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Seasonal and Genotypic Variations Influence the Biomass and Nutritional Ingredients of *Cenchrus ciliaris* Grass Forage

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ABSTRACT

High contents of protein and digestible nutrients are required in the diet of ruminants. Therefore, during, 2004-2007 a field experiment was undertaken to investigate the most promising strains of Buffel grass (*Cenchrus ciliaris*) under rain-fed conditions. Five strains of Buffel grass, included in this study (RMF-267, RMF-268, RMF-269, RMF-270 & RMF-271) were grown for four years and their data on biomass and nutrient parameters were recorded every year during two seasons viz. spring and summer. During spring season, the highest fresh biomass (24.4 t ha^{-1}) was in RMF-269 strain followed by RMF-270 and RMF-268. The lowest fresh biomass (18.1 t ha^{-1}) during this season was that of RMF-267 strain. Dry matter yield (6.93 t ha^{-1}) was at the highest in RMF-269 among other strains. Similar trend was also in monsoon season, however forage yield was higher in monsoon than in spring season due to prolonged growing period and more rainfall. RMF-268 gained higher crude protein (9.24%) and total digestible nutrients (56.5%) than in other grass strains. Post-harvest soil characteristics had non-significant difference with the original soil. It is concluded that the nutritional parameters, as well as biomass yield of Buffel grass had genotypic variation and also differ seasonally.

Key Words: Buffel; Biomass production; Forage quality; Rain-fed area

INTRODUCTION

One of the major constraints in rangeland management is lack of authentic scientific inventory of range resources (Afzal *et al*., 2007). Pakistan has a wealth of 135 million heads of livestock, which accounts for 10.8% of the GDP (GOP, 2007). *Cenchrus ciliaris* commonly named as Buffel grass (Dhaman) belongs to the family poaceae being annual/perennial growing to a height of 15-20 cm and sometimes produces rhizomes. The rhizomatous drought resistant plants are dormant during summer months and with the on set of rains, they regenerate quickly and produce heavy canopy to cover soil (Bose & Balakarishnan, 2001). It is very good pasture grass for hot and dry regions in the tropics and sub tropics, cultivated for permanent pastures in Africa and Australia. This highly nutritious grass is valued for its production of palatable forage and intermittent grazing during droughty period. Yield of some strains makes it good for forage during the wet season. A new cultivar Blue Buffel grass yields 30% more than other species. The grass is fed green and can also be turned into silage. The silage made with hay is said to increase flow of milk and impact a sleek and glossy appearance. It is also folk remedy for kidney pain tumors, sores and wounds (Siegfried, 1990).

A high percentage of protein is required in the diet of ruminants because production of milk, meat and reproduction mainly depends on protein ingredient of the animals' diet. Amount of crude protein lower than 6-7% in the animal diet may depress microbial activity due to non-availability of nitrogen in the summer. Another factor for evaluation of feeding value is digestibility, which is mainly concerned with the availability of total digestible nutrients (TDN). With respect to the contents of crude protein and TDN, Buffel grass varied at three locations in Pakistan, mainly due to difference in nutrients availability at these sites (Afzal & Ullah, 2007). However, the effect of different growing seasons and climatic conditions on the production and nutritional composition of *C. ciliaris* grass strains is not known. Therefore, this four year study was undertaken on five strains of the *C. ciliaris* grass to see their biomass production and proximate composition dynamics pattern.

MATERIALS AND METHODS

The experiment was carried out at the research area of National Agricultural Research Centre (NARC) Islamabad, Pakistan. Climate of the planting site is sub-humid, sub-

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tropical continental type with an annual rainfall of about 1000 mm, occurring in bi-model pattern mostly in late summer and winter spring periods. About 60 to 70% of total rainfall is generally received during monsoon (mid June to mid September). Monsoon rains are usually in heavy downpours and are accompanied by the thunderstorms. The remainder rainfall is received in winter mostly during December-March. June and July are the hottest months having mean maximum temperature ranging from 36 to 42°C with extremes sometimes as high as 48°C. December and January are the coldest months with mean temperature of about 3 to 3.5°C. Occasionally, the lowest minimum temperature may drop to -3.3°C (Nizami *et al*., 2007). Frost occurs from mid December to February during the days of favorable conditions i.e., clear sky, calm wind and temperature close to below the freezing point. Agro-metrological data of the study period is given in Table I, which was collected from the Water Resources Station, NARC, Islamabad. The soils are slightly alkaline with pH 8.30 and are non-saline, loamy in texture, low in organic matter and deficient in major plant nutrients except for available P (Table VI). The grasses were tested for their adaptability for three years at Rangeland Research Experimental Area, NARC, Islamabad, Pakistan. The tufts plantation of the grass was done in July, 2003 with three replications in a factorial randomized complete block design, row to row and plant to plant distance was 50 cm and plot size was 3 m × 4 m. Grass plots were maintained without irrigation and fertilizer application. Weeding and hoeing was done manually, whenever needed. Samples were collected during the months of September for quality analysis. Plant samples were collected at 50% flowering stage due to the best stage for feeding the animals.

Data on fresh and dry matter yield were collected from the same plots during, 2004 to 2007. Data were collected at 50% flowering stage. Three quadrates (1 m^2) were harvested randomly for fresh and dry matter determination (Khan, 1966). Fresh biomass was calculated with the following formula:

$$\text{Fresh biomass (t ha}^{-1}) = [\text{Fresh biomass/area (m}^2)] \times 10$$

All the plants in one square meter (1 m^2) area were clipped close to ground level and fresh biomass was weighed and samples were oven dried to a constant temperature at 70°C for 72 h. Dry samples were weighed and their dry biomass was determined. Moisture contents percentage was calculated with the following formula:

$$\text{Moisture contents (\%)} = (\text{Fresh weight- dry weight/fresh weight}) \times 100$$

Forage quality analysis: For proximate composition the samples were again oven dried at 70°C for 12 h. Grass samples were manually harvested at panicle stage. Contents of crude protein (CP) and total digestible nutrients (TDN) were calculated on the basis of chemical analyses. Samples of grass were chopped in an electric chopper, dried at 55°C and ground to 2 mm size through Willey mill. These samples were analyzed for CP and TDN by AOAC methods (Khan, 1966). Total digestible nutrients (TDN) of each grass sample were calculated by using the equation of Wardeh (1981).

Nitrogen-free extract: Nitrogen-free extract contents were determined by difference after the analysis of all the other items in proximate analysis on dry matter percent basis by using the equation of Harris (1970) as follows:

$$\text{NFE (\%)} = 100 - (\text{\% crude protein} + \text{\% crude fiber} + \text{\% ether extract} + \text{\% ash}$$

Total digestible nutrients: Total digestible nutrients (TDN) were calculated by the equation described by Wardeh (1981) as below:

$$\text{TDN (\%)} = -26.685 + 1.334\,(CF) + 6.598\,(EE) + 1.423\,(NFE) + 0.967\,(Pr) - 0.002\,(CF)^2 - 0.670\,(EE)^2 - 0.024\,(CF)(NFE) - 0.055\,(EE)(NFE) - 0.146\,(EE)(Pr) + 0.039\,(EE)^2\,(Pr)$$

Soil analysis: Soil samples were collected from 0-15 cm before starting the experiment and after harvesting grass from all the treatment plots. These samples were air dried, ground and passed through 2 mm sieve. Analysis was carried out in the laboratories of Land Resources Research Institute, National Agricultural Research Center, Islamabad. Analytical methods of US Salinity Laboratory Staff (1954) were followed or otherwise mentioned. All the calculations were made on oven dried soil weight basis. Saturated soil paste was prepared according to Method 2. Saturated soil extract was obtained by vacuum pump (Method 3a).

Soil textural analysis was done by Bouyoucos hydrometer technique (Bouyoucos, 1962). Dispersion was made with 1% sodium hexametaphosphate solution and soil texture was determined by using international textural triangle. Soil pH of the saturated paste was determined by pH meter having combination electrode after calibrating with buffer solutions of pH 7.0 and 9.0 (Method 21a). After calibrating the instrument with 0.01 N KCl, the EC_e was measured with LF-191 Conduktometer (Method 4b). Organic carbon was determined by titrating the sample containing soil, potassium dichromate and sulphuric acid using ferroin indicator (Method 24). Organic matter was determined by the following formula:

$$\text{Organic matter (\%)} = \text{Organic matter (\%)}] \times 1.72$$

Nitrogen was estimated through sulphuric acid digestion. Distillation was undertaken with micro-Kjeldhal method (AOAC, 1994). Available phosphorus contents in the soil were determined by using $NaHCO_3$ solution as extracting agent. Soil sample of 2.5 g was weighed and 50 mL Olson's reagent (0.5 M $NaHCO_3$, pH = 8.5) was added and this suspension was shaken for 30 min and filtered. Standard stock P solution was prepared by dissolving exactly 0.439 g potassium dihydrogen orthophosphate (KH_2PO_4) in half liter distilled water. Then 25 mL 7 N H_2SO_4 were added and volume was made one liter to get 100 ppm P standard stock solution. Five mL of the filtrate was used to develop color and then reading was recorded using Spectrophotometer. Soil was saturated with normal NH_4 OAC (pH 7.0). Extraction was made with same solution and extractable (available) K was determined by Jenway flame photometer (Method 18).

Agro-meteorological data: Meteorological data on rainfall, humidity, pan evaporation, wind speed, sunshine hours and temperature during the study period were obtained from Water Resources Research Institute (WRRI), National Agricultural Research Centre (NARC), Islamabad (WRRI, 2007) to elaborate and understand the experimental results under the light of climatic changes, if any (Table I).

Statistical analysis: Data of various grass species on their biomass and composition were subjected to analysis of variance (ANOVA) and data means were compared using least significant difference test (Steel *et al*., 1997).

RESULTS AND DISCUSSION

Biomass production: Various biotic and abiotic factors influence the crop growth and production (Jaleel *et al*., 2009). Similarly, a variety of biochemical and physiological mechanisms are involved in the growth and development of plants (Razmjoo *et al*., 2008). Plant growth can be measured through characters like height, tillering, branching and bearing of new leaves etc, which are ultimately translated into biomass. In this study, the highest fresh biomass (24.4 t ha^{-1}) was produced by RMF-269 followed by RMF-270 (22.5 t ha^{-1}) and RMF-268 (21.2 t ha^{-1}) during spring season (Table II). The RMF-271 gained the bottom figure (17.4 t ha^{-1}) in this regard. Dry matter yield was also higher in RMF-269. During monsoon season, similar trend was indicated (Table II). Genetic variation plays a key role in plant biomass production giving edge to some varieties over others due to their better tolerance to various stress factors (Jia *et al*., 2008; Shamsi *et al*., 2008; Rasheed *et al*., 2010). Fresh biomass and dry matter yield were more in monsoon than in spring season due to prolonged growth period and more rainfall. Higher biomass production during summer season could be attributed to increased temperature, water availability and sun-shine hours, which all favored the plant growth. Kaleem *et al*. (2010) reported better capture and utilization of temperature, light, photoperiod and soil moisture during spring than in autumn for crop growth and yield attributes.

In addition to higher yield, digestibility is one of the major qualities of forage. Apart from other factors, it also depends upon the succulence of forage. Succulence mainly depends on the moisture contents in plant tissues. Old leaves of plant tissues generally contain water from 75 to 85% of the fresh weight. Succulence of forage depends on the value of moisture contents (%). A significant difference among Buffel grass varieties was observed for their moisture contents (Table II). Higher biomass producing varieties RMF-269 and RMF-270 also had greater moisture contents. More moisture indicated more succulence and digestibility. Moisture contents in forage were higher in monsoon season than in spring, so digestibility was greater in the monsoon as compared to spring season (Ullah *et al.,* 2006). This was due to higher water availability from monsoon rains and enhanced growth of Buffel grass during summer months as compared to that in spring which was relatively drier season. Shaheen *et al.* (2010) reported an increase in the crop yields and water-use efficiency through integrated soil-water conservation and improved fertilization practices.

Forage quality parameters: Higher production of fodder is appreciable only if its quality is acceptable as well. Production of milk, meat and associated products of livestock depends upon hereditary factors by approximately 25% while 75% is dependent on the feed quality and quantity. Feeding value of forage quality is dependent on crude protein percentage and total digestible nutrients. Crude protein is very useful in the diet of rumens to keep up their milk, meat and maintaining livestock property (Afzal & Ullah, 2007). The RMF-268 had the highest crude protein (9.24%) among other strains of Buffel grass (Table III). Amount of crude protein lower than 6%-7% in animal diet may depress microbial activity due to non-availability of nitrogen in the rumen (Bose & Balakarishnan, 2001).

Forage digestibility is related to chemical composition, particularly of fiber, lignin and silica contents and to some extent of more crude protein. Crude fibre mainly consists of cellulose, hemicellulose and lignin. The lignin content generally reduces the digestibility of forage. Significant differences were depicted in crude fibre among Buffel grass species (Table III). The ash contents showed non-significant effect among the grass species (Table III). Ether extract is composed of fats, oils, waxes, organic acids, pigments, sterols and vitamins A, D, E and K. Among these, vitamins are of chief concern in animal feeding. These compounds are the components of enzymatic systems that catalyze metabolic reactions. A significant difference among grass species was observed for their chemical composition (Table III). Analysis of nitrogen free extract (NFE) indicates nitrogen free compounds like sugar, fructosans, starch, pectin, organic acids, resins, tannins, pigments and water-soluble vitamins. There was significant difference of RMF-267 with other strains by attaining the highest percentage of NFE contents (Table III). Genotypic differences in the nutritional parameters among the cultivars of other crop plants e.g. rice have been reported (Ullah *et al*., 2006).

Higher dry matter intake is related to better voluntary intake and thereby for higher nutrient intake. The TDN is the physiological equivalent of digestible energy and also is a feed-feces difference. It is an important factor in the digestibility/feeding value. The TDN value was at the highest (56.5%) in RMF-268 cultivar (Table III). So depending on the availability of crude protein and TDN, Buffel grass strain RMF-268 is the best to grow followed by RMF-270 (Afzal & Ullah, 2007).

Soil characteristics: Soil analysis indicates the potential of supplying nutrients and is required for any scientific study so that results coming out of the treatments can be well understood and explained on the basis of processes undergoing in the soil. Electrical conductivity is a parameter that indicates indirectly the total concentration of soluble

Table I: Agro-metrological data

Parameters	Rainfall (mm)				Wind speed (km day^{-1})			
Year/Month	**2004**	**2005**	**2006**	**2007**	**2004**	**2005**	**2006**	**2007**
January	87	59	54	0	33	40	23	27
February	35	184	23	94	27	60	29	37
March	0	75	52	179	37	35	35	52
April	93	14	21	3	72	64	48	51
May	7	20	41	58	93	98	44	80
June	132	73	62	141	91	108	61	84
July	190	183	493	335	85	75	95	57
August	258	270	312	456	71	61	87	47
September	24	73	13	133.13	63	52	69	42.81
October	83	68	35	0	68	45	49	33.97
November	23	4	15	13.35	45	46	36	17.31
December	30	9	124	0	41	36	36	21.01
	Pan evaporation (mm day^{-1})				**Sunshine hours**			
January	1.0	1.1	1.1	1.6	5.0	5.2	5.4	7.5
February	2.2	1.5	1.9	1.6	8.2	4.7	5.6	5.8
March	3.8	2.4	3.3	3.2	8.6	7.2	7.0	8.3
April	5.2	4.2	3.4	5.2	6.4	8.0	8.2	9.0
May	7.7	7.2	4.2	7.1	10.2	10.1	9.99	10.1
June	8.0	9.0	5.1	8.0	9.7	9.8	8.7	9.3
July	6.9	4.6	8.1	5.3	9.0	7.1	10.6	8.4
August	4.9	5.2	8.0	4.1	8.4	8.6	8.8	8.1
September	4.8	4.3	6.1	3.61	8.4	8.0	7.3	9.4
October	3.3	3.4	2.9	3.50	7.0	9.0	8.5	8.8
November	2.3	2.4	2.4	1.78	6.5	6.7	8.3	7.1
December	1.2	1.6	1.4	1.34	4.5	7.6	5.4	7.8

Parameters	Monthly average temperature (^{o}C)								Relative humidity (%)							
Year	**2004**		**2005**		**2006**		**2007**		**2004**		**2005**		**2006**		**2007**	
Month	**Max.**	**Min.**	**Max.**	**Min.**	**Max.**	**Min.**	**Max.**	**Min.**	**Max.**	**Min.**	**Max.**	**Min.**	**Max.**	**Min.**	**Max.**	**Min.**
January	16	5	15	3	97	63	96	64	96	63	92	46	96	63	92	46
February	21	5	16	5	91	49	95	67	94	51	95	67	94	51	95	67
March	29	10	22	10	94	39	95	64	87	45	92	58	87	45	92	58
April	32	16	30	12	68	36	73	38	84	51	71	37	84	51	71	37
May	36	18	33	17	51	28	54	32	90	74	58	33	90	74	58	33
June	35	22	39	20	63	45	62	28	90	69	65	43	90	69	65	43
July	36	23	33	23	74	51	86	70	57	33	88	65	57	33	88	65
August	33	22	33	22	68	64	89	68	51	28	91	70	51	28	91	70
September	34	20	33	21	77	47	87	57	58	26	90	62	58	26	90	62
October	26	13	30	14	87	46	83	39	90	52	81	29	90	52	81	29
November	25	8	24	7	91	37	84	33	89	55	89	43	89	55	89	43
December	18	5	21	1	97	58	88	33	92	63	90	49	92	63	90	49

Source: Agricultural meteorological field station at Water Resources Research Institute, NARC, Islamabad (WRRI, 2007). Altitude 518 m, Latitude 33′ 42″, Longitude 73′ 08″

salts in a soil. The ECe of a soil is increased as the soluble salts concentration increases. Electrical conductivity of the experimental soil was 0.53 dS m^{-1} (Table IV), which was within the normal limits (≤ 4 dS m^{-1}). Soil pH is the single characteristic, which elucidates an overall picture of the medium for plant growth including nutrient supply trend, fate of added nutrients, salinity/sodicity status, soil aeration, soil mineralogy and ultimate weather condition of the region. The pH value of experimental soil was 8.4 that was also within normal limit (> 8.5 to be declared as a sodic soil) and did not cause neither any detrimental effect on plant performance. Post-harvest values of soil pH were also normal and the minor observed differences were not statistically significant. According to Shaheen *et al.* (2008), Pothwar soils have slightly alkaline pH, but there is no salinity problem. The *C. ciliarls* grass rendered no effect on the soil texture (Table IV).

Nitrogen is essential for plant growth as it is a constituent of all proteins and nucleic acids and hence of all types of protoplasm. The original status of total N was very low because soil analysis indicated its numerical value just as 0.042% (Table IV). The decreasing trend in this value was due to utilization of soil nitrogen for forage production. Phosphorus plays a fundamental role in numerous enzymatic reactions that depend on phosphorylation. The pre-sowing phosphorus status of original soil (5.40 ppm) was very low that decreased to 5.38 ppm due to utilization of phosphorus by plants (Table IV). A little decrease was recorded in this respect. Potassium is important in the synthesis of amino acids and proteins from ammonium ions. The available K status of original soil was in deficient range. Post-harvest K values were almost equal to original soil value (Table IV). Organic matter in the soil comes from the remains of plants and animals. Land application of composted poultry litter results in higher crop yields as compared to un-composted one (Chaudhry *et al*., 2009). Original status of OM was very poor (0.51%). No significant variation was observed due to equal addition and

Table II: Biomass (t ha^{-1}) and moisture contents (%) of C*enchrus ciliaris* grass strains (average of 4 years, 2004-2007)

Buffel grass strains	Spring			Summer		
	Fresh biomass	Dry biomass	Moisture contents	Fresh biomass	Dry biomass	Moisture contents
RMF-267	18.1 ab *	6.38 ^{NS}	65 b	29.6 a	9.54 a	68 b
RMF-268	21.2 a	6.83	68 ab	27.9 a	8.53 a	69 b
RMF-269	24.4 a	6.93	72 a	29.9 a	10.88 a	73 a
RMF-270	22.5 a	6.07	73 a	24.6 b	7.53 b	74 a
RMF-271	17.4 b	5.16	70 a	24.1 b	7.50 b	71 ab

* Values in a column followed by same letter (s) are statistically similar at $p \leq 0.05$; ^{NS} Non-significant difference at $p \leq 0.05$

Table III: Proximate composition of C*enchrus ciliaris* strains as percent (%) of dry biomass (average of 4 years, 2004-2007)

Buffel grass strains	Dry matter	Crude protein	Crude fiber	Ash content	Ether extract	Nitrogen free extract	Total digestible nutrients
RMF-267	32.2 a	4.67 b	37.2 b	7.38 ^{NS}	3.03 b	47.7 a	55.2 a
RMF-268	35.4 a	9.24 a	36.8 b	7.08	5.56 a	41.4 b	54.5 a
RMF-269	26.2 b	4.54 b	43.8 a	8.49	3.19 b	40.0 bc	51.5 b
RMF-270	31.9 a	6.15 a	33.2 c	7.65	5.92 a	47.1 a	53.8 a
RMF-271	31.8 a	5.18 ab	38.0 b	7.89	3.98 ab	45.0 a	53.3 ab

* Values in a column followed by same letter (s) are statistically similar at $p \leq 0.05$; ^{NS} Non-significant difference at $p \leq 0.05$

Table IV: Physical and chemical characteristics of experimental soil

Soil characteristics	Pre-sowing (2004)	Post-harvest (2007)
ECe (dS m^{-1})	0.53	0.50
pH	8.30	7.40
Total N (%)	0.042	0.080
Phosphorus (mg kg^{-1})	5.40	5.38
Extractable K (mg kg^{-1})	78.5	75.7
Organic matter (%)	0.51	0.50
Sand (%)	34.2	35.1
Silt (%)	28.6	28.2
Clay (%)	37.2	36.1
Textural class	Loam	Loam

utilization of the soil organic matter by all the grass species (Table IV). Generally, the soils in Pothwar region are deficient in OM, N, P, Zn and B contents (Shaheen *et al*., 2008).

CONCLUSION

On biomass and dry matter yield basis the RMF-269 strain gave the best performance, but for forage quality RMF-268 was found the best among all *C. ciliaris* strains. So, RMF-269 should be grown with forage legume mixture, while RMF-268 could be grown on pure basis without forage legume as it contains more crude protein and total digestible nutrients. Among *C. ciliaris* strains, the RMF-268 can preferably be grown on marginal lands for fulfilling the requirements of balanced diet of the livestock.

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