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RESEARCH

Forage performance of pearl millet (*Pennisetum glaucum* [L.] R. Br.) in arid regions: Yield and quality assessment of new genotypes on different sowing dates

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ABSTRACT

The evaluation of new forage genotypes under arid conditions would contribute to solving the feed shortage problem during the summer season in arid areas and identify potential candidates for particular breeding programs. The present study evaluated the yield and quality of five multi-cut pearl millet (*Pennisetum glaucum* [L.] R. Br.) genotypes (IP19586, IP19612, IP6105, IP13150, and Shandaweel-1) affected by three sowing dates (15 May, 1 June, and 15 June) during the 2017 and 2018 summer seasons in Alexandria, Egypt. Among the new genotypes, IP13150 maintained the desirable balance between both productivity and quality. In addition to its high DM yield (3.50 t ha^{-1}), it was characterized by the highest crude protein (91.6 g kg^{-1}) and N-free extract (500.5 g kg^{-1}) contents, while it had the lowest fiber fractions. This was reflected on its organic matter digestibility (395.7 g kg^{-1}), high gas production (24.5 mL g^{-1} OM), short-chain fatty acid production (47.4 Mm), microbial protein (47.8 g kg^{-1} OM), and the highest energy values among all the genotypes. Although DM yield of the local cv. Shandaweel-1 was moderate (3.2 t ha^{-1}), it was inferior regarding all the tested quality attributes. Altering the sowing date exerted a limited effect on the studied parameters; early sowing on 15 May was superior to later sowing on 1 and 15 June. The superiority of the second cut over the first and third cuts in forage production highlights the success of pearl millet as a multi-cut crop in similar environments.

Key words: Forage grasses, forage quality, fresh yield, gas production, in vitro digestibility.

INTRODUCTION

Feed shortage is a major challenge for the expansion of livestock production to cover the increasing demand for meat and dairy products by the continuously growing population, especially in developing countries (Rai et al., 2012). There are many options to cover the gap between forage demand and supply, one of which is the adoption of high-yielding crop varieties (Hassan et al., 2014; Babiker et al., 2015). Millets are especially gaining popularity due to their high resilience to climate change effects and acceptable productivity and nutritional value (Jukanti et al., 2016).

Pearl millet (*Pennisetum glaucum* [L.] R. Br.) is an annual, warm season crop belonging to the *Poaceae* family. It is drought- and heat-tolerant and has a considerable ability to grow and yield in poor, sandy, and saline soils under arid, hot, and dry climates; this is an advantage over other popular forage grasses in the region, such as fodder maize (Jukanti et al., 2016). It is also a hydrocyanic and prussic acid-free crop, which gives it nutritional superiority over sorghum and

Sudan grass (Hassan et al., 2014). However, it is still mostly cultivated for grain production in Asia and Sub-Saharan Africa (Babiker et al., 2015). There has been increased attention recently for pearl millet as a multi-cut forage crop for fresh feeding and silage production (Jukanti et al., 2016), especially in Brazil, the Middle East, and Central Asia (Rai et al., 2012). Intensive research has been conducted in the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT) to develop new pearl millet lines and hybrids; however, they are focused on evaluating the grain yield and yield components of the new genotypes. Meanwhile, complete information on their forage potential is still lacking (Jukanti et al., 2016). Despite various breeding efforts to produce agronomical elite pearl millet genotypes, their adoption in arid regions is very limited (Divya et al., 2017). There is therefore a dire need to create a complete quantitative and qualitative profile of the new pearl millet genotypes to monitor their integration in the regional forage production systems. Pearl millet is characterized by its abundant phenotypic and genetic variability, which supports its effective use in breeding programs to develop high-yielding genotypes with specific adaptation to arid and semi-arid production systems in many parts of the world. Furthermore, as a climate-resilient crop, pearl millet has great potential as an excellent genomic resource to isolate candidate genes for tolerance to drought and heat stress (Serba et al., 2017). The evaluation of newly released genotypes in different regions of the world, especially those characterized by arid environments such as in the current study, is widely encouraged to facilitate the identification of potential parents for a particular genetic improvement program. Agronomic practices to maximize productivity and quality are still under review for fodder pearl millet. The sowing date is reported to have a significant impact on crop growth and development (Abd El-Lattief, 2011), which is then reflected on yield and quality (Radhouane, 2008). Accurate decision-making as to the sowing date is not only important to achieve the highest crop yield and quality but also to minimize the risk of crop stand failure; this decreases overall farming practice costs by eliminating labor and re-sowing costs (Santos et al., 2017). Manipulating the sowing date is also an effective climate change adaptation strategy (Dharmarathna et al., 2014). The sowing date adjustment is crucial, especially for arid and semi-arid ecosystems, because it is directly related to the challenges of the global warming hazards these regions face (Zhang et al., 2019).

Optimizing the use of any forage crop in livestock feeding requires an understanding not only of its yield and dry matter (DM) accumulation but also changes in its quality. The best forage crop is the one that maintains the balance between high productivity and optimum nutritional profile that improve animal performance. The chemical composition analysis of any given forage is essential to determine its nutritional value. However, to create a complete profile for a particular feedstuff, which truly demonstrates its nutritional value for the animal, a more precise feed evaluation technique should be adopted. In vitro techniques have been the most popular because they are easily applied, repeatable, and require few animals. Thus, the in vitro ruminal fermentation kinetics for gas production is widely used to evaluate the nutritional value of feeds (Aderinboye et al., 2016). Gas measurement is also useful to determine digestion kinetics of both soluble and insoluble fractions of feedstuffs. It also closely indicates the production of short chain fatty acids (Blümmel et al., 1999). A better understanding of the changes in yield potential and nutritional value among different cuts of multi-cut forages is needed by the farmer to estimate their importance for livestock nutrition.

The aim of the present study was to quantify the variations in productivity and quality in terms of nutritional components, ruminal fermentation, and in vitro nutrient degradability of multiple cuts of four newly introduced pearl millet genotypes compared with a local cultivar when grown on different sowing dates in the Egyptian agricultural system.

MATERIALS AND METHODS

Experimental location

Field trials were carried out at the Agricultural Research Station of the Faculty of Agriculture, Alexandria University, Alexandria, Northern Egypt, in the 2017 and 2018 summer seasons. The experimental location is characterized by its hot and dry climate with zero precipitation during the summer growing season (from May to September). The mean temperature during the summer months of the two experimental seasons (2017 and 2018) is shown in Table 1. Temperature data were measured at the El Nouzha Airport meteorological station (31º12′ N, 29º57′ E; -2 m a.s.l.), Alexandria, Egypt. The soil of the experimental location is sandy loam, Fluvents, and composed of 55.5%, 29.0%, and 15.5% sand, silt, and clay, respectively. The pH was approximately 8.29, electrical conductivity 1.33 dS m^{-1}, and 8.10% $CaCO_3$. The nutrient composition of the top 25 cm of the soil was 2.04% organic matter and 100, 75, and 450 mg kg^{-1} available N, P, and K, respectively.

Table 1. Mean temperature accumulation prior to each cut for each sowing date of the 2017 and 2018 growing seasons.

		Mean temperature accumulation during 2017 (°C)		Mean temperature accumulation during 2018 (°C)	
Sowing date	Cut	Maximum	Minimum	Maximum	Minimum
15 May	1st	38.89	26.11	40.00	25.00
	2nd	38.89	27.22	38.89	27.22
	3rd	32.78	27.22	32.22	27.22
1 June	1st	38.89	36.11	40.00	25.00
	2nd	38.89	27.22	38.89	27.22
	3rd	32.78	27.22	32.22	27.22
15 June	1st	38.89	27.22	38.89	27.22
	2nd	32.78	27.22	32.22	27.22
	3rd	36.11	36.11	33.89	27.22

Design, treatments, and management

Variations in productivity, nutrient composition, in vitro nutrient degradability, and ruminal gas production of three successive cuts of four new pearl millet (*Pennisetum glaucum* [L.] R. Br.) genotypes (IP19586, IP19612, IP6105, and IP13150) compared with the local cv. Shandaweel-1, were studied on three different sowing dates (15 May, 1 June, and 15 June 2017 and 2018). The first cut occurred at 45 d after sowing (DAS), equivalent to the vegetative growth stage, and a 30-d interval was left between each of the two following cuts up to the third cut. Seeds of the four new genotypes under study were obtained from the International Crops Research Institute for the Semi-Arid Tropics (ICRISAT). A split-split plot experimental design with three replicates was adopted in which the main plots were the three sowing dates, sub-plots were the cuts, and the sub-subplots were the five tested pearl millet genotypes.

The experimental plot consisted of four ridges with 60 cm spacing for a total area of 3.0 × 2.4 m. Seeds were drilled in rows on two sides of the ridge with the 48 kg ha^{-1} forage seeding rate recommended by the Egyptian Ministry of Agriculture and Land Reclamation. Based on soil analysis and the current recommendations for pearl millet production in the area, 120 kg N ha^{-1} N fertilizer was applied as ammonium nitrate (33.5% N) divided in three equal doses; the first dose was applied as side-banded between the ridges at sowing, and the second and third doses were applied as top dressing directly after the first and second cuts, respectively. A single 200 kg ha^{-1} P dose was added at seedbed preparation as monocalcium phosphate (15.5% P_2O_5). Due to the zero precipitation in the summer seasons, agriculture in the region mainly depends on irrigation. Thus, all plots received equal amounts of water (applied as surface irrigation) at equal intervals to prevent induced drought stress. Manual weeding was used when necessary.

Sampling and analytical procedures

At forage harvesting, plots were cut with a sickle 7 cm aboveground and total fresh yield for each cut and plot was weighed. A 1 kg representative fresh matter sub-sample was taken from each experimental plot, put in paper bags, dried at 60 ºC until constant weight was reached (approximately 72 h), and dry matter (DM) content per plot was determined (g kg^{-1}). Dry matter yield (DMY) was estimated from the values of fresh yield and DM content (t ha^{-1}). Dried sub-samples were milled to a 1-mm particle size in preparation for the analytical procedure. The technique proposed by Van Soest et al. (1991) was used to sequentially determine neutral detergent fiber (NDF), acid detergent fiber (ADF), and acid detergent lignin (ADL) with a semiautomatic ANKOM^{220} fiber analyzer (ANKOM Technology, Macedon, New York, USA). Samples were burned at 550 ºC for 3 h (AOAC, 2012) in a muffle oven to determine the crude ash (CA) content. Organic matter (OM), ether extract (EE), and crude fiber (CF) contents were also determined as described by the AOAC (2012). When the N content was determined by the Kjeldahl procedure (AOAC, 2012), crude protein (CP) was calculated as N × 6.25.

The Hohenheim gas test proposed by Menke and Steingass (1988) was used to measure in vitro gas production (GP). Cumulative gas was expressed as milliliter of gas produced per 200 mg DM and corrected for blanks. Gas *(Y)* at time (*t*) was fitted to the exponential model of Orskov and McDonald (1979) as

$$Gas\ (Y) = a + b\ (1\text{-}exp^{-ct}),$$

where *a* is GP from the immediately soluble fraction, *b* is GP from the insoluble fraction, *c* is the GP rate constant for the insoluble fraction (*b*), and *t* is incubation time.

Metabolizable energy (ME), net energy of lactation (NEL), and OM digestibility (OMD) were calculated from the amount of gas produced after incubation for 24 h with the supplementary analysis of CP, CA, and EE expressed as g kg^{-1} DM (Menke and Steingass, 1988). Short chain fatty acids (SCFA) were calculated according to Getachew et al. (2002) after which total digestible nutrient (TDN) value was calculated from the ME value (NRC, 1989). Finally, microbial protein (MP) was calculated as 19.3 g microbial N kg^{-1} OM (Czerkawski, 1986). Calculations were

$$\text{ME (MJ kg}^{-1}\text{ DM)} = 2.20 + 0.1357 \times \text{GP} + 0.0057 \times \text{CP} + 0.000286 \times (\text{EE})^2,$$

$$\text{NEL (MJ kg}^{-1}\text{ DM)} = 0.0960 \times \text{GP} + 0.0038 \times \text{CP} + 0.000173 \times (\text{EE})^2 + 0.54,$$

$$\text{OMD (\%)} = 14.88 + 0.889\ \text{GP} + 0.45\ \text{CP} + 0.0651\ \text{CA},$$

$$\text{SCFA (mM)} = (-0.00425 + 0.0222 \times \text{GP}) \times 100\text{, and TDN (\%)} = (\text{ME (Mcal kg}^{-1}\text{ DM)} + 0.45)/0.0445309.$$

Statistical procedures

The ANOVA test was performed with the Proc GLM of SAS 9.4 (SAS Institute, Cary, North Carolina, USA) for the response variables of fresh forage yield, DMY, DM, NDF, ADF, ADL, CP, N-free extract (NFE), OMD, TDN, ME, NEL, GP, MP, and SCFA. After running the statistical analysis separately for the two experimental years, the homogeneity of variance error was determined according to Hartley's test (Winer et al., 1971); it was homogeneous and data were therefore presented in a combined analysis for the two years. The response variables (V) under study were analyzed according to the following model with only replicates considered random:

$$V_{ijkl} = \mu + R_i + SD_j + R_i \times SD_j + C_k + SD_j \times C_k + R_i \times SD_j \times C_k + G_l + SD_j \times G_l + C_k \times G_l + SD_j \times C_k \times G_l + e_{ijkl},$$

where μ is the overall mean, R_i is the replicate effect (i = 1, 2, 3), SD_j is the sowing date effect (j = 1, 2, 3), $R_i \times SD_j$ is the experimental error "a", C_k is the cut effect (k = 1, 2, 3), G_l is the genotype effect (l = 1, 2, 3, 4, 5), $R_i \times SD_j \times C_k$ is the experimental error "b", and e_{ijkl} is the experimental error "c".

Significance was declared at $P < 0.05$ and means were compared by the least significant difference (LSD) test.

RESULTS

Forage yield and nutrient composition

The ANOVA test revealed that fresh forage yield, DMY, DM, NDF, ADF, ADL, CP, and NFE were significantly affected by cut and genotype ($P < 0.01$), while only the DM content was significantly variable among sowing dates ($P < 0.05$). In addition, sowing date × cut (SD × C) significantly affected fresh forage yield, DMY, and DM ($P < 0.01$), whereas SD × genotype (SD × G) significantly affected CP and NFE and C × G significantly affected fiber fractions, CP, and NFE ($P < 0.01$). Only the lignin content was significantly affected by the three-way SD × C × G interaction ($P < 0.01$).

Regarding the significant variations among the five tested genotypes for fresh forage yield, the four genotypes IP19586, IP6105, IP13150, and 'Shandaweel-1' produced the highest significant values for fresh yield with 24.3, 22.0, 22.4, and 21.2 t ha^{-1}, respectively (Table 2). The lowest significant fresh forage yield was a character of IP19612 with18.1 t ha^{-1}. Despite the acceptable amount of fresh forage (21.2 t ha^{-1}) produced by the local genotype 'Shandaweel-1', it was characterized by the least significant amount of accumulated DM (149.9 g kg^{-1}). The highest significant DM accumulation was a character of IP19586, IP6105, and IP13150. Similar to fresh yield, DMY means (Table 2) revealed that genotype IP19586 produced the highest significant amount of DMY (3.82 t ha^{-1}), which was not significantly different from IP13150 (3.50 t ha^{-1}), while IP19612 was significantly lower with 2.74 t ha^{-1} DMY.

Table 2. Variations in fresh forage yield, dry matter content, and dry matter yield among genotypes combined for the 2017 and 2018 growing seasons.

Genotype	Fresh forage yield	Dry matter content	Dry matter yield
	t ha^{-1}	g kg^{-1}	t ha^{-1}
IP19586	24.3a	157.2a	3.82a
IP19612	18.1b	151.4b	2.74c
IP6105	22.0a	156.5a	3.44b
IP13150	22.4a	156.1a	3.50ab
Shandaweel-1	21.2a	149.9c	3.18b

Means followed by different lowercase letters in the same column for each studied parameter are significantly different according to the LSD test at 0.05 level of probability.

Sowing on 15 May and 1 June produced similar fresh forage yield from the three tested cuts, while sowing on 15 June significantly decreased the fresh forage yield of the second and third cuts (Table 3). The least significant yield resulted from the third cut when sowing was on 15 June (12.7 t ha^{-1}), while the highest significant yield was 25.6 t ha^{-1} for the first cut at early sowing (15 May). The combined effect of sowing date and cut on DM accumulation was different from its effect on forage yield (Table 3). When sowing was on 15 May or 1 June, the first cut accumulated the highest amount of DM with 173.2 and 178.0 g kg^{-1}, respectively, which gradually decreased until the third cut. On the contrary, with late sowing on 15 June, DM accumulation gradually increased from the first cut (144.1 g kg^{-1}) to the third cut (183.9 g kg^{-1}). Thus, the first and second cuts were superior to the third cut for DM accumulation with early and intermediate sowing, while the third cut was superior with late sowing. In addition to the variations in the direction of the response, the variable magnitude also contributed to the significant interaction. The decrease in DM accumulation from the first to third cuts reached 24.0% and 32.0% when sowing was on 15 May and 1 June, respectively. Meanwhile, DM accumulation increased by 27.6% from the first to the third cuts of millet sown on 15 June. The differences in the response of fresh yield and DM contents to the C × SD interaction were manifested on the DMY response (Table 3) in which the first cut produced the highest significant DMY with 4.44, 3.97, and 3.49 t ha^{-1} for the three sowing dates, respectively. Meanwhile, sowing on 15 May resulted in the highest significant DMY with values of 4.44, 3.05, and 3.07 t ha^{-1} for the first, second, and third cuts, respectively.

The significant variations in the nutritional components among the tested genotypes greatly depended on the cut. Means in Table 4 illustrate that the second cut was usually characterized by better quality than the first and third cuts in terms of lower NDF and ADF contents and higher CP content. The third cut was characterized by moderate NDF, ADF, and CP contents, while the values of the three nutritional components were significantly higher for the first cut. Mean NDF content was 661.7, 628.7, and 651.7 g kg^{-1} for the first, second, and third cuts, respectively, while mean ADF content was 345.4, 320.1, and 329.2 g kg^{-1} for the three cuts, respectively. On the contrary, mean CP content reached 103.0 g kg^{-1} for the second cut, which was approximately 53.24% and 10.83% higher than the mean CP contents of the first and third cuts, respectively. Table 4 clearly shows that the genotypes IP19612 and 'Shandaweel-1' were characterized by the highest significant fiber fractions with values of 660.8 and 670.9 g NDF kg^{-1} and 341.4 and 347.7 g ADF kg^{-1} as the mean of the three cuts for genotypes IP19612 and 'Shandaweel-1', respectively. Genotypes IP19612, 'Shandaweel-1', and IP6105 produced the lowest significant CP content with means of 85.3, 77.8, and 86.9 g kg^{-1}, respectively. Meanwhile, the highest significant CP content was a character of IP19586 and IP13150 with values of 94.6 and 93.9 g kg^{-1}, respectively.

Table 3. Effect of the Sowing date × Cut interaction on the variations in fresh forage yield, dry matter content, and dry matter yield combined for the 2017 and 2018 growing seasons.

	Fresh forage yield			Dry matter content			Dry matter yield		
Cut	15 May	1 June	15 June	15 May	1 June	15 June	15 May	1 June	15 June
	t ha^{-1}			g kg^{-1}			t ha^{-1}		
1st	25.61aA	22.31aA	24.23aA	173.2aA	178.0aA	144.1cB	4.44aA	3.97aAB	3.49aB
2nd	20.22aA	20.08aA	15.30bA	150.7bA	146.8bA	158.7bA	3.05bA	2.95bAB	2.43bB
3rd	23.31aA	18.80aAB	12.74bB	131.7cB	121.1cB	183.9aA	3.07bA	2.28bB	2.34bB

Means followed by different lowercase letters in the same column and/or different uppercase letters in the same row for each studied parameter are significantly different according to the LSD test at 0.05 level of probability.

Table 4. Effect of the Cut × Genotype interaction on the variations in neutral detergent fiber (NDF), acid detergent fiber (ADF), crude protein (CP), and N free extract (NFE) contents combined for the for 2017 and 2018 growing seasons.

	NDF			ADF			CP			NFE		
Genotype	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut
	g kg^{-1}			g kg^{-1}			g kg^{-1}			g kg^{-1}		
IP19586	644.7bA	619.3bB	644.0bA	338.3bA	305.2bC	313.1bB	70.7aC	113.3aA	99.7aB	523.8bA	480.8aB	469.5aB
IP19612	682.2aA	642.6aC	657.5bB	356.9aA	327.0aB	340.2aA	61.2bC	104.4bA	90.3bB	508.6cA	448.3bC	471.0aB
IP6105	647.2bA	623.8abB	646.0bA	333.6bA	325.4aAB	321.1bB	65.8bC	102.3bA	92.7bB	519.9bA	453.2bB	468.9aB
IP13150	646.7bA	616.0bC	627.6cB	339.1bA	311.8bB	318.2bB	73.3aC	110.0aA	98.5aB	543.8aA	473.7aB	474.0aB
Shandaweel-1	687.8aA	641.7aB	683.3aA	358.9aA	331.0aB	353.3aA	65.0bC	84.9cA	83.4cA	495.2cA	443.1bC	477.2aB

Means followed by different lowercase letters in the same column and/or different uppercase letters in the same row for each studied parameter are significantly different according to the LSD test at 0.05 level of probability.

Both genotypes were also characterized by their low NDF and ADF contents, highlighting their superior nutritional composition. The means for NFE illustrated that the first cut was usually characterized by the highest significant amount of NFE (518.3 g kg^{-1} mean) against means of 459.8 and 472.1 g kg^{-1} for the second and third cuts, respectively (Table 4). Genotype IP13150 produced the highest significant NFE for the first cut (543.8 g kg^{-1}); genotypes IP13150 and IP19586 were superior to the other genotypes for NFE in the second cut with 473.7 and 480.8 g kg^{-1}, respectively, while nonsignificant variations were detected among the tested genotypes for the third cut.

Both CP and NFE varied with the G × SD interaction (Table 5). Sowing on 15 May and/or 1 June produced the highest significant CP content, while delaying sowing until 15 June significantly reduced the CP content for most tested genotypes. The opposite was reported for the variations in NFE between sowing dates with late sowing on 15 June produced the highest significant NFE content for all genotypes. Similar to the previously reported results, genotype IP13150 was generally superior to the other tested genotypes regarding both the CP and NFE contents, while IP19612 and 'Shandaweel-1' were inferior for both nutritional components.

Among all the tested nutritive components, only the ADL content was significantly affected by the three-way interaction between the three studied factors (Table 6). At each sowing date and for all genotypes, the first cut was characterized by the lowest significant lignin content compared with the second and third cuts, except for the first cut of 'Shandaweel-1' sown on 1 June. Although the direction of the effects was consistent for ADL, remarkable shifts in the magnitude of the variation were observed, which contributed to the significant interaction. The increase in the ADL content from the first to third cuts was 37.74%, 6.33%, and 35.65% for the three sowing dates, respectively. Moreover, it was observed that the later the sowing, the higher the ADL content from each cut. When the genotype IP13150 was sown early on 15 May, it produced a significantly low lignin content compared with sowing on 1 and 15 June.

In vitro nutrient degradability and ruminal gas production

The ANOVA test revealed that all the studied parameters varied significantly among the tested genotypes, while significant variations between the cuts were only detected for energy measurements (ME and NEL), OMD, and TDN. However, the effect of the sowing date was only clear in the significant three-way interaction for OMD and TDN.

Table 5. Effect of the Sowing date × Genotype interaction on the variations in crude protein (CP) and nitrogen-free extract (NFE) contents combined for the 2017 and 2018 growing seasons.

	CP			NFE		
Genotype	15 May	1 June	15 June	15 May	1 June	15 June
	g kg^{-1}			g kg^{-1}		
IP19586	101.4aA	91.9bB	80.4aC	470.8bC	486.9aB	516.4aA
IP19612	77.3bB	93.8bA	72.6bB	479.4bB	475.3bB	493.1bA
IP6105	101.0aA	93.0bB	79.9aC	454.1cC	474.9bB	513.0aA
IP13150	94.5aA	100.8aA	79.5aB	492.3aB	490.8aB	518.4aA
Shandaweel-1	79.2bB	89.8bA	72.5bB	477.1bB	479.4bB	485.0bA

Means followed by different lowercase letters in the same column and/or different uppercase letters in the same row for each studied parameter are significantly different according to the LSD test at 0.05 level of probability.

Table 6. Effect of the Sowing date × Cut × Genotype interaction on the variations in acid detergent lignin (ADL) content combined for the 2017 and 2018 growing seasons.

	15 May			1 June			15 June		
Genotype	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut
	g kg^{-1}			g kg^{-1}			g kg^{-1}		
IP19586	33.7	44.6	41.6	35.9	44.9	44.5	41.5	49.6	55.2
IP19612	36.6	47.4	50.2	34.6	36.4	38.0	43.8	57.2	67.3
IP6105	30.0	40.7	52.9	36.8	35.1	39.0	46.5	66.1	67.4
IP13150	29.4	40.9	38.7	44.3	51.4	43.7	44.9	47.1	56.3
Shandaweel-1	37.5	44.9	46.9	47.3	47.2	36.3	46.6	46.1	56.7
$LSD_{0.05}$					3.0				

Tracking the variations in OMD among the tested genotypes as affected by the SD × C interaction revealed that an almost consistent response was achieved (Table 7). Only the superior genotype IP13150 had a significantly lower third cut OMD with early sowing (15 May), while the inferior genotype IP19612 had the lowest OMD values for the first cut when sown on 1 June. The highest OMD values for all cuts of all tested genotypes were usually achieved with sowing on 15 May with means of 418.7, 432.7, and 416.1 g kg^{-1} for the first, second, and third cuts, respectively. Meanwhile, late sowing on 15 June had higher OMD values for all cuts and genotypes than sowing on 1 June. Mean OMD values for all genotypes reached 319.9, 346.64, and 345.2 g kg^{-1} when sown on 1 June and 366.8, 389.2, and 384.4 g kg^{-1} when sown on 15 June for the three cuts, respectively.

Similarly, a trend was observed for TDN as affected by the three-way interaction (Table 8). The highest significant TDN value was 421.7 g kg^{-1} reported for the first cut of IP19612 sown on 15 May, while the lowest significant value was 321.7 g kg^{-1} for the second cut of 'Shandaweel-1' sown on 1 June, showing a maximum difference of 10% in TDN data. Nonsignificant variations between the three tested cuts were observed for the same sowing date, while variations between sowing dates were more pronounced. Similar to the OMD values, early sowing on 15 May resulted in the best TDN values for all cuts with means of 411.0, 406.8, and 397.4 g kg^{-1} for the first, second, and third cuts, respectively. This was followed by late sowing on 15 June with mean TDN values of 375.1, 381.9, and 378.5 g kg^{-1} for the three cuts, respectively. However, the lowest TDN values were produced when the genotypes were sown on 1 June with means of 329.5, 336.5, and 341.1 g kg^{-1} for the three cuts, respectively.

Means of energy measurements (ME and NEL) for the three cuts of the five evaluated genotypes are shown in Table 9. Genotype IP13150 produced the highest significant amount of ME with values of 5.683, 5.442, and 5.210 MJ kg^{-1} for the first, second, and third cuts, respectively. The other four tested genotypes produced lower ME values, and 'Shandaweel-1' was especially lower for the second and third cuts with 4.831 and 4.709 MJ ME kg^{-1}, respectively. Regarding the variations between cuts, an inconsistent direction was observed, which might have contributed to the significant interaction. The second and third cuts of IP19586 produced higher significant ME values than the first cut, whereas the first cut of IP13150 and 'Shandaweel-1' produced higher ME values than the second and third cuts of both genotypes. The variation in the three cuts was not significant for genotypes IP19612 and IP6105. Similarly, the NEL production by 'Shandaweel-1' was the least significant among all the genotypes with 2.248, 2.319, and 2.390 MJ NEL kg^{-1} for the three cuts, respectively. On the contrary, IP13150 was superior for the first cut (2.790 MJ kg^{-1}) and not significantly different from the other genotypes for the second and third cuts. Inconsistent variations between the three cuts for the five tested genotypes were reported, but variations had little biological impact.

Microbial protein (MP), GP, and SCFA only varied significantly among the five evaluated genotypes. Means displayed in Table 10 reveal the superiority of IP13150 and IP6105 for MP with values of 47.80 and 47.44 g kg^{-1} OM, respectively. Similarly, the two genotypes produced the highest significant amount of gas with 24.53 and 24.19 mL g^{-1}, respectively, while the lowest GP was reported for the local genotype Shandaweel-1 (19.53 mL g^{-1}). Means of SCFA of the five studied genotypes highlight the superiority of genotypes IP13150 and IP6105 with 47.4 and 46.6 mM, respectively, compared with 42.9 mM for 'Shandaweel-1'.

Table 7. Effect of the Sowing date × Cut × Genotype interaction on the variations in organic matter digestibility (OMD) combined for the 2017 and 2018 growing seasons.

	15 May			1 June			15 June		
Genotype	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut
		g kg^{-1}			g kg^{-1}			g kg^{-1}	
IP19586	420.8	412.3	447.8	320.2	346.2	360.8	325.5	369.3	379.1
IP19612	428.5	440.6	427.3	304.7	361.7	340.9	355.1	383.7	382.7
IP6105	405.9	432.7	410.6	330.2	359.1	342.7	361.6	411.3	411.3
IP13150	437.7	463.1	387.5	323.8	353.8	337.3	430.0	417.0	411.3
Shandaweel-1	400.7	414.8	407.5	320.6	312.4	344.3	361.8	364.5	337.8
LSD$_{0.05}$					57.0				

Table 8. Effect of the Sowing date × Cut × Genotype interaction on the variations in total digestible nutrients (TDN) combined for the 2017 and 2018 growing seasons.

	15 May			1 June			15 June		
Genotype	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut
	g kg^{-1}			g kg^{-1}			g kg^{-1}		
IP19586	414.4	427.1	419.6	327.5	340.8	353.5	336.0	364.0	371.4
IP19612	421.7	410.7	408.2	323.1	340.2	340.5	361.6	376.9	375.2
IP6105	403.5	407.0	383.7	334.8	347.0	342.4	370.5	400.5	400.1
IP13150	410.7	393.9	379.0	337.6	333.0	332.1	421.5	404.2	398.8
Shandaweel-1	404.9	395.5	396.7	324.3	321.7	337.2	385.8	363.8	347.1
$LSD_{0.05}$					76.0				

Table 9. Effect of the Cut × Genotype interaction on the variations in metabolizable energy (ME) and net energy of lactation (NEL) combined for the 2017 and 2018 growing seasons.

	ME			NEL		
Genotype	1st Cut	2nd Cut	3rd Cut	1st Cut	2nd Cut	3rd Cut
	MJ kg^{-1} DM			MJ kg^{-1} DM		
IP19586	4.812bB	5.147bA	5.026bA	2.586bA	2.622aA	2.678aA
IP19612	4.989bA	5.121bA	5.097bA	2.311cB	2.603aA	2.586aA
IP6105	5.003bA	5.287bA	5.112bA	2.521bB	2.721aA	2.597aB
IP13150	5.683aA	5.442aB	5.210aC	2.790aA	2.618aA	2.525aB
Shandaweel-1	5.042bA	4.831cB	4.709cB	2.248cB	2.319bA	2.390bA

Means followed by different lowercase letters in the same column and/or different uppercase letters in the same row for each studied parameter are significantly different according to the LSD test at 0.05 level of probability.

Table 10. Variations in microbial protein (MP), gas production (GP), and total short chain fatty acids (SCFA) among the five tested genotypes combined for the 2017 and 2018 growing seasons.

Genotype	MP	GP	SCFA
	g kg^{-1} OM	mL g^{-1} OM	mM
IP19586	47.0a	20.7b	45.5b
IP19612	43.9b	20.7b	45.6b
IP6105	47.4a	24.2a	46.6a
IP13150	47.8a	24.5a	47.4a
Shandaweel-1	44.2b	19.5b	42.9c

Means followed by different lowercase letters in the same column for each studied parameter are significantly different according to the LSD test at 0.05 level of probability.

DISCUSSION

Forage yield and nutrient composition

An accurate evaluation of the multiple cuts of new pearl millet genotypes compared with the local cultivar, as affected by the different sowing dates, would support their integration in the forage production sector in the Egyptian agricultural system.

Altering the sowing date had little effect on yield and DM content of the tested genotypes, and it highly depended on the cut. The highest significant yield was from the first cut of the early sown crop (15 May), while the least significant yield was from the third cut of the late sown crop (15 June). Abd El-Lattief (2011) pointed out that 15 May was the optimum sowing date in Upper Egypt for the best pearl millet productivity compared with earlier and later dates. Soler et al. (2008) suggested photoperiod and water availability as the main determining factors for the effect of sowing date on pearl millet biomass yield, especially in rainfed farming contexts. The present study depended on a homogeneous irrigation schedule for all the treatments, which eliminated the water availability factor and supported the assumption of the photoperiod effect. According to Craufurd and Bidinger (1988), the duration of the vegetative growth stage is a main determinant of pearl millet yield. Given that pearl millet is a short-day crop sensitive to the photoperiod, progress towards flowering is accelerated with decreased day length (Soler et al., 2008). Early sowing is therefore accompanied by a prolonged photoperiod, which favors the crop and allows for a more efficient use of assimilates; this leads to the production of leafier and taller plants (Maas et al., 2007), which will directly result in higher forage production. The decision as to the optimum sowing date for pearl millet is commonly based on atmospheric and soil temperatures. Lemus (2015) reported that the optimum growth of pearl millet occurs between 32.78 and 35.00 ºC and with the best sowing dates from May to June. They added that later sowing negatively affected forage yield and should only be considered for short-term grazing or emergency forage production. The negligible differences in mean temperature accumulation prior to each cut for the three tested sowing dates in the present study (Table 1) might explain the small yield variations in response to sowing dates.

The negative relationship between CP content and fiber fractions (NDF, ADF, and ADL) was in line with findings of other researchers (Du et al., 2016). It was reported that sowing on 15 May and 1 June produced higher-quality forage (high CP and low fiber fractions) than late sowing on 15 June. This concurred with results reported by Silungwe et al. (2011) for sorghum, Sudan grass, and pearl millet. Late sowing is usually accompanied with increased temperature, which accelerates stem growth over leaf growth; this reduces the leaf to stem ratio and increases cell thickness (Wilson et al., 1991). These morphological changes are usually accompanied by decreased CP content and increased fiber and lignin deposition (Silungwe et al., 2011). This also occurred in the present study in which the later the sowing date, the higher the lignin content produced from each cut.

Regarding the variations in DM accumulation as related to sowing date and cut, the first cut of the crop sown 15 May and 1 June accumulated higher DM content than later cuts. This occurred because the first cut was done after 45 DAS and had a longer period to accumulate DM content than later cuts. However, Radhouane (2008) reported a marked decrease in the number of days after sowing to maturity with delayed sowing of pearl millet in a similar Mediterranean environment. Accelerated growth accompanied by delayed sowing was probably the cause of the increase in DM accumulation of the third cut of the crop sown on 15 June and its very low yield. The abovementioned author therefore recommended sowing pearl millet from early May to early June in the Mediterranean region.

Unlike the present study, Machicek (2018) and Makarana et al. (2018) reported the highest quality for the first cut of pearl millet compared with subsequent cuts in terms of high CP and low fiber fractions (NDF and ADF). In the present study, the second cut was usually superior in quality than the first and third cuts. In partial agreement with our results, Salama and Zeid (2016) reported that the second cut of forage grasses produced a CP content that was as high as in the first cut, while the first cut had the lowest fiber fractions and the highest NFE. Analysis of the regrowth dynamics of pearl millet after cutting would explain the fluctuations in forage quality of the different cuts. Obeng et al. (2012) identified three tillering patterns in pearl millet, synchronous and non-synchronous tillering in which tillers arise from the basal leaf bud axils and sub-terminal tillering in which the tillers arise from the axillary buds. Since the first cut in the present study occurred at the vegetative stage (45 DAS), regrowth was initiated from non-basal (terminal and axillary) tillers and characterized by a denser tiller canopy than when the crop was cut at later growth stages (Stephenson and Posler, 1984). This was reflected in a higher leaf to stem ratio for the regrowth, leading to a high-quality second cut.

Pronounced variations were detected among the evaluated genotypes for yield, DM accumulation, and nutritional components, which might be attributed to the different genetic makeup of the individual genotypes which is affected by environmental conditions (Divya et al., 2017). In an attempt to explain the variations in the nutritional value among pearl millet genotypes, Hassan et al. (2014) referred these variations to the relative contribution of leaves to total biomass in the different genotypes. The present study indicated that the genotypes characterized by higher DM content had better nutritional values, in terms of high CP, NFE and low fiber components.

In forage research, DMY should be considered to efficiently select a proper genotype for incorporation in breeding programs because of its economic significance to the crop (Stida et al., 2018). In comparison with other forage grasses, and with millets grown in other farming systems, the tested genotypes produced acceptable amounts of DMY. The DMY production of the current genotypes was as high as that produced by the superior millet hybrids tested by Rai et al. (2012). They proved to be superior under Egyptian conditions than the millet genotypes investigated by Abd El-Lattief (2011) and Ziki et al. (2019). Atis and Akar (2018) reported DMY means of 2.64 and 2.45 t ha^{-1} for wheat cut at the vegetative growth stage; these values were less than those obtained from the millet genotypes in the present study. The tested genotypes contained better amounts of CP and NFE; however, NDF and ADF contents were comparable to sorghum and Sudan grass grown under similar farming contexts (Salama and Zeid, 2016). Although their CP content was comparable to maize cut at a similar age, they exhibited lower digestibility than maize (Salama, 2019), which might be attributed to the higher fiber content in pearl millet. Results of the nutritional performance of the tested pearl millet genotypes were also comparable to those obtained under similar arid/semiarid environments in Sudan (Babiker et al., 2015) and India (Makarana et al., 2018).

In vitro nutrient degradability and ruminal gas production

Chemical composition, in vitro measurements, digestibility, and energy estimates are the most commonly used tools when ranking different forage genotypes. A clear relationship between chemical composition and in vitro fermentation traits is well documented (Salama et al., 2020). To our knowledge, the chemical composition, in vitro gas production, digestibility, and energy values of the five evaluated pearl millet genotypes has not been previously investigated.

The chemical composition and degradability of feed are among the most important determinants for the amount of GP (Blümmel et al., 1999). Gas production indicates carbohydrate fermentation to the different SCFA, mainly acetate, butyrate, and propionate (Menke and Steingass, 1988); therefore, the amount of GP is mainly influenced by carbohydrate content in feed (Deaville and Givens, 2001), while the contribution of protein fermentation to GP is relatively small, although positive (Menke and Steingas, 1988). Our GP values after 24 h ranged from 24.53 to 19.53 mL 200 mg^{-1} DM for IP13150 and 'Shandaweel-1', respectively. Pal et al. (2015) reported a negative relationship between GP and structural carbohydrates (NDF and ADF contents), characterized by a slow fermentation rate by rumen microflora. Consequently, high GP values accompanied high SCFA production of genotype IP13150, which was also characterized by high CP and NFE values and low fiber values.

Fiber is a very important component in any feedstuff that supplies ruminants with the essential energy for production and maintenance and also preserves rumen health (Du et al., 2016). However, a negative relationship between fiber fractions (NDF, ADF, and ADL) and digestibility measures of forages and feedstuffs was reported by several researchers (Pal et al., 2015; Mokoboki et al., 2019); this was similar to the findings of the present study. Specifically, the increase in lignin as a non-degradable cell wall component strongly reduces the digestibility of any feedstuff (Du et al., 2016). Digestibility is an important indicator of the nutritive value of forage. The chemical composition of the feed is closely related to its digestibility and to the expected performance of the ruminant receiving it (Salama, 2019). Digestibility in our study was positively related to the CP content. In an attempt to quantify the contribution of each of the chemical components to determine OMD in maize fodder, Salama (2019) concluded that the variations in OMD mostly depended on the variations in the CP content ($r^2 = 0.8279$). This supports our findings that treatments characterized by higher CP and lower fiber values, such as early sowing (15 May) of the genotype IP13150, resulted in the highest OMD values compared with the other treatments. The tested pearl millet genotypes exhibited OMD values comparable to other forage grasses (Salama, 2019), although lower than forage legumes (Salama and Zeid, 2016). It is evident that roughages are generally deficient in fermentable carbohydrates, which results in relatively low OMD values. The tested genotypes in our study had relatively limited OMD values, probably due to their high NDF contents. Therefore, current breeding efforts are focused on improving forage grass digestibility, which includes increasing the water-soluble carbohydrate content against the cell wall contents, especially lignin, (Capstaff and Miller, 2018); this could also be adopted in pearl millet. Despite the limited digestibility of pearl millet, its acceptable DMY production in hot and dry climates of the arid/semiarid regions, compared with other forage grasses, generally gives it a considerable advantage.

Nitrogen-free extract (NFE) represents the fraction of easily fermentable dietary carbohydrates, including starches, sugars, and fructans in the present study. High NFE in the diet enhances ruminal NH_3-N concentration and reduces urine urea secretion (Lu et al., 2019), and facilitates the production of microbial metabolites, such as SCFA. In addition, NFE provides the rumen microflora with the energy required for MP synthesis. The superior genotype IP13150 in our study was characterized by the highest significant NFE, SCFA, and MP, confirming the positive relationship between the three parameters.

Energy is a limiting factor for growth, maintenance, and production of all living organisms. It substantially determines the milk and meat production levels in livestock production systems. The main chemical components that contribute to energy supply are carbohydrates, proteins, and lipids (Hall and Eastridge, 2014); energy values are known to respond positively to the CP content and negatively to fiber content (Mokoboki et al., 2019). This explains the higher predicted ME and NEL values reported for genotype IP13150, which was characterized by low fiber and high CP contents compared with the other tested genotypes. On the contrary, 'Shandaweel-1' was characterized by low CP and high fiber fractions, which resulted in low energy values. The energy values of the tested genotypes, despite being relatively low, were usually comparable to dry roughages (Garg et al., 2012) and browse species adapted to dry conditions (Mokoboki et al., 2019). Total digestible nutrients represent the usable energy content of any forage or feedstuff. Machicek (2018) stated that cutting multi-cut crops at 45 DAS resulted in higher TDN values for the first cut than later cuts; unlike these results, the 2nd cut in our study was characterized by higher TDN values than the other cuts. This was largely attributed to the previously explained high quality of the second cut in terms of lower amounts of fiber fractions (especially NDF and lignin) and higher CP content. The reported negative relationship between TDN and NDF and lignin for forages (Jayanegara et al., 2019) supports the results of the present study.

CONCLUSIONS

Studying the feed value in terms of in vitro nutrient degradability and ruminal GP provided a deep understanding of the quality of the different cuts for the five tested genotypes; and together with yield, they provided a better profile for the pearl millet genotypes. In the present study, in vitro gas production (GP), digestibility, and energy values of the genotypes greatly depended on their chemical composition. The genotype IP13150 maintained the desirable balance between productivity and quality. In addition to its high dry matter yield, it was characterized by the highest crude protein and N-free extract contents, yet the lowest fiber fractions. This was then reflected on its high organic matter digestibility, GP, and short chain fatty acid production, along with the highest energy values among all the genotypes. Altering the sowing date generally exerted a limited effect on the studied parameters, early sowing on 15 May was superior to later sowing on 1 June and 15 June. The second cut was superior to the first and third cuts taken from the genotypes for forage production; this highlights the importance and success of pearl millet as a multi-cut crop under environments similar to those of the present study.

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