



Biomass Yield, Morphological Characteristics, Nutritional Values and *In vitro* Digestibility of Different Napier (*Pennisetum purpureum*) Cultivars

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Abstract | The study assessed eleven different Napier (*Pennisetum purpureum* Schumach) cultivars' on the basis of their morphological characteristics, biomass yield, nutrient composition and *in vitro* digestibility. These varieties are: BLRI Napier (BN)-1(Bajra), BN-2 (Arosha), BN-3 (hybrid), BN-4 (Vietnam), Pakchong, Color Napier, MarkEron, Wruk-Wona, Dwarf-Early, Dwarf-Late, and Zara. They were sourced from the BLRI fodder germplasm center, Savar Dhaka. These cultivars were planted in three blocks of eleven rows each in February 2020, with a 50 cm x 50 cm spacing between cuttings and rows. Seven harvests per year were made during the 50-day harvest interval (HI). In contrast to the summer and winter seasons, the rainy season had the best morphological characteristics of Napier grass, regardless of variety ($p < 0.001$). However, fresh biomass, DM, and CP production of Napier grass per hectare of land was significantly ($p < 0.001$) higher in summer (87.6, 11.9 and 1.22 t/ha/yr, respectively) followed by rainy (75.7, 10.4 and 1.19 t/ha/yr, respectively) and winter season (50.1, 7.25 and 1.10 t/ha/yr). Irrespective of season, Pakchong, Wruk-Wona, Zara, BN-4, Dwarf-Early, BN-1, BN-2, Colour Napier and Dwarf-Late (137-163 cm) exhibited similar plant heights, with BN-3 (133 cm) being the lowest ($p < 0.01$). In comparison to other cultivars, BN-3, BN-4, Color Napier, and MarkEron had the highest leaf-to-stem ratio ($p < 0.001$) (1.06-0.90), while Pakchong and Zara had the lowest ($p < 0.01$) ratio (0.52). Pakchong yielded more fresh and dry matter biomass, crude protein, and metabolisable energy (ME) (274 t/ha/yr vs 36.7 t/ha/yr vs 4.44 t/ha/yr vs 278 GJ/ha/yr) than Wruk-Wona, Zara, and BN-4. In Color Napier and Dwarf-Early, fresh and DM biomass, CP and ME production were the lowest (172, 25.1, 3.02 t/ha/yr and 188 GJ/ha/yr vs 175, 25.2, 2.90 t/ha/yr and 184 GJ/ha/yr, respectively; $p < 0.001$). Likewise, when compared to other cultivars, Pakchong had the lowest oxalate level ($p < 0.05$) and higher digestible organic matter (dOM; $p > 0.05$) and metabolisable energy (ME; $p < 0.05$) value. Pakchong could be suggested to the farmers based on its biomass yield and nutritional quality, whereas Zara and Wruk-Wona are also promising types of Napier cultivars.

Keywords | Napier cultivars, Biomass yield, Nutrient composition, *In vitro* digestibility, Season

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optimal nutrient content, and digestibility is exceedingly challenging. Consequently, the purpose of this investigation was to assess the annual biomass production, morphological characteristics, nutrient composition and *in vitro* digestibility of various Napier cultivars.

MATERIALS AND METHODS

LOCATION AND DURATION OF STUDY

The research was carried out during February 2020 to February 2021 for almost a year, which includes the cultivation of fodder at Fodder Research Plot, BLRI, Savar, Dhaka-1341, Bangladesh.

TOPOGRAPHY, SOIL AND CLIMATES

The study place was 4 meters above the sea level and situated at latitude 24°42'0" N and longitude 90°22'30" E. Soil of the study area had a clay-like texture. Before the experiment started, soils weighing roughly 1 kg were taken from three distinct locations and sampled down to a depth of 20 cm. After five days of drying at room temperature, the samples were thoroughly mixed to make a composite. Five composite samples (each of about 100 g) were analysed for their nutrient composition at Soil Resource Development Institute, Dhaka-1215 to determine basal fertilizer doses for Napier cultivation. The chemical composition of the soil is shown in Table 1. The climatic data of daily temperature, humidity, and rainfall were recorded (Figure 1) at study farm to study any impacts of climatic change on year-round biomass production. In general, three types of cropping system prevail in Bangladesh (Banglapedia, 2021). The seasonal variations on biomass yield, as affected by daily temperature and humidity, at Summer (16 March to 30 June), rainy (01 July to 15 October;) and winter season (16 October to 15 March) were studied during experimental period.

Table 1: Chemical composition of soil (dry soil).

Statistics	pH	OM (%) (%, DM)	N (% DM)	K (mg / 100 g)	P (ppm / g)	S (ppm / g)	B (ppm / g)	Zn (ppm / g)
Mean	5.4	1.70	0.09	0.16	5.64	1.21	0.38	3.98
SD	0.25	0.22	0.01	0.04	2.56	0.28	0.07	0.19

Abbreviations: SD: standard deviation; OM: organic matter; N: nitrogen; K: potassium; P: phosphorus; S: sulphur; B: boron; Zn: Zinc; ppm: parts per million.

DESCRIPTION OF NAPIER CULTIVARS

BLRI Napier-1 is a perennial, deep-rooted, and appetising hybrid of elephant grass (*Pennisetum purpureum*) and bajra (*Pennisetum glaucum*) (Sarker *et al.*, 2021). Also known as the Bajra Napier grass. The BLRI Napier-3 was produced via accession selection of the Napier hybrid, which is the product of interspecific crosses between common elephant

Commercialized livestock production systems are quickly becoming the norm in many tropical and subtropical countries (Kabirizi *et al.*, 2015). This is due to the fact that the income of the population in these countries is rising in tandem with the demand for animal-derived protein (Kwon *et al.*, 2020). The production of meat and milk in Bangladesh increased significantly from 2.40 to 10.7 million tons (a yearly increase of 45%) and from 1.3 to 7.7 million tons (a yearly increase of 61%), respectively, between 2010 and 2020. During the same period, the population of cattle rose from 23.1 to 24.4 million, representing a yearly growth of 10% (DLS, 2021). Deficiency of quality feedstuff and its annual fluctuations may impact on animal production and productivity (Bashar *et al.*, 2024a). Available feedstuff in Bangladesh is about 56×10⁶ ton/yr dry matter (DM), which is 24% less than the annual demand (Huque and Sarker, 2014). Such available feedstuff may produce a typical diet of ruminant animals with 7.74 MJ/Kg DM and 2.32 percent digestible crude protein (Huque and Sarker, 2014), which is not sufficient to achieve optimum animal productivity (Sarwar *et al.*, 2002). Furthermore, animals provided with low-quality roughages typically necessitate a substantial quantity of concentrate derived from grains to sustain milk and meat production. The reliance on expensive monogastric-compatible feed has led to an increase in the costs of milk and meat production, which are comparable to or greater than the worldwide market prices (Roy *et al.*, 2017). A farm-level study reported that rice straw in the diet represented 46-67%, including 23-33% green grass and 8-17% concentrate (Uddin *et al.*, 2013; Bashar *et al.*, 2024a). To mitigate the rising demand for meat and milk products, producers are searching for green grass with a high biomass yield, which could reduce production costs. One of the most promising strategies for enhancing annual fodder availability and animal productivity is the distribution and cultivation of HYV Napier among farmers. The reason for this is that Napier grass grows rapidly and produces the most biomass among tropical forages (Rusdy, 2016; Islam *et al.*, 2021; Islam *et al.*, 2023; Islam *et al.*, 2024).

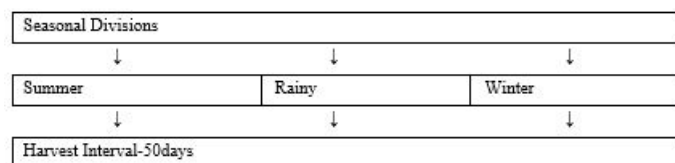
Since its inception, the Bangladesh Livestock Research Institute (BLRI) has preserved the germplasm of eleven Napier (*Pennisetum purpureum*) cultivars suitable for domestic production. These include BN-1(Bajra), BN-2 (Arosha), BN-3 (hybrid), BN-4 (Vietnam), Pakchong, Color Napier, MarkEron, Wruk-Wona, Dwarf-Early, Dwarf-Late, and Zara. In order to address annual feedstuff shortages at the farm level, BLRI distributes cuttings to farmers. Nevertheless, there is not much information regarding year-round biomass production, quality, seasonal impact of the various Napier cultivars. Therefore, advising farmers on the most prospective Napier cultivar in terms of biomass production,

grass (*Pennisetum purpureum*) and pearl millet (*Pennisetum glaucum*). It is distinguished by modest height, abundant tillers, and an improved leaf to stem ratio (Sarker *et al.*, 2021; Ahmed *et al.*, 2021). Its leaves and stems have a couple of barbs that are safe for human skin. This grass takes a while to flower, and it can be harvested 50–60 days after planting, with harvests occurring 40–45 days later. In 1889, the University of Georgia introduced the “MarkEron” dwarf cultivar of Napier grass (*Pennisetum purpureum* Schumacher). It is distinguished by a late-heading growth pattern, an abundance of leaves, and a high feed value. It also offers enhanced production potential and disease resistance. On the other hand, Pakchong is a non-GMO hybrid, developed and introduced by the Department of Livestock Development, Thailand by crossing *Pennisetum purpureum* and *Pennisetum glaucum* known as Pearl Millet Napier. It is commonly known as super Napier grass (<https://theorganicfarmer.org/super-hybrid-napier-grass-pakchong>), which may be harvested after 45 days with 16–18% CP, yielded higher biomass, grow more than 3 meter tall in less than two months (Kiyothong, 2014). BN-4 was taken from Vietnam in 2013. Pakchong was introduced at BLRI from Department of Livestock Services (DLS), Dhaka in 2015. Similarly, Color Napier and Zara were also introduced at BLRI from DLS in 2019. Wruk-Wona, MarkEron, Dwarf-Early, and Dwarf-Late were first introduced at BLRI in 2010, which is leafy with barbs.

(BN-2; Arosha), BLRI Napier-3 (BN-3; hybrid), BLRI Napier-4 (BN-4; Vietnam), Pakchong, Colour Napier, MarkEron, Wruk-Wona, Dwarf-Early, Dwarf-Late, and Zara with 3 replications. Layout of the experimental plots is presented below:

Layout of the experimental plots.

Block 1	Block 2	Block 3
Dwarf-Late	Dwarf-Early	Zara
Color Napier	Wruk-Wona	MarkEron
Dwarf-Early	Napier (BN)-4 (Vietnam)	Napier (BN)-3 (hybrid)
Wruk-Wona	Napier (BN)-1 (Bajra)	Dwarf-Late
BN-2 (Arosha)	Zara	Color Napier
Pakchong	MarkEron	Pakchong
Napier (BN)-4 (Vietnam)	Napier (BN)-3 (hybrid)	BN-2 (Arosha)
Napier (BN)-1 (Bajra)	Dwarf-Late	Dwarf-Early
Zara	Color Napier	Wruk-Wona
MarkEron	Pakchong	Napier (BN)-4 (Vietnam)
Napier (BN)-3 (hybrid)	BN-2 (Arosha)	Napier (BN)-1 (Bajra)



CULTIVATION OF NAPIER CULTIVARS

The cultivars were all grown from stem cuttings consisting two nodes each, and they were planted at 45-degree angle with two pieces each hole. Napier cuttings with 2 nodes were collected from the BLRI fodder germplasm and planted in the allocated plots in 11 rows, with a spacing of 50 cm between cuttings and rows. The total number of block was 3. The harvest interval (HI) was considered for 50 days, and the total number of harvest was 7. However, for all treatments the severity height was considered 10 cm from ground. Irrigation were started 3–7 days after transplanting and broadcasted urea at 156 kg/ha fortnightly two times after each harvest during the whole study period. Sufficient irrigation of plots was continued during the study period, using sprinkler irrigation systems. Weeds were removed manually according to infestations. In general, there are three types of cropping system prevailing in Bangladesh (Banglapedia, 2021): Summer, Rainy and Winter. Therefore, the whole experimental period was divided in to three season such as summer, rainy and winter.

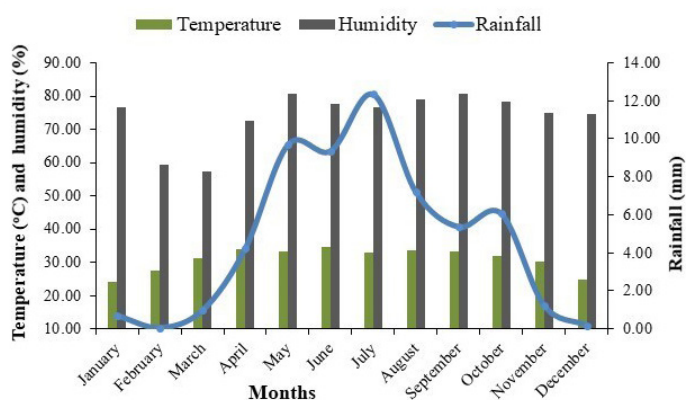


Figure 1: Average temperature (°C), humidity (%) and rainfall (mm) of experimental period.

LAND PREPARATION AND PLOT ALLOCATION

A 33-decimal land was ploughed once with a Mahindra tractor, ploughed thrice with a disc tractor, removed weeds by hand, and labeled properly by using a harrow. Before labeling, a basal doses of urea (nitrogen fertilizer), triple super phosphate, muirate of potash and Zipsam at 292, 214, 56, and 90 kg/ha were applied which was previously determined according to the soil nutrient analysis report (Table 1). The land was then divided into 33 equal plots having 4 x 4 m², separated by an alley of about 2.5 m. Plots were allocated to transplant of 11 Napier cultivars, specifically BLRI Napier-1 (BN-1; Bajra), BLRI Napier-2

PARAMETERS STUDIED

We documented data for plant height (PH), total leaves number (NL) including dead and green, length and width of leaves (LL and LW), number of tillers in each hill, number of nodes in a stem, and stem diameter (SD). On the day of harvest in every 50-day interval, Napier heights were recorded from randomly selected representative nine hills (out of 81 hills) in each plot in its natural condition, including records of heights by raising hills. The number of tillers for each hill was also recorded. To record leaf parameters, a representative Napier tiller from each hill was taken, and the leaves and nodes number, length and width of leaves, and diameter of stem were measured by using measuring tape. Fresh weight of leaves and stem were taken to determine leaf and stem ratio. The stem diameter was taken at the middle of each stem. After harvesting at 10 cm from ground, fresh biomass yield of every plot was recorded by weighing harvested fodder by a digital weighing balance. They were chopped in about 2 cm length, mixed properly by hand, and about 500 gm of fresh samples were taken with leveled in air-tight zipper bags and stored in a freezer (-20°C) for further chemical analysis.

THE DETERMINATION OF HEAT INDEX (HI)

The temperature, humidity, and rainfall data were recorded between February 2020 and February 2021. The US Department of Commerce's National Weather Service's HI chart (www.nws.noaa.gov/om/heat/heat_index.shtml) was used to generate the Heat Index, which is a indicator of how hot it actually feels when relative humidity and air temperature both are combined.

CHEMICAL ANALYSIS

Dry matter (DM), organic matter (OM), crude protein (CP) and ether extract (EE) in different Napier grass samples were estimated as per AOAC (2005). Cell wall constituents viz neutral detergent fiber (NDF) and acid detergent fiber (ADF) were measured as per Van Soest *et al.* (1991). The adiabatic bomb calorimeter (IKA* C5000) was used to measure the gross energy (GE) of feed samples.

IN VITRO STUDY

ANIMAL AND DIET: Rumen fluid was collected from two RCC (Red Chittagong Cattle) bulls which were ruminally fistulated before and fed *ad libitum* Napier grass containing 12% CP and supplied 1.5% body weight of the concentrate mixture, which consisted of 33% wheat bran, 20% khesari bran, 15% rice bran, 10% crushed maize, 10% crushed wheat, 5% soybean, 2% vitamin, mineral premix, 1% DCP, 1% salt and 3% molasses. Drinking water was supplied to the bulls *ad libitum*.

IN VITRO GAS PRODUCTION (GP) KINETICS: For *in vitro* GP kinetics, we followed a procedure, which was described by

Menke and Steingass, 1988. The GP value after 24 hours was used to estimate the dOM and ME, and Menke and Steingass's (1988) equations 43e and 12e were used to analyze the nutrients:

$$\begin{aligned} \text{dOM (\%)} &= 15.38 + 0.8453 \text{ GP}_{24} + 0.0595 \text{ CP} + 0.0675 \text{ CA} \\ \text{ME (MJ/kg DM)} &= 2.43 + 0.1206 \text{ GP}_{24} + 0.0069 \text{ CP} + 0.0187 \text{ CF} \end{aligned}$$

Where, CP stands for crude protein (g kg⁻¹ DM), CA for crude ash (g/kg DM), CF for crude fat (g/kg DM), and GP₂₄ for GP within 24 hours of incubation (ml/200 mg DM).

OXALATE, NITRATE- NITROGEN AND WATER SOLUBLE CARBOHYDRATE (WSC)

The method outlined by Rahman *et al.* (2020) was used to determine the oxalate content utilizing Smith *et al.* (1964) WSC and HPLC. Based on the nitration of salicylic acid in extremely acidic circumstances and the colorimetric analysis of the resultant colored complex, which absorbs maximum at 410 nm in basic solutions as described by Cataldo *et al.* (1975), the nitrate-N of the grass was ascertained.

STATISTICAL ANALYSIS

The response to morphological characteristics, biomass yield, nutritional quality, and *in-vitro* digestibility of different Napier cultivars were subjected to analysis statistically in an ANOVA (Steel and Torrie, 1980) of a Randomized Complete Block Design (RCBD) using GLM procedures of SPSS, version 20 for Windows (SPSS) computer software packages. The differences among mean values were compared by Tukey's Honestly Significant Difference (HSD) Test at a 5% significance level (Steel and Torrie, 1980). The following general linear model was used to investigate the overall fixed effects and interactions between effects in a 3 × 11 factorial configuration with three seasons and eleven varieties:

$$Y_{ijkl} = \mu + S_i + V_j + B_k + S_i \times V_j + E_{ijkl}$$

Where, Y_{ijk} was the dependent variable, μ was population mean, S_i = fixed effect of season *i* (*i* = 1, 2, 3; i.e., summer, rainy, winter), V_j = fixed effect of variety *j* (*j* = 1, 2, 3...11; i.e., BN-1, BN-2, BN-3 ----- Zara), B_k = random effect of blocks *n* (*n* = 3), E_{ijkl} = residual error, assumed to be normally and independently distributed and S_i × V_j were the fixed effect of *i*th season *j*th variety and their interaction, respectively.

ANIMAL ETHICS APPROVAL

The animal study protocol was approved by the Ethics Committee of Bangladesh Livestock Research Institute with protocol code AECC/BLRI 00022/25 for studies involving animals.

Table 2: Morphological characteristics of Napier cultivars.

Parameters	PH (normal; cm)	PH (raised, cm)	Leaf number	Dead LN	Green LN	LL (cm)	LW (cm)	Tiller/hill	Node/stem	SD (mm)
Season										
Summer	116 ^b	147 ^b	9.00 ^c	2.15 ^b	6.88 ^b	76.0 ^b	2.22 ^c	30.7 ^c	4.77 ^b	13.6 ^a
Rainy	136 ^a	168 ^a	11.0 ^a	2.44 ^a	8.44 ^a	88.0 ^a	3.24 ^a	37.1 ^b	6.61 ^a	10.9 ^b
Winter	99 ^c	130 ^c	10.0 ^b	1.84 ^c	8.10 ^a	63.0 ^c	2.48 ^b	44.8 ^a	3.69 ^c	10.6 ^b
Variety										
BN-1 (Bajra)	113	145 ^{ab}	10	2.01	7.88	74.0	2.43 ^c	36.2	4.68	10.2 ^c
BN-2 (Arosha)	114	144 ^{ab}	10	2.05	7.61	73.0	2.51 ^c	36.9	5.03	11.3 ^{bc}
BN-3 (Hybrid)	102	133 ^b	10	2.07	7.56	71.0	2.65 ^{bc}	33.4	3.78	11.1 ^{bc}
BN-4 (Vietnam)	115	149 ^{ab}	10	2.14	8.04	75.0	2.68 ^{bc}	39.5	4.99	11.3 ^{bc}
Pakchong	121	163 ^a	11	2.19	8.37	79.0	3.14 ^a	41.4	5.75	13.1 ^a
Colour Napier	116	141 ^{ab}	10	2.85	8.15	75.0	2.50 ^c	41.9	4.63	11.2 ^{bc}
MarkEron	110	135 ^b	9	2.08	7.25	70.0	2.45 ^c	36.5	4.61	11.8 ^b
Wruk-Wona	114	156 ^{ab}	10	2.17	8.02	72.0	2.62 ^{bc}	41.8	5.02	11.7 ^b
Dwarf-Early	121	147 ^{ab}	10	2.15	7.95	76.0	2.55 ^c	39.5	4.92	10.7 ^{bc}
Dwarf-Late	110	137 ^{ab}	10	2.18	7.63	68.0	2.46 ^c	40.2	4.32	11.6 ^b
Zara	119	153 ^{ab}	10	2.23	7.89	78.0	2.87 ^b	36.5	5.46	13.2 ^a
SEM	2.12	2.06	0.11	0.05	0.11	1.11	0.04	0.78	0.17	0.14
Season	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001
Variety	0.68	<0.01	0.31	0.88	0.30	0.17	<0.001	0.06	0.18	<0.001
S × V	0.99	0.96	<0.05	0.39	<0.01	0.26	0.06	0.45	<0.01	<0.001

Abbreviations: Summer season 1 (16 March to 30 June); Rainy season (01 July to 15 October); Winter season (16th October to 15th March); BN-1, BLRI Napier-1; BN-2, BLRI Napier-2; BN-3, BLRI Napier-3; BN-4, BLRI Napier-4; PH, plant height; LN, leaf number; SD, stem diameter; LL, leaf length; LW, leaf width; S × V, interaction effect between season and variety; SEM, standard error of the mean; ^{a-c} means with different superscripts in the same row are significantly different at $p < 0.05$ or $p < 0.01$ or $p < 0.001$.

RESULTS

MORPHOLOGY OF NAPIER CULTIVARS

The morphological characteristics of the Napier cultivars are shown in Table 2. Seasonal variations showed that morphological characteristics were optimal of Napier grass were found in rainy ($p < 0.001$), compared to summer and winter seasons, respectively, except the highest stem diameter in the summer season. Regarding varietal differences, height in normal standing was similar ($p > 0.05$), however, raised height was similar among Pakchong, Wruk-Wona, Zara, BN-4, Dwarf-Early, BN-1, BN-2, Colour Napier and Dwarf-Late varieties (137–163 cm), with the lowest in BN-3 (133 cm) ($p < 0.01$). All the varieties had a similar number of total, dead and green leaves at 9–11, 2–3, and 7–8, respectively ($p > 0.05$). With respect to leaf length, the highest was found in Pakchong, followed by Zara, (79 and 78 cm, respectively), with the lowest in Dwarf-Late (68 cm; $p > 0.05$). Similarly, Pakchong had the highest leaf width (3.14 cm), followed by Zara, BN-4, with the lowest in BN-1 and Dwarf-late (2.4 cm) ($p < 0.001$). The leaf width of BN-1, BN-2, Colour Napier, MarkEron, Dwarf-

Early and Dwarf-Late were similar ($p > 0.05$). The greater tiller number per hill (41, on average) was found in Pakchong, Color Napier, and Wruk-Wona, with the lowest in BN-3 (33) ($p > 0.05$).

Node number in stem was greater in Pakchong and Zara (5.75 and 5.46, respectively) and the lowest was in BN-3 (3.78) ($p > 0.05$). However, nodes number did not varied significantly ($p > 0.05$) among the different Napier cultivars. Similarly, Pakchong and Zara had a greater stem diameter than other varieties ($p < 0.001$); it was similar with BN-2, BN-3, BN-4, Colour Napier, and Dwarf-early, ($p > 0.05$), with the lowest in BN-1. From the morphological parameters, it could be assumed that the yielding of more biomass could be found from the cultivation of Pakchong, Zara, BN-4, and Wruk-Wona, respectively, than other cultivars.

LEAF AND STEM RATIO OF NAPIER CULTIVARS

Leaf stem ratios of different Napier cultivars are presented in Table 3. Irrespective of varieties, both leaf to stem ratios and dry matter content in leaf and stem were significantly

($p < 0.001$) affected by the season. Leaf to stem ratios were highest ($p < 0.001$) in winter season than that of summer and rainy. On the other hand, summer exhibited greater ($p < 0.001$) DM content both in leaf and stem than that of rainy and winter season.

Table 3: Leaf and stem percentage of different Napier cultivars.

Parameters	Leaf stem ratio		% DM content	
	Fresh basis	DM basis	Leaf	Stem
Season				
Summer	0.56 ^b	0.91 ^b	22.6 ^a	14.7 ^a
Rainy	0.49 ^b	0.99 ^b	20.5 ^b	10.2 ^b
Winter	1.10 ^a	2.17 ^a	20.3 ^b	10.7 ^b
Variety				
BN-1 (Bajra)	0.69 ^{ab}	1.18	20.6 ^{ab}	12.1 ^{abc}
BN-2 (Arosha)	0.72 ^{ab}	1.33	21.4 ^{ab}	12.6 ^{ab}
BN-3 (Hybrid)	1.06 ^a	1.86	21.5 ^{ab}	12.4 ^{ab}
BN-4 (Vietnam)	0.90 ^{ab}	1.73	20.6 ^{ab}	11.6 ^{bcd}
Pakchong	0.52 ^b	1.08	19.9 ^b	10.1 ^d
Colour Napier	0.95 ^{ab}	1.98	20.5 ^{ab}	10.8 ^{bcd}
MarkEron	0.90 ^{ab}	1.80	21.9 ^{ab}	13.6 ^a
Wruk-Wona	0.71 ^{ab}	1.34	20.2 ^b	12.1 ^{abc}
Dwarf-Early	0.72 ^{ab}	1.44	22.5 ^a	11.2 ^{bcd}
Dwarf-Late	0.74 ^{ab}	1.38	21.6 ^{ab}	11.9 ^{abcd}
Zar	0.52 ^b	1.06	20.2 ^b	10.4 ^{cd}
SEM	0.03	0.08	0.17	0.19
Season (S)	<0.001	<0.001	<0.001	<0.001
Variety (V)	<0.01	0.15	<0.005	<0.001
S × V	0.31	0.46	0.90	0.10

Abbreviations: Summer season 1 (16 March to 30 June); Rainy season (01 July to 15 October); Winter season (16th October to 15th March); BN-1, BLRI Napier-1; BN-2, BLRI Napier-2; BN-3, BLRI Napier-3; BN-4, BLRI Napier-4; DM, Dry matter; S × V, interaction effect between season and variety; SEM, standard error of the mean; ^{a-d} means with different superscripts in the same row are significantly different at $p < 0.05$ or $p < 0.01$ or $p < 0.001$.

Irrespective of Season, Napier cultivars exerted variation in the ratios of leaf to stem they contained, with the highest in BN-3 and the lowest in Zara and Pakchong on a fresh basis ($p < 0.01$). When expressed on a DM basis, they also followed the similar results ($p > 0.05$). Followed by BN-3, other varieties that contained a greater percentage of leaf include Colour Napier, MarkEron, BN-4, Dwarf-Late, Dwarf-Early, BN-2, Wruk-Wona and BN-1, on fresh basis, which were similar to each other ($p > 0.05$). The DM percentage of leaves in different cultivars varied ($p < 0.01$) from a range of 19.9 to 22.5, with the highest

in Dwarf-Early and lowest in Pakchong. The DM content in the leaf of BN-1, BN-2, BN-3, BN-4, Colour Napier, MarkEron and Dwarf-Late did not differ significantly ($p > 0.05$). However, the stem of MarkEron had the higher DM content, with the lower in Pakchong ($p < 0.001$). The DM content in the stem of BN-1, BN-2, BN-3, Wruk-Wona and Dwarf-Late was similar ($p > 0.05$).

Table 4: Biomass production of different Napier cultivars.

Parameters	Biomass yield (t/ha/yr)			Energy Yield (GJ/ha/yr)	
	Fresh	DM yield	CP yield	ME	GE
Season					
Summer	87.6 ^a	11.9 ^a	1.22 ^a	89.1 ^a	191 ^a
Rainy	75.7 ^b	10.4 ^b	1.19 ^a	74.8 ^b	165 ^b
Winter	50.1 ^c	7.25 ^c	1.10 ^b	54.7 ^c	111 ^c
Variety					
BN-1(Bajra)	220 ^c	31.7 ^{bc}	3.48 ^{bcd}	235 ^{bc}	494 ^{bc}
BN-2 (Arosha)	201 ^d	27.6 ^{cd}	3.34 ^{bcd}	197 ^d	430 ^{cde}
BN-3 (Hybrid)	198 ^{de}	27.8 ^{cd}	3.34 ^{bcd}	206 ^{cd}	431 ^{cde}
BN-4 (Vietnam)	236 ^b	30.2 ^{bc}	3.63 ^{bcd}	218 ^{bcd}	484 ^{bcd}
Pakchong	274 ^a	36.7 ^a	4.44 ^a	278 ^a	597 ^a
Color Napier	172 ^f	25.2 ^d	3.02 ^{de}	188 ^d	385 ^e
MarkEron	201 ^d	28.7 ^{cd}	3.30 ^{cde}	205 ^{cd}	444 ^{cde}
Wruk-Wona	243 ^b	33.5 ^{ab}	3.86 ^{bc}	249 ^{ab}	534 ^{ab}
Dwarf-Early	175 ^f	25.2 ^d	2.90 ^e	184 ^d	384 ^e
Dwarf-Late	184 ^{ef}	27.3 ^{cd}	3.39 ^{bcd}	198 ^d	416 ^{de}
Zara	239 ^b	31.9 ^{bc}	3.93 ^{ab}	250 ^{ab}	520 ^b
SEM	5.45	0.65	0.08	5.41	11.9
Season (S)	<0.001	<0.001	<0.001	<0.001	<0.001
Variety (V)	<0.001	<0.001	<0.001	<0.001	<0.001
S × V	<0.001	<0.001	<0.001	<0.001	<0.001

Abbreviations: Summer season (16 March to 30 June); Rainy season (01 July to 15 October); Winter season (16th October to 15th March); BN-1, BLRI Napier-1; BN-2, BLRI Napier-2; BN-3, BLRI Napier-3; BN-4, BLRI Napier-4; ME, Metabolisable Energy; GE, Gross Energy; GJ, gigajoule; DM, Dry matter; CP, Crude protein; S, Season; V, variety; S × V, interaction effect between season and variety; SEM, standard error of the mean; ^{a-f} means with different superscripts in the same row are significantly different at $p < 0.05$ or $p < 0.01$ or $p < 0.001$.

BIOMASS YIELD OF NAPIER CULTIVARS

Table 4 showed how the biomass and energy yield of various Napier cultivars varied by season, variety, and their interactions. The biomass yield (Fresh, DM, and CP yield) of Napier grass was found to be significantly impacted by season ($p < 0.001$), variety ($p < 0.001$) and their interaction (S×V) on statistical analysis. Irrespective of varieties, fresh biomass, DM, and CP yield (t/ha) of Napier cultivars were

Table 5: Effect of season on production performance of different Napier cultivars.

Napier Cultivars	DM yield (ton/ha)				Sig. level	CP yield (ton/ha)				Sig. level
	Summer	Rainy	Winter	SEM		Summer	Rainy	Winter	SEM	
BN-1 (Bajra)	13.21 ^a	10.94 ^b	7.57 ^c	0.84	$p < 0.001$	1.35 ^a	1.08 ^b	1.05 ^b	0.05	$p < 0.05$
BN-2 (Aroscha)	9.77 ^a	11.46 ^a	6.40 ^b	0.79	$p < 0.01$	1.02 ^b	1.37 ^a	0.94 ^b	0.07	$p < 0.01$
BN-3 (Hybrid)	9.93 ^a	10.47 ^a	7.44 ^b	0.49	$p < 0.001$	1.03 ^b	1.21 ^a	1.09 ^{ab}	0.03	$p < 0.05$
BN-4 (Vietnam)	11.41 ^a	11.11 ^a	7.71 ^b	0.67	$p < 0.01$	1.1	1.28	1.24	0.05	$p > 0.05$
Pakchong	15.57 ^a	11.30 ^b	9.85 ^b	0.88	$p < 0.001$	1.62 ^a	1.32 ^b	1.49 ^{ab}	0.05	$p < 0.05$
Color Napier	12.07 ^a	7.75 ^b	5.35 ^c	0.99	$p < 0.001$	1.15 ^a	0.98 ^b	0.88 ^b	0.04	$p < 0.01$
MarkEron	10.75 ^a	11.37 ^a	6.60 ^b	0.78	$p < 0.001$	1.07 ^{ab}	1.25 ^a	0.98 ^b	0.05	$p < 0.05$
Wruk-Wona	15.27 ^a	10.83 ^b	7.42 ^c	1.18	$p < 0.001$	1.61 ^a	1.17 ^b	1.07 ^b	0.09	$p < 0.01$
Dwarf-Early	10.46 ^a	9.04 ^a	5.70 ^b	0.73	$p < 0.001$	1.01	0.96	0.93	0.02	$p > 0.05$
Dwarf-Late	10.41 ^a	10.27 ^a	6.63 ^b	0.67	$p < 0.01$	1.01	1.28	1.09	0.06	$p > 0.05$
Zara	12.17 ^a	10.77 ^b	9.04 ^b	0.53	$p < 0.05$	1.46	1.16	1.31	0.06	$p > 0.05$

Abbreviations: Summer season (16 March to 30 June); Rainy season (01 July to 15 October); Winter season (16th October to 15th March); BN-1, BLRI Napier-1; BN-2, BLRI Napier-2; BN-3, BLRI Napier-3; BN-4, BLRI Napier-4; SEM, standard error of the mean; ^{a-c} means with different superscripts in the same row are significantly different at $p < 0.05$ or $p < 0.01$ or $p < 0.001$.

significantly ($p < 0.001$) higher in summer (87.6, 11.9 and 1.22 t/ha, respectively) followed by rainy (75.7, 10.4 and 1.19 t/ha, respectively) and winter season (50.1, 7.25 and 1.10 t/ha). Irrespective of seasons, Pakchong produced highest ($p < 0.001$) fresh (274 t/ha/yr) and DM biomass (36.7 t/ha/yr), with the lowest in Color Napier (172 and 25.1 t/ha/yr, respectively). Fresh yield of Pakchong was followed by Wruk-Wona, Zara, and BN-4 which did not differ significantly ($p > 0.05$). DM yield of Pakchong was followed by Wruk-Wona, Zara, and BN-1, which were similar ($p > 0.05$). Similarly, the CP yield was significantly ($p < 0.001$) higher in Pakchong followed by Zara, Wruk-Wona and BN-4. However, the CP yield of Zara, Wruk-Wona and BN-4 was similar ($p > 0.05$).

When considering the performance of individual cultivars throughout the course of the season, it was found that all Napier cultivars had significantly varying season-wise DM and CP yields (Table 5). Pakchong's DM yield was higher ($p < 0.001$) in summer (15.57 t/ha), followed by rainy season (11.30 t/ha) and winter (9.85 t/ha). However, in Table 5 data showed that there were no appreciable variations ($p > 0.05$) between the rainy and winter seasons. With the exception of Zara, Dwarf-Late, Dwarf-Early, and BN-4, all Napier cultivars showed a significant difference in season-wise CP yield. No significant ($p > 0.05$) differences were observed in the CP production of Pakchong over rainy and winter seasons, although the maximum CP yield (1.62 t/ha) was found in summer, followed by winter (1.49 t/ha), and rainy (1.32 t/ha).

Results showed that, Color Napier attained the lower position in aspect of production performance among the different Napier cultivar, the increment rate of DM and

CP yield of Pakchong was 31% and 32%, respectively in comparison to the yield of Color Napier grass. Whereas, DM and CP yield increment rate of Wruk-Wona (25% and 22%, respectively); and Zara (21% and 23%, respectively) were also clearly visible against Color Napier. In comparison to the yield of winter season, the increment rate of DM and CP yield in summer season were 39% and 10%, respectively whereas in rainy season the increment rate were 31% and 7.5%, respectively.

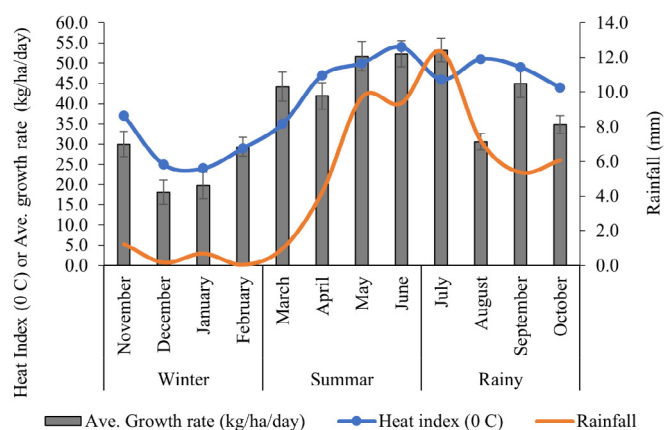


Figure 2: Effect of heat index and rainfall on average growth rate (kg/ha/day) of Napier cultivars.

The cutting number and environmental factors (Heat Index and rainfall) influenced the DM yield of various Napier cultivars (Figure 2 and 3). Figure 3 illustrates that during the second harvest, all Napier cultivars produced greater biomass than in the first harvest; nevertheless, Wruk-Wona and Pakchong demonstrated the highest biomass, reaching 10.59 and 10.02 ton/ha, respectively. Moreover, additional variables such as Heat Index (35° to 54° C), precipitation

(0.97 mm to 9.38 mm), and seasonal changes, particularly summer, also affected their biomass yield (Figure 2). As the number of cuts increased, the production of various Napier cultivars gradually diminished, with the lowest yield recorded during the sixth cut in December due to a decline in harvest index and rainfall (Figure 2).

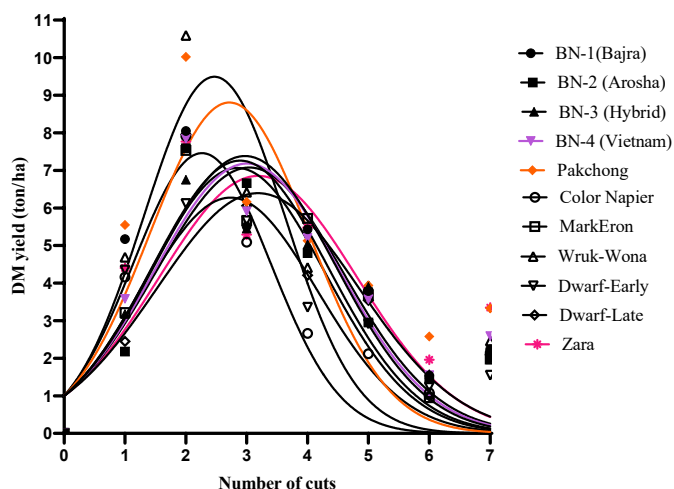


Figure 3: Cutting effect on dry matter yield of different Napier cultivars.

Both the season and varieties had significant ($p < 0.001$) effect on ME, and GE yield. Irrespective of varieties, the total ME, and GE yield per hectare land of Napier grass was significantly ($p < 0.001$) higher in summer (89.1 and 191 GJ ha⁻¹, respectively) followed by rainy (74.8 and 165 GJ/ha, respectively) and winter season (54.7 and 111 GJ/ha, respectively). Irrespective of seasons, Pakchong grass yielded significantly ($p < 0.001$) higher ME, and GE (278 GJ/ha/yr and 597 GJ/ha/yr, respectively), with the lowest in Color Napier and Dwarf-early (Table 4). The ME and GE yield of Pakchong grass was followed by Zara, and Wruk-Wona which were similar ($p > 0.05$).

Among the Napier cultivars Pakchong, Zara, Wruk-Wona, and BN-4 were found more promising than other varieties in terms of fresh, DM, CP, GE, and ME yield. The interactions of variety and season had significant effect ($p < 0.001$) on both biomass and energy yield of Napier grass.

CHEMICAL COMPOSITION OF NAPIER CULTIVARS

Table 6 shows the chemical composition of different Napier cultivars. Both season and variety had significant effect on nutritional composition of Napier grass. Irrespective of varieties, the highest DM, ADF, and GE content was obtained in Napier grass from summer season than the rainy and winter seasons ($p < 0.001$). However, the CP content was higher in winter than in other seasons ($p < 0.001$). There was no variation in OM and NDF content during summer and rainy seasons ($p > 0.05$). Regarding varieties, significantly ($p < 0.001$) greater DM was found

in Dwarf-Late (17.2%), with the lower in Zara (13.8%). Followed by Dwarf-Late, the DM content of MarkEron, BN-1, Dwarf-Early, BN-2, BN-3, Colour Napier, Wruk-Wona, Pakchong and BN-4 was similar ($p > 0.05$). Organic matter of Dwarf-Early was highest, with lowest in BN-1 ($p < 0.001$). Followed by Dwarf-Early, OM of Pakchong, Dwarf-late, MarkEron, and Zara was similar ($p > 0.05$). The highest CP was found in Color Napier and Dwarf-Late which differed significantly from other Napier varieties ($p < 0.001$). Followed by them, the level of CP in BN-2, BN-3, BN-4, Pakchong, Dwarf-Early, and Zara was similar ($p > 0.05$).

Table 6: Chemical composition and gross energy of Napier cultivars in different seasons.

Parameters	Chemical composition (% DM) and gross energy (MJkg ⁻¹ DM)					
	DM(% OM fresh)	CP	ADF	NDF	GE	
Seasons						
Summer	15.9 ^a	88.7 ^a	10.2 ^c	50.4 ^a	77.0 ^a	16.06 ^a
Rainy	13.8 ^b	88.9 ^a	11.3 ^b	48.4 ^b	77.2 ^a	15.72 ^b
Winter	15.6 ^a	88.0 ^b	15.2 ^a	44.4 ^c	74.8 ^b	15.26 ^c
Variety						
BN-1(Bajra)	15.6 ^{ab}	86.5 ^c	11.3 ^c	47.4 ^{ab}	77.5 ^{abc}	15.60 ^c
BN-2 (Arosha)	15.4 ^{ab}	88.2 ^{cd}	12.3 ^{cd}	48.1 ^{ab}	75.6 ^{bc}	15.59 ^c
BN-3 (Hybrid)	15.3 ^{ab}	88.0 ^d	12.2 ^{cd}	48.0 ^{ab}	78.6 ^{ab}	15.51 ^c
BN-4(Vietnam)	14.0 ^{ab}	88.0 ^d	12.4 ^{bc}	43.5 ^c	74.9 ^c	16.02 ^b
Pakchong	14.0 ^{ab}	88.9 ^{abcd}	12.4 ^{bc}	49.5 ^{ab}	75.9 ^{bc}	16.26 ^a
Color Napier	15.0 ^{ab}	88.2 ^{cd}	12.9 ^a	47.8 ^{ab}	73.9 ^c	15.30 ^d
MarkEron	16.5 ^{ab}	89.2 ^{abc}	11.9 ^d	50.4 ^a	76.1 ^{bc}	15.49 ^c
Wruk-Wona	14.8 ^{ab}	88.5 ^{bcd}	11.9 ^d	46.8 ^{ab}	75.4 ^{bc}	15.93 ^b
Dwarf-Early	15.5 ^{ab}	90.0 ^a	12.1 ^{cd}	47.4 ^{ab}	79.7 ^a	15.25 ^d
Dwarf-Late	17.2 ^a	89.5 ^{ab}	12.8 ^{ab}	46.7 ^b	75.1 ^{bc}	15.23 ^d
Zara	13.8 ^b	89.0 ^{abcd}	12.4 ^{cd}	49.2 ^{ab}	76.7 ^{abc}	16.28 ^a
SEM	0.22	0.15	0.23	0.40	0.31	0.05
Season	<0.001	<0.001	<0.001	<0.001	<0.001	<.001
Variety	<0.05	<0.001	<0.001	<0.001	<0.001	<.001
S × V	0.912	<0.001	<0.001	<0.01	0.10	0.11

Abbreviations: Summer season 1 (16 March to 30 June); Rainy season (01 July to 15 October); Winter season (16th October to 15th March); DM, Dry matter, BN-1, BLRI Napier-1; BN-2, BLRI Napier-2; BN-3, BLRI Napier-3; BN-4, BLRI Napier-4; DM, Dry matter; OM, Organic matter; CP, crude protein; ADF, Acid detergent fiber; NDF, Neutral detergent fiber; GE, Gross energy; MJ, megajoule; S, Season; V, variety; S × V, interaction effect between season and variety; SEM, Standard error of the mean, ^{a-c} means with different superscripts in the same row are significantly different at $p < 0.05$ or $p < 0.01$ or $p < 0.001$.

The ADF was significantly lower in BN-4 than in any other Napier varieties ($p < 0.001$). However, MarkEron contained the higher ADF content ($p < 0.001$). Followed by MarkEron, the ADF content in BN-1, BN-2, BN-3, Pakchong, Color Napier, Wruk-Wona, Dwarf-Early and Zara did not differ significantly ($p > 0.05$). Regarding NDF, the highest level was found in Dwarf-Early, with the lowest in Color Napier ($p < 0.001$). The NDF of Dwarf-Early was followed by BN-3, BN-1, Zara with similar level ($p > 0.05$). Considering GE content, the Pakchong and Zara contained the highest amount of GE, which was 16.26 and 16.28 MJ/kg DM, respectively ($p < 0.001$), whereas the lowest concentration was observed for Dwarf-Late (15.23 MJ/kg DM). However, no interaction effect between season and variety was observed for DM, NDF, and GE content except OM, CP, and ADF content.

Table 7: *In vitro* digestibility of different Napier cultivars with seasons.

Parameters	In vitro digestibility		
	GP ₂₄ (ml)	dOM (%)	ME (MJ/kg DM)
Season			
Summer	35.1 ^a	58.7 ^a	7.47 ^a
Rainy	31.7 ^b	56.3 ^b	7.14 ^b
Winter	32.8 ^b	60.4 ^a	7.55 ^a
Variety			
BN-1(Bajra)	34.2 ^{ab}	60.0	7.43 ^{ab}
BN-2 (Arosha)	31.2 ^b	57.2	7.13 ^b
BN-3 (Hybrid)	33.4 ^{ab}	59.1	7.40 ^{ab}
BN-4 (Vietnam)	31.7 ^{ab}	57.6	7.24 ^{ab}
Pakchong	35.1 ^{ab}	60.8	7.83 ^a
Color Napier	33.3 ^{ab}	59.2	7.48 ^{ab}
MarkEron	31.5 ^{ab}	56.5	7.15 ^b
Wruk-Wona	33.9 ^{ab}	58.8	7.43 ^{ab}
Dwarf-Early	32.7 ^{ab}	57.2	7.31 ^{ab}
Dwarf-Late	31.6 ^{ab}	56.9	7.27 ^{ab}
Zara	36.8 ^a	59.6	7.58 ^{ab}
SEM	0.40	0.39	0.05
Season (S)	<0.001	<0.001	<0.001
Variety (V)	<0.05	0.08	<0.05
S × V	0.15	<0.05	0.14

Abbreviations: Summer season 1 (16 March to 30 June); Rainy season (01 July to 15 October); Winter season (16th October to 15th March); BN-1, BLRI Napier-1; BN-2, BLRI Napier-2; BN-3, BLRI Napier-3; BN-4, BLRI Napier-4; GP₂₄, Gas production at 24 h; dOM, Digestible organic matter; ME, Metabolisable energy; MJ, megajoule; S, Seasons; V, Variety; S × V, interaction effect between season and variety; SEM, Standard error of the mean; ^{a-b} means with different superscripts in the same row are significantly different at $p < 0.05$ or $p < 0.01$ or $p < 0.001$.

IN VITRO DIGESTIBILITY OF DIFFERENT NAPIER CULTIVARS WITH THEIR SEASONAL EFFECT

Table 7 illustrates the *in vitro* digestibility of various Napier cultivars. Season had significant effect ($p < 0.001$) on the 24- hour gas production (GP₂₄), digestible organic matter (dOM) and metabolisable energy (ME) content of different Napier cultivars. During summer, the GP₂₄ was significantly higher (35.1 ml) followed by winter (32.8 ml) and rainy (31.7 ml) season. Nonetheless, their dOM and ME content exhibit greater efficiency ($p < 0.001$) during the winter season in comparison to other seasons. Among various Napier cultivars, Zara generated the highest ($p < 0.05$) amount of gas (36.8 ml) at 24 hours followed by Pakchong grass (35.1 ml). Irrespective of season, Pakchong had considerably higher dOM (60.8%) and ME (7.83 MJ/kg DM) compared to the other cultivars. No substantial variation ($p > 0.05$) was noted among GP₂₄, dOM, and ME throughout BN-1, BN-3, BN-4, Pakchong, Color Napier, Wruk-Wona, Dwarf-Early and Dwarf-Late. BN-2 yielded the lowest gas (31.2 ml) at 24 h, and their dOM and ME were similarly markedly reduced. An interaction effect between season and variety was seen on the dOM of Napier grass ($p < 0.05$).

WATER-SOLUBLE CHO (WSC), NITRATE-N (NO₃-N) AND SOLUBLE OXALATE

Seasons had significant effect on water soluble carbohydrate (WSC), nitrate nitrogen (NO₃-N) and gross energy (GE) content in Napier grass. The WSC content of Napier cultivars in summer was highest ($p < 0.001$), whereas lowest concentration of NO₃-N was measured in the winter season (Table 8). When comparing the Napier cultivars, the Zara contained the highest amount of WSC (141 g/kg DM; $p < 0.001$) followed by Pakchong grass (139 g/kg DM). Followed by them, the WSC content in BN-2, Color Napier, MarkEron, BN-4, BN-3 and Wruk-Wona did not vary significantly ($p > 0.05$). Irrespective of season, the soluble oxalate content was lowest in Pakchong grass (9.37 g/kg DM; $p < 0.05$) in contrast to other Napier cultivars. However, Dwarf-Late and Dwarf-Early varieties exhibited the highest concentrations of soluble oxalate, measuring 14.7 and 14.5 g/kg DM, respectively. Nonetheless, their WSC was the lowest ($p < 0.001$) and demonstrated a negative correlation. Among the Napier cultivars, there was no significant difference in the NO₃-N concentration ($p > 0.05$). However, an interaction effect between season and variety was found on the NO₃-N concentration of Napier grass ($p < 0.001$).

DISCUSSION

MORPHOLOGY OF NAPIER CULTIVARS

Napier grass is a popular and significant feed sources for small-scale farmers in the tropics and subtropics including

Bangladesh because of its high biomass yield (Mukhtar *et al.*, 2022) and for quick regrowth and propagation potentiality (Lee *et al.*, 2016). There are over 300 accessions and 140 Napier species in different gene banks worldwide (Brunk-en, 1977; Negawo *et al.*, 2017). In Bangladesh, around 11 Napier cultivars are preserved at the Bangladesh Livestock Research Institute Fodder Germplasm Bank, with some cultivars utilised at the farmer level. Being a fast-growing tropical forage (Holm *et al.*, 1979), the height of Napier grass (normal and raised) during rainy season was found greater than in other seasons due to favorable warm and moist climatic conditions of this season (Figure 1). When compared to BN-3 and other napier cultivars, Pakchong may produce substantially more biomass due to its tallest plants (Sarker *et al.*, 2019). Plant height, on the other hand, is genetically determined and mostly subject to selection, with minimal influence from environmental conditions. According to Assuero and Tognetti (2010), a combination of genetic, physiological, and environmental variables contribute to the regulation of tillering in grasses. Similarly, the number of leaves their length and width were greater during the rainy season. Particularly, higher average day-time temperature and humidity during summer and rainy might help greater growth of Napier grass than winter season. Kaur *et al.* (2017) found greater tiller number per plant and leaf length during monsoon than in autumn and winter seasons. The raised height of BN-1 was significantly ($P < 0.01$) greater than BN-3 and MarkEron (Sarker *et al.*, 2021), which were numerically comparable to the current investigation. Raised height of BN-1, BN-3, BN-4, MarkEron and, Wruk-Wona (116 to 226 cm), as reported by Sarker *et al.* (2018; 2021), is similar to the values of the present study (133 to 156 cm). The number of tillers per hill of this study (33 to 42) in BN-1, BN-3, BN-4, MarkEron, and Wruk-Wona is comparable to the values (22–50) reported by Sarker *et al.* (2018; 2021).

LEAF AND STEM RATIO OF NAPIER CULTIVARS

One important aspect influencing the nutritional richness of forages and the quality of grazing is the leaf-to-stem ratio (Annicchiarico, 2007). According to earlier research, the leaves of forage legumes add more nutrients than the stems, which are of lower quality (Rao and Northup, 2012). As the growing season lengthens, the amounts of ADF and NDF progressively rise, and as the season goes on, the various cell wall components in the plant biomass rise as well, culminating in high concentrations at maturity (Baath *et al.*, 2020). However, our findings indicated a plateau in concentrations during the summer rather than consistent rises, which was probably caused by the legumes' unpredictable development and increased biomass rather than quality (Marković *et al.*, 2012; Baath *et al.*, 2020). According to the earlier investigations, during the rainy and winter seasons, older leaves and younger leaves with less cell

wall abundance result in mild increases in cell wall fractions with higher nutritional composition. Comparable to our current work, Sarker *et al.* (2021) found that the leaves to stem ratio (LSR) varied significantly ($p < 0.001$) for the immediate impact of cultivar.

Table 8: Effect of different Napier cultivars on water-soluble carbohydrate (WSC), nitrate-nitrogen ($\text{NO}_3\text{-N}$), and soluble oxalate.

Parameters	WSC (g/kg DM)	$\text{NO}_3\text{-N}$ (g/kg DM)	Soluble Oxalate (g/kg DM)
Seasons			
Summer	137 ^a	0.86 ^{ab}	–
Rainy	131 ^b	0.94 ^a	–
Winter	127 ^b	0.75 ^b	–
Variety			
BN-1(Bajra)	123 ^{bc}	0.87	9.70 ^b
BN-2 (Arosha)	137 ^{ab}	0.75	10.2 ^{ab}
BN-3 (Hybrid	131 ^{abc}	0.81	11.3 ^{ab}
BN-4 (Vietnam)	133 ^{abc}	0.82	11.0 ^{ab}
Pakchong	139 ^a	0.76	9.37 ^b
Color Napier	137 ^{ab}	1.00	10.9 ^b
MarkEron	136 ^{ab}	0.92	12.3 ^{ab}
Wruk-Wona	129 ^{abc}	0.82	13.5 ^{ab}
Dwarf-Early	120 ^c	0.95	14.5 ^a
Dwarf-Late	121 ^c	0.95	14.7 ^a
Zara	141 ^a	0.74	9.80 ^b
SEM	1.17	0.03	0.12
Season	<0.001	<0.01	–
Variety	<0.001	0.236	<0.05
S × V	0.999	<0.001	–

Abbreviations: Summer season 1 (16 March to 30 June); Rainy season (01 July to 15 October); Winter season (16th October to 15th March); BN-1, BLRI Napier-1; BN-2, BLRI Napier-2; BN-3, BLRI Napier-3; BN-4, BLRI Napier-4; WSC, Water soluble carbohydrate; $\text{NO}_3\text{-N}$, Nitrate-Nitrogen; S, Seasons; V, Variety; S × V, interaction effect between season and variety; SEM, Standard error of the mean; ^{a-c} means with different superscripts in the same row are significantly different at $p < 0.05$ or $p < 0.01$ or $p < 0.001$.

The higher leaf proportion in Napier-3 than Napier-1 of these investigation was also comparable to another research of Sarker *et al.* (2021). Maleko *et al.* (2019) and Soumya (2011) also looked at similar effects, and their research showed that stem and leaf traits account for the differences in LSR across cultivars. Some types have robust stems and few leaves, while others have narrow stems and lots of leaves. Leaf and stem ratio of Napier varieties (0.45 to 0.86 in Napier-1, Napier-3, Napier-4, MarkEron and

Wruk–Wona), as reported by Sarker *et al.* (2018) are similar to the values of the present study (0.69 to 1.06). Halim *et al.* (2013) found that the Napier leaf-to-stem ratio varied from 0.57 to 1.63, where dwarf types yielding higher number of leaves compared to stems, whereas Soumya (2011) reported that in eight hybrid Napier cultivars, the average leaf stem varied between 0.66 and 0.92., a condition linked to full sun (Anderson *et al.*, 2008); yet, some types can also flourish in partial shade.

BIOMASS YIELD OF NAPIER CULTIVARS

Islam *et al.* (2023) said that biomass yield and crude protein concentration of Napier were 26 ton DM/ha/yr and 9.60 g/kg DM, respectively, range from 2 to 86 ton DM /ha/yr and 9.0 to 25.7 g CP/kg DM, suggesting that there is room for the productivity and nutritional value of Napier grass, such as Pakchong, to be significantly increased. Being a fast-growing tropical forage (Holm *et al.*, 1979; Bashar *et al.*, 2017) higher daily temperature and humidity (Figure 2) might favor the vegetative growth of this forage. Kaur *et al.* (2017) found greater biomass production of Napier-1 during monsoon than in other seasons of the year. A farm-level study of Napier grass indicated that annual biomass production of Napier-3 was greater than that of Napier-1 and MarkEron (Sarker *et al.*, 2021) which did not align with the current study's findings. However, biomass production of Napier differ significantly according to season, location, variety, and management techniques (Ogoshi *et al.*, 2010; Xie *et al.*, 2011; Rengsirikul *et al.*, 2011).

CHEMICAL COMPOSITION OF NAPIER CULTIVARS

A similar DM content in Napier-1, Napier-3, Napier-4, MarkEron, and Wruk–Wona of the present study (14 to 17%, fresh) was reported by previous studies (11 to 17%, fresh) (Sarker *et al.*, 2021). The CP content of them (11-12% DM) are similar to those reported by Sarker *et al.* (2018; 2021) (9 to 14% DM). Similar CP content in MarkEron and Napier-3 was reported previously (Sharker *et al.*, 2021) and the values in the present study (11.93-12.22%) are similar to the value reported for the non-drought region (Savar) (11.97%).

The nutritive value of forage is contingent upon its nutrient composition. Crude protein, structural carbohydrates, and voluntary consumption are the primary determinants of nutritional quality forages (Kebede *et al.*, 2016). Protein is a fodder quality indicator which is very much needed for ruminant growth, development, and production (Pinkerton 2005; Kebede *et al.*, 2016). The research result indicated that, the highest CP content was observed for Color Napier (129 g/Kg DM) followed by Dwarf–Late (128 g/Kg DM), Pakchong (124 g/Kg DM), and Zara (124 g/Kg DM), while BN-1 (113 g/Kg DM) gave the lowest CP content. The rate at which the CP content of Napier grass decreases in stems is known to be faster than in leaves

(Brown and Chavalimu, 1985). The higher CP content in Color Napier is due the higher leaf content found in our study than other Napier cultivars. Ruminant maintenance requires a CP concentration of roughly 60–70 g/Kg DM (Leng, 1990; Smith, 1993) whereas a dairy cows must have a CP level of 80–130 g/Kg DM in order to produce 10–15 kg milk per day (ARC, 1984; Humphreys, 1991). According to ARC (1984), about 150 g/Kg DM of CP is needed for more than 15 Kg milk production per day from a cow where the voluntary intake would decrease below the CP content of 60–70 g/kg DM (ARC, 1984; Minson, 1990). Forages with less than 80 g CP/Kg DM are considered low quality (Leng, 1990), and they would have a negative impact on rumen microbial activity (Van Soest, 1982).

The NDF content in different cultivars varied from 739 to 797 g/Kg DM. The maximum amount of NDF was observed in Dwarf–Early (797 g/Kg DM) followed by BN-3 (786 g/Kg DM), BN-1 (775 g/Kg DM) and Zara (767 g/Kg DM) with the lowest was recorded from Color Napier (739 g/Kg DM).

NDF content of Napier-1, MarkEron and Napier-3 reported by Sarker *et al.* (2018) (52-55%) was less than the findings of present study (76-79%). Our study findings were consistent with the result of Sampaio *et al.* (2009), who illustrated that tropical forage has an NDF concentration of roughly 60% DM. Kebede *et al.* (2016) said that NDF properties of ten Napier accessions grown at Holetta Research Center ranged from 728-783 g/Kg DM which were inline with our findings. The feed's quality and digestibility are directly impacted by its fiber content.. Ruminal pH, total and kinetic gas generation, endogenous enzymes, and rumen microorganisms are all impacted by the elevated NDF (Miranda-Romero *et al.*, 2020). The structural component of plants is fiber, which includes cellulose, hemicellulose, insoluble lignin, soluble pectins, and waxes (Van Soest, 1994). The rumen wall's health, rumination, saliva flow, and buffering all depend on adequate fiber, which is assessed as NDF and defined as total cell wall contents (Fox *et al.*, 1992). The high proportion of neutral detergent fiber (NDF) in napier grass typically restricts the amount of DM that ruminants can consume (Islam *et al.*, 2024). Ranathunga *et al.* (2010) said that when forage is the main source of NDF in the diet, there is a negative association between NDF and DMI. Grass with an NDF content between 500 and 600 g/Kg DM may be categorized as moderately high grade, but grasses with an NDF level beyond 600 g/Kg DM are considered low quality (Van Soest, 1982).

ADF content of different Napier cultivars ranged from 435-504 g/Kg DM. The highest ADF properties were observed in MarkEron (504 g/Kg DM) then Pakchong (495 g/Kg DM), and Zara (492 g/Kg DM) with the lowest was

recorded from BN-4 (435 g/Kg DM). Findings of our study were comparable with those [Kebede et al. \(2016\)](#), who reported the ADF content of some Napier accessions propagated at both Holetta and Areka Research Center ranged from 412-496 g/Kg DM. The higher forage ADF results in reduced digestibility dry matter as a consequence of increased lignifications of cellulose in the plants ([Depeters, 1993](#)). ADF-rich fodder has a reduced rumen cellulose digestibility, which lowers the energy available to the nursing cow for milk production. However, biomass production and nutritional composition of Napier grass are influenced by morphological characteristics of different Napier genotypes ([Tudsri et al., 2002](#)). [Islam et al. \(2024\)](#) demonstrated that the nutritional composition of Napier grass is inadequate, often failing to sustain animal production due to its low crude protein (95 g/Kg DM) and metabolisable energy (8.6 MJ/kg DM) content, alongside elevated levels ADF and NDF.

IN VITRO DIGESTIBILITY, WATER-SOLUBLE CHO (WSC), NITRATE-N ($\text{NO}_3\text{-N}$) AND SOLUBLE OXALATE

While [Bashar et al. \(2024b\)](#) reported a similar discovery of dOM and ME, the dOM and ME of Pakchong and Zara (60.8% and 7.83 MJ/kg DM and 59.6% and 7.58 MJ/kg DM, respectively) were much greater than those of the other Napier cultivars. According to [Peyraud and Delagarde \(2013\)](#), 1 kg of milk cow⁻¹ day⁻¹ is lost for every 1% drop in the dOM of grass. According to [Islam et al. \(2021\)](#), there is a 0.83-liter increase in milk output per day for every 1% rise in CP in Napier grass and a 3.5-liter increase in milk yield for every MJ increase in ME. Water soluble carbohydrate (WSC) is a primary substrate for fermentation in the rumen and other fermentative environments, leading to gas production. In the present study revealed that Zara generates significantly higher GP at 24 hours than other Napier cultivars. However the GP at 24 hours, dOM and ME content of Zara and Pakchong did not varying significantly. Higher WSC content in Zara may attributed relatively higher GP at 24 hours. Studies that quantify WSC and in vitro gas volume in forages together are limited. Our result is supported by the findings of [Berthiaume et al. \(2006\)](#) and [Cajaville et al. \(2015\)](#), who reported a favorable correlation between in vitro gas production and WSC.

Nevertheless, Napier grass has low amounts of water-soluble, highly fermentable carbohydrates (WSCs), which can be raised by adding carbohydrate supplements to increased the quality of silage and boost animal performances ([Tauqir et al., 2009](#)). However, our study found that the concentrations of water-soluble carbohydrates in different Napier cultivars ranged from 120 to 141 g/kg DM, with Pakchong and Zara possess the highest concentrations (139 and 141 g/kg DM, respectively). This was in line with the findings

of [Catchpoole and Henzell \(1971\)](#), who found that the WSC contents of the Napier grass accessions ranged from 126 to 149 g/kg DM. According to [Manyawu et al. \(2003\)](#), both varieties and cutting interval significantly affected on WSC content in Napier grass. They observed the WSC content in 6 different Napier cultivars ranged from 110-149 g/kg DM with highest in SDPN 29 variety and lowest in Bana grass. They also found WSC concentration of 91 and 133 g/kg DM, respectively in Napier grass when cut at 8 and 10 weeks interval ([Manyawu et al., 2003](#)).

Animal nutrition is significantly impacted by the oxalate and nitrate nitrogen ($\text{NO}_3\text{-N}$) content of Napier grass. The $\text{NO}_3\text{-N}$ levels in different Napier cultivars of our study ranged from 0.74 g/kg DM to 1.00 g/kg DM, with lowest in Zara (0.74 g kg⁻¹ DM) and Pakchong (0.76 g/kg DM) and highest in Color Napier. Although [Sidhu et al. \(2011\)](#) reported that the average amount of $\text{NO}_3\text{-N}$ in mature Napier grass is 0.5 g/kg DM. According to [Burrows and Tyrl \(1989\)](#), the $\text{NO}_3\text{-N}$ concentration of Napier grass from our study is at a safe level for animals because the recommended limit is 2.5 g/kg DM and forages that contain more than 4.5 g/kg DM are extremely harmful to animals. Additionally, [Adams et al. \(2019\)](#) proposed that the $\text{NO}_3\text{-N}$ content of forages that cause acute toxicity often falls between 2.3 and 6.8 g/kg DM.

According to [Das et al. \(2010\)](#) and [Rahman et al. \(2014\)](#), oxalate is known to negatively affect cattle's body condition score (BCS), calcium, and phosphorus balance. The typical DM total oxalate content of Napier grass ranges from 1 to 39 g/kg. Plants with 20 g/kg DM or more soluble oxalate may injure ruminants acutely, according to [McKenzie et al. \(1988\)](#). Some of the Napier cultivars in our study had soluble oxalate contents ranging from 9.37 g/kg DM to 14.70 g/kg DM, with lowest in Pakchong and highest in Dwarf-Early which suggests that they have no negative impacts on raising livestock in tropical and subtropical areas ([Rahman et al., 2013](#)). According to [El-khodery et al. \(2008\)](#), oxalate levels between 7% and 16.6% can result in acute poisoning and mortality. Less than 2.0% of DM intake is the safe threshold for ruminant consumption of forages containing soluble oxalate in order to prevent the development of excess Ca oxalate ([Pathmasiri et al., 2014](#)).

CONCLUSIONS AND RECOMMENDATIONS

Based on the season, the higher biomass production was observed in summer followed by rainy and winter. However, in winter the nutrient content of Napier cultivars particularly CP and ME was significantly higher than other two seasons. Among the cultivars, Pakchong was more promising in terms of yield, nutrient content, *in vitro* digestibility

and their soluble oxalate content. In addition, Wruk–Wona and Zara stands next to Pakchong.

We recommend that the top three Napier cultivars may perform better in tropical and subtropical regions due to similar climatic conditions, provided that the standard agronomical practices used in this research are followed.

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NOVELTY STATEMENTS

This study provides a comprehensive comparative evaluation of eleven Napier grass cultivars across different seasons, highlighting significant variation in biomass yield, nutritional quality, and in vitro digestibility. The identification of Pakchong as a superior cultivar in terms of biomass productivity, crude protein content, and energy value offers new insights for optimizing forage selection in tropical livestock systems.

AUTHOR'S CONTRIBUTIONS

Anamika Roy: Conceptualisation, literature search, investigation, experimentation, implementation, sample analysis, data collection and compilation, data curation, writing, review and editing; Biplob Kumer Roy: Conceptualization, literature search, establishment, experimentation, administration and implementation, data collection, compilation, data curation and analysis, writing, review and editing; Muhammad Khairul Bashar: Literature search, data curation, writing, review and editing; Md. Mostain Billah, Nasrin Sultana, and Nathu Ram Sarker: Writing, review and editing; Mohammad Al-Mamun: Conceptualisation, overall supervision, investigation, data curation, writing, review and editing. All authors have read and agreed to the published version of the manuscript.

CONFLICT OF INTEREST

There are no conflicts of interest.

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