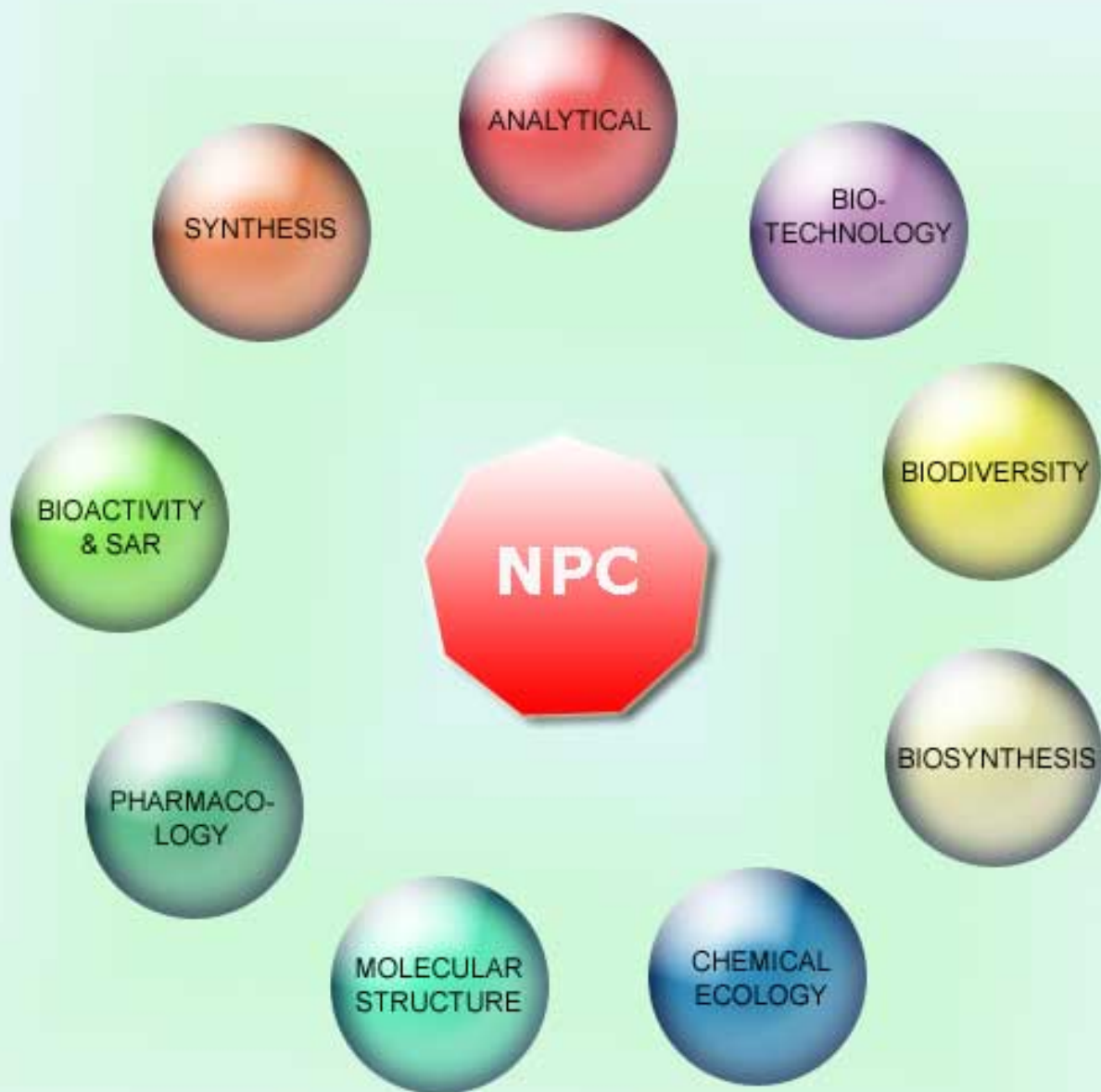


NATURAL PRODUCT COMMUNICATIONS

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



Volume 4. Issue 6. Pages 749-888. 2009
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,
Universita 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 Organica,
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
mimaki@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. Anna R. Bilia
Florence, Italy

Prof. Maurizio Bruno
Palermo, Italy

Prof. Josep Coll
Barcelona, Spain

Prof. Geoffrey Cordell
Chicago, IL, USA

Prof. Samuel Danishefsky
New York, NY, USA

Prof. Duvvuru Gunasekar
Tirupati, India

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

Prof. Stephen Hanessian
Montreal, Canada

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. Hartmut Laatsch
Gottingen, Germany

Prof. Marie Lacaille-Dubois
Dijon, France

Prof. Shoei-Sheng Lee
Taipei, Taiwan

Prof. Francisco Macias
Cadiz, Spain

Prof. Anita Marsaioli
Campinas, Brazil

Prof. Imre Mathe
Szeged, Hungary

Prof. Joseph Michael
Johannesburg, South Africa

Prof. Ermino Murano
Trieste, Italy

Prof. Virinder Parmar
Delhi, India

Prof. Luc Pieters
Antwerp, Belgium

Prof. Om Prakash
Manhattan, KS, USA

Prof. Peter Proksch
Düsseldorf, Germany

Prof. Satyajit Sarker
Wolverhampton, UK

Prof. Raffaele Riccio
Salerno, Italy

Prof. Monique Simmonds
Richmond, UK

Prof. Valentin Stonik
Vladivostok, Russia

Prof. Hiromitsu Takayama
Chiba, Japan

Prof. Karen Valant-Vetschera
Vienna, Austria

Prof. Peter G. Waterman
Lismore, Australia

Prof. Paul Wender
Stanford, USA

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. 2009 subscription price: US\$1,695 (Print, ISSN# 1934-578X); US\$1,395 (Web edition, ISSN# 1555-9475); US\$2,095 (Print + single site 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.

Acetylcholinesterase Inhibition and Antioxidant Activity of Essential Oils from *Schinus areira* L. and *Schinus longifolia* (Lindl.) Speg.

Ana P. Murray^{a,b*}, María S. Vela Gurovic^{a,b}, Silvana A. Rodriguez^a, María G. Murray^{b,c} and Adriana A. Ferrero^c

^aDepartamento de Química, Universidad Nacional del Sur, Av. Alem 1253, B8000CPB Bahía Blanca, Argentina

^bConsejo Nacional de Investigaciones Científicas y Técnicas, Argentina

^cDepartamento de Biología, Bioquímica y Farmacia, Universidad Nacional del Sur, San Juan 670, B8000CPB Bahía Blanca, Argentina

apmurray@uns.edu.ar

Received: January 23rd, 2009; Accepted: April 17th, 2009

The essential oils of *Schinus areira* L. and *S. longifolia* (Lindl.) Speg. (Anacardiaceae) have been studied for their *in vitro* anti-acetylcholinesterase and antioxidant activities. The chemical composition of the oils obtained by hydrodistillation was determined by GC-MS. Fruit and leaf oils of *S. areira* were analyzed separately. The essential oil from *S. longifolia* elicited marked enzymatic inhibition ($IC_{50} = 20.0 \pm 1.0 \mu\text{g/mL}$) and showed radical scavenger activity ($IC_{50} = 25.2 \pm 2.4 \mu\text{g/mL}$). The essential oil from *S. areira* leaves was more active than that of the fruits in both bioassays.

Keywords: *Schinus areira*, *Schinus longifolia*, Anacardiaceae, acetylcholinesterase inhibition, antioxidants, essential oils.

The genus *Schinus* L. (Anacardiaceae) is represented in Argentina by 22 species. *S. areira* L. [syn. *S. molle* L. var. *areira* (L.) DC.], commonly known as 'aguaribay', 'árbol de la pimienta' or 'pimentero', is a native species from the north-west region of Argentina. The trees are 2 to 5 metres high with evergreen leaves made up of 2-25 pairs of leaflets, and the fruit is a purplish drupe, 5-6 mm in diameter [1a-c]. *S. longifolia* (Lindl.) Speg. is also a native species, which is very common throughout the centre of the country. Its common name is 'Molle', 'Incienso' or 'Trementina'. It is a bush or tree up to 5 metres in height, frequently parasitized by different insects that produce round galls [1b], with persistent leaves and a fruit that is a lilac colored drupe, 5-6 mm in diameter [2].

S. areira is used as a purgative, diuretic, parasiticide, insecticide, vulnerary, and topical disinfectant, and for the treatment of rheumatism, stomach upsets, menstrual disorders, bronchitis and conjunctivitis [3a,b]. The essential oil of *S. areira* has shown

significant antibacterial and antifungal activity and has been reported as an allelopathic agent [3c-e]. *S. longifolia* is used in the treatment of colds, as an expectorant, laxative, emollient and for toothache [4a,b].

Acetylcholinesterase (AChE) inhibitors are used in medicine and in pest control. A few of them are in clinical use for the early stages of Alzheimer's disease (AD). Currently available approved drugs for the symptomatic treatment of AD patients inhibit AChE thus enhancing acetylcholine levels in the brain, which is associated with improvement of cognitive functions. Since these drugs are frequently associated with adverse effects, there is great interest in finding better AChE inhibitors.

A large body of evidence suggests that oxidative stress is closely related with the development and progression of a neurodegenerative disease like AD since free radical oxidative damage is extensive in the brains of AD patients [5]. Recent studies have

Table 1: Chemical composition of essential oils from *S. areira* and *S. longifolia*.

Compounds ^a	KI	Percentage		
		<i>S. areira</i> leaves	<i>S. areira</i> fruits	<i>S.</i> <i>longifolia</i>
Tricyclene	927	1.1	-	-
α -Pinene	939	5.4	3.1	0.2
Camphene	953	6.3	1.8	-
Sabinene	976	4.3	-	-
β -Pinene	981	2.7	0.8	-
β -Myrcene	991	3.4	19.3	-
2-Carene	1002	-	0.6	0.3
α -Phellandrene	1004	16.2	31.8	0.4
α -Terpinene	1017	0.3	-	-
p-Cymene	1026	5.1	3.0	-
β -Phellandrene	1031	17.6	19.9	2.3
α -Ocimene	1056	0.3	-	-
Terpinen-4-ol	1177	1.2	-	-
α -Terpineol	1188	-	-	1.7
Bornyl acetate	1285	1.4	-	-
α -Cubebene	1351	-	-	0.6
α -Copaene	1377	0.4	-	2.5
β -Caryophyllene	1418	2.0	2.0	9.0
β -Copaene	1432	-	-	0.7
Aromadendrene	1441	-	-	3.5
α -Humulene	1454	0.8	-	1.9
Alloaromadendrene	1461	0.5	-	1.1
β -Cadinene	1473	0.7	-	3.0
γ -Murolene	1477	5.1	0.9	5.0
Germacrene D	1480	-	-	1.9
β -Selinene	1485	1.5	-	7.1
α -Murolene	1499	1.3	0.9	1.9
α -Farnesene	1508	0.5	-	-
γ -Cadinene	1512	1.4	0.9	1.8
δ -Cadinene	1524	2.7	4.6	6.3
α -Elemol	1547	0.9	-	-
Germacrene B	1556	0.5	-	3.0
Palustrol	1560	0.6	1.2	-
Spathulenol	1565	0.6	-	5.1
Globulol	1575	0.5	1.1	14.2
Viridiflorol	1591	0.9	1.1	10.5
Cedrol	1595	-	-	4.0
Guaiol	1597	6.2	-	-
δ -Cadinol	1636	2.9	3.3	4.1
β -Eudesmol	1649	0.5	0.6	1.3
α -Cadinol	1653	1.0	-	-
Total		95.8	97.0	93.3

^a Compounds are listed in order of elution from a DB-5 column

shown that antioxidant therapies are safe and produce no adverse, but beneficial effects in AD patients [6]. Treatment with antioxidants appears to be an alternative approach for slowing disease progression.

Essential oils have attracted attention as potential AChE inhibitors due to several recent reports on active oils and active terpenoids [7a-l]. Inhibition of AChE by different kinds of monoterpenoids has been evaluated and several studies have been conducted to identify structure-activity relationships [7b-g,7l].

As part of our ongoing research on bioactive natural compounds [8], essential oils from *S. areira* and *S. longifolia* were analyzed for AChE inhibitory and antioxidant capacity. The essential oils from the fruits and leaves of *S. areira* were separately studied. The

oils, obtained from fresh plant material collected around Bahía Blanca city, were analyzed by gas chromatography and mass spectrometry. Their chemical composition is summarized in Table 1.

The essential oils were preliminary evaluated over TLC plates with detection of AChE inhibition [9]; all the samples showed enzymatic inhibition. Then, a spectrophotometric assay was carried out in order to quantify the enzymatic activity [10]. The results, expressed as IC₅₀ values, are summarized in Table 2. The essential oil from leaves of *S. areira* was more active (IC₅₀ = 233.8 ± 1.0 µg/mL) than that from the fruits (IC₅₀ = 487.9 ± 1.2 µg/mL), but *S. longifolia* oil exhibited the best AChE inhibition with an IC₅₀ value = 20.0 ± 1.0 µg/mL. These results are comparable with, and in many cases better than those reported in the past for other essential oils [7c,7i,7k,7l,11,12]. The AChE inhibition observed for the *S. longifolia* oil could be attributed in part to the presence of viridiflorol (10.5%), a potent inhibitor [7i]. The higher levels of active monoterpenes, like α -pinene and β -pinene, in the leaves than in the fruits of *S. areira* could explain the differences observed in activity [7f, 7l].

The antioxidant activity of the essential oils was evaluated through their ability for free radical scavenging against the stable 2,2-diphenyl-1-picrylhydrazyl radical (DPPH). All the samples gave a positive result in the preliminary assay over TLC. Then, a spectrophotometric assay was carried out and the percentage DPPH reduction was calculated for each oil. *S. longifolia* essential oil was the most active (IC₅₀ = 25.2 ± 2.4 µg/mL), with an antioxidant capacity comparable to that of the reference compound. On the other hand, the essential oil from the leaves (IC₅₀ = 38.7 ± 2.5 µg/mL) elicited a higher activity than that from the fruits of *S. areira* (IC₅₀ = 59.6 ± 3.1 µg/mL). The high percentages of oxygenated sesquiterpenes could explain the better results obtained for *S. longifolia*, even though a correlation between components and activity is not clear for these oils.

Taking into account the results obtained in our study, the essential oils from *S. areira* (leaves) and, mainly, *S. longifolia* appear as promising candidates for more detailed *in vitro* and *in vivo* studies of their AChE inhibition and antioxidant capacities. The seasonal variation of the chemical composition of these oils and their biological activities will be a matter of future studies.

Experimental

Equipment and chemicals: GC-MS were performed with a HP6890 chromatograph connected to a HP5972A mass spectrometer equipped with a capillary column (HP-5, 25 m x 0.25 mm, 0.25 μ m film thickness). The carrier gas was helium (1 mL/min). The GC oven temperature was held at 50°C for 2 min, programmed at 5°C/min to 200°C, then held at this temperature for 15 min. Mass spectra were recorded at 70 eV. GC analyses were performed on a Shimadzu G14B chromatograph with a fid detector on a DB-5 column (30 m x 0.25 mm, 0.25 μ m film thickness) with the same analytical conditions used for the GC-MS analyses. UV spectra were recorded on a GBC Spectral UV-VIS spectrophotometer. Butylated hydroxytoluene (BHT), DPPH, AChE, 5,5'-Dithiobis(2-nitrobenzoic acid) (DTNB), acetylthiocholine iodide (ATC) and eserine were purchased from Sigma.

Plant material: Aerial parts of *S. longifolia* and *S. areira* were collected in March 2006 (summer), from healthy plants from five different locations. The taxonomy of this material was determined by Dr M.G. Murray. Voucher specimens are kept in the "Herbario del Departamento de Biología, Bioquímica y Farmacia-UNS (BBB)" under the numbers MGM121 (*S. areira*) and MGM400 (*S. longifolia*).

Essential oils: Essential oils were obtained by hydrodistillation, as previously reported [13]. The chemical composition of each oil was determined by GC and GC-MS. The compounds were identified by their retention indices and by comparison of their mass spectra with those stored in MS databases (NBS75K.L MS DATA). Relative percentage amounts were obtained directly from GC peak areas.

Acetylcholinesterase inhibition: AChE used in the assay was from electric eels. The lyophilized enzyme (425 units/mg solid) was prepared in buffer phosphate (8 mM K_2HPO_4 , 2.3 mM NaH_2PO_4) to obtain 5 U/mL stock solution. Further enzyme dilution was carried out with buffer phosphate (8 mM

K_2HPO_4 , 2.3 mM NaH_2PO_4 , 0.15 M NaCl, 0.05% Tween 20, pH 7.6) to produce 0.126 U/mL enzyme solution. Samples were dissolved in the same buffer with methanol as co-solvent (2.5%). AChE solution (300 μ L) and 300 μ L of sample solution were mixed in a test tube and incubated for 30 min at room temperature. The reaction was started by adding 600 μ L of the substrate solution (0.1 M Na_2HPO_4 , 0.5 mM DTNB, 0.6 mM ATCI, pH 7.5). The absorbance was read at 405 nm for 180 s. Enzyme activity was calculated by comparing reaction rates for the sample to the blank (containing 2.5% MeOH, $n = 3$). IC_{50} values were determined with probit analysis (EPA Probit 1.4). Eserine was used as the reference AChE inhibitor. The TLC assay with detection of AChE inhibition was carried out by a previously reported method [9].

Antioxidant activity: For the preliminary test, analytical TLC on silica gel plates were developed with appropriate conditions after application of 5 μ L of oil solution (5 mg/mL, ethyl ether), dried and sprayed with DPPH solution (0.2%, MeOH). Five minutes later active compounds appeared as yellow spots against a purple background. The spectrophotometric assay was carried out at five different concentrations of the essential oils (10-100 μ g/mL in MeOH: CH_2Cl_2 95:5). A sample of 300 μ L was mixed with 2.5 mL of 0.004% DPPH methanolic solution. The absorbance was measured at 517 nm after 30 min of incubation. The percentage of DPPH reduction was calculated taking into account the absorbance of the blank solutions (2.5 mL MeOH plus 300 μ L oil solution) and the negative control (2.5 mL DPPH solution plus 300 μ L of MeOH: CH_2Cl_2 95:5). BHT was used as reference compound.

Acknowledgments - This work was financially supported by CONICET (PIP N° 6056), Universidad Nacional del Sur and ANPCYT (BID 1728/OC-AR PICT N° 25775).

References

- [1] (a) Zuloaga FO, Morrone O. (1999) *Catálogo de las Plantas Vasculares de la República Argentina II. Monographs in Systematic Botany from the Missouri Botanical Garden*. Vol. 74. St. Louis, Missouri, USA. 43-44; (b) Dimitri MJ. (1980) *Enciclopedia Argentina de Agricultura y Jardinería*. Vol. I(2). Ed. Acme S.A.C.I., Buenos Aires, Argentina; (c) Kiesling R. (2003) Dicotiledóneas Dialipétalas. In *Flora de San Juan República Argentina*. Vol II. Estudio Sigma, Buenos Aires, Argentina.
- [2] Muñoz J, Ross P, Charco P. (1993) *Flora indígena del Uruguay. Árboles y arbustos ornamentales*. Editorial Hemisferio Sur, Montevideo, Uruguay.

- [3] (a) Gupta MP. (1995) *270 Plantas Medicinales Iberoamericanas*. Bogotá, Colombia, Ed. CYTED-SECAB. 21-24; (b) Barboza GE, Cantero JJ, Nuñez CO, Ariza Espinar L. (2006) *Flora Medicinal de la Provincia de Córdoba (Argentina). Pteridofitas y antofitas silvestres o naturalizadas*. Córdoba, Argentina. Ed. Museo Botánico de Córdoba, 238-239; (c) Dikshit A, Naqvi AA, Husain A. (1986) *Schinus molle*: a new source of natural fungitoxicant. *Applied and Environmental Microbiology*, **51**, 1085-1088; (d) Gundidza M. (1993) Antimicrobial activity of essential oil from *Schinus molle* Linn. *The Central African Journal of Medicine*, **39**, 231-234. (e) Scrivanti LR, Zunino MP, Zygadlo JA. (2003) *Tagetes minuta* and *Schinus areira* essential oils as allelopathic agents. *Biochemical Systematics and Ecology*, **31**, 563-572.
- [4] (a) Waizel-Bucay J, Martínez Rico I. (2007) Plantas empleadas en odontalgias I. *Revista ADM*, **LXIV**, 173-186. <http://www.medigraphic.com/pdfs/adm/od-2007/od075b.pdf>; (b) Lahitte HB, Hurrell JA, Belgrano MJ, Jankowski L, Haloua P, Mehltreter K. (1998) *Biota Rioplatense II. Plantas Medicinales Rioplatenses*. Buenos Aires, Argentina. Ed. L.O.L.A. (Literature of Latin America). 168-169.
- [5] Valko M, Leibfritz D, Moncol J, Cronin MTD, Mazur M, Telser J. (2007) Free radicals and antioxidants in normal physiological functions and humane disease. *International Journal of Biochemistry and Cell Biology*, **39**, 44-84.
- [6] Hemachandra Reddy P. (2006) Mitochondrial oxidative damage in aging and Alzheimer's disease: Implications for mitochondrially targeted antioxidant therapeutics. *Journal of Biomedicine and Biotechnology*, 1-13.
- [7] (a) Houghton PJ, Ren Y, Howes MJ. (2006) Acetylcholinesterase inhibitors from plants and fungi. *Natural Products Reports*, **23**, 181-199; (b) Perry NSL, Bollen C, Perry E, Ballard C. (2003) *Salvia* for dementia therapy: review of pharmacological activity and pilot tolerability clinical trial. *Pharmacology, Biochemistry and Behavior*, **75**, 651-659; (c) Savelev S, Okello E, Perry NSL, Wilkins RM, Perry EK. (2003) Synergistic and antagonistic interactions of anticholinesterase terpenoids in *Salvia lavandulaefolia* essential oil. *Pharmacology, Biochemistry and Behavior*, **75**, 661-668; (d) Mills C, Cleary BJ, Gilmer JF, Walsh JJ. (2004) Inhibition of acetylcholinesterase by tea tree oil. *The Journal of Pharmacy and Pharmacology*, **56**, 375-379; (e) Miyazawa M, Watanabe H, Kameoka H. (1997) Inhibition of acetylcholinesterase activity by monoterpenoids with a *p*-menthane skeleton. *Journal of Agricultural and Food Chemistry*, **45**, 677-679; (f) Miyazawa M, Yamafuji C. (2005) Inhibition of acetylcholinesterase activity by bicyclic monoterpenoids. *Journal of Agricultural and Food Chemistry*, **53**, 1765-1768; (g) Perry NSL, Houghton PJ, Theobald A, Jennert P, Perry EK. (2000) *In-vitro* inhibition of human erythrocyte acetylcholinesterase by *Salvia lavandulaefolia* essential oil and constituent terpenes. *The Journal of Pharmacy and Pharmacology*, **52**, 895-902; (h) Mata AT, Proença C, Ferreira AR, Serralheiro MLM, Nogueira JMF, Araújo MEM. (2007) Antioxidant and antiacetylcholinesterase activities of five plants used as Portuguese food spices. *Food Chemistry*, **103**, 778-786; (i) Miyazawa M, Watanabe H, Umemoto K, Kameoka H. (1998) Inhibition of acetylcholinesterase activity by essential oils of *Mentha* species, *Journal of Agricultural and Food Chemistry*, **46**, 3431-3434; (j) Miyazawa M, Tougo H, Ishihara M. (2001) Inhibition of acetylcholinesterase activity by essential oil from *Citrus paradisi*. *Natural Product Letters*, **15**, 205-210; (k) Miyazawa M, Yamafuji C. (2006) Inhibition of acetylcholinesterase activity by tea tree oil and constituent terpenoids. *Flavour and Fragrance Journal*, **21**, 198-201; (l) Orhan I, Kartal M, Kan Y, Şener B. (2008) Activity of essential oils and individual components against acetyl- and butyrylcholinesterase. *Zeitschrift für Naturforschung C*, **63**, 547-553.
- [8] (a) Murray AP, Rodriguez SA, Murray MG. (2008) Antioxidant activity and chemical composition of essential oils from *Schinus fasciculata* (Griseb.) I.M. Johnst and *S. praecox* (Griseb.) Speg. *Natural Product Communications*, **3**, 1551-1556; (b) Rodriguez SA, Murray AP. (2008) Volatile components of *Discaria americana* Gillies & Hook (Rhamnaceae). *Natural Product Research*, **22**, 253-257.
- [9] Rhee IK, van der Meent M, Ingkaninan K, Verpoorte R. (2001) Screening for acetylcholinesterase inhibitors from Amaryllidaceae using silica gel thin-layer chromatography in combination with bioactivity staining. *Journal of Chromatography A*, **915**, 217-223.
- [10] (a) Ellman GL, Courtney KD, Andres V, Featherstone RM. (1961) A new and rapid colorimetric determination of acetylcholinesterase activity. *Biochemical Pharmacology*, **7**, 88-90; (b) López S, Bastida J, Viladomat F, Codina C. (2002) Acetylcholinesterase inhibitory activity of some Amaryllidaceae alkaloids and *Narcissus* extracts. *Life Sciences*, **71**, 2521-2529.
- [11] Ferreira A, Proença C, Serralheiro MLM, Araújo MEM. (2007) The *in vitro* screening for acetylcholinesterase inhibition and antioxidant activity of medicinal plants from Portugal. *Journal of Ethnopharmacology*, **108**, 31-37.
- [12] Orhan I, Aslan S, Kartal M, Şener B, Başer KHC. (2008) Inhibitory effect of Turkish *Rosmarinus officinalis* L. on acetylcholinesterase and butyrylcholinesterase enzymes. *Food Chemistry*, **108**, 663-668.
- [13] Murray AP, Frontera MA, Tomas MA, Mulet MC. (2005) Gas chromatography-mass spectrometry study of the essential oils of *Schinus longifolia* (Lindl.) Speg., *Schinus fasciculata* (Griseb.) I.M. Johnst. and *Schinus areira* L. *Zeitschrift für Naturforschung C*, **60**, 25-29.

Simultaneous Quantification of Eight Major Bioactive Phenolic Compounds in Chinese Propolis by High-Performance Liquid Chromatography Na Sha, Hui-Lian Huang, Jin-Qiang Zhang, Guang-Tong Chen, Si-Jia Tao, Min Yang, Xing-Nuo Li, Ping Li and De-An Guo	813
Total Phenolic Content and Antioxidant Activity of Myrtle (<i>Myrtus communis</i>) Extracts MahassineAmensour, Esther Sendra, Jamal Abrini, Samira Bouhdid, José Angel Pérez-Alvarez and Juana Fernández-López	819
Simultaneous Determination of Oxyresveratrol and Resveratrol in Rat Bile and Urine by HPLC after Oral Administration of <i>Smilax china</i> Extract Hui-lian Huang, Jin-qiang Zhang, Guang-tong Chen, Zhi-qiang Lu, Na Sha and De-an Guo	825
Composition of Essential Oils from Leaves and Flowers of <i>Stachys germanica</i> subsp. <i>salviifolia</i> (Ten.) Gams (Labiatae) and Related Secretory Structures Claudia Giuliani, Roberto Maria Pellegrino, Bruno Tirillini and Laura Maleci Bini	831
Volatile Constituents of <i>Trifolium pratense</i> and <i>T. repens</i> from N.E. Italian Alpine Pastures Aldo Tava, Daniele Ramella, Maris Grecchi, Paolo Aceto, Renato Paoletti and Efisio Piano	835
Seasonal Variation and Bioactivity in the Leaf Oil of <i>Liriodendron tulipifera</i> Growing in Huntsville, Alabama Sarah L. Miller, Heather E. Villanueva, Maria C. Palazzo, Brenda S. Wright and William N. Setzer	839
Composition and Seasonal Variation of the Essential Oil from <i>Abies sachalinensis</i> from Hokkaido, Japan Tadaaki Satou, Mariko Matsuura, Shio Murakami, Shinichiro Hayashi and Kazuo Koike	845
Compositional Variation of the Essential Oils of <i>Artemisia afra</i> Jacq. from three Provinces in South Africa - A Case Study of its Safety Adebola O. Oyedepi, Anthony J. Afolayan and Anne Hutchings	849
Chemical Variability of Essential Oils of <i>Lippia alba</i> (Miller) N. E. Brown Growing in Costa Rica and Argentina Gabriela Ricciardi, José F. Cicció, Rafael Ocampo, Daniel Lorenzo, Armando Ricciardi, Arnaldo Bandoni and Eduardo Dellacassa	853
Composition and Antibacterial Activity of Essential Oils from Leaf, Stem and Root of <i>Chrysanthemum parthenium</i> (L.) Bernh. from Iran Ali Shafaghat, Hajar Sadeghi and Khodamali Oji	859
Antibacterial Activity and Composition of Essential Oils from Flower, Leaf and Stem of <i>Chaerophyllum macropodium</i> Boiss. from Iran Ali Shafaghat	861
Composition and Antimicrobial Activity of the Leaf Essential Oil of <i>Litsea nakaii</i> from Taiwan Chen-Lung Ho, Eugene I-Chen Wang, Pei-Yeh Lee and Yu-Chang Su	865
Chemical Composition and Antimicrobial Activity of <i>Clausena indica</i> (Dalz) Oliv. (Rutaceae) Essential Oil from Vietnam Pham Thi Minh Diep, Agata Maria Pawlowska, Pier Luigi Cioni, Chau Van Minh, Le Mai Huong and Alessandra Braca	869
Acetylcholinesterase Inhibition and Antioxidant Activity of Essential Oils from <i>Schinus areira</i> L. and <i>Schinus longifolia</i> (Lindl.) Speg. Ana P. Murray, María S. Vela Gurovic, Silvana A. Rodriguez, María G. Murray and Adriana A. Ferrero	873
Potential Anti-dementia Agents in Traditional Chinese Medicine Xue-Juan Li and Hong-Yu Zhang	877

Natural Product Communications 2009

Volume 4, Number 6

Contents

<u>Original Paper</u>	<u>Page</u>
Thymol-evoked Ca²⁺ Mobilization and Ion Currents in Pituitary GH₃ Cells Ai-Yu Shen, Mei-Han Huang, Trey-Shy Wang, Hui-Ming Wu, Ya-Fei Kang and Chi-Lan Chen	749
Iridoids from <i>Spathodea campanulata</i> P. Beauvais Leaves Yaser G. Gouda	753
A New Longipinene Diester from <i>Stevia monardifolia</i> Kunth Rodrigo E. Rojas-Pérez, Ernestina Cedillo-Portugal, Pedro Joseph-Nathan and Eleuterio Burgueño-Tapia	757
A New Sesquiterpene from the Roots of <i>Vladimiria souliei</i> Jing Xu, Xiaojun Zhao, Yuanqiang Guo, Wenyuan Gao and Shuzhong Zhang	763
Asiatic Acid Derivatives Protect Primary Cultures of Rat Hepatocytes against Carbon Tetrachloride-Induced Injury via the Cellular Antioxidant System Mi Kyeong Lee, Seung Hyun Kim, Hyekyung Yang, Doo-Yeon Lim, Je-Ho Ryu, Eung Seok Lee, Sang-Sup Jew, Hyeung-Guen Park, Sang Hyun Sung and Young Choong Kim	765
A Minor, Sweet Cucurbitane Glycoside from <i>Siraitia grosvenorii</i> Zhonghua Jia and Xiaogen Yang	769
Cytotoxic Action of Triterpene Glycosides from Sea Cucumbers from the genus <i>Cucumaria</i> on Mouse Spleen Lymphocytes. Inhibition of Nonspecific Esterase Dmitry L. Aminin, Alexandra S. Silchenko, Sergey A. Avilov, Vadim G. Stepanov and Vladimir I. Kalinin	773
Inhibition of Mast Cell Degranulation by Saponins from <i>Gleditsia sinensis</i>- Structure-activity Relationships Wang Chong, Xia Yu Feng, Gao Zheng Zhen, Lu Dan and Dai Yue	777
Molecular Modeling, NOESY NMR, and the Structure of Nicandrenone Isolated from <i>Nicandra physalodes</i> (Solanaceae) Patrick F. Jonas and Geoffrey A. Cordell	783
Analysis of <i>Salvia coccinea</i> from Jamaican Populations Glenroy D. A. Martin, William F. Reynolds and Paul B. Reese	789
Antimicrobial, Antiparasitic and Cytotoxic Spermine Alkaloids from <i>Albizia schimperiana</i> Volodymyr Samoylenko, Melissa R. Jacob, Shabana I. Khan, Jianping Zhao, Babu L. Tekwani, Jacob O. Midiwo, Larry A. Walker and Ilias Muhammad	791
DMD Mediated Formal Synthesis of (±)-Coerulescine Oscar R. Suárez-Castillo, Myriam Meléndez-Rodríguez, Yaneth M. A. Contreras-Martínez, Alejandro Álvarez-Hernández, Martha S. Morales-Ríos and Pedro Joseph-Nathan	797
Globulixanthone F, a New Polyoxygenated Xanthone with an Isoprenoid group and two Antimicrobial Biflavonoids from the Stem Bark of <i>Symphonia globulifera</i> Pierre Mkounga, Zacharias T. Fomum, Michèle Meyer, Bernard Bodo and Augustin E. Nkengfack	803
Cytotoxic Effect on Cancer Cells and Structural Identification of Phenols from <i>Spatholobi caulis</i> by HPLC-ESI-MSⁿ Dan Lu, Hua He, Bin Wu and Shanjing Yao	809

Continued inside back cover