

Growth Indicators of a 48-Clone Sugar Cane Population (*Saccharum* spp.) with Forage Potential

Yoslen Fernández Gálvez¹, Redimio M. Pedraza Olivera², Yusvel Hermida Baños³, Ailsa Llanes Díaz⁴, Isabel C. Torres Varela⁵, Joaquín Montalván Delgado⁶ & Arlandy Noy Perera⁷

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ABSTRACT

The aim of this paper was to determine growth indicators in a 48-clone sugar cane population, with promising phenotypical features for forage production. The following indicators were assessed: leaf area (A), leaf area index (LA1); leaf area ratio (LAR); specific leaf area (SLA); leaf weight ratio (LWR); crop growth rate (CGR); net assimilation rate (NAR); relative growth rate in weight (RGR); biomass production speed (G); leaf area duration (LAD); and biomass duration (Z), monthly (187 - 370 days). The minimum, the mean, the maximum values, and the population variance were determined for all cutting ages and the variables assessed. The results achieved have provided quantitative values that can be used as reference for selection and assessment of forage genotypes for ruminant nutrition.

Key words/sugar cane, *Saccharum* spp., growth indicators, clones, forage potential

One of the most important factors limiting ruminant production in Cuba and other countries, is associated with poor pasture availability (quality and quantity), to feed animals, which is closely related to gradual increases of temperatures, and longer dry periods, caused by climatic change. Accordingly, there a need for new and sustainable alternatives to mitigate the negative impacts.

¹ Agronomy engineer Ing. Agronomist, Associate researcher, Territorial Station for Sugar Cane Research Central-Eastern Cuba (ETICA), Camaguey, Cuba: yoslen@eticacm.azcuba.cu

² Full researcher, Professor, Center for Animal Production Development Studies (CEDEPA) PhD, Ignacio Agramonte Loynaz University of Camaguey: redimio.pedraza@reduc.edu.cu

³ Specialist in sugar cane production, Territorial Sugar Cane Research Station (ETICA) central and eastern Cuba, based in Camaguey, Cuba: yusvel@eticacm.azcuba.cu

⁴ Specialist in science and technology, Territorial Sugar Cane Research Station (ETICA) central and eastern Cuba, based in Camaguey, Cuba: ailsa@eticacm.azcuba.cu

⁵ Associate researcher, Territorial Station for Sugar Cane Research Central-Eastern Cuba (ETICA), Camaguey, Cuba: itorres@eticacm.azcuba.cu

⁶ Associate researcher, Territorial Station for Sugar Cane Research Central-Eastern Cuba (ETICA), Camaguey, Cuba: jmontalvan@eticacm.azcuba.cu

⁷ Director, Territorial Station for Sugar Cane Research Central-Eastern Cuba (ETICA), Camaguey, Cuba: a.noy@eticacm.azcuba.cu

The inclusion of forage-producing plants that can withstand adverse conditions was one of the main actions weighed. Sugar cane has anatomical and physiological features that make it more suitable than other crops, classifying as a plant able to mitigate the effects of grass unavailability, particularly in the dry season (Fernández *et al.*, 2014).

Several studies made in Cuba (Franco, 1981; Milanés *et al.*, 1997; Molina and Lazo, 1998; Stuart, 2002; Suárez, 2006; Leyva, 2012), to determine sugar cane commercial cultivars for animal nutrition were supported by over a dozen of the main cattle raising areas of the country. However, it is important to note that the genotypes assessed and recommended were chosen based on sugar production criteria.

Today, there is a need to include genotypes that provide high nutritional values and ruminant acceptability, along with adaptation to the most adverse conditions posed by climatic change.

Lately, specific studies aimed at elucidating growth and development issues of sugar cane have been left out (Valladares *et al.*, 2015). Recently, they published a paper whose goal was to compare production and distribution of dry biomass in different parts of the plant, to provide basic knowledge of growth and more efficient use of commercial cultivars, and others, in advanced stages of development. For their part, Freire *et al.* (2010) evaluated growth indexes of 11 varieties of sugar cane with irrigation, in Brazil.

Sugar cane growth is produced by means of crop interaction with environmental factors. Thorough understanding of such interactions can be acquired through quantitative analysis of growth, and biometric measurements of the plants during their development. In turn, physiological indexes become useful tools to confirm the differences among varieties, and model growth under several environmental conditions of production and management (Keating *et al.*, 1999; Machado *et al.* 1982, cited by Freire *et al.*, 2010).

Accordingly, the goal of this paper was to determine the growth indicators of a 48-clone sugar cane population, with promising phenotypic traits to produce forage. They could also be used as reference for selection and evaluation of genotypes with forage potential for ruminant nutrition.

MATERIALS AND METHODS

This study was conducted at the Territorial Station for Sugar Cane Research, Central-Eastern Cuba, in Camaguey, municipality of Florida, 21°.31' north latitude, and 78°.04' west longitude, 57.08 meters above sea level.

The field experiment was developed on brown soils with carbonates (Hernández *et al.*, 1999). The area pH was slightly acidic (6.1 - 6.3), whereas the content of organic matter (up to 20 cm) was average (3.1 - 3.5 %). Effective depth is 45 cm.

The weather variables (Table 1) were collected at the Agricultural weather station, Florida, 250 m from the experimental area.

Table 1 Weather variables during the study

Year	Months	Stump	TMP (°C)	Rain (mm)	RD	Year	Months	Stump	TMP (°C)	Rain (mm)	RD
2007	Feb.	Plant	23.5	40.0	8	2008	Mar.	Soca	24.6	8.1	3
	Mar.		23.9	93.5	8		Apr. Abr.		24.9	30.0	5
	Apr. Abr.		25.2	0.2	1		May. May.		26.4	162.9	8
	May. May.		25.2	446.6	20		Jun.		27.3	90.8	6
	Jun.		26.8	188.4	17		Jul.		27.8	46.8	6
	Jul.		27.3	190.6	13		Aug. Ago.		27.3	141.9	12
	Aug. Ago.		27.3	212.0	12		Sep.		27.0	473.6	16
	Sep.		26.4	194.8	14		Oct.		26.1	68.0	5
	Oct.		25.8	363.3	20		Nov.		23.5	101.8	9
	Nov.		24.0	3.4	1		Dec.		22.8	55.0	10
2008	Dec.	Plant	23.7	51.9	5	2009	Jan.	Soca	22.2	8.0	2
	Jan.		22.9	0.9	1		Feb.		22.0	4.1	2
	Feb.		24.7	12.1	4						
TMP- Average mean temperature Rain.-Rainfall RD-Rain days											

A sugar cane population made of 48 clones selected from 106 clones in different stages of selection were assessed. All the clones had favorable phenotypical characteristics to produce forage. After a series of assessments based on qualitative criteria, 58 clones were discarded. The remaining clones had previously shown forage potential for ruminant nutrition.

Planting took place in February 2007, and cultural labor was performed according to the Technical Instructions for sugar cane in Cuba (MINAZ-INICA, 2007). Establishment cutting was made 12 months later, in February 2008. The experiment was made in lands with no irrigation.

It was designed in three random blocks, with three replicas. The area of each experimental unit was 36 m² (7.5 x 4.8 m), three furrows of 7.5 meters long per clone. Cultivar C86-12 was used as borderline in the experiment.

In August 2008, on day 187 (≈6 months) following the establishment cutting, three samples were taken per replica. A sample was considered a whole stem with its leaves, pods, and buds, whose growth indicator were determined, including, leaf area (A), leaf area index (LAI), leaf area rate (LAR), Specific leaf area (SLA), leaf weight ratio (LWR), crop growth rate (CGR), net assimilation rate (NAR), relative growth rate in weight (RGR), biomass production speed(G), leaf area duration (LAD), and biomass duration (Z) of the 48 clones.

Assessment was made at monthly intervals, ending in February 2009, 370 days (\approx 12 months) later. Seven assessment analyses were performed.

Growth and development were determined according to Torres (2006), using the following equations:

Leaf area: (A)

To know the total leaf area per individual, length and width of each active sheet were measured. Each sheet area was calculated by multiplying length by width by factor 0.7 (Lerch *et al.*, 1977). The sum of all the values represents each individual's leaf area, in dm^2 .

$$A = \text{ANLB} \times \text{LGLB} \times 0.7$$

ANLB = limb width, taken from the widest portion of the sheet, in cm (precision of up to 1 mm).

LGLB = limb length, from the apex to the pod insertion, in cm (precision of up to 1 mm).

Biomass production speed: (G)

It was calculated as the ration between the differences of total dry weight of an average individual and the time between the two sample collections.

$$G = (W2 - W1) / (t2 - t1) \text{ (g. day}^{-1}\text{)}$$

W1 = total dry mass of the plant, in time 1. (g)

W2 = total dry mass of the plant, in time 2. (g).

$t2 - t1$ = time interval elapsed between the two assessments (days).

Crop growth rate: (CGR)

The difference of the total dry mass of an average individual between two consecutive sample collections was divided by the land area and the time between samplings.

$$\text{CGR} = (W2 - W1) / \text{At} (t2 - t1) \text{ (g. m}^{-2} \text{ day}^{-1}\text{)}$$

W1 = total dry mass of the plant, in time 1. (g).

W2 = total dry mass of the plant, in time 2. (g).

$t2 - t1$ = time interval elapsed between the two sample collections (days).

At = Land area (m^2).

Leaf area ratio: (LAR)

Ratio between leaf area and total dry weight of an average individual

$$\text{LAR} = A / W \text{ (cm}^2 \cdot \text{g plant}^{-1}\text{)}.$$

A = Leaf area with more than 50% of active leaf sheet (cm²).

W = Total dry mass of the plant.

Determination of specific leaf area: (SLA)

Ratio between leaf area and dry mass of the surfaces.

SLA = A / Wh (cm².g leaf⁻¹).

A = Leaf area of an average individual, with more than 50% active leaf surface (cm²).

Wh = Dry mass of an individual's surfaces (g).

Determination of leaf weight ratio: (LWR)

Ratio between the surface's dry mass and the total dry mass of an average individual.

LWR = Wh / W (g leaf .g plant⁻¹)

Wh =Total dry mass of an average individual's surface (g leaves).

W = Total dry mass of an individual (g plants).

Net assimilation rate: (NAR)

The double of the difference of the total dry mass of an average individual was divided between the result of the sum of leaf areas multiplied by the time difference between sample collections.

NAR = 2 x (W2- W1) / (A1 + A2) (t2 – t1) (mg plant.cm⁻².day⁻¹).

W1 = Total dry mass of an average individual in time 1 (mg).

W2 = Total dry mass of an average individual in time 2 (mg).

A1 = Leaf area with over 50% active surface in time 1 (cm²).

A2 = Leaf area with over 50% active surface in time 2 (cm²).

t2 - t1 = Time interval during assessment (days).

Relative growth rate in weight: (RGR)

It was calculated by multiplying the net assimilation rate by the leaf area ratio.

RGR = NAR x LAR (mg. g⁻¹.day⁻¹).

NAR = Net assimilation rate (mg .cm². day⁻¹)

LAR = Leaf area ratio (cm²/g).

Determination of leaf area index: (LAI)

It was calculated by dividing the leaf area expressed in m² by the land area.

LAI = A / At (m².m⁻²).

A = Leaf area with more than 50% of active leaf surface (cm²).

At = Land area (m²).

Leaf area duration: (LAD)

It was calculated by dividing the leaf area by the double of the difference of time mediating sample collections.

$$LAD = [(A_2 + A_1) / 2] \times (t_2 - t_1) \text{ (cm}^2\text{.day)}$$

A₁ = Leaf area with over 50% active surface in time 1. (cm²).

A₂ = Leaf area with over 50% active surface in time 2. (cm²).

t₂ - t₁ = Time interval during consecutive sample collections (days).

Biomass duration: (Z)

It was calculated as the product of the semi-sum of total dry weight of an average individual by the time difference mediating between sample collections.

$$Z = [(W_2 + W_1) / 2] \times (t_2 - t_1) \text{ (g. day}^{-1}\text{)}$$

W₁ = Total dry mass of an average individual in time 1 (g)

W₂ = Total dry mass of an average individual in time 2 (g)

t₂ - t₁ = Time interval during consecutive sample collections (days).

A database with all assessment information was made. The 48 clones were processed as a population. The minimum, the mean, the maximum values, and variance were determined during each cutting age evaluated. SPSS for Windows, 15.1 (2006) was used for statistical analysis.

RESULTS AND DISCUSSION

The behavior of growth indicators for a population of 48 clones of *Saccharum spp.* with forage potential in the stump of soca at different cutting ages (Table 2) is presented below,

The leaf area (A) and leaf area index (LAI), had a very similar behavior, increasing from day 187 to 248 days of cutting age. Then, they decreased until day 309, with a slight increase on day 399, to finally drop in the last assessment.

Table 2. Behavior of growth indicators in a population of 48 clones of *Saccharum spp.* with forage potential in the soca stump, at different cutting ages.

Ages	Statistical parameters	Growth indicators										
		A	LAI	LAR	SLA	LWR	CGR	NAR	RGR	G	LAD	
187 days	Min.	19.85	0.60	10.28	33.37	0.21						
	Max.	49.92	2.73	21.24	85.55	0.35						
	μ	35.85	1.61	16.30	58.86	0.28						
	σ^2	11.89	0.61	3.37	14.78	0.04						
218 days	Min.	29.45	1.18	8.77	41.33	0.15	-11.12	0.24	3.86	1.28	7.89	6.55
	Max.	71.16	5.69	20.15	137.41	0.26	91.89	2.20	19.28	8.74	18.14	12.45
	μ	51.70	3.13	14.36	67.55	0.22	37.63	0.87	11.75	4.53	13.57	9.11
	σ^2	16.62	1.29	2.62	20.72	0.03	28.11	0.48	4.82	2.45	2.67	1.69
248 days	Min.	38.44	2.32	9.03	48.16	0.14	-16.35	0.11	1.20	0.36	10.42	8.55
	Max.	79.47	7.15	16.53	85.40	0.25	110.04	1.14	13.01	5.81	21.28	17.55
	μ	52.93	4.12	11.93	64.71	0.19	44.30	0.61	7.16	2.96	15.69	12.25
	σ^2	16.12	1.61	2.19	9.63	0.04	36.01	0.33	3.68	1.70	3.01	2.65
278 278 days	Min.	24.34	1.94	4.80	28.51	0.11	-48.77	0.15	0.99	0.50	9.42	10.87
	Max.	68.39	9.54	9.61	73.81	0.21	241.39	2.29	18.82	12.47	21.91	21.11
	μ	41.13	3.84	7.24	52.37	0.14	56.69	1.09	7.55	3.99	14.11	15.38
	σ^2	15.17	1.95	1.53	12.70	0.03	59.12	0.67	4.54	2.92	3.36	3.27
309 days	Min.	20.79	1.47	3.32	28.93	0.06	-30.81	0.01	0.05	0.02	7.32	12.08
	Max.	44.55	5.65	7.55	70.98	0.18	122.79	1.13	5.52	3.10	17.23	26.29
	μ	30.99	3.04	5.15	43.26	0.12	23.63	0.45	2.12	1.34	11.18	18.39
	σ^2	9.97	1.34	1.18	11.36	0.03	37.59	0.36	1.57	0.95	2.81	4.13
339 days	Min.	22.13	1.77	3.51	24.35	0.09	-119.91	-	-11.38	-5.82	7.10	13.97
	Max.	46.63	4.46	7.86	58.71	0.16	123.53	2.83	9.91	6.69	13.01	27.55
	μ	32.19	3.30	4.91	41.97	0.12	30.36	0.69	2.84	1.97	9.48	19.31
	σ^2	9.19	0.96	1.13	8.47	0.02	67.03	1.28	6.06	3.33	1.73	4.02
370 days	Min.	16.13	1.45	2.82	22.15	0.09	-90.15	-	-10.88	-3.77	5.93	14.54
	Max.	38.17	4.93	5.43	43.20	0.16	59.89	0.56	1.95	0.66	12.01	28.71
	μ	25.64	2.77	3.92	32.74	0.12	8.15	0.26	-0.95	-0.39	8.96	20.68
	σ^2	8.07	0.86	0.85	7.07	0.02	34.34	0.99	3.45	1.31	1.78	4.27

A: leaf area (dm²). LAI: leaf area index. LAR: Leaf area ratio (cm²/gplant). SLA: specific leaf area (cm²/gleaf). LWR: leaf weight ratio (gleaves/gplants). CGR: crop growth rate (g/m²/day). NAR: net assimilation rate (mg/cm²/day). RGR: relative growth rate in weight (mg/g/day). G: biomass production speed (g/day). LAD: leaf area duration (cm²/day). Z: biomass duration (g/day). Min. Minimum value Max. Maximum value. μ Population mean. σ^2 Population variance.

A describes the size of the assimilation organs of plants (leaves), whereas LAI refers to a plant community (Kvet *et al.*, 1970, which explains a similar behavior, by being interrelated to each other. Both indicators depend on several factors, including the climate, low temperatures, poor rainfall occurrence and deficient sunlight, causing a decrease in A and LAI in the area where the study took place (Table 1).

Leaf area ratio (LAR) and biomass duration (Z) behaved in the opposite way, LAR decreased and Z increased with cutting age. This behavior is explained by LAR's close relation with A and LAI, because it characterizes the specific size of the assimilating organ, present in an instant of time. Ortega *et al.* (1989) in a study on sugar cane cultivars, noted that the indicator decreased with age.

In turn, Z is the total dry weight of a plant in relation to time, and also an approximate value of persisting vitality (Kvet *et al.*, 1979). Biomass duration considers not only dry weight increase values, but also durability. These positive values shown by the population are truly important, as they indicate good biomass durability. Similar increases were reported by Torres *et al.* (2012)

in a study comprising three cultivars of commercial sugar cane planted in different cycles.

Specific leaf area (SLA) increased at 218 days in comparison to the previous cutting age assessed. It was followed by a decrease until day 370. This indicator measures the proportion of leaves according to their dry weight, so it could provide knowledge about thickness. Its increase means a decrease in thickness, and vice versa (Torres *et al.*, 2013). It may be inferred that the population study had a tendency to increase leaf thickness. Those results were favorable, Vazquez and Torres (1995), claimed that thick leaves can absorb light fully, using a lower percentage of light; i.e., light that goes through the leaf, so they are more efficient during photosynthesis, producing more biomass. Similar results were published by Ortega *et al.* (1989), who noted decreased SLA with age in sugar cane cultivars.

Leaf weight ratio (LWR) had a decrease since the first assessment performed, until day 309 of cutting age; then it kept a constant level, to the last age assessed. This behavior corroborated reports by Torres (2006), that during leaf development when the crop is growing fully, LWR values are high when the stage starts to decline, before maturation, and the values decrease. Therefore, time evolution of LWR is more related to maturation of sugar cane than with the characteristics of other varieties.

The crop growth rate (CGR) increased from day 218 and to day 278, then it decreased, on day 309, and then increased again on day 339, to finally decrease significantly, during the last assessment. Similar results were published by Torres (2006). Lower values were reported by Ortega *et al.* (1989) within 2.0 and 32.0 g.plant.m⁻², for CGR (day⁻¹) This indicator is very important for forage production, it represents dry matter increases in grams produced per day by the plants in one m².

The net assimilation rate (NAR), biomass production speed (G), and the relative growth rate in weight (RGR) had very irregular behaviors, first with a tendency to decrease, then increase at the next age. It was repeated three times in the study, until the three indicators ended up with a negative value in the last age assessed. The negative results achieved on day 370 may be associated to low rainfall observed in the last three months studied (Table 1), also confirmed by Armas *et al.* (1999) that drought stress triples estome resistance and decreases CO₂ input for photosynthesis, with ensuing reduction of dry matter and growth.

NAR represents the speed of plant dry weight increase per leaf area unit. This net weight increase is the result of a balance between photosynthesis and respiration (Armas *et al.*, 1988; Vázquez and Torres, 1995). Similar results were published by Ortega *et al.* (1989) and Torres (2006).

G is an important indicator to choose sugar cane cultivars with forage potential, because it tells the age at which the plant begins biomass production, and

when it can decrease. It also allows for time fluctuations. The values achieved in this study are better than the reports by Ortega *et al.* (1989) and Torres (2006) in commercial cultivar assessments for sugar production, which corroborates the high forage potential of the 48 individuals that integrated the population assessed.

Leaf area duration (LAD) increased between days 218 and 278, then it declined until day 370. This is a very important indicator to select forage cultivars, because it provides quantitative information about the time a plant keeps its assimilating surface active. Therefore, it is reasonable to have high and positive correlations between LAD and yields from different plant species (Kvet *et al.*, 1979). LAD behavior may be attributed to climatic conditions that prevailed in the area where the study was developed (Table 1). Differences in temperatures, rainfall and sunlight became more remarkable from one assessment to another. This indicator was also similar to the results achieved for A and LAI.

CONCLUSIONS

The results achieved have provided quantitative values that can be used as reference for selection and assessment of forage genotypes for ruminant nutrition.

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