Chemistry of Natural Compounds, Vol. 45, No. 6, 2009

CHEMICAL COMPOSITION AND ANTIMICROBIAL ACTIVITY OF THE ESSENTIAL OILS OF THREE *Anthemis* SPECIES FROM TURKEY

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UDC 547.913

The genus *Anthemis* L. of Asteraceae (Compositae) is represented in the Flora of Turkey by 81 taxa belonging to 51 species, 29 of which are endemic to Turkey. One of the endemic species is *A. dipsacea* Bornm. *A. pectinata* Boiss.& Reuter var. *pectinata* and *A. dipsacea* are distributed in western regions of Turkey. *A. pseudocotula* Boiss. grows wild in the western, eastern, and southern regions of Turkey. A detailed botanical description of the studied three species is presented in Flora of Turkey [1].

All of these *Anthemis* species are called as "Papatya" in Turkey, and infusions of *A. pseudocotula* are used in Turkish traditional medicine especially for hemorrhoid [2]. Papatya is a common name given to plants whose flowers resemble those of Roman and German chamomile. Many *Anthemis* spp. are used as herbal tea and for food flavoring, as well as cosmetics and in the pharmaceutical industry [3–5]. Their extracts, tinctures, salves, and tisanes are widely used as antispasmodic, anti-inflammatory, and antibacterial in Europe. The occurrence of sesquiterpene lactones, flavonoids, and essential oils in various *Anthemis* species has been reported in previous works [6–12].

To the best of our knowledge, there is no published report on the phytochemical composition and antimicrobial activity of *A. dipsacea, A. pectinata* var. *pectinata*, and *A. pseudocotula* essential oils. Therefore, we focused our study on the composition of the oils using GC and GC-MS analysis; antimicrobial activity was determined using the agar disc diffusion method.

The results of GC/MS analysis of essential oils of *A. dipsacea, A. pectinata* var. *pectinata* and *A. pseudocotula* obtained by hydrodistillation are shown in Table 1. Eighty-four components representing 88.8% *A. pseudocotula* essential oil, 75 compounds representing 85.3% *A. pectinata* var. *pectinata* essential oil, and 71 compounds representing 84.1% *A. pseudocotula* essential oil were characterized.

Although the Anthemideae is one of the chemically most investigated tribes of Asteraceae, according to literature only the essential oils of *A. montana* L. ssp. *carpatica* [6], *A. carpatica* Willd. [13], *A. nobilis* L.[14], *A. tinctoria* L. [15], *A. melampodina* auct. Non Delili [16], *A. xylopoda* O. Schwarz [17], *A. ruthenica* M.B. and *A. arvensis* L. [18, 22], *A. cretica* L. ssp. *leucanthemoides* (Boiss.) Grierson [19], *A. altissima* L. var. *altissima* [20], *A. altissima* L. [21] *A. arvensis* L. [22], *A. aciphylla* Boiss. var. *discoidea* Boiss. [23], *A. chia* L., *A. tomentosa* L., *A. auriculata* Boiss., *A. weneri* L. ssp. *weneri* Stoj.&Acht., *A. altissima* L., *A. melanolepis* Boiss., *A. tinctoria* L. var. *parnassica* and *A. cotula* L. [24], *A. triumfetti* (L.) DC. [25], *A. hyalina* DC. [26], and *A. wiedemaniana* Fish. Et. Mey. [27] have been studied so far.

To the best of our knowledge, no report exists on the essential oil compositions of *A. dipsacea*, *A. pectinata* var. *pectinata*, and *A. pseudocotula.* According to our results, the common main constituents of the essential oil from aerial parts of *A. dipsacea* were characterized by a high percentage of fatty acid [hexadecanoic acid (13.5%)], followed by sesquiterpene hydrocarbons [germacrene D (10.2%) and β-caryophyllene (5.6%)]. Fatty acid [hexadecanoic acid (9.5%)] and oxygenated monoterpenes [linalool (8.9%) and 1,8-cineole (7.5%)] were found to be the main components in the essential oil from the aerial parts *A. pseudocotula.* Fatty acids (hexadecanoic or palmitic) were also observed to be the major constituents in previously studied essential oils originating from *A. altissima*, *A. ruthenica*, and *A. arvensis* [21, 22].

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Published in Khimiya Prirodnykh Soedinenii, No. 6, pp. 751–754, November–December, 2009. Original article submitted April 6, 2008.

0009-3130/09/4506-0900

TABLE 1. Essential Oil Compositions of *A. dipsacea*, *A. pectinata* var. *pectinata,* and *A. pseudocotula*, %

Compound	RRI	1	2	3	Compound	RRI	1	2	3
α-Pinene	1032	0.4	1.9	2.4	(Z)-β-Farnesene	1668	3.9	0.7	0.1
Santolinatriene (=2,5-dimethyl-3-vinyl-1,4-hexadiene)	1043	Tr.	1.5	49.5	*trans*-Pinocarveol	1670	–	0.3	–
Camphene	1076	–	0.4	–	α-Humulene	1681	1.1	–	0.3
Undecane	1100	–	–	0.5	*trans*-Chrysanthemol	1684	0.4	0.5	–
β-Pinene	1118	Tr.	1.7	0.9	*trans*-Piperitol (=*trans*-*p*-Menth-1-en-3-ol)	1689	–	0.5	–
Sabinene	1132	0.1	0.9		γ-Selinene	1690	–	–	0.1
Myrcene	1174	–	–	0.4	Salicylaldehyde	1703	–	0.3	–
Isoamyl propionate	1195	–	1.2		γ-Muurolene	1704	0.4	–	–
Isobutyl 3-methyl butyrate (=*Isobutyl isovalerate*)	1198	–	0.2	–	α-Terpineol	1706	–	1.3	0.3
Limonene	1203	Tr.	0.3	–	Borneol	1719	0.2	3.2	–
1,8-Cineole	1213	1.9	7.5	0.6	Germacrene D	1726	10.2	1.8	1.5
Isochrysanthenone	1234	–	0.3	–	(*Z,E*)-α-Farnesene	1737	0.3	–	–
2-Pentyl furan	1244	Tr.	0.2	–	*p*-Mentha-1,5-dien-8-ol	1738	–	–	0.1
(*Z*)-β-Ocimene	1246	Tr.	–	Tr.	α-Muurolene	1740	0.6	0.4	–
γ-Terpinene	1255	Tr.	0.3	0.1	β-Bisabolene	1741	–	–	0.5
(*E*)-β-Ocimene	1266	0.4	–	Tr.	α-Selinene	1744	–	0.2	–
p-Cymene	1280	0.1	1.0	0.3	Bicyclogermacrene	1755	2.0	–	0.1
2-Methyl butyl 2-methyl butyrate	1286	–	0.2	–	*cis*-Piperitol	1758	–	0.4	–
1,2,4-Trimethyl benzene	1294	–	0.5	–	*cis*-Chrysanthenol	1764	–	0.2	–
2-Methylbutyl isovalerate	1299	Tr.	0.6	–	δ-Cadinene	1773	0.6	0.1	0.3
1,2,3-Trimethyl benzene	1355	–	0.4	–	γ-Cadinene	1776	0.5	0.1	0.1
Nonanal	1400	–	0.4	–	Myrtenol	1804	–	0.2	–
Yomogi alcohol	1403	0.2	–	0.1	Nerol	1808	–	0.1	–
Filifolone	1445	–	3.0	–	Isogeraniol	1820	–	0.4	–
1-Octen-3-ol	1452	0.1	0.3	0.1	(*E,E*)-2,4-Decadienal	1827	–	0.1	–
2,6-Dimethyl-1,3(*E*),5(*E*),7-octatetraene	1460	–	–	0.1	Hexyl octanoate	1829	–	–	0.2
Eucarvone	1465	–	0.2	–	2,6-Dimethyl-3(*E*),5(*E*),7-octatriene-2-ol	1830	–	–	0.2
δ-Elemene	1479	–	–	0.2	(*E*)-β-Damascenone	1838	–	0.1	–
α-Copaene	1497	0.1	–	0.2	*trans*-Carveol	1845	–	0.1	–
Artemisia alcohol	1510	0.1	–	–	Geraniol	1857	–	0.5	–
Chrysanthenone	1522	–	5.1	–	*p*-Cymen-8-ol	1864	–	0.2	–
Camphor	1532	–	0.4		(*E*)-Geranyl acetone	1868	–	0.1	–
β-Bourbonene	1535	0.7	–	–	Neryl isovalerate	1871	–	–	0.2
Modephene	1541	0.1	–	–	2,5-Dimethoxy-*p*-cymene	1878	–	–	0.1
(*E*)-2-Nonenal	1548	Tr.	–	–	Geranyl isovalerate	1893	–	–	1.2
β-Cubebene	1549	Tr.	–	–	Isobutyl phenylacetate	1908	–	0.1	–
Linalool	1553	–	8.9	0.2	Tetradecanal	1933	–	–	0.1
Octanol	1562	Tr.	–	–	Neryl valerate	1933	–	–	0.1
trans-*p*-Menth-2-en-1-ol	1571	–	0.8	–	α-Calacorene	1941	1.1	–	–
β-Ylangene	1580	0.2	–	0.2	1,5-Epoxy-salvial-(4)14-ene	1945	0.2	–	–
Pinocarvone	1586	–	0.3	–	Dendrolasin	1945	0.1	–	–
Bornyl acetate	1590	–	0.5	–	(*E*)-β-Ionone	1958	0.2	0.2	–
α-Guaiene	1596	–	–	0.1	2-Phenylethyl-2-methylbutyrate	1988	0.2	0.3	–
β-Copaene	1597	0.3	–	0.1	Neophytadiene	1992	–	–	0.2
β-Elemene	1600	–	0.7	–	Isocaryophyllene oxide	2001	0.3	–	–
Terpinen-4-ol	1611	–	3.0	0.3	Caryophyllene oxide	2008	3.3	1.6	1.8
β-Caryophyllene	1612	5.6	–	1.1	Isoamyl phenyl acetate	2016	–	0.1	–
Hotrienol	1616	–	0.5	–	Pentadecanal	2041	–	0.2	0.1
cis-*p*-Menth-2-en-1-ol	1638	–	0.6	–	Humulene epoxide II	2071	0.4	0.3	0.3
Thuj-3-en-10-al	1642	–	0.2	–	Caryophylla-2(12),6(13)-dien-5-one	2074	–	0.5	–
Isobornyl propionate	1655	–	Tr.	–	Tridecanol	2077	–	–	0.3
Sabinyl acetate	1658	0.6	–	–	Octanoic acid	2084	–	0.1	–
γ-Gurjunene	1659	–	–	0.1	Hexyl benzoate	2095	–	–	0.2
					Globulol	2098	0.3	–	–

TABLE 1 (continued)

Compound	RRI	1	2	3	Compound	RRI	1	2	3
Heneicosane	2100	0.4	–	–	Eudesma-4 (15), 7-dien-1β-ol	2384	0.8	–	0.4
Viridiflorol	2104	0.2	–	–	1-Hexadecanol	2386	–	–	0.2
α-Guaiol	2103	–	1.4	–	Caryophylla-2(12),6-dien-5β-ol (=Caryophyllenol II)	2392	0.3	0.7	0.5
Salviadienol	2130	–	–	0.1	γ-Dodecalactone	2396	–	–	0.6
Hexahydrofarnesylacetone	2131	0.9	0.4	0.5	α-Phenylpropanoid	2419	–	4.1	–
Spathulenol	2144	4.0	0.4	1.0	Chamazulene	2430	2.5		–
Tetradecanol	2179	0.2	–	0.7	α-Phenylpropanoid	2441	–	2.2	–
T-Cadinol	2187	3.8	–	0.4	Dodecanoic acid	2496	–	0.2	3.3
Nonanoic acid	2192	–	–	0.4	Pentacosane	2500	1.7	1.5	
Thymol	2198	–	–	0.4	γ-Costol	2533	1.6	–	–
Eremoligenol	2204	–	1.0	–	Hexacosane	2600	–	–	0.2
Copaborneol	2205	0.8	–	–	α-Costol	2604	–	–	0.2
T-Muurolol	2209	0.3	–	0.7	β-Costol	2606	–	0.3	–
Clovenol	2219	–	–	–	1-Octadecanol	2607	0.5	–	0.2
α-Bisabolol	2232	–	–	0.7	14-Hydroxy-δ-cadinene	2607	0.3	–	–
Carvacrol	2239	–	–	0.2	Tridecanoic acid	2617	–	–	0.1
trans-α-Bergamotol	2247	0.6	–	–	Phytol	2622	2.0	0.8	1.0
α-Cadinol	2255	2.0	–	0.8	Tetradecanoic acid (= Myristic acid)	2670	2.1	1.3	0.9
β-Eudesmol	2257	–	0.8	–	(*Z*)-Octadec-9-en-18-olide	2676	0.5	0.4	0.1
Alismol	2264	–	–	0.6	Heptacosane	2700	1.2	1.0	0.6
Torilenol	2278	–	–	0.3	Octacosane	2800	–	–	0.2
Selina-11-en-4α-ol	2278	1.3	–	–	Nonacosane	2900	1.2	0.7	0.7
Decanoic acid	2298	–	1.4	0.1	Hexadecanoic acid	2931	13.5	9.5	2.7
Tricosane	2300	3.1	0.5	0.2	Triacontane	3000	–	–	0.2
9-Geranyl *p*-cymene	2312	1.1	–	–	Total		84.1	82.5	85.3
Caryophylla-2(12),6(13)-dien-5β-ol (=Caryophylladienol I)	2316		0.8	–					
Caryophylla-2(12),6-dien-5α-ol (=Caryophyllenol I)	2361	–	–	0.2					

1 – *Anthemis dipsacea*, 2 – *Anthemis pseudoculata*, 3 – *Anthemis pectinata* var. *pectinata*.
RRI: relative retention indices calculated agains *n*-alkanes. Percentage calculated from FID data; Tr.: trace (<0.1%).

TABLE 2. Antimicrobial Activity of Essential Oils of *A. dipsacea, A. pectinata* var. *pectinata,* and *A. pseudocotula*

Microorganisms	Source No.	Inhibition zone, mm*			Standard antibiotics		
		A	B	C	CF20	SAM20	NS20
Gram-positive							
Enterococcus faecalis	ATCC 29212	–	–	–	17	19	Nt.
Staphylococcus aureus	ATCC 6538/P	8	10	11	24	23	Nt.
Stapylococcus epidermidis	ATCC 12228	–	–	–	12	19	Nt.
Gram-negative							
Enterococcus cloacae	ATCC 13047	–	–	–	10	17	Nt.
Escherichia coli	ATCC 11230	7	8	10	21	13	Nt.
Escherichia coli	ATCC 29998	8	8	12	22	12	Nt.
Pseudomonas aeruginosa	ATCC 27853	8	8	8	30	–	Nt.
Salmonella tyhimurium	CCM 5445	–	–	–	20	15	Nt.
Fungi							
Candida albicans	ATCC 10239	–	–	–	Nt.	Nt.	19

A: essential oil of *A. dipsacea* (20 µL); B: essential oil of *A. pectinata* var. *pectinata* (20 µL); C: essential oil of *A. pseudocotula* (20 µL); CF20: ceftazidime (20 µg); SAM20: sulbactam (10 µg)/ampicillin (10 µg); NS20: nystatin (20 µg); Nt.: not tested; –: not active. *Includes diameter of disc (6 mm).

The essential oil from the aerial parts of *A. pectinata* var. *pectinata* was characterized by a high percentage of santolinatriene (49.5%). With such a high percentage, this oil could be a good source of santolinatriene, since this compound is rarely encountered in essential oils. The monoterpene hydrocarbon (santolinatriene) was also found to be the major constituent in previously studied essential oil of *A. melampodina* [16]. It was first isolated from *Santolina chamaecyparissus* L. [28].

Results from the antimicrobial screening tests are shown in Table 2. As is clearly seen in Table 2, the essential oils of *A. dipsacea, A. pectinata* var. *pectinata*, and *A. pseudocotula* inhibited the growth of four out of eight microorganisms but had no effect on the growth of *Enterobacter cloacae* ATCC 13047, *Enterococcus faecalis* ATCC 29212, *Staphylococcus epidermidis* ATCC 12228, and *Salmonella thyphimurium* CCM 5445. *Staphylococus aureus* ATCC 6538P, *Pseudomonas aeruginosa* ATCC 27853, *Escherichia coli* ATCC 11230, and *Escherichia coli* ATCC 22998 were inhibited by the essential oils of *A. dipsacea*, *A. pectinata*, and *A. pseudocotula* at a concentration of 20 μL/disc when compared with standard antibiotics such as ceftazidime (CF20), sulbactam/ampicilin (SAM20), and nystatin (NS20) used as positive controls. A comparison of these findings with the control antibiotics CF20 and SAM20 showed that these essential oils have strong antibacterial activity against *Staphylococus aureus* ATCC 6538P, *Escherichia coli* ATCC 11230, *Pseudomonas aeruginosa* ATCC 27853, and *Escherichia coli* ATCC 29998. The oil showed activity similar to that of SAM20 against *Escherichia coli* ATCC 11230 and *Escherichia coli* ATCC 22998. However, its antibacterial effects were moderate against the remaining four bacteria. The essential oils of *A. dipsacee*, *A. pectinata* var. *pectinata*, and *A. pseudocotula* were ineffective against *Candida albicans* ATCC 10239.

Hexadecanoic acid (palmitic acid) is known to be a potential antibacterial agent [29]. Previous studies showed that linalool and 1,8-cineole are well-known antimicrobial compounds isolated from different plant species [30–34]. The essential oil of *A. pseudocotula* showed stronger antibacterial activity than the other oils.This activity may be attributed to the presence of higher concentrations of linalool (8.9%) and 1,8-cineole (7.5%) in this oil than in the other oils.

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