Pharmacochemistry

Chemical and Biological Characterization of Essential Oil from the Aerial Parts of *Salvia sclarea* L. Growing in Georgia

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The aim of the research was to study chemical composition and anti-inflammatory effect of essential oil from *Salvia sclarea* aerial parts growing in Georgia. It is well known that essential oils of *Salvia* possesses anti-inflammatory, antifungal, antioxidant, antimicrobial, cytotoxic and anti-diabetic activities. Essential oil (EO) was obtained by hydrodistillation and characterized by Gas Chromatography-Mass Spectrometry (GC-MS) technique. EO yield from areal part was 0.23%. In total, 25 different compounds of terpene nature were identified. Linalyl acetate (18.8%), linalool (14.9%) and spathulenol (9.07%) were the main components in *S. sclarea* essential oil. The anti-inflammatory activity of the essential oil was evaluated on a base of nitric oxide (NO) production measure in LPS-stimulated RAW 264.7 murine monocyte-macrophage cells. EO exhibited high anti-inflammatory activity with 74% inhibition of NO production, at 80 μg/ml dose, without any significant toxicity.

S. sclarea, Lamiaceae, anti-inflammatory activity, essential oil

The aerial parts of *S. sclarea* were collected during the flowering season (June, 2021). Dried aerial parts were hydro-distilled with three replicates for two hours with glass Clevenger-type apparatus. The concentrations of the compounds (%) were calculated by integrating their corresponding chromatographic peak areas (nonpolar column and TIC regime) using the Agilent MassHunter Software. The structure of each component was defined on the base of their mass spectral data, compared with spectral profile of reference compounds in the NIST database and the Wiley library [1,2]. For additional identification, during GC-MS analyses, the authentic samples of Linalool, Caryophyllene, Caryophyllene oxide, Humulene, α-terpineol and Linalyl acetate were

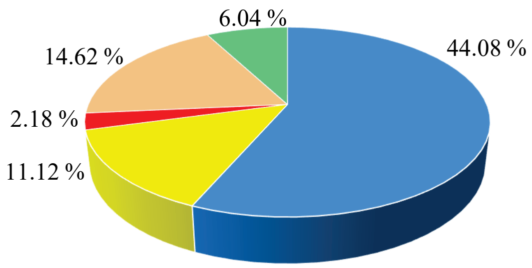

Figure. Nature of Terpenic composition of essential oil from *Salvia sclarea* (%).

Table. **Terpene composition of essential oil from *S. sclarea***

№	Compounds	RT	$RI_{(exp)}$	$RI_{(ref)}$	Essential oil content, %
1	Myrcene (MH)	7.38	980	990	1.21
2	trans-β-ocimene (MH)	8.80	1042	1038	0.5
3	β-Ocimene (MH)	9.14	1021	1037	1.04
4	Terpinolene (MH)	10.43	1012	1088	8.37
5	Linalool (OM)	11.06	1045	1090	14.9
6	Terpineol (OM)	14.05	1128	1113	0.45
7	Nerol (OM)	15.27	1225	1229	1.66
8	Linalyl acetate (OM)	16.26	1260	1234	18.8
9	Neryl acetate (OM)	19.69	1366	1361	2.97
10	Geranyl acetate (OM)	20.34	1384	1381	5.3
11	Caryophyllene (SH)	21.41	1413	1408	1.23
12	Humulene (SH)	22.44	1439	1438	0.25
13	Germacrene D (SH)	23.29	1470	1481	0.35
14	Cadinene (SH)	24.55	1540	1523	0.35
15	1,5-Epoxysalvial-4(14)-ene (OS)	25.85	1550	1554	1.16
16	Spathulenol (OS)	26.36	1559	1578	9.07
17	Caryophyllene oxide (OS)	26.44	1551	1583	1.56
18	Salvial-4(14)-en-1-one (OS)	26.67	1556	1594	0.52
19	(-)-Spathulenol (OS)	27.92	1578	1577	0.71
20	Eudesmol (OS)	28.24	1668	1650	0.73
21	Mustakone (OS)	29.03	1692	1677	0.49
22	ent-Germacra-4(15),5,10(14)-trien-1β-ol (OS)	29.24	1687	1695	0.38
23	15,16-Dinorlab-12-ene,8,13-epoxy- (O)	34.36	1877	1894	3.35
24	Epimanool (O)	38.48	2016	2056	0.42
25	Sclareol (O)	42.12	2191	2223	2.27
	Total identified				78.04

RT: Retention times were obtained from GC/MS analysis; $RI_{(exp)}$ Retention index is calculated with the help of a homologous series of n-alkanes; $RI_{(ref)}$ Retention Index of compounds from literature

used as standard compounds. The relative retention indices of all compounds were determined by the co-injection of the sample with a solution containing the homologous series of C_8–C_{30} n-alkanes. On the base of GC-MS analyses, 25 compounds were detected in *S. sclarea* essential oil. The major constituents characterised as linalool, linalyl acetate, and spathulenol. Oxygenated monoterpenes (44.08%) were abundant in *S. sclarea* aerial parts (figure), which is in agreement with the literature data [3-5]. Based on bibliography, the proportions of linalool and linalyl acetate, as major constituents were highest in *Salvia sclarea* essential oil, at the full flowering stage [6]. As it described, distillation of freshly harvested plant materials and the excess moistures, lead to production of linalool from hydrolysing of linalyl acetate and α-terpinyl acetate [7]. In our case, the essential oil was obtained from freshly harvested and rapidly dried plant material, which could explain the low constituents of linalyl acetate and linalool (18.8% and 14.9% respectively) compared to the composition of essential oils distillated from the fresh plant material. The result in Table lists the nature and quantitative ratio of identified compounds.

The anti-inflammatory activity was evaluated by assessing the inhibition of nitric oxide (NO) production by the *S. sclarea* essential oil [8]. A positive control, N-l-nitro-L-arginine methyl ester hydrochloride (L-NAME), was also used at two different concentrations: 250 µM (67 µg/mL) and 1 mM (270 µg/mL). The absorbance was read at 540 nm using a Multiskan GO plate reader (Thermo Fisher Scientific, Waltham, MA, USA). The presence of nitrite was quantified using a $NaNO_2$ standard curve. Inactivated cells (exposed to media alone) were used as a negative control and activated cells as a positive control. The essential oil exhibited high anti-inflammatory activity (74% inhibition of NO, with IC 50 - 37±3 µg/ml) at 80 µg/ml dose. No toxicity was revealed at concentration of 160 µg/ml. Based on the literature data, linalool and linalyl acetate are responsible for anti-inflammatory activity [9]. Recently revealed, that after subcutaneous injection, the *S. sclarea* essential oil exhibited a remarkable anti-inflammatory activity more evident in the carrageenan induced oedema than in the histamine-induced oedema. This activity was related to the presence of some constituents namely linalool, linalyl acetate, methyl chavicol, and terpineol [10].

ფარმაკოქიმია

საქართველოში მოზარდი ხარისვარდას (*Salvia sclarea*) მიწისზედა ნაწილების ეთერზეთების ქიმიური და ბიოლოგიური დახასიათება

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§ლაბორატორია LASEVE, ფუნდამენტურ მეცნიერებათა დეპარტამენტი, კვებეკის უნივერსიტეტი, კვებეკი, კანადა

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ნაშრომში კვლევის მიზანი იყო საქართველოში მოზარდი ხარისვარდას მიწისზედა ნაწილებიდან მიღებული ეთერზეთის ქიმიური შედგენილობისა და ანთების საწინააღმდეგო ეფექტის შესწავლა. სალბის ეთერზეთები ხასიათდება ანტიოქსიდანტური, ციტოტოქსიკური, ანთების საწინააღმდეგო, ფუნგიციდური და დიაბეტის საწინააღმდეგო აქტივობით. საქართველოში მოზარდი ხარისვარდას მიწისზედა ნაწილების ეთერზეთი მიღებულ იქნა ჰიდროდისტილაციით, მათი დახასიათება განხორციელდა გაზურ ქრომატოგრაფზე მას-სპექტრომეტრული დეტექტორის გამოყენებით. ეთერზეთის გამოსავალი იყო 0,23 %. ეთერზეთში იდენტიფიცირებულ იქნა 25 ტერპენული კომპონენტი, კერძოდ, დომინანტები – ლინალილ აცეტატი (18,8%), ლინალოოლი (14,9%) და სპატულენოლი (9,07%). ეთერზეთის ანთების საწინააღმდეგო აქტივობა შეფასდა აზოტის ოქსიდის (NO) ინჰიბირების განსაზღვრით ლიპოპოლისაქარიდით სტიმულირებული კანის ნორმალური ფიბრო-ბლასტების უჯრედებზე. ეთერზეთმა გამოავლინა NO-ს 74%-ით ინჰიბირება, 80 მკგ/მლ დოზაში, მნიშვნელოვანი ტოქსიკურობის გარეშე.

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