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Nutritional characteristics and *in vitro* digestibility by near-infrared spectroscopy of local and hybrid napiergrass varieties grown in rain-fed and irrigated conditions

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Abstract. Napiergrass can produce large amounts of biomass and its nutritive value has a significant effect on its effectiveness for animal production. However, temperature and drought stress limit its productivity. Drought-tolerant pearl millet × napiergrass hybrid (PMN) varieties were developed and produce high biomass yields. The nutritional content and digestibility of PMN is not well known, which limits its use in animal feeding. It was hypothesised that PMN hybrids are more drought tolerant and have higher nutritive value than napiergrass varieties. Four napiergrass varieties (Bana grass, Mott, MB4, and N51) and four PMN (PMN2, PMN3, 5344, 4604) were tested with or without irrigation treatment in a strip-plot design, with the objective of evaluating the nutritional value and *in vitro* digestibility of PMN hybrids and napiergrass. The forages were harvested on Day 110 of planting. Samples were hand chopped, oven-dried, ground to pass through a 1-mm screen and analysed for their nutrient content and *in vitro* digestibility using near-infrared spectroscopy. Dry matter (DM) content of PMN2 (24.3%) and PMN3 (22.9%) was significantly higher ($P < 0.05$) than 5344, Bana grass and N51 napiergrass varieties. No differences ($P > 0.05$) in acid detergent fibre, neutral detergent fibre, crude protein and metabolisable energy were found among napiergrass varieties. With no effect ($P > 0.05$) of irrigation, lignin content was highest ($P < 0.08$) in 4604 (8.2%) and lowest in 5344 (5.2%). Starch was highest ($P < 0.05$) in irrigated MB4 than both irrigated and non-irrigated 4604. Non-fibre carbohydrate content was highest ($P < 0.05$) in PMN2 (12.8%) than MB4 (8.7%). The *in vitro* true digestibility was significantly higher ($P < 0.05$) in 5344 and Bana grass (70.0% and 68.0% of DM, respectively), than PMN3 (54.5%). Rate of digestion was significantly higher ($P < 0.05$) in 5344 (4.9%/h) than PMN2 (2.7%/h), others were in between. Neutral detergent fibre digestibility (NDFD) of 5344 and Bana grass (56.7% and 53.2% of neutral detergent fibre, respectively) was significantly higher ($P < 0.05$) than PMN2 (38.0%). Although no effect of irrigation was observed, there was an interaction ($P < 0.05$) between variety and irrigation on neutral detergent fibre digestibility of napiergrass varieties. In conclusion, among four PMN varieties tested, PMN3 and 5344 has higher nutritional value and *in vitro* digestibility than PMN2 and 4604 even when grown in non-irrigated condition. Thus, PMN3 and 5344 is the preferred napiergrass variety for animal feeding, even in rain-fed farming conditions.

Additional keywords: hybrid napiergrasses, *in vitro* digestibility, nutrient content.

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Introduction

Forages are a necessary component of diets for dairy cows as they provide the fibre needed to optimise rumen function. Napiergrass is a forage that produces greater dry matter (DM) yield than other tropical grasses (Boonman 1997) and is replicated vegetatively as it has very low seed set and viability. It is a plant which has a very low self-fertilisation rate, but is an out-crossing plant species i.e. unrelated genetic materials can be introduced into its breeding line, which increases genetic diversity. Pearl millet is a major warm-season cereal grown in arid and semiarid regions of the world and is a highly cross-pollinated crop with an out-crossing rate of more than 85%. These two species cross readily producing sterile interspecies hybrids, which are vigorous (Burton 1944) and have high biomass potential (Hanna *et al.* 2004).

One of the major concerns for the cattle industry is the changing climate. The Intergovernmental Panel on Climate Change (IPCC 2013) concluded that elevated greenhouse gas concentrations are likely to lead to general drying of the subtropics by the end of this century, creating widespread stress on agriculture. Every one-degree celsius increase in seasonal temperature leads to 2.5–16.0% of direct yield loss of the major grains (IPCC 2013). Battisti and Naylor (2009) mentioned that by the end of this century there is a 90% possibility that the average summer temperature will exceed the hottest temperature on record throughout the world. Higher temperatures will also lower soil organic matter content by oxidation and further result in reduction of crop yield and land quality. These climatic changes can have serious effects for meeting food, forage, water and energy needs and maintaining the islands' fragile ecosystems. Hence, it is necessary to make major adaptational changes to develop crop varieties that are tolerant to heat and heat-induced water stress. Pearl millet is the highest drought-tolerant variety. Hence, pearl millet × napiergrass (PMN) hybrids were developed, which have high seed set and viable seeds. Additionally, with over 21 000 pearl millet accessions worldwide, there is high potential to ingress desirable traits into napiergrass. PMN hybrids can play a major role in producing high quality less water requiring forage for the cattle industry in tropical regions of the world.

Hanna *et al*. (2004) mentioned that various factors like species and growing conditions can affect the nutrient composition of forages. Also, the nutritive value of the forage has significant effects on the utilisation in animals, which in turn affect the production of animals as well as methane emission, a major greenhouse gas (Mirzaei-Aghsaghali and Maheri-Sis 2011). The nutritional content and digestibility of PMN is unknown, thus warrants its determination before making any recommendation for animal feeding. It was hypothesised that PMN hybrids will be more drought-tolerant and nutritious than napiergrass varieties for cattle. The objectives of the study were to determine the nutritional value and *in vitro* digestibility of selected napiergrass and PMN varieties grown in rain-fed or irrigated conditions.

Materials and methods

Four napiergrass varieties (Bana grass, Mott, MB4, and N51) and four PMN (PMN2, PMN3, 5344, 4604) were field tested with and without irrigation treatment in a strip-plot design, with each treatment consisting of three replicated plots at the Waimanalo Research Station of the University of Hawaii, USA. The research station is located at an elevation of 18.2 m a.s.l. where annual rainfall averages 139.7 cm. Stem cutting (~18 inches) were planted on 13 March 2013 and the trial continued for 110 days. All plots (3 m × 2.4 m) were drip irrigated at 100% pan evaporation for the first 30 days. Grasses were planted in four rows (0.6 m between each row). During the trial period, plots received 26.8 cm of rainfall and 5000 gallons of irrigation were applied (0.184 acre-inches) in irrigated treatment plots. The harvested samples were chopped and dried in a force draft oven at 55°C overnight. A representative dried sample was taken from each treatment collected separately and ground to pass through 1-mm screen using a Thomas Wiley laboratory mill. Subsamples were scanned by near-infrared reflectance (NIR) spectroscopy, an approved method of AOAC (Association of Official Analytical Chemists, Gaithersburg, MD, USA), at a certified commercial laboratory (Dairy One Cooperative Inc., Forage Laboratory, Ithaca, NY, USA; http://dairyone.com/wp-content/uploads/2014/01/23_199903.pdf and http://dairyone.com/wp-content/uploads/2014/01/22_199609.pdf, accessed 23 February 2014) for proximate composition, fibre, energy content and *in vitro* digestibility. The NIR analysis was conducted using Foss NIR Systems Model 6500 equipped with software Win ISI ii version 1.5. The laboratory uses broad-based calibrations for NIR by incorporating samples collected over several decades based on reference dataset generated using wet chemistry methods. Specific nutrients and *in vitro* digestibility parameters estimated using NIR are presented in Table 1. For predicting total digestible nutrient (TDN) and net energy, the laboratory uses the summative energy equation of NRC (1988).

Statistical analyses

The nutritional profile and *in vitro* digestibility characteristics were compared using the MIXED procedure of SAS version 9.2 (SAS Institute Inc., Cary, NC, USA) with a strip-plot

Table 1. Nutrient profile and *in vitro* digestibility parameters of forages studied

All data, except dry matter is expressed on a dry matter basis. Abbreviations: ADF – acid detergent fibre, IVTD – *in vitro* total digestibility, ME – metabolisable energy, NDF – neutral detergent fibre, NDFD – neutral detergent fibre digestibility, NDICP – neutral detergent insoluble crude protein, NFC – non-fibre carbohydrate, TDN – total digestible nutrient, Trt – treatment, Var. – variety

Variables	PMN2	PMN3	5344	4604	Local (Bana)	Mott	MB4	N51	s.e.m.	*P*-value		
										Var.	Trt	Var. × Trt
Dry matter (%)	24.2	22.9	18.5	21.6	18	20.6	20.4	17.9	0.88	<0.001	0.441	0.929
Ash (%)	8.9	11.4	14.6	11.2	14.1	12.7	13.2	11.3	0.90	0.002	0.706	0.718
Crude protein (%)	6.4	7.0	7.9	7.9	8.3	6.5	7.4	7.5	0.71	0.444	0.974	0.789
Digestible protein (%)	55.5	60.2	59.8	59.3	57.0	56.7	62.5	55.7	2.97	0.666	0.906	0.491
NDICP (%)	2.3	2.5	3.0	2.8	2.8	2.2	2.8	2.9	0.26	0.381	0.205	0.462
Starch (%)	1.6	1.4	1.7	0.7	1.7	1.3	1.8	1.2	0.24	0.079	0.157	0.011
NFC (%)	12.8a	10.6ab	10.2ab	10.8ab	10.9ab	9.6ab	8.8b	9.6ab	0.73	0.025	0.477	0.015
Soluble carbohydrate (%)	4.2	3.4	4.4	4.1	5.8	3	3.6	4.6	0.50	0.017	<0.001	0.072
ADF (%)	51.2	52.3	47.4	52.3	47.8	52.9	51.6	49.7	1.53	0.133	0.321	0.630
NDF (%)	73.3	72.5	68.9	71.7	68.4	72.2	72.2	73.0	1.41	0.143	0.631	0.344
Lignin (%)	6.6	6.5	5.2	8.2	5.5	6.2	6.3	6.6	0.64	0.084	0.288	0.817
TDN (%)	46.5	46.7	51.2	46.2	50.3	44.7	49.2	51.2	1.75	0.073	0.484	0.780
ME (Mcal/kg)	1.7	1.7	1.7	1.6	1.7	1.7	1.6	1.7	0.05	0.438	0.091	0.751
IVTD (30 h, % of DM)	54.5	59.2	70.0	59.7	68.0	59.7	63.7	63.5	2.71	0.007	0.983	0.650
NDFD (30 h, % of NDF)	38.0	44.2	56.7	43.7	53.2	44.2	50.0	49.8	2.97	0.002	0.813	0.643
Rate of digestion (%/h)	2.7	3.3	4.9	3.7	4.4	3.3	4.0	4.1	0.35	0.002	0.305	0.797

arrangement of napier varieties and treatments (with or without irrigation). Means were separated using the Tukey method. Differences were considered significant if $P < 0.05$.

Results and discussion

In general, DM content of PMN hybrids was higher than that of napiergrass varieties. Dry matter content of PMN2 (24.3%) and PMN3 (22.9%) was significantly higher ($P < 0.05$) than 5344 (18.5), local (18.1) and N51 (17.9%) napiergrass varieties. Dry matter content of non-irrigated PMN2 (24.9%) was significantly higher ($P < 0.05$) than irrigated local (16.9%) napiergrass varieties (data not presented). It was expected that the PMN hybrids would outperform napiergrass under non-irrigated conditions due to the high drought tolerance capacity of pearl millet varieties. It is possible that the 26.8 cm of rainfall during the 110-day trial period did not result in sufficient plant water stress to result in significant yield differences. Future trials will monitor plant water stress to address this issue.

Ash was lowest ($P < 0.05$) in PMN2 (8.9%) and highest in 5344 (14.6), others were in between, i.e. 5344 had the highest amount of ash. No significant difference ($P > 0.05$) in acid detergent fibre, neutral detergent fibre (NDF), crude protein and metabolisable energy were found among napiergrass varieties and PMN hybrids.

With no effect ($P > 0.05$) of irrigation, lignin content was highest ($P < 0.05$) in 4604 (8.2%) and lowest in 5344 (5.2). Though there are still unresolved mechanisms, lignin inhibits digestion of plant cell wall components (Morrison 1983). The higher the lignin the lower the digestibility of forage as lignifications within plant species is negatively associated with NDF digestibility. The *in vitro* true DM digestibility was significantly higher ($P < 0.05$) in 5344 and Bana grass (70.0% and 68.0%, respectively), than PMN2 (54.5%). The lowest lignin content in 5344 might explain the highest *in vitro* true DM digestibility in this variety. However, Dehority and Johnson (1961) and Van Soest (1994) mentioned that lignin content itself may not be the only factor influencing the digestibility, as legumes tend to have twice the lignin content than grasses at the same digestibility.

Starch was highest ($P < 0.05$) in irrigated MB4 (2.7%) than both irrigated and non-irrigated 4604 (0.76% and 0.70%, respectively). Water-soluble carbohydrate content was highest ($P < 0.05$) in Bana grass (5.9%) and lowest in Mott (3.0) and PMN3 (3.5). Non-fibre carbohydrate content was highest ($P < 0.05$) in PMN2 (12.8%) than MB4 (8.7). These non-fibre carbohydrates are digested more rapidly than fibres, yielding weaker acids as by-products. Volatile fatty acids, primarily propionate are produced from the fermentation of non-fibre carbohydrate, which are absorbed from the rumen and are also used as a source of energy for the cow. Non-fibre carbohydrate is also used by microbes to make microbial protein.

Rate of digestion was significantly higher ($P < 0.05$) in 5344 (4.9%/h) than PMN2 (2.7), others were in between. Since the rate of digestion of PMN2 is slow, it limits intake due to rumen fill, whereas, fast rate of digestion in 5344, will increase intake and the production level will also increase. The NDF values of all the grasses ranged between 68–73%, with lowest NDF in bana grass (68.4). Neutral detergent fibre digestibility (NDFD) of 5344 and Bana grass (56.7% and 53.2% of NDF, respectively), was significantly higher ($P < 0.05$) than PMN2 (38.0). Although no effect of irrigation, there was an interaction ($P < 0.05$) between variety and irrigation on NDFD of napiergrass varieties. NDFD is the measure of digestion coefficient of NDF; NDFD content of forage can have a large impact on the energy value of the diet. NDFD has to be taken into account because, as the NDF digestion coefficient changes, the dietary energy contribution from NDF also changes; and as NDFD content increases, the TDN content of forage also increases. The increase in TDN results in an increase in dietary energy content and potentially milk yield. If an indigestible portion like NDF is digested in larger amounts in varieties like 5344, animals are likely to get more nutrients; hence better performance should be expected. In general lactating dairy cows will consume more forage that is of higher energy content when forages are high in NDFD. Here 56.7% of NDF present in 5344 (a PMN variety) is digested, hence energy contribution by fibre digestion is highest in 5344, which is ~50% while it is only 38% in the case of PMN2. It can be one of the limiting factors in the use of the PMN2 variety in cattle feeding.

However, the low crude protein, high fibre and low energy density of these grasses at this age may not sustain growing beef animals and may not meet needs of brood cows in later stages of pregnancy and early lactation, or growing and lactating dairy animals. A lactating animal in its early lactation has lower DM consumption in comparison to the mid-lactation period. However, daily net energy requirement in both periods is alike (NRC 1988). This low DM consumption in the early period limits the inclusion of higher amount of grasses in their diet as energy demand is to be met with more concentrate dietary sources. In mid-lactation, there is more freedom to include additional level of grasses and still have leverage on formulating identical ration.

Conclusions

Among the four PMN varieties tested, PMN3 and 5344 had higher nutritional value and *in vitro* digestibility than PMN2 and 4604 even when grown in non-irrigated conditions. Lignin content was lowest in 5344 and highest in 4604. Lignin inhibits digestibility of other plant cell wall components in forage. The presence of a high amount of undigestible lignin and a low amount of starch can be a limiting factor in the use of 4604 while formulating cattle feed. Low *in vitro* true DM digestibility, lowest NDFD and slowest rate of digestion is the limiting factor in the use of PMN2. The nutritional characteristics of PMN2 and 4604 are similar to the other napiergrass varieties tested. Thus, for these growing conditions PMN3 and 5344 would be the two preferred napiergrass varieties for forage production for ruminant feeding systems, even in rain-fed farming conditions.

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