Evaluation and grouping of sugarcane genotypes in agreement with their physiologic characteristics types

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Abstract – The objective of this research was to evaluate physiological characteristics of sugar cane genotypes, as well as characterize them in groups according to their similarity, checking the ability of ecological adaptability of these genotypes. The work was performed in field conditions, being assessed ten sugarcane genotypes (RB855113, RB835486, RB867515, SP80-1816, RB72454, RB925345, RB855156, RB937570, RB947520 and RB925211) in a randomized block design with three replications. It were evaluated the stomatal gas flow rate (U - µ mol s^{-1}), the concentration of under-stomatal CO_2 (Ci - µmol mol^{-1}), the photosynthetic rate (A - µmol m^{-2} s^{-1}), the CO_2 consumed (Δ C - µmol mol^{-1}), the stomatal conductance (Gs - mol m^{-1} s^{-1}), the temperature gradient between leaf and air (Δ T), and the transpiration rate (E - mol H_2O m^{-2} s^{-1}), being also calculated the water use efficiency (WUE - mol CO_2 mol H_2O^{-1}) from the values of the amount of CO_2 fixed by photosynthesis and amount of water transpirated. Both univariate and multivariate data analysis were made. The genotype SP80-1816 showed better water use efficiency, combined with low stomatal conductance and transpiratory rate. The cultivar RB855113 stood out by having high photosynthetic rate, and high consumption of CO_2. The cultivar RB867515, in addition to showing high water use efficiency, also showed high photosynthetic rate. With respect to the multivariate analysis, the biotypes RB925345, RB925211, RB855156 and RB855113 are situated in different groups when compared to the others as to the physiological characteristics with respect to other genotypes with isolation in separate groups.

Keywords: *Saccharum* spp, photosynthesis, transpiration, cultivars

Avaliação e agrupamento de genótipos de cana-de-açúcar de acordo com suas características fisiológicas

Resumo – Objetivou-se com este trabalho avaliar as características fisiológicas de genótipos de cana-de-açúcar, bem como caracterizá-los em grupos de acordo com a similaridade dos mesmos, aferindo desta forma, a capacidade de adaptabilidade ecológica dos referidos genótipos. O trabalho foi realizado em condições de campo, sendo avaliados dez genótipos de cana-de-açúcar (RB855113, RB835486, RB867515, SP80-1816, RB72454, RB925345, RB855156, RB937570, RB947520 e RB925211) em delineamento de blocos casualizados com três repetições. Foram avaliados a taxa de fluxo de gases pelos estômatos (U - µmol s^{-1}), a concentração de CO_2 sub-estomática (Ci - µmol mol^{-1}) e a taxa fotossintética (A - µmol m^{-2} s^{-1}), o CO_2 consumido (ΔC - µmol mol^{-1}), a condutância estomática de vapores de água (Gs – mol m^{-1} s^{-1}), o gradiente entre temperatura da folha e do ar (ΔT), e a taxa de transpiração (E – mol H_2O m^{-2} s^{-1}), sendo calculada ainda a eficiência do uso da água (WUE – mol CO_2 mol H_2O^{-1}) a partir dos valores de quantidade de CO_2 fixado pela fotossíntese e quantidade de água transpirada. Realizou-se a análise univariada e multivariada dos dados. O genótipo SP80-1816 apresentou melhor eficiência no uso da água, aliada a baixa condutância estomática e taxa transpiratória. Já o cultivar RB855113 se destacou por apresentar elevada taxa fotossintética, bem como elevado consumo de CO_2. A cultivar RB867515, além de demonstrar alta eficiência no uso da água, mostrou também elevada taxa fotossintética. Com relação à análise multivariada, os biótipos RB925345, RB925211, RB855156 e RB855113 estão dispostos em grupos diferentes quanto às características fisiológicas quando comparados aos demais genótipos com isolamento em grupos distintos.

Palavras-chave: *Saccharum* spp, fotossíntese, transpiração, cultivares.

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INTRODUCTION

In Brazil, sugarcane crop is planted in an area exceeding five million hectares, producing over 620 million tons of sugarcane per season (CONAB, 2011). Brazil stands out as a world leader in sugar exports, as well as their use as a source of renewable energy. The main emphasis in terms of agroenergy in Brazil is the production of ethanol from sucrose produced by sugarcane fermentation (EMBRAPA, 2007), being this crop the best alternative for fuel production from the economic, energetic and environmental points of view (ANDREOLI & SOUZA, 2006). Brazil, for having area, climate and technology already developed, could supply half the world ethanol demand. However, for the country to be able to contribute with 200 billion litres of alcohol a year, it will be necessary to increase the number of alcohol refineries and the fuel production efficiency, as well as expand the area planted with sugarcane (COLOMBO, 2006).

However, it should be emphasized that the optimum sugarcane sprouting is obtained in temperatures close to 32 °C, and can be paralyzed when the temperatures are below 20 ºC (BARBIERI, 1981). Regarding sugarcane growth, according to Santos et al., (2009), in temperatures below 25 °C it is slow, between 30 and 34 °C it is maximum, and over 35 °C it becomes too slow and is virtually null in temperatures above 38 °C. Sugarcane presents C_4 metabolism, high photosynthetic efficiency and high light saturation point, thus, the greater is the light intensity, the greater is its development and sugar accumulation (BARBIERI, 1981).

Thus, in addition to the above factors, some others may influence the sprouting, the development and mainly the photosynthetic activity of sugarcane, directly or indirectly, as water deficit, heat stress (LORETO & BONGI, 1989), external and internal gas concentration (KIRSCHBAUM & PEARCEY, 1988) and light composition and intensity (SHARKEY & RASCHKE, 1981), among others. While the ability of exchanging gas by stomata is deemed to be the main limitation of the assimilation of photosynthetic CO_2 (HUTMACHER & KRIEG, 1983), it is improbable that gas exchange will limit the photosynthesis rate when interacting with other factors.

Under natural conditions, due to the variation of environmental factors, simultaneously, the assessment of the stomatal opening and gas exchange regulation mechanisms is more complex (SCHULZE & HALL, 1982). Researches of this nature are crucial to understanding the adaptive processes of agricultural species, when commercially exploited.

The objective of this work was to evaluate physiological characteristics of sugarcane genotypes, and characterize them in groups according to their similarity, as a way to check their ability of ecological adaptability.

MATERIAL AND METHODS

The work was carried out in field conditions on the Experimental Field of the Federal University of Viçosa, located at Oratórios city – MG. The soil was a Red Yellow Latosol previously fertilized according to fertilization recommendations of Cantarutti et al. (2007) and it were assessed physiological characteristics of sugarcane genotypes (RB855113, RB835486, RB867515, SP80-1816, RB72454, RB925345, RB855156, RB937570, RB947520 and RB925211) in a randomized block design, with three replications.

Dry matter of plants was evaluated at 45 and 60 days after emergence (DAE) and physiogical evaluations were conducted at 45 DAE. For this, it was used the middle third of the first fully expanded leaf with visible ligule. It was used an infrared gas analyzer (IRGA), model LCA Pro+ (Analytical Development Co. Ltd, Hoddesdon, UK). Plots were constituted by ten sugarcane plants spaced of 1.4 m and 10 m of length, each line representing a genotype, being that in all assessments made in each observation unit it were considered always ten plants in the center of each experimental unit. Each block was assessed at one day, between eight and ten hours in the morning, in order to maintain conditions homogeneous during the evaluation. It were evaluated two plants per plot.

It were evaluated the stomatal gas flow rate (U - μmol s^{-1}), under-stomatal CO_2 concentration (Ci - μmol mol^{-1}), photosynthetic rate (A - μmol m^{-2} s^{-1}), CO_2 consumed (ΔC - μmol mol^{-1}), stomatal conductance (Gs - mol m^{-1} s^{-1}), temperature gradient between leaf and air (ΔT), and transpiration rate (E - mol H_2O m^{-2} s^{-1}), being also calculated the water use efficiency (WUE - mol CO_2 mol H_2O^{-1}) from the values of the amount of CO_2 fixed by photosynthesis and the amount of transpirated water.

Data were submitted to the analysis of variance by the F test and subsequently it was applied the Duncan test, being both tests performed with probability of error of 5 % (PIMENTEL-GOMES, 1987). Data were also subjected to multivariate analysis, to group the genotypes according to their degree of similarity by the principal components method.

Principal Components Analysis (PCA) is used to evaluate the interrelationships of a large number of variables, in order to condense the most important information to a lesser number of variables, with minimal loss of information. Hence, the PCA allows verifying the discriminatory ability of the original variables in the process of forming the groups, being employed to reduce the number of indicators to two not correlated new variables, which are the main components, indicated by CP1 and CP2. The new set of variables, in order of estimation, retains the maximum information in terms of total variation. These variables will explain the better the variability expressed between the assessed individuals the smaller the number of variables that accumulate at least 70 % of the total variation (CRUZ, 1990).

RESULTS AND DISCUSSION

Genotypes RB925211, RB867515 and RB947520 showed higher dry matter accumulation than the others at 45 days after emergency (DAE). At 60 DAE, genotypes RB72454, RB835486, RB855113, RB867515, RB925211, RB937570 and RB947520 demonstrated increase in dry matter accumulation, but without statistically differing from the others (Figure 1).

The results for the physiological characteristics (CO_2 consumed - ΔC, gas flow rate - U, leaf internal CO_2 concentration - CI, transpiration - E, stomatal conductance - Gs, photosynthetic rate - A, temperature difference ΔT and water use efficiency – WUE) submitted to univariate analysis are listed in Tables 1 and 2.

The RB855113 genotype showed higher CO_2 consumption (ΔC), differing only from the RB835486 genotype. The CO_2 consumed is directly related to the photosynthetic intensity of the plant at the time of evaluation, that is, in general, the more accelerated the plant metabolism, the greater the CO_2 consumption per unit of time.

Regarding the stomatal gas flow (U), the lowest value was associated with the RB867515 genotype, and highest value was found in the RB855156.

Gas flow varies with environmental conditions, being dependent on the interactions between the mesophyll cells and stomata (BURROWS & MILTHORPE, 1976; RASCHKE, 1979). The stomatal movement is the fastest way plants possess to adjust to the environmental changes photosynthetic organs are submitted to (PASSIOURA, 1982). Propitious conditions to carbon fixation support stomatal opening, while conditions for water loss favor its closing. Stomatal opening regulation occurs by complex mechanisms (Raschke, 1979), seeming to act, however, in a sense of minimizing water loss, limiting in a less intense way the CO_2 influx (COWAN, 1982).

The RB855113 genotype showed greater internal CO_2 concentration (CI) when compared to the other cultivars. The CI in this genotype was about six times higher than in RB925345 genotype.

The genotypes SP80-1816, RB925211 and RB925345 had reduced transpiratory rate (E), where RB855156 demonstrated superior value in this feature. According to Brodribb & Holbrook (2003), reduced or elevated transpiration is associated with the capacity of stomatal opening regulation, and variations in the stomatal opening cause changes in water potential, by acting on the transpiration. The efficiency of a plant to use water relates to the perfect transpiratory rate regulation, that is, the ability to regulate stomatal opening and closing, which in turn is linked, to a lesser degree, to the photosynthetic efficiency (CONCENÇO et al., 2007).

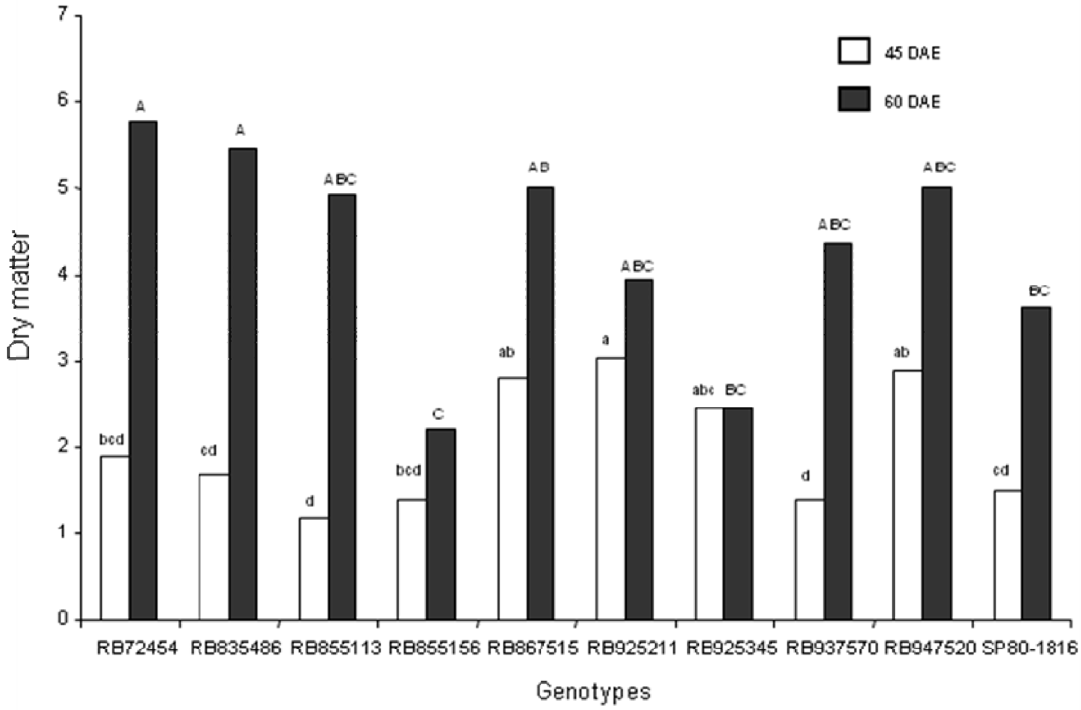

Figure 1. Dry matter accumulation in sugarcane genotypes, evaluated 45 and 60 days after emergency. Means in a given white bar followed by the same letter (lower case) do not differ significantly ($P \leq 0.05$) as determined by Duncan's multiple range test. Means in a given black bar followed by the same letter (capital letter) do not differ significantly ($P \leq 0.05$) as determined by Duncan's multiple range test**.**

It was observed a wide variation among genotypes with regard to stomatal conductance (GS), with the RB947520 genotype showing a GS two times higher than RB72454. Leaf conductance is composed in small part by epidermal cuticular conductance and, when the stomata are open, GS, which is controlled by the stomatal guard cells. Thus, the GS is proportional to the number and size of stomata, as well as to their opening diameter, features which depend on other endogenous and environmental factors (BRODRIBB & HOLBROOK, 2003). Ferreira et al. (2005), working with five sugarcane genotypes, among them the RB855113, noted that this genotype showed high stomatal density, coupled with great ostiole length. In this work, the RB855113 presented the third largest stomatal conductance, not differing from the RB947520 and RB937570 genotypes.

The processes of transpiration and CO_2 capture only occur when the stomata are open, as well as the GS. In function of the latent heat of evaporation, transpiration presents itself as a powerful cooling effect, important in regulating leaf temperature (FARQUHAR & RASCHKE, 1978).

Table 1. Physiological characteristics at 45 days after crop emergence (CO_2 consumed – ΔC, stomatal gas flow rate – U, leaf CO_2 internal concentration – CI, transpiration – E) from sugarcane genotypes. Oratórios-MG, 2008.

Cultivars	ΔC µmol mol^{-1}	U µmol s^{-1}	CI µmol mol^{-1}	E mol H_2O m^{-2} s^{-1}
RB72454	106.0ab	68.3abc	169.7abc	4.4ab
RB835486	89.3b	68.2abc	161.3abc	4.6ab
RB855113	147.7a	68.2abc	220.7a	4.3ab
RB855156	136.7ab	68.8a	123.3bc	5.8a
RB867515	104.0ab	67.8c	200.3ab	4.9ab
RB925211	115.0ab	68.2bc	97.0cd	3.0b
RB925345	137.3ab	68.1bc	38.3d	3.3b
RB937570	98.3ab	68.5ab	186.0abc	5.3ab
RB947520	98.7ab	68.0bc	146.0abc	4.8ab
SP-801816	97.3ab	68.4abc	143.0abc	2.8b
CV (%)	23.5	0.44	31.1	28.5

Means in a given column followed by the same letter do not differ significantly ($P \leq 0.05$) as determined by Duncan's multiple range test.

The RB855113 genotype showed higher photosynthetic rate (A), differing only from RB835486 and SP80-1816 genotypes. The balance and composition of incident radiation on the plant, allied to the level of carbohydrates in the leaves, can increase the respiratory rate directly or through alternative routes associated with the respiratory chain (PYSTINA & DANILOV, 2001), which could make the balance of photosynthesis even smaller and reduce the ability of mass accumulation of the plant. Several species of weeds and commercial crops alter their photosynthetic rate at different levels under the same environmental conditions (PROCÓPIO et al., 2004). The increase in CO_2 internal concentration causes linear increase on the photosynthetic rate, until it reaches the maximum photosynthesis. The greater the photosynthetic rate, the lower the CI. Genotypes RB835486 and SP80-1816, which showed the lowest values of A, had high values of CI. The elevated photosynthetic rate is related to the increased capacity of the plant to produce photosynthates and, therefore, to its greatest growth and development. Terauchi & Matsuoka (2000) report that the ideal characteristics of sugarcane cultivars could be related to the rapid growth and development in the initial phase, which corresponds to tillering occurring between 25 to 40 days after the emergency.

RB855113, RB925211 and RB867515 genotypes showed greater difference between leaf and air temperature (ΔT). The plant metabolism causes increase in leaf temperature, so that, as a rule, it is higher than the air around her. Thus, increases in metabolism may be indirectly measured as a function of temperature gradient between the leaves and air (ΔT). Usually, the difference is only one or two degrees, but, in extreme cases, can exceed 5 ºC (DRAKE & SALISBURY, 1972; ATKIN et al., 2000).

The RB835486 genotype was the less efficient in water use (WUE), differing from genotypes SP-801816 and RB947520. WUE represents the amount of CO_2 fixed for the production of dry matter in function of the quantity of transpirated water (CONCENÇO et al., 2007).

Table 2. Physiological characteristics (stomatal conductance – GS, photosynthetic rate – A, temperature difference – ΔT, and water use efficiency – WUE) from sugarcane genotypes. Oratórios-MG, 2008.

Cultivars	GS mol m^{-1} s^{-1}	A μmol m^{-2} s^{-1}	ΔT ^{O}C	WUE mol CO_2 mol H_2O^{-1}
RB72454	0.19d	33.8abc	3.2ab	7.9ab
RB835486	0.29cd	28.6bc	5.8a	5.1b
RB855113	0.44abc	47.1a	5.2a	6.9ab
RB855156	0.40abc	43.7ab	4.1ab	7.5ab
RB867515	0.32bcd	33.2 abc	6.1a	8.2ab
RB925211	0.47ab	36.9abc	4.9a	9.6ab
RB925345	0.33bcd	36.0abc	4.3ab	8.6ab
RB937570	0.47ab	31.3abc	3.6ab	6.3ab
RB947520	0.50a	31.5abc	3.2ab	9.6a
SP-801816	0.29cd	24.9c	1.8b	8.8a
CV (%)	21.6	24.1	36.6	28.4

Means in a given column followed by the same letter do not differ significantly ($P \leq 0.05$) as determined by Duncan's multiple range test.

In a general way, a plant that shows high efficiency in water use and high photosynthetic rate usually shows greater capacity for dry weight accumulation. These features are found in RB72454, RB855113, RB867515 and RB947520 genotypes at 45 DAE. Currently, the RB867515 genotype is among the most used for the production of sugar and alcohol, due to characteristics such as high productivity and high sugar levels., Terauchi & Matsuoka (2000) report that the ideal characteristics of cultivars of sugarcane were related to the rapid growth and development in the initial phase, which corresponds to tillering. So, in order to have rapid growth at this stage, it would be necessary morphophysiological characteristics that could favor the variables associated with photosynthesis, and reduce water use efficiency, provided that in a level not enough to compromise the photosynthesis.

Tables 1 and 2 show the physiological characteristics of the ten sugarcane genotypes studied which were used in multivariate statistical analysis. Table 3 shows the correlation coefficients between eight physiological characteristics the and two main components (CP1 and CP2).

The contribution intensity of the variables in the discrimination of the genotypes is directly related to the highest values of the main components CP1 and CP2. The information retained in the set of those variables concerning the first component was 86.8 %. The variables that contributed most to the discrimination of the genotypes were leaf CO_2 internal concentration and transpiration.

The information retained for the second component was 13.0 %. Overall, the main components CP1 and CP2 showed 99.7 % of reliability, allowing to select the main features. Rodella et al. (2006), verified a reliability of 83.94 % for the first two major components, from the analysis of only seven of eight leaf anatomical features from accesses of two species of *Egeria*. Ferreira et al. (2007), working with canonical variables, found accumulated information of 78.99 % for the two canonical variables.

Table 3. Correlations between the original variables concerning the physiological characteristics of sugarcane genotypes and the two main components (CP1 and CP2). Percentage of retained and accumulated information in CP1 and CP2, and ordering of the variables as its discriminatory power.

Variables	CP1	Order	CP2	Order
CO_2 consumed (ΔC)	-0.265	7	0.964	1
Gas flow rate (U)	-0.069	6	0.214	5
Leaf internal CO_2 concentration (CI)	0.999	1	0.037	7
Transpiration (E)	0.448	2	0.127	6
Stomatal conductance (Gs)	-0.011	4	0.225	4
Photosynthetic rate (A)	0.021	5	0.948	2
Temperature difference (ΔT)	0.188	3	0.265	3
Water use efficiency (WUE)	-0.426	8	-0.103	8
Retained information (%)	86.60		13.00	
Accumulated information (%)	86.60		99.71	

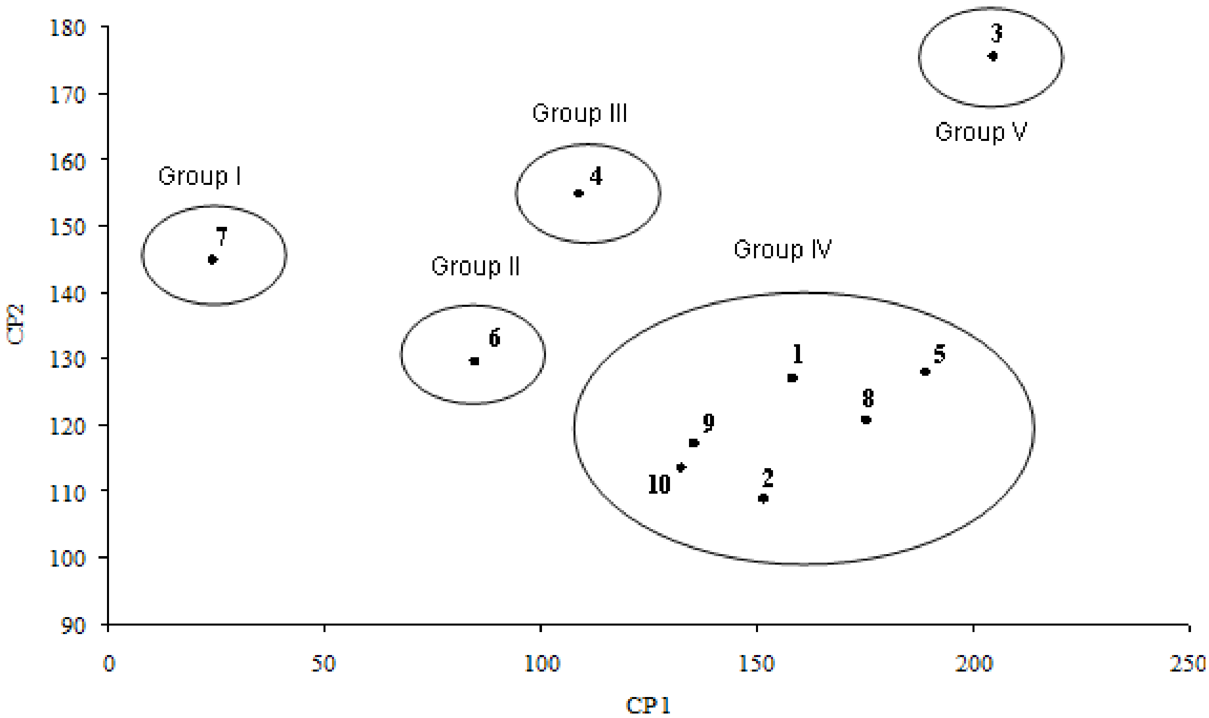

Figure 2. Graphic dispersion of sugarcane genotypes (1-RB72454, 2-RB835486, 3-RB855113, 4-RB855156, 5-RB867515, 6-RB925211, 7-RB925345, 8-RB937570, 9-RB947520 and 10-SP80 - 1816), using the first two principal components (CP1 and CP2) to the set of eight variables: CO_2 consumed, gas flow rate, leaf internal CO_2 concentration, transpiration, stomatal conductance, photosynthetic rate, temperature difference and water use efficiency.

For the ten assessed sugarcane genotypes, cluster analysis and principal component analysis of physiological variables were efficient to discriminate five groups as to the similarity (Figure 2). Genotypes RB925345, RB925211, RB855156 and RB855113 were isolated in groups I, II, III and

V, respectively, while genotypes RB72454, RB835486, RB867515, RB937570, RB947520 and SP80-1816 showed greater similarity, being grouped into only one group (IV).

Ferreira et al. (2007), working with anatomical characteristics of the leaves of five sugarcane varieties (RB867515, RB957689, RB855113, SP80-1842 and SP80-1816) subjected to multivariate analysis, observed the isolation of RB855113 in a different group from other cultivars, thus demonstrating that it presents an important feature, which is the high susceptibility to herbicides.

CONCLUSIONS

According to the results, it can be affirmed that the SP80-1816 genotype showed better water use efficiency, combined with low stomatal conductance and low transpiratory rate. Meanwhile, the RB855113 cultivar stood out showing high photosynthetic rate, with consequent high CO_2 consumption. The RB867515 cultivar, in addition to high water use efficiency, also showed high photosynthetic rate. With respect to the multivariate analysis of the data, RB925345, RB925211, RB855156 and RB855113 biotypes were isolated in separate groups, differentiating themselves from the physiological characteristics of the other six cultivars.

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