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FORAGE EVALUATION OF *BERBERIS BALUCHISTANICA* USED AS FODDER

Ghazala Shaheen, Mudassir Asrar Zaidi and Arsala Mansoor

Dept of Botany, University of Balochistan, Quetta, Pakistan; mudassirisrar@hotmail.com

ABSTRACT

This study was conducted to evaluate the mineral concentration, nutritional and anti- nutritional contents of the leaf of *Berbaris baluchistanica*. It is commonly used as a forage plant and is found in the high altitude areas of Quetta district Samples of forage were collected during the year 2004-2005 through all four seasons. Mineral contents, crude protein, crude fiber and phenolics were estimated quantitatively. The metabolic effects of this plant were checked by feeding it to sheep. Its epidermal morphology (both surfaces) was used to check the adulterants. It has predominantly anamocytic type of stomata with some of the anisocytic type. High phosphorus in foliage 0.58%, calcium 0.39%, Sodium 0.04%, Potassium 0.62% was found in spring season, while more Iron 0.05% was found in summer and autumn season. Maximum Phenolic content (1.7%), crude protein content (19.35%), crude fiber content (26.3%), carbohydrate content (10%) and medium gross energy 5.2 Macl /Kg DM were found in the autumn season. All these values gradually increased from spring to summer and reached their maximum in autumn. Significant increase ($P<0.05$), in the mineral and nutritional concentrations were found in this season. Various concentration of phenolics in different seasons were recorded which increased with the maturity of plants. *B. baluchistanica* was fed to 21 sheep (8-10 months old) and their fecal and urine samples were analyzed for crude protein, crude fiber and total nitrogen The feeding experiment has demonstrated the digestibility of *B. baluchistanica* and has led to significant increases ($P>0.05$) in the body weight of sheep in the experimental groups compared to the control group. The plant was found to be a good source of nutritional and mineral contents but its diet must be supplemented with other high energy diet.

Keywords: *Berberis baluchistanica*, forage mineral contents, forage digestibility, crude protein; crude fiber; phosphorus; calcium; sodium; potassium, Iron

INTRODUCTION

Berberis baluchistanica is an important plant because of its use in folk medicine. The roots decoction is used for the cure of internal injuries and ophthalmic diseases, beside this it is commonly used for the removal of kidney stones. In these areas, small ruminants are kept by local people who depend on cheap roughage which is generally low in nutritional value. A large number of these shrubs are being used as fodder by the grazing animals. Generally the animal health depends on quality and quantity of diet available for grazing the species present in the range site. The nutritional value of these shrubs can be evaluated from the concentrations of various minerals present in them and these can be used to assess their quality as a diet for the cattle (ARC, 1980**:** Nasrullah *et al*., 2004). On the other hand, the mineral composition of these plants is affected by a number of factors such as climate, topography and soil characteristics and anti-nutritional characteristics. Although it has been shown that plant phenolics contribute more in the herbivore's di*et al*ong with other biotic and abiotic factors (Feeny, 1976; Swain 1970; Niknam and Ebrahimzadeh 2002), the small ruminants avoid those species which contains high contents of phenolic compounds (Mole and waterman, 1987; Cork and Foley 1991; Niknam and Ebrahimzadeh 2002). In addition, is under a severe drought for last seven years which has resulted in the deterioration of the quantity and the nutritional value of foliage.Therefore this study was carried out to find fodder quality of *B. baluchistanica*.

In this study we undertook the seasonal elemental analysis, and determination of nutritional and anti-nutritional quality of leaves of dominant shrub B. baluchistanica. Feeding trials were also conducted to see its effect on animal health.

MATERIALS AND METHODS

Sample collection

Sample of forage were collected from three different sites in the Zarghoon region during two years (2004-2005) and four seasons. It was collected from the Wali Tangi area in Zarghoon range, located in the southern part of Quetta valley which lies approximately between latitude 30^{o} 39′ N and longitude 67^{o} 15′ E, with a mean elevation of approx 8000ft. Rain and snowfall dominates the winter season with mean minimum daily temperature at –16°C while the mean maximum temperature in summer is 25°C. In the Zarghoon region, scattered vegetations are still seen during winters and provide a source of livestock feed, although deciduous vegetation is more common. Samples were hand plucked from eight to ten plants to make a unit; three replicates were collected from the same

method. Samples were bought to the Botany department University of Baluchistan for laboratory analyses of of the samples. iSamples were dried at room temperature, ground to pass through 1mm screen thoroughly mixed and stored in sealed plastic bottles until analyzed.

Sample preparation and elemental analysis

Ash content was determined by igniting the known amount of plant material in muffle furnace to 650C° for at least 8 hours. Ash was cooled in desiccator at room temperature and weighed. Ash content of the samples was determined by the methods given in AOAC (1990). Samples were prepared for elemental analysis by digestion process. Phosphorus (P), Calcium (Ca), Sodium (Na), Potassium (K), Iron (Fe), Aluminum (Al), Manganese (Mn) were determined by atomic-absorption/flame-spectrophotometer AA-6105 (Shimdzu), X-ray fluorescence spectrophotometer, according to "A manual of experiment for plant biology methods (Kokichi Hinata and Teruyoshi Hashiba, 1995)" was used to analyze samples in triplicate. Results were calculated as percentages on dry weight basis.

ANTI-NUTRITIONAL ANALYSIS

Phenolics determination

a. Extraction

0.5 gm plant powder material was extracted with 100ml of MeOH-H_2O (80:20) at 30C^o in water bath for three hours (modified from Conde *et al*., (1995). The suspensions of water extraction were filtered and the aqueous solution was used for quantitative determination. The suspensions of methanolic extraction were also filtered and used for quantitative determination.

b. Quantitative determination

Total Phenolics content were determined by Waterman and Mole (1994); Niknam and Ebrahimzadeh (2002) method. In this procedure, appropriate volumes of aqueous solutions were diluted to a final volume of 17 ml by distilled H_2O then 1ml of Folin reagent and 2ml of saturated solution of sodium carbonate were added. After 30 min, the absorbance was measured at 760nm. Aqueous solutions of tannic acid (0.0–6.25µg/ml) were used as standards for plotting working curve according to Ranganna (1986). UV-Visible recording spectrophotometer (UV1601Shimadzu) was used for absorbance measurement.

Nutritional study

Carbohydrates in leaves were analyzed by Clegg's Anthrone (1956) method. Total crude protein contents was determined by Kjeldhal method and calculated by as follows

Protein = wt. of nitrogen X 6.25

Crude Fiber was analyzed by the method described by PARC (1982).

Gross energy

Foliage gross energy (GE) values were estimated, by igniting 1gm of dried sample using a Parr bomb calorimeter (Model 1266), Parr instrument Co., Moline. IL.

Animals and diet

Eight to ten month's old sheep with an average body weight of 45.6 ± 1.6 kg were used for experiment. Three sheep were used as control treatment. These sheep were obtained from Department of Livestock (Government of Balochistan) Quetta. Sheep were fed 500gm *B. baluchistanica* daily, in addition to their normal feed. During the experiment sheep were kept in individual pens. Control sheep were fed normal diet (mixed herbage), and no special treatment was accorded to them regarding their food content. There fecal and urine samples were also collected, analyzed and compared with experimental animals.

Feeding study

Feed was offered once daily at 09.00 hrs for 21 days, water was available throughout the day. Samples of feces and urine were collected before new feed was given each morning. From every daily output, 10% of fecal sub samples were collected and every three days these samples were mixed together to make one unit in order to get the mean. The same was practiced with urine sample where 20 ml of urine was collected in plastic container to which

5ml of HCl was added. Fecal samples were dried in oven at 70 C^o, grinded to pass 1mm sieve and stored at room temperature for chemical analysis. Carbohydrate, Crude protein, Crude fiber and total nitrogen were analyzed by the method given by PARC (1982). Initial and final body weight was measured to calculate the difference.

Statistical analysis

Various conversion factors have been used to determine different factors. Result are expressed in means and standard error of means (Means ±SEM). Mean values and standard deviation of elemental concentrations and other treatments were subjected to analysis of variance (ANOVA). Level of significance was checked at 0.05 levels.

RESULTS

Mineral concentrations

We observed substantial quantity of phosphorus; (0.58% DM) was found during summer season and it gradually decreased in autumn and winter season, when in winter a significant reduction to (.42% DM) was observed (Table1). The amount was above the deficiency level of phosphorus which is under 0.15% (Allen, 1989), so this species met the requirement of ruminants throughout the year. Phosphorus is a mobile elements as it may be reused within the plant, being translocated from the senescent tissues to the younger ones (Del Valle and Rosell, 2000). This amount is comparable to the recommendation of NRC (1996). Ca, another important elemental requirement for growing and finishing animals, was also met at (0.39% DM) making it a good source of calcium for the grazing animals

Table 1. Concentration of foliage elements of *B. baluchistanica*

Seasons	Phosphorus %		Calcium, %		Sodium. %		Potassium %		Iron %		Phenolics %	
Zarghoon Region												
Spring	0.58	±0.1	0.39	±0.2	0.04	±0.003	0.62	±0.03	0.04	±0.002	1.3	±0.7
Summer	0.56	±0.16	0.37	±0.1	0.04	±0.003	0.52	±0.02	0.05	±0.003	1.4	±0.5
Autumn	0.50	±0.2	0.34	±0.1	0.04	±0.002	0.37	±0.01	0.05	±0.001	1.0	±0.3
Winter	0.42	±0.2	0.32	±0.1	0.03	±0.002	0.37	±0.02	0.04	±0.002	1.7	±0.2
Mean	**0.51**		**0.35**		**0.03**		**0.47**		**0.04**		**1.3**	

Each value is mean ± standard deviation of twelve (12) determinations. ANOVA (P< 0.05), (P>0.05): Mn (0.001% – 0.006%), Al (0.001% – 0.003%)

Table 2. Average leaf nutritional value of *B. baluchistanica.*

Seasons	Fresh Weight (gm)	Dry Weight (gm)	Ash Weight (mg)	Carbo-hydrate %	Crude Protein %	Crude Fiber %	Energy (Mcal/ Kg-DM)
Zarghoon Region							
Spring	10	4.8	0.523	16.14	17.00	21.3	4.6
Summer	10	5.2	0.498	16.72	18.10	23.0	4.56
Autumn	10	5.3	0.542	18.87	19.35	26.3	5.2
Winter	10	5.0	0.531	17.77	16.52	25.4	4.52
Mean	**10**	**5.08**	**0.524**	**17.38**	**17.74**	**24**	**4.72**

Each value is mean ± standard deviation of 18 samples ANOVA (P < 0.05), (P > 0.05)

The Sodium concentration (0.04%) did not vary in three seasons but minor difference was found in winter season. The critical level of sodium 0.06% was suggested by the Anon, (1996), so this species can not fulfill the sodium requirement throughout the year for small ruminants. Nevertheless, seasonal variation did not affect sodium concentration and similar sodium deficiency is documented by (Khan et., al 2004). Same observation was noted in Potassium concentration, only in spring season 0.62%DM was found in *B. baluchistanica* leaves, which fulfill the recommended level (0.62%) as suggested by Anon (1996). The amount of iron was moderate in all the seasons while the amount of Aluminum (Al) and Manganese (Mn) were negligible.It has been reported that accumulation in high concentrations of any elements in any plant tissue without toxic effects may be a genetic characteristic which exhibits a tolerance mechanism in these plants. Although these elements are essential, they are also potentially toxic, so plants possess complex biochemistry to control them (Özcan and Bayçu 2005).

Table 3. Average animal fecal and urine output value of *B. baluchistanica*.

Localities	Carbohydrate %		Crude Protein %		Crude Fiber %		Total Nitrogen %		Body weight Kg	
Control	**F**	**U**	**F**	**U**	**F**	**U**	**F**	**U**	**I. wt**	**F. wt**
	1.2	1.1	5.20	0.2	15.0	1.1	1.1	0.2	45.8	51.2
Zarghoon Region	3.2	1.6	7.5	1.2	5.2	2.9	1.2	0.6	46.5	48.5

(F= Fecal, U= Urine, I.wt= initial weight, F.wt= Final weight)

Anti-nutritional Studies

Maximum amount of phenolics was observed in winter season (1.7%), medium amount was observed in spring season and less amount (1.0%) was found in autumn season. These results are contradicted to Niknam and Ebrahimzadeh (2002), who suggested the positive relationship between the intensity of solar radiation and the quantity of phenolics produced by plants. Generally the plants that grow in a sunny environment have high phenolic content as compared to the ones that grow in shades (Mole *et al*., 1988). Plants adapt to physiological needs and the phenolic contents are produced by reducing the photo destruction of exposed tissue. The Zarghoon region is hilly and the plant is found at high elevation, so a reduced production of phenolics might be due to the fact that the plant was found in shady areas.

Nutritional studies

The amount of ash (5.42mg/10 gm or 0.498%) was found from *B. baluchistanica* in the autumn season. It was light red to whitish in color which can be attributed to the presence of a red colored pigment in mid vein of its leaves.

High carbohydrate was found during the autumn season (18.87%), followed by the winter season (17.77%) as illustrated in Table 2. There was relatively low quantities of carbohydrates in the spring and summer seasons. The carbohydrate content of *B. baluchistanica* was remarkably higher than that of mixed herbage (4.2%), but much lower than in wheat grains (70%) Reuter and Robinson (1986).

These findings were comparable to the result of Norton (1981), when he reported that 10% carbohydrate is the minimum required amount for effective lactic acid formation, and lower amounts lead to poor fodder fermentation. Similar quantities (18%) were recorded by Dahot (1993) from *Capparis decidua*. Niknam and Lisar (2004) also found low quantities of carbohydrates from *Astragalus* species from Iran.

The highest amount of crude protein (19.35%) was found during the autumn season as well. Medium amount was found during the summer season and lesser amount was observed in the winter season. These were comparable to the standard table of feed composition in Japan (2001). The relatively high amounts of protein present in *B. baluchistanica* is able to fulfill the animal's requirement. Similar results was reported by Jones and Wilson (1987), where they concluded that in arid environments, plant may have adequate protein in relation to other nutrients when measured as concentrations of total nitrogen throughout the growing season.

High crude fiber (26.3%) was also found in autumn season, gradually decreasing in other seasons with the lowest value in spring (21.3%). Heneidy (2002) also reported high crude fiber (30–36%) DM from some palatable supplementary feed. Highest amount of gross energy was 5.2 (Mcal/Kg-DM) found in autumn season while lower amount (4.52 Mcal/Kg-DM) was observed in the winter season. Lower gross energy (3.6–3.5 Mcal/Kg-DM) was recorded by Heineidy (1992) from Western coastal region of halophytic range. Our results indicate high gross energy of the plants of this region and are comparable to Heineidy (2002), he found low energy level from coastal region, NW–Egypt. Yu *et al*., (2004) reported similar results from *Alfa Alfa.*

Feeding study

The minerals present in *B. baluchistanica* are affected by various factors which influence their intake and utilization. Generally, the dietary quality of leaves varies in nutrient content and digestibility throughout the year. Animal obtain minerals through the consumption of naturally available feed Gowda *et al*., (2004). The quality of plants as fodder is judged primarily by its consumption by animals and its nutritive content with respect to its phonological development.

When *B. baluchistanica* was fed to sheeps, the amount of carbohydrate found in feces was 3.2% and 1.6% in the urine (Table 3). This can be compared to the carbohydrates found in leaves at 18%. Approximately 20% of the

carbohydrate passed unabsorbed, which indicates the fact that these plants and less digestible. Carbohydrates are a significant part of nutrition in animal, as they are the grazing ruminant's primary source of energy.

Crude fiber was 5.2% in feces and in urine it was 2.95% (Table 3). This was much lower than the control sheep indicating that 21% out of 26% crude fiber was absorbed by the sheep. This aspect fares well for *B. baluchistanica* as according to Fonseca *et al*., (1998) and Rubenza *et al*., (2003), fiber proportion and type determine the extent and rate of feed degradation.

The amount of crude protein was at 7.5% in the feces and 1.2% in urine (Table 3). A high amount of nitrogen 1.2% in feces and 0.6% in urine was also found. Generally, the microbial population of these ruminants require about 6-8% crude protein in diet. It appears that the protein and nitrogen content of *B. baluchistanica* is sufficient for the ruminants and its microbe's requirements. Finally, relatively less body weight (2kg) increase was observed when fed *B. baluchistanica.*

Shrub species are especially important in this region having long, cold winters. As during this cold period there is little growth of herbs or grasses, the animals are forced to rely on shrubs species during this harsh season. Shrub consumption in winter varies widely depending upon the location of the site and the kind of grazing animals. While ***B. baluchistanica*** leaves are nutritious the presence of spines and thorns reduce its palatability.

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