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# Essential oil composition and nutrient analysis of selected medicinal plants in Sultanate of Oman

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# PEER REVIEW

#### **Peer reviewer**

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#### Comments

In light of main theme of the article, the authors have tackled one of the neglected aspects of medicinal plants. Mostly the local people used these plants just to cure ailments but do not use in their routine life. Because of this negligence they are deprived from essential nutrients. The manuscript is well presented and the new findings will not only benefit the local people but also the rest of world. Details on Page 426

# ABSTRACT

**Objective:** To evaluate the nutrients and essential oils of five medicinal plants, *Juniperus excelsa* (*J. excelsa*), *Dodonaea viscosa*, *Euryops pinifolius*, *Teucrium polium* (*T. polium*), and *Helianthemum lippii* that were collected from Jabal Al Akhdar, Oman.

**Methods:** Proximate parameters (moisture, dry matter, ash, crude fats, proteins, fibers, nitrogen, carbohydrates, and energy values) and nutrient analysis (K, Na, Ca, Fe, P, Mg *etc.*) were evaluated in the five medicinal plants using standard techniques. On the basis of these analysis, *T. polium* and *J. excels* were selected for essential oil analysis using a rapid solvent–free microwave extraction method and GC–MS.

**Results:** The results showed that leaves of *J. excelsa* had highest proportion of crude fats, fibers and energy value while ash was highest in *T. polium. J. excelsa* was also rich in essential minerals such as calcium, magnesium, potassium and iron while the trace elements and heavy metals composition was marginal. A rapid solvent–free microwave extraction method to extract oil from medicinal plants species showed that only *T. polium* and *J. excelsa* yielded oil. The chemical composition of essential oils showed higher proportion of delta–3–carene, limonene,  $\beta$ –eudesmol, ledeneoxide (II),  $\alpha$ –trans–bergamatene, linalyl acetate and germacrene.

**Conclusions:** *J. excelsa* and *T. polium* are a good source of proximate, minerals and essential oils, which can be considered for healthy life besides their medicinal values.

#### KEYWORDS

Proximate parameters, Essential oils, Omani medicinal plants, GC-MS

# **1. Introduction**

Plants are the primary source of medicines, food, and shelter for humans. Besides curing ailments, various plant parts like roots, stems, leaves, flowers, fruit and seeds can act as a food resource for human<sup>[1]</sup>. These are indispensable constituent of human diet supplying the body with minerals salts, vitamins and certain hormone precursors, in addition to protein and energy<sup>[2]</sup>. Plant–derived natural products can be used for the treatment of diseases, thus can act as a base for development of natural blueprint of new drugs<sup>[3]</sup>. There is high need for a constant search of new resources to alleviate hunger in developing countries. Predictions of the future needs based on the current rates of increasing population and food production emphasize the seriousness of this problem<sup>[4]</sup>.

In the present study, five medicinally important plant species were collected from Jabal Al–Akhdar, Northern Oman). Jabal Al–Akhdar has been known for its rich diversity in floral resources. It is one of the local centers of endemic plants of Sultanate of Oman and some of the Arabian countries. It is

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one of the WWF's global 200 eco-regions "Arabian Highland Woodlands and Shrublands" (World Wide Fund for Nature, Switzerland<sup>[5]</sup>. The area is bestowed with ample plant resources and its dominant vegetation is *Juniperus excelsa* subsp. *polycarpos* (*J. excelsa* subsp. *polycarpos*). It is shown to be vulnerable according to the IUCN Red List<sup>[6]</sup>. The local people of the area use it for stomachache and diabetes, in addition to wood harvesting and grazing<sup>[7]</sup>. Studies have shown its positive effects in treating dysmenorrheal<sup>[8]</sup>, cough<sup>[9]</sup>, bronchitis and colds<sup>[10]</sup>, jaundice and tuberculosis<sup>[11]</sup>, and to induce menses and expel fetus<sup>[12,13]</sup>.

Dodonaea viscosa (Sapindaceae) (D. viscosa) is also an important medicinal plant which is used for anti-inflammatory, antiviral, spasmolytic, laxative, antimicrobial and hypotensive agents<sup>[14]</sup>. The leaves were reported to possess local anesthetic, smooth muscle relaxant<sup>[15]</sup>, antibacterial<sup>[16]</sup>, antifungal<sup>[17]</sup>, antiinflammatory<sup>[18]</sup>, and anti-ulcerogenic activities. Teucrium species have been used as medicinal herbs for over 2000 years as diuretic, diaphoretic, tonic, antipyretic, antispasmodic, cholagogic and many of them are used in folk medicine<sup>[19]</sup>. Anti-inflammatory, antihypertensive and anorexic effects are other reported activities of *Teucrium polium* L (T. polium)[20,21]. In the local areas, it is used frequently for the treatment of gastrointestinal or abdominal ailments and for its hypoglycemic properties<sup>[21]</sup>. The local uses of Euryops pinifolius (E. pinifolius) and Helianthemum lippii (H. lippii) are least known, however, in other parts of the Arabian Peninsuala, these are used in analgesic and anti-inflammatory[22].

In the current study, the proximate composition and essential oil extraction of the selected medicinal plants was investigated. There are various methods available to extract essential oils; however we used microwave–assisted essential oil extraction. The method has been regarded efficient to avoid disintegration of fragile and small volatile components of oil<sup>[23]</sup>. The essential oils of *T. polium* and *J. excelsa* have been previously quantified however, looking at the variation in the composition; these have been analyzed for the first–time from the Oman along with other medicinal plants. The present study was aimed at analyzing the proximate parameters, mineral compositions and essential oils of *D. viscosa, T. polium, J. excelsa, E. pinifolius* and *H. lippii*.

#### 2. Materials and methods

#### 2.1. Sample collection and identification

The medicinal plants (*D. viscosa, T. polium, J. excelsa, E. pinifolius* and *H. lippii*) were collected from Al Jabal Al Akhdar, Oman. The plants were identified by the plant taxonomist Mr. Ghanam Salim Said and were kept under subdued light prior to proximate analysis. The dried matter obtained was ground to powder using grinder (IKA, WARKE, MF 10 B, USA). All the plants were packed in the kraft paper and herbarium sheets were prepared.

# 2.2. Proximate analysis

The proximate parameters (moisture, dry matter, ash, crude fats, proteins and fibers, nitrogen, carbohydrates and energy values) were determined using Association of Official Analytical Chemists Methods<sup>[24,25]</sup>. Determination of moisture content was done by drying samples in oven (WiseVen, WON-50, Korea) at 110 °C until constant weight was attained<sup>[24]</sup>. Nitrogen estimation was carried out by the micro-Kjeldahl (BUCHI, KjelFlex K-360, Switzerland) method with some modification<sup>[25]</sup>. The crude proteins were subsequently calculated by multiplying the nitrogen content by a factor of 6.25<sup>[25]</sup>. The energy value estimation was done by summing the multiplied values for crude protein, crude fat and carbohydrate respectively at Water Factors (4, 9 and 4). Crude fats were determined by Soxhlet apparatus using n-hexane as a solvent. The ash values were obtained by heating samples at 550 °C in a muffle furnace (Wise Therm, FHP-03, Korea) for 3 h. The carbohydrate content was determined by subtracting the total crude protein, crude fiber, ash content and crude fat from the total dry matter<sup>[24]</sup>. Crude fiber was estimated by acid-base digestion with 1.25% H<sub>2</sub>SO<sub>4</sub> (v/ v) and 1.25% NaOH (w/v) solutions[26].

#### 2.3. Macro and micro nutrients analysis

The elements profile of the selected medicinal plants were analyzed using Inductively Coupled Plasma Emission Spectrometer (ICP-OES DV 7300, Perkin Elmer, USA) equipped with Perkin Auto-Sampler with the following parameters: plasma flow rate (15 L/min), nebulizer flow rate (0.8 L/min), RF power (1500 Watts), auxilliary flow rate (0.2 L/min), sample flow rate (1.25–2.50 L/min), torch position (-3) for aqueous samples and 15 sec equilibration. The dried homogenized sample (0.5 g) was taken in Kjeldahl tube (250 mL) and digested with 20 mL of 98% sulfuric acid (Sigma Aldrich) at 370 °C to a colorless liquid. The resultant liquid was diluted with distilled water up to 100 mL and filtered using Whatman-42 filter paper.

# 2.4. Rapid solvent-free microwave extraction (SFME) of essential oils

SFME was carried out with a microwave essential oil system (MILESTONE Technologies, NEOS, Italy) with a maximum delivered power of 900 W variables in 10 W increments and 650 nm wave length. During experiment, time, pressure and power were controlled with the "easy–WAVE" software. Among these medicinal plants, we were only able to extract oil from *J. excelsa* and *T. polium*. Fresh samples of *J. excelsa* and *T. polium* (100 g each) were heated using a fix power of 400 W for 45 min at 100 °C without addition of solvents. The extraction was continued until no more essential oil was obtained. The essential oils were collected, dried over anhydrous sodium sulphate and stored at 4 °C until further used. Extractions were performed at least three times and the mean values are presented.

# 2.5. Essential oil quantification

GC-MS analysis was performed on a Perkin Elmer model (GC-MS-Clarus 600, US) instrument. Analytes were separated on a 30 m×0.25 mm non polar capillary column with a phase thickness of 0.25  $\mu$ m and interfaced with a quadrupole mass spectrometer. The injector and interface temperature were kept at 290 °C and 260 °C respectively and the temperature was programmed from 60 °C to 280 °C at a rate of 3 °C /min. Helium was used as the carrier gas with a linear velocity of 36.7 cm/s. The MS ionization voltage was 70 V.

# 2.6. Statistical analysis

Data obtained were subjected to analysis of variance (ANOVA) and duncan multiple range test (DMRT) using Statistic Analysis System (SAS 9.1). Standard deviation and average values were calculated using Microsoft Excel 2003. Statistically non–significant difference was observed at P<0.05.

# 3. Results

# 3.1. Proximate composition

In current study, it was observed that the moisture content was significantly (P < 0.05) higher in *H. lippii* followed by *T. polium* and *E. pinifolius* (Table 1). The dry mater was significantly higher in *J. excelsa* and *D. viscosa*. Ash contents were significantly higher in *T. polium*. Crude fats, fibers and energy values were significantly higher in *J. excelsa*. Nitrogen and crude proteins were significant in *E. pinifolius* while carbohydrates were higher in *D. viscosa* and *H. lippii* (Table 1).

# 3.2. Nutrient profile analysis

The composition of essential elements in the five medicinal

#### Table 1

Comparative assessment of proximate parameters of selected medicinal plants.

<u>^</u>	*	<u>^</u>		*					
Medicinal plants	Moisture content	Dry matter	Ash content	Crude fats	Nitrogen	Crude proteins	Crude fibers	CHO	Energy value
J. excelsa	$6.90 \pm 0.41^{\circ}$	$93.10 \pm 0.41^{a}$	$5.90 \pm 0.14^{d}$	$8.20 \pm 0.26^{a}$	$1.60 \pm 0.02^{b}$	$9.80\pm0.1^{\mathrm{b}}$	$31.60 \pm 0.29^{a}$	$77.80 \pm 0.36^{b}$	$389.20 \pm 1.83^{a}$
D. viscosa	$6.70 \pm 0.05^{\circ}$	$93.20 \pm 0.05^{a}$	$4.90 \pm 0.30^{d}$	$6.70 \pm 0.13^{\circ}$	$1.50 \pm 0.04^{b}$	$9.60 \pm 0.26^{b}$	$12.70 \pm 0.13^{d}$	$80.30 \pm 0.40^{a}$	$387.20 \pm 0.77^{a}$
E. pinifolius	$7.40 \pm 0.66^{b}$	$92.50 \pm 0.66^{b}$	$9.80 \pm 0.17^{b}$	$7.80 \pm 0.28^{\mathrm{b}}$	$1.90 \pm 0.01^{a}$	$12.30 \pm 0.12^{a}$	$15.60 \pm 0.05^{d}$	$73.90{\pm}0.68^{\rm c}$	$370.20 \pm 1.08^{b}$
T. polium	$7.60 \pm 0.09^{b}$	$92.30 \pm 0.09^{b}$	$15.60 \pm 0.42^{a}$	$4.90 \pm 0.01^{d}$	$1.40 \pm 0.02^{\circ}$	$9.10 \pm 0.16^{\circ}$	$20.20 \pm 0.09^{b}$	$70.80 \pm 0.45^{\circ}$	$331.50 \pm 1.79^{\circ}$
H. lippii	$8.10 \pm 0.07^{a}$	$91.80 \pm 0.07^{\circ}$	$7.80 \pm 0.23^{\circ}$	$1.10 \pm 0.05^{\mathrm{ef}}$	$1.80\pm0.01^{ab}$	$11.10 \pm 0.09^{b}$	$18.10 \pm 0.09^{\circ}$	$79.90 \pm 0.28^{a}$	$341.70 \pm 1.41^{\circ}$

CHO: Carbohydrates. The different letter(s) in each column shows values are significantly different (P<0.05) as evaluated by the DMRT. ±shows the standard deviation of mean values of three replicates.

Table 2

Non-essential nutrients/metals contents in the medicinal plants (ng/g dry weight).

Medicinal plants	Ag	Al	В	Ba	Si	Sr	Ti	V	W	Zn
D. viscosa	$0.224 \pm 0.040$	0.441±0.030	$0.720 \pm 0.010$	$0.024 \pm 0.030$	$0.168 \pm 0.040$	0.91±0.20	ND	ND	$0.162 \pm 0.050$	$0.093 \pm 0.010$
T. polium	$0.224 \pm 0.050$	$0.986 \pm 0.090$	$0.942 \pm 0.020$	$0.046 \pm 0.060$	$0.122 \pm 0.020$	1.15±0.11	0.51±0.07	ND	$0.159 \pm 0.010$	ND
J. excelsa	$0.224 \pm 0.020$	$0.887 \pm 0.070$	$0.890 \pm 0.080$	0.037±0.050	$0.151 \pm 0.050$	$0.66 \pm 0.10$	$0.05 \pm 0.03$	ND	$0.162 \pm 0.030$	$0.030 \pm 0.013$
E. pinifolius	0.224±0.010	$0.627 \pm 0.040$	0.731±0.090	$0.040 \pm 0.020$	$0.174 \pm 0.070$	$1.01 \pm 0.20$	ND	$0.028 \pm 0.010$	$0.163 \pm 0.020$	$0.084 \pm 0.010$
H. lippii	$0.224 \pm 0.030$	$0.969 \pm 0.030$	0.527±0.020	$0.053 \pm 0.030$	$0.135 \pm 0.040$	$0.75 \pm 0.02$	0.46±0.20	0.014±0.010	$0.160 \pm 0.010$	ND

ND: Not detected. The values with ± shows standard deviation of the mean values of the three replicates of each element analyzed through ICP-OES.

plants is given in Figure 1. In case of calcium (Ca), it was much higher in *T. polium* (55.7) and *H. lippii* (50.4) while in other medicinal plants like *D. viscosa* (15.7), *J. excelsa* (30.9), and *E. pinifolius* (30.2), its contents were much lower. The magnesium (Mg) content was 2.2, 5.2, 2.1, 3.6, and 4.8 in *D. viscosa, T. polium, J. excelsa, E. pinifolius* and *H. lippii* respectively. The sodium (Na) contents were higher in *E. pinifolius* (4.1) while in other plants, it was significantly lower (Figure 1). Potassium (K), on the other hand, was significantly higher in *E. pinifolius* (30.10) as compared to *D. viscosa* (10.77), *T. polium* (17.10), *J. excelsa* (13.04) and *H. lippii* (5.32). Iron (Fe) content was too low as compared to P. The Fe content was 0.266, 0.837, 0.774, 0.453 and 0.830 in *D. viscosa, T. polium, J. excelsa, E. pinifolius* and *H. lippii* respectively. The phosphorus (P) content ranged from 1.03 to 2.30 in the medicinal plants.



**Figure 1.** Composition of essential nutrients *i.e.* calcium (Ca), magnesium (Mg), sodium (Na), phosphorus (P), potassium (K), and iron (Fe) in the medicinal plants.

Values are presented as the mean values of the three replicates (Mean±SE).

In case of non-essential elemental compositions of medicinal plants, 28 different elements (silver-Ag; aluminuim-Al; barium-Ba; copper-Cu; beryllium-Be; bismuth-Bi; arsenic-As; gold-Au; cadmium-Cd; cobalt-Co; chromium-Cr; lithium -Li; nickel-Ni; lead-Pb; plutonium-Pt; Antimony-Sb; silicon-Si; tin-Sn; strontium-Sr; titanium-Ti; selenium-Se; thallium-

Table 3, continued

Tl; vanadium–V; tungestun–W) were analyzed. During analysis enough quantities of As, Au, Be, Bi, Cd, Co, Cr, Cu, Li, Mn, Mo, Ni, Pb, Pt, Sb, Se, and Sn were not detected by ICP–OES. However, appreciable quantities of Ag, Al, B, Ba, Si, Sr, Zn, Ti, V and W were observed in the medicinal plants samples of *D. viscosa*, *T. polium*, *J. excelsa*, *E. pinifolius* and *H. lippii* (Table 2). The contents of Ag were 0.224 ng/g dry weight and it was constant throughout plant samples. The Al ranged from 0.441 to 0.969 ng/ g dry weight in all medicinal plants whilst the *H. lippii* had the highest. The B was ranged from 0.527 to 0.942 ng/g dry weight, Si ranged from 0.122 to 0.174 ng/g dry weight, Sr ranged from 0.66 to 1.15 ng/g dry weight, and W ranged from 0.153 to 0.163 ng/g dry weight in medicinal plants. Ba, Ti, V and Zn were very low in their contents in all medicinal plants species (Table 2).

# 3.2. Essential oil composition

The detailed chemical profile of *T. polium* and *J. excelsa* is given in Table 3. Approximately 0.42% and 0.25% of essential oil was extracted from a 100 g of plant sample of T. polium and J. excelsa respectively. Total number of chemical constituents identified from J. excelsa were 50. The analysis showed that the contents and percentage of  $\alpha$ -Pinene (4.81%), delta-3-Carene (5.88%), DL-Limonene (49.58%), y-Cadinene (3.76%), and germacrene (9.6%), delta-Cadinene (2.31%), delta-Cadinol (2.41%) were highest in J. excelsa (Table 3). The composition of  $\beta$ -Myrcene,  $\alpha$ -Terpinene,  $\gamma$ -Elemene, and  $\alpha$ -Cadinol were ranged from 1.0% to 1.5%. The remaining 36 constituent's percentage was less than 1% (Table 3). In case of essential oil from T. polium, only 34 chemical constituents were identified and given in Table 3. The percentage composition of ledeneoxide (II) (20.47%), linalyl acetate (11.16%), β-Eudesmol (11.59%), and  $\alpha$ -trans-Bergamatene (6.81%) were higher as compared to other 30 chemicals.

#### Table 3

Chemical composition of essential oils extracted from *J. excelsa* and *T. polium*.

Compound names	Rt (min)	Ri	J. excelsa (%)	T. polium (%)
α–Pinene	4.5	932	4.81	0.45
α–Fenchene	4.7	944	0.14	-
Sabinene	5.4	971	0.05	-
β–Pinene	5.4	975	0.14	0.27
β–Myrcene	5.8	990	1.06	0.07
α–Phellandrene	6.2	1005	0.03	-
delta-3-Carene	6.4	1011	5.88	-
α–Terpinene	6.5	1016	0.08	
DL-Limonene	7.1	1032	49.58	1.05
γ–Terpinene	7.8	1056	0.61	-
α–Terpinene	8.8	1087	1.25	-
Linalool	9.2	1101	-	1.66
α–Camphenolene aldehyde	10.1	1125	0.02	0.04
trans–Limonene oxide	10.6	1137	0.03	-
Camphor	10.8	1143	0.02	-

Rt: Retention time, Ri: Retention Index. Amount of essential oil in *Teucrium polium*: 0.42% (100 g); Amount of essential oil in *Juniperus excelsa*: 0.25% (100 g). Conditions: 400 P(W), 110 °C, 45 min.

Chemical composition of essential oil	s extracted	from	J. excelsa an	d T. polium.
Compound Name	Rt (min.)	Ri	J. excelsa	T. polium
Cis-Pulegol	10.8	1144	0.03	-
4–Terpineol	12.1	1176	0.07	-
$\alpha$ –Terpinol	12.6	1190	-	0.11
Myrtenal	12.8	1195	-	0.04
β–Citronellol	14.2	1230	0.02	-
Carvone	14.7	1243	0.03	-
Linalyl acetate	15.3	1258	-	11.16
Bornyl acetate (END)	16.3	1284	0.05	0.31
2,4- decadienal (2E,4Z)	16.7	1293	0.16	-
2,4- decadienal (2E,4E)	17.6	1316	0.16	-
Delta–Elemene	18.4	1335	0.23	-
α–Cubebene	18.9	1347	0.03	-
$\alpha$ –Terpinyl acetate	18.9	1349	-	0.44
α–Copaene	19.9	1372	0.08	0.21
β–Bourbonene	20.2	1381	0.09	0.17
Gernyl acetate	20.4	1386	-	0.99
β–Elemene	20.6	1389	0.32	0.32
trans– Caryophyllene	21.6	1415	0.99	1.11
γ–Elemene	22.3	1431	1.18	_
$\alpha$ -trans-Bergamatene	22.4	1435	_	6.81
α–Cubebene	22.7	1443	0.02	_
α–Humulene	22.9	1449	0.97	0.41
Aromandrene	23.2	1457	_	1.26
EPI- bicyclosesquiphellandrene	23.3	1459	0.08	_
$\alpha$ -Amorphene	23.9	1474	0.21	0.08
Germacrene D	24.1	1477	_	0.34
Germacrene D	24.1	1478	2.44	_
β-Selenine	24.3	1482	0.06	1.34
cis-B-Guaiene	24.5	1489	_	0.57
$\alpha$ -Selenine	24.7	1492	0.19	0.52
$\alpha$ – muurolene	24.9	1498	0.36	_
Germacrene A	25.0	1501	0.17	_
β– Bisabolene	25.0	1507	_	0.32
v–Cadinene	25.5	1512	3 76	_
Delta– Cadinene	25.8	1523	2 31	2 41
$\alpha_{-}$ amorphene	26.3	1525	0.37	_
Aromandrene	26.5	1538	0.11	_
Flemol	26.9	1550	0.60	_
Cermacrene B	26.0	1553	_	0.44
Cermacrene B	20.9	1556	7 16	-
Ledeneovide (II)	27.1	1564	7.10	20.47
Nerolidol	27.5	1566	0.02	0.21
Cormagrana D. 4. al	27.5	1574	0.02	1.21
Patchulano	27.0	1520	-	1.21
() Comenbullene evide	20.0	1500	0.08	-
(-) Caryophynene oxide	28.1	1581	-	1.40
<i>c</i> Orlanger and ethyl ester	28.0	1596	0.03	-
p-Optopenone	28.9	1605	0.19	-
dalta Cadinal	29.7	1626	-	0.24
dena-Cadinol	30.2	1640	2.41	-
p-Eudesmol	30.5	1650	-	11.59
a-Cadinol	30.7	1654	1.26	1.68
Juniper camphor	32.0	1692	0.13	-
Unidentified			9.84	30.3
Total			90.16	69.7

Rt: Retention time, Ri: Retention Index. Amount of essential oil in *Teucrium polium*: 0.42% (100 g); Amount of essential oil in *Juniperus excelsa*: 0.25% (100 g). Conditions: 400 P(W), 110 °C, 45 min.

# 4.1. Proximate composition

The medicinal plants viz. D. viscosa, T. polium, J. excelsa, E. pinifolius and H. lippii were subjected to analysis of various proximate parameters such as moisture, dry matter, ash, crude fats, proteins and fibers, nitrogen, carbohydrates, and energy values. These medicinal plants/parts are used to cure various ailments in the marginal communities of Oman, therefore interest was developed to know other possible health benefits of these plants. Besides using such plants as medicine, crude fats, proteins, fibers, nitrogen, and carbohydrates can be essential for diatry intake<sup>[27]</sup>.

Plants are the medicinal intrinsic worth providing essential nutrients. Proteins and carbohydrates are important nutrients to be assessed in medicinal plants<sup>[28]</sup>. All fractions (cellulose, lignin, hemicellulose, pectin, gums and mucilage) of the fiber play a significant role treating various diseases such as diabetes, constipation, diverticulosis, cardio-vascular diseases, and fatness<sup>[28,29]</sup>. In recent years, much work has been performed for producing and evaluating various sources of proteins and fibers<sup>[29]</sup>. Proteins enclose essential amino acids and have the nutritional values for human health. Similarly, Alfawaz reported protein value 17.1-20.1, moisture 87.8-93.5, and ash 14.6-19.6 in Rumex vesicarius<sup>[30]</sup>. Medicinal plants can cope with the dual problems of diseases and malnutritions. However, this needs an extensive analysis from proximate to biochemical levels to assess the role, function and composition of these medicinal plants which might be used as duo<sup>[31]</sup>.

In current study, the leaves of *J. excelsa* was proved to have significant potential value of crude fat and fiber with high energy value. The Food and Nutrition Board recommended an intake of 20-35 g of fiber per day[32]. As a nutritive value of food, fibers in the diet are necessary for digestion and for effective elimination of wastes<sup>[32]</sup>. The nitrogen and crude proteins were significant in E. pinifolius. Alfawaz reported highest protein percentage in Rumex vesicarius<sup>[31]</sup>. Similarly, Maisuthisakul et al. performed proximate analysis of some Thailand plants[33]. According to his results, the fat content of Momordica charantia was about 1.7% which is significantly lower than that of the fat contents of J. excelsa. It is interesting to note that the total amounts of fats in J. excelsa are higher than most of the common vegetables<sup>[34]</sup>. This difference in functional properties may be attributed to the role of ameliorated soil and climatic conditions of the area[35,36]. Proximate and nutrient analysis of aerial part of selected plant species plays a decisive role in assessing their nutritional significance. As medicinal plant species are also used as food along with their medicinal benefits, evaluating their nutritional contents can help to recognize the significance of these plants species.

## 4.2. Nutrient profile analysis

Nutrients rich foods are vital for proper growth both in adults and children. The present study showed the highest Ca and Mg concentration in T. polium. Mushtaq et al. observed significantly lower Ca contents in economically important medicinal plant species viz. Chenopodium ambrosiodes (1.50%) and Achyranthes aspera (1.14%)<sup>[34]</sup>. Ca is an important component of plants performing pivotal role in growth and development of plants while its intake in human can enhance the development of bones and teeth, regulates heart rhythm, helps in normal blood clotting, maintain proper nerve and muscle functions. Its deficiency causes poor development, growth and different abnormalities<sup>[37]</sup>. In previous study, Mushtaq et al. observed significantly lower concentration of Mg in Convolvulus arvensis and Achyranthes aspera which is lesser than what we found in T. polium<sup>[34]</sup>. Mg, on the other hand, plays an important role in the human body to maintain cholesterol and control heart diseases. Mg converts blood sugar into energy.

In case of trace elements, As, Au, Be, Bi, Cd, Co, Cr, Cu, Li, Mn, Mo, Ni, Pb, Pt, Sb, Se, and Sn were not detected during this analysis. This indicates that these elements are not present in a detectable amount in the plant parts. This is beneficial to consumers, since it has been reported that some of these minerals like lead, cobalt and cadmium are highly toxic even at low concentrations<sup>[38]</sup>. It has also been reported that for many plant species Cr was proved to be toxic at 5 mg/L. In this regard, all the studied plants have very lesser concentration of Cr as compared to that of recommended level for toxicity in plants<sup>[39]</sup>. In case of Pb concentration, the suggested concentration in plant species is 2 to 6 mg/L<sup>[40]</sup>, so the analyzed plant species carries much lesser level of Pb, which further clarifies their use as food supplement. Low concentration of Ag, Al, B, Ba, Cu, Zn, Si, Sr, W, and V were observed in all the selected medicinal plants. Although low quantities of these nutrients are essential for many organisms, but they are potentially toxic and accumulate in soil over long period of time and can result in soil pollution<sup>[41]</sup>.

# 4.3. Essential oil composition

All the five medicinal plants viz. D. viscosa, T. polium, J. excelsa, E. pinifolius and H. lippii were subjected for essential oil extraction through SFME technique. Essential oils from only two medicinal plants *i.e.* T. polium and J. excelsa were extracted. The chemical constituents were investigated by using GC-MS. It was interesting to note that some of the major constituents of J. excelsa such as delta-3-Carene and germacrene B were not detected in T. polium. Linalyl acetate,  $\beta$ -Eudesmol, ledeneoxide (II), and  $\alpha$ -trans-Bergamatene, on the other hand, were not detected in J. excelsa. Similarly,

DL-Limonene was only 1.05% in *T. polium* as compared to 49.58% in *J. excelsa*. The percentage composition of  $\beta$ -Pinene,  $\alpha$ -Camphenolene aldehyde,  $\beta$ -Bourbonene,  $\beta$ -Elemene,  $\alpha$ -Humulene,  $\alpha$ -Selenine,  $\alpha$ -Cadinol, and Delta-Cadinene *etc.* was not significantly different between the two medicinal plants. Overall, 9.8% and 30.4% of the chemical constituents of essential oil of *J. excelsa* and *T. polium* could not be identified.

Emami *et al.* reported abundance of monoterpenoids whilst most of the major components of essential oil of *J. excelsa* were similar to our findings<sup>[13]</sup>. The pinene and limonenes were highest in concentrations. The composition percentage of limonene was significantly higher (49.58%) in our study as compared to other previous studies. Emami *et al.* showed 4.5%<sup>[13]</sup>; Moein *et al.* showed 4.1%<sup>[42]</sup>, and Khajjak *et al.* reported 1.46%<sup>[43]</sup>. Only Adams identified 22.7% of limonene from the plant samples collected from Greece<sup>[44]</sup>. The findings of Unlu *et al.* were in conformity with our results<sup>[45]</sup>. The plant species is same but the percentage compositions of the major components are in different proportions depending on the location of the plant.

Previous study of Afifi et al. showed the chemical constituents of T. polium collected at pre-flowering stage from different arid and semi-arid habitats of Jordan<sup>[46]</sup>. According to his results, volatile compounds such as menthone, germacrene, linalool,  $\varrho$ -cymene, carvone, and  $\beta$ -caryophyllene were significantly higher in percentage. Additionally, the samples from different locations showed qualitative and quantitative variations in the oil composition. The composition and occurrence of  $\gamma$ -Elemene and germacrene was almost similar to our results. Aburjai et al. reported 37 chemicals in the oils of T. polium extracted and identified through hydrodistillation and GC-MS<sup>[47]</sup>. The major chemical was germacrene which is also similar to our findings. This is in conformity with our results as well. Cakir et al. also studied the essential oil extraction and quantification of T. polium collected from Turkey<sup>[48]</sup>. The author reported 30 chemicals with the major constituents, germacrene and pinene. Kamel and Sandra, on the other hand, compared the essential oil composition of two varieties of T. polium growing in Egypt and Qatar, reported 64 volatile components from both the oils<sup>[49]</sup>. In their study, too, the components varied between two verities and two locations. In another recent study of Mitić et al., the chemical composition of essential oil of T. polium subsp. capitatum growing in various areas of Balkan Peninsula was reported<sup>[50]</sup>. The pinenes and germacrene were the major constituents in their study. It has been previously noted that the essential oil composition may vary greatly depending on the habitat, location, climatic conditions and soil biology<sup>[51]</sup>. This varying biospheric condition regulates the internal physiology of the plant to synthesize such diverse array of chemicals within the same species[52].

The analysis of selected medicinal plants growing in

Oman showed a wide range of significantly higher proximate parameter percentages. Among these medicinal plants, *J. excelsa* had highest fiber, proteins and fat contents while *D. viscosa* has the highest moisture and carbohydrates composition as compared to other plant species. In essential mineral compositions, *J. excelsa* has either higher or moderate amount of Mg, Ca, K and Fe. Additionally, *J. excelsa* was diverse and rich in the chemical composition of its essential oils. It had significantly higher contents of monoterpenoids specially delta– 3–Carene (5.8%), DL–Limonene (49.58%), and germacrene (9.6%). The studied medicinal plants are exposed to a constant pressure of overgrazing; therefore, besides conserving them for future generations, it is also important to further investigate their phytochemical and human health impacts.

# **Conflict of interest statement**

We declare that we have no conflict of interest.

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# **Comments**

# Background

The authors discuss here very important aspects of medicinal plants like proximate, nutrients and essential oil. The aspects are usually ignored when plants are evaluated for medicinal purposes. The other important clue for which this research should be the part of scientific investigation is because of their relation with biodiversity for which the Jabal Al–Akhdar (area from where these plants are collected) is among the WWF's global 200 eco–regions.

# **Research frontiers**

The medicinal value, essential nutrients and oil have been reported for the first time from the five medicinal plants collected from the Jabal AL Akhder.

#### Related reports

Recently few studies were carried out on the said five medicinal plants. However, no more detail is given about the nutrient composition and proximate analysis of about these medicinal plants. Moreover, the essential oil composition has been reported though they are different in quantities described here for the *T. polium* and *T. excels*. Few of which is in agreement with Emami *et al.* (2011), Mitić *et al.* (2012) and Khajjak *et al.* (2012).

# Innovations & breakthroughs

The study is perhaps the most exciting one regarding the medicinal plants collected from Oman. The analysis has been performed with most sophisticate equipment and trust worthy protocol. This kind of study could be implied for the balanced food and diet apart from their medicinal value for local peoples. The analysis has identified the nutrients from these plants for the first time. This could play major role in economic development of the farmer community.

# Applications

The present research work can be applied in curing various diseases like antimicrobial, anti-inflammatory and antispasmodic. From the current work these medicinal plants can also be checked for activity against other diseases like cancer and malnutrition.

# Peer review

In light of main theme of the article, the authors have tackled one of the neglected aspects of medicinal plants. Most of the local people used these plants just to cure ailments but do not use in their routine life. Other than this the manuscript is well presented and the new findings will be benificial not only to local people but the rest of the world.

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