |            |
| Business Money Market              | 26951.12   | EXPENDITURES TO PSNA:    |            |
| Neish Symposium acct               | 24571.29   | Meeting Advance          | \$2000.00  |
| Fortis Money Market                | 109.60     | Best Paper/Poster Awards | 500.00     |
| Fortis Advantage                   | 11476.74   | Student Travel Awards    | 2050.00    |
| TOTAL                              | \$65728.74 | Neish Symposium Speakers | 1500.00    |
|                                    | -          | TOTAL                    | \$6050.00  |
|                                    | =          | MEETING REFUND           | -\$2000.00 |
|                                    |            | NET COST TO PSNA         | \$4050.00  |

2000 BELTSVILLE MEETING FINANCIAL REPORT

#### MEMBERSHIP SUMMARY

| COUNTRY | MEMBERS (2000) | MEMBERS (2001) | MEMBER TYPE | TOTAL (2000) | TOTAL (2001) |
|---------|----------------|----------------|-------------|--------------|--------------|
| Canada  | 63             | 52             | Life        | 11           | 11           |
| Mexico  | 14             | 11             | Emeritus    | 16           | 16           |
| USA     | 227            | 206            | Regular     | 336          | 283          |
| Other   | 104            | 88             | Student     | 45           | 47           |
| TOTAL   | 408            | 357            | TOTAL       | 408          | 357          |

RECENT ADVANCES IN PHYTOCHEMISTRY SALES THROUGH PUBLISHER

| VOLUME | MEETING SITE     | EDITORS                |     | 1st VR SALE | 1S  | TOTAL SAL | ES |
|--------|------------------|------------------------|-----|-------------|-----|-----------|----|
| 25     | Delterrille      | Demos                  |     |             |     |           |    |
| 35     | Beitsville       | Rolleo                 |     | n/a         |     | n/a       |    |
| 34     | Montreal         | Romeo/Ibrahim/Varin    |     | 124         |     | n/a       |    |
| 33     | Pullman          | Romeo                  | 224 |             | 378 |           |    |
| 32     | Netherlands      | Romeo/Downum/Verpoorte |     | 330         |     | 401       |    |
| 31     | New Orleans      | Johns/Romeo            |     | 388         |     | 445       |    |
| 30     | Sault Ste. Marie | Romeo                  |     | 313         |     | 440       |    |
| 29     | Mexico City      | Romeo/Arnason/Mata     |     | 344         |     | 661       |    |
| 28     | Asilomar         | Ellis                  |     | 394         |     | 516       |    |
| 27     | Miami            | Romeo/Downum           |     | 398         |     | 505       |    |
| 26     | Fort Collins     | Stafford/Ibrahim       | 416 |             | 552 |           |    |

RECENT ADVANCES IN PHYTOCHEMISTRY SALES - BULK ORDERS

| VOLUME | MEETING SITE | EDITORS             | 1ST YR SALES | TOTAL SALES |
|--------|--------------|---------------------|--------------|-------------|
| 35     | Beltsville   | Romeo               | 38           | n/a         |
| 34     | Montreal     | Romeo/Ibrahim/Varin | 24           | 32          |

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#### **Student Award Winners**

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nearby Salk Institute for Biological Studies. Part of my motivation to join Joe's lab was the opportunity to experiment with protein engineering of natural product enzymes and pathways. Another factor was the opportunity to study the evolutionary history accompanying the functional divergence of enzymes from a unique structural viewpoint. This is one of the most fascinating features of evolution, and I welcomed a chance to increase my understanding of the interplay between primary sequence, tertiary structure, and mechanism. Although becoming a crystallographer was originally only a secondary objective for me, my experience in the lab has taught me the power of this approach for addressing difficult biological and mechanistic questions.

Since joining the lab I have worked primarily on projects related to two enzymes of the phenylpropanoid metabolic pathway in plants, namely 4-coumarate:CoA ligase (4CL) and chalcone synthase(CHS). The work I presented at the PSNA meeting involved the elucidation of the mechanistic basis for intermediate product cyclization specificity within the CHS superfamily of type III polyketide synthases (PKSs). These iterative, homodimeric enzymes are quite distinct from the much larger multi-domain type I and II PKS complexes seen in some fungi and bacteria, which resemble enzyme clusters involved in fatty acid biosynthesis.

Chalcone synthase is common to all higher plants. The crystal structure of alfalfa CHS, previously determined in our lab, has clarified many details of the well-studied CHS mechanism. Of particular interest to me is the evolutionary duplication and divergence of the CHS gene, which has given rise to a superfamily of related polyketide synthases.

One emergent type III family is that of the stilbene synthases (STSs), which share 70-90% amino acid identity with chalcone synthases. STS produces the medicinally relevant anti-fungal compound resveratrol, one of the beneficial components of red wine. CHS and STS both catalyze the sequential addition of three acetate units derived from malonyl-CoA to a p-coumaroyl-CoA starter molecule (which is derived from phenylalanine by upstream enzymes). The growing polyketide intermediate is covalently tethered to an active site cysteine, with the help of two adjacent conserved histidine and asparagine residues. Cyclization of the resulting linear tetraketide in the CHS active site occurs via an intramolecular Claisen condensation, resulting in the covalent linkage of carbon C6 to C1 (numbering from attachment to the active site cysteine). However, in the STS active site this same linear intermediate undergoes an intramolecular aldol condensation, joining C2 and C7.

STS has apparently evolved from CHS on at least three independent occasions, as exemplified by the well-characterized pine, grapevine and peanut STS subfamilies, each of which exhibit more similarity to the CHS sequences of their own species than they do with each other. Comparison of primary structures has failed to reveal an STS consensus sequence. Even 3D homology modeling based on the known structure of CHS, which has been useful in understanding the specificity differences of a number of other divergent CHS-like polyketide synthases, has failed to yield any useful insight into the structural determinants of STS specificity

To resolve this question, we have used a combination of X-ray crystallography, mutagenesis, functional characterization, and protein engineering experiments. This as yet unpublished work has resulted in the elucidation of the structural and mechanistic basis for cyclization

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specificity (aldol versus Claisen condensation) in the type III PKS superfamily. More specifically, we determined the crystal structures of stilbene synthases from two different STS subfamilies (pine and peanut), and compared them to each other and to the structure of CHS from alfalfa. Based on these observations, a functional stilbene synthase was created through mutagenesis of alfalfa CHS. Structures of this engineered STS were determined both with and without resveratrol bound. Further mutagenesis has been directed towards two objectives. First, exploration of the minimal number of mutations needed to convert a CHS to an STS highlighted the specific architectural rearrangement responsible for the mechanistic change, as well as facilitated a better understanding of how STS activity repeatedly evolved from CHS enzymes.

The ability to control type III PKS cyclization specificity is a direct outcome of this work. We are currently incorporating this knowledge into one of our long-term goals, namely the engineering of CHS-like enzymes to catalyze the biosynthesis of a diverse collection of products. Eventually, we would like to put the most interesting of these novel activities back into plants, and study their effects in the context of complex biological systems. Protein crystallography complimented by site directed mutagenesis is indeed a powerful methodology, but critical to the success of this project has been the decades of research which preceded our study, particularly the phytochemical analysis of plants, as well as the cloning and biochemical characterization of the relevant metabolic enzymes. More recently, bioinformatic analysis of the growing sequence database of related enzymes from a multitude of species has greatly facilitated our ability to ask and answer questions that address some of the most interesting mysteries remaining in phytochemistry.

**PSNA NEWS** 

# SOUTH OF THE BOARDER

#### Muy Caliente! The 2002 PSNA Annual Meeting in Mérida, México

As our new President Hector Flores mentions in his Letter, the 2002 PSNA Annual Meeting will be held in Mérida, México hosted by the Centro de Investigación Científica de Yucatán (CICY). Both myself and Víctor Loyola-Vargas, the recently selected President-Elect for 2003, are researchers at CICY. Together with all our CICY colleagues, we are delighted and honored to have the opportunity to receive the phytochemical community in our home town. In anticipation of the arrival of PSNA members and other colleagues to Mérida next August 3 to 7, I would like to provide you with some information on our institute. CICY was created in November of 1979 as part of an effort to decentralize the scientific activities from Mexico City. At that time, the main purpose of CICY was to conduct research on henequen (Agave fourcroydes), the source of the sisal fiber. For a long time (up to the early 1980s) much of the economic activity of the state of Yucatan relied on this plant. However, as the years passed research interests at CICY diversified. Today, even though Agave species, including the tequila plant Agave tequilana, still represent an important theme at CICY, studies on a variety of different plants have developed at our four Research Units that comprise the Center: Biotechnology, Experimental Biology, Natural Resources and Polymeric Materials.

CICY is located at the north sector of Mérida and occupies 6.8 hectares with more than 6000 m<sup>2</sup> of laboratory space. This area is about to expand when the construction of the new building for the Experimental Biology Unit concludes by early next year. Research programs at CICY focus on multidisciplinary studies on important tropical cropspecies, such as banana, coconut, coffee, pepper and tomato. However,

and more directly connected with phytochemistry topics, the Organic Chemistry Group at the Biotechnology Unit, is currently working on the detection, isolation, and purification of biologically active metabolites obtained from the vast local flora. At the Natural Resources Unit, researchers work on several project including the gathering and preservation of medicinal plants at the Regional Botanical Garden. These important conservation tasks are complemented by the collection of ethnobotanical information. From a more biochemical/molecular perspective, some studies are conducted on the regulation of terpenoid indole alkaloid biosynthesis in periwinkle (Catharanthus roseus) and on the synthesis of bixin, an apocarotenoid obtained from the locally important annatto plant (Bixa orellana) and used as a food colorant. Other researchers are interested in understanding the physiological roles of polyamines.

With the diversity of expertise at CICY, there will certainly be excellent opportunities for the exchange of ideas between participants at the PSNA annual meeting and the staff at CICY, which is comprised of more than 50 professors. Most of of the scientists at CICY are Mexican, but several originate from 8 other countries. CICY also offers a graduate program in Plant Sciences and Biotechnology in which more than 40 students are registered. To learn more about CICY



Mayan ruins at Chichén Itzá, near Mérida

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(and new developments on the 2002 PSNA annual meeting) please visit the CICY Web site at www.cicy.mx. However, since there is nothing better than getting a personal experience, please take the next PSNA conference as an opportunity to come south of the border and visit us. We'll be expecting you!

Felipe Vázquez-Flota Unidad de Biología Experimental Centro de Investigación Científica de Yucatán felipe@cicy.mx

# MEETINGS OF INTEREST

#### Plant Biology Canada 2002

Annual Meeting of the Canadian Society of Plant Physiologists

> June 8-12, 2002 Calgary, Alberta, Canada

Contact: Peter Facchini (pfacchin@ucalgary.ca)

#### Gordon Research Conference

Floral Scent: Biology: Chemistry and Evolution

March 3-8, 2002 Ventura, California, USA

Contact: Heidi Dobson (dobsonhe@whitman.edu)

#### 2001 Annual Meeting Review

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the belief that stilbene synthase evolved from chalcone synthase (at least 3 independent times, based on sequence data), Noel set out to try and engineer a chalcone synthase into a stilbene synthase. Guided by the recently elucidated structure for stilbene synthase, rational mutations within the active site of chalcone synthase were made which successfully produced a stilbene synthase. By this method, Noel was able to demonstrate the residues responsible for these enzymes alternate activity. Chloe Zubieta (Salk Institute, California) using similar tools tested the homology models of phenolic Omethyltransferases (OMTs). Aligning the crystal structures of chalcone O-methyl transferase, isoflavone Omethyltransferase, and caffeic acid O-methyltransferase revealed the important residues for substrate specificity and catalysis. Using this structural data, Zubieta demonstrated how it could be used as a predictive tool for other OMTs. Carl Douglas (University of British Columbia, Canada) and collaborators used two different isoforms of Arabidopsis thaliana 4-coumarate Co-A ligase, At4CL1 and At4CL2 which have different substrate specificities. Based on secondary structure predictions, domain swapping experiments revealed regions likely responsible for the difference between specificities. Georg Gross (Universitat Ulm, Germany) presented his group's studies on the biosynthesis and immuno-localization of tannins. Using an affinity-purified antibody to 1,2,3,4,6pentgalloy-lglucose PGG), Gross found the sites of deposition of PGG within the cell. Interestingly, in addition to the cell wall, PGG was also found to localize to chloroplasts. The Keynote speaker for the banquet was Joanne Chory (Salk Institute, California) who described her group's approach to addressing the questions of brassinosteroid metabolism and signaling. Activation tagging was used to find genes involved in brassinosteroid metabolism and/or signaling. A number of genes likely involved in brassinosteroid metabolism were isloated..

Using a simple fluorescent screen, Clint Chapple (Purdue University, Indiana) and co-workers have identified mutants that are deficient in sinapate ester synthesis. The reduction in sinapate esters appeared to have a corresponding increase in 4coumerate esters. Norman Lewis (Washington State University, Washington) described the extensive body of work done in his lab on lignans and polylignans. Using GUS promoter fusions and immunolocalization, appears to reveal that multiple patterns of expression of lignan biosynthetic genes are involved. Dean DellaPenna (Michigan State University, Michigan) introduced us to a new term, "nutritional genomics," referring to using existing sequenced genomes and genes to improve vitamin levels in plants. DellaPenna reported that his group has been able to increase vitamin E levels five times higher than normal levels in Arabidopsis seed. Glucosinolates have important biological activities in plant defense and also play a role in flavour compounds in Brassica crops. Barbara Halkier (Royal Verternary and Agricultural University, Denmark) reported homologous P450s involved in both glucosinolates and cyanogenic glucosides. By expressing CYP79 homologues, the glucosinolate and cyanogenic glucoside levels can be changed, resulting in an increase in the glucosinolates and a corresponding decrease in the cyanogenic glucosides. This result supports the hypothesis of the evolution of glucosinolate metabolism. Susanne Frick (Leibniz Institute of Plant Biochemistry, Germany) described the stable transformation of Papaver somniferum with alkaloid biosynbiosynthetic genes both in a sense and anti-sense arrangement. The goal of the project is not only to examine the regulation and biochemistry of the alkaloid pathways but also to, with alkaloid-free plants, test the chemico-ecological function of the morphinan and benzophenanthridine alkaloids.

Common themes for the future of phytochemistry research could be found in many of the talks, both in the symposium and non-symposium talks. Recently developed technologies that allow for highthroughput analysis of mRNA, protein, and metabolite levels are allowing for a whole new way of doing science. A "snapshot" of the vast array of expressed genes, extant protein, and metabolite levels can now be created for a plant system. Combining genomic, proteomic, and metabolomic data cannot be done using traditional methods; this mass of data must be analyzed in new ways involving computational systems. Moreover, the kind of experiments and, hence, the kinds of questions we can ask change with these technologies. It is encouraging to think that by integrating gene expression, protein level, and metabolite composition data we could better understand the functional role of previously uncharacterized genes. Even in A. thaliana, only a tiny fraction of the estimated 25,000 genes have been functionally characterized, despite the complete sequencing of the genome. As well, the positive identification of a gene will not always be enough to confirm the identity of homologous genes found in other systems, as primary structure is not sufficient to determine function. So for each species or group of organisms, there remains a great deal of work to be done.

David Bird Department of Biological Sciences University of Calgary dabird@ucalgary.ca

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#### Minutes

The 2001 Annual Business Meeting of the Phytochemical Society of North America was held on August 7 at the Westin Hotel in Oklahoma City. Rick Dixon opened the meeting and called on Hector Flores, the new PSNA President, to discuss the organization of next year's meeting. Flores indicated that the PSNA Executive Committee had approved the next meeting of the Society at a location in Mexico. One of the reasons for the support of this decision was the desire to improve the participation of more members from Mexico. The low enrollment of Mexican members in the Society was recognized as an area of weakness and seen to be in immediate need of improvement. Flores suggested that a site near Mexico City, perhaps Cuenavaca, would be an ideal location for the meeting both from the standpoint of attracting foreign participants to an interesting location with good conference facilities, as well as allowing participants from Mexico with the greatest opportunity to attend. It was also suggested that Merida in the Yucatan Peninsula might be another excellent location for the meeting. It is hoped that a meeting in Mexico might also encourage more scientists from South America to attend the conference and join the Society. The precise location of the meeting must be determined shortly for inclusion in the Society Page of the journal Phytochemistry. Norman Lewis suggested that regardless of the final decision for the location of next year's meeting, it was important to sustain the momentum that has been established in the Society over the last few years. He asked for an indication of the success of the 1994 PSNA Annnual Meeting that was held in Mexico City. In a vote, there was strong support to host the 2002 PSNA Annual Meeting in Mexico. Flores defined the tentative topic as "Phytochemistry and Molecular Biology of Medicinal Plants".

Rick Dixon raised the issue of an offer from Mamdouh Abou-Zaid to host the 2003 or 2004 Annual Meeting in Sault Sainte Marie, Ontario. Concerns were raised about whether hosting the meeting in Canada the year after it is held in Mexico, and only 4 years after the last time the last meeting in Canada (1999 at Concordia University in Montréal), was contradictory to the decision made three years ago to host the meeting four times in the United States for every time it is held in both Canada and Mexico. Other concerns were raised about returning to Sault Sainte Marie only 8 years after the meeting was last held in that city in 1995. However, concensus among those in attendance was that the Society should not pass on offers to host the Annual Meeting, especially by those who have demonstrated their capability to stage successful events, and that there is no concern about straying from a strict schedule of meeting locales based on a country-based rotation. However, it was also suggested that perhaps the meeting would be more attractive if it were held in another nearby location. Abou-Zaid said that he could organize the meeting together with colleagues in, or near, Niagara Falls, Ontario. This motion recieved strong support from those in attendance. The 2004 and 2005 Annual Meetings were proposed to be held in Clearwater, Florida (hosted by John Romeo) and Calgary, Alberta (hosted by Peter Facchini). The general themes for the next three meeting were suggested as "Phenolics" (2003), "Terpenoids" (2004), and "Alkaloids" (2005). The was strong support for these topics.

Dixon then discussed the elec-

tion of new members of the PSNA Executive Committee. Victor Loyola-Vargas (Centro de Investigación Cientifica de Yucatán. Mérida. México) was chosen as the President-Elect, and will take over as Society President after the next Annual Meeting in 2002. Charles Cantrell (USDA, ARS. NCAUR) was elected as Treasurer and will take over from Cecelia Macintosh in January 2002. A suggestion from the floor was made to nominate Jonathan Gershenzon (Max Plank Institute for Chemical Ecology, Jena, Germany) as the next President-Elect. This prompted general discussion about the Society's electoral process and raised the question about whether the Society should elect Executive Committee members residing in countries outside of North America. One grim statistic brought up by Susan McCormick was that the last election for both a new President and a new Treasurer saw the return of only 33 ballots. The question was raised about whether this indicates a lack of will or a lack of interest by PSNA members.

Cecelia Macintosh gave the Treasurer's report. She indicated that the total assets of the PSNA were down slightly (\$7,581) mostly because of the Society's commitment to its new publisher to prepurchase for a fixed quantity of the annual proceedings volume prior to its publication each year. The issue of payment methods was raised from the floor with the indication that it was sometimes difficult for those residing outside of the United States to pay thier annual membership dues in U.S. funds. Macintosh indicated that, in the past, the extra expense of a credit card option would not have made it feasible. However, there are currently several

# **STARS AND STRIPES**

# Project O.A.K. (Opportunities Alliance Network): The "Emancipation Oak" - a Living Laboratory

#### **OVERVIEW**

For the last two years, I have made an effort to bring undergraduate students to the PSNA annual meeting. There are several reasons why I chose this Societal meeting over others. Primarily, PSNA meetings are accessible for the students, as well as for me. The organization of the meeting does not lead to information overload, which I have often experienced in much larger societies. In evaluating their experiences for the last two years, students have given me positive feedback. Secondly, and equally important, the students are given the opportunity to engage in dialogue with faculty and graduate students. Attending a meeting where many graduate students present their research, has given my students a type of peer mentoring that they are not able to get directly from me. It allows them to see a different facet of science. I also come back to Phytochemistry because we feel welcomed. We have made friends and established contacts for future collaborations.

Each summer I have insisted that the students put together a presentation for the meeting. For most of the students this is the first time they have every presented any type of research. In order to have them able to put a poster together for the meeting I must turn these student into researchers and, generally, I have ten weeks to work such magic. I realize that the students are not at the same level as graduate students, but for undergraduate students who have never presented their work previously it is an invaluable experience. When I was invited to write this article, I decided to provide PSNA members with an overview of our project. Simply because often it has been a surprise to most people to see undergraduate students from a historically African-American institution participate at the meeting. So below is an overview of "Project O.A.K".

#### INTRODUCTION

On August 6, 1861, the Confiscation Act of the Emancipation Proclamation was signed by President Abraham Lincoln and declared that all slaves of rebellious states were to become chattel of the Union Army [2]. The first of five installations, the **Emancipation Proclamation became** the framework for the Thirteenth Amendment to the constitution, which was passed by Congress on December 18, 1865. This amendment fortified the Emancipation Proclamation and prohibited slavery or involuntary servitude in all territories of the United States. According to several historical accounts, Mrs. Mary Peake read the Emancipation Proclamation to the former slaves in 1863 under the branches of a live oak, which today is known as the "Emancipation Oak" (Figure 1A).

Located on the campus of Hampton University, the Emancipation Oak (Figure 1B) has been designated as a both a State and National Historical Landmark (Figure 2). Be-



Figure 1: Picture of the "Emancipation Oak" taken February 1, 1900 (Courtesy of Peabody Collection, Hampton University, Hampton, VA)

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Figure 1B: The "Emancipation Oak" on February 1, 2000



Figure 3: The "Emancipation Oak" was elevated to the status of a historic landmark in 1974

sides being the place where "all men [were] created equal", it is also recognized as the first site of organized education for black people in America. The tree provided shelter for the first group of contraband children taught by Mrs. Peake. The Emancipation Oak has gained official recognition by the State of Virginia for its importance in education. It is important that we preserve this legacy by having a living monument for future generations. We are using the Emancipation Oak as a living laboratory to teach new generations of college students and to promote plant biology at Hampton University. We are working to "immortalize" the tree by propagating it continuously in culture.

#### SIGNIFICANCE OF THE RESEARCH

In spite of its historical significance the only current efforts to propagate the tree has been the traditional methods of cutting and acorn production. We have not ignored these two methods, in fact we are trying both approaches in the laboratory. However, the main concern is that these methods would not allow for disease resistant seedlings to be produced. Therefore we are implementing somatic embryogensis to culture the seedlings such that we can select a more resistant type of tree.

Because the "Emancipation Oak" holds great importance in American history, our purpose is to use modern micro propagation techniques to assure that authentic offspring of the "Emancipation Oak" will be available for future generations. Presently, we are developing a bilateral approach to establish an undergraduate training and cellular biology research center that will develop and incorporate research with teaching and outreach (Figure 3). This training center works with basic research and addresses the overall objective of propagating seedlings from the "Emancipation Oak" from tissue culture. Although the long-term goal is to use the techniques developed from this research as a tool to detect and localize plant pathogens that affect the Emancipation Oak, this research is important because it is the first step in the development of a diagnostic test for the identification and localization of fungal pathogens.

#### THE TREE

The "Emancipation Oak" (*Quercus virginiana*) is a member of the genus Quercus in the beech family Fagaceae [3]. *Q. virginiana* is also known as a live oak or a Virginia live oak. This species is native to the southeastern United States and northern parts of South America in USDA hardiness zones 8 to 11 (Figure 4) [11]. Its horizontal branches, leathery, oval and dark-green leaves (Figure 5) can easily identify it. It is often classified separately from other oaks because it is an evergreen. The acorns are one-third covered by a deep cup.

Quercus virginiana are longlived trees, with a life span of up to 2 to 3 centuries. The exact age of the "Emancipation Oak" is not known; however, in April 1998 the tree measured 16 feet and 8 inches in girth [8]. Because (as shown above in Figure 1) the tree was already of substantial size on February 1, 1900, we estimate its age in excess of 136 years old.

#### DISEASE SIGNS AND SYMPTOMS

Trees in this genus can be propagated easily from acorns and grow well in

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Figure 3: Project O.A.K. participants, June - August 2000



Figure 4. The natural range of *Quercus virginiana* Mill.

rich, moderately moist soil or dry, sandy soil [5]. Oaks are hardy and long-lived but are not shade-tolerant and are highly susceptible to injury by leaf-eating organisms or oak wilt fungus. Oak leaf blisters, caused by the fungus Taphrina caerulescens, is a common disease affecting up to fifty species of oaks around the world [6,7,8]. Both red and live oak trees are particularly susceptible to infection, whereas white oaks are rarely infected. Most species of Taphrina are spread by dispersal of spores that are blown or splashed to newly developing tissue. These fungi have four unusual features that are used to classify them [9]. First, the assimilative mycelium is dikaryotic, which immediately distinguishes it from most other ascomycetes. Second, it produces an exposed layer of asci on the surface of the host leaf. Since there is no surrounding or supporting fungal tissue, there is nothing that can be called an ascoma. Third, the ascospores often bud in a yeast-like manner while still inside the ascus. Fourth, the asci dehisce by splitting across the tip, rather than around the bottom.

Taphrina is most often associated with peach leaf curl disease, which causes leaves to become thickened and distorted. Peach leaf curl disease is caused by Taphrina deformans and is one of the most common and widespread diseases affecting



Figure 5. Leaves from the "Emancipation Oak", *Quercus virginiana*.

peach plantings in the United States [10]. As the name implies, the most common and striking symptom of leaf curl occurs on the foliage. The fungus causes the meristematic cells at leaf margins to proliferate quickly and randomly, which results in the leaves becoming variously wrinkled, puckered and curled. Taphrina *caerulescens* is the species we have isolated from the leaves, which were showing disease symptoms. Although we have not been able to use Koch's postulate to show unambiguously that this is the cause of disease symptoms, its appearance is consistent with the symptoms.

As has been noted in the literature, *T. caerulescens* is more of an aesthetic nuisance than it's close relative *T. deformans* [9]. Leaf curl is readily controlled. There are no nonchemical controls, which have proved



Figure 6. Isolated fungus grown on oatmeal (A) and Difco PDA (B) agar.

effective (e.g. sanitation, resistance). However, effective control can be achieved with one or two fungicide treatments. Effective fungicides are chlorothalonil (Bravo and Daconil), fixed coppers (Kocide 101, Top-Cop Tribasic, Liquid Copper Fungicide) and ferbam (Carbamate WDG) [9].

#### FUTURE RESEARCH

We have been able to isolate the fungus on PDA and cornmeal agar (Figure 6). Lesions from sterilized leaves were grown first on water agar for five days. After five days blocks of agar were transferred under sterile conditions to various agar plates. Ascospores suspensions, which were obtained by flooding plates with sterile deionized water, can be isolated (Figure 7). These spores will be used in detection and localization microscope studies during Phase II of the project (Summer 2002).

Project O.A.K. (Opportunities Alliance networK) is a multiyear project that has been designed to establish an aggressive training and plant research center that has incorporated teaching and research at a historically black college, Hampton University. Although the project has focused on establishing a training center, a residual but beneficial side effect has been the use of Plant Pathology to address both a historical and biological significant purpose. As has been addressed in the review by Kaufmann and Linder [11], in order to meet the challenges of assuring sustainability of production and increasing demands for natural resources, there must be balanced and new approaches taken to tree physiology. In this paper, we have shown the effectiveness of a cross-disciplinary approach to introduce students to the complexity of tree physiology research, both to attract new students into the discipline and heighten awareness of natural resources issues. Students will be developing models to hypothesize on the effect of new combinations of factors likely to af-

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Figure 7. Isolated fungal ascospores

fect tree and fungal behavior. We will have antibodies produced toward the ascospores, such that we can do immunodetection studies. In addition to immunodetection, future plans include the use of global positioning data to track changes in atmospheric conditions and study how this data correlates to changes in Taphrina's life cycle. Our work allows students to participate in cutting-edge natural resources research. In addition to focusing on global climate changes (e.g., increase in atmospheric CO<sub>2</sub> and related climate changes on the growth and development of potential disease pathogens), participants will continue to learn molecular biology as well (e.g., RFLP and RAPDs).

Project O.A.K. is helping to add glamour to Plant Pathology, Physiology and Forestry. We are working to get plant science more deeply entrenched in the psyche of students. We are aware of the importance of our monumental ability to work on a tree of such historical significance, but we are clear on the possibility of introducing plant biology to future generations of scientists.

#### ACKNOWLEDGEMENT

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Donald Lyons and James Wise for their editorial comments and helpful suggestions.

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#### **Annual Business Meeting**

#### continued from page 20

internet-based credit card options that could work. The motion to explore these possibilities and implement a credit card option for the payment of annual membership dues was passed. The decision to raise membership dues last year from \$20 US to \$40 U.S. has apparently not had any effect on Society membership. Information contained in the Financial Report Summary (included in this issue of the Newsletter) was also discussed in detail. It was noted that this year's meeting saw a dramatic and welcomed increase in the number of students attending the meeting and requesting travel awards.

Peter Facchini presented the Secretary and Newletter Editor's report. He acknowledged the large number of positive comments about

the improvement in the quality of the Newsletter, but indicated that each issue is a monumental struggle to find PSNA members willing to contribute articles. One solution was to request that PSNA Advisors function in the writing or solicitation of at least one Newsletter article per year. This motion passed. Facchini also suggested that Arthur Neish Young Investigator Minisymposium speakers and winners of the Best Student Paper Awards be required to submit short autobiographical sketches for the Newsletter. This motion also passed. The Society's new Web site, www.psnaonline.org was promoted. It was noted that our Society is in competition for the acronym "PSNA" with the Pennsylvannia State Nurses Association and the Plastic Surgeons of

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Northern Arizona (among others); thus, the domain name "psna-online" was selected. A final suggestion was the creation of a student-based editorial board, which was agreed on as a good idea in principle and will be pursued in due course.

> Peter Facchini University of Calgary Calgary, Alberta, Canada pfacchin@ucalgary.ca



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