

**ЭФИРОМАСЛИЧНЫЕ И ЛЕКАРСТВЕННЫЕ РАСТЕНИЯ.
БИОХИМИЯ РАСТЕНИЙ**

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SCREENING STUDIES OF WOODY PLANTS AS SOURCES OF ESSENTIAL OILS**Yurii Vladimirovich Plugatar¹, Oksana Mikhailovna Shevchuk¹,
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The article presents the results of screening studies of raw materials of woody plants growing in humid subtropical conditions (“Sochi National Park”, “Arboretum” park) for the presence of essential oil. The mass fraction of essential oil was determined in fresh raw materials by hydrodistillation on Ginsberg apparatuses. The component composition of essential oils was established using a hardware and software complex based on the chromatograph “Chromatek-Crystal 5000.2”. The raw materials (freshly harvested leaves) of 52 species and cultivars of wood introduced species were studied. Essential oil was detected in 39 species. A high yield of essential oil (from 0.5% of the wet weight) was noted in *Melaleuca armillaris* (Sol. ex Gaertn.) Sm. (0.51%), *Laurus nobilis* L. (0.53%), *Callistemon salignus* cv. ‘Mauve Mist’ (0.54%), *Eucalyptus globulus* Labill. (0.57%), *Eucalyptus niphophila* Maiden & Blakely (0.66%), *Callistemon linearifolius* (Link) DC. (0.70%), *C. coccineus* F. Muell. (0.78%), *C. viminalis* (Sol. ex Gaerth.) G. Don (0.80%), *Thuja plicata* cv. ‘Zebrina’ (0.85%), *Callistemon phoeniceus* Lindl. (0.86%), *Cinnamomum camphora* (L.) J. Presl (1.2%), *C. glanduliferum* (Wall.) Meisn. (1.6%), *Eucalyptus cinerea* F. Muell. ex Benth. (1.14%), *E. globulus* subsp. *globulus* (1.67%). The highest yield of essential oil among the studied species was noted in *Umbellularia californica* (Hook. & Arn.) Nutt. – 5.10% of the wet weight.

Key words: woody plants; essential oil; mass fraction; component composition; humid subtropics**Introduction**

Essential oils are a mixture of volatile fragrant substances produced by plants during life. Being one of the varieties of highly effective biologically active substances, they have a wide spectrum of action: antioxidant, antimicrobial, expectorant, diuretic, antispasmodic, anti-inflammatory, etc. [2, 6].

More than 1000 compounds have been isolated from essential oils, which greatly expands the possibilities of their use in various industries [16]. The composition of essential oils includes hydrocarbons, alcohols, esters, ketones, lactones, aromatic compounds, etc. It is the chemical composition – the ratio of those or other components that determine the value of an essential oil and the directions of its use. To date, the properties of essential oils of about 5,000 plants have been studied, but about 300 are suitable for commercial use. Along with traditional essential oil plants, much attention is paid to studying the accumulation of secondary metabolites in the raw materials of woody plants and determining the applications of their biological activities (antioxidant, antimicrobial, anti-inflammatory, antispasmodic, etc.) [6]. The use of raw materials of woody plants has a number of advantages, among which: a significant increase in biomass; the possibility of using various types of raw materials (bark, wood, green biomass, fruits/cones) in the process of taking care of green spaces and logging, as well as when laying industrial plantations – their longevity and minimal care in the subsequent period. Essential oils of woody plants of the genera *Citrus* L., *Eucalyptus* L'hér.,

Results and discussion

When collecting plant raw materials in autumn, trace amounts of essential oil (up to 0.001%) were detected in five of the studied species: *Beilschmiedia roxburghiana* Nees, *Callistemon citrinus* (Curtis) Skeels, *Citrus limon* (L.) Osbeck and *C. sinensis* (L.) Osbeck, *Lantana camara* L. In nine species, essential oil was not obtained by hydrodistillation: *Cinnamomum daphnoides* Siebold & Zucc., *Lindera benzoin* (L.) Blume, *Lindera setchuenensis* Gamble, *Lophostemon confertus* (R.Br.) Peter G. Wilson & J.T. Waterh., *Machilus thunbergii* Siebold & Zucc., *Melaleuca styphelioides* Sm., *Platycladus orientalis* (L.) Franco, *Schinus dependens* var. *longifolia* Fenzl. ex Engler in Martius and *Syzygium paniculatum* Gaerth (fig. 1-6).



Fig. 1 Inflorescence of *Melaleuca armillaris* (Sol. ex Gaertn.)



Fig. 2 Inflorescences of *Callistemon coccineus* F. Muell.



Fig. 3 Platycladia of *Thuja plicata* cv. *Zebrina*



Fig. 4 Inflorescences of *Callistemon phoeniceus* Lindl.



Fig. 5 Leaves of *Eucalyptus cinerea* F. Muell. ex Benth



Fig. 6 Leaves of *Umbellularia californica* (Hook. & Arn.) Nutt.

A high yield (from 0.5% of the wet weight) of essential oil was noted in *Melaleuca armillaris* (Sol. ex Gaertn.) Sm. (fig. 1) (0.51%), *Laurus nobilis* L. (0.53%), *Callistemon salignus* cv. 'Mauve Mist' (0.54%), *Eucalyptus globulus* Labill. (0.57%), *E. niphophila* Maiden & Blakely (0.66%), *Callistemon linearifolius* (Link) DC. (0.70%), *C. coccineus* F. Muell. (0.78%) (fig. 2), *C. viminalis* (Sol. ex Gaerth.) G. Don (0.80%), *Thuja plicata* cv. 'Zebrina' (0.85%) (fig. 3), *Callistemon phoeniceus* Lindl. (0.86%) (fig. 4), *Cinnamomum camphora* (L.) J. Presl (1.2%), *C. glanduliferum* (Wall.) Meisn. (1.6%), *Eucalyptus cinerea* F. Muell. ex Benth. (1.14%) (fig. 5), *E. globulus* subsp. *globulus* (1.67%). The maximum yield of essential oil among all studied species was noted in *Umbellularia californica* (Hook. & Arn.) Nutt. (fig. 6) – 5.10% of the raw mass (13.04% by absolutely dry weight) (table).

Table

Dynamics of essential oil content and its component composition in the raw material of woody introduced species in humid subtropics (Park «Arboretum», Sochi National Park)

№	Species, cultivar	Yield of essential oil from wet weight, %		The main components of essential oil, %
		October, 2021 г.	May, 2022 г.	
1	2	3	4	5
Cupressaceae				
1	<i>Thuja koraiensis</i> cv. Aurea	0.20	*	α -thujone – 46.06 <i>trans</i> -sabinyl acetate – 12.82
2	<i>Thuja occidentalis</i> cv. Ellwangeriana Aurea	0.65	*	α -thujone – 48.05 β -thujone – 17.55
3	<i>Thuja plicata</i> cv. Zebrina	0.85	*	α -thujone – 84.09 β -thujone – 6.59
4	<i>Thuja standishii</i> (Gordon) Carriere	0.20	*	α -thujone – 58.29 fenchone – 12.27
5	<i>Thuja sutchuenensis</i> Franch.	0.14	*	α -thujone – 40.70 <i>trans</i> -sabinyl acetate – 11.79 hibaene – 10.8
Anacardiaceae				
6	<i>Schinus lentiscifolius</i> Marchand	0.40	*	β -pinene – 16.07 α -pinene – 12.82 bicyclogermacren – 12.83
Lauraceae				
7	<i>Apollonias barbujana</i> subsp. <i>barbujana</i> (Cav.) Bornm.	0.074	*	linalool – 28.53 methyl eugenol – 9.6
8	<i>Cinnamomum camphora</i> (L.) J. Presl	1.2	1.57	camphor – 72.23 β -selinene – 5.79
9	<i>Cinnamomum chekiangense</i> Nakai	0.30	0.34	E-nerolidol – 43.81 (E)-atlantone – 19.48
10	<i>Cinnamomum glanduliferum</i> (Wall.) Meisn.	1.6	1.00	camphor – 40.44 bornyl acetate – 8.29
11	<i>Cinnamomum sieboldii</i> Meisn.	0.05	0.10	(E)-citral – 23.70 (Z)-citral – 12.86
12	<i>Laurus canariensis</i> Willd.	0.19	0.50	1,8-cineole – 42.22 α -terpinyl acetate – 16.70
13	<i>Laurus nobilis</i> L.	0.53	0.24	1,8-cineole – 35.69 α -terpinyl acetate – 10.26
14	<i>Laurus nobilis</i> f. <i>salicifolia</i>	0.27	0.04	1,8-cineole – 30.02 linalool – 21.40 α -terpinyl acetate – 12.18
15	<i>Lindera angustifolia</i> W.C. Cheng	0.31	0.43	(Z)-ocimene – 45.01 β -eudesmol – 10.71

Continuation of the table

1	2	3	4	5
16	<i>Umbellularia californica</i> (Hook. & Arn.) Nutt. (Рис. Fig. К)	5.10	1.60	umbellulone – 36.43 1,8-cineole – 17.29
Myricaceae				
17	<i>Myrica cerifera</i> L.	0.38	0.016	1,8-cineole – 26.57 α -pinene – 14.11
18	<i>Callistemon coccineus</i> F. Muell.	0.78	0.24	1,8-cineole – 85.85
19	<i>Callistemon linearifolius</i> (Link) DC.	0.70	0.74	1,8-cineole – 74.67 α -terpineol – 11.94
20	<i>Callistemon pallidus</i> (Bonpl.) DC.	0.25	0.17	1,8-cineole – 73.61 α -terpineol – 14.77
21	<i>Callistemon phoeniceus</i> Lindl.	0.86	0.20	1,8-cineole – 81.70
22	<i>Callistemon rigidus</i> R. Br.	0.10	0.08	1,8-cineole – 68.93 α -terpineol – 16.53
23	<i>Callistemon salignus</i> cv. Mauve Mist	0.54	0.49	1,8-cineole – 77.79 α -terpineol – 11.49
24	<i>Callistemon speciosus</i> (Sims) Sweet	0.43	0.14	1,8-cineole – 65.56 α -pinene – 15.08
25	<i>Callistemon viminalis</i> (Sol. ex Gaerth.) G. Don	0.80	0.45	1,8-cineole – 62.84 α -terpineol – 9.32
26	<i>Eucalyptus cinerea</i> F. Muell. ex Benth.	1.14	1.08	1,8-cineole – 67.77 α -terpinyl acetate – 17.05
27	<i>Eucalyptus globulus</i> Labill.	0.57	0.80	1,8-cineole – 65.92 α -pinene – 9.19
28	<i>Eucalyptus globulus</i> subsp. <i>globulus</i>	1.67	1.00	1,8-cineole – 84.05
29	<i>Eucalyptus niphophila</i> Maiden & Blakely	0.66	0.55	1,8-cineole – 69.95 α -terpineol – 9.25
30	<i>Feijoa sellowiana</i> (O. Berg) O. Berg	0.013	0.028	β -caryophyllene – 10.41 (+)-spathulenol – 10.36
31	<i>Leptospermum scoparium</i> J.R. Forst. & G. Forst.	0.13	0.05	geranyl acetate – 17.93 β -eudesmol – 11.03 α -eudesmol – 10.21
32	<i>Leptospermum scoparium</i> f. <i>rubra</i> J.R. Forst. & G. Forst.	0.20	0.13	neointermedeol – 14.62 β -selinene – 10.86 α -selinene – 10.34
33	<i>Melaleuca armillaris</i> (Sol. ex Gaertn.) Sm.	0.51	0.38	methyl eugenol – 97.51
34	<i>Melaleuca ericifolia</i> Sm.	0.059	0.53	1,8-cineole – 56.09 α -pinene – 6.12
35	<i>Myrrhinium atropurpureum</i> Schott	0.051	0.09	α -pinene – 19.41 viridiflorol – 11.43
Rutaceae				
36	<i>Citrange</i> cv. Sochinsky	0.072	*	linalool – 27.32 γ -terpinene – 19.82 (Z)-ocimene – 9.96
37	<i>Citrus paradisi</i> cv. Sochinsky	0.03	*	linalool – 63.71 terpinen-4-ol – 3.97 α -terpineol – 3.90
Verbenaceae				
38	<i>Aloysia chamaedryfolia</i> Cham.	0.11	*	bicyclogermacren – 19.14 (+)-spathulenol – 8.72 (-)- β -elemene – 8.68

*Raw materials of these species have not been studied

The dynamics of the mass fraction of essential oil was studied in the raw materials of 28 plant species. In 17 species, the content of essential oil in the leaves was higher in autumn, in 9 species there was a slight increase in the content of essential oil in spring. None of the studied species and cultivars with a low mass fraction of essential oil showed high results in another season. The dynamics of the mass fraction of essential oil was practically absent in *Callistemon citrinus* (Curtis) Skeels, *C. rigidus* R. Br., *C. linearifolius* (table).

The change in the essential oil content in the freshly harvested leaves of *Umbellularia californica* is significant, which decreased from 5.10% in autumn to 1.60% in spring. Similarly, the essential oil content of *Myrica cerifera* L. and *Callistemon coccineus* decreased by more than three times in the spring. The opposite situation is with *Melaleuca ericifolia* Sm., which has essential oil content 10 times higher in spring compared to autumn (0.53% and 0.059%, respectively).

The combination of components determines the aroma of the essential oil and possible directions of practical use. The main components of the essential oil of the genus *Thuja* L. representatives are thujones (α -thujone and β -thujone), the mass fraction of which, as well as its content, varies significantly among the studied species and cultivars. The highest total thujone content (more than 90%) was found in the essential oil of *Thuja plicata* cv. 'Zebrina'. This cultivar is also characterized by the highest yield of essential oil (0.85%). There is quite contradictory information about the applications of biological activity of thujones belonging to the group of chemical compounds of terpenoids (bicyclic terpene ketones). There are data on the poisonous properties of thujones [9, 19]. At the same time, *Th. plicata* essential oil has phytoncidal activity, a tonic effect, and is effective when used in medicine as an expectorant and antiviral agent [14, 22]. The high content of α - and β -thujones determines the high antibacterial (MIC values 0.50–1.25 mg/ml) and antifungal (MIC 0.87–1.12 mg/ml) activities of thuja essential oil [36].

In this regard, *Thuja plicata* cv. 'Zebrina' may be of considerable interest as a source of valuable essential oil of pharmacological prospects. In addition, an earlier study of the component composition of the essential oil of *Th. plicata* and *Th. plicata* cv. 'Zebrina' in the conditions of the Southern coast of the Crimea and the Republic of Abkhazia, indicates a consistently high content of α - and β -thujones (up to 90%) in the raw materials of this cultivar [22].

In 16 studied species and cultivars, the main component of essential oil is monocyclic terpene 1,8-cineole (eucalyptol). The highest content of eucalyptol was found in representatives of the *Myrtaceae* family of the genera *Callistemon* L., *Eucalyptus* L., *Laurus* L., *Melaleuca ericifolia*, *Myrica cerifera* L., in particular, in the essential oil of *Callistemon salignus* cv. 'Mauve Mist', the mass fraction of 1,8-cineole reaches 77.79%, *C. phoeniceus* Lindl. - 81.70%, *C. coccineus* - 85.85% and *Eucalyptus globulus* subsp. *globulus* - 84.05%. The substantial content of eucalyptol causes antiseptic properties, high antimicrobial activity of essential oil against gram-positive and gram-negative bacteria (*Streptococcus pyogenes* Rosenbach, *Staphylococcus aureus* Rosenbach and *Pseudomonas aeruginosa* Migula) [30], is the basis for its use in pharmacology and the perfume and cosmetics industry.

The high content of eucalyptol in combination with α -terpinyl acetate is a feature of the essential oil of species and cultivars of the genus *Laurus* L. The maximum mass fraction of these components was noted in *L. canariensis* (42.22% and 16.7%, respectively). The essential oil of *L. canariensis* is also characterized by sabinene (7.59%); *L. nobilis* - α -terpineol (7.33%) and *L. nobilis* cv. *Salicifolia* - linalool and methyl eugenol (21.40% and 11.54%, respectively).

The main component in essential oil from the leaves of *Cinnamomum camphora* (L.) J. Presl and *C. glanduliferum* (Wall.) Meisn. is ketone camphor - 72.23% and 40.44%, respectively. Camphor has a wide range of biological effects, including antibacterial,

antioxidant, anti-inflammatory and insecticidal. In folk medicine, essential oil with a high content of camphor is used to treat bacterial and fungal infections [17, 20-23].

Two other representatives of the genus *Cinnamomum* accumulate essential oil of the perfume application. In the essential oil of *Cinnamomum sieboldii* Meisn. (fig. 7) the main components are monoterpene acyclic aldehyde citral in the form of two isomers of E- and Z-citrals (geranial and neral) (the total mass fraction is 36.56%). Citral has a wide industrial application, can be used as a preservative, in the food, soap and perfume and cosmetic industries; exhibits antimicrobial activity. Different isomers of citral have different aromas. The investigated essential oil of *Cinnamomum sieboldii* contains 23.7% geranium, which gives the essential oil a lemon aroma, and neral adds fruity notes [10, 26].

The essential oil of *Cinnamomum chekiangense* Nakai, due to the high content of sesquiterpenic alcohol E-nerolidol, has a woody-floral-citrus aroma. Nerolidol is used as a component of perfume compositions and food essences, as a fragrance fixative; causes antimicrobial, antioxidant, antiparasitic, skin-repellent, analgesic, anti-inflammatory and anti-cancer [3] action of essential oil. Another major component of this type of essential oil – E-atlantone - has a mucolytic effect [4].

The main component of the essential oil of *Leptospermum scoparium* J.R. Forst. & G. Forst. is geranyl acetate (17.93%), which has a floral-fruity aroma with hints of rose and geranium, and eudesmol, also present in the essential oil, gives the oil a slightly sweet woody note (Sharangi, 2021). Special aroma of essential oil of *L. scoparium* f. *rubra* (fig. 8) is woody-earthy with patchouli notes. Such aroma is due to the presence of neointermedeol (14.62%) and β - and α -selinenes (10.86% and 10.34%, respectively) in the essential oil. The last two compounds are sesquiterpenes, which are also characterized by antiseptic action [29].



Fig. 7 Leaves of *Cinnamomum sieboldii* Meisn.



Fig. 8 Inflorescences of *Leptospermum scoparium* f. *rubra* J. R. Forst. & G. Fors.

Essential oils of *Schinus lentiscifolius* Marchand, and to a lesser extent (due to the lower essential oil content), *Myrrhinium atropurpureum* Schott, are characterized by the presence of α - and β -pinenes. Pinene – bicyclic monoterpene is a feature of the essential oil of many coniferous and deciduous plants. The high content of this component causes a wide range of

biological activity of plant raw materials, manifested in anticoagulant, antitumor, antimicrobial, antimalarial, antioxidant, anti-inflammatory, anti-leishmaniasis and analgesic effects [9, 27, 38]; α -pinene has antibacterial activity against a dangerous antibiotic-resistant strain of *Staphylococcus aureus* MRSA and *Campylobacter jejuni* bacterium that causes campylobacteriosis [14]. β -pinene has disinfectant and insecticidal properties [37], it is used in the perfume industry for the synthesis of fragrant substances [33].

Apollonias barbujana subsp. *barbujana* (Cav.) Bornm., *Citrange* cv. 'Sochinsky' and *Citrus paradisi* cv. 'Sochinsky' can be distinguished as sources of linalool. It belongs to the group of chemical compounds of terpenoids and has anticonvulsant, bactericidal and antispasmodic properties [7, 9]. Linalool has a floral-citrus aroma, and is promising for use in the food and perfume and cosmetics industry [11].

The main components of *Feijoa sellowiana* (O. Berg) O. Berg are two sesquiterpenoids – β -caryophyllen and alcohol (+)-spathulenol. Caryophyllen is used to compose perfume compositions, in the production of perfumes for soap, cosmetics, in the synthesis of some fragrant substances, and spathulenol gives the essential oil a bitter-spicy aroma [15].

The main components of *Lindera angustifolia* W.C. Cheng are monoterpene hydrocarbon (Z)-ocimene (45.01%) and bicyclic sesquiterpenic alcohol β -eudesmol (10.71%). (Z)-ocimene often occur naturally in the form of mixtures of various forms. The mixture, like pure compounds, is oils with a pleasant smell. They are used in perfumes because of their sweet herbal aroma and are believed to have antifungal properties. β -eudesmol has a slightly sweet and predominantly woody aroma [12, 29].

Methyl eugenol is the methyl ester of eugenol, found in the essential oil of *Melaleuca armillaris* (Sol. ex Gaertn.) Sm. in a fairly large amount – 97.51%. Methyl eugenol is a common phenylpropanoid found in many plant species and has a clove odor. The literature provides information about its sedative, antispasmodic and antitussive properties, as well as antifungal activity [7, 9, 32]. It is used in the food industry (as a flavoring agent) and the perfumery and cosmetics industry.

The essential oil of *Aloysia chamaedryfolia* Cham. contains sesquiterpenes bicyclogermacrene (19.14%), (+)-spathulenol (8.72%) and β -elemene (8.68%), which gives the oil an earthy mushroom aroma, which makes its use in perfumery unpromising. However, these components have anti-inflammatory, antibacterial, antifungal properties and high antioxidant activity [25, 28].

Of particular interest is *Umbellularia californica* (Hook. & Arn.) Nutt., accumulating in the autumn a significant amount of essential oil (above 5%) with the major component umbellulone (mass fraction 36.43%). Umbellulone is a monoterpene ketone that has a negative effect on the human central nervous system, provokes coordination disorders and hypotension [21]. There are data on the larvicidal properties of this compound [31], which allows us to consider the essential oil *U. californica* is promising for the development of biological methods to combat harmful insects.

Conclusion

Thus, the presence of essential oil was found in 38 species and cultivars of tree introducents growing in the collection of "Arboretum" park ("Sochi National Park"), in the humid subtropical climate of the Black Sea coast of the Caucasus. 15 species have a high content of essential oil (above 0.5% of the wet weight).

The highest yield of essential oil among the studied species was noted in *Umbellularia californica* (Hook. & Arn.) Nutt. – 5.10% of the wet weight (13.04% by absolutely dry weight). The study of the dynamics of the accumulation of essential oil indicates that in most of the studied species, the content of essential oil in spring is either significantly lower or within the margin of error compared with autumn.

It is established that *Thuja plicata* cv. 'Zebrina' may be of considerable interest as a source of valuable essential oil of pharmacological application, due to the high content of α - and β -thujones.

Callistemon salignus cv. 'Mauve Mist', *C. phoeniceus* Lindl., *C. coccineus* and *Eucalyptus globulus* subsp. *globulus* are characterized by containing promising essential oil that can be used as pharmacological and perfumery-cosmetic products, due to a significant mass fraction of 1,8-cineole (eucalyptol) (84.05%). The high content of eucalyptol in combination with α -terpinyl acetate is characteristic of the essential oil of species and cultivars of the genus *Laurus* L. The maximum mass fraction of these components was noted in *L. canariensis* – (42.22% and 16.7%, respectively). The source of camphor is *Cinnamomum camphora* (L.) J. Presl and *C. glanduliferum* (Wall.) Meisn.

The essential oil of *Schinus lentiscifolius* Marchand can be used as sources of pinene (α - and β -pinene). The essential oil of *Apollonias barbujana* subsp. *barbujana* (Cav.) Bornm. and *Citrange* cv. 'Sochinsky' and *Citrus paradisi* cv. 'Sochinsky' as sources of linalool.

Cinnamomum sieboldii (lemon aroma with fruity notes), *C. chekiangense* (woody-floral-citrus aroma), *Leptospermum scoparium* (floral-fruity aroma with a sweet woody tint), *Lindera angustifolia* (woody-herbal aroma) are suitable for perfumery application, because of the unique combination of essential oil major components.

Umbellularia californica is of interest as a natural source of umbellulone, which has potential in the development of biological methods of plant protection.

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