Proceedings of the Sixteenth Conference of the International Organization of Citrus Virologists

Edited by M. E. Hilf • N. Duran-Vila • M. A. Rocha-Peña

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DEDICATIONS

David Gumpf



David Gumpf died unexpectedly on February 6, 2003, while driving home from work. His death is a great loss not only for his family, but for his numerous friends and colleagues worldwide, and for citrus pathology and citriculture.

After graduating from the University of Montana with BS (1964) MS (1966) Dave and degrees, obtained his PhD from Nebraska, and then joined the University of California in 1970. There he became distinguished member of the а Department of Plant Pathology and contributed extensively to advances in knowledge of citrus pathogens and their detection. He was active in numerous state, national and international research projects, including

BARD, and US-Spain cooperative projects on citrus virus characterization and identification. He was a major contributor to the development of serological detection procedures for *Citrus tristeza virus* (CTV) and the characterization of an international collection of CTV isolates at Beltsville, MD. Dave also served on variety of technical committees, taught graduate courses in plant pathology at UCR, and served as advisor and mentor to numerous graduate students.

In addition to his academic contributions, Dave served since 1979 as the Director of the Citrus Clonal Protection Program, a world recognized program for the development and maintenance of pathogen-free citrus bud wood. This program is supported by the University of California, the California Citrus Industry, the California Department of Food and Agriculture and the US Department of Agriculture, and benefited enormously from Dave's expertise in citrus pathogen detection, his interest and knowledge of citriculture and nursery operations, and his administrative skills. In recognition of his service he was named The Researcher of the Year by the California Nurserymen's Association in 2001, and received the Albert G. Salter Memorial Award from the California Citrus Quality Council in 2002.

Dave was a long standing member of IOCV and served as its treasurer for many years. His effective management of IOCV finances has contributed to the long range financial stability of the organization.

Listing Dave's accomplishments and contributions does not really convey the impacts he made in the lives and careers of his friends and associates. Those fortunate enough to have worked with Dave will always remember his ability to combine excellent science with an infectious enjoyment of life. His deft touch for recognizing and sharing the humorous side of life made long days and hard work pass quickly, and his visits, letters and phone calls were sure to bring smiles. He was a compassionate friend willing to help others even while dealing quietly and courageously with chronic serious health problems that persisted through most of his career.

The "Big Guy" is gone, and while grieving is certainly natural, sadness should not be his final legacy. We should savor the many good memories we have, and in tribute, follow his example of zest for science and life.

Dave is survived by his wife Jan, his brother Ralph E. Gumpf, Jr., his daughter Ann, her husband Todd and granddaughter Mallory; his son John, his wife Deanna and grandson Brady.

Steve Garnsey

Monique Garnier-Semancik (1949-2003)



Monique passed away on Saturday, May 10, 2003, at the age of 54, from a septic shock following a simple kidney stone problem, which had not received appropriate attention.

She was born on April 11, 1949, Poncharrat sur Breda, near at Chambery in the French Alps, the mountains which she liked so much; "her" mountains, as she used to say. In her early childhood, the family moved to the beautiful little town of Monflanguin, some 150 km east of Bordeaux. After her years at high school at Villeneuve sur Lot, she entered the University of Bordeaux 2, and graduated in Biology in 1971. She chose to work for her PhD in my laboratory on the replication of turnip yellow mosaic virus RNA. She

approached this problem by electron microscopy, a technique which she was going to use more and more to detect and characterize citrus pathogens. In this way, the huanglongbing (HLB) agent was shown to be a Gram negative bacterium, the lime witches' broom organism to be a phytoplasma, and *Xylella fastidiosa* to be responsible for Citrus Variegated Chlorosis (CVC). The work on CVC was the beginning of a long-lasting collaboration with Fundecitrus.

In the 1980s, claims that the HLB bacterium had been cultured made the front page. In spite of intensive efforts, Monique couldn't confirm any of these claims, and maintained that the cultures obtained were contaminants. Even though, at times, she was discouraged (as for instance at the IOCV meeting in Valencia in 1986), she never gave up, and, even today, the HLB agent remains uncultured. Monique was convinced however, that one day, the bacterium would be available in culture.

When the monoclonal antibody (MA) technology became available, Monique immediately saw the possibility of applying it to the detection of pathogens not available in culture, such as the HLB bacterium, against which 13 MAs were obtained. Unfortunately, it was eventually found that the MAs against the HLB bacterium were very specific, and none detected all the HLB strains tested. This is why we have often pointed out that MAs against the HLB pathogen could not be used for general detection purposes. Fortunately, molecular techniques became available in the meantime, and proved very fruitful for the phylogenetic and taxonomical characterizations of the non cultured citrus pathogens. For instance, the HLB agent, identified as a member of the alpha-Proteobacteriae, became soon known as *Candidatus* Liberibacter asiaticus in Asia, and *Candidatus* Liberibacter africanus in Africa. The third species, *Candidatus* Liberibacter americanus, was discovered in 2004, just one year too late for Monique to be involved.

One of Monique's clever ideas was to control phytoplasma diseases by transgenic plants expressing antibodies (plantibodies) directed against specific agents. Control of the Stolbur phytoplasma in transgenic tobacco plants was temporarily obtained, but was hindered by the difficulty of targeting the plantibodies to the sieve tubes, i.e., the site in which the phytoplasmas are localized.

Monique became also interested in host/pathogen interactions. While *Spiroplasma citri*, as a cultured organism, lent itself to molecular genetics and mutagenesis to study the genes involved in pathogenicity and insect transmission, another approach had to be used with the uncultured phytoplasmas: identification of plant genes that were differentially expressed following infection by phytoplasmas or even spiroplasmas. Indeed, such genes have been found, some are up regulated, others are down regulated. This work has become a major project in the laboratory.

The sequencing and annotation of the *Spiroplasma citri* genome was one of the most ambitious projects of the laboratory. The project is completed today, but unfortunately Monique missed the end of it.

Monique became a permanent employee of INRA in 1975 when she was appointed Research Assistant in my laboratory. In 1994, on my proposal, she became head of the laboratory. In 2001, she reached the highest scientific position at INRA, that of Research Director. She and I have been colleagues and friends for thirty years. We were not only complementary, but also synergistic. I passed over to her my conviction that both the laboratory and the orchard were equally important, a notion that seems to get lost today. I was happy to share with her my experience in citrus. She was a lively person, full of enthusiasm, always ready to go somewhere, even to Mongolia and China by train, to do something new, to take advantage of the newest techniques possible. She had a special gift to do things fast and short. But she was also a hard worker. Her administrative duties were heavy. She had planned to go back to the bench in 2004, at the end of these duties.

Monique was a faithful member of the IOCV. She participated in the last six conferences, starting in Valencia in 1986. With her, two of our papers received twice the J. Merrill and Adeline Wallace award, the first time in 1989 at the 11th IOCV conference in Orlando, for the study of the greening organism with monoclonal antibodies, and the second time in 1992 at the 12th conference in New Delhi for Koch's postulates on CVC. She authored over 50 papers in international journals, without being reluctant to have full papers published in the proceedings of the IOCV conferences.

We are all very much affected by Monique's departure. It is a small consolation to know that she passed away without realizing that she would never wake up again. She was married to Joe Semancik on April 5, 2003, in Bègles, near Bordeaux, and her last weeks down on Earth were undoubtedly the most beautiful ones in her life.

Josy Bové

PREFACE

The 16th Conference of the International Organization of Citrus Virologists began with a pre-conference tour of the Lower Rio Grande Valley of south Texas November 3-6, 2004. Twenty-four delegates were welcomed at an evening reception hosted by Dr. Ronald Rosati, Dean of the College of Agriculture & Human Sciences, Texas A & M University-Kingsville (TAMUK). After a morning of presentations on various aspects of Texas citriculture at the TAMUK Citrus Center, delegates toured the research laboratories and orchards. For dinner, a trip to the vacation resort of South Padre Island was undertaken.

On the last day, delegates toured Mission Shippers (a packer/shipper), the TexSun juice plant, and the Rio Grande Wholesale Nursery. The more technical stops included the Texas A&M Kingsville Citrus Center in Weslaco and its associated field stations. Pre-congress participants were able to see the variety collection, trees associated with the Texas Budwood Program, and some transgenic citrus. A USDA-APHIS facility in Moore Field near Mission was also visited, with demonstrations of fruit fly research and the APHIS mobile laboratory. In the evening the delegates were treated to a reception at the house of Dr. Mani Skaria. The following morning, the participants departed by luxury bus for Monterrey, Mexico, with a stop at the Centro de Biotecnologia Genomica in Reynosa, where they were hosted by Dr. Alberto Mendoza.

The main meeting of the 16th Conference of the International Organization of Citrus Virologists was held in Monterrey, Mexico, November 7-12, 2004, at the Hotel Fiesta Americana. The welcome reception on the first evening included an exquisite Mexican buffet dinner, followed by a wonderful show of Mexican singing and dancing, which presented a delightful retrospective of Mexican culture, starting with the Azteca culture from the pre-Columbian era, passing through the struggle for Independence in the 18th century, the Revolution of 1910 and ending with contemporary times.

A total of 87 delegates from 22 countries attended. At the opening ceremony on November 8, delegates were welcomed to Mexico by the Chairman of the Organizing Committee, Dr. Mario A. Rocha-Peña. Delegates were then addressed by Dr. Sebastian Acosta-Núñez, Director of the Department of Planning and Development, acting on behalf of Dr. Pedro Brajcich, the General Director of INIFAP. The Opening Address was given by Dr. Fermin Montes-Cavazos, Director of Nuevo Leon's Department of Agriculture speaking on behalf of Lic. Natividad González-Parás, Governor of the State of Nuevo Leon. The IOCV Chairman, Pedro Moreno, also addressed delegates. Day 1 and Day 2 of the scientific program were devoted to *Citrus tristeza virus* (CTV) and Citrus Sudden Death (CSD), with invited presentations from Dr. Bill Dawson (CTV) and Dr. Josy Bové (CSD). On Day 1, 12 oral presentations on CTV were made, with an evening poster session. On Day 2, a further 17 CTV papers and four CSD papers were presented, followed by the delegates viewing posters which presented more interesting research on these diseases.

On Day 3, delegates participated in a field tour to the citrus production areas of the state of Nuevo Leon. The first stop was devoted to the citrus nursery "Fondo de Aseguramineto Agrícola" in the municipality of Santiago, where delegates viewed all the management and propagation of seedlings and of scions grafted on CTV tolerant rootstocks.

The second stop was in the municipality of Allende, where delegates visited the "Saldivar Reciclados" factory which is devoted to making plastic packing boxes by recycling discarded plastic. The tour then continued with a visit to the "Distribuidora Mexicana de Cítricos", packinghouse, where the delegates witnessed all the processes involved with packing citrus fruits. During the visit the delegates enjoyed taking pictures of one another and delighted in the wonderful flavors of the diverse mandarin, grapefruit and oranges varieties.

The third stop included a visit to a citrus nursery in the municipality of Montemorelos devoted to the propagation of citrus plants on sour orange rootstock. Here the delegates viewed the beneficial effect of using mycorrhizal fungi on the growth of nursery plants.

Following this stop, delegates visited a commercial citrus grove planted with several orange and grapefruit varieties on CTV tolerant rootstocks. Here the IOCV delegates enjoyed a delicious lunch provided by Promotora Citricola del Golfo.

This informative field tour ended with a visit to a local handicrafts market at Santiago.

The conference sessions resumed on Day 4, with papers and posters on viroids, Huanglongbing, blight and stubborn. At the Business Meeting held in the afternoon of Day 4, IOCV's first three Fellows were elected, Josy Bové, Steve Garnsey and Chet Roistacher. The incoming chairman, John da Graça, announced that the new treasurer would be Robert Krueger, and he thanked the Secretary, Chet Roistacher for acting as Treasurer following the sudden death of Dave Gumpf. A minute of silence was observed in memory of lost friends (Monique Garnier, Dave Gumpf and Mort Cohen). It was decided to dedicate the 16th Proceedings to both Monique Garnier and Dave Gumpf. Other decisions made were to accept the generous offer of IVIA to set up, at no cost to IOCV, an IOCV webpage, and to move towards publication of the proceedings in an electronic form only starting with the 17th conference. A new award for the best paper on disease diagnosis and detection was created and was named the Gumpf Award. This was created with a donation from Mrs. Janice Gumpf.

Day 5 was taken up with presentations and posters describing research on several additional diseases, as well as survey methodologies and certification programs. A total of 132 papers and posters were presented during the conference.

The farewell banquet was held at the Hotel Fiesta Americana on the evening of the last day of the conference. The IOCV delegates enjoyed a delicious dinner accompanied by an excellent jazz band. The results of the election for the Chair of IOCV were then announced: Nuria Duran-Vila was named Chairperson-elect, and the Wallace Award for the best published paper was presented to M. Gandia and N. Duran-Vila for their paper from the 15th IOCV Conference titled "Effect of sequence variation on the biological properties of *Citrus exocortis viroid*".

Following the conference, 19 delegates flew from Monterrey to Veracruz to take part in a post-conference tour of the citrus areas of Veracruz state, a trip organized by Sergio Curti-Díaz. After some initial sight-seeing in the city of Veracruz, the participants were transported to the main citrus producing area around Martínez de la Torre.

This area is about 3 hours by bus from Veracruz and is slightly elevated. Most of the citrus production in this area is Persian lime with some Valencias and other types. Participants were given a view of the INIFAP field station at Ixtacuaco, which is establishing indexing facilities with the goal of providing clean stock materials to the local industry. On the second day, Sunday, the IOCV delegates played tourists and visited the Zona Arqueológica El Tajín, which is the most important in this part of México, as well as the city of Papantla, home of the famous Voladores.

On Monday, various producing citrus orchards were visited. These included the Persian lime orchard of Mr. Alemán, in which woodpocket disease was present. The group also visited the Huerta Huasteca, an orchard of Persian lime on Alemow in which cachexia was quite evident; and a visit was made to the Salas Valencia block, which showed some trees with Amachamiento, a local disorder for which no causal agent has yet been identified.

On the final day, several industry-related sites were visited, including the PROCIGO nursery, one of the first to participate in the Mexican government's certification program. In addition, the group was given excellent tours of a large modern packing plant of Grupo Exportadora B&S and a CITREX juice processing facility. The day and the tour ended with a pleasant dinner and walking tour of Martínez de la Torre, followed by a departure brindis at Ixtacuaco.

All full-length papers were reviewed by two referees. We express our thanks to the following who served as referees: S. Adkins & M. Irey (USDA, ARS, Fort Pierce, FL, USA), J. P. Agostini (INTA, Montecarlo, Misiones, Argentina), M.J.Asíns, M. Cambra, J. Guerra, P. Moreno, L. Peña, & L. Rubio (IVIA, Moncada, Spain), M. A. Ayllón (Universidad Politécnica de Madrid, Madrid, Spain), M. Bar-Joseph (S. Tolkowsky Laboratory, Volcani Center, Bet Dagan, Israel), P. Barkley (P.O. Box 46, Mulgoa, NSW 2745, Australia), C. Bock (Univ. of Florida, Fort Pierce, FL, USA), J. M. Bové (INRA & Univ. of Bordeaux, Bordeaux, France), J. V. da Graca & T. E. Mirkov (Texas A&M Univ., Weslaco, TX, USA), S. Dallot (UMR BGPI-INRA, Montpellier, France) J. A. Darós & R. Flores (CSIC-Universidad Politécnica de Valencia, Valencia, Spain), W. O. Dawson, A. Folimonov, S. Folimonova, S. M. Garnsey, T. Satyanarayana & L. W. Timmer (Univ. of Florida, Lake Alfred, Florida, USA), J. A. Dodds & J. S. Semancik (Univ. of California, Riverside, CA, USA), V. J. Febres (University of Florida, Gainesville, Florida, USA), J. Freitas-Astua (Embrapa, Centro APTA Citros, Cordeirópolis, SP, Brazil), G. A. Frías-Treviño (Corporativo Agropecuario de Nuevo León, Monterrey, Nuevo León, México), J. Hartung & R. Owens (USDA, ARS, Beltsville, MD, USA), R. F. Lee (USDA, ARS, NGR, Riverside, CA, USA), D. Lewandowski (Ohio State Univ., Columbus, OH, USA), A.V. Karasev (Univ. of Idaho, Moscow, Idaho, USA), R. Milne (Istituto di Fitovirologia Applicata, Torino, Italy), G. W. Müller (Universidade Estadual de Maringá, Paraná, Brazil), G. Pietersen (Univ. of Pretoria, Pretoria, South Africa), M. Polek (CCTEA, Tulare, CA, USA), C. Powell (Univ. of Florida, Fort Pierce, FL, USA), R. F. Rivera-Bustamante (CINVESTAV, Irapuato, Guanajuato,

México), P. Sieburth (Bureau of Citrus Budwood Registration, Winter Haven, FL, USA), R. A. Valverde (Louisiana State University, Baton Rouge, Louisiana, USA), S. P. van Vuuren. (Citrus Research International, Nelspruit, South Africa) C. Vernière.(CIRAD, La Réunion, France), W. Wintermantel (USDA, ARS, Salinas, CA, USA), and R. K. Yokomi (USDA, ARS, Parlier, CA, USA).

For virus and viroid nomenclature, we followed Rule 3.40 of the International Code of Virus Classification and Nomenclature published in the 8th Report of the International Committee on Taxonomy of Viruses (Academic Press, 2005). Species names are printed in italics and have the first letter of the first word capitalized. When the taxonomic status is uncertain, or the position within a genus is not clarified, it is considered a tentative species and its name is not italicized, although the initial letter is capitalized. Current approves species are *Citrus bent leaf viroid*, *Citrus exocortis viroid*, *Citrus viroid III*, *Citrus viroid IV*, *Citrus leaf rugose virus*, *Citrus yellow mosaic virus*, *Citrus psorosis virus*, *Citrus tristreza virus*, *Citrus leaf blotch virus*, *Satsuma dwarf virus* and *Citrus variegation virus*. Citrus cachexia viroid = Hop stunt viroid, and Citrus tatter leaf virus = Apple stem grooving virus. Citrus vein enation virus, Citrus Indian ringspot virus have not been officially assigned to a family or genus, but we have published their names in the same way as for tentative species.

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> M.E. Hilf N. Duran-Vila M. A. Rocha-Peña (Editors)

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