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Research Article

Chemical Composition and Physicochemical Properties of Essential Oil from *Myrtus communis*

Zenebe Hagos[1*], Afework Mulugeta[2], Gopalakrishnan V K[1], Krishna Chaithanya K[1], Nagaraju B[1]

[1]*Department of Chemistry, College of Natural and Computational Sciences, Aksum University, Axum, Ethiopia.*
[2]*College of Health Sciences, Mekelle University, Mekelle, Ethiopia.*

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ABSTRACT

Objective: The objective of the study was to extract the essential oil and to determine the physicochemical properties, chemical composition of the essential oil from *Myrtus communis* leaves by using GC-MS. Methods: 500 g of the powdered material (dried leaves) was subjected to hydro-distillation using Clevenger-type apparatus for 3 hours and the essential oil was dried over anhydrous sodium sulphate. The physicochemical properties essential oil such as solubility, density, specific gravity, refractive index, optical rotation, specific rotation, acid value, saponification value and ester value were determined, which are used to determine the quality of essential oils extracted from plant leaves. The chemical composition of the essential oil of *Myrtus communis* leaves was analysed using GC-MS. Results: The yield of essential oil of *Myrtus communis* leaves was 0.463 ± 0.001 g/100 gram of dry sample which is 0.463 ± 0.001 % (w/w) with pale yellow colour and pleasant odour. The essential oil of *Myrtus communis* leaves was slightly soluble in water and ethanol while it was soluble in chloroform, petroleum ether and ethyl acetate. The essential oil was subjected to GC-MS analyses, twenty eight components representing 99.949 % of the total oil composition. Conclusion: The present study indicates that the extraction of essential oil from *Myrtus communis* leaves and GC-MS analysis, the essential oil of this plant have a potential of secondary metabolites which can be exploited as a good source of bioactive substances.

Keywords: *Myrtus communis* leaves; GC-MS; Essential oil; Secondary metabolites.

INTRODUCTION

Medicinal plants are the nature's gift to human beings to make disease free healthy life. The medicinal properties of plants have been investigated in the recent scientific developments throughout the world, due to their potential activity against several diseases, without side effects and economic viability. Several bioactive compounds widely distributed in plants which have been reported to exert multiple biological effect, including antioxidant, anti-inflammatory, anti-carcinogenic etc.[1-4].

There is increasing evidence that oxidative stress leads to many biochemical changes and consequently serious disorders in the humans. Oxidative stress can end result in damage to basic biomolecules, such as DNA, proteins and lipids which leads to cytotoxic and genotoxic effects[5,6]. Generation of free radicals or reactive oxygen species (ROS) during metabolism and other activities beyond the antioxidant capacity of a biological system gives rise to oxidative stress. Oxidative stress plays a role in cardiovascular diseases, neurodegeneration, cancer and in the aging process[7]. Antioxidants are capable of stabilizing or deactivating free radicals before they attack cells. Antioxidants are completely essential for maintaining optimal cellular and systemic health and well-being. Many plant-derived substances are becoming increasingly known for their antioxidant activity[8]. It is well known that oxidative stress is involved in the pathogenesis of several diseases. However, no optimal natural antioxidant has been found for therapeutics. Thus, phenolic substances such as tocopherol (vitamin E and related compounds), various classes of flavonoids, phenolic acid, tannins, lignans, essential oils etc., are of special interest[9].

Moreover, recently there has been a profound interest in the antimicrobial properties of extracts from aromatic medicinal plants, specifically essential oils. Essential oils are volatile complex compounds which are characterized by a strong odour and are formed naturally by aromatic plants as secondary metabolites. They are rich sources of biologically active compounds[10].

Thus, plants are a good source of biologically active compounds known as phytochemicals. Phytoconstituents have been found to work as antioxidants by scavenging free radicals and many have curative potential for free radical associated disorders and they also have antimicrobial activity[11].

Myrtle (*Myrtus communis* L.) is an evergreen shrub aromatic plant belonging to the Myrtaceae family. It grows spontaneously throughout the Mediterranean area and has been used for medicinal, foodstuff and spice purposes since ancient times. The fruit and leaves are traditionally used as disinfectant, antiseptic and hypoglycemic agents[12]. In folk medicine the fruit of the plant is used in the

**Author for Correspondence: zenebe.h2005@gmail.com*

Table 1: Physicochemical properties of the essential oil of *Myrtus communis* leaves.

S.No.	Physicochemical characteristics		Results
1	Solubility in	Water	Slightly soluble
		Ethanol	Slightly soluble
		Chloroform	soluble
		Petroleum ether	Soluble
		Ethyl acetate	Soluble
2	Density at 25 ºC (g/mL)		0.894 ± 0.001
3	Specific gravity at 25 ºC		0.894 ± 0.001
4	Refractive index at 20 ºC		1.463 ± 0.001
5	Optical rotation at 25ºc in degree(º)		-24.87 ± 0.153
6	Specific rotation		-565.152 ± 3.471
7	Acid value		4.451 ± 0.710
8	Saponification value		36.465 ± 2.805
9	Ester value		32.014 ± 2.754

n = 3 and values are expressed as mean ± standard deviation (SD)

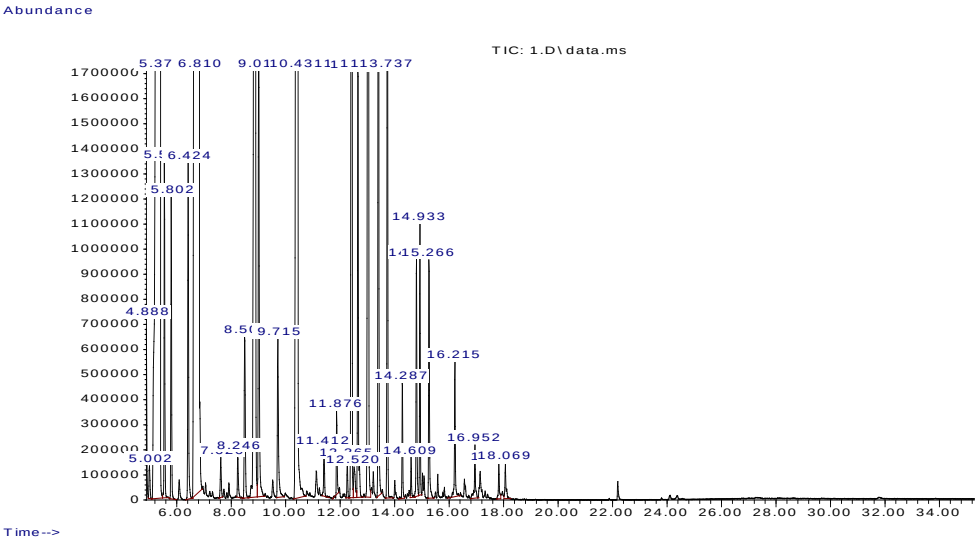

Figure 1: Gas chromatogram of the essential oil of *Myrtus communis* leaves.

treatment of various transmittable diseases such as diarrhea and dysentery, while the leaves are used as antibacterial and anti-inflammatory agents, as a mouthwash, for treatments of candidiasis, for healing wounds, as well as in the therapy of urinary diseases[13,14]. The leaves contain tannins, flavonoids such as quercetin, catechin and myricetin derivatives and volatile oils[15,16]. Essential oils are gaining remarkable interest for their potential multipurpose use as antioxidant, antibacterial, and antiseptic agent; the essential oil obtained from the leaves was used in the past for the treatment of lung disorders[17-20]. The objective of the study was to extract the oil and to determine the chemical composition, physicochemical properties of the essential oil of *Myrtus communis* leaves by using GC-MS.

MATERIALS AND METHODS

Collection of plant material

Fresh leaves were collected from healthy and uninfected *Myrtus communis* tree from Chelekot, which is about 16 km Southwest Mekelle town in January 2013. The leaves were washed under running tap water followed by using distilled water to eliminate dust and other foreign particles and to cleanse the leaves thoroughly and it was dried under shade. The plant material was authenticated and specimen herbarium was deposited at Addis Ababa University, Biology department, National herbarium of Ethiopia and it is given the voucher specimen number of 81858.

Extraction of the essential oils of the plant

The dried leaves were gridded or reduced to coarse powder using mortar and pestle. The powder material was homogenized and 500 g of the powdered material was subjected to hydro-distillation using Clevenger-type apparatus for 3 hours and the essential oil was dried over anhydrous sodium sulphate. Extraction was done five times. The percentage yield of essential oil is calculated using the formula;

$$\text{percent yield of oil (w/w)} = \frac{\text{weight of oil in gram}}{\text{weight of the sample in gram}} \times 100$$

Then it was placed in amber bottles and was stored in refrigerator at 4ºC until further investigation.

Physicochemical properties of essential oils

The physicochemical properties essential oils such as solubility, density, specific gravity, refractive index, optical rotation, specific rotation, acid value, saponification value and ester value were carried out in an essential oil of a plant, which are used to determine the quality of essential oils extracted from plant parts[21].

GC-MS Analysis

The chemical composition of the essential oil of *Myrtus communis* leaves was analysed using GC-MS (Agilent 6890N GC with 5975MSD).

RESULTS AND DISCUSSION

Essential oil yield

The yield of essential oil in the examined *Myrtus communis* leaves was 0.463 ± 0.001 g/100 gram of dry sample which is 0.463 ± 0.001 % (w/w) with pale yellow colour and pleasant odour. In the literature[23] reported yields of the hydrodistilled and Solvent Free-Micro Wave extracted essential oil of *Myrtus communis* leaves from Bainem, Northwest of Algiers harvested in November were 0.32 ± 0.06 g/100 g dry basis and 0.33 ± 0.08 g/100

Table 2: Chemical composition of the essential oil of *Myrtus communis* leaves.

RT(min)	RI (iu)	Constituents	Composition (%)
4.787	948	3-Carene (3,7,7-trimethylbicyclo[4.1.0]-3-heptene)	0.208
4.892	998	α-Terpinene (1-methyl-4-(1-methylethyl)-1,4-cyclohexadiene)	1.083
4.997	1052	Terpinolene (1-Methyl-4-(1-methylethylidene)-1-cyclohexene)	0.904
5.376	1059	Eucalyptol (1,3,3-trimethyl-2-oxabicyclo[2.2.2]octane)	33.495
5.555	948	Pinene (2,6,6-Trimethylbicyclo(3.1.1)-2-hept-2-ene)	0.695
6.807	1082	Linalool (3,7-dimethyl-1,6-octadien-3-ol)	29.217
7.617	1131	Trans-Pinocarveole	0.114
8.501	1137	p-menth-1-en-4-ol (4-methyl-1-(1-methylethyl)-3-cyclohexen-1-ol)	0.488
8.880	1143	α-Terpineol (a,a,4-trimethyl -3-cyclohexene-1-methanol)	7.158
9.017	1172	Estragole(p-allylanisole)	1.347
9.711	1228	Cis-Geraniol (z)-3,7-dimethyl-2,6-octadien-1-ol)	0.494
10.427	2157	Linalyl anthranilate	9.048
11.416	1271	(-)-trans-Pinocarveole acetate	0.128
11.879	1252	Methyl geranate (3,7-dimethyl-2,6-octadienoic acid methyl ester)	0.192
12.268	1386	Exo-2-Hydroxycineole acetate (1,3,3-trimethyl-2-oxabicyclo[2.2.2]octan-6-ol acetate)	0.090
12.426	1333	α-terpineol acetate (a,a,4-trimethyl-3-cyclohexene-1-methanol acetate)	3.639
12.520	1436	1,5,8-trimethyl-1,2-dihydronaphthalene	0.140
13.036	943	(-)-2(10)-pinene ((1s)-6,6-dimethyl-2-methylene bicycle[3.1.1]heptanes)	5.349
13.404	1361	Methyl eugenol (1,2-dimethoxy-4-(2-propenyl)benzene	2.076
13.741	1494	Caryophyllene	1.339
14.288	1579	α-Caryophyllene (2,6,6,9-Tetramethyl-1,4,8-cycloundecatriene)	0.271
14.804	1469	(+)-α-Selinene (7-Isopropenyl-4a-methyl-1-methylenedecahydronaphthalene)	0.573
14.930	1502	2-Isopropenyl-4a,8-dimethyl-1,2,3,4,4a,5,6,7-octahydronaphthalene	0.662
15.267	1574	Durohydroquinone (2,3,5,6-tetramethyl-1,4-benzenediol)	0.572
16.214	1507	Caryophyllene oxide	0.347
16.950	2026	6,6-dimethyl-4-(4-morpholinyl)-2-oxo-3-cyclohexene-1-caboxylate	0.116
17.823	1586	Geranyl isovalerate ((2E)-3,7-Dimethyl-2,6-octadienyl 3-methylbutanoate)	0.094
18.065	1746	5,6,6-trimethyl-5-(3-oxobut-1-enyl)-1-oxaspiro[2,5]octan-4-one	0.110

g dry basis respectively which are lower than the yield of the present study harvested in January 2013 from Chelekot, Southwest of Mekelle, Ethiopia. Moreover, the another researcher reported the essential oil yield of the plant leaves harvested in August from two region in Montenegro were 0.72 g/100 g dried leaves and 0.81 g/100 g dried leaves[22]. Vahid Rowshan *et al*.,[24] also reported the essential oil yield of young and mature *Myrtus communis* leaves were 0.92 % w/w and 0.48 % w/w respectively which are higher than the yield of the present study. Therefore, the yield variability in the total essential oil of *Myrtus communis* leaves growing in different areas could be due to the difference in geographic, climatic, ecological parameters. Moreover, oil yield can be affected by the maturity stage and time of harvesting of the leaves of the plant.

Physicochemical properties of the essential oil of Myrtus communis leaves

The physical and chemical characteristics such as solubility, refractive index, optical rotation, specific rotation, density, specific gravity, acid value, saponification value and ester value of the essential oil of *Myrtus communis* leaves were determined. The results are shown in table 1.

The essential oil of *Myrtus communis* leaves was slightly soluble in water and ethanol while it was soluble in chloroform, petroleum ether and ethyl acetate. The solubility difference of the essential oil in the solvents is due to the difference in polarity of the solvents and nature of the components of the essential oil since the components are terpenes and their derivatives. The slight solubility of the essential oil in water and ethanol indicates presence of oxygenated terpenes with polar ends and the solubility of essential oil in chloroform, petroleum ether and ethyl acetate is due to the presence high amount of terpenes; both terpene hydrocarbons and oxygenated terpenes.

The density and specific gravity of the essential oil at 25 ^{o}C were 0.894 ± 0.001 g/mL and 0.894 ± 0.001 respectively. These results are comparable with the results reported in literature[23] which were 0.905 ± 0.0342 g/mL and 0.905 ± 0.0342 at 20 ^{o}C.

The refractive index of the essential oil of *Myrtus communis* leaves was 1.463 ± 0.001 at 25 ^{o}C which is lower than 1.470 ± 0.021 at 20 ^{o}C as reported in literature[23]. The optical rotation of the essential oil of *Myrtus communis* leaves was -24.87 ± 0.153^{o}. This result is quite greater than the value reported in literature[23] which is 1.54 ± 0.13^{o}. The acid value, saponification value and ester value of the essential oil were 4.451 ± 0.710, 36.465 ± 2.805, and 32.014 ± 2.754 respectively. These are the characteristics of the essential oil of *Myrtus communis* leaves. These results was not compared since there is no official standard and published reports on the physicochemical properties of the essential oil of the leaves of this plant except the one which was reported by Baya Berka-Zougali *et al*.,[23] for the three physical constants of the essential oil. For that reason, it was not possible to compare on that basis.

Chemical composition of the essential oil of Myrtus communis leaves

The chemical composition of the essential oil of *Myrtus communis* leaves was analysed using GC-MS (Agilent 6890N GC with 5975MSD). The essential oil GC-MS analyses lead to the identification of twenty eight components representing 99.949 % of the total oil composition. The identified constituents of the essential oil with their retention index and respective percentages are given in table 2. The major constituents of the essential oil were 1,8-cineole which is also called eucalyptol (33.495 %) and linalool (29.217 %) followed by linalyl anthranilate (9.048 %) and α-terpineol (7.158 %). Moreover, the essential oil contain significant amount of (-)-α-Pinene (5.349 %), α-terpineol acetate (3.639 %) and methyl eugenol (2.076 %). The result of GC-MS analyses of the essential oil also showed lower amount of exo-2-hydroxycineole acetate (0.090 %) and geranyl isovalerate (0.094 %) as depicted in (Table 2).

CONCLUSION

The major components of the essential oil of the examined *Myrtus communis* leaves are eucalyptol (33.495 %). α-linalool (29.217 %), linalyl anthranilate (9.048 %) and α-terpineol (7.158 %). The essential oil of *Myrtus communis* leaves is a potential source of natural antioxidants and antibacterial compounds which are used for the treatment of various diseases caused by free radicals and microbes.

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CONFLICT OF INTEREST

There is no conflict of interest.

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