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Proximate and nutrient composition of medicinal plants of humid and sub-humid regions in North-west Pakistan

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The study was carried out to assess proximate composition and nutrient contents of five medicinal plant species collected from Northwest Pakistan. *Bupleurum falcatum* and *Valeriana officinalis* belongs to humid regions, while *Forsskalea tenacissima*, *Lavandula angustifolia* and *Otostegia limbata* belongs to sub-humid regions. Proximate analysis (total protein, fats, carbohydrate, ash, and moisture contents) were carried out following methods of Association of Official Analytical Chemists. Macronutrients (Ca, Mg, Na, K) and micronutrients (Fe, Cu, Pb, Zn, Ni, Cr, Co, Mn) were analyzed using atomic absorption spectrometry. Results revealed higher concentration of macronutrients in *F. tenacissima* and micronutrients in *O. limbata*. In conclusion, sub-humid region's species are having higher nutritional value than humid region's species.

Key words: Proximate analysis, micro and macro nutrients, medicinal plants, humid region, sub-humid region.

INTRODUCTION

Medicinal plants play a significant role in providing primary health care services to rural people and are used by about 80% of the marginal communities around the world (Prajapati and Prajapati, 2002; Latif et al., 2003; Shinwari et al., 2006). Each medicinal plant species has its own nutrient composition besides having pharmacologically important phytochemicals. These nutrients are essential for the physiological functions of human body. Such nutrients and biochemicals like carbohydrates, fats and proteins play an important role in satisfying human needs for energy and life processes (Hoffman et al., 1998; Mathews et al., 1999; Dingman, 2002).

Pakistan, being bestowed with ample wealth of plant

resources distributed along a wide range of climatic conditions. More than 1,000 plants species are being reported to carry medicinal values (Shinwari et al., 2006). These medicinal plants are used by marginal communities to cure various diseases (Latif et al., 2004; Adnan and Holscher, 2010). As various medicinal plant species are used either in the form of extract or decoction by the local people in different regions, therefore, evaluating their nutritional significance can help to understand the worth of these plants species in different ecological conditions. To do so, the present study was developed with the objective to evaluate the nutritional composition of selected medicinal plant species belongs to two different ecological conditions in Northwest Pakistan. Selected medicinal plants are *Bupleurum falcatum* L., *Forsskalea tenacissima* L., *Lavandula angustifolia* Mill., *Valeriana officinalis* L., *Otostegia limbata* Benth. (Table 1).

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Table 1. Selected medicinal plant species and their uses.

Botanical name	Family name	Local name	Part used	Habit	Collection region	Uses
<i>Bupleurum falcatum</i> L.	Umbelliferae	Zangli sowa	Whole plant	Perennial Herb	Swat	Medicinal, vegetable, fuel
<i>Forsskalea tenacissima</i> L.	Urticaceae	Chahar Mahak	Whole plant	Perennial herbs	Kohat	Medicinal, Ethno-veterinary, Fodder
<i>Lavandula angustifolia</i> Mill.	Lamiaceae	Ustokhudos	Whole plant	Evergreen shrub	Kohat	Medicinal, flavoring agent
<i>Valeriana officinalis</i> L.	Valerianaceae	Muskhe-Bala	Roots	Herb	Swat	Medicinal, Tea Condiment
<i>Otostegia limbata</i> Benth.	Lamiaceae	Phut kandu	Whole plant	Shrub	Kohat	Medicinal

Till now, little knowledge is available the proximate and elemental composition of these on these medicinal plant species.

B. falcatum is distributed in Europe and Asia, while in Pakistan it has been reported from district Dir and Swat valley (Ali et al., 2007). The roots extract (juice) of *B. falcatum* L. is used for the treatment of liver diseases (Manandhar, 2002). Key constituents are Bupleurumol, triterpenoid saponins-saikosides (saikosaponins), flavonoids (rutin) (Ishii et al., 1980). *O. limbata* is widely distributed in the Northwest Pakistan (Ali et al., 2007). Traditionally its grinded leaves mixed with other plant are used in gum diseases, curing of wounds and treatment of ophthalmia in man (Iqbal and Hamayun, 2002). It consists of 40 constituents including monoterpenes, sesquiterpenes, diterpenes and their derivatives (Hailemichael and Konig, 2004). *L. angustifolia* is native to the Mediterranean area (Ali et al., 2007). The flowering spikes are dried and grinded to extract essential oil, which is commonly used as antiasthmatic, antiseptic, antispasmodic, digestive and expectorant. It consists of twenty-three phenolic compounds including phenolic acids, hydroxycinnamoylquinic acid derivatives, glucosides of hydroxycinnamic acids and flavonoids (Claveria et al., 2007). *V. officinalis* is a well-known and frequently used medicinal herb especially for its effect as a tranquillizer particularly for those people suffering from nervous overstrains. Mostly the people crushed its dried underground parts and taken it orally. It contains over 150 - 200 chemical constituents. The biologically active components can be divided into three groups: the volatile oil, epoxy iridoid esters (valepotriates) and alkaloids (Yao et al., 2007). *F. tenacissima* is distributed in Afghanistan and Pakistan. The plant is used as a cure for cough, headache and kidney diseases (Ali et al., 2007; Ramzi et al., 2008).

MATERIALS AND METHODS

Collection sites of medicinal plants

The study was carried on five medicinal plant species collected

from two regions (Kohat and Swat) with different ecological conditions in Northwest Pakistan from May - August 2007 (Table 1). Kohat lies in the south between 32° 47' and 33° 53' N latitude and 70° 34' and 72° 17' E longitude at about 550 m above sea level. The region is sub-humid having very hot summer with mean annual temperature about 26°C and average annual rainfall about 546 mm (Khan et al., 2009). Swat valley lies in the north between 34° 40' to 35° N latitude and 72' to 74° 6' E longitude at an altitude of 2000 m above sea level. The region is humid having mild summer with average annual rainfall exceeds 1000 mm and mean annual temperature of about 18°C (Adnan et al., 2006).

Processing and sample preparation

Collected plant samples were cleaned, packed in the kraft paper and herbarium sheets were prepared. Plants were identified in the Botany Department, Kohat University of Science and Technology. Plant (whole/part) was crushed using grinding machine and powdered samples were treated onwards. 3.0 g of the powdered sample was processed for various parameters according to the Association of Official Analytical Chemists methods (AOAC, 1990).

Proximate analysis

The proximate analysis (carbohydrates, fats, proteins, moisture and ash) of all plant samples were determined by using AOAC methods. Carbohydrate was determined by difference method [100 - (Protein +Fats +moisture +ash)]. The nitrogen value, which is the precursor for protein of a substance, was determined by micro Kjeldahl method. The nitrogen value was converted to protein by multiplying to a factor of 6.25. The moisture and ash were determined using weight difference method while determination of crude lipid content of the samples was done using Soxhlet type of the direct solvent extraction method. The solvent used was petroleum ether (boiling range 40 - 60°C). All the proximate values were reported in percentage (AOCS, 2000; Okwu et al., 2004).

Elemental analysis

Macro and micronutrients were determined using Perkin Elmer; Analyst 700, single beam atomic absorption spectrometer and the data was obtained in parts per million (ppm), (1ppm=1mg/kg). Calibration curve was established using working standards for each element. Laboratory procedures for the preparation and determination of macro and micronutrients were used as outlined by Shah et al. (2009) for plant samples.

Table 2. Nutritional values of selected medicinal plants. SE represents Standard Error for three replications (n = 3).

Medicinal plant species	% Moisture	% Ash	% Protein	% Fat	% Carbohydrates
	Mean \pm SE	Mean \pm SE	Mean \pm SE	Mean \pm SE	Mean \pm SE
<i>B. falcatum</i>	7.42 \pm 0.02	4.68 \pm 0.003	5.53 \pm 0.01	1.89 \pm 0.02	80.48 \pm 0.02
<i>F. tenacissima</i>	2.97 \pm 0.01	24.38 \pm 0.01	8.591 \pm 0.01	2.66 \pm 0.04	61.397 \pm 0.06
<i>L. angustifolia</i>	6.80 \pm 0.08	7.49 \pm 0.017	6.13 \pm 0.02	6.52 \pm 0.01	73.06 \pm 0.06
<i>V. officinalis</i>	6.82 \pm 0.09	27.91 \pm 0.05	5.26 \pm 0.03	14.35 \pm 0.01	45.66 \pm 0.13
<i>O. limbata</i>	1.32 \pm 0.01	11.15 \pm 0.02	6.402 \pm 0.03	2.278 \pm 0.06	78.81 \pm 0.06

Statistical analysis

Proximate and elemental analysis was carried out three times for each parameter (carbohydrate, protein, Ca, Na, Fe, Cr etc) of a plant sample. Hence, we got three replications (n = 3) from which we derived the mean values and standard error (SE). Inter-element correlation was performed by using Excel Program (data analysis). Humid region's species (*B. falcatum*, *V. officinalis*) and sub-humid region's species (*F. tenacissima*, *L. angustifolia*, *O. limbata*) were compared by deriving averages of their parameters means of each region. These averages were added up and got percent values (Figure 1). Mathematically it can be described with the example of Calcium (Ca) as:

Average value of Ca (humid) + Average value of Ca (sub-humid) = Total

Average value of Ca (humid) * 100/ Total = Percentage of Ca in humid region

Average value of Ca (sub-humid) * 100/Total = Percentage of Ca in sub-humid region

RESULTS and DISCUSSION

Nutritional composition

The observed mean value for carbohydrates (80%) in *B. falcatum* was higher (Table 2) than the contents in *Dennettia tripetala* of 62% (Saputera et al., 2006). There are certain plants like *Croton tiglium* that can yield carbohydrates up to a low amount of 15.51% (Shah et al., 2009). These comparisons showed that *B. falcatum* is relatively a good source of carbohydrates. The protein content was not very high in all samples. A range of 5 - 8% of protein concentration between plant samples was observed, which is lower compared to other protein rich plants ranging between 23 - 33%. As an example *F. tenacissima* was observed with highest protein contents (8%), which is almost 3-fold less than produced by *Croton tiglium* of 26% (Shah et al., 2009).

Fats results demonstrated that *V. officinalis* with 14% stand highest percentage compared to other four plant samples (Table 2). *B. falcatum* has the highest mean moisture content 7%, which was in close agreement with 8.0% moisture in *Dennettia tripetala* reported earlier (Nishan, 2007). *O. limbata* have the lowest contents of 1% moisture. Results revealed that humid region's species contains more moisture compare to sub-humid region's species. It has been recommended by Pomeranz

and Clifton (1981) that ash content of nuts and seeds in the standard range of 1.5 - 2.5% are suitable for animal feeds. In our samples, the ash contents ranges between 5% (*B. falcatum*) to 28% (*V. officinalis*). This implies that all plant samples do not fall within the standard range for animal feed but can be used as ethno-veterinary in both regions.

On average, the increasing order of these nutrients among the plant samples of both regions is Carbohydrate > Ash > Protein > Fat > Moisture. Results indicated that on average moisture, ash and fat are produced more in humid region species, while carbohydrates and proteins in the sub-humid species (Figure 1a). Inter correlation among moisture, ash, protein, fats and carbohydrates (Table 4a) of both regions' species showed a moderate positive correlation (0.59) between fats and ash. Strong negative and inverse correlation of (-0.93) was observed between carbohydrates and ash.

Macronutrients

Results indicated that high concentrations of Calcium (Ca), Magnesium (Mg) and Potassium (K) have been found in *F. tenacissima*. Moreover, low concentrations of Ca, Mg and Na (Sodium) were observed in *B. falcatum* (Table 3).

It was reported that Ca concentration in plants of Swat (humid region) ranges from 287 - 20645 mg/kg (Begum, 2007). We also found that sample plant species of the humid region in our study lies within the same range. However, Ca concentration of 70375 mg/kg has been recorded for sub-humid region species (*F. tenacissima*) that was almost 3-fold higher than humid region species.

Reported concentration of Mg in humid region species ranges from 11 - 7119 mg/kg (Begum, 2007). All sample plants from both regions falls in the same range with the highest Mg concentration (3415 mg/kg) recorded for *F. tenacissima*. The identified Na concentration in the humid region species that is 77 mg/kg (*B. falcatum*) and 302 mg/kg (*V. officinalis*) in the current study falls into the reported range of 22 - 380 mg/kg Na concentration (Begum, 2007). However, *O. limbata* (sub-humid species) shows much increase in Na concentration (560 mg/kg). Out of all five investigated species, *F. tenacissima* showed highest K contents (13673 mg/kg) while *V.*

Table 3. Concentration of Macro and Micro Nutrients in ppm (1ppm = 1mg). SE represents Standard Error for three replications (n = 3).

Nutrients (ppm)		Medicinal plant species				
		<i>B. falcatum</i>	<i>F. tenacissima</i>	<i>L. angustifolia</i>	<i>V. officinalis</i>	<i>O. limbata</i>
		Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE	Mean ± SE
Macro Nutrients	Ca	8546 ± 0.17	70375 ± 0.14	10500 ± 0.33	13340 ± 0.57	24812 ± 0.83
	Mg	1404 ± 0.13	3415 ± 0.18	2192 ± 0.22	1830 ± 0.09	1950 ± 0.008
	K	10341 ± 0.44	13673 ± 0.91	11991 ± 0.42	6128 ± 0.06	10677 ± 0.03
	Na	77 ± 0.06	314 ± 0.11	375 ± 0.09	302 ± 0.1	560 ± 0.56
Micro Nutrients	Fe	176 ± 0.04	625 ± 0.08	480 ± 0.04	2787 ± 0.03	380 ± 0.12
	Cr	48 ± 0.07	45 ± 0.06	50 ± 0.001	58 ± 0.1	78 ± 0.05
	Mn	31 ± 0.005	45 ± 0.002	35 ± 0.007	269 ± 0.06	25 ± 0.09
	Cu	5 ± 0.003	8 ± 0.005	9 ± 0.002	10 ± 0.06	5 ± 0.004
	Pb	< 1	10 ± 0.02	16 ± 0.03	10 ± 0.04	4 ± 0.003
	Zn	18 ± 0.07	39 ± 0.04	23 ± 0.003	32 ± 0.01	20 ± 0.04
	Ni	26 ± 0.02	26 ± 0.008	29 ± 0.1	35 ± 0.002	44 ± 0.05
	Co	2 ± 0.006	3 ± 0.001	< 1	3 ± 0.002	8 ± 0.007
	Cd	0.73 ± 0.001	0.74 ± 0.002	< 0.50	< 0.50	0.77 ± 0.001

officinalis (6128 mg/kg) stands lowest.

On average, the increasing order of the macronutrients among the investigated plant samples of both regions is Ca > K > Mg > Na. It was observed that the concentration of Ca, Na, K, and Mg were found more in sub-humid region species compared to humid region species (Figure 1b). Strong inter-element positive correlation (0.93) was recorded between Ca and Mg (Table 4b). However, weak correlation was observed for Na with other macronutrients.

Micronutrients

Results showed that highest concentrations of Cr, Ni, Co and Cd out of investigated plant samples were found in *O. limbata*. Highest concentration of Fe, Mn and Cu were found in *V. officinalis*. Lowest values of Fe, Cu, Pb, Zn and Ni were recorded in *B. falcatum* (Table 3).

Iron (Fe) concentration was observed more in humid region's species than sub-humid region ones. The highest value of *V. officinalis* (2787 mg/kg) in our study falls within the reported range of Fe concentration (1706-8163 mg/kg) in humid region (Begum, 2007). On the other side sub-humid region species are having less concentration of Fe. The variable amount of Fe has been reported in the plants because the ability of plant to absorb Fe is tailored with the variable physical conditions of soil and aqueous arena.

In all sample plants, Chromium (Cr) showed a range of concentration as low as 45 mg/kg for *F. tenacissima* to as high as 78 mg/kg for *O. limbata*. This range comes under the reported range of 35 - 358 mg/kg recorded for the humid region (Shah et al., 2009). Adriano (1986) sugges-

ted that for many plant species Cr proved to be toxic at 10 mg/kg. Thus the studied plants have manifolds higher concentration of Cr as compared to that of recommended. Manganese (Mn) concentration of 269 mg/kg has been found as maximum in *V. officinalis*. This value is much higher than that already reported values (54 - 117 mg/kg) of Mn in humid regions (Begum, 2007).

Copper (Cu) concentration has been found maximum in *V. officinalis* (10 mg/kg). The concentration is less than the maximum amount of 33 mg/kg in the plant samples of humid region (Shah et al., 2009). For normal plant growth, 5 - 20 mg/kg concentration of Cu is adequate in plant cells. Lead (Pb) is present in maximum amount in *L. angustifolia* (16 mg/kg). This amount is much higher than the reported plants Pb concentration in the humid region (2 - 8 mg/kg) (Shah et al., 2009). Pb concentration of 2 - 6 µg/kg is sufficient for the normal plant growth (Kabata and Pendias, 2001). Pb concentration in *V. officinalis* and *F. tenacissima* is also higher than the normal concentration.

Zinc (Zn) was found at higher amount (39 mg/kg) in *F. tenacissima*, which falls in the normal range of 25 - 150 mg/kg and above the deficiency level of 20 mg/l (Jones 1972). However, *B. falcatum* (18 mg/kg) falls under the deficiency level. The upper toxic limit of Zn in most of the plants is spanning between 100 - 500 mg/l (Macnicol and Beckett, 1985). Nickel (Ni) concentration was found maximum in *O. limbata* (44 mg/kg), which falls in the recorded range of Ni for humid region species (20-289 mg/kg) (Shah et al., 2009). The phytotoxic Ni concentration is between 40 to 246 mg/l (Gough et al., 1979). Cobalt (Co) was found in maximum concentration in *O. limbata* (8ppm). Co concentrations in all plant samples are low and not considered as toxic, as other metals are

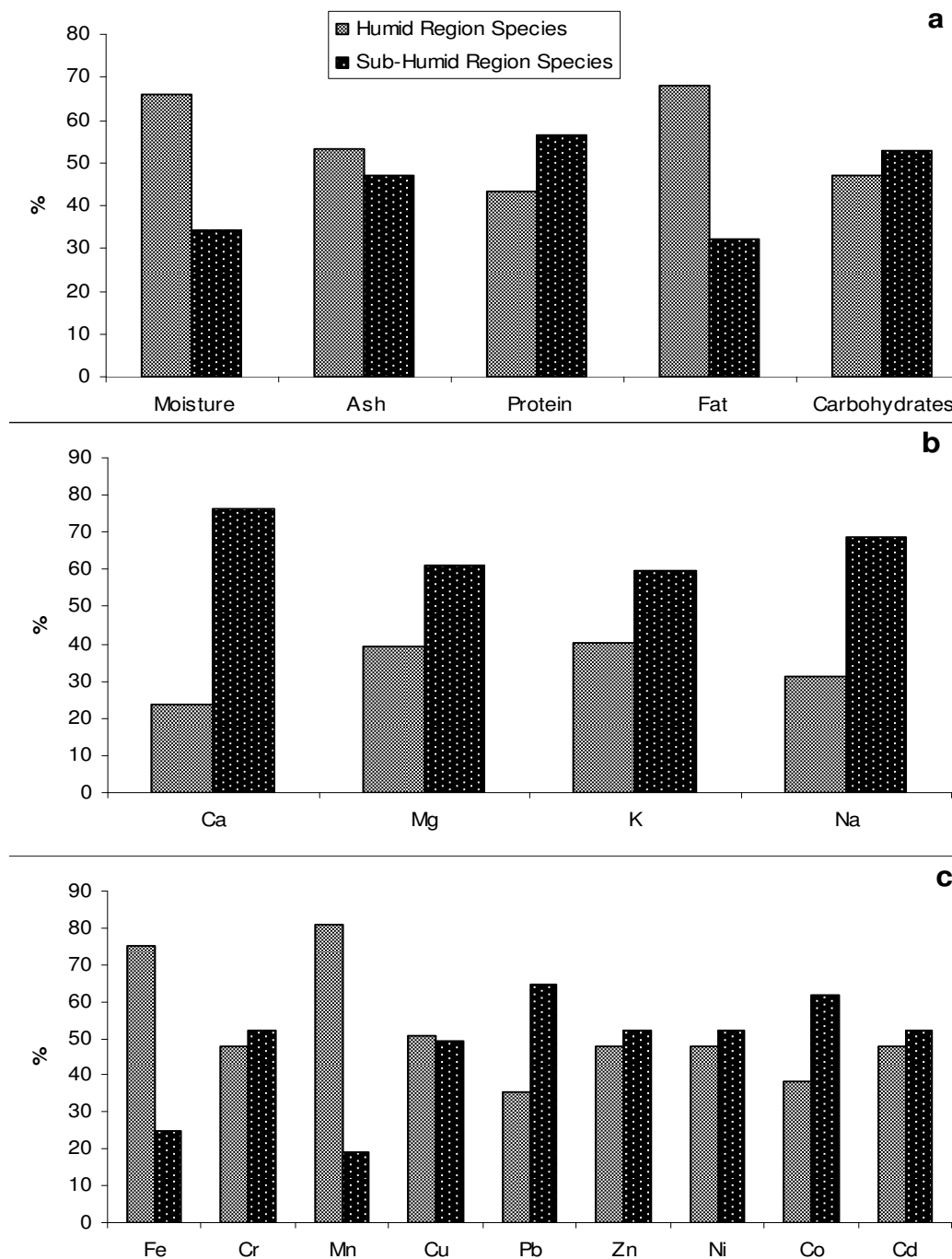


Figure 1. A comparison of nutritional (a) and elemental composition (b and c) in selected medicinal plant species of humid and sub-humid regions of Northwest Pakistan

(Broyer et al., 1972). Cadmium (Cd) concentration was very low in all sample plants; however, comparatively *O. limbata* came up with a higher value of 0.77 mg/kg. This concentration is higher than reported from humid region species (0.09 - 0.40 mg/kg) (Shah et al., 2009).

On average, the increasing order of the micronutrients

among the investigated plant samples of both regions is Fe > Mn > Cr > Ni > Zn > Pb > Cu > Co > Cd. On average Cr, Pb, Zn, Ni, Co and Cd were found more in sub-humid region species than humid region species, while Fe, Mn, Cu are vice versa (Figure 1c). Strong correlation existed between Fe and Mn, while strong

Table 4. Inter-element correlation.

4a	Moisture	Ash	Proteins	Fats					
Ash	-0.17								
Proteins	-0.63	0.31							
Fats	0.46	0.59	-0.49						
Carbohydrates	-0.17	-0.93	-0.016	-0.83					
4b	Ca	Mg	K						
Mg	0.93								
K	0.61	0.65							
Na	0.17	0.25	0.11						
4c	Fe	Cr	Mn	Cu	Pb	Zn	Ni	Co	
Cr	0.06								
Mn	0.99	0.04							
Cu	0.71	-0.36	0.67						
Pb	0.28	-0.31	0.21	0.85					
Zn	0.47	-0.38	0.41	0.64	0.45				
Ni	0.20	0.98	0.17	-0.21	-0.20	-0.28			
Co	-0.09	0.91	-0.12	-0.51	-0.47	-0.18	0.87		
Cd	-0.62	0.22	-0.61	-0.83	-0.73	-0.11	0.09	0.56	

inverse correlation was found in Cu and Cd (Table 4c).

Conclusions

In conclusion species of sub-humid region (*F. tenacissima*, *O. limbata*) contains higher nutritional value and elemental composition than humid region's species. It is also suggested that species consisting of certain elements under high toxicity level must be avoided as fodder use. However, if plants are being used for medicinal purpose, then professionals must be consulted for advices. It is thus suggested to do more studies on proximate and elemental analyses of commonly used plants by local people to test its nutritional value and toxicity level in respect of more diverse ecological conditions.

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