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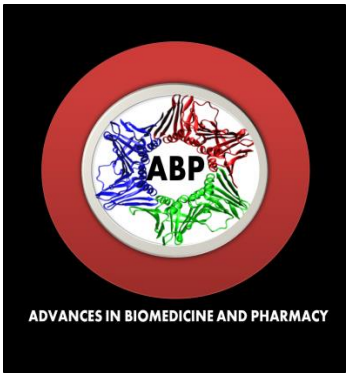

Original Article

Chemical Composition of the Essential oil of *Sambucus wightiana* Wall. ex. Wight & Arn from Kashmir Himalayas

Syed Naseer Ahmad [1]*, Manzoor A. Rather [1], Farhat Jabeen [1], Khursheed A. Bhat [1*], Mohd. A. Khuroo [2]

[1] Bio-Organic Chemistry Division, Indian Institute of Integrative Medicine, Sanat nagar, Srinagar, India.

[2] Department of Chemistry, University of Kashmir, Hazratbal, Srinagar, 190006.

*Corresponding Authors: K.A. Bhat/ Syed Naseer Ahmad

E-mail address: sdnsr18@gmail.com ; Phone: +91-9596444854; Fax: 0194-2430779

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Abstract

The essential oil composition of the fresh leaves of *Sambucus wightiana* Wall. ex. Wight & Arn growing in high altitude areas of Kashmir Himalayas is reported for the first time. A combination of Capillary GC-FID & GC-MS analysis of the essential oil led to the identification of 19 components accounting for 87.7% of the total oil composition. Monoterpene hydrocarbons dominated the oil composition accounting for 53.1% followed by oxygenated monoterpenes (27.9%); sesquiterpene hydrocarbons (4.7%) and oxygenated sesquiterpenes (2.0%). The major components were *α*-Terpinene (15.5%), (α)-Phellandrene (13.3%), (Z)-linalool oxide (13.0%); (E)- *β*-Ocimene (12.6%); 1,8-Cineole (9.0); α-Terpeniol (4.7%), *α*--Terpinyl acetate (4.0%) and Germacrene D (3.3%).

Keywords: *Sambucus wightiana*, GC-FID; GC-MS, α- Terpeniol, α- Phellandrene, (Z)-linalool oxide, (E)-β-Ocimene.

Introduction

Sambucus L. (Sambucaceae) is a small genus of approximately 25 species of deciduous shrubs, small trees and a few perennial herbs [1]. It is mainly distributed in temperate and regions of the Northern Hemisphere, but a few species occur in the Southern Hemisphere, restricted to parts of Australasia and South America [2, 3]**.** *Sambucus wightiana* (Wall. ex. Wight & Arn), is commonly known as black elder in English and locally known as "*Gandula"* in Kashmir. A gregarious herbaceous plant, up to 1.80 m high, with a perennial rootstock grows on Mountain pathway at an altitude of 2200 - 3000 meters in the Himalayas.

The fruits are used to initiate vomiting to treat stomach disorders, to wash stomach to expel poisonous substances [4]. *Sambucus wightiana* is used in folk medicine in the treatment of skin diseases. The roots, leaves and berries are reported to be used for purgative properties. The decoction of root and inner bark is an effective diuretic. It is an anti-inflammatory (especially of the bronchial tubes), expectorant, diuretic, diaphoretic and hypotensive [5]. Different extracts of *Sambucus Wightiana* have been reported to exhibit antimicrobial activity against Gram-(+), Gram (–) and various other fungal strains [6]. There are no reports on the essential oil composition of *Sambucus Wightiana*. As such, the objective of the present study was to characterize the essential oil components of *Sambucus Wightiana* which is the first report as per our literature survey from any part of the globe.

Literature survey reveals that no phytochemical investigations from any part of the globe have been previously carried out on the essential oil of *Sambucus wightiana.* As such the objective behind this research work was to characterize for the first time the essential oil components of *Sambucus wightiana* growing in Kashmir

valley using a combination of capillary GC-FID and GC-MS analytical techniques.

Materials and Methods

Plant material and extraction

As a part of our on-going research programme to develop GC-FID, GC-MS and LC-MS phytochemical profiles of various medicinal and aromatic plants of high Himalayas of Kashmir Valley, the objective behind the current research work was to study the essential oil composition of *Sambucus wightiana* using a combination of capillary GC-FID and GC-MS analytical techniques. The plant material of *Sambucus wightiana* was collected from Gulmarg (Jammu and Kashmir), India. After proper identification by Prof A. R. Naqshi (Deptt. Of Botany, University of Kashmir) a voucher specimen (No. 2107/12) was deposited in the Herbarium of the Indian Institute of Integrative Medicine, Srinagar. Fresh leaves were subjected to hydrodistillation for 3 hours, using a modified Clevenger-type apparatus. The oil yield was found to be 0.14% as calculated on fresh weight basis (v/w). The oil was dried over anhydrous Na_2SO_4 and stored in a sealed vial at 4 ^oC until gas chromatographic analysis.

GC-MS: GC-MS analysis was carried on a Varian Gas Chromatograph series 3800 fitted with a VF-5 ms fused silica capillary column (60m x 0.25mm, film thickness 0.25µm) coupled with a 4000 series mass detector under the following conditions: injection volume 0.5 µL with split ratio 1:60, helium as carrier gas at 1.0 mL/min constant flow mode, injector temperature 230 O^0C at 3^0C, oven temperature 60 C/min. Mass spectra: electron impact (EI+) mode,70 eV and ion source temperature 250°C. Mass spectra were recorded over 50-500 a.m.u range**.**

GC/FID: GC-FID was carried out on Perkin Elmer autosystem XL Gas Chromatograph 8500 series equipped with flame ionization detector (FID)and head space analyser using a fused silica capillary RTX-5 Column (30m×0.32mm×, film thickness 0.25µm) coated with dimethyl polysiloxane. Oven temperature was programmed from 60 to 280^oC at 3^oC/min, with injector temperature 230^oC. Injection volume 1µL (oil dissolved in methanol), nitrogen was used as a carrier gas (1.0 mL/min).

Results and discussion

Identification of the essential oil constituents was done on the basis of Retention Index (RI, determined with respect to homologous series of n-alkanes (C9-C24, Polyscience Corp., Niles IL) under the same experimental conditions), co-injection with standards (Sigma Aldrich and standard isolates),MS Library search (NIST 98 and WILEY), by comparing with the MS literature data [7,8].The relative percentages of the individual components were calculated based on GC peak area without using correction factors. The essential oil components identified in *Sambucus wightiana* are listed in Table 1, in order of their elution from an RTX-5 column. Capillary GC-FID & GC-MS analysis of the essential oil led to the identification of 19 components accounting for 87.7% of the total oil composition. Monoterpene hydrocarbons dominated the oil composition accounting for 53.1% followed by oxygenated monoterpenes 27.9%, sesquiterpene hydrocarbons 4.7% and oxygenated sesquiterpenes 2.0% of the total oil composition. The major components were α-Terpinene(15.5%); (*Z*)-α-Phellandrene (13.3%); (*Z*)-Linalool oxide (13.0%); (*E*)-*β*-Ocimene (12.6%); 1,8-Cineole (9.0%); *α*-Terpineol (4.7%); *α*-Terpinyl acetate (4.0%) and Germacrene D (3.3%). Some other chemical constituents with lesser quantity in the oil were (*E*)-Thujone (2.8%), Camphor (2.7%), (*Z*)-Nerolidol (1.7) and Terpinolene (1.6%).

Conflict of interest

The authors declare that there is no conflict of interest to reveal.

Table 1. Leaf essential oil composition of *Sambucus wightiana* growing in High Himalayas of Kashmir Valley.

RI Cal.	RI Lit.	Compounds	% Peak Area	Methods of identification
988	988	Myrcene	0.3	MS,RI
998	1002	*α*- Phellandrene	13.3	MS,RI,std
1012	1014	*α*- Terpinene	15.5	MS,RI,std
1033	1026	1,8-Cineole	9.0	MS,RI,std
1044	1044	*(E)-β*- ocimene	12.6	MS,RI,std
1050	1054	*γ*- terpinene	0.5	MS,RI
1062	1065	1-Octen-ol	1.3	MS,RI
1074	1067	*(z)*-linalool oxide	13.0	MS,RI,std
1081	1086	Terpinolene	1.6	MS,RI
1112	1112	*(E)*-Thujone	2.8	MS,RI,std
1140	1141	Camphor	2.7	MS,RI
1174	1174	Terpinen-4-ol	0.5	MS,RI
1186	1186	*α*- Terpineol	4.7	MS,RI,std
1348	1346	*α*-Terpinyl acetate	4.0	MS,RI,std
1417	1417	*β*-Caryophyllene	0.8	MS,RI
1454	1452	*α*- Humulene	0.5	MS,RI
1480	1484	Germacrene- D	3.3	MS,RI
1531	1531	*(Z)*-Nerolidol	1.7	MS,RI
1576	1577	Spathulenol	0.3	MS,RI
		Class composition Monoterpene hydrocarbons Oxygenated monoterpenes Sesquiterpene hydrocarbons Oxygenated sesquiterpenes Total identified	 43.3 38.0 4.6 2.0 87.9	

RI [a]; Retention indices on RTX-5column (relative to n-alkanes).
RI [b]; Retention indices on DB-5 column (reported from literature).

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