much lower sp. act. (0.02 units/mg) compared to 70–75% of the total activity and 40.6 sp. act. of enzyme I and 18–21% of the total activity and 5.2 sp. act. of enzyme II. At the moment we are not certain if this fraction is real or an artifact, although this fraction was also present in the freshly prepared crude enzyme extracts obtained from 3- and 6-day-old seedlings and the ripening seeds near maturing [unpublished data]. This fraction was not analysed any further.

Judging from the size of the peaks α-galactosidase I had *ca* 4 × the activity of enzyme II in germinating chick peas seedling. In contrast to this, however, Barham *et al.* [3] reported that the high MW forms of α-galactosidase, which are generally predominant in dormant seeds, were readily replaced by the low MW forms on germination. Thus, multiple forms of α-galactosidase in chick peas appear to function differently from those in *Vicia faba* seeds. In this respect it may be pointed out that Sephadex gel filtration of the freshly prepared crude extracts from 3- and 6-day-old germinating chick peas also showed a predominance of the enzyme I, and the activity ratio of enzyme I with respect to the enzyme II increased in favour of enzyme I [unpublished data]. Thus, the predominance of high MW form of α-galactosidase I in the germinating chick peas was thought to be real and perhaps *in vivo* conversion of the low MW species to high MW enzyme takes place during germination as the latter is *ca* × 3 the MW of the former (Table 1). However, the *in vitro* conversion of enzyme II to enzyme I as observed by Dey and Pridham [9] is possible and further work is needed to clarify this point.

A comparison of the properties of the two forms of the enzyme is shown in Table 1. The two enzymes from chick pea appear to follow the pattern reported by Dey and Pridham [4] except that in the present case the thermal stability of the low MW species of the enzyme was somewhat greater than that of high MW. Furthermore, the two forms can be distinguished from one another with respect to their MWs, thermal stability, energy of activation, ΔH and ΔS values, and responses to specific inhibitors.

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THE ESSENTIAL OIL OF *ARTEMISIA CAPILLARIS**

MITSUO MIYAZAWA and HIROMU KAMEOKA

Department of Applied Chemistry, Faculty of Science and Engineering, Kinki University, Kowakae, Higashiosaka-shi, Osaka, Japan

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Key Word Index—*Artemisia capillaris*; Compositae; essential oil; terpenoids; phenylacetylenes; phenols; fatty acids.

Abstract—Twenty-five terpenoids, 6-phenylacetylenes, 7 phenols, and 15 fatty acids were characterized in this oil. It differs considerably in composition from the oils of *A. kurromensis*, *A. maritima* and *A. fukudo*, which have α- and β-thujone as the major constituents.

INTRODUCTION

350 species of *Artemisia* occur throughout the world, about 30 of which are known in Japan. In our previous papers, [1–4], the new acetylenic compounds, 1-(2′-methoxy phenyl)-2,4-hexadiyne (*o*-methoxycapillen), capillanol, norcapillen and neocapillen, were reported in the essential oil of the stalks and leaves or extract of roots of *A. capillaris* Thunb. (Kawarayomogi). The volatile constituents from the stalks and leaves of *A. capillaris* have also been reported by Harada *et al.* [5–8], but the terpenoid, phenol and fatty acid constituents have not so far been studied. Mono- and sesqui-terpenes have been identified in other *Artemisia* species [9–13]. This

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communication describes the volatile constituents from the stalks and leaves of *A. capillaris* collected in the suburbs of Osaka-Ku in Japan.

RESULTS AND DISCUSSION

The neutral part of the essential oil of *A. capillaris*, was analyzed by various spectrometric techniques, the 33 components listed in Table 1 being identified. The relative concentrations were calculated on the basis of GLC peak areas. The essential oil contains capillen as a main component and limonene, β-pinene, β-elemene, β-caryophyllene, *ar*-curcumene and α-humulene as minor components. There are also 17 monoterpenes, 8 sesquiterpene hydrocarbons, 2 miscellaneous structures and 6 acetylenic compounds.

Among the most noteworthy features of this oil are, the presence of β-myrcene, *cis*- and *trans*-ocimene, *cis*- and *trans*-allo-ocimene and a rather large quantity of acetylenic compounds. The acyclic hydrocarbons consisted of 5 unsaturated compounds. These represented only a small portion of the oil (0.7%). Seventeen monoterpene hydrocarbons which included 5 bicyclic compounds, 5 cyclic compounds, 6 acyclic compounds and 2 aromatic compounds accounted for 8.5% of the total oil.

Table 1 lists 8 sesquiterpene compounds that accounted for 3.2% of the total oil. *Ar*-curcumene, β-caryophyllene, β-elemene and α-humulene, are the major compounds in this class (3.0%). Acetylenic compounds accounted for 82.6% of the oil. Capillen was the major compounds in this class (80.2%). Thus, the oil is comprised mostly of acetylene compounds (82.6%) with smaller quantities of several monoterpenes (8.7%), sesquiterpenes (2.5%), phenols (0.8%), miscellaneous compounds (2.1%) and fatty acids (0.2%).

The acidic part, examined after methylation was considerably less complex than the neutral part. As expected, all C_4-C_{18} straight-chain saturated fatty acids were found in *A. capillaris*, palmitic acid being the main constituent.

The compositions of the volatile fractions obtained from the seven *Artemisia* species examined particularly those of *A. fukudo*, *A. kurromensis* and *A. maritima*, are strikingly similar. The essential oils from *A. japonica* and *A. apiacea* contains ε-cadinene, β-caryophyllene, β-humulene and artemisia ketone as the characteristic components, while β-pinene, limonene, γ-terpinene, *ar*-curcumene and acetylenic compounds such as capillen, *o*-methoxycappillen, capillanol, norcapillene and capillin are present in the oil from *A. capillaris*.

EXPERIMENTAL

IR spectra were taken in liquid films, and the NMR spectra were measured on a 60 MHz apparatus with tetramethylsilane as internal standard in CCl_4. For measuring the MS spectra, a single focus apparatus was used with an ion accelerating voltage of 3500 V, an ionization voltage of 70 eV and an ionization chamber temperature of 210°. UV spectra were measured in EtOH.

Plant material and essential oil. Aerial parts of the plant were collected near the mouth of the Yamato river running through Osaka-Fu, at the end of October 1973. After steam distillation of 11.0 Kg of the *A. capillaris*, 88.3 g (0.803%) of the essential oil was obtained by the extraction of the distillate with Et_2O and by the evaporation of the solvent under N_2. The essential oil had the following characteristics: d_4^{25} 0.9719, n_D^{25} 1.5421, $(\alpha)_D^{25}$ −11.9, AV 1.82.

The essential oil (80.0 g) was treated with 5% Na_2CO_3, then with 5% NaOH, in order to separate into the neutral (79.3 g), phenolic (0.6 g) and acidic fractions (trace).

Neutral fraction. 20.0 g was chromatographed on activated alumina (300 mesh, a 2.5 × 55 cm glass tube) with *n*-hexane (300 ml), C_6H_6 (200 ml), Et_2O (200 ml), EtOAc (200 ml) and EtOH (300 ml), and divided into 5 fractions. *n*-hexane (2.5 g), C_6H_6 (15.3 g), Et_2O (1.6 g), EtOAc (0.2 g), EtOH (0.3 g). Each of the fractions was subsequently subjected to elution chromatography and/or preparative GLC (3 m × 3 mm stainless steel separation column packed with 15% of SE-30 and PEG-20M on Celite-545 (80–100 mesh)) for separation into individual components. The components thus isolated in sufficient amount were identified by IR [14], NMR, MS [15] and UV.

Phenolic fraction. The phenolic compounds (0.6 g) were isolated in the pure state by means of preparative GLC. Identification of the compounds was attained by comparing their MS, IR, NMR and R_t with those of authentic samples obtained commercially or synthetically.

Acidic fraction. The acidic fraction (trace amount) from the essential oil was reacted with ethereal CH_2N_2 overnight and subsequently examined by GC–MS.

Table 1. Chemical constituents of the essential oil of some species in the *Artemisia* genus

Compound	Method of identification	*A. capillaris*	(I)	(II)	(III)	(IV)	(V)	(VI)
Neutral fraction (monoterpenes)								
α-pinene	MS, IR	0.2	+	0.5	0.5	0.6	+	+
camphene	MS, IR	+	+	2.1		1.6	+	+
β-pinene	MS, IR, NMR	2.2		1.5	0.4		3.0	1.0
sabinene	MS, IR	+	3.0	0.5		1.5		
β-myrcene	MS, IR	0.6	+					
α-terpinene	MS, IR	0.2	+					
limonene	MS, IR, NMR	3.2					2.0	4.0
1,8-cineole	MS, IR	+	+	15.6	9.3	3.2	1.0	2.0
cis-ocimene	MS, IR	+						
γ-terpinene	MS, IR, NMR	1.4				2.2		
trans-ocimene	MS, IR	+						

Table 1 (cont.)

Compound	Method of identification	*A. capillaris*	(I)	(II)	(III)	(IV)	(V)	(VI)
p-cymene	MS, IR, NMR	0.8	+	3.2	4.3	0.2		+
terpinolene	MS, IR	+						
cis-allo-ocimene	MS, IR	+						
trans-allo-cimene	MS, IR	+						
artemisia ketone						2.5	+	+
α-thujone			40.0		31.5	1.0	+	+
β-thujone			13.0	62.0	15.5			
artemisia alcohol							1.0	
camphor			6.0	12.0	2.1	10.5		
bornyl acetate						2.5		
sabinyl acetate					11.0			
borneol						15.4		
α-terpineol	MS, IR	+						
p-cymene-8-ol	MS, IR	+						
(sesquiterpenes)								
α-copaene	MS, IR	+	+ +				1.0	1.0
β-bourbonene							+	3.0
α-bergamotene	MS, IR	+						
β-elemene	MS, IR, NMR	0.6						
β-gurjunene	MS, IR, NMR	0.1						
β-caryophyllene	MS, IR, NMR	0.9	2.0			2.9	12.0	9.0
tricyclobetivene						1.0	2.0	
α-humulene	MS, IR, NMR	0.5				4.1	4.0	16.0
β-bisabolene	MS, IR, NMR	+						
ar-curcumene	MS, IR, NMR	1.0				5.2		
ε-cadinene							46.0	32.0
δ-cadinene						2.6	7.0	7.0
γ-cadinene							8.0	2.0
farnesyl acetate							2.0	9.0
β-caryophyllene epoxide								
(acetylenic compounds)			3.0					
capillin	MS, IR, NMR, UV [7]	0.3						
capillon	MS, IR, NMR, UV [6]	+						
capillen	MS, IR, NMR, UV [5]	80.2						
norcapillen	MS, IR, NMR, UV [3]	0.1						
capillanol	MS, IR, NMR [2]	+						
o-methoxycapillen	MS, IR, NMR [1]	2.0						
(miscellaneous compounds)								
azulene	MS, IR	0.3						
3,5-dimethoxyallyl benzene	MS, IR, NMR	1.8						
phenolic fraction								
phenol	MS, IR, NMR	0.1						
o-cresol	MS, IR, NMR	0.1						
p-cresol	MS, IR, NMR	0.1						
o-ethylphenol	MS, IR, NMR	+						
m-cresol	MS, IR, NMR	+						
p-ethylphenol	MS, IR, NMR	0.1						
eugenol	MS, IR, NMR	0.3						
acidic fraction								
butyric acid	MS	+						
caproic acid	MS	+						
heptanoic acid	MS	+						
caprylic acid	MS	+						
nonanoic acid	MS	+						
capric acid	MS	+						
undecanoic acid	MS	+						
lauric acid	MS	+						
tridecanoic acid	MS	+						
myristic acid	MS	+						
pentadecanoic acid	MS	+						
palmitic acid	MS	0.1						
stearic acid	MS	+						
oleic acid	MS	+						
linoleic acid	MS	+						

(I): *A. fukudo* [13], (II): *A. kurramensis* [9], (III): *A. maritima* [10], (IV): *A. vulgaris* [11], (V): *A. japonica* [12], (VI): *A. piacea* [12].
+Trace amount.

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TERPENGLUCOSIDE AUS *SYNEILESIS ACONITIFOLIA**

FERDINAND BOHLMANN und MICHAEL GRENZ

Institut für Organische Chemie der Technischen Universität Berlin, Strasse des 17, Juni 135, D-1000 Berlin 12, Germany

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Key Word Index—*Syneilesis aconitifolia*; Senecioneae; Compositae; terpene glucosides.

Vertreter der Gattung *Syneilesis* sind bisher noch nicht auf ihre Inhaltssroffe untersucht worden. Die Wurzeln von *S. aconitifolia* enthalten neben Lachnophyllumester (**1**) [1] und dem Acetat **2** [1] ein Glucosid der Summenformel $C_{26}H_{30}O_8$. Im Massenspektrum beobachtet man allerdings nur den Peak M-H_2O. Bei chemischer Ionisation mit Isobuten erhält man jedoch den M + 1-Peak. Die säurekatalysierte Methanolyse liefert D-α-Terpineolmethylether und die Diangelicate von α- und β-D-Glucosemethylether, die als Diacetate getrennt werden. Die ^{1}H-NMR-Spektren des Naturstoffs, des 6-Acetats, des Tetraacetats und der Methylglucoside zeigen, daß der Glucosidrest in 3- und 4-Stellung mit Angelicasäure verestert ist, so daß dem Natursroff die Konstitution **3** zukommt. Die oberirdischen Teile liefern neben Germacren D (**10**) ebenfalls **3** sowie ein weiteres Glucosid, dem die analoge Konstitution **9** zukommen dürfte.

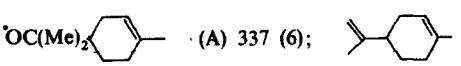
Die hier isolierten Inhaltsstoffe finden in denen der Nachbargattungen keine Analogie. **1** und **2** sind bisher nur aus der Tribus Asteraceae isoliert worden [1], während **3** und **9** unbekannt waren. Angelicaester von Glycosiden von Thymolderivaten haben wir aus *Melampodium divaricatum* isoliert [2].

EXPERIMENTELLES

Pflanzen und Herkunft. Syneilesis aconitifolia Maxim., Tribus *Senecioneae*, angezogen aus Samen vom Botanischen Garten Vilar bei Moskau, Herbar Nr. 76–306.

IR. Beckman IR 9, CCl_4; ^{1}H-NMR. Bruker WH 270, δ-Werte, TMS als innerer Standard, $CDCl_3$; MS. Varian MAT 711 und 311 A, 70 eV, Direkteinlaß. Die frisch zerkleinerten Pflanzenteile extrahierte man mit Ether–Petrol (1:2) und trennte die erhaltenen Extrakte zunächst grob durch SC (Si gel, Akt.-St. II) und weiter durch DC (Si gel, GF 254). Als Laufmittel dienten Ether–Petrol (=E–Pe)-Gemische. 300 g Wurzeln ergaben 30 mg **1**, 2 mg **2** und 100 mg **3** (E–Pe 1:1). 1 kg oberirdische Teile lieferten 15 mg **7**, 20 mg **3** und 15 mg **6**(E–Pe 1:1).

D-α-*Terpineol-β*D-O-*glucopyranosid*-3,4-*diangelicat* (**3**). Zähes farbloses Öl. IR. OH 3610; C=C CO_2R 1730, 1650 cm^{-1}. MS. M^+ —H_2O 462.262 (0.1%) (ber. für $C_{26}H_{38}O_7$ 462.260);—



(A) 337 (6);

(B) 136 (100); $C_4H_7CO^+$ 83 (65). CI- Spektrum (Isobuten). M + 1 481; —A 337; B + 1 137, (M—H_2O nicht vorhanden) 10 mg

* Mitt. 99. in Serie 'Natürlich vorkommende Terpen-Derivate', Mitt. 98. s. F. Bohlmann, D. Ehlers, C. Zdero und M. Grenz (1976) *Chem. Ber.* (im Druck).