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Fatty Acids and Elements Profile of Different Parts from Congress grass (*Parthenium hysterophorus* L.)

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Abstract

Congress grass (*Parthenium hysterophorus* L., family Asteraceae) is found generally in agricultural and vacant lands across the globe. This study investigated the fatty acids and elemental profiles of different parts including roots, stem and leaves of *P. hysterophorus* in the response to seasonal variation. The elemental analysis and fatty acid were determined in both the winter and summer seasons. The root, stem, and leaves powder were subjected to extraction with acetone, water, chloroform, diethyl ether, ethanol, methanol, and n-hexane to measure the extractive values. Among all the plant parts, leaves recorded the highest extractive value of 29%, and 28% with acetone and diethyl ether, respectively, while the root recorded the lowest (10.3%) extractive value with chloroform. Higher concentrations of saturated and unsaturated fatty acids were detected in all the plant parts in the winter collection than in the summer season. *Parthenium hysterophorus* is multi medicinal applications such as used for rheumatic pain, urinary tract infections, inflammation, diarrhea, neuralgia, and malaria. The study highlights the importance of collection time (seasons) of different parts of *P. hysterophorus* for the maximum extraction of both elements and fatty acids either saturated or unsaturated from the pharmacogenetic point of view.

Keywords: Gajar Ghaas, Asteraceae, Elemental Analysis, Medicinal Plants, Fatty Acid

INTRODUCTION

Pharmacognosy is the study of crude drugs in terms of history, cultivation, collection, structural, physical, chemical and biochemical properties, discovery, standardization as well as safe commercial uses.[1-3] Professional pharmacognosists have to evaluate various medicinal plant parts, and products derived from plant materials, investigate the crude drugs and assure the absence of adulteration.[4,5] Herbal medicines have a significant role in the traditional healing system for the cure of various ailments. The use of medicinal plants originated in Greek and gradually transferred to Asian and European countries.[3,4]

Pakistan has tremendous floristic diversity with 6000 medicinal plants of which 1572 species are used as herbal medicines.[5] About 75-80% of medicinal plants have a therapeutic effect and are used to treat various diseases in Pakistan.[6] Reliability of natural products extracted from the whole plant or plant parts is a difficult task due to their diverse composition. Plants produce useful bioactive substances such as flavonoids, alkaloids, tannins, and oils which are effective as health-promoting factors for humans.[6-10] To minimize the adverse effects of herbal medicines, their safe use might be studied to help in the identification of adulterants.[8] Medicinal plants are a rich source of secondary metabolites having health-promoting potential, and micro-chemical screening provides substantial knowledge regarding the chemical compounds which is accountable for their medicinal traits.

Nutrients play an important role in plant and animal life.[8,9] In most developing countries, the flora from a medicinal point of view remains unexplored so the system of medicines particularly brings the use of these elements for therapeutic purposes.[11] Trace elements have an important role in curative as well as in the prevention of diseases. Trace elements are a vital part of enzymatic systems which help in controlling physiological activities. They also played an important role in the metabolism of plant secondary metabolites.[12] Studies have reported the elemental profile of different medicinal plants such as *Phyla nodiflora*,[13] Ficus religiosa,[14] and *Withania somnifera*.[15]

Fatty acids are produced in various plant parts.[16] Fatty acids were identified from different plants such as *Dodonaea viscosa*[17] as well as *Sonchus asper*, and *Illicium griffithii*.[18] The therapeutic applications of fatty acids were reported by several research groups including antibacterial activity,[19] analgesic activity,[20] and antispasmodic activities.[21]

Parthenium hysterophorus commonly known as Congress Grass or Gajar Ghaas belonging to the Asteraceae family is found throughout the global agricultural and vacant lands including arid. *P. hysterophorus* is a weed of global concern with a

high threshold of tolerance against most biotic and abiotic stress zones.[22,23] Few studies reported on the chemical composition of *P. hysterophorus*. Ahmad and group[17] studied the composition of root, stem, leaves, and receptacle of *P. hysterophorus* and assessed the antioxidant potential using different assays such as 2,2-diphenyl-1-picrylhydrazyl (DPPH), ferric reducing antioxidant power (FRAP), hydroxyl radical scavenging activity (HRSA), and total antioxidant capacity (TAC) assays. The phytochemical screening of the species revealed the presence of terpenes, hydrocarbons, sterols, fatty acids, and compounds of miscellaneous chemical nature. The organ-specific maximum concentration of metabolites was β-vatirenene (root), hexadecenoic acid methyl ester (stem), and aristolene epoxide (leaf). In the present study, we investigate the elemental and fatty acid profiles of various parts including root, stem and leaves of *Parthenium hysterophorus* as affected by the season.

MATERIALS AND METHODS

Plant Collection and Preservation

The fresh plants of *Parthenium hysterophorus* were collected during the summer and winter from village Badwan, district Dir Lower, Khyber Pakhtunkhwa (Pakistan). The plant samples were identified by a taxonomist Mr. Ghulam Jilani and submitted to the University of Peshawar, Khyber Pakhtunkhwa, Pakistan (Bot. 20077 (PUP). Plant samples were washed with water to remove dust particles and separated into individual parts. Fresh plant material was used for macroscopic and microscopic features, while the rest of the plant parts were shade dried at 25-30°C and ground to a fine powder in an electric grinder. The powdered materials were used for the screening of phytochemicals, proximate and elemental analysis, fluorescence study, extractive values, and fatty acids profile.

Elemental Analysis

Each morphological part; root, stem, and leaves (0.5 g) were weighed into a 50 mL conical flask. Ten mL of nitric acid was added and kept for 24 h at 25°C. Then 4 mL perchloric acid was added to each flask and heated on a hot plate until fuming started and the color of the solution became white. The contents were cooled by adding 100 mL of distilled water. The content was filtered through filter paper (Whatman No. 42) for elemental analysis. Estimation of metals was carried out on a flame atomic absorption spectrophotometer [FAAS] (Polarized Zeeman Hitachi 2000) and a flame photometer was used to evaluate the material in triplicate (Jenway PFP7, UK). The materials of the reference elements were obtained from Merck (Darmstadt, Germany). Each metal calibration standard was made by diluting the stock solutions appropriately.

The samples were analyzed using Atomic Absorption Spectrometer for the quantitative detection of different macro and microelements following the literature.[24]

Fatty Acids

Preparation of n-hexane Extract

The finely dry powdered of each morphological part i.e., root, stem and leaves of *P. hysterophorus* (100 g) was extracted with n-hexane at room temperature for 72 h. These mixtures were then filtered using Whatman No. 42 filter paper to remove the debris and the extracts were then rotary evaporated to obtain total lipids. Procedure for preparation of Methyl Ester

To prepare the sample for GC-MC, weighed 30-40 mg of the fats from vial through top load electric balance in screw cap test tube and added 1.5 ml of methanolic NaOH (to lower the boiling point of fats) covered with a lid and boiled for five minutes in a water bath, cooled the sample and added 2.5 ml of 10% BF (Boron fluoride) solution to test tube and boiled for 30 seconds and covered with a lid to avoid evaporation. After 30 seconds removed it from heat and cooled, added 5 ml saturated NaCl solution and 1ml hexane and shook vigorously for 1-2 minutes and stored for five minutes to remove the upper oils layer with the help of a micropipette in transferred to a test tube. Again added 1ml hexane to the test tube that contained the solution and shook for 1-2 minutes vigorously and allowed to move the layer of oils in the test tube to shift to stand for some time. With the help of a micropipette removed the fixed oils layer from the test tube again into another test tube. The respective sample was passed through GC-MS and observed the fatty acids found in the samples.

GC Analysis of Fatty Acids

An Agilent 6890 GC was used to analyze the essential oil, which was equipped with an HP-5MS capillary column (30 m 0.25 mm i.d. 0.25 m) and an Agilent 5973 Mass selective detector. An EI ionization device with a 70 eV ionization energy was employed for GC-MS detection. At a flow rate of 0.8 ml/min, helium was employed as the carrier gas. The injector temperature was set at 290°C, and the column temperature was held at 50°C for 5 minutes before gradually increasing to 240°C at a rate of 4°C/min, and then to 300°C at 15°C/min for 3 minutes.

GC Analysis of Fatty Acids

Each morphological part of the root, stem and leaves of *P. hysterophorus* (100 g) were extracted with n-hexane for 72 h. The extract was rotary evaporated to obtain total lipids. Fatty acids of total lipids were trans-esterified into FAME (Fatty Acid Methyl Ester) using N-trimethyl sulfonium hydroxide (Macherey-Nagel, Duren, Germany) according to Arens et al.[25] FAME was identified on a Shimadzu GC-14A equipped with a flame ionization detector (FID) and C-R4AX chromatopac integrator (Kyoto, Japan).

RESULTS

Mineral Composition of Various Parts of *P. hysterophorus* in the Summer and Winter Seasons

The mineral composition of different morphological parts of *P. hysterophorus* in the summer season is presented in Table 1. Calcium (Ca) was absent in all the investigated parts. The stem had the highest concentration (9.95 mg/L) of Magnesium (Mg) while the root had the least (8.41 mg/L). Potassium (K) was most abundant in the leaf (13.1 mg/L5 and the least (1.26 mg/L) in the root. Cadmium (Cd) was detected in all the plant parts except the root part. Chromium (Cr) was detected in two parts roots and stem and was absent in leaf and inflorescence. The stem of *P. hysterophorus* had copper detected at 0.07 mg/L. Iron was highest (0.47 mg/L) in the leaves while it was lowest (0.15 mg/L) in the inflorescence. The maximum lead (Pb) was detected in leaves than in other parts while the inflorescence showed less the zinc (Zn) than in other parts (Table 1).

In the winter season collection, calcium was only detected in the leaves of *P. hysterophorus* at 10.4 mg/L. Magnesium was detected in all plant

Table 1. Mineral composition of morphological parts of *Parthenium hysterophorus* in the summer season

Plant part	Macro-element (mg/mL)			Macro-element (mg/mL)					
	Ca	Mg	K	Cd	Cr	Cu	Fe	Pb	Zn
Root	-	8.41	1.26	0	0.02	0.09	0.31	0.06	1.02
Stem	-	9.95	2.61	0.04	0.04	0.07	0.27	0.23	1.41
Leaves	-	9.84	13.1	0.01	0	0.08	0.47	0.62	1.07
Inflorescence	-	9.63	10.9	0.02	0	0.06	0.15	0.06	0.63
Mean value	-	9.46	6.97	0.01	0.01	0.08	0.3	0.24	1.03

Table 2. Mineral composition of various parts of *Parthenium hysterophorus* in the winter season

Plant part	Macro-element (mg/mL)			Macro-element (mg/mL)					
	Ca	Mg	K	Cd	Cr	Cu	Fe	Pb	Zn
Root	-	10	21.2	0	0	0.2	0.32	0.12	0.81
Stem	-	10.2	0	0	0.11	0.36	0.76	0.27	13.13
Leaves	10.4	10.5	3.48	0.01	0	0.18	0.34	0.14	6.03
Inflorescence	-	10.3	3.33	0.01	0.08	0.51	0.18	0.6	10.3
Mean value	2.61	10.2	6.99	0.03	0.05	0.31	9.32	0.28	7.57

Table 3. Fatty acids profile in various parts of *P. hysterophorus* in the summer season

Compounds	Percentage (%)		
	Root	Stem	Leaves
Saturated fatty acids			
Lauric acid, methyl ester	-	-	-
Myristic acid, methyl ester	-	0.01	0.01
Palmitic acid, methyl ester	0.17	0.23	0.11
Stearic acid, methyl ester	0.02	0.03	0.02
Arachidic acid, methyl ester	-	-	-
Total	0.19	0.27	0.14
Unsaturated fatty acids			
Oleic acid, methyl ester	0.06	0.06	0.06
Elaidic acid, methyl ester	-	0.01	0
Linoleic acid, methyl ester	0.5	0.34	0.43
0.Linolenic acid, methyl ester	-	-	0.18
Total	0.56	0.41	0.67
Grand total	0.75	0.68	0.81

Table 4. Fatty acids profile in various parts of *P. hysterophorus* in the winter season

Compounds	Percentage (%)		
	Root	Stem	Leaves
Saturated fatty acids			
Lauric acid, methyl ester	0	-	-
Myristic acid, methyl ester	0.01	0.01	0.04
Palmitic acid, methyl ester	0.3	0.2	0.29
Stearic acid, methyl ester	0.04	0.08	0.08
Arachidic acid, methyl ester	-	-	0.02
Total	0.35	0.29	0.43
Unsaturated fatty acids			
Oleic acid, methyl ester	0.1	0.17	0.29
Elaidic acid, methyl ester	0.01	0.01	0.02
Linoleic acid, methyl ester	0.86	1.48	1.36
Linolenic acid, methyl ester	-	0.2	0.82
Total	0.97	1.86	2.49
Grand total	1.32	2.15	2.92

parts with almost an equal amount. Potassium was detected at approximately double the value (21.1 mg/L) obtained for Mg in the root while the K was not detected in the stem. Cadmium was not detected in root and stem while the leaves and inflorescence showed similar results. Similarly, the chromium was not detected in the root and leaves while other parts showed traces. Copper was detected at the concentrations of 0.51 mg/L and 0.18 mg/L in the inflorescence and leaf parts respectively. Iron was found at 0.76 mg/L in the stem and 0.18 mg/L in the inflorescence. Lead was detected in the inflorescence and root parts at 0.6 mg/L and 0.12 mg/L levels respectively. Zinc was detected in both the stem and root of *P. hysterophorus* at 13.13 mg/Land and 0.81 mg/L respectively (Table 2).

Fatty Acids in Different Parts of *P. hysterophorus* in the Summer and Winter Seasons

In summer season, the concentration of both the saturated and unsaturated fatty acids showed variation in all the tested plant parts (Table 3). Among the tested saturated fatty acids, the lauric acid, methyl ester was not detected in any tested plant parts. The myristic acid, methyl ester was absent in the root while present in an equal amount in both stem and leaves. The concentrations of palmitic acid, methyl ester and stearic acid, methyl ester showed variation while the arachidic acid, methyl ester was not detected in all the tested plant parts (Table 4). The saturated fatty acid oleic acid, methyl ester was reported in all plant parts with an equal concentration, while the elaidic acid, methyl ester was not detected in the root, and with zero concentration in leaves. Both the linoleic acid, methyl ester and octadecadienoic acid, methyl ester varied among the different plant parts, while the linolenic acid, methyl ester was only detected in leaves compared to other plant parts (Table 3).

In the winter season, among the saturated fatty acids, the lauric acid, methyl ester was reported absent in all plant parts. The concentration of myristic acid, methyl ester, palmitic acid, methyl ester and stearic acid, methyl ester varied in different plant parts, while the arachidic acid, methyl ester was only detected in leaves (Table 4). In the case of unsaturated fatty acids, all the tested plant parts showed variation in the saturated fatty acids while the linolenic acid, and methyl ester were not detected in the root while stem and leaves showed variation in concentration (Table 4).

DISCUSSION

Our study showed variation in mineral and both saturated and unsaturated fatty acids compositions in different parts of *P. hysterophorus*

and the concentration was maximum in the winter than in the summer season. Studies have shown the differences in the composition of the elements in both root and leaves of *Elephantopus scaber*. Jabeen et al.,[26] and Kumar et al.,[27] studied the differences in the composition of the element in both root and leaves of *Elephantopus scaber* and reported that Ca, Cd, K, Mg, P, S, and Si were abundant in leaves, whereas Al, Fe, Sr, V, and Ti were more in the root while As, Cu, Rb, Sr and Zn were present equally both in leaves and root of E. scaber. The roots, stem and leaves of various medicinal plants including *Ambrosia maritime, Balanites aegyptiaca, Cymbopogon proximus, Grewia tenax, Hydnora johannis, Lepidium sativum, Nauclea latifolia, Peperomia pellucida, Senna obtusifolia* and *Tamarix aphylla* were evaluated for eleven elements (K, Zn, Ca, Cu, Mn, Fe, Cr, Br, Ni, Zr, and Rb).[28] *G. tenax* had the maximum concentration of Br (33 ppm), Ca (8107 ppm), Rb (19.9 ppm), Cu (15.9 ppm), and Zn (18.11 ppm). *Hydnora johannis* showed the highest concentration of Fe (1426 ppm) and K (15451 ppm). The maximum concentration of Ni (7.6 ppm) and Cr (13.10 ppm) was found in *Cymbopogon proximus*. *Ambrosia maritime* and *Balanites aegyptiaca* had a maximum concentration of Mn (10.6 ppm) and Zr (8.2 ppm). The elements showed seasonal variation. Lead was detected at the highest concentration of 0.620 mg/L in the leaf and 0.06 mg/L in the root. Zinc was detected at 1.41 mg/Land 0.634 mg/L in both stem and inflorescence respectively.[28]

Fatty Acids Composition of *P. hysterophorus* in the Summer and Winter Seasons

Considering the summer season (Table 3), roots contained five fatty acids of which two were saturated fatty acids namely palmitic acid (0.17%), as the major component and stearic acid (0.02%). The unsaturated fatty acids were oleic acid (0.06%) and linoleic acid (0.50%). The saturated fatty acids were lower (0.19%) in root than unsaturated fatty acids (0.74%). Stem revealed the presence of seven fatty acids; three saturated fatty acids namely myristic acid (0.01%), palmitic acid (0.23%) and stearic acid (0.03%) and three unsaturated fatty acids namely oleic acid (0.06%), elaidic acid (0.01%) and linoleic acid (0.34%). The total saturated fatty acids were detected at a lower level (0.27%) in the stem as compared to unsaturated fatty acids (0.68%) (Table 3). Leaves contained eight fatty acids, three were saturated fatty acids including myristic acid (0.01%), palmitic acid (0.11%), and stearic acid (0.02%). Five unsaturated fatty acids including oleic acid (0.06%), elaidic acid (0.00%), linoleic acid (0.43%) and linolenic acid (0.18%) were detected.

Considering the wither season (Table 4), roots contained eight fatty acids of which four were saturated namely, lauric acid (0.00%), myristic acid (0.01%), palmitic acid (0.30%), and stearic acid (0.04%) and four unsaturated namely oleic acids (0.10%), elaidic acid (0.01%). The stem comprises eight saturated and unsaturated fatty acids ester including; myristic acid (0.01%), palmitic acid (0.20%), and stearic acid (0.08%). The five unsaturated fatty acids were oleic acid (0.17%), elaidic acid (0.01%), linoleic acid (1.48%), octadecadienoic acid (0.42%) and linolenic acid (0.20%) (Table 4). Leaves contained nine fatty acids of which four were saturated fatty acids namely myristic acid (0.04%), palmitic acid (0.29%), stearic acid (0.08%), and arachidic acid (0.02%). Five unsaturated fatty acids were detected including oleic acid (0.29%), elaidic acid (0.02%), linoleic acid (1.36%), octadecadienoic acid (0.72%) and linolenic acid (0.82%). The study revealed that total saturated fatty acids were at the highest level in roots (0.35%) and the lowest level in stem (0.29%). The bioactivity is correlated due to the presence of fatty acids present in higher concentrations.

Kravtsova and Khasanov[29] investigated the fatty acid composition of roots, stem, and seeds of *Arctium lappa* and the leading compound was linoleic acid. Mozzon and co-workers studied the Elaeis oleifera for fatty acids composition, wherein stearic, palmitic, and linoleic acids were reported. The dominant fatty acid was lauric acid. Results showed that lauric acid is one of the useful fatty acids and used for acne treatment as well as helpful in the HDL (High-density lipoprotein) good blood cholesterol.[30] Mukherjee et al. observed that leaves of *Momordica cochinchinensis* had the highest fatty acids and palmitic acid was the dominant compound among the fatty acids. The fatty acids helped in the production of biodiesel. [31] Ahmad et al. studied the composition of root,

stem, leaves, and receptacle of *P. hysterophorus*. Under this study, screening of phytochemicals showed the presence of fatty acids, terpenes, hydrocarbons, and sterols, while the individual part-specific maximum concentration of bioactive compounds was hexadecanoic acid and methyl ester in the stem.[17]

CONCLUSION

Our findings provide evidence that the elemental and fatty acids compositions and concentration varied not only in different parts of a plant species but also depending on the seasons in which the plant species were collected. Fatty acid composition and elemental profiling of different morphological parts of *Parthenium hysterophorus* showed seasonal variation. Micro and macro elements and fatty acids were higher in the winter season compared to the summer season.

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CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

AUTHORS' CONTRIBUTION

All authors listed have made a substantial, direct and intellectual contribution to the work, and approved it for publication.

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None.

DATA AVAILABILITY

All datasets generated or analyzed during this study are included in the manuscript.

ETHICS STATEMENT

Not Applicable.

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