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Nutrient Characteristics of Foliage and the Availability of Water in a Rangeland near Quetta, Balochistan, Pakistan.

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Abstract: Nutrient deficiencies (primarily N, P or K) are major limitations to animal production in upland rangelands of Balochistan. Nonetheless, there is little reliable information on mineral concentrations in forage species. We assessed the mineral concentrations in different species of shrubs and perennial and annual grasses from Hazarganji Chiltan National Park along with their $\delta^{13}C$ as a time-integrated estimate of water use efficiency (WUE). The concentration of total N (15 - 30 mg g^{-1} in foliage) would meet the needs of ruminants in most of the species studied, but was substantially reduced (to below ruminant requirements) by significant concentrations (2-10 mg g^{-1}) of non-protein nitrogen. At the time of sampling (Spring), concentrations of P (1-4 mg g^{-1}) were adequate for grazing animals whereas those of K and Mg were not (mostly < 6 mg g^{-1}). Results suggested that in summer, concentrations of the main nutrients would decline further as the plants mature. Concentrations of non-protein nitrogen were related to WUE ($\delta^{13}C$) and reinforce the view that shortages of water reduce the palatability and nutritive value of most forage species in these Balochistan rangelands.

Key words: Nutrient deficiencies, Water use efficiency, Non-protein nitrogen

Introduction

Rangelands remain critical to the economy of many countries and still provide about 70% of the feed needs of domestic ruminants and 95% of that of wild ruminants (Holechek *et al.*, 1995). The daily nutrient demand (especially the N- or protein demand) of stock fluctuates in accordance with the physiological functions of the grazing animals and hence patterns of maintenance, gestation, growth, fattening and lactation (Cook and Harris, 1977) play major roles in determining daily nutrient requirements. In contrast, the chemical and nutrient composition of plants and plant communities in rangelands varies according to climate, species, soil type, phenology and other abiotic factors (Greene *et al.*, 1987). For much of the year, most range species are low in protein and available energy (Adams and Short, 1988). In arid environments, plants may seemingly have adequate protein (in relation to other nutrients) when measured as concentrations of total N, throughout the growing season (Jones and Wilson, 1987). However, such estimates are usually overestimates of the quality of available forages due to the absolute abundance of phenolics, condensed tannins (and other anti-nutritional factors) and to the relative abundance of non-protein nitrogen that is usually included in estimates of total N and thus protein. In arid environments, plants must cope with almost continual drought (Morgan, 1984) and a number of arid zone species have evolved the capacity to accumulate considerable concentrations of cytoplasmically compatible (e.g. non-polar) amino acids and other non-protein nitrogenous solutes. These help to protect the cytoplasm from desiccation when salts (e.g. K^+) accumulate in the vacuole.

Other plant adaptations to such environments include strongly developed root systems, highly sclerophyllous leaf morphologies, capacity to regulate leaf and xylem water potential to name but a few (Larcher, 1995). One of the better-studied adaptations, particularly in herbaceous species, is the capacity to regulate stomatal aperture. Knowledge of this process has been greatly improved by developments in analysis and understanding of carbon isotope discrimination when CO_2 enters the leaf (e.g. Farquhar *et al.*, 1988: Lin *et al.*, 1996). $\delta^{13}C$ has been increasingly accepted as a robust index of water use efficiency (WUE) in plants since Farquhar *et al.* (1984, 1988) elegantly described the theoretical bases for discrimination between the carbon isotopes during physical (e.g. movement of water through stomata) and biological (e.g. carbon fixation) processes. Empirical data in support of the theory is comprehensive and measurements of $\delta^{13}C$ in leaf material have become routine tests of WUE, which in turn generally increases as water availability diminishes.

Balochistan is one of the four provinces of Pakistan but is by far the largest and 93 % of its area is rangeland. The climate in much of the province is arid or even hyper-arid (van Gils and Baig, 1992) and rainfall patterns vary from Mediterranean to monsoonal. Generally, plant species in Balochistan are deficient in total digestible nutrients and in digestible protein and dry matter with respect to animal requirements (FAO, 1983). Little of the rangeland resources of Balochistan have been assessed and there is little to no information on how abiotic factors such as soil type and topography influence the nutritive value of native species. A single study of the dietary composition of the Zarchi and Tomagh rangelands suggested that most of the native species were deficient in nitrogen and phosphorus (Wahid, 1990). As the first step in an attempt to quantify and then improve range quality near Quetta, Balochistan, the aims of this study were: a) to describe the chemical composition of native shrubs and grasses (relative to nutritional needs of sheep, goats and other species of wildlife such as Ibex) and b) to relate that chemical composition to WUE (as measured by the carbon isotope composition of leaves and stems of shrubs and grasses) along a short topographic gradient. We used Hazarganji Chiltan National Park as a model for Balochistan rangelands.

Materials and Methods

This study was conducted during spring (May) 1997 at Hazarganji Chiltan National Park near Quetta (30° 07′ N, 66° 58′E, 1700 m altitude), Pakistan. The region has a Mediterranean climate, characterised by cold winter and hot dry summer. Annual rainfall varies between 250 and 300 mm and is dominated by winter snowfall. The mean maximum temperature in summer is 36 °C and the mean minimum

Table 1: Concentration of nitrogen, phosphorus and non-protein nitrogen (NPN) (mg g^{-1} oven dry weight) and potassium, magnesium and calci (% oven dry weight) in the foliage of herbaceous and perennial shrubs found along an elevation gradient in Hazarganji Chiltan Natio Park, Quetta, Pakistan.

Species	N	NPN	P	K	Mg	Ca	$\delta^{13}C$
Top							
Caragana vlicina	31.83	7.97	1.68	0.46	0.35	2.70	-27.16
Caragana ambigva	26.07	8.76	1.52	0.55	0.20	0.89	-25.28
Ephedra intermedia	11.21	4.01	0.85	0.34	0.41	2.48	-24.89
Salvia cabulica	22.75	9.64	1.62	0.54	0.42	4.41	-26.28
Sophora griffithi	24.50	8.77	1.46	0.40	0.20	1.87	-23.03
Mean	23.27	7.83	1.43	0.46	0.32	2.47	-25.34
Standard Error	3.38	0.99	0.15	0.04	0.05	0.58	0.70
Middle							
Artemisia scoparia	26.59	3.14	2.52	0.38	0.22	0.60	-28.26
Daphne oleoidee	14.88	2.23	1.62	0.49	0.24	0.89	-24.86
Ephedra nebrodensis	13.49	1.84	0.85	0.41	0.34	1.95	-24.69
Ferula ovina	24.84	7.24	2.20	0.57	0.15	2.63	-27.75
Fraxinus xanthoxyloides	23.62	8.85	2.00	0.46	0.35	0.50	-24.50
Perowskia atriplicifolia	26.77	6.35	2.26	0.35	0.21	1.59	-27.53
Prunus eburnea	17.68	6.98	1.39	0.59	0.39	1.22	-27.03
Sophora griffithi	29.56	3.28	1.49	0.36	0.36	0.75	-23.46
Mean	22.18	4.99	1.79	0.45	0.28	1.27	-26.01
Standard Error	2.13	0.94	0.19	0.03	0.03	0.26	0.82
Bottom							
Achillea santolina	19.60	3.73	3.70	0.37	0.19	1.11	-28.90
Artemisia scoparia	33.06	4.12	2.65	0.32	0.33	1.04	-27.73
Convolvulus spinosus	17.33	2.93	1.55	0.34	0.22	1.42	-24.99
Ebenus stellatus	20.83	2.87	1.16	0.75	0.20	0.90	-27.09
Nepeta glomerulosa	24.32	3.45	2.45	0.65	0.56	0.79	-26.94
Perowskia atriplicifolia	21.35	6.34	1.74	0.44	0.28	0.53	-27.38
Sophora griffithi	27.99	3.40	1.33	0.57	0.30	1.55	-24.49
Mean	23.50	3.33	2.08	0.49	0.30	1.05	-26.79
Standard Error	2.05	0.45	0.34	0.06	0.05	0.13	0.69

Table 2: Concentration of nitrogen, phosphorus (mg g^{1} oven dry weight) and potassium, magnesium and calcium (% oven dry weight) in the ster of herbaceous and perennial shrubs found along an elevation gradient in Hazarganji Chiltan National Park, Quetta, Pakistan.

Species	N	P	K	Mg	Ca	$\delta^{13}C$
Top						
Caragana ambigva	13.66	0.75	0.22	0.10	1.38	-25.03
Caragana vlicina	10.52	0.72	0.33	0.11	1.00	-26.47
Salvia cabulica	6.85	0.65	0.26	0.19	2.03	-25.76
Sophora griffithi	11.74	1.46	0.24	0.20	1.87	-22.46
Mean	10.69	0.90	0.26	0.15	1.57	-24.93
Standard Error	1.42	0.19	0.02	0.03	0.23	0.78
Middle						
Artemisia scoporia	14.36	1.71	0.24	0.13	1.23	-27.61
Daphne oleoidee	7.72	1.22	0.22	0.09	1.25	-23.40
Ferula ovina	13.14	1.68	0.35	0.26	1.56	-27.38
Fraxinus xanthoxyloides	16.81	1.39	0.24	0.12	1.29	-23.54
Perowskia atriplicifolia	13.66	1.62	0.28	0.17	1.54	-26.67
Prunus eburnea	7.02	0.98	0.32	0.18	1.95	-26.36
Sophora griffithi	19.25	0.94	0.23	0.37	1.41	-23.41
Mean	13.14	1.36	0.27	0.19	1.46	-25.13
Standard Error	1.69	0.12	0.02	0.04	0.10	0.84
Bottom						
Achillea santolina	8.42	1.17	0.39	0.10	1.79	-28.58
Artemisia scoparia	10.34	1.26	0.34	0.13	1.82	-26.07
Convolvulus spinosus	5.97	0.65	0.28	0.10	0.96	-24.82
Ebenus stellatus	9.64	0.65	0.23	0.14	1.85	-27.09
Nepeta glomerulsoa	8.59	1.30	0.46	0.56	0.79	-25.56
Perowskia atriplicifolia	10.86	1.36	0.35	0.20	1.80	-26.57
Sophora griffithi	19.60	1.01	0.32	0.24	1.94	-24.26
Mean	10.49	1.06	0.34	0.21	1.56	-26.14
Standard Error	1.63	0.11	0.03	0.06	0.19	0.65

temperature in winter is -1 °C (Kidd *et al.*, 1988). During the hot dry summers, plant growth is poor and only winter precipitation is effective in supporting plant growth. Hazarganji National Park lies on the foot slopes of the Chiltan mountain range.

The dominant plant species in the park include the perenni shrubs, *Sophra griffithi, Ferula ovina* and *Artemisia scopori* and the C_4 grasses *Cymbopogan* and *Chrysopogan* spp. A annual grasses are C_3 species. Numerous other species wer vegetative at the time of sampling (Tables 1 and 3). Many c

Table 3: Concentration of nitrogen, non-protein nitrogen (NPN) and phosphorus (mg g^{-1} oven dry weight), potassium, magnesium and calcium (% oven dry weight) and $\delta^{13}C$ in grasses along an elevation gradient in Hazarganji Chiltan National Park, Quetta, Pakistan

Species and location	N	NPN	P	K	Mg	Ca	$\delta^{13}C$
Top							
Bromus spp	10.86	1.02	1.65	0.28	0.81	2.48	-26.64
Cymbopogan jwarancusa	17.85	2.86	2.45	0.31	0.16	0.60	-13.01
Mean	14.36	1.94	2.05	0.30	0.49	1.54	-19.82
Standard Error	3.52	0.92	0.40	0.01	0.33	0.94	4.31
Middle							
Bromus spp	12.79	1.24	2.00	0.27	0.75	1.32	-27.75
Poa bulbosa	14.01	1.90	1.49	0.22	0.17	3.26	-25.78
Chrysopogon auncheris	19.25	2.27	2.20	0.36	0.13	1.28	-13.15
Cymbopogan jwarancusa	17.68	2.65	2.13	0.24	0.12	0.68	-12.22
Mean	15.92	2.02	1.96	0.27	0.29	1.64	-19.72
Standard Error	1.52	0.30	0.16	0.03	0.15	0.58	3.66
Bottom							
Chrysopogan aucheri	18.38	2.50	2.07	0.36	0.21	2.00	-12.60
Cymbopogan jwarancusa	15.76	1.52	1.81	0.25	0.14	2.15	-13.52
Mean	17.07	2.01	1.94	0.31	0.18	2.08	-13.06
Standard Error	1.31	1.49	0.13	0.06	0.04	0.07	0.29

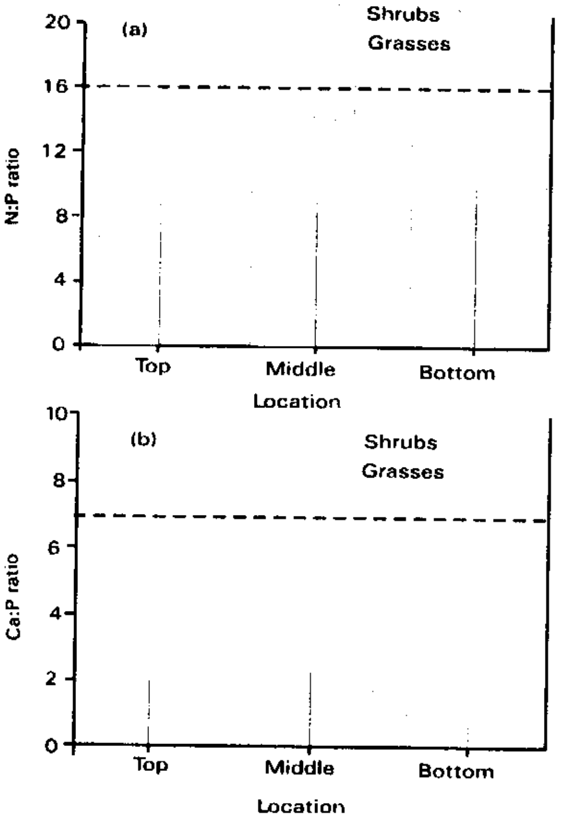

Fig. 1: Nutrient status of major life forms along an elevation gradient shown by (a) N: P and (b) Ca:P ratio of live shoots. The dashed line indicates a reference value for 'normal' nutrient ratios.

the species collected for this study have not previously been studied for their nutritive value or chemical composition. Ten to fifteen replicates of leaves and stems were taken for each species of native shrub and the same number of replicates was collected for the grasses. We sampled the flora at three points along a topographic gradient which spanned ~300 m in altitude. Plant samples were oven-dried for 2 days at 70 °C, and then ground to pass through a 1 mm sieve. Total N, P and cations were determined by acid digestion of 100 mg of plant sample at 320 °C with H_2SO_4 and H_2O_2. Diluted digest were analysed colorometrically for N and P by the procedure described by Keeney and Nelson (1982) and Murphy and Riley (1962) respectively. Cations were analysed using Atomic Absorption Spectroscopy. Plant samples were analysed for non-protein nitrogen (NPN), using the tungstic acid extraction method described by Lictira *et al.*, (1996).

Dried material was further ground for $\delta^{13}C$ analysis in a ball mill to ensure thorough homogenisation and combustion of the tissues. A 2-mg sub sample of the tissue was analysed on Tracer Mass Stable Isotope Analyser (Europa Scientific, UK) as described by Macfarlane and Adams (1998) and $\delta^{13}C$ values are expressed in parts per thousand (‰) and were calculated with respect to a Pee Dee Belemite standard $\{= [(^{13}C/^{12}C_{sample})/(^{13C}/^{12}C_{standard})-1] \times 1000\}$.

Concentrations of nutrients and chemical elements and the isotopic composition of tissues of perennial shrubs were compared between locations along the topographic gradient using paired *t* tests based on the data for 5 species at each location (top, middle, bottom). For grasses, the available data for all species were combined and tested as above.

Results

Communities at each point of the elevational gradient varied in species composition. At the lowest point, the grasses *Chrysopogon aucheri* and *Cymbopogon jwarancusa* were common amongst the dominant shrubs *Artemisia scoparia* and *Sophora griffithi*. As elevation increased, these grasses were replaced by *Bromus spp* at a lower density, and the shrubs were replaced by *Ferula ovina*, *Perowskia atriplicifolia*, *Prunus eburnea* and *Fraxinus xanthoxyloides*, again at a lower density. At the highest point of the gradient, plant density was least and much less than at lower points. *Caragana vlicin*, *Caragana ambigva* and *Ephedra intermedia* dominated with a few scattered grasses.

Analysis of foliar nutrients was characterised by high variability among species. For example, at the top of the elevational gradient, nitrogen concentrations varied more than 3-fold and calcium concentrations more than 4-fold (Table 1). There was less variation in concentrations in stems. There was no significance difference (t-test, N=5, P = 0.05) along the topographic gradient in the mean concentration of nitrogen in the shrubs and grasses. Nonetheless, concentrations of nitrogen were generally greater in foliage of shrubs than in the grasses (22 ± 2 *v.* 16 ± 2 mg g^{-1}). In shrub foliage, the concentration of non-protein nitrogen was significantly (P = 0.05) greater at the highest elevation compared with lower elevations while in grasses it was greater in the middle of the gradient. In shrubs, non-protein nitrogen contributed 30-50 % of total nitrogen at the top of the gradient and declined with elevation to 12-40 % of total N in the middle and 12-20 % at the bottom. In grasses the NPN remained within the range 20-27 % of total N at the top of the elevational gradient and between 12-18 % of the total nitrogen in the middle and bottom.

In both shrub and grasses, there was no significant difference in the mean concentration of P along the gradient. On a life-form basis, the mean N: P ratio in shrubs was between 12 and 16, while in grasses it was between 6 and 8 (Fig 1a). Concentrations of potassium were greater in shrubs than in grasses or in the shrub stems. Magnesium and calcium concentrations varied little among species or among life forms. Calcium concentrations were significantly greater in shrubs at higher elevations while in grasses they were greater at the bottom of the gradient (Tables 1, 3). At the time of sample collection, Ca: P ratios were below 2. (Fig 1b).

Carbon isotope discrimination was compared among shrubs and C_3 and C_4 grasses along the topographic gradient. Leaf carbon was generally depleted in the heavier isotope ^{13}C compared with stem carbon and both shrub components were depleted in ^{13}C relative to the C_3 grass species (Table 1- 3).

Discussion

Water availability exerts an important role on the structure (species composition, phenotypic composition) and function of many rangeland plants and plant communities. In Hazarganji Chiltan National Park, woody perennial shrubs dominated throughout the elevation gradient while grasses were generally scattered. Nearly all life forms in these communities are strongly sclerophyllous; in common with those in arid rangelands elsewhere, including those in Australia, America and Africa.

The δ^{13}C data from Hazarganji strongly suggest that more water was available at the bottom of the elevation gradient (less negative δ^{13}C). The δ^{13}C data match the measured increase in non-protein nitrogen in shrubs and grasses with elevation. Singh *et al.*, (1973) and Cyr *et al.*, (1990) reported that plants accumulate non-protein nitrogen, mostly non-essential amino acids, in response to drought. We might also expect that the concentration of non-protein nitrogen would increase while that of total nitrogen will decline as plants mature and as water availability continues to decline later in the year. A widely used measure of forage quality is the estimated concentration of 'protein' derived by multiplying the concentration of total N x 6.25. That measure will increasingly misrepresent the actual protein available to herbivores as conditions become drier and as plants mature due to the increasing abundance of non-protein and often anti-nutritional nitrogen compounds (e.g. Main, 1981). Similarly, *Ephedra* spp. accumulates large amounts of alkaloids and there were once substantial industries in Balochistan based on the extraction of these for medicinal purposes.

The low N:P ratio of both shrubs and grasses is suggestive of an N limitation (Koerselman and Mueleman, 1996) and is a common constraint of productivity in rangelands throughout the world. Nonetheless, at the time of sampling, all species, with the exception of the few annual grasses and *Ephedra nebrodensis*, had concentrations of nitrogen in foliage tha were greater than the recommended minimum for ruminant (National Research Council, 1975).

Concentrations of phosphorus and calcium were marginal fc grazing ruminants (National Research Council, 1975) except i a few species and Ca:P ratios were between 1:1 and 2: which is ideal for growth and bone formation (Underwooc 1981). All shrub and grass species had lower concentration of potassium than recommended (National Research Counci 1975) by the National Research Council (0.5-0.8 % Magnesium concentrations were also low in all species.

As noted by Minson (1990), the concentrations of nearly a nutrients in foliage of all potential forage plants vary wit season, sometimes by several-fold. Our preliminary studies o plant nutrition in Hazarganji Chiltan National Park has provide a single example of the effects of water availability on form of N in forage plants (albeit confounded by changes in specie composition along the gradient) and an overview of th variation in nutrient concentrations among species. Futur studies aimed at improving the quality and sustainability o these rangelands will need to focus on management, whic might improve the nutritional quality of the forage for stoc whilst maintaining the diversity and productivity of the plan communities. These studies should include assessment of th effects of grazing and water shortages on secondary plan metabolites that have anti-nutritional properties.

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