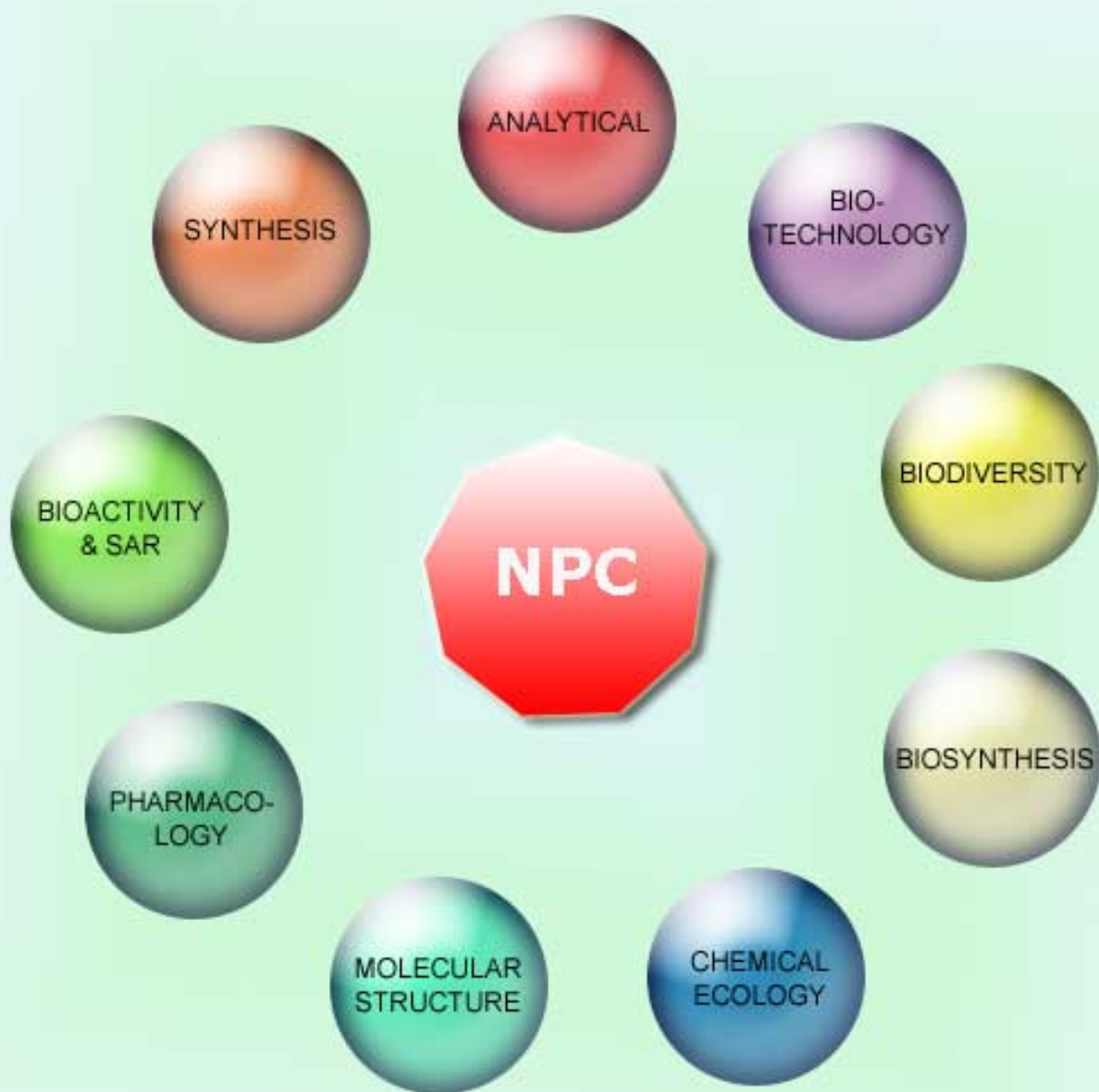


NATURAL PRODUCT COMMUNICATIONS

An International Journal for Communications and Reviews Covering all
Aspects of Natural Products Research



Volume 5. Issue 3. Pages 351-506. 2010
ISSN 1934-578X (printed); ISSN 1555-9475 (online)
www.naturalproduct.us

EDITOR-IN-CHIEF**DR. PAWAN K AGRAWAL**

*Natural Product Inc.
7963, Anderson Park Lane,
Westerville, Ohio 43081, USA
agrawal@naturalproduct.us*

EDITORS**PROFESSOR ALESSANDRA BRACA**

*Dipartimento di Chimica Bioorganica e Biofarmacia,
Università di Pisa,
via Bonanno 33, 56126 Pisa, Italy
braca@farm.unipi.it*

PROFESSOR DEAN GUO

*State Key Laboratory of Natural and Biomimetic Drugs,
School of Pharmaceutical Sciences,
Peking University,
Beijing 100083, China
gda5958@163.com*

PROFESSOR J. ALBERTO MARCO

*Departamento de Química Orgánica,
Universidade de Valencia,
E-46100 Burjassot, Valencia, Spain
alberto.marco@uv.es*

PROFESSOR YOSHIHIRO MIMAKI

*School of Pharmacy,
Tokyo University of Pharmacy and Life Sciences,
Horinouchi 1432-1, Hachioji, Tokyo 192-0392, Japan
mimakiy@ps.toyaku.ac.jp*

PROFESSOR STEPHEN G. PYNE

*Department of Chemistry
University of Wollongong
Wollongong, New South Wales, 2522, Australia
spyne@uow.edu.au*

PROFESSOR MANFRED G. REINECKE

*Department of Chemistry,
Texas Christian University,
Forts Worth, TX 76129, USA
m.reinecke@tcu.edu*

PROFESSOR WILLIAM N. SETZER

*Department of Chemistry
The University of Alabama in Huntsville
Huntsville, AL 35809, USA
wsetzer@chemistry.uah.edu*

PROFESSOR YASUHIRO TEZUKA

*Institute of Natural Medicine
Institute of Natural Medicine, University of Toyama,
2630-Sugitani, Toyama 930-0194, Japan
tezuka@inm.u-toyama.ac.jp*

PROFESSOR DAVID E. THURSTON

*Department of Pharmaceutical and Biological Chemistry,
The School of Pharmacy,
University of London, 29-39 Brunswick Square,
London WC1N 1AX, UK
david.thurston@pharmacy.ac.uk*

HONORARY EDITOR**PROFESSOR GERALD BLUNDEN**

*The School of Pharmacy & Biomedical Sciences,
University of Portsmouth,
Portsmouth, PO1 2DT U.K.
axuf64@dsl.pipex.com*

ADVISORY BOARD

Prof. Berhanu M. Abegaz
Gaborone, Botswana

Prof. Viqar Uddin Ahmad
Karachi, Pakistan

Prof. Øyvind M. Andersen
Bergen, Norway

Prof. Giovanni Appendino
Novara, Italy

Prof. Yoshinori Asakawa
Tokushima, Japan

Prof. Lee Banting
Portsmouth, U.K.

Prof. Julie Banerji
Kolkata, India

Prof. Anna R. Bilia
Florence, Italy

Prof. Maurizio Bruno
Palermo, Italy

Prof. Josep Coll
Barcelona, Spain

Prof. Geoffrey Cordell
Chicago, IL, USA

Prof. Cristina Gracia-Viguera
Murcia, Spain

Prof. Duvvuru Gunasekar
Tirupati, India

Prof. A.A. Leslie Gunatilaka
Tucson, AZ, USA

Prof. Kurt Hostettmann
Lausanne, Switzerland

Prof. Martin A. Iglesias Arteaga
Mexico, D. F., Mexico

Prof. Jerzy Jaroszewski
Copenhagen, Denmark

Prof. Leopold Jirovetz
Vienna, Austria

Prof. Teodoro Kaufman
Rosario, Argentina

Prof. Norbert De Kimpe
Gent, Belgium

Prof. Karsten Krohn
Paderborn, Germany

Prof. Hartmut Laatsch
Gottingen, Germany

Prof. Marie Lacaille-Dubois
Dijon, France

Prof. Shoei-Sheng Lee
Taipei, Taiwan

Prof. Francisco Macias
Cadiz, Spain

Prof. Imre Mathe
Szeged, Hungary

Prof. Joseph Michael
Johannesburg, South Africa

Prof. Ermino Murano
Trieste, Italy

Prof. M. Soledade C. Pedras
Saskatoon, Canada

Prof. Luc Pieters
Antwerp, Belgium

Prof. Om Prakash
Manhattan, KS, USA

Prof. Peter Proksch
Düsseldorf, Germany

Prof. Phila Rahaevelomanana
Tahiti, French Polynesia

Prof. Satyajit Sarker
Wolverhampton, UK

Prof. Monique Simmonds
Richmond, UK

Prof. Valentin Stonik
Vladivostok, Russia

Prof. Winston F. Tinto
Barbados, West Indies

Prof. Karen Valant-Vetschera
Vienna, Austria

Prof. Peter G. Waterman
Lismore, Australia

INFORMATION FOR AUTHORS

Full details of how to submit a manuscript for publication in Natural Product Communications are given in Information for Authors on our Web site <http://www.naturalproduct.us>.

Authors may reproduce/republish portions of their published contribution without seeking permission from NPC, provided that any such republication is accompanied by an acknowledgment (original citation)-Reproduced by permission of Natural Product Communications. Any unauthorized reproduction, transmission or storage may result in either civil or criminal liability.

The publication of each of the articles contained herein is protected by copyright. Except as allowed under national "fair use" laws, copying is not permitted by any means or for any purpose, such as for distribution to any third party (whether by sale, loan, gift, or otherwise); as agent (express or implied) of any third party; for purposes of advertising or promotion; or to create collective or derivative works. Such permission requests, or other inquiries, should be addressed to the Natural Product Inc. (NPI). A photocopy license is available from the NPI for institutional subscribers that need to make multiple copies of single articles for internal study or research purposes.

To Subscribe: Natural Product Communications is a journal published monthly. 2010 subscription price: US\$1,695 (Print, ISSN# 1934-578X); US\$1,695 (Web edition, ISSN# 1555-9475); US\$2,095 (Print + single site online); US\$595 (Personal online). Orders should be addressed to Subscription Department, Natural Product Communications, Natural Product Inc., 7963 Anderson Park Lane, Westerville, Ohio 43081, USA. Subscriptions are renewed on an annual basis. Claims for nonreceipt of issues will be honored if made within three months of publication of the issue. All issues are dispatched by airmail throughout the world, excluding the USA and Canada.

Insecticidal, Mutagenic and Genotoxic Evaluation of Annonaceous Acetogenins

Olga Álvarez Colom^a, Analía Salvatore^b, Eduardo Willink^b, Roxana Ordóñez^{a,c}, María I. Isla^{a,c}, Adriana Neske^{a*} and Alicia Bardón^{a,c}

^aFacultad de Bioquímica, Química y Farmacia, Universidad Nacional de Tucumán, Ayacucho 471, Tucumán (4000), Argentina

^bEstación Experimental Agroindustrial Obispo Colombres, Av. William Cross 3150, Las Talitas, Tucumán (4101), Argentina

^cINQUINOA – CONICET, Ayacucho 471, Tucumán (4000), Argentina

aneske@fbqf.unt.edu.ar

Received: October 27th, 2009; Accepted: December 22nd, 2009

Annonaceous acetogenins represent a class of bioactive compounds whose primary mode of action is the inhibition of NADH-ubiquinone oxidoreductase (Mitochondrial Complex I). Given the potential pesticidal use of these compounds, we evaluated the effects of seven acetogenins: squamocin (**1**), molvizarin (**2**), itrabin (**3**), almuñequin (**4**), cherimolin-1 (**5**), cherimolin-2 (**6**), and tucumanin (**7**) isolated from *Annona cherimolia* Mill. against *Ceratitis capitata* Wiedemann (Tephritidae). These acetogenins did not display insecticidal action at 250 µg of treatment per g of adult diet. However, the oviposition capacity of *C. capitata* females was significantly altered by some of the acetogenins at this concentration. The most potent compounds were itrabin, molvizarin and squamocin. Moreover, significant differences were detected in the preference of oviposition sites when itrabin and squamocin were spread on the surface of artificial fruits at doses of 30 µg/cm². Additionally, we investigated the mutagenic effects displayed by itrabin, as well as the phytotoxic and genotoxic action of squamocin and itrabin. Both compounds displayed slight phytotoxic and genotoxic effects on roots of *Allium cepa* at 2.5 µg/mL though no mutagenic effects were detected at 0.25, 0.5 and 2.5 µg/mL on *Salmonella typhimurium* strains TA98 and TA100.

Keywords: *Ceratitis capitata*, plant metabolites, *Salmonella typhimurium*, *Allium cepa*.

The Annonaceae, a large family of tropical plants, has been intensely investigated over the last 20 years, largely because of the discovery of the annonaceous acetogenins (ACG), a group of C32/C34 fatty-acid derived natural products. Most of the acetogenins are compounds possessing 1-3 THF rings, a γ-lactone (either saturated or unsaturated), and a long unbranched aliphatic chain. Currently, these natural products are regarded as the most potent antitumor compounds of recent years [1], acting as potent inhibitors of complex I (NADH: ubiquinone-oxidoreductase) in the mammalian mitochondrial electron transport system [2a].

Regarding the insecticidal action of acetogenins, it has been reported that many of these compounds are toxic to *Leptinotarsa decemlineata* (Coleoptera) and *Myzus persicae* (Homoptera) adults [2b], as well as to nymphs of the German cockroach [1]. Previous results from our laboratory indicated that the acetogenins

from *Annona cherimolia* and *A. montana* produce larvicidal effects [2c,2d].

The Mediterranean fruit fly, *Ceratitis capitata*, attacks a wide variety of hosts in subtropical and temperate regions of Argentina, causing serious economic damage [2e]. Continuing with our search for natural insecticides from native South Americans plants, we evaluated the adult mortality, the oviposition capacity of females, the egg viability, and the larval mortality of F1, as well as the alterations on the ovipositional behavior produced by seven ACGs of *A. cherimolia* seeds, squamocin (**1**), molvizarin (**2**), itrabin (**3**), almuñequin (**4**), cherimolin-1 (**5**), cherimolin-2 (**6**) and tucumanin (**7**) on *C. capitata*. Additionally, the phytotoxic, genotoxic and mutagenic effects displayed by itrabin, as well as the phytotoxic and genotoxic action of squamocin, the two most active ACGs on *C. capitata*, were investigated.

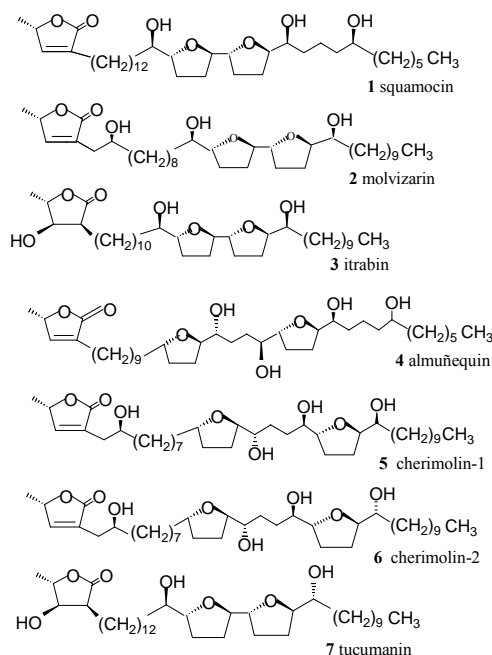


Figure 1: Annonaceous acetogenins employed in the experiment.

Annonaceous acetogenins: Squamocin (1), molvizarin (2), itrabin (3), almuñequin (4), cherimolin-1 (5), cherimolin-2 (6), and tucumanin (7) (Figure 1) were obtained and purified from seeds of *A. cherimolia* [2f]. Their structures were determined by comparison of their spectroscopic features with data previously reported in the literature.

Table 1: Oviposition-deterrent activity of ACGs on *C. capitata*.

Compounds (30 µg/cm ²)	N° eggs laid on the treated substrate (T) ^a	N° eggs laid on the nontreated substrate (C) ^a	%OI
Molvizarin	297.0 ± 2.8a	294.0 ± 11.3a	1.0
Itrabin	163.0 ± 9.8b	281.5 ± 3.5a	-42.1
Almuñequin	183.0 ± 4.2a	190.0 ± 9.9a	-3.9
Cherimolin-1	259.0 ± 2.8a	240.0 ± 14.1a	-4.6
Cherimolin-2	226.7 ± 4.3a	213.7 ± 19.6a	6.1
Squamocin	203.5 ± 9.2b	248.3 ± 11.3a	-18.0

Numbers represent Mean ± SD, n=3. ^aMeans within a row followed by the same letter are not significantly different ($P > 0.05$, Tukey multiple range test).

Oviposition-deterrent activity: To facilitate data interpretation, an oviposition index was defined as %OI = 100 T/C-100, where T is the number of eggs laid in the treated substrate, and C is the number of eggs deposited in the nontreated substrate. This index takes negative values for oviposition deterrents, and positive values for oviposition attractants. As shown in Table 1, the compounds that significantly deterred oviposition of *C. capitata* were itrabin and squamocin (%OI = -42.1 and -18.0, respectively) at 30 µg/cm².

Oviposition capacity and effects on the offspring: Oviposition capacity was significantly affected by the treated diets. The bioassay demonstrated that females from the first generation that fed on 250 ppm of treated diets with molvizarin, itrabin and squamocin oviposited

around one-fourth (0.15 ± 0.01 , 0.15 ± 0.01 and 0.20 ± 0.01 , respectively) the number of eggs of the control females (Table 2). The remaining ACGs, almuñequin, tucumanin, cherimolin-1 and cherimolin-2, reduced the oviposition capacity to around half the number of eggs of control females (0.34 ± 0.05 , 0.33 ± 0.06 , 0.33 ± 0.06 and 0.40 ± 0.10 , respectively). Eggs viability was not affected by the treatments and, therefore, no significant differences were registered in the hatching percentage of treatments in relation to control (Table 2). Treatments with molvizarin, itrabin, squamocin, and cherimolin-2 produced low but significant larval mortalities of 13.9 ± 1.3 , 13.5 ± 3.6 , 18.2 ± 1.3 , 12.9 ± 2.7 %, respectively, as shown in Table 2.

Table 2: Oviposition capacity and effects on the offspring of *C. capitata* produced by ACG.

Compounds	Volume of eggs ^a (mL)	(%) Hatch ^a	Larval Mortality ^a (%)
Control	0.63 ± 0.06a	97.0 ± 1.0a	7.6 ± 0.8a
Molvizarin	0.15 ± 0.01b	97.0 ± 1.7a	13.9 ± 1.3b
Itrabin	0.15 ± 0.01b	96.0 ± 0.6a	13.5 ± 3.6b
Squamocin	0.20 ± 0.01b	98.0 ± 1.2a	18.2 ± 1.3c
Almuñequin	0.34 ± 0.05c	97.0 ± 1.0a	7.0 ± 1.0a
Tucumanin	0.33 ± 0.06c	97.0 ± 1.0a	11.8 ± 2.1a
Cherimolin-1	0.33 ± 0.06c	97.0 ± 1.5a	12.9 ± 2.7b
Cherimolin-2	0.40 ± 0.10c	98.0 ± 0.6a	11.2 ± 3.4a

^aMean ± SD. Means within a column followed by the same letter are not significantly different ($P > 0.05$, Tukey multiple range test)

Table 3: Phytotoxic and genotoxic action of squamocin and itrabin on *Allium cepa* roots.

Itrabin (µg/mL)	Root elongation ^a (cm)	Mitotic index ^a (%)	Anaphasic aberration ^a	Micronucl ei number ^a
0	2.6±0.2	59±9a	1±1a	-
0.25	2.2±0.3	57±4a	ND	-
0.5	2.3±0.1	52 ±3a	2±1a	-
2.5	1.8±0.2	44 ±5b	3±1a	1±1a
Squamocin (µg/mL)				
0.25	2.2±0.2	41.4 ±5b	3±1a	-
0.5	1.9±0.1	34.2 ±7c	5±2b	-
2.5	1.7±0.1	30.7 ±3c	7±2c	2±1a

^aMean ± SD. Means followed by the same letter are not significantly different ($P > 0.05$, Tukey multiple range test). ND: not determined.

Allium cepa test: Macroscopic analysis of the treated roots revealed neither modification in consistency and shape nor tumors, hooked or twisted roots. The rate of inhibition of root elongation was around 30% for the maximum concentration (2.5 µg/mL) of itrabin and squamocin. The mitotic index (MI) is a parameter that estimates the frequency of cellular division. Its reduction could be due to either the inhibition of DNA synthesis or a blocking in the G₂ phase of the cell cycle, preventing the cell from entering mitosis. Squamocin produced a decrease between 30 and 50% of the MI values at the doses tested (0.25-2.5 µg/mL) (Table 3). Moreover, itrabin and squamocin induced anaphasic aberrations, as shown in Table 3.

Salmonella typhimurium test: Itrabin was tested for its mutagenic potency at 0.25, 0.5 and 2.5 µg/plate with and without S9 mix on both *S. typhimurium* TA98 and

Table 4: Mutagenic effects displayed by itrafin on *Salmonella typhimurium* strains TA98 and TA100.

Dose ($\mu\text{g}/\text{plate}$)	Mutagenicity relation (MR)			
	-S9		+S9	
	TA 98	TA 100	TA 98	TA 100
0.25	1.2	0.98	0.95	1.10
0.5	1.07	0.82	0.98	0.97
2.5	1.06	0.90	1.10	1.15
DMSO control	1.10	0.90	0.78	1.20
Positive control	2.26	2.91	2.89	3.01

MR: Revertants number in ACG plate/ Revertants number in control plate. Results are expressed as the mean number of revertants (standard error of triplicate plates).

TA100 strains. Non significant differences in the revertants number were detected for the treated experiments compared with control, as shown in Table 4. Our results are in agreement with the earlier studies reported about the mutagenicity of ACGs [2b].

The influence of chemical agents on the ovipositional behavior of insects can be used to control pests as well as vector insects. Some natural products display oviposition-deterrent activity against different insects. Factors that affect the oviposition behavior of some tephritid fruit flies include host-plant odor as well as the physical and chemical characteristics of the oviposition substrate, such as size, shape, color, and presence of deterrent chemicals. *C. capitata* has a wide geographical distribution and is considered one of the most important pests in the world affecting many species of commercially valuable fruits, including citrus. Therefore, substances that affect their behavior can be used in the development of pest-control agents.

This is the first report on the effects of acetogenins of *A. cherimolia* on *C. capitata*. The alterations in the capacity and behavior of oviposition of females produced by treatments with 250 ppm of itrafin, molvizarin and squamocin, together with the low genotoxicity and no mutagenicity, are promising results for the design of a natural pest control formulation employing the cited ACGs.

Experimental

Purification and identification of acetogenins: Dried and powdered seeds of *A. cherimolia* were percolated with methanol. The methanol extract was further partitioned between CHCl_3 and H_2O . The CHCl_3 extract was processed by chromatographic techniques, including silica gel columns and high performance liquid chromatograph employing the methodology previously reported [2c]. Structural determination of acetogenins was assessed by spectroscopic data in comparison with previously reported data [3a-3e, 4a].

Test insects and diet: A colony of *C. capitata* was initiated with pupae obtained from infested oranges from the northwest of Argentina. Adults were fed on

artificial diets made of water and a mixture of sugar and yeast hydrolysate (3:1). They were maintained in a rearing room with a photoperiod of 12L:12D, at $24\pm 2^\circ\text{C}$ and $60\pm 10\%$ relative humidity.

Oviposition-deterrent activity: Artificial fruits (oviposition substrates) were prepared by boiling a mixture of peach juice (500 mL), agar (15 g), and sodium benzoate (one teaspoonful, as preservative) and pouring the preparation into cylinders wrapped in plastic (Rolopac). After cooling, the surface of the wrapped cylinders was pricked with a needle and $30\ \mu\text{g}$ of the test compound was deposited per cm^2 . Control cylinders were impregnated only with the solvent that was then removed by evaporation. Three groups of *C. capitata* adults were selected from the laboratory colony. Each group, consisting of 7 male-female pairs, was placed in a small cage and covered with voile (a light, almost transparent cloth made of silk). Two agar cylinders (sample and control) were placed over the voile, and females oviposited on either one or the other according to their preference. After 4 days, eggs were gently rinsed from the agar and counted. All assays were conducted in triplicate [4b].

Evaluation of the oviposition capacity and effects on the offspring: Newly emerged adults (1:1, male:female) of *C. capitata* were sexed and placed in a cage of wooden frames and canvas lined with a mosquito net in order to mate and lay eggs. Insects were fed on diets treated with $250\ \mu\text{g}$ of each pure ACG per g of diet. An identical cage was set for a control experiment in which the diet did not contain the treatment. The number of adults was registered daily. Oviposition substrates were provided in the cages. After 5 days the eggs laid by females in the treated substrate (T) and nontreated substrate (C) were collected, the volume of eggs determined and the eggs placed in Petri dishes with larval diet. Dishes were placed in a rearing chamber under controlled temperature and humidity (25°C and 80% RH). After 3 days the number of emerged larvae was determined and, after 18 days, the number of pupae was quantified.

Plant genotoxicity test (*Allium cepa* test): For the *A. cepa* (onion) root anaphase aberration assay and the *A. cepa* root micronuclei assay [4c], equal sized young bulbs of common onions were used. All onion bulbs were kept in commercial mineral water for 48 h until root emergency. Six onion bulbs were exposed to 0.25, 0.5, and $2.5\ \mu\text{g}/\text{mL}$ of each ACG (suspended in a 0.1:10 DMSO-water mixture) for 24 h in the dark. Then the roots of 3 bulbs were cut up and fixed for 24 h in a 1:3 acetic acid-ethanol solution. The remaining 3 bulbs were kept in tap water for another 24 h (recovery time), and then the roots were fixed, as indicated previously.

Finally, all roots were stored in 70% ethanol. In the negative control experiment, 3 onion bulbs were exposed only to a 0.1:10 DMSO-water solution and they were submitted to the procedures previously described for treated bulbs. Three onion bulbs were exposed to a 10^{-4} M aqueous solution of methyl methanesulfonate in a positive control experiment. Macroscopic parameters such as length of roots, number of tumors, and hooked and twisted roots were registered and compared with control experiments to determine the phytotoxic action of ACG. Mitotic index (1000 cells per slide) was used to evaluate cellular division rate. Anaphasic aberrations (bridges, laggard chromosomes, and fragments; 800 anaphasic cells per sample) and micronuclei formation (5 slides, 1000 cells per slide) were the microscopic parameters assessed as indicators of DNA damage. For 40X microscopic observations, chromosomes were stained by the following procedure. Tips were cut from roots to be further hydrolyzed in 1 M HCl at 60°C for 10 min before staining in Schiff's reagent for 15 min. A portion of stained tip was immersed in a drop of 45% acetic acid, placed on a clean slide, and covered with a coverglass in order to have a single layer of cells.

Mutagenicity assay: The mutagenic effects of ACG were assayed according to the Ames test using *Salmonella typhimurium* strains TA98 and TA100 [4d], with and without metabolic activation (500 μ L of S9 mix fraction). The tested strains were cultured in Oxoid Nutrient Broth for 12 h. Different concentrations of ACGs (0.25 - 0.5 μ g/plate) in DMSO solution, were

added to 2 mL of soft agar containing L-hystidin (0.05 mM), D-biotin (0.5 mM), and 0.1 mL of bacterial culture. Then, this mixture was poured onto a plate containing minimum medium (Oxoid N°2). The plates were incubated at 37°C for 48 h, and the His⁺ revertant colonies were manually counted. S9 fraction was obtained from liver of Sprague-Dawley rats pretreated with a mixture of polychlorinated biphenyls (Araclor 1254). All experiments were analyzed in triplicate. A sample was considered to be mutagenic when the number of revertant colonies was at least twice the negative control yield and showed a significant response in analysis of variance. For the positive control experiment, the direct-acting mutagen 4-nitro-*o*-phenylenediamine (NPD, 5 μ g/plate) and the indirect-acting mutagen isoquinoline (IQ, 0.1 μ g/plate for TA98 and 0.5 μ g /plate for TA 100) were incorporated.

Statistical analysis: The results are reported as mean \pm SEM. The differences in the mean values were evaluated by analysis of variance (ANOVA). The Tukey test was used for all pair wise multiple comparisons of groups. In all statistical analyses, values of $P > 0.05$ were considered not significant (Statistix 7.1 2000).

Acknowledgments - This work was supported by grants from Consejo de Investigaciones de la Universidad de Tucumán (CIUNT) and Consejo Nacional de Investigaciones Científicas y Técnicas (CONICET), Argentina.

References

- [1] Alali F, Liu X, Mc Laughlin JL. (1999) Annonaceous acetogenins: recent progress. *Journal of Natural Products*, **62**, 504-540.
- [2] (a) Londerhausen M, Leicht W, Lieb F, Moeschler H, Weiss H. (1991) Molecular mode of action of annonins. *Pesticide Science*, **33**, 427-438; (b) Guadaño A, Gutiérrez C, de la Peña E, Cortes D, González-Coloma A. (2000) Insecticidal and mutagenic evaluation of two annonaceous acetogenins. *Journal of Natural Products*, **63**, 773-776; (c) Álvarez Colom O, Neske A, Popich S, Bardón A. (2007) Toxic effects of annonaceous acetogenins from *Annona cherimolia* (Magnoliales: Annonaceae) on *Spodoptera frugiperda* (Lepidoptera: Noctuidae). *Journal of Pest Science*, **80**, 63-67.; (d) Álvarez Colom O, Barrachina I, Ayala Mingol I, Gonzalez Mas MC, Moya Sanz P, Neske A, Bardón A. (2008) Toxic effects of annonaceous acetogenins on *Oncopeltus fasciatus*. *Journal of Pest Science*, **81**, 85-89; (e) Aluja M. (1995) Future trends in fruit fly management. In McPherson BA, Steck GJ (Eds) *Fruit fly pests: a world assessment of their biology and management*, Florida, USA, St. Lucie Press, 309-320; (f) Barrachina I, Neske A, Granell S, Bermejo A, Chahaboune N, El Aoued N, Álvarez Colom O, Bardón A, Zafra Polo MC. (2004) Tucumanin, a β -hydroxy γ -lactone bistetrahydrofuranic acetogenin from *Annona cherimolia*, potent inhibitor of mitochondrial complex I. *Planta Medica*, **70**, 866-868.
- [3] (a) McCloud TG, Smith DL, Chang CJ, Cassady JM. (1987) Annonacin, a novel, biologically active polyketide from *Annona densicoma*. *Experientia*, **43**, 947-949; (b) Folker Lieb MN, Wachendorff-Neumann U, Detlef W. (1990) Annonacins and annonastatin from *Annona squamosa*. *Planta Medica*, **56**, 317-319; (c) Yu GL, Ho DK, Cassady JM. (1992) Cytotoxic polyketides from *Annona densicoma* (Annonaceae). 10,13-*Trans*-13,14-erythro-densicomacin, 10,13-*trans*-13,14-threo-densicomacin and 8-hydroxyannonacin. *Journal of Organic Chemistry*, **57**, 6198-6202; (d) Zeng L, Wu FE, Gu ZM, McLaughlin JL. (1995) Murihexocins A and B two novel mono-THF acetogenins with six hydroxyls, from *Annona muricata* (Annonaceae). *Tetrahedron Letters*, **36**, 5291-5294; (e) Rieser M, Gu Z, Fang XP, Zeng L, Word KV, McLaughlin JL. (1996) Five novel monotetrahydrofuran ring acetogenins from the seeds of *Annona muricata*. *Journal of Natural Products*, **59**, 100-108.
- [4] (a) Álvarez Colom O, Neske A, Chahaboune N, Zafra-Polo MC, Bardón A. (2009) Tucupentol, a novel mono-tetrahydrofuranic acetogenin from *Annona montana*, as a potent inhibitor of mitochondrial complex I. *Chemistry & Biodiversity*, **6**, 335-340; (b) Socolsky C, Fascio ML, D'Accorso NB, Salvatore A, Willink E, Asakawa Y, Bardón A. (2008) Effects of *p*-vinylphenyl glycosides and other related compounds on the oviposition behavior of *Ceratitis capitata*. *Journal of Chemical Ecology*, **34**, 539-548; (c) Fiskesjo G. (1993) The *Allium* test in wastewater monitoring. *Environment Toxicology Water Quality*, **8**, 291-298.; (d) Maron DM, Ames BN. (1983) Revised methods for the *Salmonella* mutagenicity test. *Mutation Research*, **113**, 173-215.

Two New Carthamosides from *Carthamus oxycantha*

Zahid Hassan, Viqar Uddin Ahmad, Javid Hussain, Aqib Zahoor, Imran Nafees Siddiqui, Nasir Rasool and Muhammad Zubair 419

A New Lignan Dimer from *Mallotus philippensis*

Nguyen Thi Mai, Nguyen Xuan Cuong, Nguyen Phuong Thao, Nguyen Hoai Nam, Nguyen Huu Khoi, Chau Van Minh, Yvan Vander Heyden, Ngo Thi Thuan, Nguyen Van Tuyen, Joëlle Quetin-Leclercq and Phan Van Kiem 423

Tectone, a New Antihyperglycemic Anthraquinone from *Tectona grandis* Leaves

Nivedita Shukla, Manmeet Kumar, Akanksha, Ghufraan Ahmad, Neha Rahuja, Amar B. Singh, Arvind K. Srivastava, Siron M. Rajendran and Rakesh Maurya 427

A Semi-quantitative FIA-ESI-MS Method for the Rapid Screening of *Hypericum perforatum* Crude Extracts

Anna Piovan, Raffaella Filippini and Rosy Caniato 431

Free and Bound Cinnamic Acid Derivatives in Corsica Sweet Blond Oranges

Eric Carrera, Mohamed Vall Ould El Kebir, Camille Jacquemond, François Luro, Yves Lozano and Emile M. Gaydou 435

Antioxidants from Tropical Herbs

Rasyidah Razab and Azlina Abdul Aziz 441

The Antitumor and Immunostimulating Activities of Water Soluble Polysaccharides from *Radix Aconiti*, *Radix Aconiti Lateralis* and *Radix Aconiti Kusnezoffii*

Tingting Gao, Hongtao Bi, Shuai Ma and Jingmei Lu 447

Seasonal Variation in the Leaf Essential Oil Composition of *Zanthoxylum clava-herculis* growing in Huntsville, Alabama

Lauren C. Eiter, Henry Fadamiro and William N. Setzer 457

Supercritical CO₂ Extraction of Essential Oils from *Chamaecyparis obtusa*

Yinzhe Jin, Dandan Han, Minglei Tian and Kyung-Ho Row 461

Variability of the Essential Oil Content and Composition of Chamomile (*Matricaria recutita* L.) affected by Weather Conditions

Beáta Gosztola, Szilvia Sárosi and Éva Németh 465

Acaricidal Activity against *Tetranychus urticae* and Chemical Composition of Peel Essential Oils of Three *Citrus* Species Cultivated in NE Brazil

Claudio Pereira Araújo Júnior, Claudio Augusto Gomes da Camara, Ilzenayde Araújo Neves, Nicolle de Carvalho Ribeiro, Cristianne Araújo Gomes, Márcilio Martins de Moraes and Priscilla de Sousa Botelho 471

Essential Oil Composition, Antioxidant Capacity and Antifungal Activity of *Piper divaricatum*

Joyce Kelly R. da Silva, Eloísa Helena A. Andrade, Elsie F. Guimarães and José Guilherme S. Maia 477

Chemical Composition, Toxicity and Larvicidal Activity of the Essential Oil from the Whole Plant of *Acalypha segetalis* from South-West Nigeria

Sherifat A. Aboaba, Olapeju O. Aiyelaagbe and Olusegun Ekundayo 481

Review/Account

Toxicity of Non-protein Amino Acids to Humans and Domestic Animals

Peter B. Nunn, E. Arthur Bell (the late), Alison A. Watson and Robert J. Nash 485

Natural Product Communications

2010

Volume 5, Number 3

Contents

<u>Original Paper</u>	<u>Page</u>
(R)-(-)-Linalyl Acetate and (S)-(-)-Germacrene D from the Leaves of Mexican <i>Bursera linanoe</i> Koji Noge, Nobuhiro Shimizu and Judith X. Becerra	351
Three New Insecticidal Sesquiterpene Polyol Esters from <i>Celastrus angulatus</i> Shaopeng Wei, Minchang Wang, Zhiqin Ji, Baojun Shi, Shengkun Li and Jiwen Zhang	355
Triterpenoids from Aerial Parts of <i>Glochidion eriocarpum</i> Vu Kim Thu, Phan Van Kiem, Pham Hai Yen, Nguyen Xuan Nhiem, Nguyen Huu Tung, Nguyen Xuan Cuong, Chau Van Minh, Hoang Thanh Huong, Trinh Van Lau, Ngo Thi Thuan and Young Ho Kim	361
Identification of Sakurasosaponin as a Cytotoxic Principle from <i>Jacquinia flammea</i> Alberto Sánchez-Medina, Luis M. Peña-Rodríguez, Filogonio May-Pat, Gloria Karagianis, Peter G. Waterman, Anthony I. Mallet and Solomon Habtemariam	365
Vasoconstrictor and Inotropic Effects Induced by the Root Bark Extracts of <i>Anthocleista schweinfurthii</i> Nadège Kabamba Ngombe, Dibungi T. Kalenda, Joëlle Quetin-Leclercq and Nicole Morel	369
Hydroxylation of Diosgenin by <i>Absidia coerulea</i> Ying Zhao, Ling-Mei Sun, Xiao-Ning Wang, Tao Shen, Mei Ji and Hong-Xiang Lou	373
Dibromotyrosine and Histamine Derivatives from the Tropical Marine Sponge <i>Aplysina</i> sp. Elena A. Santalova, Vladimir A. Denisenko and Valentin A. Stonik	377
<i>In vitro</i> Inhibitory Activities of Lauraceae Aporphine Alkaloids Ericsson David Coy Barrera and Luis Enrique Cuca Suárez	383
Leishmanicidal activity of racemic \pm 8-[(4-Amino-1-methylbutyl)amino]-6-methoxy-4-methyl-5-[3,4-dichlorophenoxy]quinoline Angélica P. Isaac-Márquez, James D. McChesney, N.P. Dammika Nanayakara, Abhay R. Satoskar and Claudio M. Lezama-Dávila	387
Insecticidal, Mutagenic and Genotoxic Evaluation of Annonaceous Acetogenins Olga Álvarez Colom, Analia Salvatore, Eduardo Willink, Roxana Ordóñez, María I. Isla, Adriana Neske and Alicia Bardón	391
Water-retentive and Anti-inflammatory Properties of Organic and Inorganic Substances from Korean Sea Mud Jung-Hyun Kim, Jeongmi Lee, Hyang-Bok Lee, Jeong Hyun Shin and Eun-Ki Kim	395
A Phenethyl bromo ester from <i>Citharexylum fruticosum</i> Seru Ganapaty, Desaraju Venkata Rao and Steve Thomas Pannakal	399
New 2-(2-Phenylethyl)chromone Derivatives from the Seeds of <i>Cucumis melo</i> L var. <i>reticulatus</i> Sabrin R. M. Ibrahim	403
Phenolic Compounds in Different Barley Varieties: Identification by Tandem Mass Spectrometry (QStar) and NMR; Quantification by Liquid Chromatography Triple Quadrupole-Linear Ion Trap Mass Spectrometry (Q-Trap) Kamilla Klausen, Anne G. Mortensen, Bente Laursen, Kim F. Haselmann, Birthe Møller Jespersen and Inge S. Fomsgaard	407
Effect of <i>Cleome arabica</i> Leaf Extract Treated by Naringinase on Human Neutrophil Chemotaxis Hamama Bouriche and Juegen Arnhold	415

Continued inside backcover