In vitro activity of 20 essential oils against selected dermatophyte species

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The *in vitro* activity of 20 chemically defined essential oils (EOs) obtained from *Boswellia sacra, Citrus bergamia, Citrus limon, Citrus medica, Cynnamomum zeylanicum, Eucalyptus globulus, Foeniculum vulgare, Helicrysum italicum, Illicium verum, Litsea cubeba, Mentha spicata, Myrtus communis, Ocimum basilicum,Origanum majorana,Origanum vulgare, Pelargonium graveolens, Rosmarinus officinalis, Santalum album, Satureja montana, and Thymus serpillum* was assayed against clinical animal isolates of *Microsporum canis*, *Trichophyton mentagrophytes*, *Trichophyton erinacei*, *Microsporum gypseum* and *Trichophyton terrestre*, main causative agents of zoonotic and/or environmental dermatophytoses in humans. Single main components present in higher amounts in such EOs were tested, also. Different dermatophyte species showed remarkable differences in sensitivity. In general, more effective EOs were *T. serpillum* (MIC range 0.025%-0.25%), *O. vulgare* (MIC range 0.025%-0.5%) and *L. cubeba* (MIC range 0.025%-1.5%). *F. vulgare* showed a moderate efficacy against geophilic species such as *M. gypseum* and *T. terrestre*. Among single main components tested, neral was the most active (MIC and MFC values ≤ 0.25%). The results of the present study seem to be promising for an *in vivo* use of some assayed EOs.

Keywords: (3-8) Essential oils, dermatophytes, antimycotic activity, ringworm, *in vitro* sensitivity.

Superficial mycoses in both humans and animals are mainly caused by dermatophytes. These fungi are able to invade keratinized tissue and produce infections that are generally restricted to the corneous layer of skin, hair and nails. Three broad ecological groups of dermatophyte species are recognized, namely anthropophilic, zoophilic and geophilic. In general, zoophilic and geophilic

species cause lesions in humans that are more inflammatory than those induced by anthropophilic species [1]. Human infection is acquired by contact with soil for geophylic species, and with infect animals or fomites for zoophilic dermatophytes.

Tinea capitis and *Tinea corporis*, as well as inflammatory *Tinea capitis* such as favus and kerion (characterized by dry yellow encrustations and hair follicle suppurative infection, respectively) can be caused by zoophilic dermatophytes, such as *Microsporum canis*, *Trichophyton mentagrophytes* and *Trichophyton erinacei* [2, 3]. Main animal reservoirs for the above mentioned species are cats, rodents and hedgehogs, respectively. However, a large number of pet animals can become infected and transmit dermatophytes to humans [4].

Among geophilic species, *Microsporum gypseum* is the dermatophyte more frequently involved in human mycoses. It has been associated with *Tinea corporis* [5] and with favic lesions and kerion of the scalp [2,6], while *Trichophyton terrestre* has rarely been identified as causal agent of *Tinea capitis* [7].

In Europe *M. canis* is the commonest agent for overall. Its incidence is increasing, and it is the dominant agent in southern Europe, with countries such as Austria, Spain, Italy, and Greece reporting the highest numbers and proportions of *M. canis* cases [8], even if *T. mentagrophytes* and *T. erinacei* frequently occur [3,9].

A number of anti-dermatophytic drugs are available. However their side-effects and their decreased sensitivity lead to an extension of the treatment and can open the way to alternative care. In recent years research in aromatic and medicinal plants and particularly in their essential oils (EOs) has attracted many investigators. Several studies have shown evidence of the huge potential of these natural products as antifungal agents, justifying the current use of these compounds in a number of pharmaceutical, food, and cosmetic products [10]. Therefore EOs are considered promising natural products for the development of broad-spectrum, safe and cheap antifungal agents.

Furthermore EOs have been used in veterinary medicine both as monotherapy and associated in mixtures with good outcomes, sometimes better than results from conventional treatments [11, 12, 13].

Aim of the present paper was to evaluate the *in vitro* activity of 20 chemically defined EOs against isolates of *M. canis*, *T. mentagrophytes*, *T. erinacei*, *M. gypseum* and *T. terrestre* obtained from dermatologically diseased animals.

The choice of the EOs was done on the basis of our previous studies and on their availability on the market. The chemical composition of the tested essential oils is reported in Table I. Their aromatic profile is quite different since the plant material is belonging to different botanical families. In the majority of the samples there is a good amount of monoterpene hydrocarbons and oxygenated monoterpenes, with the exception of *C. limon* and *C. medica*, where monoterpene hydrocarbons are the predominant class of

compounds (92.50% and 96.20%) with limonene as main compound (59.20% and 92.20%, respectively). *I. verum* is characterized by high amount of phenyl propanoids (94.20%, due esclusively to (E)-Anethole), also present in *C. zeylanicum* even if in lower amount (80.14%). *P. graveolens* and *E. globulus* show high % of oxygenated monoterpenes (86.16 % and 90.45%, respectively). Only the EO of *S. album* among the others is rich in oxygenated sesquiterpenes (88.79%). However the chemical composition of tested EOs was substantially consistent with the literature data, except for *L. cubeba*. In fact the EO used in the present study, although containing lower amounts of neral, geranial [18] and citral [19] in comparison with the published data, showed a good antidermatophyte activity. At the best of our knowledge, there is only one paper dealing with the anti *M. canis* activity of this EO [20], whilst there is no report about its efficacy against other dermatophyte species.

The selected EOs showed a variable degree of antimycotic activity at tested dilutions, with MICs ranging from 0.025% to > 10%. MIC and MFC values varied among the different dermatophytic species tested. In general terms most effective EOs were *T. serpillum*, *O. vulgare* and *L. cubeba* which had an overall MIC range of 0.025% to 0.25%, 0.025% to 0.5% and 0.025% to 1.5%, respectively. *F. vulgare* showed a moderate efficacy against geophilic species such as *M. gypseum* and *T. terrestre.* Among all tested fungal species *M. canis* had the lowest MIC and MFC values.

Among conventional drug tested, terbinafine was found to be the most effective against *M. canis*, *M. gypseum*, *T. terrestre* and *T. erinacei*, while the best results against *T. mentagrophytes* were achieved using voriconazole. More detailed data are reported in Table II.

Neral was the most effective compound among the single main components tested, showing MIC and MFC values ≤ 0.25%. This compounds was present only in *L. cubeba* EO. Thymol, carvacrol, eugenol, geranial, geraniol and fenchone showed a satisfactory activity on the majority of the tested fungal species. Limonene, p-cymene, α pinene and γ terpinene did not yield any antifungal effect at 10%. Results are reported in detail in Table III.

The overall sensitivity to EOs of examined fungal species showed some marked differences except for zoophilic *Trichophyton* species (*T. mentagrophytes* and *T. erinacei*), which showed a quite homogeneous overall sensitivity pattern. To the best of our knowledge there are not studies including alongside such animal/human pathogens. *T. serpillum* (rich in thymol, 52.61%) presented the lowest MIC value against all fungi, except for *M. gypseum*. For this last species *O. vulgare* gave the best results both in terms of MIC and of MFC. Furthermore *T. serpillum* showed the best results when tested versus *Trichophyton* species. This feature would be due to the large amounts of thymol that had antifungal efficacy in concentrations ranging from 0.05% to 2.5%. Both

Microsporum species and zoophilic *Trichophyton* were sensitive to *L. cubeba* EO, probably due to the large amounts of neral and geranial. *O. vulgare* had a good antimycotic activity against *Microsporum* species tested, confirming the good efficacy of carvacrole.

T. terrestre showed a peculiar sensitivity pattern; it resulted sensitive to *T. serpillum* even if MFC value obtained was quite high in respect of other examined fungi. The most striking finding was the low sensitivity to *L. cubeba* EO, probably due to the poor activity of geranial versus this fungal species.

The MIC values obtained with conventional antimycotic drugs were in substantial agreement with the literature data [21-24].

Several EOs were tested against both geophilic and zoophilic fungi; these latter have been chosen on the basis of their broad host range and their zoonotic potential. Our data confirm anti dermatophyte efficacy of *T. serpillum, O. vulgare* [25, 26] and *L. cubeba* EOs [20].

The results of the present study seem to be promising for a possible *in vivo* use of these EOs, alone and/or as a mixture. The topical administration of EOs mixtures appeared to be effective in veterinary medicine [12-13] allowing the use of lower concentrations of each component and showing a better antimycotic activity in comparison with the ingredients used alone.

The usage of these natural products could be advisable for a topical administration in combination with systemic treatment. In particular *T. mentagrophytes* appeared resistant to all synthetic antimycotic drugs except for voriconazole, while MIC and MFC values for *T. serpillum, L. cubeba* and *O. vulgare* demonstrated a good antimycotic efficacy. This finding was partially referable to *M. gypseum* also, fully sensitive to terbinafine and voriconazole only, and inhibited at low concentrations of *O. vulgare*, *L. cubeba*, *T. serpillum* and *F. vulgare* EOs.

The differences in drugs/remedia sensitivity when tested against distinct dermatophyte species were remarkable, so reiterating the capital role of the correct identification of etiological agents of ringworm to set up a proper treatment.

Some of the used EOs were highly effective against the tested dermatophytes. Further clinical studies are needed to evaluate the possibility of applying these natural products to the treatment of dermatophytosis, providing an effective and safe additional therapeutic option.

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Experimental

Essential Oils - *Boswellia sacra*, *Citrus bergamia*, *Citrus limon*, *Citrus medica*, *Cynnamomum zeylanicum*, *Eucalyptus globulus*, *Foeniculum vulgare*, *Helicrysum italicum*, *Illicium verum*, *Litsea cubeba, Mentha spicata*, *Myrtus communis*, *Ocimum basilicum*, *Origanum majorana*, *Origanum vulgare*, *Pelargonium graveolens*, *Rosmarinus officinalis*, *Santalum album*, *Satureja montana*, and *Thymus serpillum* EOs were employed for all *in vitro* studies. EOs 20% solutions were prepared in sweet almond oil (*Prunus dulcis* Mill. Flora Srl., Lorenzana, Pisa, Italy). The above mentioned EOs were supplied by Flora srl (Lorenzana, Pisa, Italy).

GC-MS analysis - Volatile constituents of each EO were analysed by GC-MS as previously reported (Pistelli et al. 2012). Briefly, a CP-3800 gas chromatograph equipped with HP-5 capillary column (30 m X 0.25 mm; coating thickness, 0.25 mm) and Varian Saturn 2000 ion trap mass detector were employed. Analytical conditions were as follows: injector and transfer line temperature, 220 and 240 °C respectively; oven temperature, programmed from 60 to 240 °C at 3°C/min; carrier gas, helium at 1 ml/min; injection, 0.2 ml (10% hexane solution); split ratio, 1:30. Identification of the constituents was based on comparison of the retention times with those of authentic samples, comparing their linear retention indices relative to the series of n-hydrocarbons, and on computer matching against commercial and home-made library mass spectra built up from pure substances and components of known oils and MS literature data [14,15].

In vitro assay - The *in vitro* antimycotic activity of EOs was evaluated on clinical isolates of *M. canis,*

T. mentagrophytes, *T. erinacei*, *M. gypseum* and *T. terrestre*, respectively. The dermatophytes were cultured by affected cats (*M. canis* and *T. mentagrophytes*) and dogs (*T. erinacei*, *M. gypseum* and *T. terrestre*) and maintained on Sabouraud dextrose agar (SDA). The effectiveness of EOs was assessed by means of a microdilution test carried out as previously described using a semisolid malt extract medium (MEA) with 1% agar [16]. Portions of approximately 1 mm^3 non-sporulating mycelia from SDA were used as fungal inocula in 24- wells plates (Pbi International, Milano, Italy). Stock solutions at 20% of all the chemically defined EOs were diluted in culture medium to obtain concentrations ranging from 10% to 0.01%.

Control cultures were achieved using medium alone and medium supplemented with 0.5% sweet almond oil. Plates were incubated at 25°C for about ten days or until the full development of mycotic growth in control wells was observed. Portions of the inocula where growth was not visually noticed were removed, twice washed in sterile saline and then seeded onto solid MEA plates to evaluate the viability of the fungi. All tests were performed in quadruplicate. Further controls were achieved using griseofulvin (Sigma Aldrich, Italy), itraconazole (Janssen Cilag, Italy) terbinafine (Sandoz, Italy), voriconazole (Sigma Aldrich, Italy) and posaconazole (Sigma Aldrich, Italy) at concentrations from 160 mg/kg to 0.02 mg/kg following the procedure described by Mancianti et al. (1997) [17].

Single main components present in higher amounts in EOs were separately tested *in vitro* as well at concentrations ranging from 10% to 0.02%. So anethol, carvacrol, p-cymene, 1,8 cineole, thymol, eugenol, citronellol, geranial, neral, geraniol, fenchone, linalool, menthol, menthone, α pinene, γ terpinene and limonene were assayed against all dermatophytes as described above. All tests were carried out in quadruplicate. All these components were provided by Sigma-Aldrich (Italy). The viability of the inocula was evaluated as described above.

References

[1] Weitzman I, Summerbell RC. (**1995**) The dermatophytes. *Clinical Microbiology Reviews* **8**: 240–59.

[2] Isa-Isa R., Arenas R., Isa M (**2010**) Inflammatory tinea capitis: kerion, dermatophytic granuloma, and mycetoma *Clinics in Dermatology* **28**:133-136.

[3] Sidwell RU, Chan I, Francis N, Bunker CB. (**2014**) *Trichophyton erinacei* kerion barbae from a hedgehog with direct osculatory transfer to another person. *Clinical Experimental Dermatology* **39**: 38-40.

[4] Monod M, Fratti M, Mignon B, Baudraz-Rosselet F. (**2014**) Dermatophytes transmitted by pets and cattle *Revue Medicine Suisse* **10**: 749-753.

[5] Jeske J, Lupa S, Seneczko F, Glowacka A, Ochecka-Szymanska A. (**1999**) Epidemiology of dermatomycoses of humans in central Poland. Part V. Tinea corporis. *Mycoses* **42**: 661-663.

[6] Ilkit M. (**2010**) Favus of the scalp: an overview and update. *Mycopathologia* **170**:143-154.

[7] Contet-Audonneau N., Grosjean P., Razanakolona L.R., Andriantsinjovina T. and Rapelanoro R. (**2006**) Tinea capitis in Madagascar: a survey in a primary school in Antsirabe. *Annales de Dermatologie et de Vénéréologie* **133**: 22-25.

[8] Hay R.J., Robles W., Midgley G.,and Moore M.K. (**2001**) Tinea capitis in Europe: new perspective on an old problem *Journal of the European Academy of Dermatology and Venereology* **15**:229-233

[9] Drouot S., Mignon B., Fratti B., Roosje P. and Monod M. (**2009**) Pets as the main source of two zoonotic species of the *Trichophyton mentagrophytes* complex in Switzerland, *Arthroderma vanbreuseghemii* and *Arthroderma benhamiae*. *Veterinary Dermatology* **20**: 13-18.

[10] Lang G. and Buchbauer G., (**2012**) A review on recent research results (2008–2010) on essential oils as antimicrobials and antifungals. A review. *Flavour and Fragrance Journal* **27**:13-39.

[11] Pisseri F., Bertoli A., Nardoni S., Pinto L., Pistelli L., Guidi G., Mancianti F. (**2009**) Antifungal activity of tea tree oil from *Melaleuca alternifolia* against *Trichophyton equinum*: an in vivo assay. *Phytomedicine* **16**:1056-1058.

[12] Mugnaini L., Nardoni S., Pistelli L., Leonardi M., Giuliotti L., Benvenuti M. N., Pisseri F., Mancianti F. (**2013**) A herbal antifungal formulation of *Thymus serpillum*, *Origanum vulgare* and *Rosmarinus officinalis* for treating ovine dermatophytosis due to Trichophyton mentagrophytes. *Mycoses* **56**:333-337.

[13] Nardoni S, Mugnaini L, Pistelli L, Leonardi M, Sanna V, Perrucci S, Pisseri F, Mancianti F. (**2014**) Clinical and mycological evaluation of an herbal antifungal formulation in canine Malassezia dermatitis. *Journal de Mycologie Médicale* **24**:234-240.

[14] National Institute of Standards and Technology (**1999**) NIST/EPA/NIH Mass Spectral Library, Pc Version 1.7. Perkin Elmer, Norwalk.

[15] Adams RP. (**2001**). Identification of Essential Oils by Capillary Gas Chromatography/Mass Spectroscopy. Allured Publ, Corp., Carol Stream, IL, USA.

[16] Mugnaini L, Nardoni S, Pinto L, Pistelli L, Leonardi M, Pisseri F, Mancianti F. (**2012**) In vitro and in vivo antifungal activity of some essential oils against feline isolates of *Microsporum canis*. *Journal de Mycologie Médicale* **22**:179-184.

[17] Mancianti F., Zullino C., Papini R. (**1997**) Itraconazole susceptibility of feline isolates of *Microsporum canis*. *Mycoses* **40**:313-315.

[18] Si L.,Chen Y., Han X., Zhan Z., Tian S., Cui Q., Wang Y. (**2012**) Chemical composition of essential oils of *Litsea cubeba* harvested from its distribution areas in China. *Molecules* **17**: 7057-7066.

[19] Lalko J. and Api A.M., (**2006**) Investigation of the dermal sensitization potential of various essential oils in the local lymph node assay. *Food and Chemical Toxicology*. **44**:739-746.

[20] Nardoni S., Tortorano A.M., Mugnaini L., Profili G., Pistelli L., Giovanelli S., Pisseri F., Papini R. and Mancianti F. (2015) Susceptibility of *Microsporum canis* arthrospores to a mixture of chemically defined essential oils: a perspective for environmental decontamination. Zeitschrift für Naturforschung c DOI 10.1515/znc-2014-4105.

[21] Perea S., Fothergill A.W., Sutton D.A., M.G. Rinaldi (**2001**) Comparison of in vitro activities of voriconazole and five established antifungal agents against different species of dermatophytes using a broth microdilution method. *Journal of Clinical Microbiology*. **39**: 385-388.

[22] Gupta A.K., Kohli, Batra R. (**2005**) In vitro activity of posaconazole, ravuconazolo, terbinafine, itraconazole, and fluconazole against dermatophyte, yeast and non-dermatophyte species. Medical Mycology. **43**: 179-185

[23] Sabatelli F., Patel R., Mann P.A., Mendrick C.A., Norris C.C., Hare R., Loebenberg D., Black T.A. and Mc Nicholas P.M. (**2006**) *Antimicrobial Agents and Chemotherapy*. **50**: 2009-2015.

[24] Adimi P, Hashemi SJ, Mahmoudi M, Mirhendi H, Shidfar MR, Emmami M, Rezaei-Matehkolaei A, Gramishoar M, Kordbacheh P. (**2013**) *Iranian Journal of Pharmacological Research*.**12**:537-545.

[25] Inouye S, Uchida K, Nishiyama Y, Hasumi Y, Yamaguchi H, Abe S. (**2007**) Combined effect of heat, essential oils and salt on fungicidal activity against *Trichophyton mentagrophytes* in a foot bath. *Nihon Ishinkin Gakkai Zasshi*.**48**: 27-36.

[26] de Souza E.L., de Barros J.C., de Oliveira C.E., da Conceição M.L.(**2010**) Influence of *Origanum vulgare* L. essential oil on enterotoxin production, membrane permeability and surface characteristics of *Staphylococcus aureus*. *International Journal of Food Microbiology*. **137**:308-311.

Compounds	LRI§	L.c.	M.s.	S.a.	P.g.	I.v.	C.z.	F.v.	C.b.	O.m.	O.v.	S.m.	E.g.	H.i.	M.c.	R.o.	T.s.	O.b.	C.l.	C.p.
α-Thujene	932													7.24						
Tricyclene	938														49.04					
α-Pinene	940	1.12						2.24					1.98			37.89			2.30	
a-Fenchene	951													1.17						
Camphene	955															5.36				
Sabinene	978									3.22									2.50	
β-Pinene	981		1.00						2.81					1.00		5.01			13.70	
6-Methyl-5-hepten-2-one	990	1.38																		
Myrcene	993	1.09								1.55	2.20					1.63			1.80	2.20
α-Phellandrene	1006							1.29												
α-Terpinene	1019									4.73	2.05	1.17								
o-Cymene	1026															1.14				
p-Cymene	1028							1.67	3.75	4.17	9.33	8.96	6.73	1.10	2.66		15.25			
Limonene	1032	10.84	1.65			2.90		1.76	27.06	2.13				6.97	5.94	3.26			59.20	92.20
1,8-Cineole	1036		5.21									1.01	89.76	2.25	28.95	22.01		5.90		
(E)-β-Ocimene	1053																			
γ-Terpinene	1062								3.45	7.90	5.25	6.06					2.92		10.80	
cis-Sabinene hydrate	1072									3.16										
Terpinolene	1090									1.53										
Fenchone	1090						17.40	18.39												
trans-Sabinene hydrate	1098									12.83	1.78						3.76			
Linalool	1102	1.66			3.90				16.83			3.11			1.46			46.00		
Camphor	1148															7.57				
Menthone	1154		16.46		1.07															
Citronellal	1155	1.70																		
Isomenthone	1164				3.48															
neo-Menthol	1166		11.18																	
Borneol	1169											2.05				2.04	1.56			
Menthol	1178		39.03																	
4-Terpineol	1180									17.61							2.41			
α-Terpineol	1192									2.73										
unknown																	1.69			
Methyl chavicol (= estragol)	1198							2.89										1.10		

Citronellol	1231				44.46														
Thymol methyl ether	1232																1.72		
Pulegone	1237		3.29																
Neral	1242	31.97																	1.00
Carvone	1248											1.90							
para-Anisaldehyde	1258							1.92											
Geraniol	1259	1.48			13.73					2.73									
Linalool acetate	1260								34.47	3.22		1.19							
Geranial	1276	36.89																	1.50
Citronellyl formate	1280				7.26														
(E)-Anethole	1283					94.20		65.01											
Isobornyl acetate	1285														3.32		1.60		
Thymol	1290											2.57				52.61			
Safrole	1291						1.36												
Menthyl acetate	1294		6.94																
Geranyl formate	1298				1.92														
Carvacrol	1299									20.84	65.94	47.10							
Eugenol	1361						64.77										11.50		
Neryl acetate	1368													31.83					1.00
β-Elemene	1392																2.20		
β-Caryophyllene	1418	2.21	2.09				2.46			1.72	3.72	3.60		3.11		4.06	6.77		
trans-α-Bergamotene	1437																3.60		
α-Guaiene	1440																1.10		
Aromadendrene	1445				2.86														
Cinnamyl acetate	1446						1.12												
unknown														2.61					
Isoeugenol	1449						14.01												
Neryl propanoate	1454													5.12					
(E)-b-Farnesene	1458																		
β-Santalene	1461			1.42															
Germacrene D	1481													3.43			3.50		
ar-Curcumene	1484													5.56					
trans-Muurola-4(14),5-diene (=Bicyclosesquiphellandrene)	1492													2.11					
Valencene	1493													1.68					

Bicyclogermacrene	1495	0.15															1.00			
α-Muurolene	1499									1.36										
a-Bulnesene	1505																2.00			
β-Bisabolene	1509											1.53						1.10		
trans-γ-Cadinene	1513													1.14			2.80			
δ-Cadinene	1523											1.42				1.02				
Citronellyl butyrate	1532				1.19															
Selina-3,7(11)-diene	1542			1.00																
Elemol	1553			4.69																
Geranyl butyrate	1564				1.11															
(E)-2-Phenyl ethyl tiglate	1585				1.19															
5-epi-7-epi-a-Eudesmol	1606													5.47						
1,10-di-epi-Cubenol	1614																1.00			
epi-10-γ-Eudesmol	1627			4.87																
γ-Eudesmol	1634			5.16																
tau-Cadinol	1640																5.80			
β-Eudesmol	1649			5.52																
(Z)-Citronellyl tiglate	1658				1.15															
Valerianol	1658			14.35																
7-epi-α-Eudesmol	1664			5.92																
(Z)-α-Santalol	1672			27.09																
(Z)-trans-α-Bergamotol	1691			2.00																
Geranyl tiglate	1696				1.19															
(Z)-β-cis-Santalol	1705			2.13																
(Z)-β-trans-Santalol	1710			10.75																
Benzyl benzoate	1764						2.74													
(Z)-Lanceol	1768			1.16																
Monoterpene hydrocarbons		4.42	7.08	-	-	3.03	3.36	8.23	38.80	25.40	34.60	19.79	9.07	19.16	61.22	56.52	21.68	2.03	92.50	96.20
Oxygenated monoterpenes		50.37	76.69	-	86.16	-	1.30	19.37	57.74	63.00	58.10	62.09	90.45	35.47	31.11	36.67	64.14	56.10	4.90	0.50
Sesquiterpenes hydrocarbons		39.57	10.52	5.63	7.77	-	3.59	-	1.62	4.90	5.80	11.88	0.33	29.40	1.18	4.38	9.20	20.00	2.60	1.90
Oxygenated sesquiterpenes		0.33	1.52	88.79	4.14	-	0.38	-	0.46	0.90	0.20	1.10	0.14	9.35	-	0.25	-	7.90	-	-
Phenyl propanoids		-	-	-	-	94.20	80.14	67.90	-	-	-	0.16	-	-	-	-	-	12.70	-	-
Others		1.38	0.18	-	1.19	-	4.09	1.92	0.12	0.20	1.00	0	-	-	-	-	0.79	0.20	-	0.60
Unknowns		-	-	-	-	-	-	-	-	-	-	-	-	2.61	0.34	0.12	1.69	-	-	-

Total		96.85	95.99	94.42	99.26	97.23	92.83	97.42	98.64	98.28	98,44	95.02	99.99	94.25	93.85	97.94	97.31	99.20	100.00	99.20

EOs	M. canis	M. canis	M. gypseum	M. gypseum	T. mentagrophytes	T. mentagrophytes	T.terrestre	T.terrestre	T. erinacei	T. erinacei
Litsea	0.025	0.25	0.25	0.5	0.25	0.5	1.5	5	0.25	2.5
Feniculum	0.25	1.5	0.5	1.5	1.5	2	1.5	3	3	3
Illicium	3	7.5	1	2	2	2.5	1.5	5	3.5	5
Pelargonium	0.25	0.5	1.5	2.5	0.75	2	0.75	3	0.75	2
Eucaliptus	0.1	0.5	0.5	0.5	0,5	1	1,5	3	0.5	0.5
Satureja	0.5	3	2	5	2	7.5	3	7.5	2	7
Origanum majorana	0.5	4	2	7.5	1	6	2	7.5	1.5	6
Menta spicata	2	5	3	7.5	3	5	3	5	3	5
Santalum	7,5	>10	7,5	>10	7,5	>10	10	>10	7.5	>10
Cynnamomum	4.5	5	7,5	10	7,5	>10	10	>10	7.5	>10
Citrus bergamia	4	5	5	7.5	5	7.5	5	7.5	7.5	>10
Boswellia sagra	5	>10	7,5	>10	7,5	>10	10	>10	8	>10
Origanum vulgare	0.025	0.05	0.025	0.05	0.5	0.5	0,25	1.5	0.5	1
Thymus serpillum	0.025	0.05	0.25	0.5	0.1	0.1	0,1	1	0.2	0.5
Elicriso	5	>10	10	>10	10	>10	10	>10	10	>10
Mirto	2	3	3	5	1,5	5	3	6	2	5
Basilico	1	5	3	5	2,5	5	3	6	2.5	5
Cedro	4	5	5	7.5	7,5	10	10	10	8	10
Limone	2.5	7.5	2.5	10	5	7.5	7,5	10	5	7.5
Rosmarino	2.5	7.5	2.5	7.5	5	7.5	5	7.5	1.5	1.5
C. berg fant	1	2	1	2	1	2	5	7.5	5	>10
C. berg castagn	10		10		10		5	>10	>10	>10
C.berg Femm	5		>10		>10		5	>10	>10	>10
Tangerine	>10		>10		>10	>10		>10		>10
Paradisi	>10		>10		>10	>10		>10		>10
Griseofulvin	1		40		160				2	
Terbinafine	0.0156		0.16		16				0.01	
Itraconazole	0.125		32		32				0.25	
Posaconazole	0.0625		8		16				8	
Voriconazole	0.0625		3.2		3.2				0.25	
	mg/l									