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PHYTOCHEMICAL ANALYSIS OF *PORTULACA OLERACEA* AND *PORTULACA QUADRIFIDA* EXTRACTS USING GAS CHROMATOGRAPHY-MASS SPECTROMETRY

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ABSTRACT

Objective: The two plant species *Portulaca oleracea* and *Portulaca quadrifida* are commonly known as purslane and chickweed, respectively. They are typically consumed as salad or pickle. Traditional systems of medicine from Africa and China have described these plants belonging to family Portulacaceae as remedies against a host of diseases. Recent pharmacological investigations have revealed the importance of these plants as sources of antioxidants, essential fatty acids, and even antimicrobial agents. The objective of this study was phytochemical analysis and comparison of ethanolic extracts of these two species of Portulaca.

Methods: The ethanolic extracts of both the species were prepared using Soxhlet extraction and were analyzed using gas chromatography coupled with mass spectrometry (GC–MS). Furthermore, the ethanolic extracts of fresh and dried whole plant of *P. oleracea* and seed of *P. oleracea* were studied.

Results: The phytochemical constituents of ethanolic extracts of *P. oleracea* and *P. quarifida* were found to be quite different from one another and contained beneficial polyunsaturated fatty acids, alkaloids among other beneficial chemical species.

Conclusion: The results of the study could be further used by researchers to assess the beneficial properties of both these species for *in vitro* and *in vivo* experiments.

Keywords: Portulaca oleracea, Portulaca quadrifida, Gas chromatography-mass spectrometry.

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INTRODUCTION

Portulaca oleracea L. commonly known as Purslane belongs to the family Portulacaceae. This plant might have originated in Asia and is now grown in Africa, Asia, and Mediterranean region [1]. It is found to grow as a weed in turfgrass or field crop [2,3]. It is used in salads, soups, and pickles for its sour taste. Conventionally, it has been described to be beneficial for the treatment of diseases related to intestine, liver, stomach, cough, shortness of breath, arthritis, as well as for burns and headache. It is also used for diuretic treatment, as a purgative, emollient, and as anti-inflammatory agent [4]. Chinese traditional medicine describes the use of Purslane to resolve toxins, stanch bleeding, cool blood, and the aerial parts of the plant for the treatment of eczema, dysentery, and diarrhea [5]. Not only traditional systems of medicine but also modern investigative techniques have revealed that the Purslane weed has noticeable nutritive value. The phytochemical screening of Purslane has suggested that the plant has a higher content of beta-carotene and ascorbic acid than some of the traditional nutritive plant crops. Furthermore, the plant has been reported to be a storehouse of omega-3 fatty acids such as alpha-linolenic acid [6]. As such, the plant can be said to be a major source of dietary antioxidants and nutrients [7].

Another plant belonging to family Portulacaceae and widely distributed around Asia and Africa is *Portulaca quadrifida* commonly known as chickweed [8]. The plant has been traditionally used in parts of Africa for medicinal purposes to treat asthma, cough, urinary discharges, inflammations and ulcers, abdominal complaints, and hemorrhoids [9]. It has also been demonstrated to possess antifungal activity against *Aspergillus fumigatus* and *Candida albicans* [10]. The leaves of the plant are used in salad or in preparation of soup [11]. Preliminary phytochemical screening of *P. quadrifida* ethanolic extract has been reported to contain polyphenols such as flavonoids and alkaloids. [12]. Since detailed analysis of phytochemical constituents for *P. quadrifida* were not found to be reported earlier, this study might help investigate this aspect in comparison with phytochemical constituents of *Portulaca oleracea* collected from Maharashtra state of Western part of India.

METHODS

Collection of specimen

The plants of *P. oleracea* and *P. quadrifida* were harvested from local fields of district Sangli, state Maharashtra in February–March. The specimens were authenticated by Dr. Dhanaji S. Pawar, Associate Professor, Department of Botany, M. H. Shinde Mahavidyalaya, Tisangi. For obtaining the dry powder of the *P. oleracea* whole plant, the plants were dried in shade and seeds from some of the *P. oleracea* plants were separated. The dried whole plant of *P. oleracea* was powdered using mortar pestle.

Extraction using Soxhlet apparatus

For each of the four specimens (namely the *P. oleracea* fresh whole plant, *P. quadrifida* fresh whole plant, powder of dried *P. oleracea* whole plant, and seeds of *P. oleracea*), 20 g specimen was extracted in 200 mL of ethanol for 5–6 cycles. The extracts were then evaporated to dryness. The extracts were reconstituted in ethanol for further analysis.

Gas chromatography-mass spectrometry (GC-MS) analysis

The GC system comprised a PerkinElmer GC Clarus 500 system with an autosampler. The capillary column used for GC system was of methyl silicone 25 m in length, 0.2 mm inner diameter, and 0.33 μ m film thickness. The carrier gas used was helium at a constant flow rate of

1 mL/min with 2 μ L injection volume at a temperature of 260°C. Initial temperature of oven was 100°C and was increased at a rate of 10°C/min to 280°C for 10 min. The temperature in the interphase was 280°C and temperature of the source in detector was 180°C [13]. The spectrum data obtained were searched against the database of National Institute of Standard ad Technology.

RESULTS AND DISCUSSION

Previous studies have reported that Purslane is a rich source of antioxidants. The observed antioxidant property can be attributed to the presence of Vitamin A, Vitamin C, B-complex vitamins, glutathione, and β -carotene among other phenolic compounds. It also provides high quantity of dietary minerals such as potassium, magnesium, iron, calcium, and phosphorus [4]. Another beneficial property of Purslane is the presence of omega-3 fatty acids which are a precursor to specific group of hormones and provide protection against cardiovascular diseases by decreasing the thickness of blood [4,6]. However, the distribution of nutrients varies among the different parts of the plant such as in the leaves and stem [14,15]. Furthermore, variation in the nutritive content has been reported in the different varieties of Purslane depending on the various growth conditions [16]. This study aimed to investigate and compare the phytochemicals present in ethanolic extract of fresh sample and dry powder of P. oleracea whole plant, seeds, and whole plant of P. quadrifida collected from the local fields of Sangli district, state of Maharashtra.

The GC–MS data obtained for the ethanolic extract of fresh whole plant of *P. oleracea* were as mentioned below along with the major identified phytochemicals (Table 1).

The above data reveal the presence of methyl esters of polyunsaturated fatty acid 6,9,12-octadecatrienoic acid, phenylmethyl ester, and other methyl esters such as cyclopropanepentanoic acid, 2-undecyl-, methyl ester, trans. 1-(5-bicyclo[2.2.1]heptyl)ethylamine is an alkaloid which has may pharmaceutical applications and has been tested for antibacterial and antiviral properties [17].

The GC obtained for the ethanolic extract of fresh whole plant of *P. quadrifida* contained major constituents identified as Table 2.

The ethanolic extract of fresh whole plant of *P* quadrifida was found to contain methyl esters of fatty acids 9-octadecenoic acid, tetradecanoic acid, hexanedioic acid, and octadecanoic acid. 3,7,11,15-tetramethyl-

2-hexadecen-1-ol is a diterpenoid commonly called as phytol is commercially used as a precursor for synthesis of Vitamin E and Vitamin K1 and has pharmaceutical properties [18]. The sample also contained acyclic alkanes such as 2-methylhexacosane and triacontane.

The dried powder of fresh whole plant of *P. oleracea* was extracted using ethanol and analyzed by GC coupled to MS as shown in Table 3.

The ethanolic extract of dried whole plant of *P. oleracea* contained esters of cyclopropanepentanoic acid, hexanedioic acid, octadecanoic acid besides, phosphoric acid, dibutyl 3-trifluoromethyl-3-pentyl ester, n-nonadecanol, trans-2-dodecen-1-ol, 2-methyl-Z,Z-3,13-octadecadienol, and 9,12,15-octadecatrienal. As compared to the results of fresh whole plant *P. oleracea*, the content of phosphoric acid, dibutyl 3-trifluoromethyl-3-pentyl ester, and cyclopropanepentanoic acid was found to be lower in the dried whole plant sample (Table 1).

The GC obtained for the ethanolic extract of seeds of *P. oleracea* was obtained as below along with the major identified phytochemicals (Table 4).

The ethanolic extract of seeds of *P. oleracea* was found to contain esters of fatty acids such as 10-octadecenoic acid, tetradecanoic acid, 9,12-octadecadienoic acid, and octadecanoic acid. Tributyl phosphate has been reported to possess cytotoxic activity [19].

CONCLUSION

The results of the GC-MS analysis of ethanolic extracts of P. oleracea and P. quadrifida indicate the presence of many useful compounds such as polyunsaturated fatty acids including ω -3 and ω -6 fatty acids, alkaloids, and terpenoids. Although the results of GC-MS analysis of P. oleracea were not similar to previous reports, it must be remembered that the growth conditions and time of harvest play an important role in the determining the phytochemicals and their amount [20]. Since the plants were harvested from fields where they were neglected as weed, the growing conditions might not have been conducive for expression of high amounts of essential fatty acids in the plant. Portulaca quadrifida GC-MS analysis was not published earlier, and hence, this analytical report might further help researchers study its use as a source of nutrition or other pharmacological activities. Earlier, in vivo investigation on the effective doses of P. oleracea whole plant extract indicated that dosage levels of 500 mg/kg body weight could be used to study its pharmacological activity and dosage higher than 400 mg/kg body weight could provide

Table 1: Identified phytochemicals using MS coupled to GC of ethanolic extract of fresh whole plant of P. oleracea

Peak No.	R.T. (min)	Area (%)	Molecular formula	Molecular weight	Name
1	9.701	16.44	C ₀ H ₁₇ N	139	1-(5-Bicyclo[2.2.1]heptyl) ethylamine
2	11.378	26.48	$C_{14}^{9}H_{28}F_{3}O_{4}P$	348	Phosphoric acid, dibutyl 3-trifluoromethyl-3-pentyl ester
3	14.288	27.98	C ₆ H ₇ N ₃ O ₂	153	Imidazole, 2-amino-5-[(2-carboxy) vinyl]
4	15.993	23.7	$C_{20}H_{38}O_2^2$	310	Cyclopropanepentanoic acid, 2-undecyl-, methyl ester, trans
5	16.629	5.39	$C_{25}^{20}H_{36}^{30}O_{2}^{2}$	368	6,9,12-Octadecatrienoic acid, phenylmethyl ester, (Z, Z, Z)

MS: Mass spectrometry, GC: Gas chromatography, P. oleracea: Portulaca oleracea

Table 2: Identified phytochemicals using MS coupled to GC of ethanol	lic extract of fresh whole plant of P. quadrifida
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Peak No.	R.T. (min)	Area (%)	Molecular formula	Molecular weight	Name
1	7.434	2.05	$C_{12}H_{18}O_2$	194	Tricyclo[4.3.1.1 (3,8)]undecane-3-carboxylic acid
2	11.379	2.73	$C_{12}^{12}H_{27}^{10}O_{4}^{2}P$	266	Tributyl phosphate
3	13.386	6.25	$C_{20}^{12}H_{40}^{2}O^{4}$	296	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
4	13.472	2.72	$C_{23}^{20}H_{30}^{40}N_2O$	350	1-Benzoylamino-5-piperidinyl-1-phenylpentane
5	13.841	3.76	$C_{19}H_{36}O_{2}$	296	9-Octadecenoic acid, methyl ester
6	14.845	1.15	$C_{16}^{15}H_{32}^{30}O_{2}^{2}$	256	Tetradecanoic acid, 12-methyl-, methyl ester
7	15.943	6.58	$C_{14}^{10}H_{26}^{2}O_{4}^{2}$	258	Hexanedioic acid, mono (2-ethylhexyl) ester
8	15.985	18.02	$C_{27}^{14}H_{56}^{20}$	380	2-methylhexacosane
9	16.199	2.51	$C_{30}^{27}H_{61}^{30}Br$	500	Triacontane, 1-bromo-
10	17.643	1.94	$C_{38}^{50}H_{76}^{10}O_{3}$	580	Octadecanoic acid, 2-(octadecyloxy) ethyl ester

MS: Mass spectrometry, GC: Gas chromatography, Portulaca quadrifida: P. quadrifida

Table 3: Identified phytochemicals using mass spectrometry coupled to gas chromatography of ethanolic extract of dried whole plant of P. oleracea

Peak No.	R.T. (min)	Area (%)	Molecular formula	Molecular weight	Name
1	11.378	2.9	$C_{14}H_{28}F_{3}O_{4}P$	348	Phosphoric acid, dibutyl 3-trifluoromethyl-3-pentyl ester
2	12.054	2.69	$C_{10}^{14}H_{40}^{20}O^{3}$	284	n-Nonadecanol-1
3	13.384	4.05	$C_{12}H_{24}^{10}O$	184	Trans-2-Dodecen-1-ol
4	14.839	4.85	$C_{8}^{11}H_{19}^{12}N$	129	2-Pentanamine, N-ethyl-4-methyl
5	15.988	7.98	$C_{20}H_{38}O_2$	310	Cyclopropanepentanoic acid, 2-undecyl-, methyl ester, trans
6	16.549	2.2	$C_{19}^{20}H_{36}^{30}O^{2}$	280	2-Methyl-Z, Z-3,13-octadecadienol
7	16.587	0.99	$C_{18}^{10}H_{30}^{10}O$	262	9,12,15-Octadecatrienal
8	16.624	1.01	$C_{22}H_{42}O_{4}$	370	Hexanedioic acid, diisooctyl ester
9	18.899	35.16	$C_{38}^{22}H_{76}^{42}O_{3}^{4}$	580	Octadecanoic acid, 2-(octadecyloxy) ethyl ester
10	28.006	8.07	$C_{39}^{30}H_{80}^{30}O_{2}^{3}$	580	Octadecane, 1,1'-[(1-methyl-1,2-ethanediyl) bis (oxy)]bis-

Table 4: Identified phytochemicals using mass spectrometry coupled to gas chromatography of ethanolic extract of seeds of P. oleracea

Peak No.	R.T. (min)	% Area	Molecular formula	Molecular weight	Name
1	11.377	4.05	$C_{12}H_{27}O_{4}P$	266	Tributyl phosphate
2	12.059	1.85	$C_8^{12}H_{19}^{12}N$	129	2-Pentanamine, N-ethyl-4-methyl-
3	14.839	3.4	$C_{19}H_{36}O_2$	296	10-Octadecenoic acid, methyl ester
4	14.94	2.24	$C_{16}^{19}H_{32}^{30}O_{2}^{2}$	256	Tetradecanoic acid, 12-methyl-, methyl ester
5	15.943	5.4	$C_{17}^{10}H_{32}^{32}O_{1}^{2}$	252	13-Heptadecyn-1-ol
6	15.986	31.03	$C_{27}^{17}H_{54}^{52}O_{4}Si_{2}$	498	9,12-Octadecadienoic acid (Z, Z)-, 2,3 bis[(trimethylsilyl) oxy]
			2, 01 1 2		propyl ester
7	18.9	4.36	$C_{20}H_{42}O_{2}S$	346	Di-n-decylsulfone
8	20.751	5.48	$C_{26}^{20}H_{44}^{42}O_5^{2}$	436	Ethyl iso-allocholate
9	26.65	5.97	$C_{38}^{20}H_{76}^{44}O_{3}^{5}$	580	Octadecanoic acid, 2-(octadecyloxy) ethyl ester

significant hepatoprotective activity in mice [21]. Similar dosage levels have also been reported for *in vivo* pharmacological activities of *P. oleracea* and *P. quadrifida* extracts in mice which indicated the presence of antinociceptive and muscle relaxant activities [22]. These reports along with the present findings of phytochemical constituents could help investigators design experiments accordingly.

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AUTHOR'S CONTRIBUTION

The work of collection of plant material, preparation of extract, and analysis of GC–MS results was done by Trupti Durgawale. Dr. Chitra C. Khanwelkar has guided her and reviewed her work. Pratik Durgawale has worked on drafting of the manuscript and analysis of GC–MS results.

CONFLICTS OF INTEREST

The authors declare that they have no conflicts of interest.

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