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significant inhibition in parasites growth. IC<sub>50</sub> values of 8.0; 17.0; 54.6 ng/ml indicated that *Callistemon rigidus, Citrus aurantifolia* and *Citrus sinensis* followed the same order of activity as shown in the *in-vivo* study. However, the oils ability to prolong the survival of highly parasitemic mice was not the same in all the animals. *Callistemon rigidus, Citrus aurantifolia* and *Citrus sinensis* prolonged the survival time of animals between 18.0 and 29.6 days, 16.2 and 20.2 days and 3.8 and 8.6 days respectively, compared to the control group that survived till 15.6 days. Temperature of the animals monitored showed that the three oils lowered the temperature of treated infected animals as compared to the control. Our results suggest that *Callistemon rigidus*, and *Citrus aurantifolia* are promising new antimalarial agents.

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Synergistic antibacterial activity of Bunium persicum and Cuminum cyminum essential oils Oroojalian F<sup>1</sup>, Kasra- Kermanshahi R<sup>2</sup>, Azizi M<sup>3</sup> <sup>1</sup>Microbiology section, Biology Department, Science Faculty of Isfahan University, Isfahan, Iran; <sup>2</sup>Microbiology section, Biology Department, Science Faculty of Azzahra University, Tehran, Iran; <sup>3</sup>Department of Horticulture, College of Agriculture, Ferdowsi University, Mashhad, Iran

Phytomedicines derived from plants as single or poly herbal preparations have shown great promise in the treatment of infectious diseases. Many studies have been carried out to date in order to extract essential oils (EOs) for screening their antimicrobial activity. However, there has not been sufficient research on the combinations of these products for their antimicrobial activity. In this study, the EOs of Bunium persicum (Kerman or black Zireh, i.e. balck cumin) and Cuminum cyminum (cumin) were obtained by hydrodistillation of the plants. The EOs were subsequently analysed by gas chromatography (GC) and gas chromatography-mass spectrometry (GC/MS). The main constituents of Bunium persicum were  $\gamma$ -terpinene (44.2%), cuminaldehyde (16.9%),  $\gamma$ -terpinen-7-al (10.5%) and p-Cymene (8%). Furthermore, the main constituents of Cuminum cyminum were found to be Cuminal (36.31%), cuminic alcohol (16.92%), γ-terpinene (11.14%), safranal (10.87%), p-cymene (9.85%) and  $\beta$ -pinene (7.75%). EOs of the plants were evaluated for their individual and combined antibacterial activities against several pathogenic bacteria, namely Staphylococcus aureus, Bacillus cereus, Escherichia coli O157H7, Salmonella enteritidis and Pseudomonas aeruginosa. Results revealed promising antibacterial activities against most pathogens using broth microdilution method. Maximum activity of the tested essential oils were observed to be against Staphylococcus aureus and Bacillus cereus and their Minimum Inhibitory Concentrations (MICs) were determined to be 31.2 and 125 µg/ml respectively. Combinations of the essential oils showed an additive antibacterial effect against most tested pathogens especially Pseudomonas aeruginosa.

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Chemical composition and acetylcholinesterase inhibition of volatile oils from *Marlierea racemosa* (Vell.) Kiaersk. (Myrtaceae) collected in two different areas of the Brazilian Atlantic Rain Forest

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Recent studies demonstrate the ability of terpenoids, found in several volatile oils, in inhibiting the acetylcholinesterase activity [1]. Myrtaceae species are known as producers of volatile oils and they have been investigated for innumerous biological activities but not for their potential as acetylcholinesterase activity inhibitors. Thus, the aim of the present work was to analyze the volatile oil composition in specimens of *Marlierea racemosa* (Vell.) Kiaersk. growing in two areas of the Atlantic Rain Forest (Cananéia and Caraguatatuba, SP, Brazil) and their antiacetylcholinesterase activity. Volatile oils were extracted from dried leaves by hydrodistillation for 4h. The component identification was performed by GC/MS. Acetylcholinesterase activity was measured through colorimetric analysis [2]. The major constituent in both groups was spathulenol (25% in Cananéia and 32% in Caraguatatuba). However, volatile oils of the Cananéia plants also presented monoterpenes

(41.2%), while in the Carguatatuba plants these compounds were detected in traces. The oils from the plants collected in Cananéia were able to inhibit the acetylcholinesterase activity up to 75% whereas for the oils from the Caraguatatuba group the maximal inhibition achieved was 35%. These results suggested that the monoterpenes are more effective in the inhibition of acetylcholinesterase activity than sesquiterpenes as these compounds are present in higher amounts in the *Marlierea racemosa* (Vell.) Kiaersk. plants collected in Cananéia. *Acknowledgements: FAPESP, Capes.* References: 1. Miyazawa, M., Yamafuji, C. (2005) J. Agric. Food Chem. 53:1765 – 1768. 2. Rhee, I.K. et al. (2001) J. Chromat. 915:217 – 223.

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Analysis of the volatile constituents of Cornus mas (Cornaceae) fruits
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There have been no previous references about volatile compounds in Cornus mas fruits. The aim of this study is important since Cornus mas fruits are widely used for the preparation of beverages, liqueurs and distillates. 1 kg of fruits was subjected to steam distillation according to the procedure suggested by the Third European Pharmacopoeia. The attribution of the volatiles was  $1\% \ v/w \ (0.001 \ ml)$  and the fraction was collected in pentane. The analysis was carried out in a Shimadzu GC 2010 gas chromatographer. Split type sample introduction, column HP-5 MS (30 m x 0.25 mm, 0.25  $\mu$ m), injection temperature 240°C, [50°C(1 min), (4 °C/min) - 220 °C (5 min)]. The GC-MS analysis was carried out in a GC/MS QP 2010 mass analyzer (70 eV, ion source temp. 220 °C). The existence of at least sixty two constituents in the whole fraction was confirmed. Up to now, about the 80% of the existing compounds was identified [monoterpenic compounds (14.02%), sesquiterpenic compounds (11.48%), diterpenes (2.18%), aliphatic derivatives (31.74%), aromatic derivatives (7.21%), furan derivatives (10.06%), cyclohexane derivatives (3.31%)] [1 – 5]. References: 1. Jennings N., Shibamoto T. (1980) Quantitative Analysis of Flavor and Fragrance Volatiles by G. Capillary Gas Chromatography. Academic Press. N. York. 2. Adams R.P. (1989) Identification of Essential Oils by Ion Trap Mass Spectroscopy. Academic Press. N. York. 3. Adams R.P. (1995) Identification of Essential Oil Components by Gas Chromatography/Mass Spectrometry. Allured Publishing Corporation. Illinois USA. 4. Adams R.P. (2007) Identification of Essential Oil Components by Gas Chromatography/Mass Spectrometry. Allured Publishing Corporation. Carol Stream. 5. Spectral Library Nist.

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Molecular characterization and analysis of the volatile oils of two endemic Portuguese species: Angelica lignescens and Melanoselinum decipiens Mendes MD¹, Trindade H¹, Figueiredo AC¹, Pedro LG¹, Barroso JG¹, Fontinha SS²
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Angelica lignescens and Melanoselinum decipiens were considered as the same species, M. decipiens, until 1998 [1]. However, chemical and morphological studies with these species revealed they were actually different and nowadays it is accepted that A. lignescens is endemic from the Azores archipelago while M. decipiens is endemic from Madeira [1]. Molecular data and analysis of the volatile oils of 12 individuals of A. lignescens and 3 individuals of M. decipiens were studied to determine whether volatile components could be used as taxonomical markers and to examine the relationship between molecular and chemical markers. RAPD markers were used in order to assess the genetic relationships between the individuals of the two species. Volatiles were extracted by distillation-extraction and analyzed by GC and GC-MS. On the basis of their genetic similarities, RAPD analysis grouped the samples into two main genetically distinct clusters, corresponding to the two species under study. Limonene (57 – 86%) was the main component of A. lignescens volatile oils, while  $\beta$ -pinene (38 – 47%), sabinene (1 – 33%) and bornyl acetate (not detected-23%) dominated M. decipiens volatile oils. Chemi-

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