(+)-Globulol as a new sesquiterpene alcohol from *Angelica sylvestris* L.

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The chemical composition of the essential oil from the roots of *Angelica sylvestris* L. was studied. The structure of the major component, the tricyclic hydroazulene sesquiterpene (+)-globulol, which was isolated from a natural object for the first time, was established based on X-ray diffraction analysis.

Key words: essential oil analysis, *Angelica sylvenstris* L., sesquiterpenes, globulol, X-ray diffraction analysis.

The useful properties of plants of the *Angelica sylvestris* L. (wild angelica) species belonging to the *Apiaceae* (Umbelliferae) family have long been known.[1—3] The active components of these plants include coumarins,[4—6] flavonoids,[7,8] polyacetylene compounds,[9] and also an essential oil, which was found to contain (+)-limonene, α-pinene, β-pinene, furfural (in roots),[10,11] and the sesquiterpene alcohol bisabolangelone (in fruits).[12] Substantial contents of essential oils (0.4—0.6%) have also been found in other species of the *Angelica* genus, namely, *A. archangelica* L.[13] and *A. glauca*.[14]

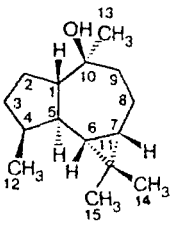
The present communication is devoted to the composition of the essential oils (EO) from the underground organs of individual specimens of *Angelica sylvestris*, occurring at different stages of development (ontogenesis), *viz.*, young nonflowering (virginal) and adult flowering (generative) specimens.

The EO were isolated by the hydrodistillation method modified for small samples.[15] This method can be employed when only 5.0 g of the raw material is available; thus, EO of an individual plant can be isolated and analyzed. The overall content of EO was 0.9—1.3% relative to the air-dry substance of a root.

The isolated compounds were identified by GLC.

The determined component compositions of the EO from the underground organs of *A. sylvestris* are summarized in Table 1. The qualitative composition of the EO in all of the plants studied was roughly the same, and the quantitative ratio of the components (both groups and individual components) varied insignificantly. It should be noted that the amount of sesquiterpene alcohols tends to decrease on passing from young (virginal) to ripe (generative) individuals, whereas the accumulation of hydrocarbons follows the opposite dynamics.

Sesquiterpene alcohols (70—76%) constitute the main group of compounds present in the EO from *A. sylvestris*. The hydroazulene alcohol (+)-globulol (**1**), which was isolated from natural sources for the first time, proved to be the major component of these alcohols and of the EO as a whole.



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The structure of this compound was determined based on physicochemical characteristics and NMR spectra. Thus the melting point and the ^{1}H NMR spectrum of compound **1** virtually coincide with published

Table 1. Contents of the essential oil components in the roots of *Angelica sylvestris* as a function of the age (in percent of the EO weight)

Component	Age state	
	virginal (*v*)	generative (*g*)
Monoterpene hydrocarbons	0.56±0.01	1.01±0.29
α-Pinene	0.39±0.02	0.43±0.13
Δ^3-Carene	—	0.21±0.01
Limonene	0.15±0.02	0.36±0.11
Monoterpene alcohols	0.43±0.07	1.01±0.30
α-Terpineol	0.43±0.07	0.53±0.16
Methyl ether of thymol	—	0.48±0.07
Acetates of monoterpene alcohols	0.74±0.12	1.75±0.45
Bornyl acetate	0.13±0.05	0.43±0.10
Citronellyl acetate	0.28±0.03	0.85±0.21
Geranyl acetate	0.18±0.03	0.24±0.03
Sesquiterpene hydrocarbons	14.65±0.75	21.03±1.59
Caryophyllene	0.95±0.07	1.29±0.21
β-Farnesene	1.57±0.16	1.86±0.22
Germacrene D	3.10±0.16	1.64±0.21
β-Bisabolene	3.93±0.40	4.04±0.51
α-Murolene	—	1.59±0.16
δ,γ-Cadinenes	2.85±0.21	4.27±0.28
Sesquiterpene alcohols	75.90±1.28	70.33±2.59
Nerolidol	3.00±0.26	4.33±0.61
β-Cedrol	1.93±0.12	2.38±0.15
(+)-Globulol	46.36±1.14	35.64±0.66
α-Bisabolol	6.50±0.33	8.58±1.40

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Table 2. Data of the ^{13}C NMR spectra of globulol enantiomers

Atom	δ		Atom	δ	
	(+)-1	(−)-1		(+)-1	(−)-1
C(1)	56.6 d	56.8 d	C(9)	44.4 t	44.4 t
C(2)	26.5 t	26.6 t	C(10)	75.4 s	75.3 s
C(3)	34.4 t	34.3 t	C(11)	19.1 s	19.2 s
C(4)	36.2 d	36.2 d	C(12)	15.6 q	15.5 q
C(5)	39.4 d	39.5 d	C(13)	28.5 q	28.5 q
C(6)	26.0 d	26.0 d	C(14)	15.9 q	15.9 q
C(7)	20.00 d	19.98 d	C(15)	20.01 q	20.01 q
C(8)	28.2 t	28.2 t			

data.[16–18] The optical rotation $[\alpha]_D^{20}$ of the globulol enantiomer that we isolated is +43.13° (*c* 0.8, $CHCl_3$), which coincides, except for the sign, with the value found for (−)-globulol from *Eucalyptus globulus* Labill (blue gum).[19] The carbon chemical shifts in the ^{13}C NMR spectrum of globulol isolated from *Angelica sylvestris* roots and *Eucalyptus globulus* leaves are given in Table 2. It should be noted that the difference between the carbon chemical shifts of enantiomers is slight and falls within the experimental error; the greatest difference is observed for the C(1) and C(10) atoms (Δδ 0.2).

The X-ray diffraction data confirm the structure of (+)-globulol isolated from *A. sylvestris* roots. The molecular structure is shown in Fig. 1. The bond lengths in the molecule are close to the statistical mean values.[20] The five- and seven-membered rings of the molecule are *trans*-joined. The cyclopentane moiety has an envelope shape; the C(4) atom deviates by −0.642(9) Å from the plane containing the other atoms of this ring (with a maximum deflection of 0.016(3) Å). The conformation of the seven-membered ring is one of the most stable conformations of cycloheptane and is defined as a chair (its energy is only ~4 kJ mol^{-1} higher than that of the twist-chair conformation).[21] We found about a dozen structures of the aromadendrane type in the Cambridge Structural Data Bank.[22] Apoaromadendrone and (+)-4-hydropalustrol[23] are the closest analogs of the compound studied here. Despite the difference in the side groups, the geometry and conformations of the rings in the three compounds are generally similar.

In addition to (+)-globulol, the EO from *A. sylvestris* contain sesquiterpene alcohols β-cedrol, α-bisabolol, and nerolidol (Table 1). These compounds were found in *A. sylvestris* for the first time.

The second largest group of compounds in the EO is the group of sesquiterpene hydrocarbons (15—21%), comprising β-bisabolene, δ,γ-cadinenes, germacrene, β-farnesene, α-murolene, and caryophyllene.

Slight amounts of other compounds were also identified: monoterpene hydrocarbons (α-pinene, Δ^3-carene, and limonene), monoterpene alcohols (α-terpineol and methyl ether of thymol), and monoterpene alcohol acetates (bornyl acetate, citronellyl acetate, and geranyl acetate).

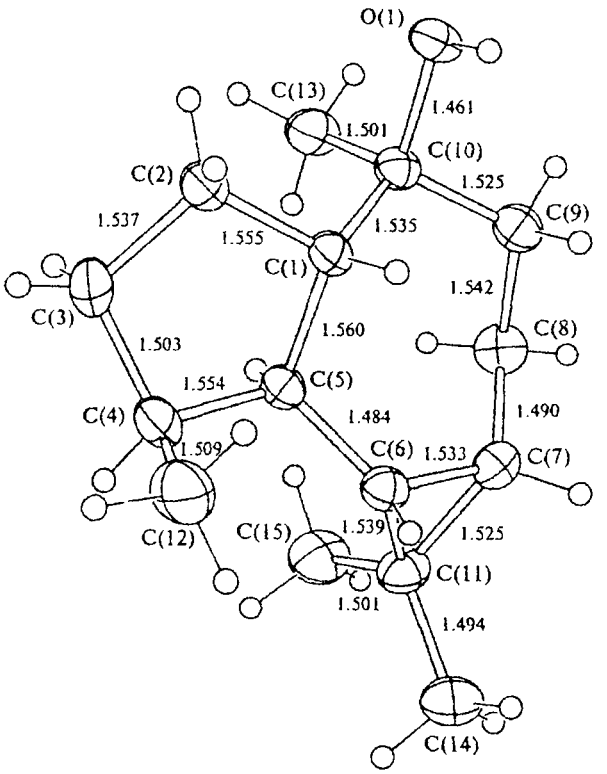

Fig. 1. Overall view of the molecule of globulol from *A. sylvestris*. The errors of determination of bond lengths are within 0.004—0.007 Å.

Thus, the composition of the EO of the underground organs of *A. sylvestris* was studied in detail. Eighteen individual compounds including 8 monoterpene and 10 sesquiterpene derivatives were isolated and identified. The sesquiterpene alcohol (+)-globulol was isolated for the first time from natural objects.

Experimental

The material for the study was gathered in large-grass meadows at the axial part of the Salairskii mountain range (Novosibirsk region, Toguchinskii area). The underground organs of *A. sylvestris* were marked and each root was dried separately. To study the dynamics of accumulation of EO components (procedure ***A***), 25 virginal and 25 generative individual plants were gathered. Some of the roots were gathered and dried together without grading into age states in order to isolate individual substances (procedure ***B***).

NMR spectra were recorded on a Bruker AC 200 instrument (200.13 MHz for ^1H and 50.32 MHz for ^{13}C) in $CDCl_3$ solutions. The signal multiplicity in the ^{13}C NMR spectra was determined using the standard procedure for recording the spectra in the JMODXH mode. The X-ray diffraction study of (+)-globulol **1** was carried out on a Syntex $P2_1$ diffractometer using Cu-Kα radiation and a graphite monochromator.

The optical rotation was determined on a Polamat-A instrument at a wavelength λ of 594 nm.

The refraction index was determined on a URF-454 B refractometer. GLC was carried out on a Chrom-41 instrument with a flame ionization detector using nitrogen as a carrier gas (flow rate 2 mL min^{-1}), a 50 m×0.2 mm capillary column, and polymethylsiloxane as the stationary phase; the evaporator temperature was 160 °C and the column temperature was 60 °C. Compounds were identified based on the retention time and by adding reference compounds to the EO (procedure *A*) or to fractions of the pentane extract (procedure *B*). The products were isolated by column chromatography on KSK silica gel (SiO_2, 100—250 mm) (pentane, pentane—Et_2O 50 : 1, 20 : 1, 10 : 1).

Isolation of EO from the underground parts of *A. sylvestris*. *A.* An individual air-dry root was powdered, weighed, and placed in a 1-L flask, either whole or as a portion (4.5—7.0 g) if the root was too large. Then 600 mL of water was added to the flask, a setup was assembled as described previously[15] (the setup was modified by the addition of one more reflux condenser), and the contents were heated to boiling. The extraction was carried out for 2.5 h after the first drops of condensate had appeared in the receiver. The resulting essential oil was weighed in a glass ampule to determine the weight proportion of the EO in the air-dry root and analyzed by GLC (the ampules with EO samples were stored sealed). The results are presented in Table 1. The general characteristics of the EO from the underground organs of *A. sylvestris* were as follows: d_4^{20} 0.9535; n_D^{20} 1.494; $[\alpha]_D^{24}$ +20.03° (*c* 7.6, $CHCl_3$); the essential number is 16.648; the acid number is 6.827.

B. A 1400 g sample of crushed air-dried roots of *A. sylvestris* was extracted with pentane (2×900 mL). The combined pentane extract was concentrated to give 14.6 g (1.04%) of an oil, which was distilled *in vacuo* to give three fractions, 1.25 g (b.p. 90—140 °C), 3.58 g (b.p. 140—170 °C), and 3.66 g (b.p. 170—200 °C). The bottom residue (6.12 g) was not analyzed. The distillation products were additionally separated by column chromatography on SiO_2. Elution of the first and second fractions by pentane gave monoterpene and sesquiterpene hydrocarbons, respectively; subsequent elution with ether gave acetates of monoterpene alcohols (fraction 1) and sesquiterpene alcohols (fraction 2); the latter were additionally separated by gradient elution. The predominant component of the second fraction was (+)-globulol **1**, which crystallized in the eluent as long needle crystals. The mother liquor was separated from the crystalline precipitate and analyzed by GLC. Recrystallization of the precipitate from EtOH afforded pure (+)-globulol **1**, 1.57 g (43.85% of the fraction weight, 10.75% of the weight of the pentane extract, and 0.11% of the weight of the air-dry roots). (+)-Globulol (**1**) thus isolated had the following characteristics: m.p. 88.0—88.5 °C (from EtOH), $[\alpha]_D^{20}$ +43.13° (*c* 0.8, $CHCl_3$). Gradient elution of the third fraction also gave sesquiterpene alcohols.

Isolation of (−)-globulol from *Eucalyptus globulus* Labill. leaves. The eucalyptus leaves were gathered in the Botanical Garden of the Siberian Branch of the RAS. A 300-g portion of crushed air-dry *E. globulus* leaves was extracted under the same conditions as underground organs of *A. sylvestris* (pentane, 2×200 mL). The combined pentane extract was concentrated (weight 2.43 g) and separated by column chromatography. Only the fraction containing (−)-globulol was collected (TLC monitoring by comparison with (+)-globulol) to give 0.04 g (1.6% of the extract weight and 0.01% of the weight of the leaves). After evaporation of the solvent, the residue, (−)-globulol, crystallized, m.p. 88.5 °C (from EtOH), $[\alpha]_D$ −48.30° (*c* 2.45, $CHCl_3$).

X-ray diffraction study of (+)-globulol. The crystals are trigonal, a = 13.438(5), b = 13.438(5), c = 6.807(3) Å, V = 1064.5(7) $Å^3$, space group $P3_2$, Z = 3, $C_{15}H_{26}O$, μ = 0.471 mm^{-1}, d_{calc} = 1.041 g cm^{-3}. The intensities of 1154 independent reflections with $2\theta < 120°$ were measured by the $\theta/2\theta$-scanning method. The absorption correction over the crystal cutting was applied (transmission 0.94—0.97). The structure was solved by the direct method using the SHELX-86 program. The positions of the H atoms were determined geometrically. The final refinement of the structure parameters was performed by the least-squares method in the full-matrix anisotropic approximation (the H atoms were not included in the refinement) by the SHELXL-93 program to wR_2 = 0.1198 for F^2 and R = 0.0486 for 839 $F_0 > 4\sigma$ (145 parameters were refined). The coordinates and the equivalent thermal factors for nonhydrogen atoms are presented in Table 3.

Table 3. Coordinates (×10^4) and equivalent thermal factors (×10^3/$Å^2$) of nonhydrogen atoms in (+)-globulol

Atom	*x/a*	*y/b*	*z/c*	U_{equiv}
C(1)	841(3)	8217(3)	7652(6)	44(1)
C(2)	2011(4)	8915(4)	8733(8)	64(1)
C(3)	2466(4)	8078(5)	9018(9)	81(2)
C(4)	1967(5)	7245(4)	7340(8)	72(2)
C(5)	713(3)	7012(3)	7282(6)	46(1)
C(6)	34(3)	6430(3)	5499(6)	48(1)
C(7)	−1244(3)	6064(3)	5413(7)	51(1)
C(8)	−1790(4)	6302(3)	7118(7)	58(1)
C(9)	−1286(3)	7595(4)	7524(7)	54(1)
C(10)	−179(3)	8189(3)	8719(6)	45(1)
C(11)	−887(4)	5151(3)	5392(7)	59(1)
C(12)	2610(4)	7723(5)	5439(10)	91(2)
C(13)	−352(4)	7716(4)	10767(6)	64(1)
C(14)	−1013(5)	4565(4)	3467(9)	83(2)
C(15)	−1122(5)	4377(4)	7130(8)	82(2)
O(1)	92(2)	9376(2)	9008(4)	50(1)

The absolute configuration of (+)-globulol could not be established unambiguously, the Flack parameter being equal 0.1(6). Note that refinement of the enantiomeric structure results in a somewhat worse Flack parameter (0.3(6)).

In the crystal, the globulol molecules are linked through hydrogen bonds, O(1)—H...O(1) (O—H 0.82 Å, O(1)...O(1) 2.76 Å, H...O(1) 1.950 Å, O(1)—H...O(1) 169°), to give infinite chains twisted around a 3_2 threefold axis.

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References

1. *Rastitel'nye resursy SSSR. Tsvetkovye rasteniya, ikh khimicheskii sostav, ispol'zovanie* [*USSR Plant Resources. Flowering Plants, Their Chemical Composition, and Application*], Nauka, Leningrad, 1988, 71 (in Russian).
2. Ya. I. Khadzhai and V. E. Sokolova, *Farmakologiya i toksikologiya* [*Pharmacology and Toxicology*], 1960, **23**, 37 (in Russian).
3. M. M. Glukhov, *Medonosnye rasteniya* [*Melliferous Plants*], Meditsina, Moscow, 1974, 294 pp. (in Russian).
4. L. Horhammer, H. Wagner, and W. Eyrich, *J. Naturforsch.*, 1963, **18**, 639.
5. Y. Fujita, *J. Japan Bot.*, 1966, **43**, 65.

6. A. G. Valutskaya, I. N. Gus'kova, and E. V. Tyurina, *Rastit. resursy* [*Plant Resources*], 1972, **8**, 547 (in Russian).
7. L. Horhammer, H. Wagner, and H. Gotz, *Arch. Pharm.*, **291**, 44.
8. E. Yu. Vinokurova, in *Novye lekarstvennye preparaty iz rastenii Sibiri i Dal'nego Vostoka (Tez. dokl.)* [*New Medicinal Preparations from Siberian and Far Eastern Plants*], Tomsk, 1989, 33 (in Russian).
9. F. Bohlmann, M. Grenz, and Ch. Zdero, *Naturally Occurring Acetylenes*, London, New York, 1973, 547 p.
10. A. F. Costa and J. C. do Vale, *Bol. Escola Farm. Univ. Coimbra*, 1954, **13/14**, 68; *Chem. Abstrs.*, 1959, **53**, 5419g.
11. P. Rovesti, in *Riv. Ital. Essenze, Profumi Piante Offic., Oill Vegetali, Saponi*, 1954, **36**, 162; Chem. Abstrs., 1954, **48**, 9718b.
12. L. Novotny, Z. Samek, and F. Sorm, *Tetrahedron Lett.*, 1966, **30**, 3541.
13. A. Chatterjes, A. Chapravarty, and R. Mukherjee, *J. Ind. Chem. Soc.*, 1967, **44**, 110; *Chem. Abstrs.*, 1967, **67**, 5631z.
14. S. S. Chandhary, P. Vistuva, and K. L. Handa, *Indian Soap J.*, 1959, **24**, 288; *Chem. Abstrs.*, 1959, **53**, 22753.
15. *Metody biokhimicheskogo issledovaniya rastenii* [*Methods for Biochemical Investigation of Plants*], Ed. A. I. Ermakov, Agropromizdat, Leningrad, 1987, 430 pp. (in Russian).
16. J. A. Marshall and J. A. Ruth, *J. Org. Chem.*, 1974, **39**, 1971.
17. G. Büchi, S. W. Show, T. Matsuura, T. L. Popper, H. H. Rennhard, and M. Schach v. Wittenan, *Tetrahedron Lett.*, 1959, No. 6, 14.
18. R. Faure, A. R. P. Ramanoelina, O. Rakotonirainy, J. P. Bianchini, and E. M. Gaydou, *Magn. Reson. Chem.*, 1991, **29**, 969.
19. L. Dolejs, O. Motl, M. Soucek, V. Herout, and F. Sorm, *Coll. Czech. Chem. Commun.*, 1960, **25**, 1483.
20. F. H. Allen, O. Kennard, D. G. Watson, L. Bramer, A. G. Orpen, and R. Taylor, *J. Chem. Soc., Perkin Trans. 2*, 1987, 1.
21. U. Burkert and N. L. Allinger, *Molecular Mechanics*, Am. Chem. Soc., Washington, 1982, 364 pp.
22. F. H. Allen and O. Kennard, *Chemical Design Automation News*, 1993, **8**, 31.
23. A. R. Overbeek, G. J. Olthof, N. Putten, and H. Schenk, *Cryst. Struct. Commun.*, 1979, **8**, 473; J. C. Braekmann, D. Daloze, C. Stoller, and J. P. Declercq, *Bull. Soc. Chem. Belg.*, 1989, **98**, 869; *Chem. Abstrs.*, 1989, **112**, 198822s.

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