Agronomic Performance and Hydrocyanic Acid Potential (HCN-p) of Single and Three-Way Sorghum-Forage Hybrids and DeKalb Hybrid SX-17[1]

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ABSTRACT

Eleven sorghum forage entries, including five single crosses, five three-way hybrids and DeKalb hybrid SX-17 were evaluated for yield of green forage (GF), yield of dry forage (DF), crude protein (CP) content, CP yield, height, leaf area, number of tillers, leaf-stem ratio and hydrocyanic acid potential (HCN-p) at Isabela, Puerto Rico. Four cuttings were made. The second harvest, 112 days after planting, had the highest DF yield, and the lowest CP content. When data for all harvests were combined, the single cross of AT×624 × Common sudangrass, three-way hybrid (AT×624 × BRhodesian) × Common sudangrass and single cross hybrid ARhodesian × Common sudangrass produced the highest DF yields. These were significantly higher than those of DeKalb hybrid SX-17. The highest DF yield was that from single cross hybrid AT×624 × Common sudangrass (17,303 kg/ha in 217 days), which exceeded by 11% and 13% the 2nd and 3rd highest producers. Based on orthogonal comparisons, single crosses were on the average superior to three-way hybrids in yield and most of the other characteristics studied. Three-way hybrids had a lower average HCN content (250 p/m) than single crosses (285 p/m). These values are slightly higher than the 200 p/m level at which HCN toxicity may begin to affect animals. Results indicate that any of the three top hybrids of this study would be an excellent choice for farmers desiring high forage production under irrigation in Puerto Rico.

INTRODUCTION

Few reports are available in Puerto Rico concerning the use of forage sorghums, in spite of their great potential for production throughout the year. In studies conducted in the Lajas Valley, Vázquez et al. (18) reported dry forage yields of over 35 tons per hectare with "Millo Blanco" (MB), a local forage-sorghum variety. Sotomayor-Ríos and Telek (14) obtained dry forage yields of 17 tons per hectare from hybrid CK-60 × MB, with an average crude protein content of 17.6% during a 211-day period at Isabela. On the basis of cuttings at a 30-day interval Sotomayor-Ríos and Santiago (16) reported dry forage yields of 20 tons per hectare in 140 days or 143 $kg \cdot ha^{-1} \cdot day^{-1}$, with an average crude protein content of about 14.3%. Morales (10) evaluated a series of hybrids of forage-sorghum and sorgo-sudan at Isabela, and reported yields of over 40 tons of dry forage/ha yearly when harvested at a 45-day interval.

Although forage sorghums are excellent in terms of high forage yields

[1] Manuscript submitted to Editorial Board November 2, 1981.

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over short life cycles, one of the objections to their use is the possibility of poisoning livestock with hydrocyanic (prussic) acid, which may be released during the digestive process from dhurrin present in the forage (4).

According to McBee and Miller (9) considerable genetic variation exists among sorghums in inherent levels of dhurrin or HCN-p. It has been found that N fertilization increases dhurrin levels (9) and that young regrowth is more likely to be toxic to animals than mature crops (4). Many other factors such as soil fertility, stress, genetic variation, age and location of tissue in the plant influence the level of HCN-p in sorghums (2, 4, 6). Levels of HCN tend to increase when growth is slowed by injury and lack of water (2, 6).

The literature concerning the use of three-way sorghum hybrids, either for forage or grain purposes, is very limited (11, 12). As early as 1969, Ross (12) stated that three-way hybrids could have a definite yield advantage over single crosses, but since then no additional information has become available to verify this statement. The development of three-way hybrids involves an additional generation, a factor which needs to be evaluated in economic terms in comparing them to single crosses. If three-way forage hybrids prove capable of producing yields superior to those of single crosses over several consecutive harvests in the tropics, their development might well be justified.

The objectives of this study were to compare a series of agronomic traits of five single and five three-way crosses and DeKalb hybrid SX-17 sorghums and to determine whether differences exist among them in terms of their HCN-p after four consecutive harvests in the tropics.

MATERIALS AND METHODS

In the summer of 1978, hand crosses were made among 2 male sterile (A,B) lines and 2 restorers. The female parents were AT×624 and ARhodesian sudangrass; the latter was developed from PI 156549 (*S. arundinaceum*) and Combine Kafir 60 (CK-60) by Craigmiles in Georgia (3). Males were Common and Greenleaf sudangrass from now on referred to as Common and Greenleaf. Single crosses were made as A×R, A×B and three-way crosses as (A×B)×R and (A×B)×B. In some cases crosses among Common sudangrass and the sterile lines resulted in sterile crosses. These instead were used in various combinations for the development of three-way hybrids. Sets of five single crosses, five three-way hybrids and DeKalb hybrid SX-17 were tested at the Isabela experiment farm of MITA, ARS-SEA-USDA, in northwestern Puerto Rico. The farm is situated 128 m above sea level, and ambient temperatures range from 18° to 31° C. The soil is a Coto clay (Oxisol) with a pH of about 5.8 (15). The 10 hybrids and DeKalb SX-17 were machine planted on July 26, 1979, with a population density of about 100,000 plants/ha. Immediately after planting, propazine[3] [2-chloro-4-6-bis-(isopropylamino)-s-triazine], was applied to all plots at 2 to 3 kg a.i./ha.

The experimental design was randomized complete blocks with four replications. Plots consisted of two rows spaced 101 cm apart and 6 m in length. Sampling area was 5 m^2. The first cutting was at 60 days after planting; subsequent cuttings were made approximately every 52 days thereafter. The experiment was discontinued after the 4th harvest. At planting, and after each cutting, a 15-5-10 fertilizer, equivalent to about 83 kg N/ha, was applied to all plots. Overhead irrigation was applied whenever necessary.

Before each cutting, plant height from the ground to the midpoint of the last leaf and number of tillers and leaf area were measured from 8 plants/plot chosen at random. Leaf area was determined by removing the second leaf from the top of the whorl, following the relationship maximum length × maximum width × 0.747 × number of leaves/plant based on the procedure developed by Stickler et al. (17). Green forage (GF) and dry forage (DF) yields were calculated for each harvest. Samples were analyzed for dry matter and crude protein (CP) contents and HCN-p at the MITA laboratories.

Leaf tissue was analyzed for HCN-p with the spectrophotometric method described by Gorz et al. (5) with modifications in sample preparation. The third leaf from the distal end of the plant was sampled prior to each cutting, according to the suggestion of Benson et al. (1). Samples were obtained at random over the length of the blade, excluding the midrib with an ordinary hole puncher. Fifteen discs 6 mm in diameter were obtained from each treatment and placed in tared 10 ml vials, and sealed. The sample was weighed and 5 ml of distilled water added to each vial. Samples were then autoclaved for 60 min at 37° C to achieve hydrolysis and extraction of dhurrin. After autoclaving, the vials were cooled and 1 ml of 0.1 *N* NaOH was added. Absorbance values were then determined at 330 nm, adjusting dilutions to register an absorbance in the range of 0.2 to 0.5.

To convert the absorbance readings to HCN-p values in p/m, the following equation was used:

$$p/m\ HCN = \frac{(A_{330})\ (DF)\ (VE)\ (27.03)}{(fr\ wt)\ (27.9)}$$

[3] Trade names in this publication are used only to provide specific information. Mention of a trade name does not constitute a warranty of equipment or materials by the Agricultural Experiment Station of the University of Puerto Rico, nor is this mention a statement of preference over other equipment or materials.

where

A_{330} is the absorbance of the base-diluted sample at 330 nm,
DF is the dilution factor if the sample had to be diluted,
VE is the volume (ml) of distilled water used to extract the tissue,
27.03 is the formula weight of HCN (μg/μmole), fr wt is the fresh weight (g) of the extracted tissue, and 27.9 is the extinction coefficient (ml/μmole) at 330 nm of p-HB in 0.1 *N* NaOH solution.

The method is nondestructive and does not involve enzymatic hydrolysis of dhurrin. The HCN-p values calculated in this way are on a fresh weight basis.

TABLE 1.—*Means and coefficient of variation (C.V.) in yields of green forage, dry forage, and crude protein; crude protein content; and height, leaf area, number of tillers, leaf/stem ratio and hydrocyanic acid potential in five single crosses, five three-way forage-sorghums and DeKalb hybrid SX-17 across four harvests at Isabela, Puerto Rico*

Characteristic	Mean and coefficient of variation (C.V.) at each harvest			
	Harvest 1	Harvest 2	Harvest 3	Harvest 4
Green forage yield (kg/ha)	24,189	30,668	17,429	18,625
C.V. (%)	8.67	9.33	11.96	14.66
Dry forage yield (kg/ha)	3,544	4,482	2,571	2,828
C.V. (%)	9.31	9.23	12.48	15.37
Crude protein (%)	14.12	11.29	12.18	12.40
C.V. (%)	10.75	8.95	10.02	5.89
Crude protein yield (kg/ha)	500	504	313	350
C.V. (%)	13.24	14.24	16.71	17.59
Height (cm)	215	259	181	184
C.V. (%)	2.72	5.84	5.03	3.91
Leaf area (cm^2)	5,552	6,855	3,775	4,016
C.V. (%)	7.03	13.12	8.63	9.00
Tillers, No.	1.81	3.38	5.99	6.48
C.V. (%)	14.53	14.58	13.38	13.40
Leaf/stem ratio	0.4246	0.4179	0.4260	0.4306
C.V. (%)	10.45	11.30	8.75	8.46
HCN-p (p/m)	268	280	245	260
C.V. (%)	8.32	11.63	9.86	14.21

Data were subjected to analysis of variance, orthogonal comparisons, and significant differences were identified with Duncan's multiple range test.

RESULTS AND DISCUSSION

Table 1 shows the means and coefficient of variation (C.V.) in GF, DF and CP yield; CP content, height, leaf area, leaf/stem ratio and HCN-p across the eleven hybrids at each harvest. The forage yields were usually highest at harvest 2. Yields of GF and DF in harvests 3 and 4 were very similar. In general, the highest plant height and leaf area were obtained in harvest 2, in agreement with the yield data.

Table 2 shows the mean GF, DF and CP yields, and the DM and CP contents of the eleven hybrids across the four harvests. The single cross hybrid of AT×624 × Common produced the highest GF yield, although this yield was not significantly different from those of (AT×624 × BRhodesian) × Common and ARhodesian × Common. The GF yields ranged from 114,300 to 70,304 kg/ha, the three-way hybrid (ARhodesian × (AT×624 × Greenleaf) having the lowest value.

The single cross hybrid of AT×624 × Common produced the highest DF yield with 17,304 kg/ha, although this value was not significantly different from those of (AT×624 × BRhodesian) × Common and (AT×624 × BRhodesian) × Greenleaf. The DF yields of the top producing hybrids are excellent, comparing well with those of some of the best perennial tropical grasses. At Isabela, Sotomayor-Ríos et al. (13) studied the effect of harvest intervals on 10 forage grasses. At the 30-day interval, the best yielder, Transvala digitgrass (*Digitaria decumbens* Stent), produced about 29,000 kg/ha/year, whereas in the present study AT×624 × Common sudangrass produced almost 60% of that yield in only 217 days. Its daily DF yield was about 80 $kg \cdot ha.^{-1}$.

The three-way hybrid (AT×624 × BRhodesian) × Greenleaf had the highest DM content (16.77%), which was significantly superior to the remaining hybrids. The three-way hybrid (AT×624 × Common) × BT×624 exhibited the lowest DM content. Crude protein contents ranged from 13.06 to 11.68% and no significant differences were observed among the 11 hybrids. Similar results were obtained by Sotomayor and Santiago (16) when nine sorghum hybrids were evaluated at Isabela. The CP yields of the 11 hybrids ranged from 2,004 to 1,360 kg/ha; the single cross hybrid of AT×624 × Common was highest. This yield was not significantly different from those of (AT×624 × BRhodesian) × Common, ARhodesian × Common, AT×624 × Greenleaf and (AT×624 × BRhodesian) × Greenleaf.

Table 3 shows the mean height, leaf area, number of tillers, leaf/stem ratio and HCN-p values of the eleven hybrids across the four harvests. The single cross hybrid AT×624 × Common was the tallest, although not significantly different from the remaining hybrids except (AT×624 × Common) × BT×624, AT×624 × BRhodesian, and ARhodesian × (AT×624 × Greenleaf).

The three-way hybrid (AT×624 × BRhodesian) × Common and the single cross hybrids AT×624 × Common and AT×624 × Greenleaf had

TABLE 2.—*Mean yields of green, dry forage and crude protein; dry matter and crude protein contents in five single crosses and five three-way forage-sorghums and DeKalb hybrid SX-17 across four harvests at Isabela, Puerto Rico*[1]

Hybrid	Green forage yield	Dry forage yield	Dry matter	Crude protein yield	Crude protein
	kg/ha	*kg/ha*	%	*kg/ha*	%
AT×624 × Common	114300 a	17303 a	15.13 bc	2044 a	11.81 a
(AT×624 × BRhodesian) × Common	109200 ab	15377 ab	14.08 bc	1924 abc	12.51 a
ARhodesian × Common	101300 abc	14662 bc	14.47 abcd	1834 abcd	12.51 a
AT×624 × Greenleaf	97972 bcd	14730 bc	15.03 bc	1722 abcde	11.68 a
(AT×624 × BRhodesian) × Greenleaf	93348 cde	15657 ab	16.77 a	1954 ab	12.47 a
DeKalb Hybrid SX-17	89400 cde	12624 cd	14.12 bc	1553 cde	12.29 a
AT×624 × (ARhodesian × Greenleaf)	84248 def	12408 d	14.72 bc	1620 bcde	13.06 a
ARhodesian × BT×624	81248 ef	11190 d	14.65 bc	1503 de	12.61 a
AT×624 × BRhodesian	79824 ef	11480 d	14.38 bc	1389 e	12.10 a
(AT×624 × Common) × BT×624	78272 f	10705 d	13.67c	1432 e	13.37 a
ARhodesian × (AT×624 × Greenleaf)	70304 f	10823 d	15.30 b	1360 e	12.56 a
$\bar{X}$	90856	13425	14.76	1667	12.45
$\bar{X}$ Single Crosses	94299	14016	14.73	1698	12.14
$\bar{X}$ Three-way Hybrids	87074	12994	14.92	1658	12.79
C.V. (%)	10.82	11.26	6.27	15.20	9.26

[1] Data are means across four replicate plots and four harvests. In each column, means followed by one or more letters in common do not differ significantly (P = .05), according to Duncan's multiple range test.

TABLE 3.—*Mean height, leaf area, number of tillers, leaf/stem ratio and hydrocyanic acid potential (HCN-p) in five single crosses and five three-way forage-sorghums and DeKalb hybrid SX-17 across four harvests at Isabela, P.R.*[1]

Hybrid	Height	Leaf area	Tillers	Leaf/stem ratio	HCN-p
	cm	*cm*^{2}	*No.*		*p/m*
(AT×624 × BRhodesian) × Common	221 a	7096 a	4.73 abc	0.3360 efg	269 cd
AT×624 × Common	220 a	6737 a	4.60 abc	0.5393 b	253 de
AT×624 × Greenleaf	220 a	6647 a	5.06 ab	0.5074 bc	340 a
ARhodesian × Common	218 a	5261 b	5.37 a	0.3802 de	320 ab
DeKalb Hybrid SX-17	215 ab	5166 b	4.78 abc	0.4893 c	223 e
AT×624 × (ARhodesian × Greenleaf)	212 ab	3109 e	5.54 a	0.3588 def	219 e
(AT×624 × BRhodesian) × Greenleaf	209 ab	3978 d	4.07 bcd	0.4034 d	208 e
ARhodesian × BT×624	206 ab	4854 bc	4.25 bcd	0.3717 de	287 bcd
(AT×624 × Common) × BT×624	202 b	5593 b	2.96 e	0.6913 a	247 de
AT×624 × BRhodesian	200 b	4141 cd	3.39 de	0.3022 fg	224 e
ARhodesian × (AT×624 × Greenleaf)	180 c	2875 e	3.82 cde	0.2927 g	307 abc
$\bar{X}$	209	5041	4.42	0.4247	263
$\bar{X}$ Single Crosses	213	5528	4.53	0.4201	285
$\bar{X}$ Three-way Hybrids	205	4530	4.22	0.4164	250
C.V. (%)	4.75	10.82	14.80	9.79	11.24

[1] Data are means across four replicate plots and four harvests. In each columns, means followed by one or more letters in common do not differ significantly (P = .05), according to Duncan's multiple range test.

the greatest leaf areas. Their values were significantly different from those of the remaining hybrids including DeKalb hybrid SX-17, which had about 37% less leaf area than the top hybrid, (AT×624 × BRhodesian) × Common. These results correspond to those of Liang et al. (8), who utilized the same procedure of Stickler et al. (17). Leaf area, along with plant height and number of tillers/plant, is an additional characteristic of importance influencing yield.

Three-way hybrid AT×624 × (ARhodesian × Greenleaf) and single cross hybrid ARhodesian × Common had the highest number of tillers, although not significantly superior to most of the remaining hybrids. The coefficient of variation associated with number of tillers was large in this experiment and may explain the nonsignificant difference.

A high leaf/stem ratio indicates good forage quality. Sorghum genotypes having a larger proportion of leaves are more nutritious and will produce better hay than those with a low ratio. The three-way hybrid (AT×624 × Common) × BT×624 had the highest leaf/stem ratio (0.6913), significantly different from the remaining hybrids. The single cross hybrids, AT×624 × Common and AT×624 × Greenleaf, were the 2nd and 3rd best in terms of their leaf/stem ratios. These two hybrids were also excellent DF producers.

The potential HCN content of all the hybrids studied exceeded the "threshold of danger" of 200 p/m (7) at each harvest. The three-way hybrid (AT×624 × BRhodesian) × Greenleaf exhibited the lowest HCN content, while AT×624 × Greenleaf had the highest (340 p/m). The value cited is an approximation, representing the concentration of HCN at which it may become toxic to ruminant animals (7, 9).

Wolf and Washko (19) studied the distribution and concentration of HCN-p in several parts of a sorghum-sudangrass hybrid at three growth stages: 50-, 120-, and 155 cm height. Leaf blade minus midrib was high in HCN at all growth stages. Midrib, sheath, and stem portions decreased in HCN concentration with maturity. These authors attributed the decrease of HCN potential of the entire plant with age to the proportional increase in weight of the low-HCN parts. Our best producing hybrids, even those having HCN values exceeding the "threshold of danger" of 200 p/m in the leaves, can be recommended for green-chop feeding of animals, where the entire plant is consumed. According to the findings of Wolf and Washko (19), the animals fed green chop will be consuming a high proportion of low-HCN plant parts such as the stem, sheath and leaf midrib.

The present results could be of value in the selection of low-HCN cultivars; however, extensive research is needed to determine the "threshold of danger" applicable to the tropics.

Table 4 shows the statistical significance of the orthogonal comparisons

TABLE 4.—*Statistical significance (F values) or orthogonal comparisons for 11 hybrids (five single crosses versus five three-way hybrids and five single crosses and five three-way hybrids versus DeKalb hybrid SX-17) across four harvests at Isabela, P.R.*

Comparison	Green forage yield	Dry forage yield	Crude protein yield	Height	Leaf area	No. of tillers	Leaf/stem ratio	HCN-p
Single cross versus three-way hybrids	104.40**[1]	73.13**	4.09*[2]	110.00**	538.44**	35.71**	1.11	219.19**
Single cross and three-way hybrids versus DeKalb hybrid SX-17	2.65	19.73**	14.24**	25.50**	3.73	21.80**	17.29**	129.97**

[1] (p 0.01).

[2] (p 0.05).

and means for the 11 hybrids (single cross versus three-way hybrids and single cross + three-way hybrids versus DeKalb hybrid SX-17). On the average, single crosses were superior to three-way hybrids in terms of GF, DF and CP yields, height, leaf area, and number of tillers. The three-way hybrids averaged higher than the single crosses in HCN content. The single cross and three-way hybrids were superior to DeKalb hybrid SX-17 in terms of DF and CP yields. DeKalb hybrid SC-17 exceeded the average combination of single cross hybrids and three-way hybrids in terms of plant height, leaf area, and HCN content.

The results indicate that these genotypes offer an excellent opportunity to farmers, interested in the utilization of forage sorghum hybrids under irrigation in Puerto Rico. The high DF yields and rapid regrowth of these hybrids, plus the possible availability of seed, could be decisive factors in more extensive utilization of forage sorghums in the beef and dairy industries in Puerto Rico.

RESUMEN

Cinco híbridos sencillos, cinco híbridos triples y el híbrido comercial DeKalb SX-17 se evaluaron en términos de forraje verde (GF), materia seca (DM), proteína bruta (CP), altura, área foliar, número de vástagos, razón hoja-tallo y potencial de ácido prúsico (HCN-p) durante 217 días en Isabela, Puerto Rico. Se llevaron a cabo 4 cortes, el primero a los 60 días y los siguientes cada 52 días, aproximadamente. Se utilizaron las yerbas sudán "Greenleaf" y "Common" como progenitores masculinos y las líneas androesterériles "AT×624" y "ARhodesian sudangrass" como progenitores femeninos. En adición, se utilizaron las líneas "BT×624" y "BRhodesian sudangrass" como mantenedoras de las respectivas líneas A. El segundo corte registró la mayor producción de forraje seco (DF) y el menor contenido de CP. Al combinar los cuatro cortes, el híbrido sencillo AT×624 × Common, el híbrido triple (AT×624 ×BRhodesian) × Common y el híbrido sencillo ARhodesian × Common fueron los mayores productores en términos de DF, resultando significativamente superiores al híbrido comercial DeKalb SX-17. La producción de DF del híbrido sencillo AT×624 × Common (17,303 kg/ha en 217 días) fue superior en 11 y 13%, respectivamente, con relación a los híbridos que resultaron segundo y tercero en producción de DF. A base de las comparaciones ortogonales, los híbridos sencillos fueron en promedio superiores a los híbridos triples en producción y otras características estudiadas. Los híbridos triples registraron en promedio menos HCN-p (250 ppm) al compararse con los híbridos sencillos (285 ppm). Estos valores son ligeramente superiores al nivel de 200 ppm, nivel sobre el cual el HCN-p puede ser tóxico para el ganado. Los resultados indican, que los híbridos que registraron los rendimientos más altos tienen magníficas potencialidades para usarse como forrajeras en la Isla.

La producción de DF del híbrido sencillo AT×624 × Common de 17,303 kg/ha en un período de 217 días o de 80 kg·ha^{-1} $día^{-1}$ con una CP de 12% es excelente, especialmente cuando la yerba se puede cortar en los trópicos a intervalos cortos.

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