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Aims

The *Journal of Essential Oil Research* (JEOR) is a scientific journal devoted entirely to all facets of pure and applied studies on essential oils or plant volatiles, excluding those of a purely agricultural or horticultural nature. The main areas of emphasis of the journal are:

Analytical Chemistry
 Biological Activity
 Biotechnology

Chemical Composition
 Chemical Synthesis
 Chemosystematics

Microbiological Activity
 Plant Biochemistry/Biosynthesis
 Toxicology

The *Journal of Essential Oil Research* is international in scope, as can be seen by the composition of the Editorial Board. The goal of this Board is the timely publication of papers of a high standard of technical merit and scientific quality. All manuscripts submitted for publication in the JEOR will be formally reviewed by no less than two members of the scientific community who are regarded as authorities in that field. To be considered as a subject for publication, the manuscript must contain information on the aromatic principles of a plant or its isolate, or must be directed toward furthering our knowledge of the aromatic plant and animal kingdoms. This journal will serve as a forum for the publication of formally refereed manuscripts devoted to the field of essential oils and plant volatiles. Consequently, concise contributions on the experimental or theoretical investigations of some facet of essential oils, aromatic plants, or plant and animal interactions are invited for publication.

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INTRODUCTION TO YOUR GUIDE TO THE ISEO CONFERENCE

The 42nd International Symposium of Essential Oils (ISEO 2011) being held September 11–14, 2011 in Antalya, Turkey is the latest iteration of a symposium series that has been organized annually in Europe for the last 41 years, in order to stimulate cooperation among scientists for the advancement of research and development into the science of essential oils and aroma chemicals. The only other ISEO organized in Turkey (the 28th Symposium) was held in 1997 in Eskisehir, and we are gratified to once again be back in this unique country.

The first ISEO was held in 1969 in Leiden, Netherlands, as an informal meeting of four active scientists in order to discuss common analytical and associated problems concerning the research on essential oils. Participants of that meeting were Prof. Baerheim-Svendsen and Dr. Jan Karlsen, both from University of Leiden, Dr. Hefendehl from the University of Freiburg, and Prof. Karl-Heinz Kubeczka, at that time from the Technical University of Karlsruhe in Germany. In subsequent years, the meetings were held in connection with the annual congresses of the Society of Medicinal Plant Research (GA) until ISEOs were organized independently. The increase in the number of ISEO participants from 1974 in Freiburg, Germany with 15 participants, to 2009 in Savigliano, Italy with 252 participants has been nothing short of remarkable.

This 42nd symposium is expected to attract global attention in the International Year of Chemistry of the United Nations preceded by the 59th International Symposium and Annual Meeting of the Society for Medicinal Plant and Natural Product Research (GA 2011) and Herbal Products in Animal Health and Nutrition Symposium (HERBAN 2011) this past week at this very same venue. This will give most of the participants a feast of natural products chemistry for two weeks in early September, at an excellent venue where business and leisure can be combined for pleasure and satisfaction. Antalya is an attractive spot blending history and culture and a favorite holiday resort with numerous five-star hotels and holiday villages. As you are hopefully experiencing, September is a fine time for Antalya with a favorable and pleasant climate.

Antalya with its rich diverse and fragrant flora is an ideal venue to enjoy the symposium on essential oils. Being a Mediterranean town, it is home to citrus production, and many spice and condiment plants are wild crafted, grown and processed in this region. A strong rose growing and processing spot of Turkey, Isparta is the neighboring province.

The meeting will get together essential oil scientists and aroma therapists working in academia, trade, industry and regulatory affairs dealing with various aspects of essential oils, aromatic plants and aroma chemicals to enjoy a rich, varied and attractive scientific and cultural program.

For the first time in the history of ISEO symposia, abstracts of the meeting being provided to each attendee, gratis, courtesy of *The Journal of Essential Oil Research (JEOR)*. Abstracts were peer-reviewed by the Permanent Committee Members of ISEO before acceptance. We are thankful to *JEOR* for this gesture and hope that the abstracts will both fuel your interest in this year's symposium and find a permanent place on your reference shelf.

We wish ISEO 2011 participants a successful and enjoyable symposium!

Prof. Dr. K. Hüsnü Can Başer
President of the Organising Committee of ISEO 2011

Prof. Dr. Luigi Mondello
Editor-in-Chief, JEOR

PLENARY LECTURES

[PL-001]

The New EU Flavouring Regulation and the Safety Evaluation of Flavouring Substances in the EU

J. C. Demyttenaere

EFFA—European Flavour Association

Abstract

The new flavouring regulation, i.e. Regulation (EC) No. 1334/2008 [1] was published at the end of 2008 and entered into force on Jan. 20, 2009. However the real application was more recent, starting from Jan. 20, 2011. According to this new flavouring regulation, a “List of Flavouring Substances” for use in the EU internal market had to be adopted by the end of 2010 at the latest, but is expected to be published only by the end of this year (2011). In 2004 the European Food Safety Authority (EFSA) began a safety review of more than 2,000 flavouring substances, which is still ongoing. This list (which will become part of a broader “Union List” of flavourings) includes many substances that have been reviewed and recognized as safe at the global level by various regulatory and assessment bodies (e.g. JECFA, US FDA, FEMA GRAS, and the Council of Europe Committee of Experts). However, for a large number of the materials, the outcome of the final evaluation by the EFSA has not been published yet or remains outstanding. This presentation will provide an update on this safety evaluation of flavouring substances by the EFSA (“Where we are today”) and highlight other important aspects of the new flavouring regulation, which have an impact on the flavour business. Specifically, the impact on natural labeling resulting from the difference between the new flavouring regulation and the old “Flavour Directive” 88/388/EC [2] will be discussed from a B2B (business to business) perspective.

Key Words

Flavouring regulation, safety evaluation, flavouring substances list, natural labeling

[PL-002]

Essential Oils from Fungi?

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Abstract

Higher plants show a long history as sources of all kinds of essential oils, while fungi invariably are associated with contamination and spoilage. However, the traditional plant sources suffer from drawbacks, while particularly the most developed fungi, the Basidiomycetes, combine complex biochemistry and easy bio-processing. Their natural habitat forced the development of potent hydrolases and redox enzymes to degrade lignin, the most recalcitrant organic material. Contact of these powerful enzymes with mono- and sesquiterpene hydrocarbons generates mixtures of allylic oxidation products which may be used as commercial flavours and fragrances.

Using deuterio-labeled precursors, novel pathways running through epoxides or endoperoxides were recently found involved in the conversion of β -myrcene to the rose flower constituent perillene (woody, flowery citrus

odour) by the oyster mushroom, *Pleurotus ostreatus*. Ocimene was likewise converted to rose furane (caramel odour). The key enzyme catalyzing the reaction of valencene to nootkatone (grapefruit odour) was identified as a dioxygenase of *Pleurotus sapidus*. This enzyme was isolated, purified, partially sequenced and actively expressed in *E. coli* as a recombinant host. The identity of intact hydroperoxides of valencene as intermediates was proven by microchemical and spectrometric methods.

The biotechnological conversion of easily available precursors to potent natural flavours is industrial reality. However, the lipophilic and cytotoxic terpene hydrocarbons provide particular challenges. The increasing availability of genome data will enable the construction of heterologous hosts expressing combinations of key enzymes of terpene pathways. This may ultimately result in sustainable and inexhaustible biotechnologies to produce “essential oils” using fungal sources.

Key Words

Biotransformation, basidiomycete, β -myrcene, ocimene, perillene, rose furan, valencene, nootkatone, dioxygenase, terpene peroxide.

[PL-003]

Volatile Components of the *Marchantiophyta* (liverworts): Chemical Diversity, Bioactivity and Biotransformation

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Abstract

Liverworts are taxonomically placed between algae and pteridophytes, and there are 6,000 species. They are not damaged by fungi, insects, earthworms or mammals because of the presence of potent pungent and bitter substances. Almost all liverworts possess beautiful cellular oil bodies. Several hundred new compounds have been isolated from liverworts and more than 40 new carbon skeletal sesqui- and diterpenoids have been found in this class, together with new bi- and bisbibenzyls and acetogenins. Except for a few sesqui- and diterpenoids, almost all mono-, sesqui- and diterpenoids are enantiomers of those found in higher plants. The different species in the same genus of *Frullania* produce normal sesquiterpenoid or its enantiomer. Some *Lepidozia* species elaborate both normal sesquiterpenoid and its enantiomer. Many isolated volatile components show characteristic scents, hot taste and bitterness, allergenic contact dermatitis, cytotoxicity against cancer cell lines, antimicrobial and antifungal, insect antifeedant against African army worm and mortality against mosquito larva, neurotrophic and piscicidal activity, inhibition of NO production and plant growth inhibitory activity. The isolated volatile components were biotransformed by several fungi to obtain oxidated and/or hydrogenated compounds which showed different bioactivity from that of the substrates. The chemical diversity of volatile components and their bioactivity and biotransformation are surveyed in this discussion.

Key Words

Liverworts, volatile components, sesquiterpenoids, diterpenoids

[PL-004]

Advanced Chromatographic Techniques for the Analysis of Essential Oils and Related Products

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Abstract

The last decades have witnessed widespread application of multidimensional (comprehensive) chromatographic techniques, capable of affording enhanced resolving power for the separation of multi-component mixtures, with matrix interfering compounds, or trace-level components. Recent advances in both hardware and column technology, easiness of full automation and the likelihood of straightforward hyphenation to mass spectrometry have definitely concurred to make GC- and LC-based separation methods the focus of current research in most research fields. However, the great complexity of essential oils and related products often overwhelms the separation capability of any one-dimensional technique, and along with the increasing amount of information required has strongly stimulated the development of multidimensional (comprehensive) chromatographic techniques.

In this contribution, an overview of the different comprehensive GC and LC approaches is presented, together with selected applications to challenging separation tasks. In such platforms, the use of MS detection represents a third, added dimension, delivering the complementary information needed for confident elucidation of the structures of unknown analytes.

Regarding comprehensive GC, the most recent trends toward the development of a cryogen free, chip-based pneumatic modulator will be presented, while for comprehensive LC, recent developments towards the implementation of UHPLC methods for high efficiency analyses, as well as miniaturization of the system to attain lower limits of detection and solvent/time consumption will be discussed.

Key Words

Comprehensive chromatography, GC x GC, LC x LC.

[PL-005]

High Performance TLC – Get the Picture of Essential Oils

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Abstract

Thin layer chromatography (TLC) has a long tradition of being used as a simple, inexpensive, and flexible tool for rapid identification of complex mixtures. Many students of science have been introduced to the principles of separation science by some form of TLC when experimenting with inks, spinach or other plant juices, or substances squeezed out of citrus peel. Nowadays most people would probably associate “real” chromatographic work with either High Performance Liquid Chromatography (HPLC) or - in the field of essential oils - Gas Chromatography (GC), while TLC is viewed as out dated.

High Performance TLC (HPTLC) is today's approach to planar chromatography which in many aspects can be seen as orthogonal to separation in a column/capillary. Resting on the foundation of well-established science HPTLC is a modern concept including the use of software controlled instrumentation, a rigorously standardized methodology, and validated methods. In contrast to its classical form, HPTLC offers highly reproducible information in the form of electronic images, which make the visual or software aided comparison of multiple samples a very easy task. HPTLC analysis of essential oils can provide pictures, called fingerprints, which are complementary

to GC data and thus provide another window for looking at the same sample with different separation principles and detection modes.

Traditionally classical TLC is described in all monographs for essential (and fatty) oils found in the European Pharmacopoeia as a method of identification. In practice these methods are hardly used. If replaced by HPTLC methods, which are fine-tuned to bring out similarities and differences between samples, identification of essential oils is a simple task. Within about 30 minutes up to 20 samples can be analyzed in parallel. Specific HPTLC fingerprints are not only suitable for reliable identification of a given sample but also enable detection of adulteration with substances that are not easily separated by GC such as fatty oils. Fingerprints may also be useful to establish quality specifications beyond GC profiles to help trace origins, compare multiple batches, screen for artificial mixtures, or distinguish oils obtained from related plant species. HPTLC should therefore be valued as a powerful tool for quality control particularly in the context of cGMPs for products that contain essential oils as raw materials.

Due to the planar separation principle all samples and their separated components remain static on the HPTLC plate once chromatography is finished. This opens the door for various and multiple detection modes, of which capturing images under UV and/or white light with and/or without chemical derivatization is only one. Classical scanning densitometry or image based video densitometry enable quantitative evaluation of separated compounds. The combination with bio assays (effect directed analysis) can help identify the presence of biologically active substances in essential oils. With commercially available interfaces even mass spectrometric identification of compounds on the HPTLC plate is possible. All this makes HPTLC also a great research tool.

[PL-006]

The Role of Volatile Organic Compounds in Plant-Insect Interactions

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Abstract

All plants are able to emit volatile organic compounds (VOCs) and the content and composition of these molecules show both genotypic variation and phenotypic plasticity. The biochemistry and molecular biology of plant VOCs is vast and complex, including several biochemical pathways and hundreds of genes. The primary functions of airborne VOCs are to defend plants against herbivores and pathogens, to attract pollinators, seed dispersers, and other beneficial animals and microorganisms, and to serve as signals in plant-plant communication. In some plants, released VOCs may also act as wound sealers.

Chemically, plant VOCs belong to the large group of terpenoids (homo-, mono-, sesquiterpenoids), fatty acid derived C₆-volatiles and derivatives, phenylpropanoid aromatic compounds (like methyl salicylate, MeSA, and indole), as well as certain alkanes, alkenes, alcohols, esters, aldehydes, and ketones. Today more almost 2000 volatile compounds have been isolated from more than 90 plant families, constituting approximately 1% of all plant secondary metabolites.

The composition of VOCs emitted by plants also depends on the mode of damage such as single wounding, continuous wounding, herbivore feeding, and egg deposition. Some VOCs emitted after insect feeding can serve as repellents to the attacking insect itself as a direct defence, as well as attractants to the natural enemies of the attacking insect as indirect defences. An herbivore-induced VOC blend may comprise more than 200 compounds. In addition to attracting the natural enemies of the egg and larval stages, herbivore-induced plant volatiles (HIPVs) can also decrease the oviposition rates of the attacking herbivores and thus can be considered both direct and indirect defense systems.

This paper describes the current advances in the role of volatile organic compounds in plant-insect interactions.

[PL-007]

South Africa's Aromatic Flora: Reflecting on a Decade of Research

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Abstract

South Africa is considered to be the third most biodiverse area in the world and harbors 10% of the world's total flora. Several aromatic plants reside in this extra-special botanical biome on the southern tip of Africa. Three indigenous species (*Pelargonium graveolens*, *Agathosma betulina* and *Eriocephalus tenuifolius*) have been commercialized and are incorporated into flavor and or fragrance formulations.

In 2000 an ambitious program was initiated to explore the essential oils of South African aromatic plants. Our research has revolved around three integrating themes:

1. *Composition and biological activity*: Taking leads from traditional medicine practices, several plants used by the ethnic groups of South Africa have been studied to provide a scientific basis for their use in healing rites. Synergy has formed an important component of our research and several examples will be presented to illustrate how volatile molecules may interact to enhance activity.

2. *Chemotaxonomy*: The complexity of essential oils make them ideally suited for multivariate data analysis and remains a valuable independent test of taxonomy, which is often based on morphology alone. Using classical GC/MS data in tandem with vibrational spectroscopy, examples from Asteraceae and Lamiaceae will be presented to illustrate how essential oil data has assisted to unravel taxonomic problems.

3. *Bioprospecting*: Flavor and fragrance companies remain under strong consumer pressure to produce novel and innovative products. Like the pharmaceutical industry, the flavor and fragrance industries turn to nature for guidance and inspiration. Several multinational companies have explored the aromatic flora of South Africa, and the tremendous opportunity, as well as the hurdles associated with bioprospecting, will be highlighted.

Although the research topics have been presented as separate entities, our approach has been to integrate the various themes in a multidisciplinary approach to explore and catalogue the composition and biological properties of South Africa's aromatic flora.

ORAL PRESENTATIONS

[O-001]

Stereochemical Structure and Thermal Change of Frankincense Constituents in Hexane Extract

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Abstract

Frankincense has been used as an important fragrance source since ancient times, and its importance is now increasing. One type of frankincense contains diterpenes as main constituents, along with octyl acetate and octanol. We evaluated the odor of frankincense by focusing on the difference in odor between its steam-distilled oil and hexane extract. As a result, the important odor components were found to be incensole and its derivatives, which have not been recognized previously as important odor compounds. Many studies on the structure of incensole derivatives have been reported, but the relative stereochemistry of the structures is not yet clear. We investigated the exact structures of these compounds by NMR spectroscopy. The epoxide of incensole was isolated as pure crystals, and its structure was determined by single-crystal X-ray diffraction. The stereochemical configurations of the other incensole compounds, relative to epoxide, were elucidated through synthetic studies. Furthermore, incensole and its acetate in hexane extracts were found to convert gradually to their epoxides, although the compounds in their pure forms did not undergo this change. These results indicate that incensole epoxides are not original constituents of frankincense, but rather secondary products.

Key Words

Frankincense, incensole, incensole epoxides, single-crystal X-ray diffraction.

[O-002]

Comparative Study of Natural Polymers Encapsulated Fragrances

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Abstract

Comparative study of natural polymers encapsulated fragrances was investigated. Natural polymers such as gelatin and acacia were prepared with and without chitosan to encapsulate fragrances containing peppermint oil using coacervation phase separation and spray drying technique. The different concentrations of fragrances and natural polymers of core to wall ratio of 2:1, 1:1 and 1:2 were studied on percent yield, morphology and odor released properties. The percent yields of gelatin-acacia coacervation coated with and without chitosan as wall

forming were between 37.26 and 39.61, and 10.05 and 25.13, respectively. The morphologies of particles were investigated by scanning electron microscope. The particle sizes were measured by MasterSizer, LA-950V2, Kyoto, Japan. The particle sizes distribution of gelatin-acacia coacervation coated with and without chitosan ranged 13.90-18.55 and 24.24-93.93 μm , respectively. The prolonged odor release properties of fragrances from formulations were evaluated by electronic nose (E-nose). The chitosan coated gelatin acacia coacervation wall forming of core to wall ratio of 1:2 gave the best entrapment efficiency with significantly prolong the odor release when compared to control oil.

Key Words

Gelatin-acacia coacervation, chitosan, electronic nose, fragrances.

[O-003]

Enhanced Collection of Essential Oil Components by Using a Multidimensional GC-prep System

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Abstract

The request for terpene compounds has greatly increased in the last decades due to their capability to enhance/provide pleasant flavour and fragrance. Furthermore, the continuous efforts to find new flavours and fragrances necessitate an effective collection system to provide these compounds from natural sources. GC coupled to preparative system has proven to be a valid tool for the collection of volatile and semi-volatile compounds but often this procedure is time-consuming, difficult to optimize and frequently characterized by low recovery values requiring a high number of collections. Furthermore, in some cases it is very difficult to collect pure compounds, due to the partial or total coelutions, by using low-efficiency wide-bore or packed GC columns. To overcome these problems an MDGC system has been coupled to a preparative system with the purpose of increasing the amount of volatile compounds that can be collected in each single GC run due to their higher sample capacity. The wide-bore columns in both chromatographic dimensions allowed the possibility to inject high sample amounts of neat essential oil, maintaining an acceptable efficiency level. Even if in some cases the sample amount overloaded the primary apolar column, compromising the separation of the target compound, the heart-cut of this fraction on the second dimension polar column allowed the purification of the compound before the collection. This option has proven to be very useful, reducing the relative time and cost. The method has been validated in terms of recovery collection for different amounts of volatile compounds.

Key Words

MDGC, Prep-GC, terpene.

[O-004]

Chemotaxonomic significance of the volatile components from *Thysananthus* and *Porella* species

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Abstract

Thysananthus and *Porella* are liverwort genera classified within class Jungermanniopsida. The genus *Thysananthus* contains about ten species, growing in rain and montane cloud forests throughout the tropics, whereas *Porella* comprises about 60 species with a worldwide distribution. The taxonomy of both liverwort genera based on plant morphology is still confusing. One of the outstanding futures of the Marchantiophyta is their chemical composition, which is characterized by great complexity. This complexity can be used for chemosystematic classification of liverworts (4). The aim of this work was the exploration of chemosystematic relevancies within four *Thysananthus* species from Thailand and Malaysia, as well as twenty-nine *Porella* species on the basis of their sesqui- and diterpenoids composition. The received data were statistically verified through PCA and CA analysis indicating variability of the chemical composition depending on liverwort species. The analysis of the volatile components of *Thysananthus* species showed that among all species *T. retusus* is most homogeneous, and its chemical composition is quite different from the other species. Noteworthy is the chemical heterogeneity of *T. convolutus*, which correlates with the degree of dentation of leaves and underleaves. *Porella* is a liverwort genus which is chemically a very diverse group. The chemical data suggest that the pungent chemotype which comprises *P. arboris-vitae*, *P. canariensis*, *P. obtusata*, *P. gracillima*, *P. roellii*, and *P. vernicosa* deserves recognition as a separate section of *Porella* and named *Porella* section Vernicosae Ludwiczuk, Gradstein, Nagashima & Asakawa. These data are with agreement with morphological and DNA-based studies.

Key Words

Liverworts, chemosystematics, *Thysananthus*, *Porella*, sesquiterpenoids, diterpenoids.

[O-005]

Chemical Compositions and Antifungal Activity of the Essential Oils of *Cinnamomum* spp. of Sarawak, Malaysian Borneo

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Abstract

The essential oils of the bark, twig and leaves of five *Cinnamomum* spp. (*C. burmanii* Blume, *C. parthnoxylon* (Jack) Meisn, *C. microphyllum* Ridl., *C. javanicum* Blume, *C. zeylanicum* Blume) were obtained by hydrodistillation methods and analyzed using GC and GC/MS. The oils were evaluated for their antifungal activity against five dermatophytes (*Trichophyton rubrum*, *T. mentagrophytes*, *Microsporum canis*, *M. gypseum*, *M. audouini*) and four strains of yeast (*Candida albicans*, *C. tropicalis*, *C. glabrata*, *C. parapsilosis*). The major constituent of the bark and twig oil of *C. javanicum*, *C. parthnoxylon* and *C. burmanii* was methyl(E)-cinnamate, while for *C. zeylanicum* and *C. microphyllum* the major constituent was cinnamaldehyde. For the leaf oils, benzyl benzoate was the major constituent in all species studied except for *C. Zeylanicum*, where eugenol appears as the major constituent. In order to evaluate the effectiveness of the antifungal activities of the essential oil, the antifungal activities of 10 standard compounds which were dominant in the oils of *Cinnamomum* spp. were also investigated. Most of the oils displayed overwhelming antifungal activities with minimum inhibitory concentration below 0.63 µg/µL. The essential oils of *C. zeylanicum* and *C. parthnoxylon* showed the strongest activity against all the fungi tested, with minimum inhibitory concentration of 0.04 µg/µL. The presence of cinnamaldehyde, eugenol, methyl(E)-cinnamate and benzyl benzoate as the major constituents in the species studied might be responsible for the observed antifungal activities either individually or in combination since the standard compounds used also showed strong antifungal activities againsts all the dermatophytes and yeast tested.

Key Words

Cinnamomum spp., essential oil, antifungal activity, cinnamaldehyde, eugenol, methyl(E)-cinnamate, benzyl benzoate.

[O-006]

Development of a Botanical Termiticide—Production, Chemistry, Bioactivity and Registration of *Eremophila mitchellii* Essential Oil

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Abstract

Eremophila mitchellii Benth. wood is resistant to termite attack in its natural environment; however, the properties conferring such resistance were unknown. Extensive research has established that the essential oil from the heart wood of this species provides much of the observed resistance. A detailed analysis of the wood oil was carried out by a combination of GC-FID, GC/MS, LCMS and NMR spectroscopy. After fractionation by preparative HPLC, eight components of the wood oil were characterized and accounted for >80% of the oil. There are four main compounds (composing more than 70% of the whole oil): eremophilone, 8-hydroxy-1(10)-

dihydroeremophilone, 9-hydroxy-7(11),9-eremophiladien-8-one and 9-hydroxy-1,7(11),9-eremophilatrien-8-one. A survey of wild populations of *E. mitchellii* established the oil's physical and chemical properties, which were then used to establish a declaration of composition and quality control standards for production batches of the oil. The insecticidal properties of *E. mitchellii* oil were initially evaluated against two species of termites, *Nasutitermes exitiosus* and *Coptotermes acinaciformis*. The steam-distilled wood oil exhibited toxicity, repellency and antifeedant activity against both species. Bioassay-guided fractionation determined that only two compounds, eremophilone and 8-hydroxy-1(10)-dihydroeremophilone, were found to be the key actives responsible for the oil's observed activity. Mammalian and environmental toxicity data was also required for the successful registration of eremophilone oil as an active constituent with the Australian Pesticide and Veterinary Medicines Administration. This is the first natural termiticide to receive such registration in Australia.

Key Words

Eremophila mitchellii, termiticide, composition, mammalian toxicity, environmental toxicity.

[O-007]

Effect of Warm Air Flow and Reduced Pressure on Antibacterial Activity of Essential Oil Vapors

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Abstract

Among biological effects of essential oils (EOs), *in vitro* antibacterial activity is one of the most studied and proven. However, their practical utilization is still quite rare, partially because of negative side effects like phytotoxicity and strong odors. Our aim was to investigate warm air flow and reduced pressure as possible cofactors for reduction of time and dose of EOs vapors treatment and hence mitigation of negative side effects. Three EOs, namely *Cinnamomum zeylanicum*, *Origanum vulgare* and *Ocimum basilicum*, were tested *in vitro* against three bacteria: *Staphylococcus aureus*, *Salmonella enteritidis* and *Pseudomonas aeruginosa*. The results show that warm air flow containing EO applied for very short time (2 x 15 s; 40 °C, 0.4 m/s) is still able to inhibit bacterial growth, moreover in concentrations even 40 times lower in comparison with microatmosphere method. Similarly, application of EO vapors under reduced pressure (1.7 kPa) was able to significantly shorten time needed for growth inhibition of tested bacteria, e.g. from 60 min under normal pressure, to 5 min under reduced pressure in the case of *O. vulgare* against *S. aureus* and *S. enteritidis*. In conclusion, except *O. basilicum*, which was not active at all, activity of *C. zeylanicum* and *O. vulgare* EO vapors was improved by both warm air flow and reduced pressure against bacterial strains tested.

Key Words

Antimicrobial, fumigant, fumigation, vapour, vacuum, mild heat.

Repellent Activity of Essential Oils of Two *Monarda* Species against *Aedes aegypti*

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Abstract

Mosquitoes are vectors of pathogens that can cause serious diseases such as Malaria, Dengue Fever, Yellow Fever, Rift Valley Fever, and Chikungunya. There are no vaccines available for many of these diseases, and management and control of vector populations, including personal protection, are the primary means used to protect human and animal populations. Insecticides, especially pyrethroids, have been the primary chemicals used to control insect pests in agriculture and public health. However, frequent use of insecticides can lead to non-target effects and the development of insecticide resistance. Therefore, there is a continuing need to explore natural products that are safe and can be used effectively for managing populations of these vectors. Our research program is aimed at identifying natural insecticides with low mammalian and environmental toxicity. We evaluated 21 different essential oils as topical repellents against *Aedes aegypti* L. using a “cloth patch assay” test using DEET as a standard. The constituents of these essential oils were analyzed by GC and GC/MS using the in-house Baser Library. *Monarda bradburiana* Beck and *M. fistulosa* L. oils showed the highest repellent activity with minimum effective dosages of 0.055 ± 0.036 mg/cm² and 0.078 ± 0.027 mg/cm², respectively. The minimum effective dose of DEET was 0.039 ± 0.014 mg/cm². Systematic bioassay-guided fractionation of both *Monarda* essential oils was performed to identify active repellent compounds using an *in vitro* Klun & Debboun (K&D) module bioassay system. Individual pure compounds were tested with both cloth patch and K&D module bioassays.

Key Words

Repellent activity, *Aedes aegypti*, essential oil, personal protection, *Monarda bradburiana*, *Monarda fistulosa*.

[O-009]

Analytical Study of Commercial Frankincense Essential Oil: the Quest for the Odor Active Constituents

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Abstract

Frankincense, also known as *olibanum*, is a gum resin obtained from trees of the *Boswellia* species (Burseraceae). In Occident, it is mainly used as incense burned during religious ceremonies, but in Asia, it is also a popular remedy in traditional medicines. The chemical composition of frankincense is rather complex and has been described in many publications. Apart from triterpenoids, the volatile constituents represent about 5-15% of the resin, which can be hydrodistilled to furnish the frankincense essential oil, an important raw material in the flavour and fragrance industries, very appreciated for its nice characteristic spicy base note. The composition of this essential oil depends on the *Boswellia* species, and is usually dominated by common monoterpenes (e.g. alpha- and beta-pinene, and limonene) and/or octyl acetate, accompanied by other mono-, sesqui- and diterpenic compounds. Indeed, the diterpenoids incensole and its acetate are restricted to the *Boswellia* genus and are often used as biomarkers for quality control of frankincense. A recent review in *Flavour and Fragrance Journal*, on the volatile constituents of frankincense pointed out that “despite the wide range of data reported on frankincense resin and extracts, no information regarding the chemosensory contribution of individual constituents to the specific odour of frankincense resin can be drawn.” Therefore, in order to bring some information on this topic, we performed a detailed investigation on a commercial sample of somalian frankincense essential oil by a combination of distillations, preparative chromatographies, GC, GC/MS and GC/O experiments.

Key Words

Frankincense, *Boswellia* species, odor active compounds.

Enhancement of Antifungal Activity of Clove Oil against *Aspergillus niger* on the Surface of Rubberwood (*Hevea brasiliensis*) Using Low-pressure RF Plasma

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Abstract

An attempt to effectively control growth of *Aspergillus niger* commonly found on the surface of rubberwood (*Hevea brasiliensis* (Willd. ex A. Juss.) Mull. Arg.) by using low concentration of clove oil (10 µL/mL) both *in vitro* (broth dilution method) and *in vivo* (dip method) was enhanced by means of low-pressure RF plasma at various powers (50, 100, 150 and 200 W) for 10 min. Colony germination after low-pressure plasma treatment was evaluated *in vitro* on agar mixed with clove oil. In the *in vivo* assays, rubberwood specimens were dip-treated in clove oil before exposition to low pressure plasma. Control treatments were done in the same way but without plasma treatment. The results showed that growth of *A. niger* on agar mixed with clove oil was completely inhibited using low-pressure plasma treatment at 200 W for 10 min. Furthermore, plasma treatment at 200 W for 10 min of rubberwood specimen dip-treated with clove oil could extend the protection against mold growth on the surface of rubberwood from 14 days (control) to at least 12 weeks at 25°C. This study has demonstrated a good potential of using low-pressure plasma treatment to enhance antifungal activity of essential oils to control molds on wood.

Key Words

Clove oil, *Aspergillus niger*, rubberwood, plasma.

[O-011]

The Results of Susceptibility Testing of *Staphylococcus aureus* by Broth Microdilution Method Are Strongly Affected by Thymoquinone Vapours

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Abstract

Antimicrobial activity of essential oils and their compounds has been studied intensively and also the interest in the activity of these volatiles in vapour phase has been increasing recently. However, there are no reports considering the influence of vapours on the results of broth microdilution testing method (MD). In our study, we demonstrated that thymoquinone (TQ) vapours affect the results of antimicrobial susceptibility tests obtained by the standard MD. The minimum inhibitory concentration (MIC) values of p-cymene against various strains of *Staphylococcus aureus* were almost equal to that of TQ (4-8 mg/L) when tested simultaneously on one microtiter plate (MP), whilst no antimicrobial activity of p-cymene was detected when tested separately. TQ was therefore examined for possible interference with adjoining wells on MP. A series of twofold dilutions was prepared and each concentration was added in the first column of separate MP. The remaining columns contained Mueller-Hinton broth as positive control. The MIC of TQ against *S. aureus* ATCC 29213 obtained by this procedure was 16 mg/L and surprisingly, TQ at concentrations of 32-512 mg/L caused inhibition of bacterial growth in one to four adjoining wells containing no antimicrobial agent. To prevent this affection, we designed a modified MD method with application of sealing membrane. Our results indicate that antimicrobial testing of volatiles with other compounds on one MP using the standard MD can lead to false positive results. This finding could be of crucial importance for further testing of volatile compounds by MD.

Key Words

Thymoquinone, vapour, microdilution, *Staphylococcus aureus*, essential oil, volatile, vapor, p-cymene.

[O-012]

Involvement of Glutamatergic Pathway and Vanilloid System in Antinociception Induced by the Essential Oil of *Boesenbergia pandurata* in Mice

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Abstract

The transient receptor potential vanilloid-1 (TRPV1) of the vanilloid system mediates chemical and thermal pain, whose stimulation elicit glutamate release. We have demonstrated that the essential oil of *Boesenbergia pandurata* Schltr. (BPEO) possesses antinociceptive activity against thermal- and chemical-induced nociception. Therefore, the possible involvement of the glutamatergic or vanilloid system in antinociception induced by BPEO in both models of nociception was explored. In order to determine whether BPEO antagonizes capsaicin-induced pain in the mouse paw, a capsaicin-induced paw licking test was performed. Animals were pretreated (p.o.) either with vehicle (10 mL/kg), capsazepine (0.17 mmol/kg) or BPEO (10, 30, 100 and 300 mg/kg) 1 h before capsaicin (1.6 µg/paw, 20 µL) injection. Animals were observed individually for 5 min post capsaicin injection and the amount of time the animals spent licking the injected paw was recorded with a chronometer and considered as an indication of nociception. In the glutamate-induced paw licking test, animals were administered (p.o.) either with vehicle (10 mL/kg) or BPEO (10, 30, 100 and 300 mg/kg) 1 h before glutamate (10 µmol/paw, 20 µL) injection. Mice were observed individually from 0 to 15 min following glutamate injection and the nocifensive responses recorded. Results from this study indicated that the administration of BPEO significantly reduced the pain-related behaviours induced by intraplantar administration of both capsaicin and glutamate in a dose-dependent manner. These results suggested that BPEO-induced antinociceptive activity was possibly related to its ability to block or inhibit the vanilloid receptors, and thus the release of glutamate into the system.

Key Words

Antinociceptive, glutamatergic, vanilloid, essential oil, *Boesenbergia pandurata*, mice.

[O-013]

Composition and Antimicrobial Activities on Cinnamon Essential Oil

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Abstract

Cinnamon (*Cinnamomum aromaticum* Nees) is a popular and traditional Chinese spice, extensively consumed for cuisine and medicinal usages. The essential oil was extracted from dried cinnamon bark through steam distillation. GC/MS analysis showed that major tentative chemical constituents of essential oil was *trans*-cinnamaldehyde (24.07%), α -copaene (11.31%), 3-methoxycinnamaldehyde (11.08%), δ -cadinene (6.51%), α -muurolene (5.20%) and verticilol (3.62%). Antimicrobial experiments revealed that essential oil had effective and selective antimicrobial activities against four bacteria (*Bacillus subtilis*, *Escherichia coli*, *Pseudomonas fluorescense*, *Staphylococcus aureus*) and four fungi (*Alternaria alternata*, *Fusarium semitectum*, *Penicillium expansum*, *Trichothecium roseum*) *in vitro*. The essential oil was more effective against fungi than against bacteria.

Key Words

Cinnamon, essential oil, chemical constituents, antimicrobial activities.

[O-014]

Ecological Production of Lavenders in Cuenca Province (Spain): a Study of Yield Production and Quality of the Essential Oils

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Abstract

We present a description of the ecological agriculture of different lavenders in the Cuenca province (Spain). We focused on the important aromatic crops of *Lavandula angustifolia* Mill. (lavender) and on three varieties of *Lavandula x intermedia* Emeric ex Loisel. (lavandin) named abrial, grosso and super. A description of yield production and essential oil qualities obtained in our experimental fields are presented. Lavandin var. super showed the best essential oil quality, with good yield production as well. The current market of fragrant and medicinal plants claims raw materials with confirmed quality, with good practices of harvesting, transformation and manufacture by means of ecological production.

Key Words

Ecological production, lavandin, lavender, essential oil, linalool, linalyl acetate.

Biologically Active Volatile Components of Some French Polynesian, Indonesian and Japanese Liverworts

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Abstract

Oil bodies of liverworts have been known to contain lipophilic terpenoids and aromatic compounds, several of which show interesting biological activities. In the course of study of biologically active metabolites from liverworts, we found that the crude extracts of the French Polynesian *Mastigophora diclados*, unidentified French Polynesian and Indonesian *Frullania* species and Japanese *Porella perrottetiana* indicated cytotoxicity against HL-60 and KB cell lines. *M. diclados* also exhibited radical scavenging activity on DPPH and antimicrobial activity against *Bacillus subtilis* and *Staphylococcus aureus*.

1) *M. diclados* indicated the presence of a new (+)-drimenol together with five known herbertane sesquiterpenoids, (-)-alpha-herbertenol, (-)-herbertane diol, herbertane dimers, mastigophorene A, (-)-mastigophorene C and (-)-mastigophorene D, and (-)-ent-pimara-8(14),15-dien-19-oic acid and two eudesmanolides, (-)-diplophyllolide A and (-)-diplophyllin.

2) Unidentified *Frullania* species (French Polynesian) produced germacranolides, tulipinolide and costunolide.

3) Unidentified *Frullania* species (Indonesian) elaborated 3-alpha-[4'-methoxybenzyl]-5,7-dimethoxyphthalide, 3-alpha-[3'-methoxy-4',5'-methylenedioxybenzyl]-5,7-dimethoxyphthalide, 3-methoxy-3',4'-methylenedioxybibenzyl and 2,3,5-trimethoxy-9,10-dihydrophenanthrene.

4) *P. perrottetiana* biosynthesized 7-oxo-pinguisenol-12-methyl ester, perrottetianal and 4-alpha,5-beta-epoxy-8-epi-inunolide.

All of the herbertanes showed cytotoxicity against HL-60 and KB cells at 1.4-12.8 microgram/mL and radical scavenging at 1.9-23.2 microgram/mL and antimicrobial activity against both bacteria at MIC 2-100 microgram/mL.

Key Words

Liverworts, cytotoxicity, radical scavenging, antimicrobial.

[O-016]

Volatile Components from the Selected Japanese Liverworts: a Chemosystematic Approach

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Abstract

Classification of liverworts is very difficult because of their small gametophytes. It has been demonstrated that the secondary metabolites, such as lipophilic terpenoids, aromatic compounds and acetogens in their cellular oil bodies can assist in their taxonomic differentiation. The pattern of terpenoids and aromatic compounds often depends not only on the developmental stage, season and altitudinal distribution, but also on sexual forms of the same species and collection from the different locations. Chemical knowledge of liverworts might serve to delineate not only chemical but evolutionary relationships within the Marchantiophyta at the genus or family level. At present, more than 500 species of liverworts were collected in Japan and each species was crushed and extracted with ether, followed by filtration through a Pasteur pipette packed with silica gel and evaporation of solvent to obtain the crude oil which was analyzed by TLC and GC/MS. In the present paper, we report chemosystematic approach to the Aneuraceae, Fossombronia, Pelliaceae, Hymenophytaceae and Metzgeriaceae belonging to Metzgeriales; Jungermanniaceae, Lophoziaaceae, Gymnocoleaceae, Plagiochilaceae, Scapaniaceae, Isotachidaceae, Trichocoleaceae, Ptilidiaceae, Balantiopsilaceae, Schistochilaceae, Lepidoziaceae, Herbertaceae, Radulaceae, Pleuroziaceae, Frullaniaceae belonging to Jungermanniales; and Aytoniaceae and Marchantiaceae belonging to Marchantiales. Homomonoterpenoids, 1,4-dimethyl-azulene, barbatane, herbertane, brasilane, drimane, myltaylane, pacifigorgiane, pinguisane, 2,3-secoaromadendrane, striatane, trifarane, sacculatane, fusicicane, dolabellane, and bibenzyls are significant chemical markers of each genus and family.

Key Words

Liverworts, chemosystematics, Metzgeriales, Jungermanniales, Marchantiales.

[O-017]

Isolation of Enantiomeric Compounds from Essential Oils by Preparative Gas Chromatography-Fraction Collector System (PGC-FCS) and Evaluation for Biological Activities

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Abstract

The separation of a few milligrams of a pure substance from a complex mixture of biological origin is often a matter of paramount importance in studies of volatile metabolites. The aim of our studies was isolation and identification of volatile constituents with subsequent investigation for their biological activity: antibacterial, anticandidal, and antifungal. Specific applications studied in our work included the separation of important volatile enantiomeric compounds from essential oils using a PGC-FCS. The most striking advantage of a temperature programmed separation by PGC-FCS was the simultaneous separation of up to six target compounds from the mixture. This study constituted part of TUBITAK (The Scientific and Technological Research Council of Turkey) project on enantiomeric compound isolation from essential oil by a Preparative Fraction Collector (PFC) system.

The increasing demand for the separation and production of enantiomerically pure compounds has become one of the most important analytical tasks that we perform. Due to the enormous separation power of capillary gas chromatography, we isolated terpene enantiomers from selected essential oils with high purity that ranged 95-99%. This system allowed us to separate and recover sufficient quantities of individual target terpenoids, which were further separated using various chiral columns. After separation, we subjected purified compounds to biological assays for antifungal activity against plant pathogens, anticandidal and antibacterial activities using agar dilution/diffusion, micro-dilution broth assays in order to determine the biological activity of each isolated compounds.

Key Words

Essential oil, enantiomer, isolation, preparative GC, biological activity.

POSTER PRESENTATIONS

[P-001]

Adaptation of Uzbekistan Dill Plant in Egypt Environment

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Abstract

Adaptation and comparison study was done on the two varieties of dill plant (*Anethum graveolens* L.), Local and Uzbekistan (243) during two successive seasons 2009–2010 in the farm of the Horticulture Research station – El-Kanater El Khairia. The data showed that Uzbekistan variety excelled the Local one in vegetative growth (plant height, number of branches, fresh and dry weights of aerial parts of plant). Also, it predominated in number and weight of umbels, and further it gets flowers a month early than the local one. According to the volatile oil extracted from the different cuts and seeds of two varieties, the data indicate the Uzbekistan variety exceeds in oil percentage the local one. GC analysis shows that the chemical composition of the volatile oil from herb and seed contain carvone, linalool, myrcene, limonene, and caryophyllene. The data also illustrated that Uzbekistan variety contains a higher percentage (40.8%) of carvone, while the local variety contains 35.2%.

Key Words

Uzbekistan dill, adaptation, Egypt environment, GC analysis.

[P-002]

Supercritical CO₂ Extraction of Turkish Bergamot Peel Oil

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Abstract

Bergamot (*Citrus bergamia* Risso et Poit.) is cultivated on the coast of the Mediterranean region of Turkey. Bergamot peel oil is widely used in the food, cosmetics and pharmaceutical industries across the world. In this study, the bergamot samples were collected from southern Turkey (Antalya) in December 2010 and their peel oil was obtained by SCFE method. The effect of operation conditions was observed in a series of experiments at 313.2 K, 323.2 K and 333.2 K temperatures, and 90, 100 and 110 Bar pressures, respectively.

The compositions of the oily extracts have been analyzed by gas chromatograph (GC) and gas chromatography/mass spectrometer (GC/MS). In total, 51 components have been identified in the oil. The main monoterpenes of the oil were limonene (16.43%), β -pinene (2.1%), myrcene (0.9%). The major sesquiterpenes were (E)-caryophyllene

(0.4%), β -bisabolene (0.3%) and *trans*- α -bergamotene (0.3%). The most important oxygenated components of the oily extract were found as follows: aldehyde components; neral (0.3%); alcohol components; linalool (43.6%) and ester components; linalyl acetate (27.9%), neryl acetate (0.9%), and geranyl acetate (0.5%). Overall hydrocarbon contents of the oily extract obtained by the SCFE method were found to be 25.3%. The contents of monoterpenes and sesquiterpenes of the SFE extract were found as 24.3% and 1.1% respectively. The of sum oxygenated components was 74.5%, while the other groups of compounds were as follows: carbonyl compounds (0.5%), alcohols (44.3%) and esters (29.7%).

Key Words

Bergamot peel oil, *Citrus bergamia*, Rutaceae, essential oil composition, supercritical CO₂ extraction.

[P-003]

Extraction and Chemical Components on Essential Oil from Kushui Rose

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Abstract

Kushui rose is a hybrid of *Rosa sertata* Rolfe and *Rosa rugosa* Thunb., and occupies more than 60% of rose production in China. The major region of production distributes in the Yongdeng county of Gansu province in northwest China. The plants are adapted to the sandy region with long and dry growing season and have been cultivated over 200 years. The essential oil is rich in citronellol and geraniol, a significant difference from that of *Rosa damascena* Mill., which was extracted by steam-distillation. The optimum conditions were as follows: half opening flowers treated with 20% NaCl, distilled for 3 h, distillation rate at 250 mL/h, the ratio of liquid to solid 4:1. Under these conditions, the yield was up to 0.03984%. The chemical constituents of the essential oil were identified by GC/MS. Among the small quantity of detected volatiles, there were 26 higher alcohols, 14 esters, 2 ethers, 15 aldehydes and ketones, 9 alkanes, 5 terpenes and 1 furan. Higher alcohols made up about 77.37% of the total level and this group was mainly composed of citronellol (70.54%) and geraniol (14.99%). Esters made up about 14.37%, of which citronellyl acetate made up 75.71% and geraniol acetate 11.13%. Ethers made up about 4.26%, of which eugenol methyl ether made up 96.24%. Alkanes made up 3.07% and this group was mainly composed of 1-hexadecylene (63.19%) and pentadecane (16.61%). Aldehydes and ketones made up 1.15% and terpenes 0.40%. Considering all the volatiles detected, higher alcohols and esters are the main contributors to the essential oils.

Key Words

Kushui rose (*Rosa sertata* $\times$ *Rosa rugosa*), essential oil, extraction, chemical components.

[P-004]

Comparative Essential Oil Composition of Two Subspecies of *Teucrium stocksianum* Boiss. from Iran

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Abstract

The hydrodistilled essential oils obtained from aerial flowering parts of *Teucrium stocksianum* subsp. *stocksianum* (TSS) and *T. stocksianum* subsp. *gabrielli* (TSG) from Iran were analyzed by capillary GC and GC/MS and identified. The oil analysis of two subspecies led to the identification of 65 compounds that represented 93.3% and 95.1% of the total oil compositions, respectively. Sesquiterpenoids (52.9%) constituted the main compounds in the essential oil of TSS represented mainly by *cis*-sesquisabinene hydrate (12.0%) followed by epi- β -bisabolol (6.6%), guaialol (5.4%) and β -eudesmol (4.4%), whilst monoterpenoids (61.2%) were found to be the major proportion of the oil composition of TSG, represented by α -pinene (23.0%), β -pinene (13.0%), myrcene (6.3%) and sabinene (6.3%). The principal component in both taxa (TSS and TSG) was α -pinene (22.0% and 23.0%) and β -pinene (6.5% and 13.0%), respectively. epi- α -Cadinol, myrcene and sabinene, which were detected as principal compounds of TSG, were characterized in lower amounts (< 1.5%) in the oil of TSS. Seven components were identified in the oil of TSS corresponding to 25.9% of total oil which was totally absent in the oil of TSG, out of which *cis*-sesquisabinene hydrate (12.0%), guaialol (5.4%) and β -eudesmol (4.4%) were in considerable amounts. In the previous study on the oil composition of *T. Stocksianum*, camphene (20.6%) was reported to be the main component and this may be attributed to the presence of intraspecific chemotypic differentiation.

Key Words

Teucrium, essential oil, α -pinene, epi- α -cadinol, Iran.

[P-005]

Chemical Composition of Essential Oils of *Pinus peuce* Griseb. from Macedonian Flora

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Abstract

Pinus peuce Griseb. (Pinaceae), commonly called Macedonian pine or Balkan pine is an endemic conifer which is spread in the southern and western parts of the Macedonian territory. This conifer is native to Baba Mountain (Pelister) and Nidze, as well as on Shara Mountain where it can be found in small population. The essential oils from the needles, branches without needles and branches with needles were obtained by hydrodistillation in a Clevenger apparatus for 4 h, using 20 g plant material and ranged in yields 0.3-1.0 %, 1.3-1.7 % and 0.7-1.3 %, respectively. The chemical composition of the essential oils was analyzed by gas chromatography system equipped

with FID detector and mass spectrometer, as well as capillary flow technology. HP-5ms capillary column was used. Onehundred-seven components were identified. Among different classes of components that are present in *Pinus peuce* essential oils, the most abundant constituents were terpene hydrocarbons, both monoterpenes and sesquiterpenes. The major components were monoterpenes α -pinene (10.1-39.9%), β -pinene (3.6-16.4%), limonene + β -phellandrene (1.9-35.3%) and bornyl acetate (1.4-14.4%) and sesquiterpenes *trans*-(E)-caryophyllene (1.2-8.4%) and germacrene D (3.0-22.9%). Nowadays, there are no evident data that relate to chemical composition of essential oils of *Pinus peuce* from Macedonia, and but few references are concerned with the composition of the essential oils isolated from needles and branches from *P. peuce* that grows on the territory of Greece, Serbia and Montenegro.

Key Words

Pinus peuce, Macedonian pine, essential oil, gas chromatography/mass spectrometry.

[P-006]

Chemical Characterization of *Aloysia gratissima* (Gillies & Hook) Tronk. Essential Oil (Verbenaceae) from Brazil

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Abstract

The Verbenaceae family has 100 genres that grow in tropical and subtropical regions. *Aloysia gratissima* is a species belonging to this family and it is popularly known as “Alfazema-do-Brazil.” It presents with aromatic and medicinal properties as stomachic, anti-cancer, anti-rheumatic and anti inflammatory activity. The aim of this work was to study the chemical composition of the aerial parts (leaves and inflorescences) from *A. gratissima* essential oil from Brazil (Southeast). Leaves and inflorescences of *A. gratissima* were collected in a private property in Paty-do-Alferes district (Rio de Janeiro State) and dried at room temperature ($28^{\circ}\text{C} \pm 2$) in shade conditions. Essential oil was extracted by hydrodistillation (Clevenger-type apparatus, 4 h) and analyzed by GC/MS. Identifications of compounds were made by matching their mass spectra and Kovat's indices values with known compounds reported in the literature. Seventeen compounds were found in this oil. High contents of β -pinene (25.7%) and *trans*-pinocamphone (16.4%) were observed. These compounds were cited in literature as present in *A. gratissima* essential oil, but some articles on *A. gratissima* samples collected in other countries show sabinene and β -caryophyllene as the main compounds. These results show that Alfazema-do-Brazil can present different chemical compositions, likely because of influence of climatic and geographical conditions, or it can be evidence that the sample from Paty-do-Alferes would be considered a different chemotype of this species.

Key Words

β -pinene, *trans*-pinocamphone, bioprotection, Alfazema do Brasil, *Aloysia gratissima*.

[P-007]

Chemical Differences in the Volatiles of Three Varieties of Wild Garlic (*Allium vineale* L, Alliaceae)

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Abstract

The medical use of some of the plants of the genus *Allium* L. has been well known since ancient times and its therapeutic use is approved. Generally, it is concerned that health-related functions are mostly attributed to the rich content of many sulfur-containing compounds that produce a characteristic flavour formed during storage and processing. However, wild growing *Allium* species are not investigated in detail from any point of view. With respect to this, in the present study the chemical composition of volatile fractions of the bulb of *A. vineale* L. var. *vineale*, *A. vineale* L. var. *compactum* (Thuill.) Lej. & Courtois and *A. vineale* L. var. *capsuliferum* (Koch) Cheshmednev are investigated. The chemical composition of volatiles formed after cutting of bulbs is analyzed by GC/MS with static headspace sampling. In *A. vineale* var. *compactum* the main compounds were methyl allyl disulfide (27.8%) and allyl disulfide (26.6%), and in *A. vineale* var. *capsuliferum* allyl disulfide (22.8%), methyl allyl disulfide (20.6%) and 1-propenyl disulfide (17.2%). In *A. vineale* var. *vineale* the main compound was allyl disulfide (32.2%). Although the chemotaxonomic significance of detected compounds is demonstrated, the investigated species could be used as potential substitutes for garlic, for which a number of complications (prolonged bleeding, side effects including halitosis, nausea, hypotension, headache and bloating) and drug-herb interactions with hypoglycaemics, cardiovascular medications and MAO inhibitors have been reported.

Key Words

Allium vineale, varieties, volatile compounds, headspace-GC/MS.

[P-008]

Hairy Root Cultures and the *In vitro* Production of Volatiles: an update

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Abstract

Hairy roots are differentiated in *in vitro* cultures obtained by plant infection with the soil bacterium *Agrobacterium rhizogenes*. The obtained transformed roots are fast growing systems, with a stable production capacity, a larger biomass increase than their cell suspension culture counterparts, and the advantage of growing without exogenous phytohormones. Over the last sixteen years, our work has focused mainly on the volatiles production by hairy roots of yarrow (*Achillea millefolium* L.), anise (*Pimpinella anisum* L.), dill (*Anethum graveolens* L.) and lovage (*Levisticum officinale* W.D.J. Koch), and more recently foxglove (*Digitalis purpurea* L.), potato (*Solanum*

tuberosum L.) and tomato (*Solanum lycopersicum* L.). On a regular basis, hairy root cultures are kept in batch culture in darkness, or different photoperiod conditions, at 24°C, 80 rpm., and subcultured every month. Separately or combined, the culture medium composition, nitrogen sources and ratios, illumination conditions, addition of precursors or elicitors, induction of regenerants, culturing in bioreactor and in two-phase systems and biotransformation have been explored in order to increase and/or modify the hairy root cultures volatiles production. Hairy roots biosynthetic ability is correlated with the “rooty” phenotype, showing higher yields and both qualitative and quantitative differences between their volatiles and those from parent plant roots, or other parent plant parts. Currently, studies are being carried out to establish nematode-specific hairy root co-cultures, aiming to create phytochemical testing models for nematocide research.

Key Words

Hairy roots, volatiles, plant biotechnology.

[P-009]

Comparison of Essential Oil Composition of *Thymus* Species by Ultrasound-assisted SPME Method with a New Nanoporous Fiber

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Abstract

Nanoporous silica (LUS-1) particles were silylated with hexamethyldisilazane (HMDSA) to be used as a highly porous fiber coating material for solid-phase microextraction (SPME). The nanomaterial was immobilized onto a copper wire by the use of epoxy glue for fabrication of the SPME fiber. An ultrasound assisted SPME method with the nanoporous HMDSA-LUS-1 fiber was applied to the study of the essential oil composition of *Thymus pubescens* Boiss. et Kotschy ex Celak, *Thymus daenensis* subsp. *lancifolius* Celak and *Thymus kotschyanus* Boiss & Hohen. The first sample was collected from Aleshtar, Lorestan province, Iran, and the two others from Aligoodarz, Lorestan province, Iran. The samples were dried and powdered and 0.5 g of each was irradiated by ultrasound radiation in a thermostated (70°C) water bath. The volatile components were collected by the nanoporous fiber from the sample headspace in a capped glass vial and directly injected into a GC/MS injection port for analysis. A simplex method was used for optimization of four different parameters affecting the efficiency of the extraction. The number of components identified by the proposed method was 10 and those identified by a commercial PDMS fiber and a hydrodistillation technique were 14 and 16, respectively. However, the efficiency of the nanoporous fiber for extraction of important biocides such as carvacrol and thymol was significantly higher. Carvacrol constituted 90.7%, 48.1% and 32.9% of the adsorbed components on the nano fiber and thymol constituted 0.26%, 28.9 and 62.8% of them for the three thymus samples, respectively.

Key Words

Nanoporous silica, solid-phase microextraction, *Thymus pubescens*, *Thymus daenensis* subsp. *lancifolius*, *Thymus kotschyanus*.

[P-010]

Skullcap (*Scutellaria lateriflora* L.) Essential Oil Composition from North American and Australian Provenances

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Abstract

The North American skullcap (*Scutellaria lateriflora* L., Lamiaceae) is used in phytotherapy as a mild sedative and anti-spasmodic agent. It has a long history of substitution involving other *Scutellaria* species and hepatotoxic *Teucrium* species. Skullcap is known to contain a range of flavonoids but also contains essential oil at a low concentration. The only publication detailing the composition of the essential oil from *S. lateriflora* was published in 1988. Rather surprisingly, this paper stated that this endemic North American species “grows in Northern Iran (near the Caspian Sea)” where the material used for the analysis was collected. Given the species’ history of substitution problems, we determined the composition of essential oil distilled from three authentic *S. lateriflora* samples (cultivated in North America and Australia; authenticated by morphology and matched to authentic reference material by LC-MS). The steam-distilled oils from the three authentic samples (yield 0.01-0.02% w/w) were dominated by 1-octen-3-ol (29-68%), acetophenone (6-20%), β -caryophyllene (2-19%), linalool (two samples, 6-8%) and α -humulene (1-7%). These results are at odds with those reported for the Iranian oil sample, which was dominated by δ -cadinene, calamenene and β -elemene. Headspace GC/MS analysis of five authentic *S. lateriflora* samples was also at odds with the oil composition of the Iranian sample with four compounds (3-methyl butanal, 2-methyl butanal, styrene and 1-octen-3-ol) comprising > 50% of all samples. We conclude that authentic *S. lateriflora* essential oil is characterized by high levels of 1-octen-3-ol and acetophenone.

Key Words

Scutellaria lateriflora, skullcap, 1-octen-3-ol, essential oil composition, headspace analysis.

[P-011]

Chemical Composition of the Essential Oil from Aerial Parts of *Prangos latiloba* Korov

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Abstract

The genus *Prangos* consists of about 30 species. In Iran, fifteen species of this Umbelliferae family plant exist, among which five are endemic. The essential oil composition of some *Prangos* species has been the subject of several investigations. Medicinal applications have been reported for some *Prangos* species as: emollient, carminative, antifungal, antioxidant, antibacterial, cytokine release inhibitor and anti-HIV. In this work, hydrodistilled volatile oil from crushed dry aerial parts of *Prangos latiloba* Korov. (Umbelliferae) at flowering stage from Sabzevar (Iran), were studied by GC and GC/MS. The air-dried aerial parts of the plant was yielded 0.26 (w/w) clear yellowish oil. Forty-two components were identified in the aerial parts oil that contain 90.7% of the compounds. The main

components in this oil were hexadecanoic acid (5.9%), spathulenol (3.2%), α -bisabolol (3.1%) and α -curcumene (2.0%). A comparison of chemical composition of volatiles of *Prangos latiloba* with previous studies on the same genus showed variation of the major components.

Key Words

Prangos latiloba Korov., Umbelliferae, essential oil, hexadecanoic acid, spathulenol, α -bisabolol.

[P-012]

GC-TOF Analysis of *Pelargonium graveolens*: Comparison between the Volatiles Found in the Headspace, Essential Oil and Solvent Extract

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Abstract

Pelargonium graveolens L'Herit is a plant synonymous with the commercially sold rose geranium essential oil. However, it is not a parent plant of the cultivar from where this oil is derived. *Pelargonium graveolens* is an aromatic pelargonium, with a distinctive minty aroma, indigenous to Grahamstown in the Eastern Cape and the Limpopo provinces of South Africa. Leaves were harvested from *Pelargonium graveolens* growing in its native environment. Headspace analysis was performed at 250 rpm (35°C) using a 50/30 μ m DVB/CAR/PDMS SPME fiber with 30 min exposure. This was analyzed by GC-TOF. The essential oil was produced by using a Clevenger type apparatus, while the solvent extract used dichloromethane as the solvent. The essential oil and extract were analyzed through injection using GC-TOF. Results obtained from the headspace analysis showed > 20 peaks with isomenthone accounting for 21% of the total peak area in the chromatogram. The essential oil and solvent extract showed one major peak for isomenthone which accounted for > 80% and 68% of the total peak area, respectively. Minor peaks present in the essential oil were consistent with previous research, including myrcene (1%), limonene (0.5%), p-cymene (0.7%) and menthone (2%). Both the headspace sample and the essential oil showed minor peaks of α -phellandrene, ocimene and β -bourbonene. In conclusion, using SPME with GC-TOF to analyze the headspace of this aromatic pelargonium displayed the major constituents present in the essential oil along with extracted volatiles not present in the essential oil. The DCM solvent only extracted the more abundant lipophilic constituents in the leaves.

Key Words

Pelargonium graveolens, essential oil analysis, SPME, headspace analysis.

[P-013]

Chemical Composition of the Leaf Essential Oil of *Litsea glutinosa* (Lour.) C.B. Robinson from Central India

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Abstract

Litsea glutinosa (Lour.) C.B. Robinson (Indian Laurel) is an evergreen, medium-sized tree belonging to family Lauraceae that grows in the forests of central India. Its bark is one of the most popular native drugs considered to be capable of relieving pain, arousing sexual power and good for stomach. It is a mild astringent and also used for treatment of diarrhoea and dysentery. Leaves possess antispasmodic, antimicrobial properties and are also used as demulcent and emollient. The essential oil obtained by hydrodistillation from the fresh leaves of *L. glutinosa* was investigated by GC and GC/MS. Forty-eight components comprising 92.10% of the oil were identified. The major constituents of the oil were phytol (15.42%), caryophyllene (11.48%), thujopsene (10.17%), (E)-beta-ocimene (8.04%), β -myrcene (4.62%), germacrene D (4.58%) and caryophyllene oxide (4.18%).

Key Words

Litsea glutinosa, Lauraceae, essential oil composition, phytol, (E)-b-ocimene, caryophyllene.

[P-014]

Chemical Composition of the *Ligusticum mutellina* Flowers Essential Oil Obtained in Deryng and Clevenger Apparatus and by Use of SPME

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Abstract

Ligusticum mutellina (L.) Crantz [syn. *Mutellina purpurea* (Poir.) Thell., *Meum mutellina* (L.) Gaert.] belongs to Apiaceae family. Genus *Ligusticum* is widespread and complex. It consists of about 60 species growing in Asia, Europe and North America. Among these, 35 species are endemic in China. In nature *Ligusticum mutellina* grows on the Alpine pastures, among the mountain pine or on the glades. It is the herb typical of Carpathian Mountains and of Polish Tatra Mountains. The inflorescences of this aromatic plant were collected in Poland. The essential oils from *L. mutellina* inflorescences were obtained in the Deryng apparatus, Clevenger apparatus and using SPME. The received essential oils were analyzed by GC. Analysis revealed differences in the essential oils quantitative composition. The essential oil obtained in the Deryng apparatus contained as a main constituents sabinene (23,97%), alpha-pinene (9,97%), myrcene (8,86%), *trans*-sesquisabinene hydrate (8,55%) and *cis*-sesquisabinene hydrate (6,89%). Sabinene (19,16%), alpha-pinene (12,58%), *trans*-sesquisabinene hydrate (9,01%), myrcene (7,82%), *cis*-sesquisabinene hydrate (7,50%) and alpha-bisabolol (6,73%) dominated in the essential oil from

Clevenger apparatus. SPME microextraction gave sabinene (23,0%), metacresol (17.37%), alpha-pinene (16.96%) and myrcene (10,88%) as major constituents.

Key Words

Ligusticum mutellina, essential oil, Mutellina purpurea, Deryng apparatus, Clevenger apparatus, SPME.

[P-015]

Effect of Extraction Parameters on the Supercritical CO₂ Extraction of *Inula viscosa* (L.) Aiton

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Abstract

Supercritical fluid extraction (SFE) has been demonstrated to be a valuable alternative since it requires less solvent and shorter extraction time and is capable of extracting the mainly labile compounds under mild conditions. Supercritical carbon dioxide (SC-CO₂) was used as solvent for extraction from aerial parts of *Inula viscosa* (L.) Aiton. Several parameters influencing the extraction process have been identified and studied, such as pressure (200, 300, 400 bar), temperature (30°C, 35°C, 40°C, 50°C), flow rate (2, 4, 6 mL/min), and particle size (ground and full leaves). The amount extracted was about 55-80 mg/g of dry leaves, which was quite noticeable. The experimental results showed that pressure has significant effect on the yield. The extraction yield increased as pressure was increased; however, temperature had a varying effect on the extraction yield. A higher extraction yield was obtained for ground leaves as compared to full leaves and a flow rate of 2 mL/min gave greater yield than 4 and 6 mL/min, respectively.

Key Words

Inula viscosa, supercritical CO₂ extraction, essential oils.

[P-016]

Phytochemical Studies on Essential Oils of the Plants of Myrtaceae family

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Abstract

Using steam distillation method, essential oils of some of the Eucalyptus species related to the Myrtaceae family grown in some areas of Iran, i.e, Fars province, were extracted and analyzed by GC/MS to identify their constituents. To separate active constituent, i.e., 1,8-Cineole, crystallization at low temperature was applied. Results showed that the species *E. flocktoniae* Maiden, *E. bosistoana* F. Muell. *E. transcontinentalis* Maiden and *E. longicornis* (F. Muell.) Maiden contain higher amounts of essential oils and *E. viridis*, *E. tereticornis* Sm. and *E. eremophila* (Diels) Maiden were rich in 1,8-cineole.

Key Words

Eucalyptus, Myrtaceae, essential oils, active constituent.

[P-017]

Chemical Composition of Essential Oil and *In vitro* Antibacterial Activity from *Hypericum scabrum* in Northern Iran

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Abstract

The chemical composition of the essential oil obtained from the leaves of *Hypericum scabrum* L. by hydro-distillation was analyzed by GC/MS. A total of twenty compounds, representing 93.8% of the oil were identified. The major constituents of the essential oil were n-nonane (6.3%), α -pinene (44.8), β -pinene (2.2%), α -camphene (4.3%), camphor (1.6%), δ -verbenol (2.7%), p-mentha-1,5-dien-8-ol (4.2%), verbenone (3.52%), germacrene-D (3.2%) and verbenene (1.2%). The antibacterial activity of the essential oil was evaluated using disc diffusion method. The essential oil exhibited moderate activity against *Proteus mirabilis*, *Enterobacter cloacae* and *Staphylococcus aureus* at minimum inhibitory concentration (MIC) of 1000 μ g/mL. The antibacterial activity of the oil suggests its potential use as a remedy for food-borne diseases.

Key Words

Hypericum scabrum, antibacterial activity, essential oil

[P-018]

Essential Oil Variability of *Hypericum perforatum* L. Growing Wild in Albania

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Abstract

The essential oils obtained by hydrodistillation from the aerial parts of wild plant of *Hypericum perforatum* L. collected in 10 different localities in Albania, were obtained by steam distillation and analyzed. The essential oils investigated were characterized by a high content of non-terpene compounds and a low content of monoterpenes. All data were statistically processed with principal component analysis and cluster analysis. Both oils were characterized by the presence of many components which could have numerous applications in food, pharmaceutical and perfume industries. The main conclusion from the above data is that genetic and environmental factors both play a role in determining the chemical variability of essential oils of the *Hypericum* species studied.

Key Words

Hypericum perforatum, essential oil composition, GC/MS, cluster analysis.

[P-019]

A Simple, Reliable and Efficient QSRR-chemometrics-based Model for Estimation of Retention Indices of *Eryngium rosulatum* P.W. Michael ined. as a Thorny Wild Plant Based on Related Molecular Structures

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Abstract

One of the most ubiquitous challenges for scientists is the theoretical evaluation of experimental parameters to validate and improve their ability. This work is a part of our comprehensive investigation to correlate the experimental and calculated retention indexes of the oil separated from *Eryngium rosulatum* P.W. Michael ined. The structures of all organic compounds were drawn into the HYPERCHEM program and optimized using semi-empirical AM1 method, applying a gradient limit of 0.01 kcal/Å as a stopping criterion for optimized structures prior to the geometry optimization step. Then molecular descriptors were calculated for each compound by the DRAGON software on the minimal energy conformations. The Stepwise SPSS was used for the selection of the variables that resulted in the best fitted models. Five descriptors (ATS6e, R5e, BENm4, HATSe, GATS6e) are considered to account for the effect of solute structure on the retention index. After the variables selection, the

MLR method used for building the regression models. The statistical figures obtained by the proposed model are RMSEP = 64.7, REP = 4.2 and SEP = 67.8. In the final step, generated models were used to predict the retention indices for a diverse set of test compounds.

Key Words

HYPERCHEM, AM1 method, retention index, *Eryngium rosulatum*, essential oils, QSRR.

[P-020]

Comparison of Artificial Neural Network with Multivariate Linear Models for Prediction of Retention Behaviors of *Echinops kebericho* Mesfin Essential Oil

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Abstract

Echinops is one of the genera classified under the family Asteraceae. It is claimed that the roots of this plant are effective to repel snakes, reduce stomachache as well as curing the intestinal diseases in cattle. According to a data set containing the components of essential oil from *E. kebericho*, a quantitative structure–retention relationship (QSRR) study, has been carried out on the gas chromatographic retention indices (RIs) with different multivariate calibration methods feed-forward artificial neural networks (ANN) with back-propagation learning rule, and linear correlating model multiple linear regression (MLR) by using molecular structural descriptors. Appropriate models with low standard errors and high correlation coefficients were obtained. The Stepwise SPSS was used for the selection of the variables that resulted in the best fitted models. Five descriptors are considered to account for the effect of solute structure on the retention index. After variables selection, the MLR method was used for building the regression models. A 3-layer feed-forward network formed by five input layer consisting of a number of neurons equal to the number of descriptors, 1 output neuron and a number of hidden units fully connected to both input and output neurons, were adopted in this study. After variables selection, 83 compounds were randomly divided into two training and test sets. Comparing the two used methods showed that ANN has better prediction ability than MLR. Results obtained showed that nonlinear model (ANN) can accurately simulate the relationship between structural descriptors and the retention indices of the molecules in data set.

Key Words

Echinops kebericho, artificial neural networks (ANN), retention index (RI), essential oil.

[P-021]

Chemical Composition of the Essential Oil from Flowers of *Haplophyllum perforatum* by using Headspace Solid Phase Microextraction-GC/MS

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Abstract

The genus *Haplophyllum* (family Rutaceae) consists of 18 species in Iran. Among them, *H. tuberculatum*, *H. robustum*, and *H. Perfolatum* are the most common species and are widely grown in areas of different climates. The present report deals with identification of the volatile constituents of flowers of *Haplophyllum perforatum* in Semnan province, Iran, by using headspace solid phase microextraction-gas chromatography mass spectrometry (HS-SPME-GC/MS). The search for the concerned active compounds has led to isolation of nine constituents (95.9%) in flower oil. The chemical profile was consisted of three monoterpene hydrocarbons (60.5%), two oxygenated monoterpenes (6.6%), one sesquiterpene hydrocarbons (10.8%), one non-terpenoid hydrocarbon (6.6%), and two oxygenated sesquiterpenes (10.3%). Accordingly, monoterpene hydrocarbons were the major constituents, and a ranking order of the groups of constituents as MH>SH>OS>OM=NH was observed in the volatile flower oil. Sabinene was the most abundant constituent (52.7%), followed by *trans*-caryophyllene (10.8%), (2E,6E)-farnesyl acetone (10.3%), hexadecanoic acid (5.1%), β -pinene (5.0%), and *cis*-sabinene hydrate (4.9%).

Key Words

Haplophyllum perforatum, essential oil, HS-SPME-GC/MS, sabinene.

[P-021]

Quantitative structure-retention relationship modeling study of *Eugenia brasiliensis* volatile oil compounds based on GA-MLR and SW-MLR techniques

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Abstract

Eugenia brasiliensis Lam. is a tree to 12-15 m high and native to southeastern Brazil from Santa Catarina to Bahia States where it is known as grumixama. The analysis of the essential oil of this plant has been the subject of a report. However, in this work a comparative study concerning genetic algorithm multiple linear regression (GA-MLR) and stepwise multiple linear regression analysis (SW-MLR) techniques for understanding quantitative structure-retention relationship (QSRR) behavior of the oil constituents has been conducted by using the molecular descriptors. The structures of all organic compounds were drawn into the HYPERCHEM program and optimized using semi-empirical AM1 method, applying a gradient limit of 0.001 kcal/Å as a stopping criterion for optimized structures. Then molecular descriptors were calculated for each compound by the DRAGON

software on the minimal energy conformations. Generated models were used to predict the retention indices for a set of test compounds. The results indicated that the GA-MLR method proved to be superior of SW-MLR. In the training set, the prediction power of GA-MLR was very high and also it performed better in estimation the RI values for the test set. These QSPR approaches may be a useful to screen in new compounds candidates from larger compound libraries to be further evaluated. The prediction results are in very good agreement with the experimental values.

Key Words

Eugenia brasiliensis, genetic algorithm multiple linear regression (GA-MLR), stepwise multiple linear regression (SW-MLR).

[P-022]

Essential oil composition of *Kalakia marginata* (Apiaceae) from Iran

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Abstract

The essential oil composition of monotypic genus *Kalakia* Alava, belonging to Apiaceae from Iran was investigated for the first time. Hydrodistillation of the aerial parts of *Kalakia marginata* (Boiss.) Alava at fruiting stage gave a yellow oil in 0.76% (w/w) yield, based on the dry weight of the plant. The essential oil was analyzed by GC and GC/MS and identified. Twenty-four components were characterized accounting for 97.9% of the total oil. (2E)-decenal (73.5%), n-decanal (12.0%) and (2E)-decen-1-ol (3.8%) were determined as principal compounds.

Key Words

Kalakia marginata, Umbelliferae, essential oil composition, Iran.

[P-023]

Validated GC/MS method for quick and simultaneous determination of α -pinene, cineole, linalool and eugenol in different essential oil containing plants

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Abstract

Fixed oils and essential oils have been widely used in medicine for treatment of a number of diseases. The GC/MS is a technique through which we can quantitate all the constituents of an oil in a comparatively more precise manner. The one drawback with GC/MS is its long run time as compared to other analytical techniques like HPLC. In the present work we have developed and validated a quick and simultaneous method for determination of four

volatile oil constituents, α -pinene, 1,8-cineole, linalool and eugenol. These constituents (either individually or in combination) are generally present in major amounts in most of the essential oil containing medicinal plants like *Elettaria cardamomum* Linn., *Cinnamomum zeylanicum* Blume, *Syzygium aromaticum* L. and *Coriandrum sativum* L. The advantage of this method is short elution time (only 15 min) and better separation of each component. The method was developed on a GC/MS system (Agilent 7890A series) equipped with split-splitless injector and CTC-PAL auto sampler attached to an apolar HP-5MSTM capillary column (30 m x 0.25 mm i.d. and 0.25 μ m film thickness). Helium was used as carrier gas. Mass spectra were acquired in EI mode (70 eV); in m/z range 30–600. The retention times of α -pinene, 1,8-cineole, linalool and eugenol were found as 3.87 ± 0.07 , 5.9 ± 0.05 , 7.128 ± 0.09 and 14.78 ± 0.12 respectively. The method was validated as per the ICH guidelines for linearity, precision, accuracy, robustness, LOD and LOQ. This developed method has been used for analysis of oil of different plant species and was found satisfactory.

Key Words

α -Pinene, cineole, linalool, eugenol, GC/MS.

[P-024]

Creation of Nature-identical Cinnamon, Nutmeg and Pimento Oils

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Abstract

The Amendments of IFRA (International Fragrance Association) have stated certain restrictions for the usage of 4-allyl-1,2-dimethoxybenzene, CAS 93-15-2 (methyl eugenol), and 4-allyl-1,2-methylene dioxybenzene, CAS 94-59-7 (safrole), in fragrance compounds. Contributions from essential oils can be very significant. *Cinnamomum zeylanicum* Nees (cinnamon leaf oil), *Myristica fragrans* Houtt (nutmeg oil) and *Pimenta officinalis* Lindl. (pimento berry oil) were analyzed by GC/MS. Amounts of methyl eugenol and safrole obtained correlate with the data collected in the previous researches. Studies were made in order to develop nature-identical essential oils mentioned above that do not contain either methyl eugenol or safrole. The derived nature-identicals were examined and compared by GC/MS. Although the results showed differences in some constituents, only trace amounts (< 0.1%) of safrole and methyl eugenol were observed. In conclusion, nature-identicals resembling essential oils olfactively, which can be used without any restrictions, were created.

Key Words

Methyl eugenol, safrole, GC/MS, IFRA, *Cinnamomum zeylanicum*, *Myristica fragrans*, *Pimenta officinalis*.

[P-025]

Study of essential oil of clove buds for its nutraceutical activities

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Abstract

Through past and present time, human beings have been need of food and medicine. In industrialized societies where many new diseases are developing and due to economical problems, especially in poverty areas, need of having safe food and natural drugs seems to be more needed. In this work the essential oil of clove buds was extracted by steam distillation method and analyzed by GC/MS. Essential oil of clove is 15.5% of the buds weight and had 92% of eugenol. In the present work 50-85% eugenol was observed. Other constituents were: caryophyllin, furfural, alpha and beta sesquiterpene and also eugenin. Studies on rats showed this oil as having antidiabetic effects, and can be used as a nutraceutical, since clove bud is also used as fragrance in the food industry. Antidiabetic effects were observed using aloxan to make diabetic model in rats. It is observed that the oil is processing antidiabetic effects.

Key Words

Clove buds, essential oils, antidiabetic, nutraceuticals.

[P-026]

Chemotaxonomic evidence suggests that *Eriocephalus tenuifolius* is the source of Cape chamomile oil and not *E. punctulatus*

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Abstract

The distinct blue essential oil obtained from *Eriocephalus* sp. (Asteraceae) is known in commerce as Cape chamomile and has become an important ingredient in flavour and fragrance formulations. To date, it is commonly accepted that Cape chamomile oil is obtained from *Eriocephalus punctulatus*. Samples of *E. tenuifolius* DC (n = 35) and *E. punctulatus* DC (n = 17) were collected from different localities in South Africa and Lesotho. The hydrodistilled essential oil was analyzed by GC/MS. The data was exported to MetAlign for spectral alignment and differentiation. The mid-infrared red (MIR) data was processed using principal component analysis (PCA), partial least squares (PLS) and orthogonal projections to latent structures discriminant analysis (OPLS-DA) algorithms. The GC/MS results showed the presence of 2-methyl butyl isobutyrate and isobutyl isobutyrate as characteristic compounds in the commercial oil and *E. tenuifolius* oils. *Eriocephalus punctulatus* oil was devoid of these compounds with piperitone, camphor, yomogi alcohol, pogostol and 1,8-cineole occurring in high quantities not observed in the commercial oil. MetAlign successfully discriminated between *E. punctulatus* and *E. tenuifolius* oil and the data together with multivariate analysis showed that the commercial oil was closely related to *E. tenuifolius* oil. Using the MIR data, an OPLS-DA model was constructed which convincingly discriminated

between *E. tenuifolius* and *E. punctulatus* oil. The developed model further predicted the botanical origin of the commercial oil to be *E. tenuifolius*. The statistical performance of the model was excellent with $R^2X = 0.897$; $R^2Y = 0.991$ and $Q^2Y = 0.987$.

Key Words

Eriocephalus punctulatus, *E. tenuifolius*, Cape chamomile oil, MetAlign, MIR, OPLS-DA.

[P-027]

GC/MS finger print of an Essential Oil used as Antibacterial agent and Analgesic

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Abstract

Clove oil is distilled from the flowers, stems and leaves of the clove tree (*Eugenia caryophyllus* (Spreng.) Bullock et S.G. Harrison, Family: Myrtaceae). Official literature showed that it contains not less than 85% and not more than 95% w/w of phenolic substance. A method has been developed for the fingerprinting of different essential oil constituents of clove (*Eugenia caryophyllus*) in hydrodistilled oils. Separation was performed on HP5 MS column (5% phenyl polymethyl siloxane; 30 m x 0.25 mm i.d. and 0.25 μ m film thickness); the detector was an EI/CI MSD. Helium at a flow-rate of 1 mL/min was used as a carrier gas. The eugenol was found to be eluting at the retention time 4.8 ± 0.02 mins. The content of different constituents of clove oil was identified by the newly developed method. The developed GC/MS method has been found satisfactory enough for the separation and identification of different constituents present in the clove oil.

Key Words

GC/MS, clove oil, eugenol.

[P-028]

Hydrodistilled volatile oil from stems of *Eryngium creticum* Lam. in the marginal brackish regions of Semnan province by using gas chromatography combined with mass spectrometry

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Abstract

The present report deals with the chemical composition of the volatile oil from stems of *Eryngium creticum* Lam., a growing wild plant in Iran. The genus *Eryngium* belongs to Apiaceae family and consists of 9 thorny species in Iran. Among them, *E. bungei* Boiss., *E. caucasicum* Trautv., as well as *E. billardiarei* Heldr. ex Boiss., are the most common species worldwide. Our samples were collected from different brackish and hill slope locations

during the flowering stage in Semnan province, Iran. The oil was obtained by using a modified Clevenger-type apparatus, and the respective analyses were performed by means of GC and GC/MS instrumentations. It was obtained as a clear yellowish color liquid involving 0.18-1%. Seventeen components were identified constituting 91.4% of the oil composition. The major components of the oil were found to be bornyl acetate (28.4%), camphor (17.8%), α -pinene (12.1%), germacrene D (9.4%), borneol (8.6%), and α -thujene (4.2%). Based upon the chemical profile, the essential oil was mainly characterized by the presence of higher amounts of oxygenated monoterpenes.

Key Words

Eryngium creticum, GC/MS, hydrodistillation, bornyl acetate, essential oil.

[P-029]

Quality and quantity of the flower essential oil of *Ferulago angulata* Boiss. plants growing at different altitudes

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Abstract

Variability in the quality and quantity of the flower essential oil of *Ferulago angulata* Boiss. plants growing at different altitudes of Hesar, Markazi province, Iran (2100, 2500 and 2800 m) was investigated in the present study. Essential oils were obtained by hydrodistillation of dried flowers and analyzed by GC and GC/MS. The highest (2.11%) and lowest (1.71%) yields of essential oil (w/w) were observed at the middle and higher altitudes, respectively. Changes in altitude also resulted in significant variations in the chemical composition of studies essential oils, especially from the quantitative point of view. Samples from the plants of higher altitudes (2800 m) were rich in (Z)- β -ocimene (40.13%), allo-ocimene (15.51%) and α -pinene (12.64%), while plants of the middle (2500 m) and lower (2100 m) altitudes contained the high levels of α -pinene (23.75%), bornyl acetate (16.34%) and (E)-verbenol (16.33%), and α -pinene (24.00%), (Z)- β -ocimene (18.86%) and γ -terpinene (11.44%), respectively.

Key Words

Ferulago angulata, essential oil, (Z)- β -ocimene, α -pinene.

[P-030]

Variation in the essential oil content and composition of flowers of *Ferulago angulata* Boiss. populations growing wild in Iran

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Abstract

The present study was aimed to investigate the essential oil content and composition of flowers of 5 wild growing *Ferulago angulata* Boiss. (Apiaceae) populations collected from different parts of Iran including Dalahoo and Mersad (Kermanshah province), Sisakht (Kohgiluyeh va Boyer-Ahmad province), and Sefidkhani and Besri (Markazi province). For this purpose, the air-dried samples were separately subjected to hydrodistillation for 3 h and their essential oils chemically analyzed. Considerable variation was found among the oil content of studied populations (w/w) with the mean value of 1.45% and the highest and lowest levels in samples from Mersad (3.01%) and Besri (0.53%), respectively. According to GC and GC/MS analyses, monoterpenoids were identified as the principal fraction of all essential oils and α -pinene (16.9-32.7%), (Z)- β -ocimene (13.5-20.9%), allo-ocimene (5.8-8.1%), bornyl acetate (2.3-12.0%) and γ -terpinene (0.9-13.0%) as the most abundant components.

Key Words

Ferulago angulata Boiss, Apiaceae, essential oil, α -pinene, (Z)- β -ocimene.

[P-031]

Chemical analysis of essential oils from stems, flowers and fruits of *Ferulago angulata* (Schlecht.) Boiss. from Iran

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Abstract

Ferulago angulata (Schlecht.) Boiss. is a popular perennial aromatic plant of celery family (Apiaceae) with some medicinal properties. In this study, different parts of wild growing plants gathered from Kerend, Kermanshah province, Iran including stems, flowers and fruits were separately hydrodistilled for 3 h using a Clevenger-type apparatus and their essential oils investigated qualitatively by means of GC and GC/MS. Based on the dry weight, the oil content of air-dried stems, flowers and fruits was calculated to be 0.3%, 1.7% and 2.3% (w/w), respectively. There was considerable variation among the chemical profiles of studied samples, although they were principally made up of monoterpene hydrocarbons. While the oil from stems was consisted mainly of bornyl acetate (26.9%) and α -pinene (14.0%), (Z)- β -ocimene (50.6% and 45.9%) and allo-ocimene (20.9% and 16.7%) constituted the major components of essential oils from fruits and flowers, respectively.

Key Words

Ferulago angulata, essential oil, bornyl acetate, (Z)- β -ocimene, allo-ocimene, α -pinene.

[P-032]

Essential Oil Analysis of *Cichorium intybus* L. from Iran

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Abstract

Cichorium intybus L. commonly known as Chicory or Kasni has a long history of medicinal herbal use and is especially of great value for its tonic effect upon the liver, cardio and digestive tract. In this study, the composition of volatile oil from overground parts of *C. intybus* has been surveyed. Two samples of *C. intybus* aerial parts were collected at flowering stage in central Iran (Kashan, Isfahan province). Volatile compounds were obtained by a two-step extraction of hydrodistillation and liquid-liquid extraction. Distillate was extracted with pentane. The organic layer was separated, dried over anhydrous sodium sulphate and concentrated to 0.5 mL under reduced pressure. The concentrated extract had a strong odor and yellow color. The separation and identification of the oil components were carried out using a combination of GC-FID and GC/MS. The analysis of the oil revealed the presence of twenty components identified in the aerial parts of this herb. The main components were carvacrol (50.1%), thymol (13.3%), cinnamic aldehyde (12.4%), camphor (4.4%), carvone (4.1%), linalool (3.9%) and α -terpineol (2.1%).

Key Words

Cichorium intybus, carvacrol, thymol, GC/MS.

[P-033]

Comparative Investigation of the Essential Oils of *Sannantha leratii* (Schltr.) Peter G. Wilson and *Sannantha pinifolia* (Labill.) Peter G. Wilson from New Caledonia

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Abstract

Sannantha leratii and *Sannantha pinifolia* (Myrtaceae) are two endemic plants from New Caledonia. In this study, the essential oils from the flowering branches of *S. leratii* and *S. pinifolia* growing wild were subjected to a comparative investigation by GC and GC/MS. The plant material from *S. leratii* was collected in the southern region of the main island (Grande Terre), where the plant grows abundantly. By contrast, *S. pinifolia* is widespread on the eastern coast and in the central area of the island. On steam distillation, the oil yields were 0.19% for *S. leratii* and 0.08% for *S. pinifolia*. More than 50 compounds were identified in both oils, representing more than 90% of the total. Both oil samples showed remarkable similarity with respect to the major peaks. The oils were rich in monoterpene and sesquiterpene hydrocarbons. α -Pinene (21,1% and 14,4%), β -pinene (4,7% and 2,0%), p-cymene (11,3% and 9,3%), limonene (6,5% and 2,4%), 1,8-cineole (5,7% and 2,0%), γ -terpinene (12,0% and 15,8%), β -caryophyllene (3,7% and 10,0%), aromadendrene (6,1% and 1,4%), bicyclogermacrene (1,6% and 3,7%), *cis*-calamenene (1,8% and 6,6%) and caryophyllene oxide (3,3% and 1,0%) were common to both oils and were the principal constituents. Nevertheless, the investigation revealed some characteristic components found in the oil from one species but not in the other. These chemical markers were borneol and 1,10-di-epi-cubenol for *S. leratii* and, respectively, α -cubebene, β -bourbonene, δ -cadinene and *trans*-cadin-1,4-diene for *S. pinifolia*.

Key Words

Sannantha leratii, *Sannantha pinifolia*, Myrtaceae, essential oil composition, α -pinene, p-cymene, γ -terpinene, β -caryophyllene, *cis*-calamenene.

[P-034]

Volatiles components of *Daucus crinitus* from Algeria

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Abstract

In the present work, we investigated the chemical composition of the Algerian *Daucus crinitus* Desf. essential oils extracted by hydrodistillation (HD) from separated organs (roots, stems, leaves, flowers and umbels) and the volatile fractions extracted by Head-Space Solid Phase Micro-Extraction (HS-SPME) from the same plant materials. In all cases the analysis was carried out using GC and GC/MS and Carbon-13 NMR for essential oils.

In accordance with the literature, the main volatiles of *D. crinitus* separated organs extracted by HD and

HS-SPME were phenylpropanoids, aliphatic compounds and hydrocarbon sesquiterpenes such as isochavicol isobutyrate (tr-84.1%), isochavicol 2-methyl butyrate (tr-17.8%), isochavicol (tr-34.7%), dodecanal (tr-55.4%), pentadecane (tr-51.6%) and heptadecane (tr-20.7%). Therefore, our studies showed that two types of essential oils were produced by the plant: root oils were dominated by aliphatic compounds (87.0-90.1%) while the aerials part oils were characterized by phenylpropanoids (43.1-88.6%). Moreover, HS-SPME analysis showed a more precise distribution of volatiles in the studied organs: oxygenated aliphatic compounds (44.3-84.0%) were well represented in roots, hydrocarbon aliphatic compounds in leaves and stems (31.3-88.7%) and phenylpropanoids in flowers and umbels (47.9-64.2%).

Finally, the study demonstrated that HS-SPME extraction could be considered as an alternative technique for the investigation of volatiles from aromatic plants.

Key Words

Daucus crinitus, essential oil, HS-SPME, isochavicol derivatives.

[P-035]

Composition and chemical variability of essential oil of *Limbarda crithmoides* (L.) Dumort from Corsica

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Abstract

The genus *Limbarda* (Asteraceae family) is widespread all over the world. In Corsica, it comprises only one species: *Limbarda crithmoides* (L.) subsp. *longifolia*. *L. crithmoides* is a perennial plant commonly widespread on the ponds and briny water. Two studies described the chemical composition of aerial part essential oils from Greece, Malta, Spain and central Italy. The aim of the work was to investigate the chemical composition of the essential oils of *L. crithmoides* harvested in Corsica using a combination of techniques including GC/RI, GC/MS and RMN.

Integrated analysis of *L. crithmoides* oils allowed the identification of 65 components accounting for 91.2-96.4 g/100 g of the total oils. Among them, the main components were para-cymene (34.6-15.5 g/100 g), 2-methoxy-para-cymenene (13.5-28.5 g/100 g), 2,5-dimethoxy-para-cymenene (6.3-10.3 g/100 g), α -phellandrene (1.4-6.6 g/100 g), thymyl methyl oxide (1.3-13.3 g/100 g) and α -pinene (0.2-13.1 g/100 g). To our knowledge, it is the first report of both para-cymenene derivatives as natural products. Since, the ¹H-NMR spectral data of both para-cymenene derivatives were reported in the literature, we reported for the first time their ¹³C-NMR spectral data.

In addition, statistical analysis has been performed in order to obtain a better insight of the chemical variability of *L. crithmoides* oil originated from 12 localities of Corsica. Two main groups were identified, they were discriminated by the amount of α -Pinene which was higher in GI sample oils (6.9-13.1 g/100 g) than in GII sample oils (0.2-0.8 g/100 g). The clustering samples seem to be linked to the salinity level of the harvest localities.

Key Words

Limbarda crithmoides, Essential oil, Chemical variability, GC/MS, NMR, para-cymenene derivatives.

[P-036]

Chemical composition of essential oils from *Asteriscus graveolens* (Forssk.) Less.: Identification of *cis*-8-acetoxychrysanthenyl acetate as new natural component

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Abstract

Asteriscus graveolens (Forssk.) Less., is an endemic species from Morocco growing wild throughout the Sahara, especially in sandy clay pans. It has been used in Saharan folk medicine as a stomachic, for treating fever, gastrointestinal tract complaints, cephalic pains, bronchitis and as an anti-inflammatory. Only two studies have been published concerning the chemical composition of *A. graveolens* essential oil under synonyms (*Nauplius graveolens* and *Bubonium graveolens*). The aim of this work was to investigate the essential oils of leaves, flowers, stems and full aerial parts using GC, GC/MS and NMR. In this study, the spectral data of two nerolidol derivatives: 6-oxocyclonerolidol and 6-hydroxycyclonerolidol were reassigned using 1D and 2D NMR. These two compounds can be considered as chemical markers of this genus. The structure of a monoterpene diester bearing to the chrysanthenane skeleton, *cis*-8-acetoxychrysanthenyl acetate was reported for the first time using GC/MS(IC), 1D and 2D NMR. Finally, 50 components were identified in essential oils of *A. graveolens* amounting to 94.4%, 98.2%, 94.2% and 93.0% of the total oil from aerial parts, leaves, flowers and stems, respectively. The aerial part leaf and stem oil was characterized by high content of oxygenated sesquiterpenes with 6-oxocyclonerolidol and 6-hydroxycyclonerolidol as major components and the flower essential oil was dominated by the new monoterpene compound *cis*-8-acetoxy chrysanthenyl acetate.

Key Words

Asteriscus graveolens, essential oil, 6-oxocyclonerolidol and 6-hydroxycyclonerolidol, *cis*-8-acetoxy chrysanthenyl acetate, GC/MS, NMR.

[P-037]

Identification of carvone chemotypes of *Lippia scaberrima* by headspace analysis

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Abstract

Lippia scaberrima Sond. is an aromatic shrub of semi-arid, summer-rainfall grassland. The phytoremediation strategy currently followed at some of the South African goldmines involves the identification of indigenous species that have naturally established on disturbed soils and subsequent evaluation of their potential for use in tailing rehabilitation. *L. scaberrima*, which produces a carvone-rich essential oil with antifungal properties, was one of such species identified. The aim of this study was to screen and continuously monitor volatile emissions of *L. scaberrima* to identify good carvone-producing chemotypes without a lengthy distillation process. Three sampling techniques were evaluated and compared in spring and summer. The GC profiles of the essential oils obtained through hydrodistillation were compared to the volatile profiles obtained using two head-space techniques i.e. solid phase micro-extraction (SPME) and thermal desorption from Tenax traps. The GC profiles resulting from the three sampling methods were similar, but both qualitative and quantitative variability was observed after using different SPME fibres. Although small quantitative differences in the terpene constituents were observed, all three methods were found to be suitable for identifying carvone chemotypes. These rapid headspace methods are useful tools for on-site monitoring of aromatic plants used for mine rehabilitation and other purposes.

Key Words

Carvone, tailing, phytoremediation, headspace.

[P-038]

Biological activities of the essential oil of *Cymbopogon jawarancusa*

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Abstract

The chemical composition of essential oil isolated by hydrodistillation from *Cymbopogon jawarancusa* Schult. was analyzed by GC-FID, GC/MS, ¹³C-NMR and assessed for anticancer and antimicrobial activities. GC/MS analysis led to the identification of 17 constituents representing 97.8% of the total oil. The major constituents were piperitone (58.7%), elemol (18.6%), α -eudesmol (10.0%) and α -pinene (2.6%). The essential oil and its major constituents isolated by silica gel chromatography were subjected to screening for possible anticancer activity at different concentrations (10 μ g, 30 μ g, 50 μ g and 100 μ g), against four human cancer cell lines of varying tissue origin, like THP-1 (Leukemia), A-549 (Lung), HEP-2 (Liver) and IGR-OV-1 (ovary) by SRB assay. The oil showed appreciable anticancer effects even at 10 μ g and 30 μ g against THP-1 (Leukemia), A-549 (lung) and HEP-2 (liver) cancer cell lines. In addition to anticancer activity, antimicrobial activity of oil was screened using Agar diffusion method against six test microorganisms, namely *Staphylococcus epidermis*, *Bacillus subtilis*, *Staphylococcus aureus*, *Proteus vulgaris*, *Pseudomonas aeruginosa*, *Aspergillus niger* and *Penicillium chrysogenum*. The zones of inhibition were measured after 24 h incubation for bacteria at 37°C and 72 h for fungi at 28°C, using 3.5 mg

of oil. Minimum inhibitory concentration and minimum biocidal concentration of oil was determined by dilution method. The oil showed strong antimicrobial and anticancer activity against aforesaid seven test microorganisms and human cancer cell lines, respectively.

Key Words

Cymbopogon jawarancusa, essential oil, piperitone, anticancer activity, antifungal activity, antibacterial activity, MIC, MBC.

[P-039]

Insecticidal effects of *Thuja occidentalis* (Cupressaceae) essential oil on *Lasioderma serricorne* (Col.: Anobiidae)

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Abstract

In order to find recyclable, environmentally friendly and easily accessible insecticides, the essential oil of *Thuja occidentalis* L. (Cupressaceae), was used against the adult stages of *Lasioderma serricorne* (F.). Analysis of *Thuja occidentalis* essential oil used for insect fumigation by phase gas chromatography revealed the presence of 22 compounds including α -thujone (49.64%), fenchone (14.06%), and β -thujone (8.98%). The experiment was conducted in 6 replications using a completely randomized design of factorial experiment. The essential oil was prepared by water distillation method. Experiment was carried out at $30 \pm 2^\circ\text{C}$ and $60 \pm 5\%$ R.H. under dark conditions. Concentrations are included 0.00375, 0.00493, 0.00650, 0.00855, 0.01125 $\mu\text{L/L}$ air and a control (untreatment), after 24 h, respectively. At the concentration of 0.015 $\mu\text{L/L}$ air essential oil caused 100% mortalities of *Lasioderma serricorne* (F.) adults. A value of 50% lethal concentration on adults was 0.00687 $\mu\text{L/L}$ air for *Lasioderma serricorne* (F.), respectively.

Key Words

Thuja occidentalis, *Lasioderma serricorne*, essential oil, fumigation, mortality, Lc50.

[P-040]

Chemical composition and antifungal activity of the *Myrtus communis* and *Pistacia lentiscus* essential oils of Mediterranean regions in laboratory medium and strawberry fruit

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Abstract

The aim of this work is to study the chemical composition and antifungal activity of essential oils isolated from leaves of *Myrtus communis* L. and *Pistacia lentiscus* L. growing in North Algeria against two postharvest pathogenic fungi of strawberry fruit: *Aspergillus niger* and *Penicillium* sp. The chemical compositions of the essential oils were analyzed using Gas Chromatography/Mass Spectrometry (GC/MS). The essential oil of *M. communis* was characterized by the presence of 1,8-cineole (47.0%) and geraniol (25.2%) as principal chemical components. The essential oil of *P. lentiscus* consisted mainly of myrcene (15.2%) and 1,8-cineole (15.0%). The *in vitro* antimicrobial activity of *P. lentiscus* and *M. communis* essential oils against *A. niger* and *Penicillium* sp. were evaluated using standard agar-disk diffusion assay. The results obtained, followed by measurements of Minimal Inhibitory Concentrations (MICs), showed promising inhibitory effect against both pathogens tested. Oil of *M. communis* was the most active (Φ 59.5 mm; Φ 46 mm) against *Penicillium* sp. and *A. niger*, respectively. *P. lentiscus* oil was tested on strawberry fruit, experimentally inoculated with *A. niger* and *Penicillium* sp., separately at a concentration of 107 spores/mL. Results demonstrated that *P. lentiscus* oil exerted an antifungal effect against the pathogens tested. This effect was evident from 24 h of incubation; showing significant differences against untreated samples. The data suggest a possibility that these essential oils could be used as natural preservatives for improving the shelf life of strawberry fruit.

Key Words

Essential oils, *Aspergillus niger*, *Penicillium* sp, antifungal activity, strawberry.

[P-041]

Essential oils from tea leaves provide oral hygiene and prevent oral bacterial growth

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Abstract

Tea has long been known to have antibacterial, antifungal and antiviral properties but there are less data on its effects against oral bacteria, particularly putative periodontal pathogens. Oral health and hygiene as well as salivary biochemical composition are important factors responsible for prevention of gum and dental disorders such as chronic periodontitis. In this research, the inhibitory effect of aqueous extracts and essential oils from leaves of white, green and black tea on growth of *Streptococcus mutans* and other bacteria was determined using a standard susceptibility agar dilution technique. The effects on adhesion of *S. mutans* to glass were also studied.

The lowest inhibitory concentration for black, green and white tea were 0.8 mg mL⁻¹, 4 mg mL⁻¹ and 6 mg mL⁻¹ respectively. Essential oils from some other medicinal herbs including *Thymus*, *Mentha piperita* and *Origanum vulgare* were also tested for their antioxidant and antibacterial activities. Selected extracts were tested against some bacteria such as *Candida albicans* and a few strains of *Enterobacteriaceae* found in plaque samples. It was shown that the inhibitory concentration of all other extracts was ≥ 4 mg mL⁻¹. They all showed moderate to high activity against oral microorganisms *in vitro*. According to the results obtained and taking into consideration the various side effects known for chemical antibacterial mouth washes, it is suggested that many herbal extracts could be used almost unlimited on a daily basis to highly improve oral hygiene and to support other attempts including brushing and use of dental floss.

Key Words

Tea essential oil, antimicrobial, dental caries, oral bacteria, *Streptococcus mutans*.

[P-042]

Chemical profiles and antioxidant activity of essential oils extracted from the leaves, stem and stem bark of *Parkia biglobosa* (Jacq) Benth.

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Abstract

Parkia biglobosa (Jacq.) Benth. (Mimosaceae) is widely used for the treatment of hypertension, diabetes, leprosy, toothache and diarrhea in Nigeria. The leaves, stem and stem bark of the plant were subjected to hydro-distillation to obtain essential oils which were analyzed by high resolution GC/MS. The essential oils extracted from the leaf, bark and stem contained 36, 27 and 18 constituents accounting for 89.97%, 96.32% and 99.9% of the total oil, respectively. Thirty-four compounds were identified while two compounds were unidentified in the leaf essential oil. The major components are limonene (15.96%), hexadecanoic acid (12.48%) and farnesene (10.21%). The GC/MS identified 27 compounds in the stem bark oil, and the predominant compound was 3-furan acetic acid (81.39%). Seventeen compounds were identified in the stem oil, in which the major compounds were caryophyllene oxide (13.77%), β -caryophyllene alcohol (12.13%), α -terpineol (10.82%) and β -caryophyllene (8.13%). Some of these major compounds have been reported to have biological activities. The antioxidant activities of the oils were determined by evaluating their scavenging activities against DPPH, ABTS and nitric oxide radicals. The three assays were concentration-dependent with varying strong antioxidant potentials. The antioxidant activity of the stem oil is not significantly different ($P < 0.05$) from those of vitamin C and rutin, used as standard drugs in this study. The presence of terpenoids and phenolic compounds identified in the essential oils has validated the medicinal use of this plant.

Key Words

Parkia biglobosa, essential oil, antioxidant, free radicals, nitric oxide.

[P-043]

Chemical profiles and antioxidant activities of essential oils extracted from leaf and stem of *Jatropha curcas* L.

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Abstract

Jatropha curcas L. (Euphorbiaceae) is a medicinal plant for the treatment of skin diseases, cancer, diabetes, and piles. The leaves and stem were subjected to hydrodistillation and essential oils obtained were analyzed by high resolution GC/MS. The leaf and stem of the plant contain 14 and 20 compounds accounting for 90.34% and 95.80% of the total oils, respectively. The major monoterpenes in the stem oil were α -pinene (13.52%), β -terpinene (10.59%) and menthol (5.40%) while the major sesquiterpenes were γ -cadinene (5.49%), methyl carvacrol (5.13%), β -caryophyllene (3.18%). The prominent sesquiterpene compounds in the leaf oil were γ -cadinene 14.46%, germacrene D (10.87%), α -aromadendrene 8.32% and caryophyllene oxide 7.95%, while the principal monoterpenoid components were menthol (12.38%), β -citral (11.14%), limonene (6.20%) and α -pinene (5.69%). Antioxidant activity of each oil was examined using DPPH, ABTS, nitric oxide and TBARS free radicals scavenging assays. The four assays were concentration-dependent with varying strong antioxidant potentials. The IC_{50} value of DPPH radical scavenging activity of the leaf and stem essential oils were 3.10 ± 0.01 and 8.80 ± 0.03 mg/mL, while that of ABTS radicals were 6.20 ± 0.02 and 4.6 ± 0.02 mg/mL, respectively. In the TBARS assay, the leaf and stem oils could inhibit 50% lipid damage by 11.50 ± 0.02 and 9.50 ± 0.01 mg/mL, while the IC_{50} value for the nitric oxide free radical were 3.64 ± 0.03 and 3.17 ± 0.02 mg/mL, respectively. The antioxidant activity for the stem and leaf oils on nitric oxide radical were not significantly different ($P < 0.05$) from those of vitamin C and rutin used in this study.

Key Words

Jatropha curcas, essential oil, major compounds, antioxidant, free radicals.

[P-044]

Antimicrobial activities and chemical composition of *Artemisia aucheri* Boiss seeds essential oils

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Abstract

This study was carried out to evaluate antimicrobial activities of seed extracts and essential oil of *Artemisia aucheri* Boiss (Asteraceae) and the chemical composition of seed essential oil. Antimicrobial activity tests were carried out using disc diffusion methods against five bacterial species. The seed essential oil was analyzed by GC/MS. The results showed that essential oil of *Artemisia aucheri* seed was effective against *E. coli*, *S. aureus* and *L. monocytogenes*. The identified essential oil constituents were decane, α -cymene, 1,8-cineol, linalool, α -mentha-8-ol, triene, borneol, lavandulol, bornel acetate, chrysanthenyl acetate, dehydro aromadenderene, and caryophyllene oxide. Most of these compounds are reported in other *Artemisia* species. Variation on composition of *Artemisia aucheri* may be due to the role of environmental parameters on plant growth and metabolite biosynthesis.

Key Words

Artemisia aucheri, essential oil, antimicrobial, seed.

[P-045]

Antifungal activity of the essential oils of cultivated species of *Labiatae* against *Fusarium* sp.

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Abstract

Antifungal activity of essential oils from different cultivated species of *Labiatae* (*Lavandula latifolia* Medik., *L. angustifolia* Mill., *L. x intermedia* Emeric ex Loisel., *Salvia officinalis* L., *S. lavandulifolia* Vahl. and *Thymus vulgaris* L.) against three species of *Fusarium* (*F. solani* (Mart.) Sacc., *F. oxysporum* Schlecht. and *F. moniliforme* Sheld.) has been tested on artificial growth media by the dilution method. *T. vulgaris* essential oil exhibited the most significant antifungal activity (EC_{50} ranged from 0.22 to 0.35 mg/mL). *L. x intermedia* and *L. latifolia* presented also activity, which is similar against *F. oxysporum* (EC_{50} = 0.54 and 0.60 mg/mL, respectively) and against *F. solani* (EC_{50} = 0.62 and 0.65 mg/mL), but lower against *F. moniliforme* (EC_{50} = 0.84 and 0.86 mg/mL). *L. angustifolia*

and both species of *Salvia* presented less inhibitory effect. Chemical compositions of oils were conducted by GC/MS. The main constituents of the essential oil of *T. vulgaris* were thymol (23.5%) and carvacrol (21.4%); the species of *Lavandula* genus were distinguished by their high content in linalool (33.24%, 32.1%, 30.6%) and linalyl acetate (17.2%, 17.1%, 28.1%) in *L. latifolia*, *L. angustifolia* and *L. x intermedia* respectively. Also of note was the concentration of camphor in the two most active species of the genus (17.2% in *L. latifolia* and 10.1% in *L. x intermedia*). 1,8-cineole appeared as the main compound in *S. lavandulifolia* (22.3%) and *S. officinalis* (16.5%) oils, while high content of α -thujone (16.5%) was found in the latter species.

Key Words

Labiatae, essential oil, antifungal activity, *Fusarium*.

[P-046]

Antimicrobial activities of essential oils from some Nigerian local spices

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Abstract

Antimicrobial activities of essential oils from six species from southeast Nigeria: *Zingiber officinale* (Roscoe), *Allium sativum*, (Linn), *Monodora myristica* (Dunal) A. Rich, *Xylopi aethiopica* (Dunal) A. Rich, *Piper guineense* (Schum & Thum), and *Tetrapleura tetraptera* (Taub) were investigated on four fungi and three bacteria using the agar diffusion method. The essential oils were obtained by steam distillation from locally sourced fresh samples. Results of the antifungal tests indicated that the oils showed no inhibition against *Aspergillus* and *Penicillium* species. However, the oils were active against *Candida albicans* in the order of *A. sativum* > *Z. officinale* > *P. guineense* : *M. myristica* > *X. aethiopica* > *T. tetraptera*. For *Candida stellotooides*, inhibition was in the order of *A. sativum* > *M. myristica* > *P. guineense* > *Z. officinale* > *X. aethiopica* > *T. tetraptera*. The antibacterial tests showed that *A. sativum* gave the highest inhibition against two strains of *Escherichia coli* (strains 5210 and 1099), while *Z. officinale* showed the least inhibition. *Staphylococcus aureus* was more significantly susceptible to all the oils, with *A. sativum* also being the most potent. The MBC results obtained were the same as the MIC in most samples. These activities were comparable to those of standard drugs. These results lend credence to the use of these spices, in addition to other culinary purposes, in 'light soups' for the treatment of common cold, amidst other medicinal uses; and in curing and preservation of meat and fish, and may also be exploited for pharmaceutical purposes.

Key Words

Essential oils, spices, antifungal, antibacterial activity.

[P-047]

Antioxidant and acetylcholinesterase inhibitory activity from *Citrus aurantium* L., *Citrus limon* (L.) Burm. f., *Cupressus sempervirens* L., *Eucalyptus globulus* Labill., *Foeniculum vulgare* Mill. and *Thymus vulgaris*

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Abstract

Medicinal and aromatic plants are a part of Morocco's natural heritage, viewed as a potential mean of combating desertification and improving rural economy. To evaluate the potential use of local plants, and/or their extracts as natural bioactive products, it is important to assess their biological properties. *Citrus aurantium* L., *Citrus limon* (L.) Burm. f., *Cupressus sempervirens* L., *Eucalyptus globulus* Labill., *Foeniculum vulgare* Mill. and *Thymus vulgaris* L. essential oils were obtained by steam distillation from a local Moroccan company, and analyzed by GC and GC/MS. The antioxidant activity was evaluated by the TBARS, TEAC, DPPH, reductive power and ORAC methods. The AChE inhibition was likewise measured. *C. aurantium* oil was dominated by linalool (59%) and linalyl acetate (23%) and that of *C. limon* by limonene (99%). α -Pinene (49%), limonene (32%) and δ -3-carene (18%) were *C. sempervirens* oil main compounds. *E. globulus* oil main components were 1,8-cineole (38%) and limonene (55%). *trans*-Anethole (75%) was the main oil component in *F. vulgare* and p-cymene (24%), borneol (16%), carvacrol (16%) and thymol (12%) dominated *Thymus vulgaris* oil. *T. vulgaris* oil was the most effective for preventing lipid peroxidation and scavenging free radicals with IC₅₀ of 0.1 mg/mL (TBARS), 0.01 mg/mL (TEAC) and 0.2 mg/mL (DPPH). *T. vulgaris* essential oil showed also the best results with the reductive power and ORAC methods. AChE IC₅₀ inhibitory activity was of 0.1 mg/mL (*E. globulus*), 0.2 mg/mL (*T. vulgaris*), 0.3 mg/mL (*C. sempervirens*), 0.8 mg/mL (*C. limon*) and 1.2 mg/mL (*F. vulgare*). Evaluated main oil components showed IC₅₀ of 0.1 mg/mL (carvacrol, 1,8-cineole and borneol) and 0.6 mg/mL (limonene).

Key Words

Citrus aurantium, *Citrus limon*, *Cupressus sempervirens*, *Eucalyptus globulus*, *Foeniculum vulgare*, *Thymus vulgaris*, Morocco, essential oil, antioxidant, acetylcholinesterase inhibitors.

[P-048]

Essential oil from Mexican desert plants as antibacterial agents in agriculture

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Abstract

The North American desert ecosystem plays a very important role, not only for the wide area occupied but also for its specific characters and species diversity. Diverse efforts to explain the evolution and phylogenetic that have created the peculiarity of these deserts have been performed. On the other hand, to generate new uses and applications of native plants, researches have been carried out that relate the plants with biological activities. Far from being abandoned and unproductive areas, deserts are biologically, economically and culturally dynamic. In this work, essential oils were extracted from Mexican desert plants; Gobernadora [*Larrea tridentata* (DC.) Coville], Hojasen [*Flourensia cernua* DC.] , Mexican Oregano [*Lippia berlandieri* Schauer], Sangre de Drago [*Jatropha dioica* Sesse] and Damiana [*Turnera diffusa* Willd. ex Schult] essential oils were characterized physically and chemically; and antioxidant activity was evaluated by DPPH and ABTS tests. Antifungal activity against fungal plant pathogens: *Alternaria alternata*, *Colletotrichum gloeosporioides*, *Rhizoctonia solani*, *Fusarium oxysporum*, *Sclerotinia sclerotiorum* and *Slerotium cepivorum* was assessed a concentration (1 oil : 500 diluents). All oils showed antioxidant activity of 100% inhibited the growth of microorganisms in 100%, result excellent materials for use as antifungal agents against phytopathogens. This research leads to applications of these antimicrobial oils in agricultural products.

Key Words

Essential oil, Mexican desert, *Larrea tridentata*, *Flourensia cernua*, *Larrea tridentata*, Oregano, *Jatropha dioica*, *Turnera diffusa*.

[P-049]

Chemical composition, olfactory analysis and antibacterial activity of *Thymus vulgaris* L. chemotype “thujanol”

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Abstract

In continuation of our research about various chemotypes of *Thymus vulgaris* L., “thujanol-one” was examined. The aim of this investigation was the analysis of the composition and antibacterial activity against some selected microorganisms. The used essential *T. vulgaris* L. ct “thujanol” oil was cultivated in the South of France and analyzed by GC-FID and GC/MS on polar and apolar columns. The following major compounds could be detected: *cis*-sabinene hydrate (32.7%), limonen-10-ol (9.7%), terpinen-4-ol (6.5%) and *trans*-sabinene hydrate (5.5%). The values are in accordance with existing literature. The oil was olfactorily evaluated by two professional perfumers and two aroma chemists. The odor was characterised as aromatic, herbal-balsamic and soft camphorous. The antimicrobial activity of the essential oil was tested against eleven different genera of Gram-positive and Gram-negative bacteria by using serial broth dilution method. It could be verified that the essential oil possesses high antimicrobial activity against both strains of Gram-positive *Staphylococcus aureus* and Gram-negative *Escherichia coli*, as well as three strains of *Salmonella abony*, but less against two strains of *Pseudomonas aeruginosa* and *Ps. fragi*.

Key Words

Thymus vulgaris ct “thujanol,” antimicrobial activity, *Staphylococcus aureus*, *Escherichia coli*, *Salmonella abony*, *Pseudomonas aeruginosa*.

[P-050]

Studies on the “negramina” plant (*Siparuna guianensis* Aubl-Monimiaceae), a source of essential oils and insect repellent

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Abstract

The negramina plant (*Siparuna guianensis* Aubl –Monimiaceae) is rich in essential oil. Studies were performed: occurrence in natural areas, popular usage, crude leaf extract yield and bioactivity of negramina in southwestern Mato Grosso (MT-sw), Brazil, during 2003-2009. In MT-sw the negramina occurs in savana, grassland and mixed in thin wooded areas. The size of the plant varies from shrubs to trees clustered, according to the soil. It has important usefulness for local people. Its leaves, fruit and bark are used for medicinal purposes, mystics and repellency of insects. The plant provides essential oil with strong and lasting odors. Leaves, when rubbed on the body, impregnate the odor and repel insects. Smoke from bonfires met with green branches and leaves of negramina also prevent hematophagous nearby. In mystical rites, according to local culture, the odors and smoke from negramina keep away evil spirits. The old residents use the plant against headache (washing head with leaf aqueous of the plant) and sinusitis (inhaling the plant volatile substances). Women make intimate wash with water by boiling the leaves. The yield of crude metabolic extract mass (EMe) derived from fresh leaves of negramina (FFN), was: 2.1998% EMe towards mass FFN and 5.8467% EMe towards dried leaf mass (FDN). It was found in nursery conditions, which the possible arrangements of ten species of medicinal plants did not affect allelopathically to negramina, but this affected others: inhibited the growth of catuaba (*Anemopaegma arvense*- Bignoniaceae) and less intensely, also the node-to-dog (*Heteropterys aphrodisiaca* - Malpighiaceae). Negramina's potential needsto be studied further

Key Words

Medicinal plant, essential oil plant, insect repelent potential, allelopathy induction, Mato Grosso state, Brazil.

[P-051]

Antifungal activity of essential oils from Citrus spp. against fungi growing on bread

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Abstract

The antifungal activity of eight essential oils obtained in a Citrus juice plant, located in the State of São Paulo, Brazil, was evaluated. The oils were obtained in different steps from industrial juice processing of orange and tahiti acid lime. The oils were named Orange Peel, Orange Oil Phase Essence, Orange Peel Oil Five Fold, Orange Terpenes, Citrus Essence, Tahiti Lime Oil Phase, Tahiti Lime Peel Oil e Tahiti Lime Waterphase. The composition of oils was obtained by gas chromatography and mass spectroscopy. Seven fungal species were used in the evaluation (*Penicillium* sp., *Aspergillus* sp., *Penicillium citrinum*, *Aspergillus flavus*, *Eurotium* sp., *Eurotium chevalieri*). The species were isolated from bread samples collected in the market and which showed fungal growth after a period of storage in two different temperatures (15°C and 30°C). The minimum inhibitory concentration (MIC) was evaluated for each oil and species growing in 100 µL of PDA (Potato Dextrose Agar) broth maintained at 25°C for 18 h. Fungal growth was monitored by esterase activity. Esterase production was verified by the conversion of fluorescein diacetate to the fluorescent dye fluorescein (Chand et al., 1994) according to Hadacek & Greger (2000) methodology. The fluorescence readings were performed by the reader Victor X3 (PerkinElmer) operating with excitation and emission of 485 nm and 635 nm, respectively. Inhibition curves showed that MIC values were obtained and that all oils had antifungal activity against fungal species. The intensity of inhibition was dependent of oil and species.

Key Words

Antifungal activity, bread, Citrus essential oil, juice processing.

[P-052]

Blood pressure lowering action of essential oil component from *Ocimum basilicum*

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Abstract

Ocimum basilicum L. belongs to the family Lamiaceae and commonly known as Basil (Tulsi). It is a widespread plant cultivated around the world. In Indo-China, the ashes of the roots are suggested as a remedy for skin disease. The plant is used as aromatic, antimicrobial, astringent in dysentery, while the leaves are antipyretic. The seeds are laxative, particularly in case of habitual constipation. The juice of the leaves and flowers are a treatment of cough. A decoction may be given after parturition as emmenagogue and febrifuge. The leaves are carminative, antispasmodic and sedative. Preparations of basil are used for supportive therapy for feeling of fullness and flatulence, for the stimulation of appetite and digestion, and as diuretic.

In anaesthetized rats, methanolic extract, fractions, and pure compound eugenol (0.3-3.0 mg/kg) produced dose-dependent fall in blood pressure and heart rate. These effects were not blocked by atropine (1 mg/kg) and eugenol did not modify pressor response of norepinephrine which rules out the possibility of cholinergic stimulation or α -adrenergic blockade. In spontaneously beating atria, eugenol caused decrease in force and rate of contractions. These effects remain unaltered in the presence of atropine. In rabbit aorta, eugenol caused relaxation of norepinephrine and potassium induced contractions in a concentration-dependent manner. These results suggest that the direct relaxant action of eugenol on myocardium and on blood vessels may be responsible for its hypotensive and bradycardiac effects observed in the *in vivo* studies.

Key Words

Ocimum basilicum, Lamiaceae, Basil (Tulsi), hypotensive, bradycardiac.

[P-053]

Chemical composition and antimicrobial activity of the essential oil of *Ocimum basilicum* L. growing in Iran

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Abstract

Ocimum basilicum L. is an aromatic medicinal plant belonging to the Lamiaceae family. In this study, we investigated the main chemical constituents of volatile oils from leaves of *O. basilicum* and antimicrobial screening of native plant in the northern region of Iran. The essential oil from the leaves of *O. basilicum* L. were analyzed by GC/MS and also evaluated for antimicrobial activity against Gram-positive (*Staphylococcus aureus*, *Bacillus* spp.) and Gram negative (*Escherichia coli*, *Pseudomonas aeruginosa*, *Salmonella typhi*, *Klebsiella pneumoniae*, *Proteus mirabilis*) bacteria and a pathogenic fungus (*Candida albicans*). The disc diffusion method was used to evaluate the zone of microbial growth inhibition at various concentrations of the oil. The MIC and MBC values of the oil were determined. The oil was found to have an antimicrobial effect. GC/MS analysis of the oil revealed 31 components. The oil was dominated by monoterpenes, which accounted for 92.48%. This monoterpene fraction was characterized by a high percentage of eugenol (62.8%). The other major monoterpenes were methyl eugenol (11.91%), *cis*-ocimene (7.27%), *trans*-ocimene (1.04%), α -pinene (7.17%) and camphor (4.15%). The major ses-

quiterpenes of the oil were germacrene D (5.15%) and *trans*-caryophyllene (1.11%). The minor sesquiterpenes were α -farnesene (0.21%) and β -bisabolene (0.34). It can be concluded that the essential oil of *O.basilicum* demonstrated appreciable antimicrobial activity.

Key Words

Antimicrobial activity, *Ocimum basilicum*, native herbs.

[P-054]

Relationship between Emotional Behavior and Brain Distribution of Inhaled Essential Oil

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Abstract

The influence of essential oil on emotional behavior requires further scientific clarification. Here, we introduce our recent research about the influence of essential oil and its underlying mechanism on emotional behavior. In inhaled administration, it is thought that the effect is the result of two routes. One is neurological transmission through olfactory receptors, while the other is parenteral transmission through the bloodstream. Neurological transmission is thought to be an action of short duration. Therefore, it has been thought that the influence on emotional behavior occurs by parenteral transmission. We researched the mechanism of action by using the essential oils from *Alpinia zerumbet* and *Abies sachalinensis*. A time-dependent potentiation of the anxiolytic-like action of essential oils from *Alpinia zerumbet* was observed from 0 to 150 min. From these results, with respect to inhaled administration of essential oil, it was suggested that the effects are observed because the components reach the brain through a parenteral route. In addition, direct neurological transmission might play a role in mediating the effects of essential oil.

Key Words

Emotional behavior, brain distribution, inhalation, *Alpinia zerumbet*, *Abies sachalinensis*.

[P-055]

The Bioactive Essential Oil of *Stachys lavandulifolia* Vahl. subsp. *lavandulifolia*

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Abstract

The subcosmopolitan genus *Stachys* comprises about 300 species throughout the world and one of the largest genera of the Lamiaceae. Turkey is one of the richest countries in *Stachys* diversity and it is represented by 83 species at a level of 48% endemism. *Stachys lavandulifolia* Vahl. subsp. *lavandulifolia* is widely used by the people of south Anatolia. An infusion of the flowers is used for the treatment of gastrointestinal and respiratory disorders. In folk medicine it is also well known as an appetizer, carminative, stimulant, diuretic and against sore throat. Aerial parts of *S. lavandulifolia* collected from Antalya were hydrodistilled to obtain an essential oil which was analyzed simultaneously by GC and GC/MS systems. Thirty-seven compounds representing 98.3% of the oil were characterized. β -phellandrene (27%), α -pinene (18.5%) and germacrene-D (13%) were found to be major components of the oil. Anticandidal and antibacterial effects of the oil were evaluated against several pathogenic *Candida* and bacteria clinical and standard strains by using microdilution broth method. The oil showed good inhibitory effect against *Candida tropicalis* (MIC of 0.0937 mg/mL). *S. aureus* and *Salmonella typhimurium* also inhibited by the oil at a concentration of 0.375 mg/mL, supporting its antimicrobial use.

Key Words

Stachys lavandulifolia, Lamiaceae, antibacterial, essential oil.

[P-056]

Antibacterial activity of volatile constituents of Citrus peel oil from Algeria; application in *Sardina pilchardus*

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Abstract

The essential oils (EOs) extracted by hydrodistillation from *Citrus* peels of *C. limonum* Risso, *C. sinensis* Pers. and *C. aurantium* L. were analyzed by gas chromatography coupled with mass spectrometry (GC/MS). The major constituents were limonene (77.37%) for *C. sinensis* EO; linalyl acetate (37.28%) followed by linalool (23.36%) and citral (10.45%) for *C. aurantium* EO and limonene (51.39%), β -pinene (17.04%), γ -terpinene (13.46%) and α -pinene (3.07%) for *C. limonum* EO. EOs were screened for their ability to inhibit the growth of *Staphylococcus aureus*, *Pseudomonas aeruginosa*, *Salmonella enteritidis* and *Escherichia coli* using the standard agar-disk diffusion

assay. Results obtained from disk-diffusion method, followed by measurements of Minimal Inhibition Concentration (MIC), indicated that *C. sinensis* was the most active, with the lowest MIC value against pathogenic bacterias. EOs were tested in *Sardina pilchardus* stored at $4\pm 2^{\circ}\text{C}$, experimentally inoculated with foodborne pathogen at level of 5×10^5 cfu/g. All bacteria counts in treated samples were less ($p < 0.05$) compared to controls at different intervals during storage. The results of the bioassays, together with the chemical profile of the EOs, support the possibility of using all EOs as potent natural preservatives to contribute in the reduction of experimentally inoculated pathogenic bacteria in fish

Key Words

Citrus essential oils, GC/MS, antibacterial activity, CMI, *Sardina pilchardus*.

[P-057]

Evaluation of antifungal activities of essential oils extracted from *Inula viscosa*

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Abstract

Several studies have revealed that *Inula viscosa* (L.) Aiton contains active principles that could have potent bioactive properties and pharmacological functions. Different extraction processes of active principles from *Inula viscosa* are carried out in this work: hydrodistillation, steam distillation and supercritical fluid extraction (SFE) with CO_2 . The obtained yields in the extraction process were 7.32% (SFE), 0.336% (steam distillation), 0.163 % (hydrodistillation). The yield of CO_2 extract was potentially the highest. Moreover, the steam distillation extract was analyzed by GC/MS to determine its composition. It was shown to contain iso-costic acid (56.71%), fukienol (14.6%) and Nerolidol (0.56%). The antifungal properties of steam distillation extract were investigated with antifungal tests against two fungal species. The extracted oil showed positive activities against these fungal species with the various concentrations 3120 $\mu\text{g/mL}$, 1560 $\mu\text{g/mL}$ and 780 $\mu\text{g/mL}$.

Key Words

Inula viscosa, antifungicidal activity, essential oil, supercritical fluid extraction, steam distillation.

[P-058]

***Achillea biebersteinii* Essential Oil: A Fragrant Herb used in Saudi Traditional Medicine**

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Abstract

Aromatic plants and their essential oils are valuable natural products used in many fields, including perfumes, cosmetics, phytotherapy, aromatherapy and nutrition. *Achillea biebersteinii* Afan. (Family: Asteraceae), locally known as althufra, is a native to Arabian peninsula, including Saudi Arabia. The aerial parts of the plant in the form of decoction are commonly used by folkloric practitioners for the treatment of various illnesses, including abdominal pain and as a diuretic, wound healing, curing diarrhea and jaundice. The aim of this study was to determine the possible protective effect of the essential oil (obtained by hydrodistillation) of *A. biebersteinii* on carbon tetrachloride CCl₄ induced liver injury and oxidative stress in Wistar albino rats. The animals were administered the *A. biebersteinii* (I.P) at 0.2 mL/kg body weight for 7 consecutive days. GOT, GPT, ALP, GGT and bilirubin were estimated in serum; whereas, non-protein sulfhydryl (NP-SH), total protein (TP) and malondialdehyde (MDA) were estimated in liver tissue as markers of oxidative stress. Histopathological assessment was also done on liver tissue. CCl₄-induced liver toxicity was evident by an elevation of liver marker enzymes and a fall in the activities of NP-SH, TP and an increased level of MDA concentration. *A. biebersteinii* administration for 7 days prevented the CCl₄ induced hepatic injury and oxidative stress. In conclusion, it was observed that essential oil of *A. biebersteinii* protects the liver against CCl₄ induced hepatic injury through its potent antioxidant activity in rats, and substantiates its use in liver disorders in Arab traditional medicine. Results will be presented.

Key Words

Achillea biebersteinii, essential oil; CCl₄, hepatoprotection, oxidative stress.

[P-059]

Effect of diethyl maleate on toxicity of linalool against two stored product insects in laboratory conditions

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Abstract

Many plant secondary metabolites, such as alkaloids, monoterpenoids or phenylpropanoids, are toxic to insects; in addition, essential oil extracted from plants have been widely investigated for pest control properties, with some proving to be toxic in insect pests. In this research, the effect of diethyl maleate on the toxicity of monoterpenoids such as linalool was studied against two most common stored-product insects: *Callosobruchus maculatus* and *Rhyzopertha dominica*. Diethyl maleate was combined in mass ratios (1:8 and 1:4) with acetone used and applied on *C. maculatus* and *R. dominica* adult. Five concentrations of linalool were tested with four replicated at 24 h and 48 h with 30 adult insects in each replication. After 24 h of exposure, the LC₅₀ values were estimated to be 23.6 and 31.01 µl/L air and after 48 h were 15.07 and 21.84 µl/L, respectively for each insect. Linalool with synergist after 24 h of exposure, the LC₅₀ values were estimated to be 11.93 and 13.07 µl/L air and after 48 h were 7.38 and 7.93 µl/L, respectively for each insect. It was found that synergist is able to block the specific system of enzymes involved in selection of tolerance in susceptible generations. Diethyl maleate (DEM) was shown as an inhibitor of glutathione S-transferase (GST) activity. These results show diethyl maleate decreasing doses of linalool and enhancing the activity of linalool.

Key Words

Diethyl maleate, linalool, LC₅₀, synergism, *Rhyzopertha dominica*, *Callosobruchus maculatus*.

[P-060]

A comparison between insecticidal efficacy of a novel pellet formulation and an obsolete one, Phostoxin

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Abstract

Due to insect resistance to common fumigants such as Phostoxin, and some arguments about the genotoxicity of this pellet, production of alternative pellets with same application seems necessary. The aim of this work was comparison between insecticidal efficacy of Phostoxin and a novel pellet based on eucalyptol, botanical constituent, and poly(vinyl alcohol) against *Tribolium castaneum* (Herbst), *Callosobruchus maculatus* F. and *Rhyzopertha dominica* F. in 28±2 °C, 24 h and 48 h exposure time. Mortality percent in 24 h ± S.E. of 0.25 and 0.3 gr Phostoxin pieces in 159 L barrels against more tolerant (*R. dominica*) and more susceptible (*T. castaneum*) species were 33.3±4.4, 90±4.4, 45±2.8 and 100, respectively. Mortality percent in 24 h ± S.E. of 3 and 4 g novel pellets (with 3:5 dose) in 20 L casks against more tolerant (*T. castaneum*) and more susceptible (*C. maculatus*) species were 0, 8.33±4.4, 35±2.8 and 48.33±4.4, respectively. In general, Phostoxin pellets were more effective than novel pellets, but results showed that eucalyptol-based pellets have potential toxicity to surrogate instead of obsolete Phostoxin pellets.

Key Words

Phostoxin, eucalyptol, PVA, fumigant toxicity, stored products pests.

[P-061]

Chemical composition and antimicrobial activity of the essential oil from aerial parts of *Origanum libanoticum* Boiss. growing wild in Lebanon

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Abstract

The family Lamiaceae includes about 260 genera and 7,000 species widely distributed in the Mediterranean area. The genus *Origanum*, one of the most economically important of this family, is characterized by extensive morphological and chemical diversity: based on morphological characters, Ietswaart has divided it into 42 species and 49 taxa. Several studies have shown that *Origanum* species possesses antimicrobial, antifungal, antioxidant and insecticidal properties, and they are widely used all over the world as very popular spices. *Origanum libanoticum* Boiss. is a rare plant growing wild in Lebanon. No report on the essential oil of this species has been found in the literature so far. Therefore, as a continuation of our research on plants from Lebanon, we report on the chemical composition of the essential oil of aerial parts of *O. libanoticum* collected on July 2008 in Laklouf, Lebanon. The oil from air-dried and ground aerial parts of the plant was isolated by hydrodistillation for 3 h, using a Clevenger-type apparatus according to the method recommended in the European Pharmacopoeia. The GC and GC/MS analyses on a HP 5MS column evidenced 82 constituents, representing 87.3% of the total oil. The most abundant compounds were thymol methyl ether (10.0%), (E)- β -caryophyllene (7.7%), linalool (6.5%), carvacrol (4.1%), caryophyllene oxide (4.0%), carvacrol methyl ether (2.8%) and thymol (2.0%). The *in vitro* activity of the essential oil against some microorganisms in comparison with gentamicin by the broth dilution method was determined. The oil exhibited a good activity as inhibitor of growth of Gram-positive bacteria.

Key Words

Origanum libanoticum, essential oil, antibacterial activity.

[P-062]

Composition and antifungal activity of the essential oil of *Artemisia aucheri* Boiss. from Iran

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Abstract

Artemisia aucheri Boiss. is a shrub from the Asteraceae family that spread all over Iran. In traditional medicine, *A. aucheri* is a plant with astringent properties, disinfectant, antimicrobial, antiparasite and antioxidant activity. The aim of this study was to evaluate the antifungal potential of the oils from *Artemisia aucheri* against four post-harvest pathogenic fungi (*Rhizopus stolonifer*, *Penicillium digitatum*, *Aspergillus niger*, and *Botrytis cinerea*) which can reduce the shelf life of some fruits. The essential oil was extracted through hydrodistillation of air-dried samples, and its chemical composition was determined through GC/MS. Antifungal assays were carried out *in vitro* using potato dextrose agar (PDA) plates. Minimum inhibitory concentration (MIC) and minimum fungicidal concentration (MFC) of the essential oil were determined. GC analyses resulted in the identification of 39 components, representing 96.3% of the constituents of the oil. The major constituents of *A. aucheri* were linalool (41.6%), geranyl acetate (11.4%) and (E)-citral (10.7%). The essential oil of *A. aucheri* had great potential of antifungal activity against all of the four fungi tested. *A. aucheri* oil showed promising inhibitory effects at concentrations of 1,800 $\mu\text{L L}^{-1}$, against *B. cinerea* and *R. stolonifer*, whereas they had antifungal effects against *P. digitatum* and *A. niger* at 2,400 $\mu\text{L L}^{-1}$. The essential oil of *A. aucheri* showed fungicidal effect against *B. cinerea* and *R. stolonifer* with the MFC of 2,400 $\mu\text{L L}^{-1}$. No fungicidal effect was observed against *A. niger*, even at high concentrations (2,400 $\mu\text{L L}^{-1}$).

Key Words

Artemisia aucheri, antifungal activity, post-harvest fungi.

[P-063]

Essential oil composition and the evaluation of antimicrobial activity of a new species: *Hypericum sechmenii* Ocak & Koyuncu from Eskişehir-Turkey

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Abstract

The aerial parts of *H. sechmenii* Ocak & Koyuncu were collected at full flowering stage from Eskişehir, Arayit Mountain (1750-1820 m). Essential oil of the plant was obtained by hydrodistillation and analyzed by GC and GC/MS. Essential oil yield was 0.06%. Sixty components (90.5%) of the oil were identified. The major essential oil

components of *H. sechmenii* were germacrene D (14.9%), β -elemene (13.2%) and bicyclogermacrene (7.4%). The *in vitro* antimicrobial activity of essential oil was evaluated through disc diffusion assay. Stock solution of essential oil was prepared with DMSO at the 318 mg/mL concentration level. Each disc (5 mm in diameter) contained 10 μ L of stock solution of essential oil. Antimicrobial activity tests were performed with four Gram-positive (*Bacillus cereus*, vancomycin-resistant *Enterococcus* sp., *Staphylococcus aureus*, methicillin-resistant *S. aureus*), three Gram-negative (*Escherichia coli*, *E. coli* O157:H7, *Pseudomonas aeruginosa*) bacteria and two yeasts (*Candida albicans*, *Candida glabrata*). However, the essential oil of *H. sechmenii* did not show antimicrobial effect against tested strains.

Key Words

Hypericum sechmenii, essential oil, antimicrobial activity.

[P-064]

Volatile Compounds of Five Endemic *Aristolochia* Species from Turkey

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Abstract

The genus *Aristolochia* (Aristolochiaceae) consists of about 500 species throughout the world. They are used for stomach ache, abdominal pain, rheumatism, intestinal worms, wounds and skin diseases and cholera. Furthermore they are known as abortifacient, emmenagogue, sedative, analgesic, anticancer, anti-inflammatory, antifeedant, muscle relaxant and antihistaminic. In Turkey, *Aristolochia* is represented by 27 species and 15 of these are endemic. In the present study, aerial parts of *A. stenosiphon* Davis & Khan, *A. poluninii* Davis & Khan, *A. guichardii* Davis & Khan, *A. krausei* P.H. Davis and *A. baseri* Malyer & Erken were hydrodistilled to obtain their essential oils and they were analyzed by GC-FID and GC/MS systems, simultaneously. The main constituents of the oils are as follows: *A. stenosiphon*: spathulenol 20.9%, hexadecanoic acid 19.1%, bornyl acetate 7.1%, limonene 6.4% and α -pinene 5.7%; *A. poluninii*: hexadecanoic acid 38.4%, caryophyllene oxide 11.6% and spathulenol 9.3%; *A. guichardii*: β -pinene 20.9%, α -pinene 10.6%, hexadecanoic acid 8.3% and caryophyllene oxide 5.2%; *A. krausei*: hexadecanoic acid 37.1%, bornyl acetate 6.2%, caryophyllene oxide 6.1% and β -bisabolene 5.1%; *A. baseri*: spathulenol 16.3%, caryophyllene oxide 12.6%, bornyl acetate 9.5%, hexadecanoic acid 7.5% and β -caryophyllene 6.8%.

Key Words

Aristolochia, essential oil, GC-FID, GC/MS.

[P-065]

Fungal disease management of button mushrooms through application of essential oils

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Abstract

Natural products, including essential oils, have been investigated as alternative biocides to minimize the use of synthetic fungicides. Mushroom production is heavily reliant on a limited number of synthetic crop protection products to control outbreaks of fungal pathogens. In this study, the application of essential oils (*Thymus vulgaris*, *Cymbopogon citratus*, *Lippia citriodora*) and their major components were evaluated for their ability to control fungal pathogens of *Agaricus bisporus*. Initially, *in vitro* assays were conducted using nutrient medium supplemented with essential oil or terpenoids against *Mycogone perniciosus* (wet bubble disease), *Cladobotryum dendroides* (cobweb disease) and *Trichoderma* sp. The *in vitro* effects of the oils on the growth of *Agaricus* were also established. Thyme oil was found to substantially inhibit the growth of the pathogens while demonstrating lower toxicity towards *Agaricus*. Subsequently, *in vivo* preventative and curative studies were conducted using all three oils against *Mycogone* and *Cladobotryum* by inoculating mushroom casings with the respective pathogens. Although thyme oil was highly effective when applied as a preventative treatment, the curative application of the oils was less effective. This was attributed to the high spore loads applied (10^6 spores/mL). Such high spore loads would be unlikely to occur in a commercial environment. Thyme oil successfully controlled disease development when the spore load was reduced (10^3 spores/mL). Commercial trials to establish the effect of the oils on mushroom yield indicated that the oils did not affect the yield, taste or texture of the mushroom and could be an option for fungal control.

Key Words

Button mushroom, thyme oil, *Agaricus bisporus*, cobweb disease, wet bubble.

[P-066]

Antioxidant properties, essential oil compositions, phenolic and flavonoid contents of *Elionurus hensii* from Congo

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Abstract

Elionurus hensii K. Schum. (Family: Poaceae) is a perennial plant found in the tropical zones of Congo, Gabon and Angola. Infusions of *E. hensii* are used in Congo to treat aches and influenza-like conditions. To the best of our knowledge, there has been only one report on the essential oil composition, but no data was available on the antioxidant activities of *E. hensii* essential oil and solvent extracts. The objective of this study was to characterize the chemical composition of *E. hensii* essential oil. The antioxidant activities of this species were also evaluated to suggest it as a new potential source of natural antioxidants. Analysis of the chemical composition of *E. hensii* essential oil was carried out using GC and GC/MS. The aerial part essential oil was dominated by oxygenated monoterpenic compounds with two menthane alcohol stereoisomers as the main compounds, while root essential

oil was dominated by oxygenated sesquiterpenes and the major component was aristolone. The amounts of total phenolics and flavonoids in the solvent extracts (dichloromethane, ethyl acetate and methanol) were determined spectrometrically. The essential oil did not reveal any significant antioxidant properties regarding DPPH and ABTS•+ scavenging ability, and was only slightly active for Fe³⁺ reducing power. Conversely, the antioxidant activity of aerial part extracts increased in the order: ethyl acetate > methanol > dichloromethane; whereas the root antioxidant activity increased in the order: methanol > ethyl acetate > dichloromethane. Finally, a relationship was observed between the antioxidant activity potential and total phenolic and flavonoid levels of the extract.

Key Words

Elionurus hensii, essential oil, solvent extract, antioxidant activity, GC/MS, DDPH, ABTS.

[P-067]

Biological Activity and Constituents of the Essential Oil from *Gypsophila bicolor* Flower, from Iran

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Abstract

The plants of genus *Gypsophila* (Caryophyllaceae) are well known by their medicinal, decorative and industrial application. This genus comprised of about 36 species is described in the flora of Iran, among which 18 are endemic. The species of *Gypsophila* are studied for saponins, flavonoids and sterols. *Gypsophila saponins* have anticarcinogenic properties, including direct cytotoxicity, immune- modulating effects, and normalization of carcinogen induced cell proliferation. The chemical composition of the flower of *Gypsophila bicolor* Grossh. (Caryophyllaceae) growing in Iran was obtained by hydrodistillation and analyzed by GC and GC/MS. Twenty-two constituents representing (98.5%) of the flower oil were identified of which germacrene-D (21.2%), p-cymene (20.6%), bicyclogermacrene (17.6%), γ -dodecadienolactone (13.7%) and terpinolene (9.4%) were major components. The antimicrobial effect of flower essential oil from *Gypsophila bicolor* was studied according to the agar diffusion cup method. The test microorganisms used were the Gram-positive bacteria *Bacillus pumilus*, *B. subtilis*, *Staphylococcus epidermidis* and *S. aureus*; Gram-negative bacteria *Pseudomonas aeruginosa*, *Escherichia coli* and *Salmonella abony*, the yeast *Saccharomyces cerevisiae* and *Candida albicans* and the fungi *Penicillium* sp., *Aspergillus niger*, *Botrytis cinerea* and *Rhizopus nigricans*. The essential oil had a mean effect on the Gram-positive and Gram-negative bacteria and had a substantial fungicidal effect on the fungi under study.

Key Words

Gypsophila bicolor, Caryophyllaceae, essential oil composition, germacrene-D, p-cymene, bicyclogermacrene, γ -dodecadienolactone, sabinyl acetate, terpinolene, antimicrobial effect.

[P-068]

Ovicidal effect of essential oil vapours of *Mentha pulegium* L. (Labiatae) and *Thuja orientalis* L. (Cupressaceae) plants against two-spotted spider mite, *Tetranychus urticae* Koch (Acari: Tetranychidae)

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Abstract

The two-spotted mite *Tetranychus urticae* Koch is an economically important pest of greenhouse and ornamental plants in the world. This pest is commonly controlled by application of synthetic acaricides. The number of confirmed resistant mite species to synthetic pesticides has continued to rise. Spider mites have evolved resistance to >80 acaricides to date, and resistance has been reported from >60 countries. The risk of developing pest resistance and the high cost-benefit ratio of synthetic pesticides pushed research towards investigating alternative pesticides. In the search for alternatives to conventional pesticides, essential oils and extracts from aromatic plants have been widely investigated. In the present study, the acaricidal potential of two essential oil vapors from *Mentha pulegium* and *Thuja orientalis* plants were tested against the egg of *T. urticae*. Leaves of *M. pulegium* and *T. orientalis* at flowering stage were collected from various localities of Tehran. Air-dried plant materials were distilled for 8 h using a circulatory Clevenger-type apparatus. The essential oil was dried over anhydrous sodium sulphate and stored at +4°C until tested. The amounts of the essential oils applied were 0.1, 0.5, 1.5, 14 µL in each vial with 140 mL capacity corresponding 0.7, 3.57, 10.71, 100 µL/L air. The essential oil vapours of these two plant species tested were toxic to egg of *T. urticae*. In all cases, mortality of mite eggs was increased with increases the concentration. The essential oil vapor of two plants species had the highest mortality 100 µL/L air dose respectively and at 24 h of exposure.

Key Words

Ovicidal, essential oil, *Mentha pulegium*, *Thuja orientalis*, *Tetranychus urticae*.

[P-069]

Chemical Composition and Antibacterial Activity of *Helichrysum armenium* DC. Growing Wild in Two Localities from Iran

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Abstract

The genus *Helichrysum* Mill. is represented in the flora of Iran by 19 species of which eight are endemic. Some species of *Helichrysum* are used as medicinal plants. In this work, the aerial part of *H. armenium* DC. was collected on July 2010 in Khalkhal area (sample A) at an altitude of 1600 m near Lonbar village, and sample B was collected from Ardabil area at an altitude of 2100 m in northwest Iran. The aerial part of samples were air-dried at room temperature for ten days then subjected to hydrodistillation for 4 h, using a Clevenger-type apparatus. The analysis of oils was performed using GC and GC/MS. Twenty-three constituents representing 97.3% of the essential oil of sample A and twenty-one components (96.8%) of oil from sample B were identified. The oil of sample A was characterized by a higher amount of pulegone (51.9%), piperitenone (31.8%), mentone (6.5%) and thymol (4.7%);

whereas, the main components of the oil from sample B were piperitenone (34.9%), pulegone (31.8%), β -cubebene (13.6%), α -pinene (7.9%), mentone (4.5%) and thymol (3.9%). The antibacterial activity of the oil from leaf was assessed against *Staphylococcus aureus* and *Escherichia coli*. A significant antibacterial activity was determined with the agar diffusion method. The results indicated a moderate activity on *S. aureus* and *E. coli*.

Key Words

Compositae, *Helichrysum armenium*, pulegone, piperitenone, β -cubebene.

[P-070]

Chemical Composition and Antimicrobial Activity of Essential Oils of *Matricaria chamomilla* L. Grown in Neyshabur in Iran

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Abstract

Matricaria chamomilla L. genus is one of the important plants in Iran, some of which are endemic. In this study *Matricaria chamomilla* L. was collected from Neyshabur in Iran, and the chemical constituent of the essential oil of *Matricaria chamomilla* L. was determined. Aerial parts (200 g) were subjected to hydrodistillation in a Clevenger-type apparatus until there was no significant increase in the volume of the oil collected (2.5 h). The yield of the yellow oil was 0.9% (w/w). The essential oil was analyzed by GC and GC/MS. Identification of the components was based on GC retention indices computer matching with Wiley GC/MS library, and by comparison of the fragmentation patterns of the mass spectra with those reported in the literature. Forty-two components were identified, constituting more than 83% of the oil. p-Cymen-8-ol (0.71%), azulene (0.41%), p-cymene (1.1%), 1,8-cineole (2.1%), artemisia alcohol (0.2%), β -elemene (0.86%), cis- β -farnesene (0.93%), trans- β -farnesene (5.20%), borneol (0.82%), γ -cadinene (0.43%), δ -cadinene (0.22%), spathulenol (9.42%), γ -eudesmol (1.50%), α -bisabolol oxide B (7.00%), α -bisabolol oxide A (8.50%), α -bisabolol (5.00%), and germacrene D (0.82%) were major components in *Matricaria chamomilla*. The oil was evaluated against three strains of bacteria (Gram-positive and Gram-negative) and yeasts. The oil showed mild to moderate antimicrobial activity associated mainly with Gram-positive and Gram-negative bacteria and yeasts.

Key Words

Matricaria chamomilla L., Neyshabur, essential oil, antimicrobial activity, chemical constituent.

[P-071]

Chemical composition and *in vitro* antifungal and antioxidant activity of the essential oil and methanolic extract of *Chenopodium album*

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Abstract

In recent years there has been an increasing interest in the use of natural substances, and some questions concerning the safety of synthetic compounds have encouraged more detailed studies of plant resources. Essential oils, and volatile products of plant secondary metabolism, have a wide application in folk medicine and preservation, as well as in fragrance industries. The antimicrobial properties of essential oils have been known for many centuries. The chemical composition and the *in vitro* antifungal and antioxidant activity of the essential oil and the methanolic aerial parts extracts of *Chenopodium album* L., a medicinal plant growing in Egypt, have been studied. More than 20 constituents were identified in the oil by GC/MS. Results of the antifungal activity test indicated that the methanolic extract inhibited the *in vitro* growth of different *Aspergillus* species, *in vitro* antioxidant properties of the essential oil and the methanolic extract were determined by DPPH (2,2-diphenyl-1-picrylhydrazyl), hydrogen peroxide (H₂O₂) scavenging activity and ferric reducing power assays and compared to those of the synthetic antioxidant BHT and BHA. Due to their antifungal and antioxidant properties, the essential oil and the methanolic extract of *Chenopodium album* may be used as natural preservative ingredients in food and/or pharmaceutical industries.

Key Words

Essential oil, antifungal, *Chenopodium album*.

[P-072]

Assessment of some Sudanese essential oils against the tick *Hyalomma anatolicum anatolicum* larvae

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Abstract

Six essential oils from Sudanese plants (*Boswellia papyrifera* (Del. ex Caill.) Hochst., *Citrus maxima* (Burm. f.), *Cuminum cyminum* L., *Cyperus rotundus* L., *Cymbopogon nervatus* (Hochst.) Chiov., and *Cymbopogon schoenanthus* (L.) Spreng.) were assessed against the tick *Hyalomma anatolicum anatolicum* larvae. The most potent one being *Cuminum cyminum* with 100% mortality at 3.125% concentration, followed by *Cymbopogon schoenanthus* at 12.5%. At 25% all the oils except *Cymbopogon nervatus* achieved 100 % mortality, while all others achieved 100% mortality at 50% concentration. The lethal concentration that kills 50% (LD50) of the population ranged between 1.1742% for *C. cyminum* and 4.7443% for *C. Nervatus*. That of 99% ranged between 7.7812 to

92.3325% for *C. cyminum* and *C. nervatus* respectively.

Key Words

Sudanese, essential Oils, ticks.

[P-073]

In vitro* antimicrobial essential oil of *Melaleuca genistifolia* and *Acokanthera spectabilis

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Abstract

GC/MS investigation of the essential oils isolated from the leaves of *Melaleuca genistifolia* Smith (Myrtaceae) and the flowers of *Acokanthera spectabilis* Hook F. (Apocynaceae) revealed that: *M. genistifolia* leaves contain more than 28 compounds, including anisyl acetone: 67.4%, di-isooctylphthalate: 10.4%, varatraldehyde: 5.73%, benzothiazole: 2.74%, 3, 4-dimethoxy cinnamaldehyde: 2.66%, and *cis*-isoelemicin: 1.96% as the main components. While the minor ones were represented by cinnamic acid methyl ester: 0.62%, linalool: 0.53%, *trans*-methyl isoeugenol: 0.32%, anisole: 0.21%, eugenol: 0.17%, and α -terpineol: 0.16%. Concerning the flowers of *Acokanthera spectabilis*, GC/MS analysis resulted in identification of thirty-two compounds. The major components of the oil were represented by di-isooctylphthalate: 27.64%, benzylalcohol: 18.18%, benzoic acid: 3.5%, epoxy linalol: 2.7%, methyl salicylate: 2.5%, benzothiazole: 1.92%, cinnamic acid: 1.86%, geranyl linalool: 1.78%, and linalool: 1.27%. While the minor compounds (less than 1%) were represented by myrtenyl acetate: 0.79%, neryl acetone: 0.77%, geranyl phenyl acetate: 0.6%, linalool oxide: 0.5%, and 8-hydroxy linalool: 0.48%. The results of the *in vitro* assays using a variety of essential oils extracted from *M. genistifolia* leaves and *A. spectabilis* flowers revealed a particularly antibacterial effect and affecting the viability of a broad spectrum of bacteria and yeast such as *Staphylococcus aureus*, *Aspergillus niger*, *Escherichia coli* and *Candida albicans*. In large scale, this investigation may be of interest in the pharmaceutical application of *M. genistifolia* and *A. spectabilis* oils.

Key Words

Melaleuca genistifolia, *Acokanthera spectabilis*, essential oils.

[P-074]

Efficiency of *Thuja orientalis* and *Artemisia campestris* extracts to control of potato leaf roll virus (PLRV) in potato plants

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Abstract

Potato leaf roll virus (PLRV) was identified by enzyme-linked immunosorbent assay (ELISA) using PLRV polyclonal antibodies. Extracts from virus infected potato leaves was found to react with PLRV antibodies but not with antibodies of PVY, PVX, PVA and PVM. Extracts from *Thuja orientalis* and *Artemisia campestris* at 6 g/L was found to inhibit multiplication of PLRV by 81.72% and 63.6% respectively. Lower effect on the virus (57.8% and 45.7% of inhibition) were observed by the two extracts in plants generated from infected tubers. Extract application before virus inoculation protected the plants from infection for 10 days by *Artemisia* and 8 days by *Thuja*. No harmful effect was detected on the treated plants as determined by foliage dry weight.

Key Words

Potato leaf roll virus, potato, virus control, plant extracts.

[P-075]

Antioxidant and DNA Protecting Activities of *Mentha pulegium* L. from Southeast Flora of Turkey and its Hydrodistilled Essential Oil Composition

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Abstract

This study is designed to determine the chemical composition and antioxidant activities of the hydrodistilled essential oil obtained from aerial parts of *Mentha pulegium* L. GC and GC/MS analysis of the essential oil resulted in the determination 15 different compounds, representing 99.99% of total oil. Oxygenated monoterpenes (62.0%) were the main group of volatiles and this is followed by monoterpene hydrocarbons (32.3%). Sabinene (39.0%), 1,8-Cineole (28.5%) and Piperitone (24.3%) were determined as major compounds of the oil. Also, the essential oil was screened for its possible antioxidant activity by DPPH (0.16 mmol TE/g) and ABTS free radical scavenging (0.18 mmol TE/g), reducing power (0.23 mmol AAE/g), metal chelating (0.77 mmol EDTAE/g dw, β -carotene/linoleic acid (24.4%)) and DNA nicking assays. In DNA nicking assay, essential oil (20-40 μ g/mL) exhibited strong DNA protecting activity. The essential oil can be used as a natural antioxidant.

Key Words

Mentha pulegium, essential oil, antioxidant activity, DNA protecting activity, GC/MS.

[P-076]

Antibacterial Activity of the Volatile Oil Constituents from Leaf and Stem of *Achillea tenuifolia* Lam. from Iran

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Abstract

The genus *Achillea* is represented in Iran by 19 species including seven endemics. This genus is one of the most important genera of the Compositae family. *Achillea tenuifolia* Lam. (Compositae) with small yellow flowers and several times pinnately divided leaves in worm shape has been known for many years in folk medicine. Plant material (leaf and stem) was collected in the Khalkhal–Ardabil road area, at an altitude of 1650 m in northwest Iran. Plant materials were air dried; 150 g of leaf and 200 g of stem were subjected to 3 h of hydrodistillation in a Clevenger-type apparatus, separately. The hydrodistilled essential oils were analyzed by GC and GC/MS methods. Twenty-one constituents representing (92.2%) of the leaf oil were identified of which limonene (25.2%), α -pinene (14.4%), caryophyllene oxide (6.5%), α -gurjunene (6.3%), bornyl acetate (5.5%) and δ -cadinene (4.4%) were major components. The main components of the stem oil were limonene (23.6%), α -pinene (13.4%) and spathulenol (6.4%). Antibacterial activity of the oils was evaluated by the disc diffusion method using Mueller-Hinton agar for bacteria. The both oils showed inhibitory effects on *Escherichia coli* and *Salmonella typhi*.

Key Words

Achillea tenuifolia, Compositae, essential oil composition, limonene, α -cadinol.

[P-077]

Chemical Composition and Biological Activity of the Oil of *Bilacunaria aksekiense* A. Duran sp. nov

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Abstract

Bilacunaria aksekiense A. Duran sp. nov (Apiaceae) is a new species belonging to one of the most diversified families in Turkey. Fruits and aerial parts of *B. aksekiense* were separately subjected to hydrodistillation to yield the oils (0.55% and 0.21%, respectively). Essential oils were analyzed by GC/FID and GC/MS techniques.

In the fruit oil, β -caryophyllene (41.2%), bicyclogermacrene (9.0%), germacrene B (8.2%), α -humulene (7.0%), and caryophyllene oxide (5.6%) were identified as the major constituents. The main constituents in the oil from aerial parts were caryophyllene oxide (18.1%), β -caryophyllene (9.1%), myrcene (8.7%), (Z)- β -ocimene (6.5%), β -phellandrene (5.7%), and spathulenol (5.6%). The oils were evaluated for antifungal activity against the strawberry anthracnose-causing fungal plant pathogens *Colletotrichum acutatum*, *C. fragariae* and *C. gloeosporioides* using the direct bioautography assay. The essential oils showed no antifungal activity up to a concentration of 160 μ g/spot compared to commercial antifungal standards. In addition, *B. aksekiense* oils were subsequently investigated individually for toxicity against *Aedes aegypti* first instar larvae in a high throughput bioassay. Both oils showed 100% mortality at 500 ppm, 250 ppm and 125 ppm and only the oil from aerial parts gave 40% mortality at 62.5 ppm. Both oils showed weak adulticidal activity against *Ae. aegypti*. It appears that the *B. aksekiense* oils may play a role in plant defense against insects.

Key Words

Bilacunaria aksekiense, essential oil, GC/MS, antifungal, adulticidal activity.

[P-078]

Fumigant Activity of Essential Oil of *Thuja orientalis* L. against *Lasioderma serricorne* F.

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Abstract

The use of synthetic insecticides in agriculture as a pest control method, in order to increase yields and protect stored products, may involve serious health hazards for mammals. Essential oils were introduced as low toxic agents on mammals and non-targeted insects in insect pests control programs. In this research fumigant toxicity of essential oil from *Thuja orientalis* L. (Cupressaceae) was studied against 7-14 day-old adults of *Lasioderma serricorne* F. (Coleoptera: anobiidae). Bioassays were carried out at $27 \pm 1^\circ\text{C}$ and $60 \pm 5\%$ R.H. under dark conditions adopting a complete randomized block design. Five concentrations of essential oil were tested in four replications with 20 adult insects in each replication. The essential oil was obtained from the fresh leaves and branchlets by water steam distillation using a Clevenger apparatus. The chemical composition was analyzed by GC/MS. α -Pinene (35.2%), α -cedrol (14.6%), and Δ -3-carene (6.3%) were identified as main constituents. Mortality of adults was recorded at different concentrations ranged from 8 to 22.85 $\mu\text{L/L}$ air after 48 h. The LC_{50} value was 43.86 $\mu\text{L/L}$ air. A direct relationship between mortality rate and dose was detected. The same trend was observed between mortality percent and exposure time as well. The findings of current study suggest that essential oil could also be used as insecticide against the cigarette beetle, *L. serricorne*.

Key Words

Essential oil, *Thuja orientalis*, *Lasioderma serricorne*, fumigation, mortality, LC_{50} .

[P-079]

Antimicrobial, antioxidant and antimutagenic activity of clove oil

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Abstract

In view of the multiple traditional, industrial and medicinal uses of plant essential oils, it is imperative to build up better understanding regarding their mechanism of action and novel application in disease management. The oil of *Syzygium aromaticum* L. (clove, family, Myrtaceae) has been shown to have various biological activities. Therefore, the clove oil and the major active compound eugenol were tested for their antimicrobial, antioxidant and antimutagenic activities against drug resistant clinical strains of *Candida albicans*, MRSA and ESBL producing bacterial isolates. Oil and eugenol showed varied activity against the above group of problematic pathogens. Virulence factors of *S. aureus* (haemolysin, coagulase) were inhibited. Similarly, the virulence of *C. Albicans*, such as germ tube formation and proteinase activity, were also inhibited. On the other hand antioxidant activity was also revealed by DPPH assay. The oil showed no sign of mutagenicity but showed positive antimutagenic potential against direct acting mutagens in *Salmonella typhimurium* using Ames test. On the basis of the present study it is concluded that clove oil interact with different targets of the cell and exert multiple bioactivities. Therefore, multiple targets of these oils have made them effective agent for multiple uses. Further, *in vivo* evaluation of clove oil is needed to explore therapeutic potential against infectious diseases.

Key Words

Clove oil, antimicrobial, antioxidant, antimutagenic activity, drug resistance, microbial virulence.

[P-080]

Antioxidant activity and chemical composition of essentials oils from *Aloysia herrerae* and *Minthostachys mollis* from Perú

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Abstract

The leaves of two Peruvian medicinal plants, *Aloysia herrerae* Moldenke ("Cedroncillo", Fam. Verbenaceae) and *Minthostachys mollis* (Kunth) Griseb. ("Muña", Fam. Lamiaceae), were collected in Cusco and Lima, respectively. Their essential oils were obtained by hydrodistillation and their chemical composition investigated by gas chromatography/mass spectrometry (GC/MS, Agilent 7890A/5975, with capillary column DB-5MS (60 m x 250 µm x 0.25 µm), split 1:50, injector T=250°C, detector T=280°C, carrier gas He (1.0 mL/min), oven temperature ramped from 45°C to 275°C, 95 min. total run time). The main constituents of the essential oils were: sabinene (37.1%), limonene (25.8%), citronellal (7.4%), and cineole (7.3%) for *Aloysia herrerae*; and menthone (37.1%), isomenthone (25.8%), piperitone (7.4%), and pulegone (7.3%) for *Minthostachys mollis*. The essential oil of *A. herrerae* exhibited stronger antioxidant in vitro in the DPPH test ($EC_{50} = 7.4$ mg/mL) than *M. mollis* ($EC_{50} = 29.9$ mg/mL). Infusion of the leaves of *A. herrerae* and *M. mollis* are traditionally used in Perú because of their seda-

tive and antispasmodic activities. Also, the essential oils of several species of the genus *Aloysia* have application in the pharmaceutical and cosmetic industries. Because of the very pleasant smell of *A. herrerae* essential oil, we are developing a completely natural air freshener by mixing it with the white moss *Sphagnum maguellanicum*, with the purpose of adding value to this natural product, which grows in the highlands of Perú and is being commercialized in a sustainable way by the local communities.

Key Words

Aloysia herrerae, *Minthostachys mollis*, antioxidant, essential oil, Perú.

[P-081]

Antioxidant activity of essential oil of *Lallemantia iberica* in flowering stage and post-flowering stage

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Abstract

In this study, the essential oils from the arial parts of *Lallemantia iberica* Fisch. et C.A. Mey. (Lamiaceae), harvested in two stages (flowering and post-flowering) from plants cultivated at the Institute of Medicinal Plants (ACECR), in Hashtgerd, Iran, were obtained by hydrodistillation using a Clevenger type apparatus (yield 0.3% w/w). The chemical components of the essential oils were examined by GC and GC/MS. Thirty-six components were characterized in samples collected at the flowering stage, comprising 97.4% of the oil with β -cubebene (19.6%), linalool (18.7%), spathulenol (18.0%), β -caryophyllene (11.1%), geraniol (3.5%), and bicyclogermacrene (3.5%) as the major constituents. The oil comprised monoterpenes (33.9%) and sesquiterpenes (63.5%). Thirty-nine components of the oil of post-flowering stage comprising 96% of the oil consisted of caryophyllene oxide (38.8%), linalool (15.2%), germacrene-D (7.0%), β -caryophyllene (5.6%), β -bourbonene (5.0%), geraniol (4.3%) as the main constituents. The oil comprised 26.5% monoterpenes and 69.2% sesquiterpenes. The studied oils showed antioxidant activities in the following *in vitro* assays: DPPH radical scavenging, Ferric Reducing Power Assay (FRAP).

Key Words

Lallemantia iberica, Lamiaceae, essential oil composition, GC, GC/MS, antioxidant activity, DPPH, FRAP.

[P-082]

Antibacterial Activity and Chemical Composition of Essential oil from *Achillea wilhelmsii* in Northern Iran

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Abstract

The extraction of essential oils of leaves of *Achillea wilhelmsii* K. Koch is obtained by hydrodistillation and analyzed by gas chromatography coupled with mass spectrometry (GC/MS) and gas chromatography with flame ionization detection (GC-FID) for determining their chemical composition and identification of their chemotypes. The antibacterial screening was carried out by disc diffusion method. Their antibacterial activity was studied *in vitro* on three bacterial strains: *Proteus mirabilis*, *Enterobacter cloacae* and *Staphylococcus aureus*. The essential oil yield of the studies was 1.86%. The major component was α -pinene (3.23%); other predominant components were camphene (7.75%), 1,8-cineole (9.29%), camphor (51.86%), borneol (1.62%), spathulenol (1.47%), caryophyllene oxide (3.18%), caryophylla-4 (1.95%) and 2-naphthalenemethanol (2.48%). The result of this study showed that the essential oil of *A. wilhelmsii* exhibited varied range of antimicrobial activity against the tested organism including Gram-positive and Gram-negative bacteria, which is comparable to standard antibiotic effect.

Key Words

Achillea wilhelmsii, antibacterial activity, essential oil.

[P-083]

In vitro demonstration of *Juniperus virginiana* essential oil effects on dermo-epidermal junction

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Abstract

The Virginia cedar, *Juniperus virginiana* L., is a conical tree which grows in the southern United States. Volatile molecules are mostly present in the wood and the corresponding essential oil is obtained by its distillation. Beside its aromatic nature, nothing is really demonstrated in cosmetic applications. Thus, we thought it could be interesting to study and to evaluate its actions on normal human skin, especially on the dermo-epidermal junction.

The dermo-epidermal junction (DEJ) is a specialized acellular cutaneous zone of the upper dermis matrix that separates it from the epidermis. This basement membrane exhibits more than simple structural and filtering functions; it provides anchorage for adjacent basal keratinocytes and transmits informations to neighboring. The spatial and functional integrity of the epidermis such as terminal differentiation, barrier function or cellular communication depends directly on the qualities of DEJ. However, with aging, this structure is less and less organized and effective. DEJ is mainly composed of glycoproteins, laminins (particularly laminin 5), collagen IV, collagen VII and proteoglycans. We focused on nidogen and perlecan that organize them spatially. Using skin explants, we demonstrated that the Virginia cedar essential oil could enhance expression and deposition of the two proteoglycans. Immunolabellings, also quantified, proved the real efficacy of the oil.

According to these results, *J. virginiana* essential oil was demonstrated efficient on skin cells compartments. It could induce structural modifications implicated in skin anti-aging cosmetic strategies.

Key Words

Juniperus oil, dermo-epidermal junction.

[P-084]

***In vitro* demonstration of *Juniperus virginiana* essential oil effects on epidermis**

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Abstract

The Virginia cedar, *Juniperus virginiana* L., is a conical tree which grows in the southern United States. Volatile molecules are mostly present in the wood and the corresponding essential oil is obtained by its distillation. Beside its aromatic nature, the cedar essential oil was highly prized in aromatherapy to enhance blood and lymphatic flows. Nothing is really demonstrated in cosmetic applications. Thus, we thought it could be interesting to study and to evaluate its actions on normal human skin. Because skin is an external organ, it is particularly exposed to several stresses. We especially focused on dehydration because water is essential for cell metabolism. During epidermal differentiation process succeeding to barrier function establishment, dehydration is a current phenomenon when the horny layer is defective. Using different cellular models (keratinocytes and fibroblasts monolayer cultures and skin explants) we demonstrated two mechanisms implicating cedar essential oil in skin hydration maintenance. First, we showed that it could preserve cell viability from hydrous stress. When applied after the stress, it repaired the cellular induced damages. Secondly, treated explants showed a better cohesion between keratinocytes. The cedar essential oil enhanced expression of Claudin-1 that is constitutive of the tight junctions, responsible for water retention and cellular cohesion. According to these results, the *J. virginiana* essential oil modifies skin cells compartments and could induce cellular responses and reparation mechanisms. Considering the numerous effects of external stress and the slowing down cells adaptation due to aging, this oil could be presented as a new anti-stress active ingredient.

Key Words

Juniperus, oil, epidermis.

[P-085]

(E)-Methyl cinnamate-rich Rhizome Oil of Bornean *Alpinia latilabris* Ridl: Chemical Characterization and Antimicrobial Property

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Abstract

The chemical composition of the essential oil hydrodistilled from the dried rhizomes of Bornean *Alpinia latilabris* Ridl. (Zingiberaceae) was analyzed by GC-FID and GC/MS. The major compound identified was (E)-methyl cinnamate in remarkably high percentage (98.0%). Comparisons were made with the rhizome essential oils of *A. galanga* (L.) Willd. (9.9%), *A. malaccensis* var. *nobilis* (94.3%) and Peninsular Malaysian *A. latilabris* (0.29%), which led to the discovery that the oil constituents of the Bornean *A. latilabris* closely matched the rhizome essential oil of *A. malaccensis* var. *nobilis* based on the extraordinarily high content of (E)-methyl cinnamate in both species. This has also given rise to the possibility that the Bornean *Alpinia* species have been mislabelled by previous workers. Eucalytol, α -terpineol, d-limonene, camphene, α -pinene and β -pinene appear in all *Alpinia* species and were detected in the Bornean *Alpinia* species in this study. The rhizome essential oil of Bornean *A. latilabris* exhibited promising antimicrobial activity toward *Aspergillus niger* (1.25 μ g/mL; 20.0 $\pm$ 0.2 mm) and *Streptococcus faecalis* (0.63 μ g/mL; 15.0 $\pm$ 0.6 mm). The results illustrate the potential efficacy of plant-based natural products in controlling microbes.

Key Words

(E)-methyl cinnamate, *Alpinia latilabris*, hydrodistillation, *Aspergillus niger*, *Streptococcus faecalis*.

[P-086]

Chemosystematic importance of the essential oil compositions in *Dracocephalum* (Lamiaceae) species from Iran

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Abstract

The genus *Dracocephalum* L. belongs to the mint family (Lamiaceae) and is represented in the flora of Iran by eight annual and perennial species. Infusions of the aerial flowering parts of the plant are locally used as a general tonic, as well for treatment of cold and gastrointestinal disorders. The air-dried hydrodistilled essential oils obtained from aerial flowering parts of nine populations from seven Iranian *Dracocephalum* species was analyzed by GC and

GC/MS and identified. In order to evaluate interspecific chemical relationships, all of the characterized essential oil components were subjected to cluster and principal component analyses. Cluster analysis of the percentage composition of compounds resulted in the recognition of three distinct groups. *D. aucheri* and *D. moldavica* with sabinene and neral as the major oil components, respectively, were completely separated from the compact cluster formed by other species i.e., *D. surmandinum*, *D. subcapitatum*, *D. polychaetum*, *D. multicaule* and *D. kotschyi*, in which limonene and perilla aldehyde were found to be the principal components. The chemotaxonomic relationships among the taxa of the genus *Dracocephalum* have been discussed in detail.

Key Words

Dracocephalum, cluster analysis, PCA, essential oil, chemotaxonomy, Iran.

[P-087]

Use of *Satureja montana* essential oil in protection of crop products

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Abstract

Inhibition of *Cladosporium cucumerinum* on cucumber (*Cucumis sativus*) L. peel and natural microorganisms on poppy (*Papaver somniferum* L.) seeds by *Satureja montana* L. essential oil (EO) was investigated. Chemical composition was analyzed by SPME-GC; main components were carvacrol (66.4%) and p-cymene (22.3%). Pieces of cucumber (2 cm), inoculated on five points onto peel, and 0.1 g of naturally contaminated poppy seeds were treated with the same manner. First treatment was with filter paper infused with EO in the bottom of glass Petri dishes (250 µL/L of air) whole time of incubation. Second treatment of products was by warm air flow containing EO (2 x 15 s; 40°C, 0.3 m/s) before incubation. All Petri dishes were sealed by parafilm and incubated in 25°C for 48 h. After incubation, numbers of visible colonies on cucumbers were counted. Contamination of poppy seeds was evaluated by standard plate count method. In the first treatment, cucumber tissues were damaged. Second treatment made no damage on cucumber tissues and minimum inhibition dose, which totally inhibit growth of *C. cucumerinum*, was 12 µL. Dose of 6 µL inhibited growth of *C. cucumerinum* partially. It seems that warm air treatment is good possibility for treating fruit or vegetables. Contamination of non-treated poppy seeds was 6.4 x 10⁴ CFU/g, while after first treatment the contamination was 1 x 10² CFU/g. There was no significant difference in contamination between non treated and warm air (with EO) treated seeds.

Key Words

Cucumis sativus, *Papaver somniferum*, *Cladosporium cucumerinum*, essential oil, warm air, *Satureja montana*, antimicrobial activity, seed treatment.

[P-088]

Effect of Extraction Method and Solvent on Antioxidant Capacity of Ground Cinnamon

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Abstract

It is well known that extraction method and solvent are important factors in determining the chemical composition of botanicals. Ground cinnamon bark was extracted by soxhlet in 100% methanol and 100% ethanol, by overnight extraction in 100% methanol and 50% acetone, and by Accelerated Solvent Extraction (ASE) in 100% methanol, 50% methanol, and 75% acetone. Total phenolic content (TPC), oxygen radical absorbance capacity (ORAC), and relative DPPH· scavenging capacity (RDSC) were determined for each extraction method. TPC ranged 79.8 mg to 173.6 mg gallic acid equivalent (GAE/g), with the highest value obtained from ASE using 50% methanol. ORAC was between 660.9 and 4593.0 μmol Trolox equivalent (TE)/g among tested methods and solvents. Soxhlet extraction with 100% methanol yielded significantly higher ORAC value ($P < 0.05$) than other methods and solvents. RDSC ranged from 179.0 to 915.7 μmol TE/g, and ASE using 50% methanol or 75% acetone had the highest values compared to the other methods ($P < 0.05$). The results demonstrate that extraction solvent and method have a significant effect on available phenolic content and antioxidant capacity of cinnamon ($P < 0.05$). Overall, ASE with 50% methanol or 75% acetone was the best antioxidant extraction method, except in the ORAC assay in which soxhlet extraction yielded the highest value. The extraction methods in this study may be applied to future research on the antioxidant capacity of botanicals.

Key Words

Cinnamon, antioxidant, solvent, method.

[P-089]

Chemical Composition and Bioautography-guided Evaluation of *Thymus rasitatus* Essential Oil for its Biological activity

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Abstract

Thymus rasitatus Klokov (Lamiaceae) is an endemic species of Kazakhstan. Aerial parts of *T. rasitatus* were subjected to hydrodistillation to yield the oil (0.58%). Chemical composition of the oil was analyzed by GC-FID and GC/MS techniques. More than half of the oil was constituted by linalool (52.5%). p-Cymene (12.6%) and α -terpineol (9.9%) were found to be other major compounds of the oil. The oil was subjected to investigation for antioxidant activity by β -carotene bleaching test, Trolox equivalent and DPPH tests. Guided isolation through bioautography on TLC using 1,1-diphenyl-2-picryl-hydrazyl radical (DPPH•) as a detection reagent led to the detection of two antioxidant compounds from the oil. These compounds subsequently were identified as thymol and carvacrol by means of GC-FID and GC/MS.

Key Words

Thymus rasitatus, essential oil, GC/MS, antioxidant activity.

[P-090]

Bioactivity and Characterization of essential oils from *Geijera parviflora* (Rutaceae) (Lindley) (Wilga Bush): a native bush medicine from Australia

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Abstract

For ceremonial applications, Aboriginal Australians traditionally baked the leaves of the Wilga bush (*Geijera parviflora*), then ground them to a powder and mixed this with other plant material before smoking to induce a form of intoxication. Medicinally, leaves were also used to numb pain, particularly if suffering a toothache. Essential oils have been extracted and characterized previously, with four main chemotypes identified. Despite this, no effort has previously been made to investigate bioactivities or to relate the chemical composition to the traditional medicinal uses of the oil. In the work presented here, essential oils from many specimens were extracted by hydrodistillation of the leaves and fully characterized in our laboratory using a range of techniques, including TLC bioautography. The composition of the oils was investigated by GC/MS and compared with the compositions of previously published chemotypes. Research here aims to broaden the known traditional applications of the plant and to establish new uses for the essential oil. Antifeedant/larvicidal activity was evaluated; along with antifungal, anti-inflammatory, antimicrobial and free radical scavenging ability. Results indicate that essential oils

extracted from the Wilga bush exhibit a range of bioactivities not necessarily related to traditional use. In this regard, further research may help to reveal mechanisms responsible for the local anaesthetic and mind-altering effects described previously.

Key Words

Geijera parviflora, coumarin, Wilga, bioautography

[P-091]

Ancient Essential Oils: Exploring the Effects of Age on Bioactivity

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Abstract

It seems reasonable to expect that the molecular composition and bioactivity of essential oils will undergo changes after protracted storage times, reflecting, among other things, the effects of differential evaporation, photo-oxidation or temperature. These effects may produce significant changes despite careful handling and storage during the ensuing storage period. Long-term effects are not expected to be consistent across a range of oils, which makes it relatively difficult to predict successful preservation or changes in activity as benign or otherwise. Here we compare the composition and bioactivity of oils prepared by hydrodistillation from a range of common aromatic plants over 75 years ago and subsequently stored at room temperature with oils from the same species recently hydrodistilled in our laboratory or sourced from commercial suppliers. Tests for bioactivity focus on free radical scavenging, antioxidant, antimicrobial and antifungal activities across a range of essential oils; using DPPH, FRAP, broth dilution/disc diffusion and a core transplant method, respectively. TLC and TLC-bioautography is used to further correlate effects of likely changes in chemical composition with bioactivity of discrete components identified following characterization using GC/MS. Results may serve as a useful guide for appropriate preservation of commercial essential oils.

Key Words

Ancient oils, preservation, bioactivity.

[P-092]

Modulation of Nitric Oxide Levels in *Leishmania amazonensis* by *Piper clausenianum* Essential Oil

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Abstract

Leishmaniasis is one of the most neglected parasitic tropical diseases spread worldwide. Approximately 350 million people live in areas of active transmission of leishmaniasis, with about 2 million estimated new cases occurring every year. Efforts to improve the therapeutic arsenal against leishmaniasis have led to the search for new and cheap range of drugs, including natural product compounds. Some essential oils (EO) have shown inhibitory action against human parasites related to neglected tropical diseases such as *Leishmania amazonensis*. This work evaluated the ability of a nerolidol-rich essential oil from the leaves of *P. clausenianum* in modulating the nitric oxide levels in *L. amazonensis* infected macrophages. The results have suggested that the production of NO from infected macrophages treated for 24 h with the EO increased 17.2%, an increase greater than the one observed when infected cells were treated with INF γ , which is a well-known NO inductive cytokine. Furthermore, an increase of 20.5% in NO production could be seen when EO and INF γ were used together, reinforcing the idea of a synergistic effect. The experimental parasite-host cell interaction experimental revealed that the infected macrophages percentage was substantially reduced in 31.25%, among infected macrophages treated with the EO. Taken together, our results point to the nerolidol-rich essential oil from *P. clausenianum* with a strong potential in the development of new alternatives aiming the treatment and the management of leishmaniasis.

Key Words

Essential oil, Piperaceae, Leishmania, nitric oxide, *Piper clausenianum*.

[P-093]

Antioxidant activity of the essential oil and various extracts of *Paederia foetida* L.

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Abstract

This study was designed to examine the chemical composition and *in vitro* antioxidant activity of the essential oil and various extracts (hexane, chloroform, methanol) of *Paederia foetida* L. and GC/MS analyses of the essential oil resulted in the identification of compounds. The samples were subjected to a screening for their possible antioxidant activities by using 2,2-diphenyl-1-picrylhydrazyl (DPPH), β -carotene-linoleic acid, reducing

power, 2,21-Azino-bis-(3-ethyl benzothiazoline)-6-sulfonic acid (ABTS) and metal chelating assay. In the first case, the IC_{50} value of the *P. foetida* essential oil was determined. Among the extracts, the strongest activity was exhibited by the polar sub-fraction of the methanol extract in the β -carotene-linoleic acid system, DPPH, ABTS method. Among the extracts prepared with various solvents, a correlation was observed between the polarity and antioxidant activity. On the other hand, the major compound of the essential oil exhibited marked antioxidant activity. The amount of total phenolics and vitamin C was highest in the polar. Particularly, a positive correlation was observed between the total phenolic content and the antioxidant activity of the extracts. As estimated from the results, amounts of phenolic compounds were less in hexane and chloroform extracts than in the others. In conclusion, antioxidant potentials of polar and non-polar solvents attributed to their high phenolic contents. In both systems, the antioxidant capacities of BHT, ascorbic acid, curcumin and α -tocopherol were also determined in parallel experiments.

Key Words

Paederia, DPPH, ABTS.

[P-094]

Extraction, composition and antimicrobial activity *in vitro* on essential oil of Hops

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Abstract

Hops are major economic crops in northwest China. Essential oil Hop (*Humulus lupulus* cv. Tsingtaodahua) was extracted by steam distillation. The conditions of extraction were optimized by Response Surface Methodology (RSM). The maximum yield (0.47%) of essential oil was obtained with a liquid-solid ratio of 30:1, for 2.78 h, flow rate at 250mL/h. The composition was analyzed by gas chromatography/mass spectrometry (GC/MS). A total of 195 volatile components were identified; major components included β -pinene (31.693%) followed by α -humulene (14.615%), caryophyllene (5.680%), isocaryophyllene (4.566%), methyl *cis*-4-decenoate (4.292%), isopentyl isobutyrate (3.249%) and 9,12,15-octadecatrienoic acid, methyl ester,(zzz) (3.059%). The essential oils displayed an effective antimicrobial activity *in vitro*. Among four tested bacterial strains (*Bacillus subtilis*, *Escherichia coli*, *Pseudomonas fluorescence*, *Staphylococcus aureus*), the maximum inhibition was found for *S. aureus* and *B. subtilis*, then followed *P. fluorescence*, and the minimum was *E. coli*. The minimum inhibitory concentration (MIC) was 2.5, 2.5, 5.0, 20uL/mL against *B. subtilis*, *S. aureus*, *P. fluorescence*, and *E. coli*, respectively. Among four tested fungal strains (*Alternaria alternata*, *Fusarium semitectum*, *Penicillium expansum*, *Trichothecium roseum*), the maximum inhibition was found for *T. roseum*, then followed *A. alternata*; the minimum was *F. semitectum* and *P. expansum*. The MIC was 0.25, 2.5, 0.5, 0.5 uL/mL against *T. roseum*, *A. alternata*, *F. semitectum*, and *P. expansum*, respectively.

Key Words

“Qingdaodahua” Hops (*Humulus lupulus* L), essential oil, extraction, composition, antifungal activity.

[P-095]

Chemical composition and antibacterial effect of the essential oil *Hyssopus officinalis* L. (Lamiaceae) from Montenegro

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Abstract

The present study describes chemical composition and antibacterial effect of the essential oil of *Hyssopus officinalis* L. Plant samples were collected in northern part of Piperi (near Podgorica) in October 2009. A herbarium specimen was deposited at the Herbarium of the Department of Biology, University of Montenegro. Herb material was submitted to hydrodistillation in a Clevenger-type apparatus for 2 h according to Yugoslav Pharmacopoeia IV. The obtained oil was dried over anhydrous sodium sulphate, measured, poured in hermetically sealed dark glass containers and stored in a freezer at -4°C until analyzed by GC/MS (Shimadzu 2010+). Forty-three compounds accounting for 100% of the extracted essential oil were identified. The predominant constituents of the oil were limonene (36.45%), methyl eugenol (24.27%), 1,8-cineole (10.41%), β -pinene (4.13%), pinocamphone (3.14%) and spathulenol (1.67%). Standard disc diffusion method was used for evaluation of antibacterial effects of essential oil against 5 strains of Gram-negative (*P. aeruginosa*; *E. coli*; *P. vulgaris*; *K. pneumoniae*; *Citrobacter* sp.), and 4 strains of Gram-positive bacteria (*S. aureus*; *E. faecalis*; *B. subtilis*; *S. pyogenes*). The data obtained indicated that hyssop oil has a general antibacterial effect (inhibition zone was in range from 22 mm to 53 mm), especially against *S. pyogenes*, *B. subtilis*, *P. vulgaris* and *E. coli*. Moreover, inhibition of dehydrogenase activities (DHA) of *S. aureus* was determinate on different oil concentrations.

Key Words

Hyssopus officinalis L, essential oil, chemical composition, antimicrobial activity, dehydrogenase activity.

[P-096]

Essential oil composition and antimicrobial activity of the various plant parts of *Warburgia salutaris*

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Abstract

Although the antimicrobial activity and essential oil composition of various South African plants has been widely reported, the characterization of the essential oil of *Warburgia salutaris* (Bertol.f.) Chiov. (Canellaceae) has been neglected. This aromatic plant found in various parts of South Africa is used for the treatment of various conditions, especially upper respiratory infections. The essential oils of both the bark and leaves obtained by hydrodistillation were analyzed using GC/MS/FID and the identification of the compounds carried out by comparing

both the mass spectra from various libraries (NIST; Mass Finder, Flavour) and retention indices. The head space volatiles (SPME) of the bark were also analyzed using GC/MS/FID. The essential oil yield of the leaf (0.26%, w/w) was higher in comparison to the bark (0.036%, w/w). Quantitative and qualitative differences were obtained in essential oil composition of the bark and leaves, as well as the head space analysis of the bark. However, some of the constituents were common in the three samples investigated. More than 85% of the total oil could be characterized. Limonene (19.3%), myrcene (18.1%), linalool (8.1%), nerolidol (6.7%) and drimenol (6.0%) were the major compounds in the bark oil, while the leaves had predominately myrcene (23.8%) and E-(β)-ocimene (21.8%) as major constituents. The head space analysis of the bark also showed the predominance of myrcene (41.9%). The antimicrobial activity of the leaf oil was evaluated *in vitro* using the microdilution technique against *Mycobacterium smegmatis* and noteworthy activity was obtained with a minimum inhibitory concentration of 0.5 mg/mL.

Key Words

Warburgia salutaris, essential oils, GC/MS, head space analysis.

[P-097]

***In vitro* antimicrobial activity of main components of essential oils in vapor phase**

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Abstract

Main components of 20 essential oils (EO), which proved to have high antimicrobial activity in previous screening tests, were chosen for further testing. Their antimicrobial activity in vapor phase was tested against three bacteria and three fungi strains (*Staphylococcus aureus*, *Salmonella enteritidis*, *Pseudomonas aeruginosa*, *Alternaria alternata*, *Aspergillus niger*, *Penicillium digitatum*). Fast screening method utilizing quartered Petri dishes was applied. Minimal inhibition concentration (MIC) of main component was compared with MIC of corresponding essential oil. In general MIC of main essential oil components was higher in comparison to MIC of essential oils. The most significant increase of MICs was observed firstly for “allyl disulfide x EO of *Allium sativum* L.” and “isothiocyanate x EO of *Armoracia rusticana* G.Gaertn., B.Mey. & Scherb.”, e.g. allyl disulfide did not inhibit growth of any bacteria and MIC for fungi ranged 256–512 μ L/L; meanwhile EO of *Allium sativum* did inhibit growth either of bacteria or fungi (MIC 30–512 μ L/L). On the other hand, several components proved to be more effective against microorganisms compared to corresponding EOs, e.g. terpin-4-ol had lower MIC against bacteria (256 μ L/L) when compared with EO from *Melaleuca alternifolia* Cheel, which did not inhibit the bacteria growth.

Key Words

Essential oils, main components of essential oils, antimicrobial activity, vapor phase.

[P-098]

Chemical composition and antibacterial activity of essential oils of different populations of *Artemisia spicigera* in Northwest of Iran

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Abstract

Artemisia spicigera K.Koch is a perennial shrubby herb and is generally distributed in Armenia and also in the Middle Anatolia. This species traditionally has been used in medicines. The essential oil of *A. spicigera* was obtained by hydrodistillation from eight populations collected from different regions of East and West Azerbaijan and analyzed by gas chromatography/mass spectrometry (GC/MS). The antibacterial activity of the oils was investigated against four Gram-positive and four Gram-negative bacteria. Sixty-four compounds were identified that the main ones including 1,8-cineole, camphor, α -thujone, camphene, β -thujone and p-cymene. The studied populations have different yielding regarding essential oil components. MIC of essential oil was evaluated from 4 μ L mL⁻¹ against *Staphylococcus subtilis* to 42 μ L mL⁻¹ against *Enterobacter aerogenes*. This study demonstrates the occurrence of camphor/1,8-cineole chemotype of *A. spicigera* in western regions of Iran. The finding showed also the studied oils have good antibacterial activity, thus have great potentiality to be used as a natural health product.

Key Words

Artemisia spicigera, essential oil, antibacterial activity.

[P-099]

Chemical composition, antibacterial activity and cytotoxicity of essential oils of *Tanacetum parthenium* in different developmental stages

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Abstract

Tanacetum parthenium Schultz Bip. (Asteraceae) is an aromatic perennial plant, widely distributed in the northern hemisphere. This species traditionally has been used in insecticides, cosmetics, balsams, dyes, medicines and preservatives. The essential oil of *T. parthenium* was obtained by hydrodistillation in three developmental stages and analyzed by gas chromatography/mass spectrometry (GC/MS). The antibacterial activity of the oils was investigated against four Gram-positive and four Gram-negative bacteria. The oil was tested for cytotoxicity against THP-1 cells using the Trypan blue assay. Twenty-nine components were identified in the essential oil: the highest amount was extracted at the flowering stage. The main component, in the flowering stage, was camphor (18.94%) and other major components were bornyl acetate (18.35%), camphene (13.74%), bornyl isovalerate (3.15%), borneol (10.93%), juniper camphor (6.23%) and β -eudesmol (2.65%). MIC of essential oil was evaluated from 4 μ L mL⁻¹ against *Staphylococcus subtilis* to 38 μ L mL⁻¹ against *Enterobacter aerogenes*. Toxicity assay showed that the oil has no significant toxicity at 5-15% v/v concentrations on THP-1 cells. This study demonstrates the occurrence of camphor/bornyl acetate chemotype of *T. parthenium* in western regions of Iran. The finding showed also the studied oils have relatively good antibacterial activity without significant toxicity, thus have great potentiality to be used as natural health product.

Key Words

Antibacterial activity, essential oil, developmental stages, toxicity, *Tanacetum parthenium*.

[P-100]

Evaluation of antibacterial and antioxidant activity of *Syzygium cumini* leaf essential oil

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Abstract

The *Syzygium cumini* (L.) Skeels member of the Myrtaceae is of wider interest for its medicinal applications than for its edible fruit, commonly called jambolan. The chemical composition and the *in vitro* antibacterial and antioxidant activity of the essential oil and the methanolic leaf extracts of *S. cumini* have been studied. More than 15 constituents were identified in the oil by GC/MS. The antioxidant activity of *S. cumini* leaf extracts was investigated using the 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical-scavenging and ferric-reducing antioxidant power (FRAP) assays and compared to those of the synthetic antioxidant BHT and BHA. Results of the antimicrobial activity test indicated that the methanolic extract had high antimicrobial activity. Due to their antimicrobial and antioxidant properties, the essential oil and the methanolic extract of *S. cumini* may be used as natural preservative ingredients in food and/or pharmaceutical industries.

Key Words

Leaf essential oils, *Syzygium cumini*, Myrtaceae, antioxidant.

[P-101]

Biomedical potential of *Salvia nemorosa* L. (Lamiaceae) essential oil

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Abstract

Although different species of genus *Salvia* and their essential oils, especially *S. officinalis* L., are widely used in food flavoring, pharmaceuticals and in perfumery, there is little data related to the biological effects of *S. nemorosa* L. However, these results are concerned only with the activity of leaf extracts and chemical composition of the essential oil. Regarding to this, in the present study, the antimicrobial activity of *S. nemorosa* essential oil towards 13 bacteria and six fungi is tested. Antioxidant effects of the investigated essential oil (EO) in three model systems are also assayed. Main compounds in the EO were E-caryophyllene (22.9%) and caryophyllene oxide (18.0%). Although exhibiting mild antibacterial activity, notable are the effects on multiresistant strains of *Salmonella typhi*, *Shigella sonnei*, *Staphylococcus aureus* and *S. epidermidis*. Similar effects are exhibited on all tested fungi (*Candida albicans*, *Microsporium canis*, *Trichophyton mentagrophytes*, *T. rubrum*, *T. tonsurans*, and *Epidermophyton floccosum*). However, the investigated essential oil exhibited strong antioxidant activity, comparing to tert-butylated hydroxytoluene (BHT), used as a positive control. The IC₅₀ value in the DPPH test system was 0.98 µL/mL for EO and 5.37 µL/mL for BHT. Similarly, the 50% of inhibition of lipid peroxidation (LP) in the Fe²⁺/ascorbate

system of induction was 0.58 $\mu\text{L/mL}$ for EO and 37.04 $\mu\text{L/mL}$ for BHT, respectively. In the $\text{Fe}^{2+}/\text{H}_2\text{O}_2$ system of induction, 1.21 $\mu\text{L/mL}$ of EO was needed to inhibit the LP for 50%, comparing to the 66.67 $\mu\text{L/mL}$ of BHT.

Key Words

Salvia nemorosa, essential oil, antimicrobial activity, antioxidant effects.

[P-102]

Efficacy of plant essential oils on the control of *Botrytis cinerea* and *Penicillium expansum* on four cultivars of apples

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Abstract

Antifungal activity of essential oils was identified by recent research as an alternative strategy to reduce dependency on synthetic fungicides in post-harvest fruits and vegetables. The efficacy of essential oil treatments was assessed on apples cvs. Golden Delicious, Granny Smith, Red Chief and Royal Gala on post-harvest control of *Botrytis cinerea* and *Penicillium expansum*. The essential oils from basil (*Ocimum basilicum* L.), fennel (*Foeniculum vulgare* Mill.), lavender (*Lavandula officinalis* Chaix), marjoram (*Origanum majorana* L.), oregano (*Origanum vulgare* L.), peppermint (*Mentha piperita* L.), rosemary (*Rosmarinus officinalis* L.), sage (*Salvia officinalis* L.), savory (*Satureja montana* L.), thyme (*Thymus vulgaris* L.) and wild mint (*Mentha arvensis* L.) were tested at different concentrations. Apples were artificially inoculated with a spore suspension of each pathogen and essential oil emulsions were applied after 12 h incubation time. Results showed that, at the same concentration, treatments containing savory and thyme essential oils were statistically more efficient in controlling both pathogens than the other essential oil treatments tested. Moreover they were more efficient on apples cvs. Granny Smith and Red Chief than on cvs. Golden Delicious and Royal Gala. It was observed that a high efficacy of pathogen control could cause a high damage to the carposphere of apples if essential oil concentration is high. The most susceptible fruits to this side effect were those of cv. Golden Delicious, followed by those of cv. Granny Smith. It is possible to conclude that the efficacy of essential oil treatments and its possible phytotoxicity on apples were at least partially cultivar-dependant.

Key Words

Antifungal activity, apple, *Botrytis cinerea*, essential oil, *Penicillium expansum*, post-harvest rot.

[P-103]

Efficacy of plant essential oils on the control of post-harvest rots of stone fruits

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Abstract

Recent research recognized the possible use of essential oils against post-harvest pathogens of fruits and vegetables. The efficacy of essential oil treatments was assessed on apricots cvs. Kyoto and Tonda di Costigliole, nectarines cvs. Big Top and Nectaross and on plums cvs. Italia and TC Sun against *Botrytis cinerea*, *Monilinia fructigena* and *Monilinia laxa*. The essential oils from basil (*Ocimum basilicum* L.), fennel (*Foeniculum vulgare* L.), lavender (*Lavandula officinalis* Chaix), marjoram (*Origanum majorana* L.), oregano (*Origanum vulgare* L.), peppermint (*Mentha piperita* L.), rosemary (*Rosmarinus officinalis* L.), sage (*Salvia officinalis* L.), savory (*Satureja montana* L.), thyme (*Thymus vulgaris* L.) and wild mint (*Mentha arvensis* L.) were tested at different concentrations. Fruits were artificially inoculated with a spore suspension of each pathogen and essential oil emulsions were applied after a 12 h incubation time. Results showed that the treatments containing savory essential oil were statistically more efficient on the control of the tested pathogens than the other treatments performed. Moreover they were more efficient on plums than on apricots and on nectarines; treatments containing essential oils from basil and oregano obtained similar results but only on apricots cv. Tonda di Costigliole. Essential oils treatments at elevated concentrations presented a high efficacy of pathogen control after 15 days of storage, but could induce severe damage to the carposphere of apricots and subsequent contaminations after 30 days of storage. It is possible to conclude that the efficacy of essential oil treatments and its possible phytotoxicity on stone fruits depends both on species and on carposphere texture.

Key Words

Antifungal activity, *Botrytis cinerea*, essential oil, *Monilinia fructigena*, *Monilinia laxa*, post-harvest rot, stone fruit.

[P-104]

Chemical composition, antioxidant and antimicrobial activities of volatiles from *Plectranthus barbatus*, *P. neochilus* and *P. ornatus* (Lamiaceae)

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Abstract

The wide range of ethnobotanical uses of *Plectranthus* species (Lamiaceae) led to a growing interest in this genus. In order to contribute to a better knowledge of *Plectranthus* chemistry, volatiles of three species, included in the sub-genus *Calceolantus*, were investigated and their antioxidant and antimicrobial activities evaluated. Volatiles were obtained by distillation-extraction and analyzed by GC and GC/MS. The volatiles from *P. barbatus* Andrews, *P. neochilus* Schltr. and *P. ornatus* Codd exhibited a complex chemical composition. Monoterpene hydrocarbons were dominant (12–77%), with α -thujene, α -pinene, sabinene, 1-octen-3-ol and β -pinene being the major constituents. Sesquiterpene hydrocarbons (5–45%) were the second most representative group of components in *P. barbatus* and in *P. neochilus*, but in *P. ornatus* no relevant difference was noted between this group and the oxygen-containing compounds. The volatiles of the three species were tested for preventing lipid peroxidation and scavenging free radicals using TBARS and DPPH methods, respectively. Volatiles from *P. ornatus* and *P. neochilus* had a medium anti-lipid peroxidation activity, but volatiles from *P. barbatus* showed a very poor activity. All volatiles showed low radical scavenging activity. The antimicrobial activity of the volatiles from these three *Plectranthus* species was evaluated by the disc diffusion method against Gram-positive and Gram-negative bacteria and against a yeast strain. All the volatiles tested revealed a low to medium antimicrobial activity. The volatiles composition may explain the low antioxidant and antimicrobial activities obtained, once monoterpenes hydrocarbons were the major constituents and this type of compounds generally do not show significant biological activities.

Key Words

Plectranthus barbatus, *Plectranthus neochilus*, *Plectranthus ornatus*, Lamiaceae, volatiles, biological activity.

[P-105]

Effects of harvesting time and duration of extraction on essential oil composition of *Origanum vulgare* L.

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Abstract

In this research, samples of *Origanum vulgare* L. were collected in areas of the Kojour region of northern Iran, during the flowering stage, in June 2008. The sampling was conducted in three time intervals: morning, noon and evening. All samples were air-dried in the shade. Distillation was carried out by hydro-distillation using a modified Clevenger apparatus twice (2 and 4 h). Oils were analyzed by GC and GC/MS. Based on dry weight of the herb, oil percentage of morning, noon and evening samples were 1.0%, 1.1% and 1.0% after 2 h and 0.9%, 0.9% and 0.8% after 4 h of distillation, respectively. It shows that morning samples yield the most essential oil contents. Among 35 identified compounds of the essential oils, thymol, γ -terpinene, 1,8-cineole, p-cymene and carvacrol were the major compounds in the first two hours of extraction; while after four hours of distillation, *trans*-caryophyllene, germacrene D, bicyclogermacrene and decane were the most important essential oils compounds.

Key Words

Origanum vulgare, essential oil, harvesting time, duration of extraction.

[P-106]

The effects of organic and chemical fertilizers on yield and essential oil percentage of vegetative parts of *Ocimum basilicum* L.

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Abstract

The cropping of medicinal plants could positively contribute to the income of organic farms, as the guidelines for good agricultural practice for medicinal and spice plants demands products which are not contaminated by chemicals. Basil (*Ocimum basilicum* L.) is a medicinal and vegetable crop which is widely cultivated through the world. To evaluate the basil response to organic and chemical fertilizers, an experiment was conducted at the Research Farm of Faculty of Agriculture, Ferdowsi University of Mashhad, Iran, in 2009. A complete randomized block design with six treatments and three replications was used. The treatments were: 1- control (no fertilizer), 2- cow manure, 3- sheep manure, 4- chicken manure, 5- vermicompost and 6- NPK fertilizers. The results showed that organic manures compared to NPK fertilizers and control treatment significantly increased many of traits, e.g. plant height, leaf yield, fresh and dry matter under vermicompost were higher than other treatments. The highest essential oil yield was obtained under cow manure treatment. The third and the first cuttings had the maximum and the minimum of leaf yield, fresh and dry shoot yield, respectively. Essential oil percentage in the first cut was significantly more than other cuts, but essential oil yield was the highest in the third cut because this cut produced

the highest leaf yield. There was no significant difference between NPK fertilizers and control treatment amongst many studied traits. At a glance, the organic fertilizers could be an appropriate alternative for chemical fertilizers to achieve ecological production of basil.

Key Words

Cow manure, sheep manure, vermicompost, ecological input, *Ocimum basilicum*.

[P-107]

Effect of soil nutrients on the essential oil composition of *Salvia sclarea* L

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Abstract

The soil factor affecting the quality of essential oil from *Salvia sclarea* L. was analyzed. A field experiment was conducted with two pot trials. Complete analysis of the soils was made, including organic matter (Walkley-Black method) and total Nitrogen (Kjeldahl method). Climatic conditions as temperature and rainfall had been recorded. Plants growing on soil with high organic content (22.7%) and high Nitrogen (1.0%) were compared with those plants growing in poor organic (1.5%) and Nitrogen (0.08%) soil (percentages expressed in g/g). Growth of all samples was around the same, as was their phenological development. Aerial parts including flowers, leaves and stems were collected and subjected to hydrodistillation in a Clevenger-type apparatus. GC and GC/MS analysis determined qualitative and quantitative differences between the plants. A total of 58 compounds have been identified. Whereas 56 compounds belong to essential oils analyzed from plants growing on poor soil, only two compounds were exclusive from organic soil plants. The main constituents of the aerial parts were identified as linalool (23.5-26.2%), linalyl acetate (20.6-25.2%) and α -terpineol (6.0-7.6%) in concordance with consulted bibliography. The organic and Nitrogen content of the soil seemed to exert an influence on the chemical composition of the oils of this species, especially in the number of compounds identified which is larger in relatively poor soils. A more exhaustive study needs to be carried out in order to confirm if the biosynthesis of these compounds could be influenced by the availability of organic matter and nitrogen.

Key Words

Salvia sclarea, essential oil, soil nutrients, nitrogen, organic content.

[P-108]

Effects of salt stress and silicon nutrition on yield and essential oil of three accessions of fennel

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Abstract

Fennel (*Foeniculum vulgare* Mill.) is an important essential oil medicinal plant. Salinity is one of the most severe environmental factors influencing crop growth in Iran. Results of the beneficial effects of silicon in enhancing plants' tolerance to stress in several crops have been widely described. One study was carried out using a factorial experiment based on randomized complete block design with three replicates to study the effect of salinity stress and silicate on growth parameters, essential oil and yield of fennel. The experiment consisted of three factors, including salinity (0, 40 and 80 mmol L⁻¹), silicon (0, 0.5 and 1 mmol L⁻¹) and accession (Esfahani, Hamedani and Mibodi). The results showed that salinity stress significantly affected fresh and dry plant weight, floret number per plant, seed yield and fruit essential oil. However, an increase in the salinity leads to a reduction in shoot fresh and dry weights, floret number per plant, and seed yield, but application of the silicon increases all mentioned parameters. Among accessions, Hamadani yield had more reduction than the two other accessions in saline conditions, whereas Esfahani was less affected. Furthermore, silicon in saline condition decreased the effect of salinity stress more in sensitive accession. Using the salinity stress seems to increase the essential oil percentage in fennel. Essential oil percentage was the highest in 40 mmol L⁻¹ salinity treatment, whereas the lowest was in the control condition. Application of silicon in saline condition had no significant effect on the essential oil percentage.

Key Words

Foeniculum vulgare, floret number, accession.

[P-109]

Effect of Three Organic Material (Super Absorbent Polymer, Mineral Zeolite and Manure) in Quantity Characteristics of Essential Oil at sweet Basil (*Ocimum basilicum* L. var. Keshkeny Levelu)

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Abstract

In this research, we studied the effect of three organic materials on the percentage and amount of essential oil of leaf and shoot and the relationship between essential oil and chlorophyll in sweet basil (*Ocimum basilicum* L.). This research was randomized completed blocks (RCBD) with ten treatments and four replications. Treatments include: super absorbent polymer (SAP) (in three levels 0.1%, 0.2% and 0.3% V/V), mineral zeolite (2%, 4% and 6% V/V) and manure (5%, 10% and 15% V/V) mixed with soil of pots and a control (free). The results indicated

that SAP did not have a positive effect on the percentage and amount of essential oil. However, there was a positive trend for zeolite 4% and 6%. In zeolite 6% the percentage and amount of essential oil of leaf was 57.7% and 48.6%, both respectively higher than control. The best results were obtained for manure; accordingly, manure 5% had the highest percentage and amount of essential oil of leaf (0.687 and 0.057 mL/100 gr dry leaf, respectively) and the highest percentage of essential oil for shoot was in manure 15% (0.383 mL/100 gr dry shoot) and the manure 10% had the highest amount of essential oil of shoot (0.102 mL/100 gr dry shoot). The percentage and amount of essential oil of leaf in manure 5% was 62.4% and 62.8% respectively more than control. There was no positive relationship between percentage and amount of essential oil and chlorophyll concentration. According to the results, manure is the best organic material for production of essential oil in sweet basil.

Key Words

Ocimum basilicum, essential oil, super-absorbent polymer, zeolite, manure.

[P-110]

Effect of different essential oils on growth of the blue and green mould of orange fruit: an *in vivo* assay

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Abstract

This research was carried out to investigate the antifungal effects of the essential oils of Shiraz Thyme (*Zataria multiflora* Boiss), Eucalypt (*Eucalyptus leucoxylon* L.) and Ajowan (*Trachyspermum ammi* L.) against *Penicillium digitatum* and *P. italicum* on the Orange fruit of the 'Washington Navel' type. The investigated treatments included 0, 200 and 400 microliter concentrations of three hydrodistilled essential oils of Shiraz Thyme, Eucalypt and Ajowan in the forms of spray alone, mixture and floating for 10 and 20 min. This investigation was performed by the completely random design with three replications each containing four fruits. Major components of Shiraz thyme, eucalypt and ajowan oil, respectively, are: carvacrol, thymol, p-cymene, linalool, α -Pinene; 1,8-cineol, p-cymene, α -Pinene, α - phellandrene, δ -terpinen; and thymol, δ -terpinen, p-cymene. The results during one month in incubator with 10°C indicated that the highest preventing effect from the growth of the green mould is related to the essential oils of Ajowan and Eucalypt with 200 and 400 microliter concentrations, respectively. The essence of Shiraz Thyme was not effective on the green mould and the highest preventing effect of the essential oils was related to the mixture of the three essential oils treatment. Having applied the essential oils in the floating position of the fruit, the highest prevention effect was related to Shiraz Thyme with 400 microliter concentration for 20 min. Using different concentrations of essential oils in the form of spray and joint form indicated that these essences are not effective in controlling blue mould significantly.

Key Words

Essential oil, orange, green mould, blue mould.

[P-111]

Analysis of genetic diversity and relationships among ancient sweet *Osmanthus* trees using ISSR markers

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Abstract

The essential oils from different groups of sweet osmanthus (*Osmanthus fragrans* (Thunb.) Lour.) were usually different. The study on genetic diversity and relationships among different sweet osmanthus groups would help reveal the hereditary basis of fragrance diversity, and ancient sweet osmanthus trees were great study materials for it. They may provide more objective germplasm genetic information than new cultivated materials for their low possibility of inbreeding. In this study, interspecific relationships of 45 ancient sweet osmanthus trees from 6 provinces and 2 wild species of sweet osmanthus were evaluated by using ISSR (inter simple sequence repeat) marker with *Osmanthus yunnanensis* (Franchet) P.S. Green and *Osmanthus fordii* Hemsl as contrast species. The results showed that ancient sweet osmanthus trees had abundant genetic diversity. Fourteen ISSR primers produced a total of 148 bands, of which 131 were polymorphic. The percentage of polymorphic bands was 88.51%. The effective number of alleles, Nei's genetic distance and Shannon index were 1.8851, 0.2410 and 0.3683, respectively. The cluster analysis showed that ancient sweet osmanthus trees were closely correlated with geographical locations. And trees from the same place or in similar color often clustered together. The genetic distance between Asiaticus Group and others was far.

Key Words

Essential oil, hereditary basis, sweet osmanthus, ancient tree, genetic diversity, ISSR marker.

[P-112]

Chemical analysis of oil of *Pelargonium* var Rose as part of marketing it as geranium essential oil crop

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Abstract

There is a world demand for geranium oil from the rose scented South African plant (*Pelargonium* spp. Rose geranium). There is a desperate need in South Africa for suitable small farm crops with low risk, alternative choices for commercial farmers, and for drought tolerant crops. The Natural Products Industry is estimated to grow at 15% in demand worldwide. Due to ignorance regarding chemical composition, the industry has not bloomed in South Africa the way it was expected. There are only a few commercial farmers that are successful in South Africa, while *Pelargonium* species are indigenous to South Africa. There is a growing demand for this and it is considered

an option for small farm enterprises in South Africa and other countries which are importing huge volumes of essential oils each year. It is used for toothpaste, perfume, soap, washing powder, cool drinks, sweets and ices. South Africa's share of the world exports in essential oils stands at 1.03%. South Africa has unique indigenous oils that need to be developed and marketed. The success and experience of producers who have access to chemical analysis and relating data proved successful and formed part of the marketing strategy. In order to get the products up to a world standard and sustainable quantity, the chemical database of geranium oils in South Africa has to be coordinated and all information made available to new entrants. This study compares the quality of oils with ISO standards and the price offered by different buyers.

Key Words

ISO Standards, essential oil, geranium, chemical analysis, small farmer enterprises, indigenous.

[P-113]

Evaluation the Effect of Preservative Solutions on Vase Life of Gladiolus

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Abstract

Gladiolus is a plant belonging to the Iridaceae family, which is used as a cut flower and garden plant. Considering the limitation of the production areas of this flower, identifying the effective factors is necessary to increasing the post-harvest life for possibility of dispatch to far places. In such cases, an experiment has been done in a completely random design by using five preservative solutions: sucrose 3%, tap water, three types of plant essences (*Rosmarinus officinalis* L, *Origanum vulgare* and *Teucrium polium*). The effectiveness of treatment has been investigated on the longevity life of the flower, percentage of wilting flowers, vase life (days after treatment until senescence and petal abscission), flower relative weight and florets abscission. The results of this research showed that treatment by sucrose and essences causes increased vase life followed by quality of treated flowers. The lowest vase life has been observed in normal water (7 days) treatment. Sucrose and tap water treatments caused floret abscission, but essences treatment has played an effective role in controlling it.

Key Words

Gladiolus, vase life, preservative solutions, longevity.

[P-114]

Oil yield and composition of two genotypes of *Mentha spicata* L. grown in Kenana, Sudan

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Abstract

The objective of this study is to assess the oil yield and composition of two genotypes of *Mentha spicata* L. namely Balady and Crispa genotypes grown at Kenana experimental field, White Nile State. Hydrodistillation of the leaves gave 3.1% and 2.9% essential oils for Balady and Crispa genotypes respectively. Genotype Crispa gave higher herbage yield (2.341 ton/fed) and oil yield (68.618 L/fed) compared to genotype Balady, which yielded 1.7 kg/fed herbage and 52.809 L/fed oil. The essential oil obtained by hydrodistillation of the leaves from the two genotypes was analyzed by GC and GC/MS. Nine components representing 93.5% and 93.4% of the oil of Balady and Crispa genotypes were respectively identified. Carvone (64.9–74.3%) is the major component of Balady and Crispa oils, respectively, followed by limonene (8.3–19.7%) which was higher in the oil of Balady genotype. On the other hand dihydrocarveol (0.7–1.4%); 1,8-cineole (3.5–4.7%) and β -pinene (1.7–2.9%) were slightly higher in Crispa oil. This study demonstrated that the genotype Crispa could be a suitable genotype for production of high herbage and oil yields with high carvone content under the prevailing environment of the Kenana area.

Key Words

Mentha spicata, Balady genotype, crispa genotype, carvone, herbage yield.

[P-115]

Selection of salinity resistance of native land race of cumin through tissue culture

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Abstract

A large number of all new drugs introduced in developed countries have been derived from medicinal plants. With the increase of saline cultivated areas and desirable land deficiency, increased importance is being given to identify plants resistant to salinity. The selection resistance plant to salinity in the germination stage in hydroponics media as a cost-effective technique that requires shorter time. In this study, the effects of five solutions of different NaCl concentrations, namely, 0, 5, 100, 150 and 200 mM NaCl on 8 native land race of cumin (*Cuminum cyminum* L.), were examined in factorial experiments based on completely randomized design with three replications. The assessed land race included Shiraz, Abade, Kerman, Jirooft, Birjand, Sabzevar, Torbat-e-Heydareyeh and Jajarm. Explants of each land race incubated in MS substrate containing 1 mg/L-1 NAA and 2 mg/L-1 kinetin

and different NaCl concentrations. Germination rate had effected by salinity. Germinating rate was decreased by increase of salt concentration. There is no significant difference between 150 and 200 mM NaCL. The highest callus formation was obtained in the control treatment. The results of individual land race response to various concentrations are different, so that the highest callus formation was obtained at Birjand, followed by Shiraz and Kerman. In 150 mM NaCl, however, Kerman and Torbat had the highest callus formation and in 200 mM NaCl no callus formation was found in some land races.

Key Words

Cumin, tissue culture, salinity, cultivar.

[P-116]

The effect of different levels of phosphorus fertilizer together with phosphate bio-fertilizer (Barvar 2) on yield, essential oil content and chamazulen percentage of *Matricaria recutita* L.

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Abstract

In order to study the effect of the fertilizer of triple super phosphate (in three levels: 0, 50 and 80 kg of pure phosphorus per hectare) with or without phosphorus bio-fertilizer (Barvar 2) on yield and essence production of *Matricaria recutita* L., an experiment was executed in the form of factorial on the basis of randomized complete design in three replications at Research Farm of Agricultural Faculty of Shahed University in Tehran in 2008. In this experiment, phenologic attributes (the time of appearance of the first bud and flower, number of flowers, moist weight of flowers) and physiologic attributes (amount of essential oil, chamazulen percentage) were studied. The results showed that there was a significant difference at the level of 1% between the treatments of phosphorus fertilizer with a view to yield (number of flowers and dry weight of flower) and the amount of 40 kg phosphorus together with bio-fertilizer with production of 452.93 flowers, and 7.74 g of dry flower had the highest yield as compared with the other fertilizer treatments. Also the treatment of 40 kg of phosphorus together with bio-fertilizer with production of 0.53 mL essential oil and 15.81% chamazulen (an effective ingredient) in 152.52 of dry flower (m²) had the highest yield of essential oil and chamazulen percentage, therefore, for an experiment 40 kg per hectare of phosphorus together with 80 kg of nitrogen with the seed inoculated with phosphorus biologic fertilizer of Barvar 2 is advised.

Key Words

Barvar 2, phosphorus, essential oil, chamazulen.

[P-117]

Composition of the Essential Oils of *Salvia tomentosa* Mill. Populations in the Flora of the Marmara Region

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Abstract

Salvia is the largest genus of the family Lamiaceae with ca. 900 species distributed around the world. Its center of origin is considered to be southwest and central Asia (Hedge, 1992). *Salvia* is represented in Turkey by 97 species including 4 subspecies and 8 varieties. The rate of endemism in Turkey is 52.5% with 51 species (Ipek and Gurbuz, 2010). *Salvia tomentosa* Mill. has a wide distribution in Turkey and its dried leaves are sold in local markets for consumption as herbal tea. We have distilled essential oils from *S. tomentosa* samples collected from 20 localities by water distillation and analyzed them by GC and GC/MS techniques. Oil yields in the samples varied 0.7% to 3.5% and the main components were characterized as α -pinene (1.8% to 38.9%), β -pinene (1.9% to 35.8%), 1,8-cineole (1.5% to 39.2%) and camphor (1.5% to 40.9%).

Key Words

Essential oil, Labiatae, *Salvia tomentosa* Mill., populations, variation.

[P-118]

Determination of Yield and Quality Characteristics of the Oils of *Lippia citriodora* L. Harvested at Different Times

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Atatürk Bahçe Kültürleri Merkez Araştırma Enstitüsü-Tıbbi ve Aromatik Bitkiler Bölümü-Yalova-Turkey

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Abstract

Lippia (Verbenaceae) is represented by 200 species in Central and South America, consisting of herbs, shrubs and small trees (Terblanche and Kornelius, 1996). *Lippia citriodora* L. (Syn. *L. triphylla* (L'her.) Kuntze, *Aloysia triphylla* (L'Her.) Britton) has lemon scented leaves used as tea and the plant is cultivated in the temperate regions throughout the world (Chevallier, 1996). This plant is not cultivated in Turkey. The present study was aimed at cultivating it in the ecological conditions of Yalova and to compare essential oil content and composition of the materials harvested at different times. The results have indicated that oil yields vary between 0.83% and 1%, and percentages of the main components in the oils, according to first and the second harvests were found as follows: limonene (37.0-17.8%), geranial (17.6-32.4%) and neral (12.3-18.1%).

Key Words

Essential oil, *Lippia*, quality, Verbenaceae, yield.

[P-119]

The effects of different drying methods (natural method, oven and microwave) on drying time, essential oil content and composition of Basil (*Ocimum basilicum* L.)

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Abstract

Drying is one of the important pre-processing methods of medicinal plants after harvest, in that it has a major affect on quantity and quality of their active substances. To determine the effect of different drying methods on drying time, essential oil content and composition of Basil (*Ocimum basilicum* L.), this experiment was carried out, in 2009. The experimental design was completely randomized block design with three replications; treatments were: two temperatures: 50 and 70°C, six microwave powers: 100, 180, 300, 450, 600 and 900 w and drying in both shaded and sunny areas. The drying process was continued until the mass of the sample reduced to a moisture content of about 0.10% on a dry basis or 10% on a wet basis. The results indicated that different treatments of drying had a significant effect on drying time and essential oil content. Minimum and maximum drying times (4.1 min and 48 h respectively) obtained at 900 w microwave power and drying in shaded area. The maximum essential oil content (1.3%) obtained at drying in shaded area and minimum content (0.3%) obtained at drying in 450, 600 and 900 w. Microwave powers had a negative effect on main compositions of essential oil (geranial and methyl chavicol) but drying in shaded area preserved them. According to the results, because of suitable essential oil content and composition of drying in shaded area, this method is recommended for the drying of Basil.

Key Words

Basil (*Ocimum basilicum* L.), drying, essential oil content and composition.

[P-120]

Effect of sowing date and seeding rate on essential oil content and composition of German chamomile (*Matricaria recutita* L.)

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Abstract

In order to study the effect of sowing date and seeding rate on essential oil content and composition of chamomile (*Matricaria recutita* L.), prepared from Slovakia, an experiment was conducted. The experimental design was split-plot in the basic of randomized complete blocked design (RCBD) with three replications. Main plots consisted of three sowing dates (6 Nov, 5 Mar, and 4 Apr) and sub-plots included three seeding levels (0.2, 0.4 and 0.8 g/m²). Evaluated traits were essential oil content and percent of β -farnesene, α -bisabolol oxide B, α -bisabolol, chamazulene, and α -bisabolol oxide A. The results showed that spring sowing (March and April) produced chamomile with a high level of essential oil content. On the basis of the results, the highest essential oil content (0.63% w/w) was obtained from the plots were sown on March 5 with 0.4 g/m², but the highest α -bisabolol and chamazulene content (75.99% and 17.31% respectively) were obtained from the plots sown on April 4 with 0.2 g/m² and 0.4 g/m². According to the results, because of the high percentage of essential oil and desirable content of α -bisabolol and chamazulene, the most suitable sowing date and seeding level in Mashhad condition was March 5 with 0.4 g/m² seeds, respectively.

Key Words

German chamomile, *Matricaria recutita*, sowing date, seeding rate, essential oil content and composition.

[P-121]

The effects of different drying methods (natural method, oven and microwave) on drying time, essential oil content and composition of Savory (*Satureja hortensis* L.)

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Abstract

To determine the effect of different drying methods on drying time, essential oil content and composition of Savory (*Satureja hortensis* L.), this experiment was carried out in 2008. The experimental design was completely randomized block design with three replications, and treatments were: two temperatures: 50°C and 70°C, six microwave powers: 100, 180, 300, 450, 600 and 900 w and drying in both shaded and sunny areas. The drying process was continued until the mass of the sample reduced to a moisture content of about 0.10 on a dry basis or 10% on a wet basis. The results indicated that different treatments of drying had a significant effect on drying time and essential oil content. Minimum and maximum drying times (4.5 min and 96 h, respectively) obtained at 900 w microwave power and drying in shaded area. The maximum essential oil content (3%) obtained at drying by 70°C and drying in shaded area and minimum content (0.9%) obtained at drying in sunny area. 100 and 300 w microwave powers had an average content of essential oil of 2.3%. Maximum carvacrol content (63.89%) was obtained at 300 w microwave drying. Maximum γ -terpinene content (28.18 %) was obtained at drying by 70°C but had little difference with 50°C, 100 and 300 w. According to the results, because of reduction of drying time and suitable essential oil content and composition in drying by low microwave powers, these methods counseled.

Key Words

Satureja hortensis L., drying, essential oil content and composition.

[P-122]

Effects of leaf processing and drying methods on the quality and quantity of the essential oil of *Ocimum basilicum* L. cv. Maria Bonita

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Abstract

Ocimum basilicum L. cv. Maria Bonita (Lamiaceae) is rich in essential oil with a high content of industrial interest compound. The present work aimed to study the influence of leaf processing (whole and ground leaves) in two drying methods (oven-drying at 38°C and at room temperature using moisture dryer) on the qualitative and quantitative chemical analysis of the essential oil of *O. basilicum* L. cv. Maria Bonita. A completely randomized design was used with 4 treatments and 6 replicates. The essential oil obtained by hydrodistillation of the leaves was analyzed by GC and GC/MS techniques. The results showed that the drying methods and processing of the leaf influenced the content and chemical composition of the essential oil. Statistical difference was observed in oil content between oven drying (1.07%) and moisture dryer (0.87%). Essential oil content in whole leaves dried in oven (1.07%) was higher than in grounded leaves (0.86%). The essential oil showed predominance of oxygenated monoterpenes linalool and geraniol, but the concentration from these compounds did not change significantly within the conditions available. The oven-dried conserved the aroma and the green color of the leaves, preserving the original cultivar characteristics. For higher yield of the essential oil of *O. basilicum* L. cv. Maria Bonita oven-drying and hydrodistillation from whole leaves is recommended.

Key Words

Basil, essential oil, medicinal plants, chemical composition.

[P-123]

Effect of manure source on the quality and quantity of the essential oil of *Plectranthus neochilus* Schlechter

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Abstract

Organic fertilizer applications in medicinal aromatic plants normally modify positively the vegetal production and essential oil. The present work studied biomass production and essential oil content, yield and chemical composition of *Plectranthus neochilus* Schlechter in different organic manure sources. The scions after their acclimatization were cultured in 10 L pots under protected environment. The experiment was done in 4 treatments, 4 replications and each parcel had five pots (one plant per pot⁻¹). The treatments were: no organic manure (control); cattle manure; chicken manure; and organic manure. Plants were harvested after 120 days of culturing. The essential oil obtained by hydrodistillation of the leaves was analyzed by GC and GC/MS. The organic sources promoted differences between treatments related to biomass production, biomass distribution, essential oil yield

and chemical composition. The total plant biomass using chicken manures was 12 times more than the control. Cattle and organic manure were 3.7 and 1.5 times than the control, respectively. The different organic manures did not change the volatile oil content, but the yield had significant differences. The chemical composition of essential oil showed expressive change in the quantity and quality of compounds. Chicken manure provides the major biomass and essential oil productivity in *P. neochilus*.

Key Words

Essential oil, chemical composition, medicinal plant, fertilizer.

[P-124]

Investigation of Variability of Essential oil, Polyphenol Content and Antioxidant Activity of *Satureja hortensis* L. During Flowering Period in Rize Condition

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Abstract

This study was conducted to investigate the changes of essential oil content and antioxidant activity of *Satureja hortensis* L. during the flowering stage in 2010 from plants cultivated under Rize ecological conditions. The plantation was established with natural cuttings of *S. hortensis*. Three cutting stages (pre, full, post flowering) were done and dried by oven-drying at 35°C. The analysis of essential oil in the leaves and flowers of *S. hortensis* were carried out using a Clevenger apparatus for 3 h. The determination of total polyphenol compounds in the aqueous extracts of the plant were spectrophotometrically determined at 700 nm by Folin-Ciocalteu's colorimetric method using Folin-Ciocalteu reagent and expressed as catechin (mM) equivalents. Antiradical activity of the extracts of savory parts was measured by utilizing DPPH method and expressed as EC50 (mg/mL), the concentration necessary for 50 % reduction DPPH. In the research, mean of essential oil content was varied between 3.4 % and 2.6 %. The highest essential oil was obtained from full flowering stage. Total phenolic content of the extracts ranged from 341±9 to 400±12 µM. Also, antiradical activities of these extracts during the development stage (pre, full, post flowering) were recorded as 26±4 µM, 23±4 µM and 30±5 µM catechin equivalent, respectively. The extract of full flowering period for *S. hortensis* compared to their other extracts showed the highest phenolic concentration and antiradical activity.

Key Words

Antioxidant activity, essential oil, polyphenol content, Rize, *Satureja hortensis* L.

[P-125]

Investigation on drought resistance of cumin (*Cuminum cyminum* L.) at different growing stages

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Abstract

Cumin is one of the most important herbal drug crops used in traditional foods. It needs low water for growth cycle, and grows in arid and semi-arid regions of Iran. This experiment has been done to determine drought resistance at different growing stages in RCB design with three replications. Treatments were complete irrigation (I1), twice irrigation (First irrigation and irrigation at stem elongation stage=I2), twice irrigation (First irrigation and irrigation at flowering stage=I3) and twice irrigation (First irrigation and irrigation at seed formation stage=I4). Seed yield, biological yield, harvest index and modified stress susceptibility index were studied. The highest seed yield and biological yield were related to complete irrigation treatment. There was no significant difference among treatments for harvest index. The highest value for modified stress susceptibility index was related to drought stress at stem elongation stage. So, it is advisable that irrigation at stem elongation stage should be done to produce acceptable cumin at low irrigation conditions.

Key Words

Cumin, *Cuminum cyminum*, drought, yield.

[P-126]

Seed germination response to cold stratification period and temperature in Iranian coriander

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Abstract

In order to examine seed germination responses of Iranian coriander (*Coriandrum sativum* L.) to cold stratification and temperatures, a factorial experiment was performed in a completely randomized design with three replications under controlled conditions. The seeds were kept for 0, 1, 5, 10 and 15 days at cold stratification (4°C) and then each germinated at 20°C and 25°C in a germinator. At 20°C, by increasing cold days from 0 to 5, seed germination percentage was increased and then decreased, so that the highest seed germination was obtained in 5 cold days (91%) and had significant difference compared to the control (60%). Also at 25°C, the maximum seed germination percentage was attained in 5 days by 95%.

Key Words

Iranian coriander, seed germination, stratification, temperature.

[P-127]

Determination of Interrelationships between Carvacrol and other Some Important Constituents of Essential Oil in Oregano (*Origanum onites* L.) by Path Analysis

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Abstract

Carvacrol is the most predominant component in the many essential oils of family Labiatae which includes *Origanum onites* L. The aim of this study was to determine the relationships between carvacrol and some other constituent contents of essential oil in oregano by path analysis. The research was conducted at the Southeastern Anatolia Agricultural Research Institution experimental field during 2002-2004. In the results of path analysis, the highest positive and significant correlation ($r = 0.396$) was recorded between carvacrol and linalool contents, while α -pinene (-0.404), 1,8-cineole (-0.633) and borneol (-0.253) rates had significant but negative effect on carvacrol content. On the other hand, the highest positive and direct effect (67.53%) on carvacrol content was shown by linalool content, while highest negative and direct effect contribution to carvacrol was made by α -pinene content (42.21%). According to the results of this research, the linalool and α -pinene contents can be taken into consideration as selection criteria to improve a high carvacrol content chemotype in breeding programs of oregano

Key Words

Oregano, essential oil, constituents, carvacrol, path analysis.

[P-128]

Variability in the essential oil content and composition of different *Satureja bachtiarica* Bung. populations, cultivated in a common environment

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Abstract

Satureja bachtiarica Bung. is a perennial aromatic plant belonging to the Lamiaceae family. The plant is endemic to Iran and grows wild in the cold mountains of the west, southwest and center of the country at a range of elevations from 1100-3500 m. In this study, stem cuts of *S. bachtiarica* plants were collected in November and December from several individual plants of eight different locations covering its natural habitats. Stem cuts were rooted and planted in the field at March on separate lines. Aerial parts of the plants were collected at the full flowering stage from each line and essential oils of air-dried samples were isolated by hydrodistillation and their components identified by means of GC-FID and GC/MS. According to the results, there was considerable variation in the essential oil content (0.3-3% w/w) and composition of studied populations. Three different chemotypes including carvacrol (67-86%), thymol (60.6-84.1%) and thymol/carvacrol (in total 67-86%) were identified.

Key Words

Satureja bachtiarica Bung., chemotype, essential oil, carvacrol, thymol.

[P-129]

The herbaceous *Conobea scoparioides* obtained by hydroponics and its essential oil

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Abstract

Conobea scoparioides Cham. & Schltdl. (Scrophulariaceae) is an herbaceous known as “pataqueira,” occurring in the wetlands of the Brazilian Amazon. Its essential oil has excellent fragrance, shows high antioxidant capacity and has an oil content of 3.0% (dry weight plant). The main constituents are thymol, methylthymol and α -phellandrene, which represent about 90.0% of the oil. Fragrance oil is being regarded with interest by the industry. However, its economic exploitation has not yet consolidated, since its management in the dry season is very difficult. In summer the “pataqueira” disappears from the wet areas and its seeds only germinate again in the beginning of the rains. The plant was subjected to NFT hydroponics treatment, whose results were highly satisfactory, with production of 4 kg/m² of plant fresh weight at 4 months of growth. The oil yield increased to 3.2% with the hydroponics. For analyzing the chemical composition of “pataqueira” oil, samples were used from spontaneous growth and from the hydroponics treatment (four months of cultivation). Hydroponic cultivation did not alter qualitatively the oil composition. On the other hand, about the quantitative analysis the content of major constituents was greatly modified. From the spontaneous growth were obtained 12.9% of α -phellandrene, 40.0% of methylthymol and 38.5% of thymol, while from the hydroponic treatment these values were 25.0%, 33.8% and 32.1%, respectively. It is estimated that in the young plant (four months of culture) α -phellandrene has a higher percentage due to its being biosynthetically the precursor of thymol and methylthymol.

Key Words

Conobea scoparioides, Scrophulariaceae, hydroponic cultivation, essential oil, thymol, methylthymol, α -phellandrene.

[P-130]

Geographic variation and phenotypic plasticity of volatile oil compounds

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Abstract

Phenotypic variation and plasticity of volatile oil compounds (VOC) has been shown to exist across several taxonomic groups because of the influence of biotic factors. However, little is known about the specific abiotic factors that shape VOC and plant genetic diversity across ecogeographic gradients and taxonomic groups. The objective of this research is to investigate if specific environmental factors are better predictors of phenotypic plasticity of VOC. A meta-analysis approach has been utilized to analyze data on essential oil content and ecogeographic variables across the peer reviewed literature from various disciplinary domains. The aim of this analysis is

to develop a general theory of ecogeographic variation and its impact on VOC diversity over space and time. The results of this work will help guide future inquiry into intraspecific diversity of plants utilized for fragrance and to facilitate the exploration of new locations for collection and investigation of novel chemotypes.

Key Words

Phenotypic plasticity, volatile oil compounds, ecogeography, meta-analysis.

[P-131]

Effect of different drying methods on essential oil content and composition of *Lippia citriodora* L.

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Abstract

The effect of different drying air temperatures and methods (oven 30°C, 40°C and 50°C; sun and shade) on the content and chemical composition of the essential oil of *Lippia citriodora* L. flowering tops was studied. The essential oils from fresh and dried samples were isolated by steam distillation in a Clevenger apparatus and analyzed by gas chromatography/mass spectrometry (GC/MS). This investigation was performed by the completely random design with 3 replications. The analysis of the oils by GC/MS revealed that the main components geranial, neral, limonene, spathulenol and bergamotene showed significant variations with drying methods. Totally, the highest and lowest of oil yield was obtained at 30°C and sun drying, respectively (1.13% and 0.58%). The increase in oven air temperature resulted in a decrease of essential oil content and composition, so minimum oil yield was achieved at 50°C (0.6%).

Key Words

Drying methods, essential oil content and composition, *Lippia citriodora* L.

[P-132]

Effect of planting date and plant density on seed and essential oil yield and water use efficiency of fennel (*Foeniculum vulgare*)

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Abstract

In order to study the effect of planting date and plant density on essential oil production, seed yield and water use efficiency of fennel (*Foeniculum vulgare* Mill.), an experiment was conducted in Agricultural Research center of Islamic Azad University, Birjand branch, Iran in 2008. The experimental design was split-plot based on randomized complete block with three replications. Main plots consisted of three planting dates (19 March, 8 and 29 April) and sub plots included three density levels (6.7, 10 and 20 plants/m²). The results show that the effect of planting date, plant density and their interactions on seed and biomass yield per plant and seed, biomass

and essential oil yield per hectare were significant. Delay in planting date from 19 March to 29 April declined seed, biomass and essential oil yield per hectare. The highest essential oil yield was related to planting date of 19 March, in spite of essential oil percentage increasing with late planting. Also this treatment had the highest seed wue (138.74 g/m³) and essential oil wue (2.85 ml/m³). Increase of plant density from 6.7 to 20 plants/m² reduced seed and biomass yield per plant 45.8% and 42.5%, respectively but increased seed, biomass and essential oil yield per hectare and wue of seed and essential oil. Totally planting in 20 March with 20 plants/m² are recommended for fennel production in Birjand, Iran.

Key Words

Ennel, essential oil, wue, yield, planting date, plant density.

[P-133]

Effects of Harvesting time and duration of extraction on Essential oil compositions of *Artemisia annua* L.

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Abstract

In this research, samples of *Artemisia annua* L. were collected in area of the Kojour in small province of Nowshahr, during the flowering stage. The sampling was conducted in three time intervals: morning, noon and evening in June 2008. All samples were air-dried in the shade. Extraction carried out by hydrodistillation for two different times (2 and 4 h). Analysis was evaluated by GC and GC/MS. Based on dry weight of the herb, oil percentages of morning, noon and evening samples were 1.52%, 1.25% and 0.99% after 2 h and 1.00%, 0.98% and 0.75% after 4 h of distillation, respectively. It shows that morning samples have the most essential oil contents. Among 35 identified compounds of the essential oils, camphor, α -pinene and 1,8-cineole were the major components in the first two hours of distillation; while after 4 h of distillation, *trans*-caryophyllene, germacrene-A and α -pinene were the main components.

Key Words

Artemisia annua, essential oil, harvesting time, duration of extraction.

[P-134]

The study on the Fungicidal Effects of some Essential Oils on Tangerine *Penicillium digitatum* control Under *In vivo* Conditions

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Abstract

In this study, antifungal activity of some essential oils extracted from *Foeniculum vulgare* L., *Bunium persicum* B. Fedtsch., *Carum copticum* Benth. & Hook. f., *Zingiber officinale* Roscoe and *Mentha piperita* L. were investigated against *Penicillium digitatum* of mandarin (*Citrus reticulata* Blanco). This experiment was randomized completely design with 4 replications and 3 observations. Used treatments include *F. vulgare*, *B. persicum*, *C. copticum*, *Z. officinale* and *M. piperita* essential oils (0, 200, 400, 600 and 800 µL/L). Results indicated on the fruit showed that wild caraway essential oil in applied concentrations in compared to other essential oils perfectly inhibited growth of *P. digitatum* fungus on the mandarin fruit. Wild caraway essential was in low concentrations 200 and 400 µL/L positive effects on some qualitative parameters (TSS, content vitamin C, titrable acidity, content decay and weight loss). But *Z. officinale* essential oil had the lowest effect on the shelf life of mandarin fruits. Results of the present paper confirmed antifungal effect of *B. persicum* essential oil on fruit post-harvest. However, more studies are required to recommendation of *B. persicum* essential oil as a commercial and natural antifungal for increase post-harvest on horticultural crops.

Key Words

Essential oils, tangerine, *Penicillium digitatum*, wild caraway.

[P-135]

Seasonal variation on the chemical composition of the essential oils of *Salvia lavandulifolia* Vahl

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Abstract

The essential oils extracted by hydrodistillation from the aerial parts of *Salvia lavandulifolia* were analyzed by GC and GC/MS. In order to study their seasonal variation, samples were collected every two months throughout a year. Moreover, distillations from different plant parts in full bloom were compared. Soil and climatic conditions where plants grew were also characterized. It was found that both yield and chemical composition of the essential oils were quite variable according to the phenological state. The highest yield was obtained just after bloom time, and the lowest in late winter. A total of thirty-seven compounds were identified in the essential oil corresponding to the full bloom season, being the 95.8% of the total essence. The main compounds were α -pinene (4.7-11.6%), 1,8-cineole (16.5-53.3%), camphor (2.3-7.9%) and viridiflorol (3.0-13.6%). Statistical analysis showed significant differences in these compounds throughout the year, and results obtained by Pearson Correlations indicated a strong negative correlation (-0.905) between 1,8-cineole and viridiflorol. Results comparing essential oils from different aerial parts showed important differences both in chemical composition and yield production. Further-

more, this work has been developed with data from a single year and further investigations should be done in the following seasons, including genetic studies to verify the influence of genetic heritage and abiotic factors in the *S. lavandulifolia* essential oil composition

Key Words

Salvia lavandulifolia, essential oil, seasonal variation, edaphoclimatic conditions, α -pinene, 1,8-cineole, camphor, viridiflorol, Pearson Correlations.

[P-136]

Effects of salinity on essential oils of two species of *Lavandula* by soilless culture

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Abstract

Salinity is a factor affecting the development and concentration of *Lavandula* essential oils. *Lavandula* are typical Mediterranean plants; soil and climate factors, growth model and type and concentration of essential oils vary depending on the species. The experiment was carried out with *Lavandula dentata* L. and *L. angustifolia* Goodwin creek grey under substrate (peat: fiber coir; 2:1 vol:vol) crop. Two treatments were evaluated using standard nutrient solution to 1.6 and 2.7 dS m⁻¹ of EC (Treatment T0 and T1, respectively) and two more with 2.7 and 3.6 dS m⁻¹ with the addition of Cl (T2 and T3, respectively). The results suggest a reduction in growth and an increase in the concentration of essential oils in relation to salinity. This behavior was different in the two species of the same genus

Key Words

Salinity, soilless crop, cultivar, pot.

[P-137]

Difference of growth traits, essential oil content and composition between diploid and induced tetraploid plants of basil medicinal plant (*Ocimum basilicum* L.)

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Abstract

The effect of colchicine induced tetraploidy on some growth traits and essential oil content and composition of *Ocimum basilicum* L. ($2n = 48$) was studied. A significant increase in the essential oil content was found in C1 generation and the tetraploids have the potential of producing 69% more oil than the diploid parents. The chemical composition of diploid and tetraploid basil, have been examined by GC and GC/MS. A field experiment was conducted to investigate the effect of ploidy levels on some of the growth traits such as: leaf area, fresh weight, dry weight, leaf long and wide, leaf thickness, period of flowering. This study demonstrated that in tetraploid basil revealed significant increases in dry and fresh weight, leaf wide and thickness, essential oil content of areal parts. In the other hand increase in ploidy level causes significant decrease in leaf long, seed germination percent and rate, as compared with diploid parent. It could be concluded that genomic multiplication can confer enhanced production and/or qualitative improvement in the biochemical profile of secondary metabolites and tetraploidy seems a promising approach for essential oil increasing in basil.

Key Words

Basil, *Ocimum basilicum*, tetraploidy, growth traits, essential oil content and composition.

Seasonal variations in oil yield of *Eucalyptus* species and hybrids in India

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Abstract

The natural hybrids between *E. citriodora* Hook. and *E. torelliana* F. Muell. were identified thorough morphological genetic markers viz. absence and presence of lignotuber, aroma of crushed leaves, colour of lower epidermis of cotyledon and leaf size. The hybrids and the parents were planted during 1987 at the Forest Research Institute, Dehradun, India. The open-pollinated progenies of inter-specific reciprocals were further planted in different agro climatic conditions to understand genotype and environmental interactions. Pertinently, though, a majority of eucalypts have seriously been eclipsed with gall insect caused by *Leptocybe invasa*, these hybrids have so far not been infested with the insect. Eucalypt oil plays a signification role in perfumery and pharmacy industry. *E. torelliana* has broad leaves with a dense crown but hardly produces oil. On the other hand, *E. citriodora* has narrowed and long leaves and known for oil production. The hybrids being new gene recombinants of two different species produced oil with pungent smell that varied in color. The seasonal variation for oil yield was analyzed by collecting fresh leaves of *E. citriodora*, *E. torelliana* and F1 inter-specific hybrids of *E. citriodora* x *E. torelliana* and *E. torelliana* x *E. citriodora* during rainy, summer and winter seasons. Oil from the leaves was extracted through steam distillation. The maximum oil was extracted during rainy seasons. The color and odor for parents and hybrids had variations, which is being tested independently. However, efficacy per se for medicinal purposes needs to be tested.

Key Words

E. citriodora, *E. torelliana*, morphological genetic markers, interspecific hybrids, reciprocal natural hybrids.

[P-139]

Comparison among different parsley accessions

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Abstract

Parsley (*Petroselinum crispum* [Mill.] Nyman.), family Apiaceae, is a very important aromatic plant the utilization of which goes back more than 2,500 years. In the present research five accessions were compared in an experimental field trial carried out in 2010-2011 adopting a scheme of randomized blocks, with four repetitions. Four of these accessions were from commercial origin, while the fifth was a wild accession utilized in a small mountain village in the province of Trento, named Brentonico, located at 700 m a.s.l.. According to local folks, the origin of this parsley, popular as "Fresh parsley," goes back to the time of the Napoleonic Wars and the French invasion of Trento (1803), and recently a local breeder has been trying to broaden its utilization. In the trial the four commercial cultivars and the wild accession were characterized for the morphological and productive traits and for the yield and quality of the essential oil obtained by hydrodistillation and analyzed with HPLC. In the present work, the productive and qualitative results obtained by the trial will be presented.

Key Words

Parsley, essential oil, wild accession.

[P-140]

Impact of environmental effects on the quality of genipi liquors from Southern French Alps

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Abstract

Root colonization by arbuscular mycorrhizal fungi of *Artemisia umbelliformis* L. was conducted in greenhouse conditions for 6 months of inoculation. Essential oils components were analyzed by GC/MS, showing that the alpine microbial inoculum increased significantly the percentage of E- β -ocimene and reduced those of E-2-decenal and (E,E)-2-4-decadienal. These results seem to indicate an influence of alpine microbial inoculum on essential oil production. A second study investigated 6 different clones cultivated in 5 natural sites in the Southern Alps of France. After 6 months, takings were macerated in pure ethanol and analyzed by GC and GC-O, using different extraction techniques (HeadSpace, SPME, ITEX, INDEX). Sites and inoculation seem to have an important influence on the aroma quality of the liquor because of the differences showed by the incoming of new results. The forthcoming results should give important indications for the cultivation of quality genipi in natural conditions.

Key Words

Artemisia umbelliformis, genipi, GC/MS, extraction, aroma, liquor.

[P-141]

Compositions of the essential oils of *Teucrium cavernarum* and *Teucrium paederotoides*, two endemic species from Turkey

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Abstract

The essential oils from aerial parts of *Teucrium cavernarum* P. H. Davis and *Teucrium paederotoides* Boiss & Hausskn. (Lamiaceae), both of which are endemic species in Turkey, were isolated by steam distillation and analyzed by gas chromatography (GC) and gas chromatography/mass spectrometry (GC/MS). Plants were collected during the flowering period from Karaman (*T. cavernarum*) and Gaziantep (*T. paederotoides*) provinces of Turkey. Thirty compounds were identified in the oil of *T. cavernarum* and seventy-three compounds were identified in the oil of *T. paederotoides*. The major components were characterized as β -caryophyllene (32.9%), germacrene D (20.7%), caryophyllene oxide (14.1%) and bicyclogermacrene (6.3%) for *T. cavernarum* and germacrene-D (20.8%), pulegone (9.5%), bicyclogermacrene (9.2%), hexadecanoic acid (7.9%) and spathulenol (6.5%) for *T. paederotoides*, respectively.

Key Words

Essential oil, endemic, GC, GC/MS, *Teucrium cavernarum*, *Teucrium paederotoides*, Turkey.

[P-142]

Seasonality effects on *Cordia verbenacea* D.C. transcriptome and essential oils target metabolome

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Abstract

Cordia verbenacea DC. (Boraginaceae), a Brazilian native species, produces active principles in the essential oils from leaves, such as E-caryophyllene and α -humulene that exhibit anti-inflammatory activity. The current study aimed to investigate the effects of seasonality on the concentration of essential oil active components and on the transcriptome of four genotypes of *C. verbenacea*. Leaves from pot grown plants of the four genotypes (1, 2, 3, 4) were harvested at fixed dates in the four seasons (spring, summer, autumn and winter) in the morning (8:00 to 10:00 AM) and afternoon (5:00 to 6:00 PM). Leaf material was employed for chemical and transcriptional

analyses. Active compounds were identified by GC/MS and quantified by GC/FID. Whole-genome transcriptome was determined by cDNA-AFLP. In all four investigated genotypes for both sampling points, higher concentrations of the active compounds were found in the summer, ranging from 0.15 to 0.65 mg/mL for E-caryophyllene and from 0.013 to 0.48 mg/mL for α -humulene. The contents of both substances were lower in the spring (0.06 to 0.23 mg/mL) and winter (0.06 to 0.27 mg/mL) for E-caryophyllene and α -humulene (0.03 to 0.012 mg/mL; 0.004 to 0.019 mg/mL), regardless of the diurnal period. Thirty-four differentially expressed transcript-derived fragments (TDFs), with molecular weights ranging from 1150 to 130 bp, were identified in the sampling conditions. Fragments corresponding to ten distinctly regulated transcripts were cloned and are currently being sequenced. The association of chemical and molecular analyses will provide important tools to elucidate the biosynthesis of economically important compounds in *C. verbenaceae*.

Key Words

Cordia verbenacea, essential oils, transcriptome, metabolome.

[P-143]

A Comparison between the Yield and Essential Oil Parameters of *Thymus vulgaris* Population and a Culture Material (Varico III)

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Abstract

There are limited studies on Thyme species yield and essential oil parameters conducted in Central Anatolian climatic conditions. Although there is no natural distribution in the wild flora of Turkey, *Thymus vulgaris* L. is still the most important species of the genus Thyme. Population obtained from different locations of *T. vulgaris* and a cultivar (Varico III) were studied in terms of several yield and essential oil parameters during the years of 2009-2010 in Ankara. Field experiments were designed at complete randomized block design with four replications. Two cuttings were recorded each year from the species. The first cuttings were in early July, while the second took place in mid-October. Total yield of green herb, drug herb and drug leaf of the population forms of *T. vulgaris* were 2512-2872 kg/da, 863-1039 kg/da and 458-682 kg/da, respectively. Average essential oil ratio and total essential oil yield were also detected as 2.670-2.585% and 22.39-24.63 L/da. Varico III (cultivar) had a total of 2514-4048 kg/da green herb, 854-1272 kg/da drug herb and 412-624 kg/da drug leaf yield. The average essential oil and total essential oil yield were determined to be 3.588-2.783% and 31.64-37.75 L/da. Thymol content ranges of the two cuttings were 66.93-57.78% and 58.15-41.52% for the population and 64.87-34.20% and 74.86-44.76% for the Varico III. P-Cymene, γ -terpinene, carvacrol and β -Caryophyllene were the main components.

Key Words

Thymus vulgaris, Varico III, fresh and drug herb yield, drug leaf yield, essential oil content and components, thymol.

[P-144]

The Yield and Essential Oil Characteristics of Different *Origanum* Species

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Abstract

The genus *Origanum* is represented by 23 species and 32 taxa in Turkey while there are 41 species and 52 taxa in the world. *Origanum vulgare* L. subsp. *hirtum* (Link) Ietsw. and *Origanum onites* L. are the main species of the export products in Turkey. In this research, two *O. vulgare* subsp. *hirtum* [one population and a cultivar, Carva (*O. vulgare* subsp. *hirtum* x *O. vulgare* L. subsp. *viridulum* (Martini-Donos) Nyman)] and an *O. onites* population material's yield and essential oil characteristics were examined during the years 2009 and 2010. The research was conducted in the ecological conditions of Ankara and two cuttings were studied each year. It was also possible to get the third cutting from all the species in the same growing season. Total yield of green herb were recorded as ranges of 4372-6955 kg/da, 5274-6785 kg/da and 1887-2872 kg/da, respectively. Total drug herb and drug leaf yields were as follows; 1537-2110 kg/da, 1522-1917 kg/da, 440.9-804.2 kg/da and 870-1083 kg/da, 512-957 kg/da, and 241-510 kg/da. Average essential oil content and total essential oil yield were also determined as 4.565-4.682%, 6.088-5.432%, 2.020-2.565% and 70.36-97.63 L/da, 92.58-105.6 L/da, 8.70-21.33 L/da. Although carvacrol chemotypes are common in the natural flora of Turkey, the main component of the population *O. vulgare* subsp. *hirtum* and *O. onites* were isolated as 'thymol' (55.41-54.02%, 55.95-49.39% and 65.07-69.83%, 53.17-62.40%). The main component from Carva was detected as 'carvacrol' (74.18-43.34%, 79.03-70.39%). γ -Terpinene, p-cymene, α -terpinene and β -caryophyllene were other major components.

Key Words

Origanum vulgare subsp. *hirtum*, Carva, *Origanum onites*, fresh and drug herb yield, drug leaf yield, essential oil content and components, thymol, carvacrol.

[P-145]

Evaluation of essential oil in Fennel (*Foeniculum vulgare* Mill) accessions under water limited condition

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Abstract

To examine effect of water limited condition on essential oil production of Fennel (*Foeniculum vulgare* Mill), eight accessions originated from the central region of Iran were evaluated under field conditions using two irrigation regimes. The experiment was carried out using a split plot on the basis of randomized complete block design with three replicates. The main plot included two irrigation regimes, one and two weeks' interval, and accessions

assigned in the subplots. The results indicated that two-week compared with one-week interval irrigation regime significantly increased essential oil volume and essential oil weight but significantly decreased essential oil yield as a result of reduction of plant biomass production in this condition. The accessions were significantly ($P < 0.01$) different for essential oil content in both irrigation regimes. Essential oil volume of accession ranged from 5.44 to 19.44 and 9.33 to 27.89 (mL/100g⁻¹ seed weight (SW)) in weekly and two-week irrigation regimes respectively. The highest essential oil weight was measured under two-week irrigation regime condition (2.03 g 100g⁻¹ SW), while the weekly irrigation regime plants were characterized by the lowest accumulation level (0.39 g 100g⁻¹ SW). However essential oil yield of accession ranged from 0.11 to 0.28 and 0.13 to 0.59 (g per plant) in weekly and two-week irrigation regimes, respectively. Essential oil characteristics (volume, weight and yield) showed positive correlation with wet and dry plant biomass productions, while essential oil volume and weight showed negative correlation with plant seed yield.

Key Words

Foeniculum vulgare, essential oil content, irrigation regimes.

[P-146]

Effects of nitrogen and iron on yield and essential oil of Garden Thyme

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Abstract

Garden thyme (*Thymus vulgaris*) L. is a pleasant smelling perennial shrub, which grows in several regions in the world. It is native to the Western Mediterranean region and southern Italy. It is commonly known that the composition of the essential oil determines the specific aroma of plants and flavor of condiments. The main constituents of thyme include thymol, carvacrol and flavonoids often thought to have antifungal, antibacterial, anti-flatulent and anti-worm characteristics. Nutrients as one of the most important environmental factors affect growth and development of medicinal plants. A field experiment was conducted to evaluate the effect of nitrogen (N) and iron (Fe) fertilizers on qualitative and quantitative characteristics of garden thyme. Nitrogen (0, 75 150 kg/ha) and Fe (0 and 5 ppm FeSO₄ applied as spraying and 20 kg/ha chelated iron applied in soil) were applied in a split plot experiment design based on randomized complete block design with three replications. The results showed that nitrogen significantly affected the shoot fresh and dry weight, height, yield, essential oil percentage, leaf nitrogen and iron. An increase in the nitrogen leads to an increase in shoot fresh and dry weights, height, yield, essential oil percentage and leaf nitrogen. However, application of iron did not affect all mentioned parameters clearly. The result showed N increased essential oil percentage. Essential oil percentage was highest in 150 kg/ha nitrogen, whereas the lowest were in the control condition. Application of iron decreased the essential oil percentage.

Key Words

Thymus vulgaris, nitrogen, FeSO₄.

[P-147]

Seed germination of an important plant producing medicinal essential oil in Brazil, Copaiba (*Copaifera langsdorffii* Desf - Fabaceae)

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Abstract

The Copaiba (*Copaifera langsdorffii* Desf - Fabaceae) is a tree species occurring in Brazil, the source of the oil for Copaiba. This essential oil has extensive use in folk medicine in humans and animals. The oil has anti-inflammatory and wound-healing potential, according to scientific studies. The occurrence of Copaiba was common in tropical forests, but deforestation reduced the density of mature trees, leaving but few specimens. It's too small to get the index of mature plants from naturally germinated seeds. For this reason many regions no longer have this species. An alternative is reforestation. For this to be viable, specific techniques must be developed. This study aimed to evaluate the germination of Copaiba subjected to two treatments: T1 (without scarification or natural) and T2 (mechanical scarification) breaking through the cuticle to break dormancy. The two treatments were conducted under experimental conditions with 200 seeds in 10 random blocks by researchers at the University of Mato Grosso, Cáceres (MT), Brazil, January–April 2006. The results show that seeds subjected to treatment T2 provided a germination rate six times higher (59.50%) than those from T1 (9.50%). This highly significant difference between treatments revealed that the mechanical scarification (T2) is the best way to break the dormancy of the seeds of Copaiba. As a consequence, it will be more successful in producing seedlings of this species for purposes of reforestation, seeking to make possible in the future, the continued production of the oil of Copaiba.

Key Words

Copaiba essential oil, folk medicine, seed germination, Mato Grosso state, Brazil.

[P-148]

Effect from natural aging in the viability of seed germination of Copaiba (*Copaifera langsdorffii* Desf - Fabaceae) plant production essential oil containing medicinal properties

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Abstract

The Copaiba (*Copaifera langsdorffii* Desf-Fabaceae) is a plant from which folk medicine uses an essential oil with medicinal properties. It is important to be included in reforestation efforts for commercial exploitation and prevention of extinction. Its seeds have dormancy, hindering its germination, but may extend its physiologi-

cal pós-maturação viability. To assess the effect of natural aging in the germination of seed viability of Copaiba, seeds were collected soon after physiological ripeness (Aug-Sept 2005) under native trees located in the “Flecha” community, Cáceres (MT), Brazil. Then the seeds were stored in paper envelopes under stable conditions of temperature, light and moisture. The experimental study was deployed on random blocks with four treatments (monthly sowing, on the fifth day of: January-T1, February-T2, March-T3 and April-T4, 2006), using 200 seeds at a time. The results of germination were T1: 72%, T2: 68%, T3: 53% T4: 45%. Germination sowing highest occurred in January (T1), decrease in the following seeding and with worse outcome in April (T4). The differences between the pairs of T1 to T4 treatments were highly significant ($p = 0.006$), T2-T4, significant ($p = 0.019$). In the other pairs (T1-T2, T1-T3, T2-T3 and T3-T4) differences were not significant ($p > 0.5$), according to the Tukey test. Aging ($T_x = 1, 2, 3, 4$) seeds did decrease the germination (y), whose best fit trends toward the polynomial regression model of third degree ($R^2 = 1$; $y = 0, 003x^3 - 0,235x^2 + 0,455x + 0,47$) and then the linear model ($R^2 = 0.95$; $y = 2,096x + 0.835$)

Key Words

Seed aging effect of germination, medicinal and essential oil plant, Mato Grosso state, Brazil.

[P-149]

Development of an NaCl-tolerant line in *Tanacetum cinerariaefolium* (Trevir.) Schultz-Bip through shoot organogenesis of selected callus line

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Abstract

Plants were regenerated successfully through shoot organogenesis of a NaCl-selected callus line of *Tanacetum cinerariaefolium* (Trevir.) Schultz-Bip developed through stepwise increase in NaCl concentration in MS medium. Increasing NaCl level concentration (0, 5, 10, 15, 20, 25, 30, 35, 40, 45 mM) from low level to high level was found to be a better way to isolate NaCl-tolerant callus line, since direct transfer of callus to high saline medium was detrimental to callus survival and growth. Among different media and growth regulator treatments, MS media containing 1 mgL⁻¹ BA and 1 mg L⁻¹ NAA or 1 mgL⁻¹ BA, 2 mgL⁻¹ NAA and 0.5 mgL⁻¹ GA3 for shoot organogenesis in selected callus line and B5 medium supplemented with 2 mgL⁻¹ NAA showed best response for root regeneration. As increasing NaCl concentrations (from 0 to 45 mM) the ability of shoot and root regeneration were decreased. The selected callus line showed significance increase in proline content and decrease in pyretrine content. Based on growth performance and proline content (20 mM in callus line and 35 mM in shoot culture) could be considered as NaCl-tolerant line showing all positive adaptive features towards the salinity stress. Further studies about agronomic performance of obtained plants under saline soil conditions are necessary for understanding the genetic stability of the induced salt tolerance plants.

Key Words

Acclimatization, callus induction, *in vitro*, NaCl-tolerant callus line, salinity stress, *Tanacetum cinerariaefolium*.

[P-150]

Seasonal variability of essential oils of *Thymus mastichina* L. from PNTI (Portugal)

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Abstract

Thymus mastichina L. is an endemic plant from Iberian Peninsula and is widespread in Portugal. The composition of its essential oils can vary according to the geographic origin as reported by Salgueiro. There are three chemotypes in Portugal: 1,8-cineole; 1,8-cineole/linalool and linalool. In this work three different wild populations of *T. mastichina* were collected during the flowering (May) and vegetative (October) phase at Parque Natural do Tejo Internacional (PNTI), in Center-East of Portugal. Essential oils from the aerial parts of the plants were extracted by hydrodistillation and analyzed by gas chromatography coupled with flame ionization detector and ion trap mass spectrometry. Essential oil yields (volume per dry weight) were very good in all the samples and increased from the flowering to vegetative phase in populations from places 2 and 7, but population from place 8 decreased. The major compound was 1,8-cineole, accounting for, on average, 28.25–34.40% in flowering and 51.26–53.42% in vegetative phase, followed by borneol (11.14–13.31% in flowering and 8.53–14.15% in vegetative phase), α -terpineol (9.07–11.24% in flowering and 7.05–9.97% in vegetative phase). As expected, this is a 1,8-cineole chemotype.

Key Words

Thymus mastichina L., essential oil, Portugal, hydrodistillation.

[P-151]

Molecular profiles and essential oil compositions of 24 varieties from *Citrus medica* L.

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Abstract

The aim of this work is to investigate the genetic and chemical diversity of the *Citrus medica* L. varieties. For this purpose, the polymorphism of 27 nuclear and cytoplasmic genetic markers of 24 citron varieties (*C. medica*) were studied using five markers represented by three universal microsatellite primers amplifying chloroplast SSR (cpSSR) and three markers of *C. mitochondria*. The chemical compositions of leaf essential oils were also reported to assess their interest in the chemotaxonomy studies. Most of the citron varieties showed two chloroplastic haplotypes (A and B). These two haplotypes were distinguished only by the Ccmp5 marker. Haplotype A included five varieties and haplotype B contained eight varieties. This group of 13 genotypes is considered a cluster of true

citrons. All other varieties were characterized by different cytotypes without any common allele with haplotypes A and B at the level of the chloroplastic genome. The nuclear genome diversity confirms the closely parentage of the 13 citrons (haplotypes A+B) and the hybrid status of other varieties. Genetic analysis allowed us to identify the true citrons and revealed a possible sexual crossing between citron varieties. The essential oil compositions are qualitatively close but differed greatly from a quantitative viewpoint. Indeed, nine varieties exhibited a similar composition with the same five major compounds, whereas the fifteen other varieties exhibited a specific chromatographic profile. Finally, the results on essential oil composition are not in agreement with DNA marker diversity; nevertheless, the analysis of volatile components was informative for specific phenotype characterization.

Key Words

Citrus medica, nuclear marker, leaf essential oil, diversity, GC, GC/MS.

[P-152]

Identification and investigation of features and compatibility of different masses of basil (*Ocimum basilicum*) at Shahre Ghods, Iran

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Abstract

In order to identify and investigate features and compatibility of different masses of basil (*Ocimum basilicum* L.), this study was carried out in 2009 at University Azad of Shahre Ghods Branch in Iran. The field experiment was carried out in a randomized complete block design with three replications. Factors were masses of basil (masses of Karaj, Shahriar and Varamin) and essential oil from flowering branches produced by the hydrodistillation method. Our results showed that masses of basil significantly affect stem diameter, biological yield, flowering branch yield, essential oil percentage, essential oil yield. The highest essential oil percentage (0.54%) was achieved by mass of Karaj but maximum of stem diameter (7.62 mm), biological yield (3389 kg ha⁻¹), flowering branch yield and essential oil yield (2/12 L per hectare) were achieved by mass of Shahriar. It was thus concluded that mass of basil influenced on the quantity and quality features in basil and mass of Shahriar had a good potential for gain of further essential oil yield. Therefore, yield of essential oil is different between masses of medicinal plants, and it is better that such studies be further validated by conducting them for other masses and cultivars of medicinal plants.

Key Words

Basil (*Ocimum basilicum*), Shahriar mass, flowering branch, essential oil yield.

[P-153]

Essential oils from *Lavandula luisieri* (Rozeira) Rivas-Martinez grown at a Montado ecosystem

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Abstract

Montado is a *Quercus* spp. woodland (holm- or cork-oak woodlands), resulting from an agro-silvo-pastoral land use system. This typical southwestern European and northwestern African ecosystem is classified as a habitat of community interest for conservation by the European Union and shows high biodiversity. *Lavandula luisieri* (Rozeira) Rivas-Martinez [*L. stoechas* subsp. *luisieri* (Rozeira) Rozeira in Agron] is a small aromatic shrub and an Iberian Peninsula endemic, frequent among the Montado species. The present study is part of a program to evaluate the essential oil properties from aromatic plants of the Montado Field Station of Herdade da Ribeira Abaixo, located on the eastern slope of Serra de Grândola (Baixo Alentejo, Portugal). A total of eleven lavender collective samples were collected in different locations in the field station, in four consecutive years. The essential oils were isolated from lavender flowering parts, and analyzed by GC and GC/MS. *L. luisieri* essential oils were obtained in a yield 0.1-0.7% (v/w). Fifty-one components were identified, representing 71-91% of the total essential oils. A remarkable quantitative variability was detected in the essential oils, which were mainly constituted by 1,8-cineole (1-36%) and the characteristic necrodane derivative, *trans*- α -necrodyl acetate (1-20%), only present in this lavender species. 5-Methylene-2,3,4,4-tetramethylcyclopent-2-enone (2-16%), α -pinene (1-9%), camphor (tr-8%) and 3,5-dimethylene-1,4,4-trimethylcyclopentene (2-7%) were also present in high relative amounts. The variability in the relative amounts of *trans*- α -necrodyl acetate, 1,8-cineole and camphor was also previously reported for plants collected in different regions of Portugal and Spain.

Key Words

Lavandula luisieri, Lamiaceae, essential oils, Montado, oak forest, *Quercus* forest, cork oak woodlands.

[P-154]

Composition of Essential Oil of *Ajuga bombycina* Boiss

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Abstract

The genus *Ajuga* L. consist of about 90 species, mostly distributed in the north temperate zone of the old world. The genus also occurs in South Africa and Australia. In the flora of Turkey *Ajuga* is represented by 14 species and 27 taxa. Some *Ajuga* species have been widely used for their diuretic, antipyretic, tonic, diaphoretic, astringent properties in the Turkish folk medicine. *Ajuga bombycina* Boiss., used in this study, was collected from two different localities in Alanya-Hadim (Sample A) and Alanya-Kale (Sample B) in Turkey. They were investigated for their essential oil compositions. The hydrodistilled essential oils were analyzed both by gas chromatography (GC-FID) and gas chromatography/mass spectrometry (GC/MS). β -Pinene (25.7% and 14.3%), germacrene-D (23.5% and 32.6), α -pinene (16.5% and 12.0%) and β -caryophyllene (5.9% and 10.8%) were found as major components in the oil of sample A and sample B, respectively. Results were in correlation with our previous studies.

Key Words

Ajuga bombycina, Lamiaceae, GC-FID, GC/MS.

[P-155]

Variability in essential oil composition of *Piper dilatatum* L.C. Rich

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Abstract

Twelve samples of the aerial parts of *Piper dilatatum* L. C. Rich yielded essential oils from 0.2–1.2% and their volatile constituents were analyzed by GC and GC/MS. In total, 141 components were identified, covering more than 90% of the composition. Sesquiterpenes, both hydrocarbons and oxygenated, were the most highly represented classes, the former ranging from 31.5% to 87.7% and the latter varying from 1.8% to 49.4%. The monoterpene hydrocarbons were represented secondarily in the oils, ranging from 0.6% to 42.7%. Using hierarchical cluster analysis, the oils were divided into seven chemotypes whose main constituents were: (E)-caryophyllene, α -cadinol and germacrene D (chemotype A); spathulenol, bicyclogermacrene and (Z)- β -ocimene, (chemotype B); spathulenol, germacrene D and (E)-nerolidol, (chemotype C); germacrene D, limonene, α -phellandrene and bicyclogermacrene

(chemotype D); β -elemene, germacrene D and β -pinene (chemotype E); curzerene, p-cymene and α -eudesmol (chemotype F); and (Z)- α -bisabolene, curzerene and germacrene D (chemotype G). The chemical composition of oils typically varies with season, plant age and soil composition. We think that the analyzed oils showed qualitative and quantitative variation influenced by environmental conditions of collection sites. We have seen that *Piper* oils from the Amazon present terpenoids and phenylpropanoids as major constituents, with the predominance of one over another. The essential oils of *P. dilatatum* containing only mono- and sesquiterpenes as its major components, is further chemotaxonomic evidence of this dichotomy in the *Piper* genus. At the same time, its chemical types may have ecological significance in the management and utilization of the species.

Key Words

Piper dilatatum, Piperaceae, essential oils, chemotypes, hierarchical cluster analysis, mono- and sesquiterpenes.

[P-156]

Link between terpenoid emissions and mango fruit yield and quality

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Abstract

Infestation of mango (*Mangifera indica* L.) leaves by the mango gall fly (*Procontarinia*) may be linked to the emission of terpenoid volatiles. Lenticel discoloration is a cosmetic condition that develops post-harvest as a result of volatile emissions from the fruit. These factors result in reduced fruit yield and consumer resistance, respectively. For this study, three commercial mango cultivars that display varying gall fly and lenticel susceptibility were selected to investigate a possible volatile trigger to gall fly attraction, as well as lenticel formation. Chemometric models were used to analyze metabolomic data and identify biomarkers for susceptibility. Orthogonal partial least square analysis (O-PLS) allowed two sets of data to be correlated and was used to predict data for new observations, derived from samples not included in the model. Susceptibilities of cultivars to gall fly infestation were found to be associated with the terpene emissions of mango flush leaves. A chemometric model identified α - and β -pinene, as well as camphene, as biomarkers for susceptibility to gall fly attack. The developed model can be a useful tool to predict the susceptibilities of new cultivars intended for cultivation. The pinene terpenes were also linked to vulnerability of the fruit to lenticel discoloration. Taking this into account, pack house practices can be adapted to prevent buildup of volatiles in the atmosphere surrounding harvested fruit.

Key Words

Mango gall fly, lenticel discolouration, volatiles, susceptibility, chemometrics.

[P-157]

Chemical variation and phytoremediation potential of *Helichrysum splendidum*

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Abstract

Helichrysum splendidum Less., an indigenous South African shrub, has potential for phytoremediation, since some *Helichrysum* species are known to be metal tolerant. Phytoremediation involves plants that naturally inhabit the polluted environment. The essential oil compositions of natural populations were investigated for potential commercial application to provide for local beneficiation in mining regions. Plants were harvested from different localities to study the seasonal chemical variability. Oils were isolated by hydrodistillation and analyzed by gas chromatography. The analyses revealed the presence of compounds known to possess antimicrobial activities. Seasonal changes and growth stage were found to influence the chemical compositions of the oils. The antifungal activities of the essential oil were evaluated against various fruit and grain crops including *Penicillium* from cactus pear, *Lasiodiplodia theobromae* and *Alternaria alternata* from avocado, *Fusarium oxysporum* from maize and *Penicillium digitatum* from citrus. The *in vitro* assays were performed using different concentrations of oil, and in some cases, a 100% inhibition of fungal growth was observed. The results of this investigation may contribute to the development of new antifungal agents for fungal control. Currently the effects of metal contamination on the essential oils are being investigated, since stress induced by contamination may increase secondary metabolite yields.

Key Words

Helichrysum, essential oils, fungal pathogens, phytoremediation

[P-158]

Study of changing in the essential oil composition of Valerian (*Valeriana officinalis* L.) due to different sowing date and planting density at Iran

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Abstract

Valerian (*Valeriana officinalis* L.) is a medicinal plant that is very a useful sedative for headache. In order to investigate of changing in the essential oil composition of Valerian (*Valeriana officinalis* L.) due to different sowing dates and planting density under conditions of Pakdasht, Iran, this study was conducted at the Experimental Farm on Abureyhan Campus, University of Tehran, Iran. The experimental design was a split plot on the basis of completely randomized block design with four replicates. The main factors, including sowing dates (10 August, 1 September and 20 September) and subfactors, including planting densities (40,000, 80,000 and 120,000 plants ha⁻¹), were studied. The volatile constituents of the root part of cultivated *Valeriana officinalis* were isolated by steam distillation and analyzed by the GC and GC/MS systems that were identified the 87 compositions. The results showed that sowing date and planting density significantly affected essential oil percentage, camphene, bornyl acetate, valerenal and valeranone ($p \leq 0.01$). The highest essential oil percentage (2.2%), camphene

(5.8%), bornyl acetate (30.33%), valerenal (17.05%) and valeranone (3.7%) were provided by the 40,000 plants ha⁻¹ planting density and 20 September sowing date. It was thus concluded that sowing date and planting density are the main factors affecting quantity and quality yields of Valerian. Our finding may give applicable advice to commercial farmers and medicinal and aromatic plants researchers for management and concern of planting density strategy and careful estimating of sowing dates for an increase of quantity and quality yields in medicinal and aromatic plants farming.

Key Words

Valerian, essential oil, sowing date, planting density.

[P-159]

Genetic diversity and chemistry of essential oils from *Baccharis dracunculifolia*, native species from Brazil

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Abstract

Baccharis dracunculifolia DC. (Asteraceae) is a medicinal plant, native from Brazilian Cerrado and Atlantic Forest, widely used in a folk medicine and also as a source of green propolis. Knowing that essential oil production is influenced by genetics and environmental factors, four populations from two distinct geographic regions, Southeast (Botucatu and Delfinópolis) and Midwest (Itahum and Dourados), were evaluated in order to verify the genetic and the chemical profile of essential oils from populations. The essential oils were analyzed by GC-FID and GC/MS and the genetic diversity by SSR. The results obtained by Principal Component analysis showed variability in essential oils. E-nerolidol was the main component of essential oil from Botucatu (28.3%) and Delfinópolis (38.6%) while in Dourados and in Itahum it was E-nerolidol (26.1% and 19.7%) and spathulenol (22.6% and 23.5%). High percentages of β -pinene (12.2%, 5.3%, 8.9%, 12.8%) and limonene (13.6%, 10.6%, 9.8%, 11%) were observed in leaf essential oils from Botucatu, Delfinópolis, Dourados and Itahum. The total genetic diversity was relatively high ($H_t = 0.663$), being that most occurrences were at own populations ($H_s = 0.644$). The interpopulation diversity ($D_{ST} = 0.019$) was not significant, suggesting no structuring among populations. These results were confirmed by UPGMA, showing the existence of just one group of population, forming a hierarchical pattern of genetic similarity. Thus, we concluded that, because there is no genetic divergence among the populations, the divergences at chemical profiles of essential oils can be attributed to environmental factors due to the local of populations' occurrence.

Key Words

Baccharis dracunculifolia, essential oil, molecular markers.

Antifeedant activity of nootkatone and its derivatives on the settling behaviour of the peach potato aphid *Myzus persicae* (Sulz.)

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Abstract

Aphids are pests that cause damage in agriculture which inhibits the growth of plants and buds, causes string ties to weaken, and increases susceptibility to frost and infection with different pathogens. Peach-potato aphid *M. persicae* is considered one of the most harmful pests. It is a cosmopolitan and extremely polyphagous vector of more than 100 plant virus species and it has developed resistance to almost all available aphicides. The purpose of our study was finding the natural compounds which possess good antifeedant activity against *M. persicae* and can be applied as alternatives to conventional neurotoxic chemicals in pest insect control. We evaluated the behavioral effect of biologically active sesquiterpenoids isolated from plants. Nootkatone and its structural derivatives obtained in biotransformations process were tested against *M. persicae*. This bicyclic sesquiterpene ketone, nootkatone, was chosen as a starting material for the enzymatic reactions because it is known as the potential insecticide against larvae of *Drosophila melanogaster* and a strong repellent to termites (*Coptotermes formosanus*). Aphid settling was assessed using the half-leaf choice-test: compounds were applied on one half of the leaf, the other side of the midrib was treated with ethanol and acted as a control. The data were analyzed using a statistic program. In the group of studied compounds we can see that almost all the derivatives of nootkatone with additional oxygen functions in the structure possess good antifeedant activity. The most active compounds are nootkatone, dihydroxynootkatone, triol, 9-hydroxynootkatone and derivatives with oxirane ring.

Key Words

Antifeedant activity, nootkatone, peach potato aphid, natural products.

[P-161]

Evaluation of Essential oil and Secon metabolites difference of valerian (*Valeriana officinalis* L.) under sowing dates and planting densities in Iran

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Abstract

This study was conducted on experimental field of Abureyhan Campus, Tehran University in the Pakdasht zone in Iran during 2005-2006. The experimental design was a split factorial on the basis of completely randomized block design with four replicates. The main factor including sowing dates (10 August, 1 September and 20 September) and subfactor including planting densities (40,000, 80,000 and 120,000 plants ha⁻¹) were studied. The volatile constituents of the root part of cultivated *V. officinalis* were isolated by steam distillation and analyzed by GC and GC/MS systems that identified 87 compositions. The results showed that sowing date and planting density significantly affected essential oil percentage, camphen, bornyl acetate and valerenal ($P \leq 0.01$) and the highest of all plant values were provided by the 40,000 plants ha⁻¹ planting density and 20 September sowing date. It was thus concluded that sowing date and planting density are the main factors effective on quantity and quality yields of valerian. Our finding may give applicable advice to commercial farmers and medicinal and aromatic plants researches for management and concern of planting density strategy and careful estimate of sowing date for the increase of quantity and quality yields in medicinal and aromatic plants farming.

Key Words

Sowing date, planting density, essential oil, Valerian, Didrovaltrate and Valernic acid.

[P-162]

The Effects of Extraction Time and Different Harvest Stages on the Essential Oil Content and Composition of *Foeniculum vulgare* Mill. cv. Soroksari Cultivated in Iran

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Abstract

Fennel (*Foeniculum vulgare* Mill. cv. Soroksari), is an aromatic as well as medicinal crop. The changes in bio-active components in the fruits of *F. vulgare* relate to harvest time during different maturation stages (immature, premature, and mature) and germplasm. In order to study the effects of distillation time of essential oil (60, 90, 120, 180 min) and stages of maturity, chemical composition of the essential oil were measured. Essential oil of different fruits were obtained by hydrodistillation. Analysis of variance showed that extraction time of essential

oil and maturation stages of fruits has a significant effect on the essential oil of fennel. Therefore, the highest essential oil (6%) was measured after 180 min and the lowest level (5.09%) after 60 min; also, with fruit maturity essential oil content declined. The essential oils of *F. vulgare* were characterized by gas chromatography (GC) and gas chromatography/mass spectrometry (GC/MS). In total 16 compounds were identified in the oil, and the main characteristic of the oils is the high content of E-anethole, fenchone and α -pinene. The amount and percentage of major E-anethole fennel essential oils combined in different periods were different. Maximum E-anethole content (72.72%) was observed after 90 min and the lowest (58.52%) after 60 min, respectively. This may be critical for the quality control of essential oils of *F. vulgare*. Our findings determined appropriate extraction time of essential oil of fennel fruit most suitable for obtaining the essential oil quantity and quality time is 90 min at the immature stage of fruit.

Key Words

Fennel, extraction time, E-anethole, Apiaceae, aromatic plants.

[P-163]

Effects of abiotic environmental conditions on composition of α -pinene enantiomers and amount of essential oil in *Juniperus communis* L.

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Abstract

Unripe cones and leaves of *Juniperus communis* L. were sampled from 11 habitats which differed by light conditions and soil characteristics across Lithuania. Essential oils of leaves and cones from 110 samples (trees) each were isolated by hydrodistillation in European Pharmacopoeia apparatus for 2 h. α -Pinene enantiomers were separated by chiral-phase capillary GC and identified by matching their retention times to optically pure analytical standards. The average contents of essential oils varied between habitats from 0.28–0.52% in leaves and from 0.91–1.62% in cones. The contents of essential oils in leaves and unripe cones were by 29% and 14% higher in shaded habitats than in open ones, respectively. However, significant negative correlation ($r = -0.69$, $p < 0.05$) between light conditions and the contents of essential oils was established in leaves only. No correlations were established between the contents of essential oils and soil characteristics in either leaves or cones. Ratio of (1R)-(+) and (1S)-(-) enantiomers of α -pinene varied from 2.62–4.36 in leaves and from 2.22–5.74 in cones. No light effect was established on the ratio of α -pinene enantiomers in the essential oils of cones and leaves. However, significant correlations ($r = -0.60$ and $r = -0.69$; $p < 0.05$) were observed between the ratio (1R)-(+)/(1S)-(-) of α -pinene enantiomers in leaves and the contents of organic nitrogen and humus in soil, respectively. The study has revealed that abiotic environmental conditions affect the contents of essential oils and ratio of α -pinene enantiomers in leaves more than that in unripe cones of *J. communis*.

Key Words

Juniperus communis L., essential oils, α -pinene enantiomers, environmental conditions.

[P-164]

Preparative isolation of monoterpenes isomers from citral-rich essential oil of *Pectis brevipedunculata* essential oil by High Speed Countercurrent Chromatography (HSCCC)

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Abstract

Many plant species in Brazil are known as lemongrass due to the citric fragrance of their volatile compounds. *Pectis brevipedunculata* Sch.Bip., a Brazilian ornamental aromatic grass, is one of these species. Its EO content is characterized by the high content of citral (3,7 dimethyl-2, 6-octadienal) up to 90.0%. Citral is one of the most important constituents of citrus flavors, used as a taste enhancer, widely used in foods or as an odorant in perfumes. This monoterpene aldehyde fraction is normally composed by a mixture of the 2 geometric *cis*- and *trans*-isomers, geranial and neral in a 3:2 ratio that has been related to many biological activities. Fresh samples of aerial parts of *P. brevipedunculata* were collected in Rio de Janeiro and were submitted to hydrodistillation for 2 h in a Clevenger-type apparatus. The obtained EO was analyzed by GC and GC/MS. The EO (1 mL) was purified through a HSCCC equipment using hexane: acetonitrile (1:1) as solvent system. Isocratic elution was conducted with the acetonitrile lower phase as stationary phase. Rotation of the coil was 860 rpm, and the upper phase was pumped at a flow rate of 1.0 mL/min. Fractions (200) of 2 mL each were collected. The collected fractions were analyzed by TLC, GC and GC/MS. HSCCC is a useful tool to separate of non-polar compounds due to high percentage of mass recovery and the high purity level and quantity of the isolated isomers neral/geranial (100.0%), nerol/geraniol (96.0%), geraniol (85.97%), geranial (91.0%) and neral (81.0%).

Key Words

Pectis brevipedunculata, essential oil, citral, HSCCC, isomers.

[P-165]

Volatile constituents of Blue Moon & Blue Perfume, classified blue type HT rose flower

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Abstract

The volatile constituents of Blue Moon and Blue Perfume species, which were classified blue type, depend on its scent in HT roses, were analyzed using Aromascope technology and solvent extraction method followed by GC/MS analysis. Amongst total 156 components were identified from Blue Moon, geraniol, β -citronellol, nerol, 1,3-dimethoxy-5-methylbenzene (DMMB) and β -dihydroionol were qualified as the main odor components. On the other hand, amongst total 136 components analyzed from Blue Perfume, geraniol, β -citronellol, nerol, and geranial with fresh citrus lemon like note were identified. From both flowers, three components were newly identified, 2-isopropyl-4-methyl-thiazole (1) with a characteristic pungent vegetable tropical notes, (Z)-yuzu lactone (2), and methyl epijasmonate (3). And the small addition of compound (1) to rose type fragrance showed better effect and also highly contributed for Blue Moon and Blue Perfume notes in ground. And then a small addition

of compound (3) to a rose type fragrance also enhanced the natural scenting effect, resulting in a performing rose fragrance. Several components identified from both flowers had some asymmetric carbons in their molecules, leading to analyzing their chirality. For the first time, the enantiomers ratio of linalool, *trans*-nerolidol, theaspiranes and β -dihydroionol could be clarified by using MDGC/MS. The results were followings in both rose flowers. The ratio of (S)-isomer vs (R)-isomer of linalool was 8:92. Only (3S)-enantiomer was detected for *trans*-nerolidol. The ratio of (2S)-isomer vs (2R)-isomer in theaspirane A and theaspirane B were about 96:4. Finally, no absolute stereochemistry determined for β -dihydroionol, only (+)-isomer was present.

Key Words

Rose, Blue Moon, Blue Perfume, volatile constituents, enantiomer.

[P-166]

Authenticity assessment of citrus essential oils

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Abstract

Multidimensional Gas Chromatography (MDGC) with a chiral column in the second dimension, and Gas Chromatography Combustion Isotope Ratio Mass Spectrometry (GC-C-IRMS) represent powerful analytical tools for the quality assessment of the genuineness of Citrus essential oils. In fact, in addition to the conventional analytical methods (GC, GC/MS, HPLC, etc.) these techniques can be applied to reveal sophistications of different nature, such as the subtle addition of single natural components of different botanical sources. Enantio MDGC is the most powerful tool to determine with high accuracy the enantiomeric distribution of chiral volatile components, avoiding interferences. GC-C-IRMS is applied to determine the isotopic ratios of selected components, which are biogenetically related to the plant metabolism. These values are characteristic of the botanical source of the plant material, and often related to its geographic origin. These two techniques have been successfully applied to determine the enantiomeric excess (EE) and the isotopic ratio ($\delta^{13}\text{C}$) of selected volatile components (monoterpene and sesquiterpene hydrocarbons, aldehydes and alcohols) in numerous samples of authentic cold-pressed and distilled Citrus oils, of different geographic origin and commercial oils. The results of the two analytical approaches are compared. The genuineness assessment was accomplished based on the range of authenticity determined by the EE and $\delta^{13}\text{C}$ from genuine samples of secure origin. The results attained are often complementary, thus providing additional information for the evaluation of natural, natural-identical or synthetic mixtures. Useful information on the natural occurrence of chiral compounds and on the naturally occurring $\delta^{13}\text{C}$ values in authentic oils are provided.

Key Words

Enantio-MDGC, GC-C-IRMS, citrus oils.

[P-167]

Chemical Composition of Essential Oils from Different Parts of Endemic *Bupleurum* in Turkey

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Abstract

The chemical constituents of the essential oils from the flowers, fruits and roots of *Bupleurum heldreichii* Boiss. & Bal., *Bupleurum sulphureum* Boiss. & Bal., *Bupleurum turcicum* Snogerup, *Bupleurum pauciradiatum* Fenzl and *Bupleurum lycaonicum* Snogerup were analyzed by GC and GC/MS systems simultaneously. The major components in the oils from flowers, fruits and roots of *B. heldreichii* were germacrene D and hexadecanoic acid, respectively. Main constituents in the oils from flowers, fruits and roots of *B. sulphureum* were undecane and carene, respectively. The major components in the oils from flowers, fruits and roots of *B. turcicum* were heptanal and pentacosane, respectively. Main constituents in the oils from flowers, fruits and roots of *B. pauciradiatum* were germacrene D, β -pinene and spathulenol, respectively. In the oils from flowers, fruits and roots of *B. lycaonicum*, the principal compounds were tridecane and spathulenol, respectively.

Key Words

Bupleurum heldreichii, *Bupleurum lycaonicum*, *Bupleurum pauciradiatum*, *Bupleurum sulphureum*, *Bupleurum turcicum*, essential oil composition.

[P-168]

Essential Oil Composition from Different Parts of Some *Bupleurum* species from Turkey

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Abstract

The compositions of the essential oils from different parts of *Bupleurum intermedium* Poir., *Bupleurum lancifolium* Hornem., *Bupleurum cappadocicum* Boiss., *Bupleurum falcatum* L. subsp. *cernuum* (Ten.) Arcang., *Bupleurum croceum* Fenzl and *Bupleurum gerardii* All., including roots, flowers and fruits, were investigated by GC and GC/MS systems, simultaneously. Methyl linoleate, germacrene D and undecane were major compounds in the oils from flowers, fruits and roots of *B. intermedium*. The main constituents of oils from flowers, fruits and roots of *B. lancifolium* were spathulenol, hexacosane and hexadecanoic acid, respectively. Main constituents separated in the oils from flowers, fruits and roots of *B. cappadocicum* were heptanal and undecane, respectively.

The oils from flowers, fruits and roots of *B. falcatum* subsp. *cernuum* contained, as main components, α -pinene and amyl furan. The main constituents of oils from flowers, fruits and roots of *B. croceum* were germacrene D, undecane and hexadecanoic acid. Undecane and hexanal were major compounds in the oils from flowers, fruits and roots of *B. gerardii*.

Key Words

Bupleurum cappadocicum, *Bupleurum croceum*, *Bupleurum falcatum* subsp. *cernuum*, *Bupleurum gerardii*, *Bupleurum intermedium*, *Bupleurum lancifolium*, essential oil composition.

[P-169]

The Effects of Extraction Time on the Essential Oil Content and Composition of Lemon verbena (*Lippia citriodora*) Cultivated in Iran

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Abstract

Lemon verbena (*Lippia citriodora* Kunth) is used worldwide for the sensorial and medicinal properties of its leaves and essential oil. Extraction time of essential oil is one of the important factors affecting quality and quantity of the essential oil of medicinal plants, thus selecting the optimum extraction times is very important in medicinal plants. In order to investigate the influence of extraction time on the essential oil content and composition this plant, an experiment was carried out in the College of Agriculture of Tarbiat Modares University in 2010. Four extraction times used were: 60, 90, 120 and 180 min. Essential oil was obtained by water distillation method using a Clevenger-type apparatus. Results showed that extraction time did not have significant effects on essential oil content but highest essential oil content (0.73%) was obtained at 120 min time. Based on the results of essential oil analysis by gas chromatography (GC) and gas chromatography/mass spectrometry (GC/MS), 21 major compounds were observed in Lemon verbena essential oil. Geranial (29.65%), neral (23.16%) and limonene (15.38%) were known essential oil compositions. The highest geranial was observed at 60 min time.

Key Words

Extraction time, essential oil, Lemon verbena, medicinal plant.

[P-170]

Biotransformation of Unsaturated Terpenoid Lactone with the p-Menthane System

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Abstract

Being interested in the functionalization of lactones, we have studied the possibility of their biotransformation with the fungi strains as biocatalysts. The hydroxylation of the substrate was the common transformation of saturated lactones 1, 2 and halolactones 3, 4. However, in some fungi cultures, the epoxidation of double bond was also observed. Here we present the results of microbial transformations of racemic unsaturated lactone with the p-menthane system. Several biocatalysts were screened to check their ability to transform this substrate. Three of them (*Absidia cylindrospora* AM336, *Absidia glauca* AM177 and *Syncephalastrum racemosum* AM105) showed the ability of hydroxylation of the studied lactone. The hydroxy group was introduced in allylic positions to give lactones 2 and 3 as products. The composition of product mixture was dependent on fungi strains. The reaction progress was checked by thin layer and gas chromatography. The structures of obtained products were confirmed by spectral data (¹H NMR, ¹³C NMR, HSQC, COSY and IR). The optical purity of hydroxylactones was determined by chiral gas chromatography.

Key Words

p-Menthane lactones, biotransformation, hydroxylation.

[P-171]

Lipase-catalyzed kinetic resolution of 1-(6,6-dimethylbicyclo[3.1.0]hex-*trans*-3-yl) ethanol

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Abstract

Biocatalysis is an effective tool for the structural modification of bioactive, natural and synthetic compounds. Enzymes, especially lipases (EC 3.1.1.3), are very useful in the preparation of a broad spectrum of compounds in their optically active forms. These enzymes possess wide substrate specificity, generally high chemo-, regio- and stereoselectivity, and in many cases perform under mild conditions. Their ability to catalyze hydrolysis, as well as transesterifications, is well recognized and has been described in a number of published papers on lipases. The first step to obtain pure diastereoisomers of designed compound was the chemical synthesis of substrates for biotransformation (alcohol 4 and acetate 5) followed by lipases-mediated transesterification in anhydrous conditions, and enzymatic hydrolysis in a biphasic system. Reaction of 1 with N-bromosuccinimide gave bromohydrine, which, after treating with silver oxide, is transformed to the bicyclic acetyl derivative 2. Reduction of ketone 3 with LiAlH₄ led to the desired alcohol 4. Acetate (R,S)-5 was obtained from a mixture of alcohol (R,S)-4 in a one-step reaction in a pyridine medium with the addition of acetyl chloride. Several kinds of lipases were used and

after screening, an appropriate enzyme was selected and influence of solvent and temperature were investigated. In both cases bioconversion to the one form of acetate or alcohol were observed followed by separation of pure diastereoisomers. Synthetic and biotransformation details of the applied procedures and olfactory properties of new derivatives will be presented.

Key Words

Lipases, biotransformation, stereochemistry, terpenoids.

[P-172]

**Studies on Essential Oil Bearing Plants from Mozambique.
Synthesis of Rose Oxide**

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Abstract

The *cis/trans*-rose oxide was prepared from citronellal (3,7-dimethyloct-6-enal) isolated from *Eucalyptus citriodora* via sequential treatment with tetrabutylammonium borohydride (TBABH₄) in dichloromethane (DCM) to give citronellol (3,7dimethyloct-6-en-1-ol). Dye-sensitized photooxidation with simultaneous reduction of the hydroperoxides with TBABH₄ gave a mixture of the alkenediols 3,7-dimethyl-7-octene 1,6-diol and 3,7-dimethyl-5-octene-1,7diol in 91% yield. These diols were converted into *cis/trans*-rose oxide by a two-stage treatment with p-toluenesulfonic acid and sulfuric acid in a one pot reaction giving 69% overall yield.

Key Words

Citronellal, photooxidation, *cis/trans*-rose oxide.

[P-173]

Investigation of the essential oils from peels, blossoms and leaves from *Citrus aurantium* L. growing in Greece

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Abstract

Several varieties of *Citrus aurantium* L. (sour orange) are important for their essential oils, which are used as flavoring agents in foods and drinks, in perfumes and also in cosmetology. *Citrus aurantium* extracts have also been employed in herbal traditional medicine as a stimulant, or as an appetite suppressant. The tree is widely used mostly as a Citrus rootstock in all Citrus-producing areas of the world, rather than for its fruit. Furthermore, in Greece and Cyprus it is also used as a popular ornamental tree. The aim of the present study is the determination of the yield and composition of *C. aurantium* essential oils from local Greek cultivars. Fruits, blossoms and leaves were collected from *C. aurantium* trees growing in the district of Thessaloniki, Greece. The fresh plant material—fruit peels, leaves and blossoms, respectively—were subjected to hydrodistillation using a modified Clevenger distillation apparatus. The essential oils yield was estimated at 2.70% (mL/100 g) for the peels, 0.12% (mL/100 g) for the blossoms and 0.22-0.46% (mL/100 g) for the leaves. The GC/MS analysis of the essential oils revealed that monoterpene hydrocarbons was the main group of constituents in all the obtained oils. Limonene was the predominant component in peels oil (94.6%). Linalool was found to be the main constituent of blossom oil, whereas linalool and linalyl acetate accounted for more than 50% in the essential oil from leaves.

Key Words

Citrus aurantium, essential oils, leaves, peels, blossoms.

[P-174]

Use of Lemongrass Essential Oil Fractionated in Perfumery

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Abstract

Lemongrass (*Cymbopogon citratus* (DC.) Stapf) is native to Southern India and Sri Lanka and is an important aromatic grass, principally for its essential oil. It is mainly used in the perfumery and cosmetics industries. Unlike most market samples of lemongrass essential oil, the sample developed in Brazil by Natura showed slight solubility in alcohol 96%. The aim of this work was to solve this insolubility using fractionated distillation. The trials were done using typical organic lemongrass (Ecocert certification) collected in the Turvo community, state of Parana, and submitted to vacuum fractioned distillation. The essential oil was fractionated in 11 fractions (plus residue). Based on their composition (analyzed by gas chromatography/mass spectrometry) and solubility, the fractions were combined maintaining the mass proportion and olfactory features. Through these results it was possible to

conclude that the insolubility problem is not caused only by waxes and gums, since the removal of residue was not enough to obtain a soluble sample, but also because of the presence of beta-myrcene and a low concentration of citral. Therefore, a last experiment was performed removing only the beta-myrcene without distilling all the oil (making a terpeneless oil). The final reconstituted essential oil (85.94% of citral and 0.16% of beta-myrcene) is clearly soluble in a concentration of 1-5% in ethyl alcohol 96% with a pleasure of fruity, sweet, tea and lemon juice odor. This shows that the fractionated distillation after the complete distillation seems to be a good approach to eliminate these by-products and guarantee a good enough product.

Key Words

Cymbopogon citratus, essential oil, fractionament, citral and perfumery.

[P-175]

Chemical Composition of Essential Oil of *Syzygium Aromaticum* (L.) Merr. & Perry Acquired on Municipal Market in Pedreira, São Paulo, Brazil

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Abstract

Syzygium aromaticum (L.) Merr. & Perry (clove tree) belongs to Myrtaceae family. It is endemic in the North Moluccas (Indonesia) and was of old cultivated on the islands of Ternate, Tidore, Bacan and the West coast of Halmahera. Dried flower buds present essential oil that has eugenol as the main compound. Eugenol acetate and β -caryophyllene are also found in this essential oil. It is popularly known in Brazil as cravo-da-Índia and this spice is commonly used in Brazilian traditional culinary and perfumery, due to its characteristic aroma and pungent flavor. Clove is used in folk medicine and as a phytomedicine as anesthetic, antifungal, antibacterial and antioxidant. The aim of this work was to study the chemical composition of commercial samples of clove acquired in a municipal market in the Pedreira district (São Paulo, Brazil). The essential oil of dried flower buds of clove was extracted by hydrodistillation (Clevenger-type apparatus; 4 h) and analyzed by GC/MS. Identifications of chemical compounds were made by matching their mass spectra and Kovat's indices (KI) values with known compounds reported in the literature. Seventeen compounds were characterized in the oil. Major compounds were eugenol (60%) followed by β -caryophyllene (9.7%) and 1,8-cineole (9.4%). This result shows that commercial samples of clove present the phytochemical quality required for their use in folk medicine.

Key Words

Clove tree, eugenol, hydrodistillation, β -caryophyllene, 1,8 cineole.

[P-176]

Prospection of Aromatic Plants from Dense Ombrophilous Forest in Paraná State, South Brazil

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Abstract

The great plant biodiversity in Brazil, particularly in the Atlantic Forest, represents a great potential for the development of sustainable new agricultural products, essential oils among them, for the benefit of both small farmers and the aroma and fragrance industries. This work had as its main objective the identification of aromatic native species from the dense Ombrophilous Forest of Paraná State Coast region with potential application. Fifty-one samples of different plant species were collected from the Natural Reserve of Cachoeira river, belonging to the Society for Research and Protection of Wild Life in Antonina, Paraná State. The species were identified at the Botanical Museum of Curitiba and the essential oil extracted from fresh leaves by hydrodistillation in a Clevenger type apparatus for 2.5 h. The essential oils were analyzed by GC and GC/MS. From the species collected, 37 presented essential oil, most of them belonging to Myrtaceae, Lauraceae and Piperaceae families. Some substances were found in high content (> 30%) in several species, namely spathulenol (47.0%) in *Xylopia brasiliensis* Spreng., caryophyllene oxide (41.7%) in *Copaifera trapezifolia* Hayne, eugenol (40.0%) and β -bisabolene (34.8%) in *Ocimum gratissimum* L., (E)-nerolidol (35.8%) in *Piper corcovadense* C.DC., decanal (52.7%) in *Peperomia emarginella* C.DC., and benzyl benzoate (73.7%) in *Endlicheria paniculata* (Spreng.) J.F.Macbr. From the data collected, it was found that some species could be cultivated as raw materials for the aroma industry.

Key Words

Ombrophilous dense forest, essential oils, Brazil biodiversity, spathulenol, nerolidol.

[P-177]

Chemical composition of volatile oil phase recovered from the processing of orange juice

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Abstract

Frozen concentrated orange juice (FCOJ) is produced by concentrating juice filtered through vacuum evaporators in multiple stages. During the process some aromatics are removed. Low molecular weight compounds, such as aldehydes, alcohols and terpenes, are recovered in a retrieval system of essential oils which consists of a fractional column and a series of condensers in which two products are obtained: the oil phase and aqueous phase that are re-added to FCOJ in different proportions, to achieve the original flavor lost during the process. The identification of eleven volatile organic compounds in the oil phase was obtained from injection in GC/FID analysis, LRI and with injection of standards. The limonene (91.58%) was a major compound and is also one of the most important contributors to orange flavor. The concentration of some important compounds was observed in recovered concentrate as compared to their essential oil contents. The terpene hydrocarbons were the predominant class of volatile compounds in orange oil phase and are among the largest contributors of the orange aroma note.

Key Words

Citrus, aroma, limonene.

[P-178]

Volatile constituents of two *Labiatae* species from Iran

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Abstract

Salvia and *Stachys* are considered as the largest genera of the family Lamiaceae. The oils obtained by hydrodistillation of the aerial parts of *Salvia brachyantha* (Bordz.) Pobed. and *Stachys byzantina* C. Koch grown in Iran were analyzed by GC/MS. Forty-five compounds representing 95.06% of the oil of *S. brachyantha* were identified, of which 1,8-cineole (10.2%), γ -muurolene (7.5%), caryophyllene oxide (6.7%), spathulenol (6.2%) and elemol (5.9%) were the major constituents. Sesquiterpenes comprised 48.09%, while the monoterpene fraction was 35.8% of the total oil; 11.1% nonterpenoids were also found in the oil. Furthermore, twenty-five components constituting 94.3% of the total oil of *Stachys byzantina* were identified, containing 61.8% monoterpenes, 20.5% sesquiterpenes and 12.0% nonterpenoid compounds. 1,8-Cineole (16.4%), hexadecanoic acid (7.4%), camphor (6.8%) and borneol (5.7%) were found as the main components among 25 constituents in the oil of *S. byzantina*.

Key Words

Volatile constituents, Labiatae, *Salvia*, *Stachys*, 1, 8-cineole, hexadecanoic acid.

Volatile oil yields from fallen and fresh leaves of *Eucalyptus* species cultivated in Nigeria

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Abstract

Eucalyptus trees are common in northern Nigeria and have various uses, especially for its oil. The quality, quantity and some analysis of the oil from the fallen and dried leaves of two *Eucalyptus* species namely *Eucalyptus citriodora* Hook (Myrtaceae) and *Eucalyptus tereticornis* SM (Myrtaceae) were evaluated. These leaves were extracted by hydrodistillation followed by gas chromatography (GC) and thin layer chromatography (TLC) analysis. Solubility, specific gravity, evaporation on filter paper and refractive index of the oils were also determined. The yields were 3.5% and 2.3% for *E. citriodora* and *E. tereticornis*, respectively, for fresh leaves and 2.8% and 1.6% for the fallen leaves, respectively. Oil from fresh *E. citriodora* leaves was colorless with strong lemon scent and that from fallen leaves pale yellow and less lemon-scented. The main constituents identified by TLC analysis were citronellal, citronellol, cineole and geraniol. The quantitative chromatographic analysis revealed citronellal and citronellol as major constituents of *E. citriodora* and cineole as the main constituent of *E. tereticornis*. Oils from fallen leaves were found to contain less oil than fresh leaves. Citronellal from *E. citriodora* was more in fallen leaves than fresh leaves. In fresh leaves, the content of citronellol was more. Cineole was found to be a major constituent of *E. tereticornis*, while *E. citriodora* contained chiefly citronellal and citronellol. These findings show that the fallen leaves, like the dried leaves, also contain valuable constituents that are very useful and potential sources of raw materials for the pharmaceutical and perfumery industries.

Key Words

Eucalyptus species, fallen leaves, fresh leaves, oils, GC, TLC, physical properties.

[P-180]

Essential oil from *Philodendron fragrantissimum*, an Araceae from Amazonia, Brazil

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Abstract

Anthurium and *Philodendron* are the most representative genera of the Araceae family, with *Philodendron* alone represented by more than 700 species, which are characterized by the presence of alkyl resorcinols allergens in the leaves and stems. Until now, only two *Philodendron* species, namely *P. acutatum* Schott. and *P. imbe* Schott., had their essential oils studied. In the Amazon region of Brazil, the roots of different species of *Philodendron* are used in the traditional medicine. Among them, *Philodendron fragrantissimum* (W.J.Hook.) G.Don, locally known as cipó-tracuá with its very aromatic roots, is used for bath and fumigation. In the present work, we describe the chemical composition of the essential oil obtained from the adventitious roots of *P. fragrantissimum*. The plant was collected in August 2010 at Oriximiná, Pará State, Brazil, and a voucher specimen was deposited at the Herbarium of the Instituto Nacional de Pesquisas da Amazônia under number 237609. The essential oils from adventitious roots were obtained by hydrodistillation in a modified Clevenger-type apparatus for 4 h. The oil was analyzed by GC and GC/MS. Thirty-five substances were detected and the major components were β -caryophyllene (29.9%), limonene (15.8%), α -selinene (15.7%), β -selinene (11.5%), α -pinene (3.4%), α -copaene (3.2%), β -pinene (2.5%), δ -cadinene (2.9%) and γ -eudesmol (2.9%). To the best of our knowledge, this is the first report on the chemical composition of the essential oil from *P. fragrantissimum*. The high content of β -caryophyllene was not observed in the other oils studied so far and might be considered as a tool in taxonomy studies.

Key Words

Philodendron fragrantissimum, Araceae, essential oil composition, β -caryophyllene, medicinal plants.

[P-181]

Essential oil composition of *Artemisia verlotiorum* from Northern Austria

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Abstract

Studying the aromatic flora of Eastern Austria, we reported the composition of the essential oils from Chinese Mugwort, *Artemisia verlotiorum* LAMOTTE (Asteraceae). This plant presents a pleasant aroma and is similar to the very common *A. vulgaris*, but flowers late in autumn. It was introduced to Central Europe during the 20th century. Leaves produced about 0.6% (v/dw) essential oil, the stems less than 0.05%. The main oil compounds in the leaf oil were the monoterpenes α -thujone (36.3%), β -thujone (6.2%) and 1,8-cineole (6.0%), and the sesquiterpenes β -caryophyllene (13.1%) and germacrene D (8.3%). Assuming the same response in the FID as for the internal standard, the contents of α -thujone, β -caryophyllene and germacrene D were estimated to 1100, 399 and 252 $\mu\text{g/g}$ dried leaves, respectively. The stem oil gave a fingerprint of mainly sesquiterpenes: β -caryophyllene (22.5%), germacrene D (18.5%), E- β -farnesene (17.9%), caryophyllene oxide (6.4%) and α -humulene (4.3%). In the stems these compounds were each lower than 35 $\mu\text{g/g}$ dry weight. α -Thujone as a major component has already been reported in various *A. verlotiorum* oils. There are also oils rich in borneol and/or camphor or in myrcene. In the present Austrian oil, these compounds were very low. Vegetative plants were collected on a field border near Schrems, Northern Lower Austria. The plants were separated into stems and leaves, dried in the ambient air and hydrodistilled. The obtained oils were diluted with hexane and analyzed by GC and GC/MS using biphenyl as internal standard and the mass spectra and retention indices for compound identification.

Key Words

Artemisia verlotiorum, α -thujone, oil composition.

[P-182]

Essential Oil Compositions of Two *Chaerophyllum* Species Growing in Turkey

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Abstract

The genus *Chaerophyllum* (Apiaceae) encompasses 15 species, three of which are endemic to Turkey. *Chaerophyllum* species, especially *C. macropodum* Boiss., are used as food in herby cheese called as *Van otlu peyniri*. Therefore, it is important to characterize the aroma composition of these species. This study reports the essential oil compositions of the aerial parts of *C. crinitum* Boiss. and *C. macrospermum* (Sprengel) Fisch. et Mey. collected from Bitlis and Hakkari in Turkey, respectively. The aerial parts of the plants subjected to hydrodistillation by using Clevenger apparatus were analyzed by GC and GC/MS simultaneously. The main constituents of the essential oil of *C. crinitum* were found to be (E)- β -ocimene (38.1%), terpinolene (12.7%), α -pinene (5.5%), p-cymene

(5.3%), limonene (5.3%) and p-cymen-8-ol (2.2%). Terpinolene (21.4%), myristicin (18.9%), p-cymen-8-ol (11.9%), limonene (6.1%), α -pinene (4.6%), spathulenol (4.4%), and α -p-dimethyl-styrene (3.6%) were identified as major constituents for *C. macrospermum* essential oil.

Key Words

Essential oil, *Chaerophyllum crinitum*, *Chaerophyllum macrospermum*, Apiaceae.

[P-183]

Essential Oil Composition of *Grammosciadium pterocarpum* Boiss. Growing in Turkey

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Abstract

The genus *Grammosciadium* DC. comprises eight taxa in Turkey (*G. daucoides* DC., *G. cornutum* (Nábek) C.C.Towns., *G. macrodon* Boiss., *G. pterocarpum* Boiss., *G. platycarpum* Boiss. & Hausskn. ex Boiss., *G. confertum* Hub.-Mor. & Lamond, *G. schischkinii* (V.M.Vinogr. & Tamamsch.) V.M.Vinogr., and *G. haussknechtii* Boiss.), three of which (*G. schischkinii*, *G. haussknechtii* and *G. confertum*) are endemic to Turkey. All the species of the genus are Irano-Turanian element, except *G. confertum*. *Grammosciadium pterocarpum* Boiss. is distributed throughout east, south and central Anatolia in Turkey, and northwest Iran. Its altitudinal range changes between 1100 m and 2800 m. *G. pterocarpum* is an erect, branched, glabrous perennial plant with leaves linear to narrowly ovate-oblong. The fruits are oblong in shape and winged up to 2.5 mm. Volatile constituents obtained from crushed fruits and aerial parts of *G. pterocarpum* by micro-distillation methods were analyzed by GC and GC/MS systems, simultaneously. Fifteen components representing 99.7% of the fruit oil were identified, whereas twenty-one components were detected representing 97.4% of the aerial parts' oil. Main constituents of the fruit oil were found to be linalool (68.4%) and β -pinene (22.0%), while caryophyllene oxide (55.1%) and β -caryophyllene (15.3%) were found as major in the aerial parts of plant.

Key Words

Grammosciadium pterocarpum, micro-distillation, GC, GC/MS.

[P-184]

Use of vibrational spectroscopy as a quality control method for assessment of geranium oil

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Abstract

Rose-scented geranium obtained from *Pelargonium graveolens* L'Hér. (Geraniaceae) is a high value oil used extensively in flavor, fragrance and pharmaceutical formulations. The oil is variable in composition with "Bourbon geranium" (Reunion Island) regarded as the highest quality geranium oil. The quality assessment of geranium oil is generally determined by the level of seven major volatile constituents (geraniol, citronellol, geranyl formate, citronellyl formate, linalool, isomenthone and guaia-6,9-diene) using gas chromatographic (GC) techniques. Vibrational spectroscopy together with chemometric algorithms were investigated for use as fast and reliable alternatives in the quality assessment of geranium oil. Geranium oil samples ($n = 70$) obtained from different suppliers (South Africa, Egypt, India, Reunion Island, China and Madagascar) were analysed using GC/MS as reference method. The mid- and near-infrared spectral data of the oils was recorded and partial least squares (PLS) multivariate calibration models developed by regressing the spectra on the GC/FID reference values. The developed calibration models showed very good correlation with coefficients of > 0.90 between the predicted and reference values. Geraniol ($r^2 = 0.99$ and 0.98), citronellol ($r^2 = 0.98$ and 0.97), linalool ($r^2 = 0.97$ and 0.97), citronellyl formate ($r^2 = 0.96$ and 0.95) and geranyl formate ($r^2 = 0.96$ and 0.95) had even higher co-efficients of > 0.95 for MIR and NIR, respectively. Generally, the error parameters (SEE and SEP) after external validation were low ($< 1.0\%$) for all compounds guaranteeing reliable predictions. The results demonstrate the potential use of both MIRS and NIRS as alternative methods in the quality assessment of geranium oil.

[P-185]

Volatile oil composition from *Porophyllum ruderale* (Jacq.) Cass. subs. *runderale* in different phenological stages

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Abstract

Porophyllum ruderale (Jacq.) Cass. ssp *runderale*, known as “couve-cravinho” or “arnica paulistana,” is a ruderal, aromatic and medicinal herb used in folk medicine as wound healing, antimicrobial, anti-inflammatory and analgesic. The plants were cultivated in two biogeographically different regions, Brazilian Cerrado (Mogi-Guaçu, SP) and the Atlantic Rain Forest (Instituto de Botânica-IBt, São Paulo, SP). Plant material (aerial parts) was harvested at three different phenological stages (i.e. vegetative, flowering, and fruiting), and their volatile oils were analyzed by GC/MS. In all phenological stages, the main components were *cis*- β -ocimene, limonene and β -pinene. In the Mogi-Guaçu plants, there was an increase in the concentration of *cis*- β -ocimene and a decrease in the concentration of limonene during the fruiting stage, whereas the inverse was observed in IBt ones. The concentration of the hydrocarbon 1-undecene decreased when the plants were fertile. The presence of myrcene was observed in flowering (Mogi-Guaçu) and fruiting (IBt) at low concentration. There was almost no variation in the relative amounts of all main components with the phenological stages and, the highest yield was obtained for the plants from Mogi-Guaçu during the fruiting stages.

Key Words

Arnica, phenological stages, essential oil, Brazilian Cerrado, Atlantic Rain Forest.

[P-186]

Analysis of chiral coumarins and furocoumarins in Citrus essential oils by means NP-HPLC

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Abstract

Coumarins and psoralens (furocoumarins) play an important role for the characterization of Citrus essential oils, and in fact have received considerable attention for their chemical, biological and pharmacological properties. Some of the coumarins and furocoumarins present in Citrus essential oils have, in their structure, chiral centers, hence a separation of enantiomers could be an important tool to improve knowledge on these oils. However, up to now the enantiomeric distribution of coumarins and psoralens in citrus oils has never been reported. The recent introduction of new chiral stationary phases for liquid chromatography contributed to the development of efficient methods for the separation of enantiomers. In this work, the use of a column packed with cellulosic-based chiral stationary phase (250 x 4.6mm, 5 μ m), has provided an excellent baseline separation of the enantiomers in Citrus

essential oils. Chiral coumarins and furocoumarins, used to optimize the NP-HPLC method, were obtained by synthetic process and natural compound were isolated from lemon (oxypeucedanin, oxypeucedanin hydrate, byakangelicol, byakangelicin) and grapefruit (epoxybergamottin and epoxyaurapten) essential oil residues by preparative RP-HPLC. The NP-HPLC method optimized in this work, allowed to determine the enantiomeric distribution of chiral furocoumarins oxypeucedanin hydrate, byakangelicin, oxypeucedanin, byakangelicin in lime and lemon essential oils, and of epoxyaurapten and epoxybergamottin in grapefruit essential oil. Essential oils obtained during a whole production season using different extraction technologies were analysed to detect possible modification of the enantiomeric ratios. In addition oils from different geographical origin were also studied.

Key Words

Enantiomeric HPLC separation, chiral coumarins and furocoumarins, Citrus essential oils.

[P-187]

Essential oil compositions of 22 volatile oils from 8 Nigerian herbs utilized as anti-malarial in ethno-medicine

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Abstract

In our quest for active metabolites in natural products which are lesser known, we report volatile compounds from eight herbs utilized as anti-malarial in southwest Nigerian ethno-medicine. Most abundant volatiles from GC/MS analyses of twenty-two essential oils procured from different parts [leaf, bark, fruit and root parts] of eight herbs obtained in good yields [0.05-1.26%] are reported. The eight plants and their families with parts studied and number of compounds in each are *Alstonia boonei* De Wild. [Apocynaceae]- leaf (14), bark (14), root (12); *Dialium guinense* Willd. [Leguminosae]- leaf (25), bark (21), fruit (18); *Nauclea latifolia* S.M. [Rubiaceae]- leaf (25), bark (11), root (21); *Morinda lucida* Benth. [Rubiaceae]- leaf (15), bark (15), root (14); *Ficus exasperata* Vahl. [Moraceae]- leaf (9), bark (19), root (25); *Anogeissus leiocarpus* (DC.) Guill. & Perr. [Combretaceae]- leaf (11), bark (13), root (14); *Lonchocarpus cyanescens* Benth. [Leguminosae]- leaf (10), bark (12); *Harungana madagascanensis* Lam. ex Poiret [Hypericaceae]- leaf (11). Identified compounds constitute 80-100% of the oil. Each oil contain compounds which range from 9 to 25. They are terpenoid, non-terpenoid hydrocarbons and oxygenated compounds. Reports on most of these valuable medicinal plants are new and rare in literature.

Key Words

Essential oils, hydro distillation, families, terpenoids, antimalaria, GC and GC.MS, ethno-medicine.

[P-188]

Essential oil composition of three Compositae and one Labiatae species commonly utilized as rabbit feed: an indication of preferences observed during consumption

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Abstract

We observed in our quest for active metabolites from plants that rabbits show varied attitudes and preferences when consuming *Aspillia africana* (Pers.) C. D. Adams, *Chromolaena odorata* (L.) R.M.King & H.Rob. and *Syndrella nodiflora* (L.) Gaertn. (Compositae) and *Hyptis suaveolens* (L.) Poit. (Labiatae), which are utilized as alternate rabbit feeds in Nigeria. This may be due to the chemical compositions of each plant, including differences in their levels of nutrients, palability and odor. This study is targeted at examining chemical compositions of both leaf and stem essential oils, likely responsible for the self-selection of the four plants by their consumers. Respectively *Aspillia africana* contains 7 compounds in leaf, and 9 compounds in stem essential oils; *Chromolaena odorata* have 13 in leaf and 13 in stem; *Syndrella nodiflora* consist of 6 in leaf and 12 in stem, while *Hyptis suaveolens* is made up of 15 compounds in leaf and 11 compounds in stem essential oils. The dominant compounds in leaf and stem respectively of each are: *Aspillia africana* [α -bisabolol & + nerolidol]; *Chromolaena odorata* [caryophyllene oxide & heptadecen-5-yne], *Syndrella nodiflora* [Z-13-docosenamide & + limonene] and *Hyptis suaveolens* [1,8-cineole & 9-octadecenamide]. The chemical compositions of the eight essential oils from the four plants vary also the procured % yield of the essential oils range from 0.14% to 0.43% yields.

Key Words

Aspillia africana, *Chromolaena odorata*, *Syndrella nodiflora*, *Hyptis suaveolens*, Compositae, Labiatae, essential oil, GC and GC/MS, hydrodistillation, terpenoids.

[P-189]

The application of chemometrics for the identification of antimicrobial compounds from essential oils

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Abstract

Chemometrics is an indispensable tool in elucidating patterns in complex chemical matrices. Multivariate data analysis is well suited for comparing complex spectral data, identifying patterns of co-occurrence of compounds and to assist in elucidating chemical compositional differences between samples. Correlating metabolomic data with pharmacological activities of natural products enables the researcher to identify the active fraction or compounds avoiding the laborious workflow of the reductionist approach. The aim of this study was to use chemometrics as a tool for the possible prediction of antimicrobially active compounds from essential oils based on GC-MS data alone. The composition of 160 commercial essential oils was analysed by GC-MS and their antimicrobial activity were determined with a quick, sensitive microplate MIC method against various pathogens. The GC-MS chromatograms were exported to MetAlign software for baseline correction and peak alignment. Subsequently the data was exported to Simca-P+ (12.0) for chemometric analysis. Only 4.14% ($R^2X(\text{cum}) = 0.0414$; $Q^2(\text{cum}) = 0.976$) of the phytochemical variation of samples was responsible for the difference between samples with noteworthy (MIC values ≤ 2 mg/mL) and moderate ($2 \text{ mg/mL} < \text{MIC values} \leq 6 \text{ mg/mL}$) activity against *Bacillus cereus*. An S-plot was constructed to identify putative biomarkers. Several chromatographic regions were identified that may be responsible for the noteworthy activity of the samples. Preliminary data indicates that eugenol and β -pinene present in the oils may be the compounds responsible for the antimicrobial activity against *Bacillus cereus*.

Key Words

Chemometrics, Biomarker identification, Metabolomics.

[P-190]

Volatile Oil Constituents and Antimicrobial Activity of *Tanacetum punctatum* (Desr.) Grierson Growing Wild in Two Regions in Iran

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Abstract

The genus *Tanacetum* (Family: Compositae) is represented in Iran by twenty-six species including 12 endemics. Some species of *Tanacetum* are used as medicinal plants. In this work, the aerial part of *Tanacetum punctatum* (Desr.) Grierson was collected on July 2010 in Khalkhal area (sample A) at an altitude of 1600 m near Anavis village, and sample B was collected from Ardabil area at an altitude of 2250 m in northwest Iran. The aerial part of samples were air-dried at room temperature for twelve days then subjected to hydrodistillation for 4 h, using a Clevenger-type apparatus. The analysis of oils was performed by using GC and GC/MS. Twenty-six constituents

representing 92.3% of the essential oil of sample A and twenty-two components (94.8%) of oil from sample B were identified. The oil of sample A was characterized by higher amount of camphor (21.9%), piperitenone (17.8%), α -pinene (12.2%), mentone (6.5%) and thymol (4.7%). Whereas, the main components of the oil from sample B were camphor (24.9%), pulegone (12.8%), β -cubebene (11.6%), α -pinene (7.9%), mentone (4.5%) and thymol (3.9%). The antibacterial activity of the oils from samples was assessed against two microorganisms, *Staphylococcus aureus* and *Escherichia coli*. A significant antibacterial activity was determined with the agar diffusion method. The results indicated a high activity on *Staphylococcus aureus* and *Escherichia coli*.

Key Words

Compositae, *Tanacetum punctatum*, camphor, piperitenone, α -pinene, β -cubebene

[P-191]

Composition of volatile oil and antioxidant activity of the oil and methanolic extracts of *Ferula microcolea* Boiss.

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Abstract

The essential oils of *Ferula microcolea* Boiss. collected from west of Iran during the flowering stage, were obtained by hydrodistillation and analyzed by gas chromatography (GC) and gas chromatography/mass spectrometry (GC/MS). Under the optimum distillation and analysis conditions, 22 constituents (mainly monoterpenes) were identified in *Ferula microcolea* which represented 93.6% of the oil. The main constituents were α -pinene (27.3%), β -pinene (16.4%), nonanal (8.7%), β -caryophyllene (8.5%) and thymol (6.7%). The samples were also subjected to screening for their possible antioxidant activity by using 2,2-diphenyl-1-picrylhydrazyl (DPPH) and β -carotene-linoleic acid assays. In the first case, the free radical scavenging activity of polar sub-fraction of methanol extract was superior to all other extracts ($IC_{50} = 34.3 \pm 0.3 \mu g/ml$). nonpolar sub-fraction of methanol extract exhibited stronger activity than the essential oil. In the case of the linoleic acid system, oxidation of the linoleic acid was effectively inhibited by the polar sub-fraction of methanol extract, while the oil and nonpolar sub-fraction of methanol extract were less effective.

Key Words

Ferula microcolea, antioxidant activity, essential oil.

[P-192]

Essential oils composition and antioxidant properties of three *Thymus* species

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Abstract

The essential oils of three wild-growing *Thymus* species collected from west of Iran during the flowering stage, were obtained by hydrodistillation and analyzed by gas chromatography (GC) and gas chromatography/mass spectrometry (GC/MS). Under the optimum extraction and analysis conditions, 44, 38 and 38 constituents (mainly monoterpenes compounds) were identified in *T. kotschyianus* Boiss. & Hohen, *T. eriocalyx* (Ronniger) Jalas and *T. daenensis* Celak subsp *lancifolius* (Celak) Jalas which represented 89.9%, 99.7% and 95.8% of the oils, respectively. The main constituents were thymol (16.4-42.6%), carvacrol (7.6-52.3%) and γ -terpinene (3-11.4%). Antioxidant activity was employed by two complementary test systems, namely 2,2-diphenyl-1-picrylhydrazyl (DPPH) free radical scavenging and β -carotene/linoleic acid systems. Antioxidant activity of polar sub-fraction of *T. daenensis* subsp *lancifolius* was found to be higher than those of the others in DPPH assay while nonpolar sub-fraction of *T. eriocalyx* has most antioxidant activity in β -carotene/linoleic acid test (19.1 ± 0.1 μ g/mL and $96.1 \pm 0.8\%$ inhibition rate, respectively).

Key Words

Thymus, antioxidant activity, essential oil.

[P-193]

Some Theoretical Studies on the Gas Chromatography/Mass Spectroscopy Study of an Essential Oil from an Iranian *Salix* species

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Abstract

The essential oils from *Salix aegyptiaca* L. were obtained by hydrodistillation. The oils were analyzed by capillary gas chromatography (GC), using flame ionization and capillary gas chromatography/mass spectrometry (GC/MS) detection. Forty-nine components were identified in the oil of *S. aegyptiaca* based on the most stable conformations the structural properties of *S. aegyptiaca* were calculated by using some semi-empirical quantum mechanic methods. All calculations were performed in gas phase, and the HOMO-LUMO energy levels, dipole moment, surface, volume and the charge density on all atoms for all compounds were determined in this study. It is a good agreement between the retention times the structural properties for the essential oil compounds.

Key Words

Theoretical study, Salix, GC/MS.

[P-194]

Hybrid nanomaterial for stabilizing *Rosmarinus officinalis* L. essential oils in biointerface applications

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Abstract

The aim of the present study was to develop a stabilization method for *R. officinalis* L. essential oil using a new hybrid organic/inorganic nanomaterial, in order to obtain an efficient antipathogenic strategy for biofilm related fungal infections. The essential oil microwave assisted extraction was performed in a Neo-Clevenger type apparatus and its chemical composition was settled by GC-MS analysis. The fungal strains were isolated from different clinical specimens and were identified by Vitek II automatic system. Fe₃O₄/oleic acid nanoparticles were obtained by a Massart method. High resolution transmission electron microscopy (HR-TEM) was used as a primary characterization method. The nanoparticles were used to achieve a core/shell/coated-shell based on essential oil nanosystem for covering treatment of the inert substrata represented by glass coverslips. The obtained modified surfaces were subsequently used for the in vitro study of the fungal biofilm development. The biofilm architecture was assessed by advanced microscopy techniques - confocal laser scanning microscopy (CLSM) and atomic force microscopy (AFM). The results are recommending the hybrid nanomaterial (core/shell/coated-shell) for the stabilization of *R. officinalis* essential oils, which proved a significant antibiofilm activity, highlighting the opportunity of using them for the developing of efficient antibiofilm strategies as coating modified biomaterials.

Key Words

R. officinalis essential oil, GC-MS, biointerface, antibiofilm strategy, advanced microscopy techniques.

[P-195]

Antimicrobial Effects of Aetheroleum *Pini pumilionis*, Aetheroleum *Melaleuca* and their Main Components on Airborne Microbes

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Abstract

Airborne microbes are a still underestimated cause of risk for human health. Actually, all currently used air disinfectants have disadvantages. Consequently, it seems to be worthwhile to investigate new, eco-friendly, unobjectionable substances with a potential for air-disinfection. Although the antimicrobial (antibacterial and antifungal) effects of the essential oils of *Pinus pinaster* Aiton (Pinaceae) and *Melaleuca alternifolia* Cheel (Myrtaceae) are well known, very little was known yet about the antimicrobial effects of their scents on airborne microbes. In the present study, essential tea-tree oil and the essential oil of mountain pine were tested in several concentrations

for their antimicrobial effect on airborne microbes. In addition the antimicrobial effect of the main components of these oils (terpinen-4-ol, γ -terpinene, α -terpinene, p-cymene, 1,8-cineole, terpinolene and terpineol (α , β , γ), α -2,5-dimethylstyrene, respectively) and mixtures of the main components were examined in the same way. With each of the tested essential oils, main components and mixtures thereof, a reduction of the total microbial count in the air of the testing rooms could be achieved. With the spraying of Aetheroleum *Pini pumilionis* an average reduction of total germ count of 23.4% was detected. With an average reduction of 24.5%, essential tea-tree oil was likewise effective. For the tested single components, 1,8-cineole (average reduction of total germ count of 44.2%), α -2,5-dimethylstyrene (43.8% reduction) and terpineol (35% reduction) were the most successful ones.

Key Words

Antimicrobial Effects, Aetheroleum *Pini pumilionis*, Aetheroleum *Melaleuca*, airborne microbes.

[P-196]

Survey on Chemical Analysis and Antibacterial effect of *Carum copticum* L. (Zenyan) essential oil on the growth of *Listeria monocytogenes* in Broth Medium

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Abstract

The aim of the present work was first to evaluate the antimicrobial effect of *Carum copticum* L. (Zenyan in Persian) essential oil (extracted from an aromatic, grassy plant grown in Iran, being popular as an anti-vomiting, diuretic, carminative agent) on *Listeria monocytogenes*, a prevalent pathogen, and secondly, to determine the chemical composition of the essential oil by GC/MS. The antimicrobial effect of *Carum copticum* L. was evaluated using disk diffusion method as a preliminary step, and microdilution method which is based on optical density measurements. The half-fold dilutions were made from stock solution of the essential oil by using Tryptone Soya Broth in order to determine minimum inhibitory concentration (MIC) for the bacterium using the Bioscreen C device. The applied technique uses 100 well microtitre plates and collects turbidometric growth data. Results showed that *Carum copticum* essential oil exhibited significant antibacterial effect on *Listeria monocytogenes* in the concentration of 686 ppm. According to the results obtained by GC/MS analysis around twenty compounds were identified in the essential oil, the most of which included thymol, p-cymene, and γ -terpinene. In conclusion, edible plants can be a potential source for inhibitory substances for some foodborne pathogens. Owing to the strong antibacterial activity of *Carum copticum*, it can be used as an effective alternative for food preservation. Considering the low amount of the essential oil used for growth inhibition, probably it will not exhibit undesirable effects on organoleptic properties of foods.

Key Words

Antimicrobial, *Carum copticum*, essential oil, *Listeria monocytogenes*.

[P-197]

Effect of Cattle and Liquid Manures on Essential Oil and Antioxidant Activities of Celery (*Apium graveolens* L.) Fruits

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Abstract

This study was carried out to evaluate the essential oil (EO) of celery (*Apium graveolens* L.) and its antioxidant activities under organic fertilizer conditions [cattle manure (CM), liquid manure (LM) and their interactions]. Generally, treatments with organic fertilizers had a positive effect on EO content (%) and yield (mL plant⁻¹). Limonene was the major component of the EO extracted from celery fruits (30.4–37.5%). Limonene increased at all levels of CM + LM compared with CM treatments. Celery EOs obtained from CM + LM treatments had a positive effect on free radical scavenging activities (FRSA) compared with those obtained from CM treatments.

Key Words

Cattle manure, celery, *Apium graveolens*, essential oil, free radical scavenging activities, liquid manure.

[P-198]

Influences of silicate dissolving bacteria with feldspar on growth and essential oil of Rue (*Ruta graveolens* L.)

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Abstract

The experiment was consummated at the Experimental Farm, National Research Centre (NRC), Egypt during 2004/2005 season to evaluate the effect of silicate dissolving bacteria (SDB) + feldspar [as a natural source of important elements i.e. potassium (K)] by different levels, 0.0, 25, 50 and 100 g L⁻¹ on rue (*Ruta graveolens* L.) plant. Adding SDB + feldspar improved fresh and dry weights (FW and DW) of herb and essential oil (EO) content (% or g plant⁻¹). On the other hand, the main constituents of essential (Undecan-2-ol and Nonan-2-one) were increased but the relative level for various constituents increased or decreased under SDB + feldspar treatments.

Key Words

Rue, *Ruta graveolens*, silicate-dissolving bacteria, potassium, feldspar, fresh weight, dry weight, essential oil.

[P-199]

Effect of lemon essential oil on the developmental stages of *Trialeurodes vaporariorum* West

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Abstract

Trialeurodes vaporariorum West (Homoptera: Aleyrodidae), is a destructive pest of many ornamental and greenhouse crops throughout the world. In this research, effect of essential oil derived from Citrus limon peel [*Citrus aurantifolia* (Christm.) Swingle] on mortality of eggs, first-instar nymphs and on adult oviposition, of *T. vaporariorum* (Westwood) was determined under laboratory conditions. Five concentrations of essential oil, 0.002, 0.004, 0.008, 0.016 and 0.032 (µl/mL), were applied in fumigant toxicity experiments. Greater mortality was observed with increasing dose of essential oil. First-instar nymphs were more sensitive to essential oil treatments compared with eggs and adults that reduced the survival rate of *T. vaporariorum* by 58%, 70% and 56% after treatment of eggs, nymphs and adult, respectively. Based on this study, essential oil derived from *C. aurantiifolia* could be used as effective and environmentally sustainable bioinsecticides for the control of *T. vaporariorum*.

Key Words

Trialeurodes vaporariorum, lemon essential oil, fumigation.

[P-200]

A Study on Antioxidant Activity, Essential Oil and Polyphenol Contents of *Hypericum perforatum* L. Growing Wild From Different Altitudes of Ayder Plateau in Rize

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Abstract

This study was carried out to determine the changes in the essential oil content and antioxidant activity of *Hypericum perforatum* L. from three different altitudes (100, 275, 600 m) of Ayder Plateau, Rize, Turkey. Materials for extraction were collected during the full flowering periods in August 2010. The aerial parts of *H. perforatum* were dried in shadow at room temperature and for essential oil analyses 30 g of powdered leaves and flowers were hydrodistilled with a Clevenger type apparatus for 4 h. Also, the extraction process for powdered leaves and

flowers was performed according to Auddy protocol and the determination of total polyphenol compounds in the aqueous extracts but not on essential oil of these samples were spectrophotometrically determined at 700 nm by Folin-Ciocalteu's colorimetric method using Folin-Ciocalteu reagent and expressed as catechin (mM) equivalents. Antiradical activity of the extracts of *Hypericum* parts was measured by utilizing DPPH method and expressed as EC50 (mg/mL), the concentration of antioxidant, required for 50% scavenging of DPPH radical in the specified time period. In the research, mean of essential oil content was varied between 0.2% and 0.35%. For the total polyphenolic contents, the values of the different altitudes (100, 275 and 600 m) were recorded 489 ± 8 μ M, 510 ± 8 μ M and 524 ± 10 μ M catechin equivalent, respectively. And there were no statistically differences between the antiradical activities of the extracts of the different altitudes (100, 275, 600 m) that was recorded as 18 ± 3 μ M, 18 ± 3 μ M, and 20 ± 4 μ M catechin equivalent, respectively.

Key Words

Antioxidant activity, Ayder Plateau, essential oil, *Hypericum perforatum* L., polyphenol content.

[P-201]

Antioxidant properties of *Mentha pulegium* essential oil

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Abstract

Mentha pulegium L., commonly known as 'naena' in Persian, belongs to the genus *Mentha* in the family Labiatae (Lamiaceae). The genus *Mentha* includes 25-30 species such as spearmint, peppermint, curled mint wild mint, and corn mint. It grows extensively in wet places, riverbank, and marshy areas. In folk medicine, *M. pulegium* is known for its pungent taste having post digestive effect and hot potency. Various species of *Mentha* have many constituents in common and all have been held in high dietary medical repute. In Persian traditional home recipes, dried powdered leaves is added to yoghurt to make a pleasant drink in hot summer days. Its essential oil is used to flavor toothpaste, confectionery, and chewing gum and also to perfume soaps, even in modern industry. It has been shown that spearmint has antifungal, antiviral, antimicrobial, insecticide, antioxidant, antiamoebic, antihemolytic, allergenic, CNS depressant, antihelmintic and antiancyclostomiasis activity. In the present study, we have distilled the essential oils of three mint species collected from different locations in northern Iran. The antibacterial activity of each sample was then compared to the common strong antibiotic, ampiciline. The results showed that Gram-positive bacteria were in general more sensitive to the essential oils than Gram-negative bacteria. *Streptococcus mutans* was the most sensitive strain, while *Pseudomonas aeruginosa* was the most resistant. Interestingly, free radical-scavenging activities of the essential oil were comparable to the butylated hydroxyl toluene (BHT) and lower than the extract.

Key Words

Oil, mint, antibacterial, *Mentha pulegium*.

[P-202]

Effect of Nitrogen Application on Growth and Essential Oil Production of Peppermint (*Mentha piperita* L.)

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Abstract

Peppermint (*Mentha piperita* L.) plants were grown in Investigative Field of Ferdowsi University of Mashhad in Iran (59°N latitude), and evaluated for their response to nitrogen rates. The form of applied nitrogen was urea in solid form in the soil. Plants were measured for total dry weight, final plant height, nodes number, inter-node length, total leaves number, and essential oil content in flowering stage. Rates of nitrogen applied were 0 (control), 75, 150, and 225 Kg nitrogen/ha. Each treatment had three replicates. It was established that plant dry matter, plant height, internode length, leaves number, node number, and essential oil content were increased with increasing nitrogen rates (with the exception at 225 Kg nitrogen/ha). With the increased fertilizer rates, the essential oil yield of the treated plants increased compared to the control. The highest and lowest essential oil content were shown in the plants that received 150 and 0 Kg nitrogen/ha, respectively.

Key Words

Mentha piperita, nitrogen Fertilizer, essential oil, growth.

[P-203]

Antifungal Activity of some Plant Extracts and Essential Oils for Citrus Fungal Diseases Control

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Abstract

The antifungal activity of essential oils and extracts were tested against *Penicillium* sp. and *Aspergillus niger*. The essential oils of fennel (*Foeniculum vulgare* Mill.) and caraway (*Carum carvi* L.), and also plant extracts of fennel, caraway and sage (*Salvia officinalis* L.) were used, too. The essential oil treatments were performed at two levels (0.25 and 0.5%) and extract treatments applied at 4 levels (25%, 50%, 75% and 100%). All treatments were showed different activities in control of fungi growth. A significant decrease in mycelial growth was obtained with the addition of 0.25% and 0.5% in each of two essential oils in PDA medium. *Carum carvi* essential oil inhibited mycelial growth of two fungi tested at 0.5% level. Among of plant extracts, *Carum carvi* was had highest inhibition with highest concentration, which was 100% (v/v) level. The results indicated that the *Carum carvi* extract inhibited growth of *Aspergillus niger* (53.68%) and *Penicillium* sp. (88.56%) compared to control.

Key Words

Antifungal activity, essential oil, plant extract, *Foeniculum vulgare*, *Carum carvi*, *Salvia officinalis*, *Aspergillus niger*, *Penicillium* sp.

[P-204]

The effects of the essential oils of several medicinal plants on *Botrytis cinerea* and *Rhizopus stolonifer* rot fungus post-harvest control under *in vivo* conditions

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Abstract

Antifungal activity of *Foeniculum vulgare* Mill., *Cuminum cyminum* L., *Thymus vulgaris* L. and *Mentha piperita* L. essential oils against *Botrytis cinerea* and *Rhizopus stolonifer* of strawberry were investigated under lab conditions. Fennel essential oil perfectly inhibited growth of *Rhizopus* at concentration higher than 600 µL/L in PDA medium. Cumin essential oil perfectly inhibited growth of *Botrytis* at 400 µL/L concentration in PDA medium. Mean separate test (Duncan, $p < 0.01$) indicated essential oils of fennel (76.29%) and cumin (74.85%) have the higher inhibition percentage on growth of *Rhizopus* and *Botrytis* fungus, respectively. Percentage of fungus spur germination was the lowest in medium fennel and cumin essential oils and was the highest in *Thymus*. As for GC and GC/MS analyses, the main component in cumin oil was cumin aldehyde and in fennel oil was anethole. Both these components have strong antifungal effects.

Key Words

Botrytis cinerea, *Rhizopus stolonifer*, cumin, fennel, essential oil, post-harvest.

[P-205]

The essential oil composition of *Cymbocarpum marginatum* and *C. erythraeum* (Apiaceae) from Iran

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Abstract

The chemical composition of the essential oils from the aerial parts of *Cymbocarpum marginatum* Boiss. (*Kalakia stenocarpa* (Bornm. & Gauba) Alava) and *C. erythraeum* (DC.) Boiss. (Apiaceae), endemic of Iran, was analyzed by GC/MS. The oil yields obtained by hydrodistillation were 0.3% and 0.4% w/w, respectively. Methods of identification were comparison of relative retention indices and mass spectra with standards. The aerial parts of *C. marginatum* were collected around Karaj, in central Iran. Fifty-one components, accounting for 92.8% of the oil, were identified. The main class of the compounds was found to be aliphatic aldehydes (71.1%), and the major constituents were 3-dodecen-1-al (37.1%), decanal (18.0%) and 2-decenal (6.7%). 2-Methylene cyclopentanol (7.7%) was a main components in the oil. The aerial parts of *C. erythraeum* were collected from around Tabriz (East Azerbaijan Province, northwestern Iran). In *C. erythraeum*, 33 components were identified.

The major constituents were 2-dodecen-1-al (29.4%), 2-tridecanal (13.7%), nonane (9.1%), α -pinene (8.6%), 2-decenal (5.2%), and 1,8-cineole (3.8%). As far as we know, the essential oil composition of the herbs has not been described previously.

Key Words

Essential oil composition, aerial parts, *Cymbocarpum marginatum*, *C. erythraeum*, Iran, Apiaceae, GC/MS, hydrodistillation, aliphatic aldehydes.

[P-206]

The pharmacologic evaluation of Thymoquinone of *Nigella sativa* oil in the treatment of diseases

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Abstract

Nigella sativa L. is an annual plant which belongs to southwestern Asia. It grows up to 20-30 cm, with divided and linear (not thread-like) leaves. Its flowers are delicate, and usually pale blue and white with 5-10 petals. The fruit is a large and inflated capsule composed of 3-7 follicles and each contains numerous seeds. The seed is used as a spice. *Nigella sativa* has been used for medicinal purposes for centuries as a herb and sometimes as an oil in Asia, Middle East, and Africa in order to treat ailments including asthma, bronchitis, rheumatism and related inflammatory diseases, increasing milk production in nursing mothers, promoting digestion and fighting parasitic infections, health, stomach and intestinal health, kidney and liver functions, circulatory and immune system support, as an analgesic, anti-inflammatory, anti-allergic, antioxidant, anticancer, antiviral and for general well-being. Its oil has been used to treat skin conditions such as eczema and boils and cold symptoms. Medicinal plants widely are used in traditional medicine in order to avoid major effects caused by chemical drugs. The black seeds of *N. sativa* contain non-volatile (30%) and volatile (45-40%) oils. And thymoquinone is the main component of this oil. In different countries the black seed is used in traditional medicine in order to treat diseases such as hypertension, decrease pressure blood, nutraceutical, fat diseases, reduction asthma and diarrhea, rheumatism and infectious diseases against nematodes and cestodes (tape worms), anti-malarial, and tumor and cancer disinfectant. This article reviews the pharmacology of the oil of TQ of Ns in treating diseases.

Key Words

Nigella sativa, thymoquinone, biological activity.

[P-207]

Essential oil constituents of *Blepharocalyx salicifolius* (Kunt) O. Berg from Brazilian savannah

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Abstract

Maria Preta [*Blepharocalyx salicifolius* (Kunt) O. Berg - Myrtaceae] is a 20 m tree occurring in many states of Brazil. Leaves are used as medicinal to treat diarrhea, bronchitis, and arthritis. *B. salicifolius* grows in a wide variety of habitats along its geographical area, presenting extreme morphological variations, also reflected in its essential oil composition. As part of a research effort to study the aromatic flora from Cerrado (a savannah-like area in central Brazil), the objective of this work was to evaluate the yield and chemical composition of the essential oil from *B. salicifolius*. Leaf samples were collected in April 2010, at the University of Brasília Água Limpa farm, Distrito Federal, Brazil. The essential oil was extracted by steam distillation using a Linax Co. model D1 equipment and analyzed by GC and GC/MS. Both mass spectra and linear retention indices on DB-5 were used for compound identification. Forty-two volatile oil constituents were identified in *B. salicifolius* leaves, representing 90.5% of the total composition. The major detected constituents were p-cymene (25.9%), γ -terpinene (12.5%); caryophyllene oxide (9.6%) and α -pinene (9.0%). To the best of our knowledge, this is the first chemical evaluation of this species collected in the Cerrado area, and its essential oil composition differed from data of plants from other areas.

Key Words

Cerrado, essential oil, *Blepharocalyx salicifolius*.

[P-208]

Chemical composition and antibacterial activity of the essential oil of flowering aerial parts of *Achillea millefolium* L. of Northern Iran

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Abstract

Achillea millefolium L. (Asteraceae) has been used by the rural healers of northern Iran in traditional medicine as a sedative, antiseptic and anti-inflammatory. With the aim to investigate essential oil composition and antibacterial activity, essential oil was obtained from flowers by hydro-distillation (Clevenger-type apparatus), aqueous and ethanolic extracts were obtained by percolation, and their antibacterial activities were measured by disc diffusion and well methods, against 6 Gram-positive and Gram-negative bacteria. Results showed that terpinolene (81%), borneol (4.2%), β -pinene (3.5%), chamazulene (2.9%), p-mentha-3,8-diene (2.4%) and *cis*-thujone (1.2%) were the major constituents. Ethanolic extract showed strong effect against the tested bacteria but aqueous extract had moderate and weak effect. Gram-positive bacteria such *Staphylococcus aureus* and *Bacillus cereus* were the most sensitive bacteria, whereas *Salmonella typhimurium* and *Escherichia coli* were the resistant bacteria especially in well method ($P < 0.05$). According to previous studies and our results, probably showing the terpinolene, borneol, β -pinene and chamazulene were the most effective constituents of *A. millefolium* essential oil. In conclusion *A. millefolium* L. was found to be a good candidate as antibacterial plant for future *in vivo* and clinical research.

Key Words

Essential oil, *Achillea millefolium*, chemical composition, antibacterial, Iran.

[P-209]

Chemical composition and antimicrobial activity of essential oil obtained from uvaia (*Eugenia pyriformis* Cambess)

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Abstract

The essential oils extracted from the peel, seed and full fruit of the uvaia (*Eugenia pyriformis* Cambess, Myrtaceae) were analyzed for their physicochemical composition and evaluated for their antimicrobial activity. Higher yield of essential oil was obtained from the peel (0.23%), followed by the whole fruit. The lowest yield was found in the oil extracted from the seed. The Microbiological results obtained from the study on strains of *Escherichia coli* ATCC25922, *Staphylococcus aureus* ATCC 6538, *Pseudomonas aeruginosa* ATCC 27853 and *Enterococcus faecalis* ATCC99212 proved their bacteriostatic performance. The essential oil was active against Gram-negative bacteria *P. aeruginosa* (MIC=267 µg/mL) compared to Fosfomycin (MIC=128 µg/mL). This result is quite significant since, in general, the essential oils are more active against Gram-positive bacteria. The volatile organic compounds present in essential oil were identified by using the data obtained from their GC/MS analysis and LRI. Twenty-five compounds were identified, the principal being monoterpenes containing limonene (11.0%), which was the major compound, followed by α -terpineol (6.1%) and terpinen-4-ol (0.9%), and caryophyllene oxide (18.7%).

Key Words

Pseudomonas aeruginosa, caryophyllene oxide, Myrtaceae, volatile, compound.

[P-210]

Antibacterial activities of *Black pepper (Piper nigrum L.)*, and *Cinnamon (Cinnamomum zeylanicum Blume)* on *Escherichia coli* ATCC 8739 in laboratory medium

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Abstract

The present study was designated to evaluate the antimicrobial effects of two plant extracts including *Black pepper (Piper nigrum L.)*, and *Cinnamon (Cinnamomum zeylanicum Blume)* on *Escherichia coli*, a prevalent pathogen. The antimicrobial activity of the extracts were evaluated using disk diffusion method as a preliminary step which was followed by microdilution method. The mentioned extracts were introduced into TSB Broth at ten concentrations from 50% (v/v) to 0.097 % (v/v) in order to determine minimum inhibitory concentration (MIC), using the Bioscreen C device, which is based on optical density measurements. The applied technique uses 100 well microtitre plates and collects turbidometric growth data. To produce inhibition profiles, the concentrations were tested for the bacterium. Results revealed that both extracts indicated inhibitory effects, showing MIC of 3.125% (v/v) and 6.25% (v/v) for Black pepper (*Piper nigrum*), and, Cinnamon (*Cinnamomum zeylanicum*) respectively. These findings support the idea that Black pepper and Cinnamon extracts, investigated in this survey could be suitable for use as a natural active ingredient in natural and low-cost drugs against *Escherichia coli*.

Keywords: Antimicrobial, Cinnamon, Black pepper, extracts, *Escherichia coli*

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[P-211]

Inhibitory effects of *Capsicum annuum* L., *Curcuma longa* L., and *Piper nigrum* L. plant extracts on the growth of *Listeria monocytogenes* ATCC 19115 in laboratory medium

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Abstract

The present study aimed at evaluating the antimicrobial effects of Red pepper (*Capsicum annuum* L.), Turmeric (*Curcuma longa* L.) and Black pepper (*Piper nigrum* L.) on *Listeria monocytogenes* using disk diffusion method as a preliminary step and microdilution method. The extracts were introduced into TSB Broth at 9 concentrations from 50% (v/v) to 0.195 % (v/v) in order to determine minimum inhibitory concentration (MIC) for *Listeria monocytogenes* using the Bioscreen C device, which is based on optical density measurements. The applied technique uses 100 well microtitre plates and collects turbidometric growth data. To produce inhibition profiles, the concentrations were tested for the bacterium. Results indicated that using Red pepper showed great antibacterial activity and completely prevented the growth of *Listeria monocytogenes*. Turmeric extract had moderate antimicrobial effect, showing MIC of 6.25 % (v/v), but Black pepper extract could not prevent the growth of *Listeria monocytogenes*. In short, edible plants can be a potential source for inhibitory substances for some foodborne pathogens. Among the extracts studied Red pepper and Turmeric were effective on *Listeria monocytogenes* so they can be used as chemical antibacterial alternatives in foods.

Keywords: Antimicrobial, Red pepper, Black pepper, extract, Turmeric, *Listeria monocytogenes*

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[P-212]

Local knowledge and potential revenue of oil products from native trees in Mali

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Abstract

This research is a PhD project and aims to explore the socio-cultural knowledge of communities for improving the production of vegetable oil from local forest tree species and enhance the nutritional and economic potential of these species. The research will be conducted in two agro-ecological zones in Mali: the Sudanian zone (Sikasso) and the Sahel region (Mopti). The research is based on the main assumption that there is a potential of oil tree species and local knowledge untapped that can improve food security and living conditions of populations. A quantitative ethno-botanical approach is adopted to identify local knowledge on oils forest tree species. The economic potential will be assessed through a combination of participatory focus groups, structured and semi-structured socio-economic interviews with all the actors involved in the value chain: local producers, rural assemblers, wholesalers, distributors, transformers, retailers and consumers. A SWOT-analysis will identify strengths, weaknesses, opportunities and threats related to local and regional oil products market channels, upon which recommendations will be formulated.

Keywords: local knowledge, native oil tree, ethnobotany, value chain, revenue, Mali

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[P-213]

Attractiveness to three *Bactrocera* species (Diptera: Tephritidae) of 30 essential oils in New Caledonia

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Abstract

Tephritid Fruit flies of the genus *Bactrocera* are ones of the worst pests of fruit and vegetable crops in the world. The dramatic reduction of the use of chemical insecticides pushes to consider some olfactive stimuli acting as attractants or repellants to be used against these significant pests. These substances being able to be used in IPM running control systems. In New Caledonia, a French Over-Seas Country in the South Pacific, three economic fruit fly species occur: the two endemic *Bactrocera curvipennis* (Froggatt) and *B. psidii* (Froggatt) and the invasive *B. tryoni* (Froggatt) also called the Queensland fruit fly (Mille 2008, Mille & Hancock 2009). These three species are currently reared in the laboratory. Thirty essential oils commercially available were tested regarding their activity to unmated and mated fruit fly females (Robacker 2007, Mille 2010). As shown in the following table, within the 30 essential oils tested, only 11 showed some attractiveness and repulsiveness to tested fruit flies.

Keywords: *Bactrocera*, odours, essential oils, attractant, repellent, trap, pest control

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[P-214]

Analysis of essential oils of aerial parts of *Ducrosia anethifolia* (DC.) Boiss. and *Ducrosia flabellifolia* Boiss. from Iran

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Abstract

The essential oils of *D. anethifolia* Boiss. and *D. flabellifolia* Boiss. have been investigated by capillary gas chromatography mass spectrometry (GC/MS) detection. Thirty two components were identified in the oil of *D. anethifolia* with dodecanal (28.8%), n-decanal (21.1%), 2E-Tridecen-1-al (15.78%) and 2E-dodecenal (13.4%) as the main constituents. Thirty three components were identified in the oil of *D. flabellifolia* with n-decanal (32.8%), dodecanal (32.6%), n-decanol (4.3%) and 2E-Tridecen-1-al (3.3%), as the main constituents.

Keywords: *Ducrosia anethifolia*, *Ducrosia flabellifolia*, Apiaceae, essential oil composition

References:

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[P-215]

Composition and cytotoxic activities of essential oil from *Libanotis transcaucasica* Schischk. growing in Iran

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Abstract

The chemical compositions of the water-distilled essential oil of *Libanotis transcaucasica* Schischk. was determined by GC and GC-MS. Eighty-four compounds were identified by comparison of retention indices and mass spectra with published data. The sesquiterpene hydrocarbon, germacrene B (20.2%) was the main component identified in this oil. Furthermore, isospathulenol (11.0%), germacrene-D (9.2%) and kessane (5.5%) were the compounds occurring in abundance. In addition, cytotoxic activity assessed on 3 human cancer cell lines (HeLa, LS180 and Raji). The volatile oils displayed moderate cytotoxic activity in all human cancer cell lines. The highest and the lowest effects were observed on Raji and LS180 cell lines.

Keywords: *Libanotis transcaucasica*, Essential oil composition, cytotoxic activity

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Constituents of the essential oil of two endemic *Hypericum* species (*H. asperulum* Jaub. & Spach and *H. dogonbadanicum* Assadi) from Iran

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Abstract

In this research, the essential oil content and composition of two species of *Hypericum* were investigated. Essential oils from aerial parts of *Hypericum asperulum* Jaub. & Spach and *Hypericum dogonbadanicum* Assadi were obtained by hydrodistillation. *Hypericum asperulum*, has not been subjected to any previous phytochemical analysis. The oils were analysed by capillary gas chromatography, using flame ionization and mass spectrometric detection. Fifty-two components were identified in the oil of *Hypericum asperulum*, with β -caryophyllene (29.1%), spathulenol (17.3%), (Z)- β -ocimene (5.4%) and α -pinene (4.6%) as the main constituents. Seventy-four compounds were identified in the oil of *Hypericum dogonbadanicum*, with α -Pinene (12.8%), Limonene (8.2%), β -Pinene (4.7%), α -Terpineol (4.2%), Borneol (4%) and Camphene (3.9%) as the main components.

Two first major compounds of *H. asperulum* were β -caryophyllene (29.1%) and spathulenol (17.3%). *Hypericum* species can be divided in two groups, based on the monoterpene and sesquiterpene content with emphasis on β -caryophyllene and α -pinene as the major and characteristic compounds. In the light of this conclusion and despite the morphological characters, *H. dogonbadanicum* could be placed in the α -pinene group, and *H. asperulum* in the β -caryophyllene group.

Keywords: *Hypericum asperulum*, *Hypericum dogonbadanicum*, Essential oil composition

References:

Adams RP (2001) Identification of essential oil components by gas chromatography/quadrupole mass spectroscopy. Illinois: Allured Publishing Corporation.

[P-217]

Comparison of essential oil ratio in some cultivated medicinal plants with wild species

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Abstract

Medicinal plants are one of the most important plant groups in nature which have the various medical properties. As we know, effect of this plants is related to the compositions and their percentage in herb and growth location is one of the important factor in the production and value of essential oils. This study was conducted to compare essential oil value of two groups of plant. The first group contains *Achillea millefolium* L., *Mentha longifolia* Huds. and *Ziziphora tenuior* L. collected from Ahar region before flowering stage as wild species and the second group includes *Apium graveolens* L., *Mentha longifolia* and *Thymus persicus* (Ronniger ex Rech.f.) Jals collected from the cultivated fields of around Tabriz city. The essential oil ratio of plants were obtained at the laboratory of Agricultural Faculty of Islamic Azad University, Shabestar branch during 2010. The specimens were dried in cool shadow and in the temperature of the lab room. The essential oil content was determined from aerial parts of plants and using a Clevenger-type apparatus and samples were distilled by water. The results showed that, mean of essential oil ratio in wild species were in *A. millefolium* (0.8%), *M. longifolia* (1.1%) and *Z. tenuior* (1.5%) and in cultivated species were *A. graveolens* (0.33%), *M. longifolia* (1.4%) and *T. persicus* (0.8%).

Keywords: Cultivated species, essential oil ratio, medicinal plants, wild species

[P-218]

Evaluation of essential oil and heavy metals percentage in some domestic medicinal plants with wild species

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Abstract

This study was investigated to compare essential oil ratio of domestic medicinal plants and wild species in two separated groups. The first group contains *Achillea millefolium* L., *Mentha longifolia* Huds. and *Ziziphora tenuior* L. collected from Ahar region (Gazanghah) before flowering stage as wild species and the second group includes *Mentha piperita* L., *Mentha longifolia* collected from the cultivated fields of around Tabriz city. The essential oil ratio of plants were obtained at the laboratory of Agricultural Faculty of Islamic Azad University, Shabestar branch during 2011. The specimens were dried in cool shadow and in the temperature of the lab room. The essential oil content was determined from aerial parts of plants and using a Clevenger-type apparatus and samples were distilled by water. The results showed that, mean of essential oil ratio in wild species were in *A. millefolium* (0.9%), *M. longifolia* (1.2%) and *Z. tenuior* (0.87%) and in cultivated species were *M. piperita* (1.7%), *M. longifolia* (1.2%).

Keywords: *Achillea millefolium*, *Mentha longifolia*, *Ziziphora tenuior*, essential oil ratio, medicinal plants, wild species

[P-219]

New volatile compounds of wild *Allium* species from Middle Asia

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Abstract

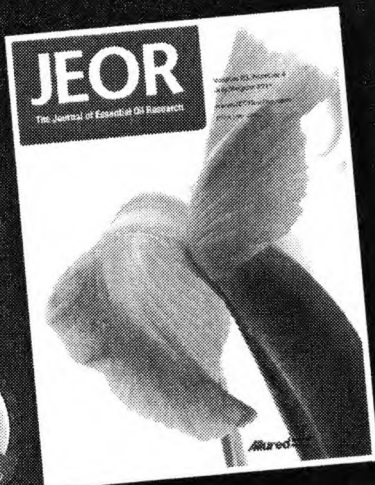
The mountainous areas of Middle and Southwest Asia are a rich source for wild growing *Allium* plants. For instance, *Allium tripedale* TRAUTV. belonging to the subgenus *Nectaroscordum* is naturally growing in the northwest of Iran and neighbouring countries. Leaves have a very strong and hot taste and are widely used by the local population as spicy vegetable. As a major constituent of the bulb, (+)-S-(1-butenyl)-L-cysteine sulfoxide ('homoisoalliin' **1**) could be identified. As volatile compounds, di-(1-butenyl)-disulphide **2** and the cepaene-like compounds di-(1-S-sulphoxymethyl-butyl)-disulphide **3**, 1-S-sulphoxymethyl-butyl-1'-S-sulphoxy-1-butenyl-butyl-disulphide **4** and 1-S-sulphoxymethyl-butyl-1'-S-sulphoxybutyl-butyl-disulphide **5** could be identified by various MS experiments [1]. Primary products resulting from the alliinase reaction of homoisoalliin seem to be highly unstable and were rapidly converted to the volatile compounds listed above. A further rather interesting species of this area is *Allium stipitatum* REGEL. Pyridinyl containing sulfoxides could be identified for the first time in the genus *Allium* [2]. It can be assumed that these compounds were converted into pyridinyl-*N*-oxides inside the plant. Corresponding *N*-oxides were also monitored after alliinase reaction giving the very typical odour of this species. Beside pyridinyl derivatives, the cysteine sulfoxide marasmin could be observed also yielding volatile sulphur compounds after alliinase reaction with at least three sulphur atoms.

References: J. Kusterer and M. Keusgen, *Cysteine sulfoxides and volatile sulfur compounds from Allium tripedale*. JAFC **58** (2), 1129 (2010). J. Kusterer, A. Vogt, and M. Keusgen, *Isolation and identification of a new cysteine sulfoxide and volatile sulfur compounds from Allium subgenus Melanocrommium*, JAFC **58**(1), 520 (2010).

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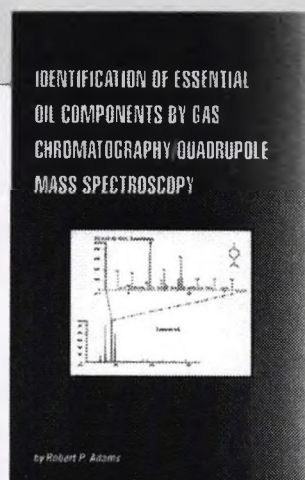
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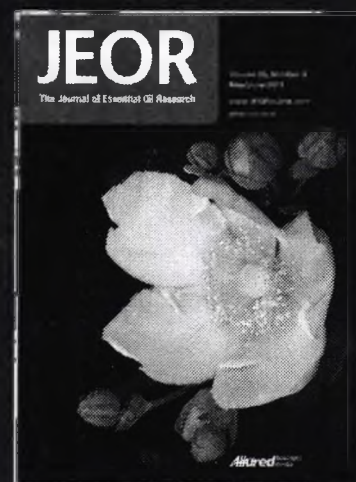
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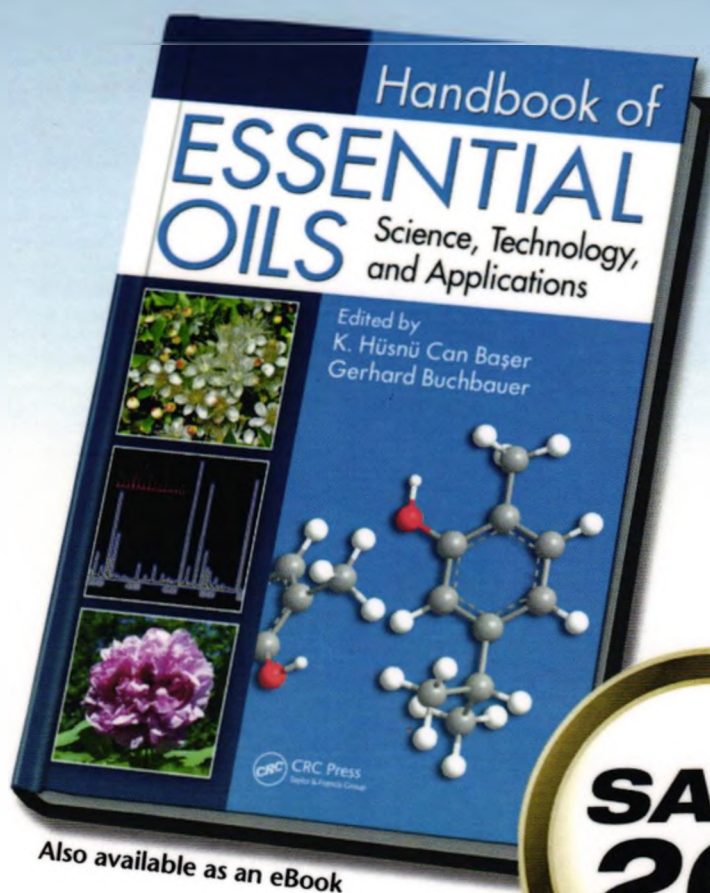
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