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Research Paper

The use of ethephon for improving shoot development of buds from different positions of BRS Capiaçú elephant grass culms

El uso del etefón para mejorar el desarrollo de brotes en diferentes posiciones del culmo de pasto elefante Capiaçú

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Abstract

Use of ethephon on buds from different positions on the culm of BRS Capiaçú elephant grass was evaluated to improve development of shoots during planting. The experiment compared buds originating from the apex and base of the BRS Capiaçú culm without ethephon application, cut culms with buds immersed for 1 min in ethephon solution (0.650 L ethephon/ha) and application of 150 L ethephon/ha directly on the bud on the standing culm. The position of the bud in the culm and the application of ethephon positively influenced the development and gaseous exchange of BRS Capiaçú. Buds immersed for 1 min in ephethon showed better development, CO₂ assimilation rate (A), transpiration (E) and stomatal conductance. Plants growing from buds from the base of the culm produced more leaves, with higher biomass and higher transpiration, It is recommended to use buds from the base of the BRS Capiaçú culm for propagation when using ethephon.

Keywords: Cenchrus purpureus (Schumach.) Morrone, forage, phytohormone, stomatal conductance, transpiration.

Resumen

Se evaluó el uso de etefón en diferentes posiciones en el culmo de pasto elefante BRS Capiaçú para mejorar el desarrollo de los brotes durante la fase de establecimiento. El experimento comparó brotes originados del ápice y la base del culmo BRS Capiaçú sin aplicación de etefón, porciones de tallo cortadas con brotes sumergidos durante 1 min en solución de etefón (0.650 L de etefón/ha) y la aplicación de 150 L de etefón/ha directamente en el brote en el culmo en pie. La posición del brote en el culmo y la aplicación de etefón influyeron positivamente en el desarrollo y el intercambio gaseoso de BRS Capiaçú. Los brotes sumergidos durante 1 min en etefón mostraron mejor desarrollo, tasa de asimilación de CO_2 (A), transpiración (E) y conductancia estomática. Las plantas que crecen a partir de yemas de la base del culmo produjeron más hojas, con mayor biomasa y mayor transpiración. Se recomienda utilizar yemas de la base del culmo de BRS Capiaçú para la propagación cuando se utiliza etefón.

Palabras clave: Cenchrus purpureus (Schumach.) Morrone, conductancia estomática, fitohormona, forraje, transpiración.

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Introduction

The elephant grass (Cenchrus purpureus (Schumach.) Morrone) cultivar 'BRS Capiacú' (BRS Capiacú) stands out from other elephant grass cultivars for its resistance to damping off, ease of mechanical harvesting and erect and dense clumps. It is a tall cultivar, reaching up to 4 m height in favorable conditions, planted from vegetative cuttings that produce plants with a high number of tillers from the axillary and basal buds. It has high regrowth rates that support up to 4 cuts during the year and it is widely recommended and adopted in Brazil. Unlike other pasture grasses, it needs to be replanted every 3 years to sustain high productivity (Lisboa et al. 2019a).

BRS Capiaçú can be used to produce good quality silage or as fresh chopped green feed for livestock (Pereira et al. 2016). This forage produces more dry matter at a lower cost than corn and sugarcane, and its silage is a cheaper alternative for pasture supplementation in the dry season (Gonçalves et al. 2022; Alves et al. 2022). Bud selection is an important decision at planting because the position of the bud in the culm can influence sprouting, due to the lower amount of lignin in tissues (Kalanzi and Mwanja 2023; Santos et al. 2023) and higher amount of auxins that control cell elongation (Taiz et al. 2017). The first sprouting of the culms occurs when soil moisture and temperature conditions allow the meristems in buds to initiate enzymatic and hormonal metabolism (Lisboa et al. 2019a). Exogenous application of phytohormones can support more uniform plant development and a greater number of tillers in the formation of clumps (Chang and Williams 2010).

The use of the phytohormone Ethephon, which releases ethylene, can improve production. The exogenous application of ethylene can promote better expansion of the aerial part of plants in the early stages of crop development. Ethylene is a gaseous hormone that is naturally synthesized in plants and acts in cell expansion, flowering and fruit maturation (Chang 2016; Chang and Williams 2010; Taiz et al. 2017). Ethylene was observed to promote greater tillering in the initial phase of development of sugarcane (Lisboa et al. 2019b) when investigating the responses of exogenously applied ethylene to sugarcane buds.

The root system can be influenced by ethylene, promoting greater root growth with increased nutrient and water uptake, so plants can withstand a longer period of water stress (Chen et al. 2021). Phytoregulators can also cause internal changes in tissues and physiology, promoting better response in gas exchange parameters, including internal carbon concentration of the substomatal chamber, stomatal conductance, and leaf transpiration (Pan Rui et al. 2022). Photosynthesis requires a high carbon dioxide (CO₂) concentration in the mesophyll cells. Exogenously applied ethylene can promote partial stomatal closure, which can reduce stomatal conductance and transpiration and increase the internal concentration of CO2, making the photosynthetic process more efficient (Marin and Nassif 2013).

Farmers are seeking increased tillering in forages during the formation of clumps at the time of planting or after grazing to support greater dry mass production/area and greater tolerance to abiotic stresses. The exogenous application of ethephon for increased tillering is a possible strategy for better establishment of elephant grass.

Sugar degradation occurs naturally in cellular metabolism to obtain the chemical energy (adenosine triphosphate) (Taiz et al. 2017). When the ethephon molecule enters the cell, in the slightly alkaline cellular environment, it promotes the decomposition into ethylene, phosphate, and chloride ions (Zhang Wei and Wen Chi-Kuang 2010). The presence of ethylene within the cell stimulates the storage of these free sugars (sucrose and glucose-6-phosphate) as starch. When the grass seedlings are removed from ethylene, this starch is converted back into sugars, which are then consumed during the lateral bud sprouting process, increasing tillering in the early stages of forage development.

The objective of this work was to evaluate the use of ethephon in buds originating from different positions on the BRS Capiaçú culm on CO₂ assimilation rate/area (A), transpiration (E), stomatal conductance (gs), internal concentration of CO, in the substomatal chamber (ci), efficient use of water (EUW), stomatal density (DEM), stomatal functionality (FUNE), length of the aerial shoot (APL), number of fully expanded leaves (NL) and dry mass of the aerial shoot and roots (DMAP and RDM).

Materials and Methods

The experiment was carried out in August 2023 at the Faculty of Agricultural Sciences at the State University of São Paulo (UNESP), Dracena, São Paulo (21°29'10.24" S, 51°31'41.29" W; 411 masl).

The experimental design was a completely randomized 2 × 3 factorial scheme with 4 replications. The first factor tested was apex and base location of the bud on the BRS Capiaçú culm, and the second factor was ethephon application levels (no application, buds immersed for 1 min in an ethephon solution at a concentration of 0.650 L/ha and application with ethephon solution at a concentration of 0.650 L/ha directly on the bud on the standing culm using a volume of 150 L/ha, giving a total of 6 treatments.

The 10 L capacity pots were filled with 0-0.3 m soil classified as Hypoferric Red Latosol (Santos et al. 2013) (Table 1).

The soil was fertilized following the requirements of the BRS Capiaçú crop according to van Raij et al. (1996). A 5 cm culm fragment with 1 bud from each treatment was planted at a depth of 5 cm, 1 in each pot. The pots were placed in a greenhouse covered with light-diffusing plastic film and 70% black shade screen on the sides. During the experiment, the average maximum daily temperature was 28.5 °C and the minimum was 17.2 °C, and the relative humidity varied between 70-80%. All pots were irrigated until they reached field capacity and weed removal was carried out as needed.

The first fully expanded fresh leaf was selected from the apex of each plantlet from shoots from the apex and base of the culm for gas exchange analysis using the Infra-Red Gas Analyzer (IRGA), BioScientific Ltd, model LC-Pro) 1,200 µmol/m²/s of photosynthetically active radiation (PAR) provided by LED lamps. After stabilization of leaf gas exchange, the following parameters were determined: CO₂ assimilation rate expressed per area (A expressed as µmol CO₂/m²/s); transpiration (E expressed

as mmol H₂O/m²/s); stomatal conductance (gs expressed as mol H₂O/m²/s) and internal concentration of CO₂ in the substomatal chamber (ci expressed as µmol/mol) with 380 ppm of CO, and chamber temperature of 28 °C. For the determination of efficient use of water (EUW) the following formula was applied:

$$EUW = \frac{A}{E}$$

where:

EUW=efficient use of water;

A=CO₂ assimilation rate; and

E=transpiration.

On the same leaf, an abaxial epidermal impression was taken using cyanoacrylate ester (Segatto et al. 2004) to determine DEM and FUNE according to Castro et al. (2009).

After 30 days, plant height (PH), leaf length and leaf width were measured in mm with a graduated ruler. Leaf number (NL) was counted and leaf area (LA) was determined by multiplying leaf length and width. The correction factor $[LA = (leaf length \times leaf width)]$ \times 0.75], was applied according to Simões et al. (2017). To determine dry mass aerial part (DMAP), the green matter was cut close to soil level and root dry mass (RDM) was determined after washing the soil from the roots with running water. The fresh samples were dried in an oven with air circulation at a constant temperature of 65 °C until constant weight.

Statistical analysis

For statistical evaluation, the variables were submitted to normality tests using the Shapiro-Wilk test, followed by analysis of variance using the F test (P<0.05). Means were compared using the Tukey test at 5% probability (Banzatto and Kronka 2013). A Pearson correlation and principal component analysis (PCA) was performed using the R Studio statistical program (R Core Team 2019).

Table 1. Soil chemical attributes.

pН	OM	P	K	Ca	Mg	H+A1	Al	SB	CEC	V%	m%
CaCl ₂	g/dm³	mg/dm³				r	nmolc/dn	1^3			
4.0	4.0	1.0	0.3	6.0	2.0	33	13	8.0	41	20	61

SB=Sum of the bases: V%=Base saturation: m%=Aluminium saturation.

Results

A significant difference was observed for mode of application of ethephon on A, where immersion of the buds in the solution resulted in an increase of 24% (Table 2).

A showed a significant correlation with E, gs and EUW, and also with DMAP and NL (Figure 1), with a significant linear regression (Figure 2).

Buds from the base of the BRS Capiaçú culm had approximately 17% higher transpiration than the buds from the apex and use of ethephon increased transpiration by approximately 19% (Table 2). Buds from the apex without ethephon were negatively correlated with the traits measured, while the buds at the base of the culm and the treatments with ethephon were positively correlated to the traits measured (Figure 3).

Table 2. Mean values of CO₂ assimilation rate (A-μmol CO₂/m²/s); transpiration (E-mmol H₂O/m²/s); stomatal conductance (gs-mol H₂O/m²/s), internal CO₂ concentration in the substomatal chamber (ci-μmol/mol) and efficient use of water (EUW) of BRS Capiaçú shoots.

Position	A	Е	gs	ci	EUW
Apex	11.70a	1.41b	0.073a	188.67a	8.27a
Base	13.15a	1.71a	0.084a	174.56a	7.50a
LSD	2.26	0.17	0.012	26.33	1.00
P-value	0.1928ns	0.0024**	0.1001ns	0.2754ns	0.1268ns
Application					
Absence	11.28b	1.44b	0.069b	172.95a	7.72a
Immersion	14.78a	1.77a	0.092a	169.61a	8.44a
On the bud	11.21b	1.48b	0.074ab	202.29a	7.50a
LSD	3.37	0.26	0.018	39.18	1.49
P-value	0.0216*	0.0081**	0.0143*	0.0913ns	0.2660ns
P-value P x A	0.1494ns	0.5289ns	0.1828ns	0.4280ns	0.3732ns
CV%	21.25	13.06	18.55	16.90	14.82
OM	12.42	1.56	0.079	181.62	7.88

OM=overall mean; CV=Coefficient of variation; LSD=Least significant difference; **=significant at 1% probability level (P<0.01); *=significant at 5% probability level (P<0.05). Means followed by the same letter are not statistically different.

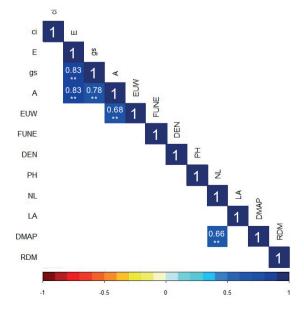


Figure 1. Significant Pearson's correlation among BRS Capiaçú shoot variables.

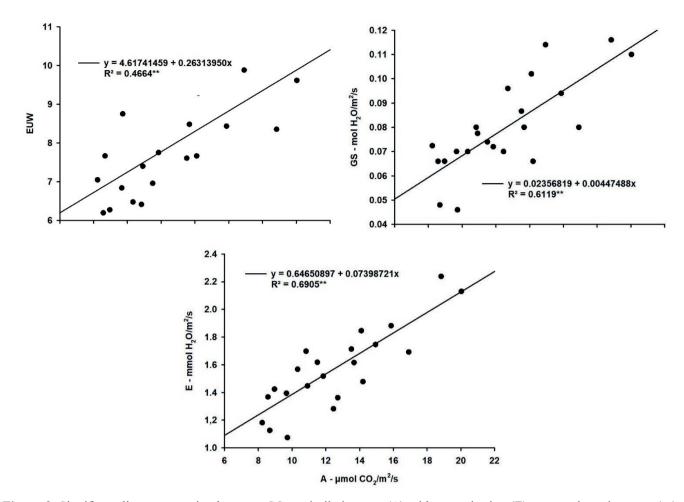


Figure 2. Significant linear regression between CO₂ assimilation rate (A) with transpiration (E); stomatal conductance (gs) and efficient use of water (EUW) of BRS Capiaçú shoots.

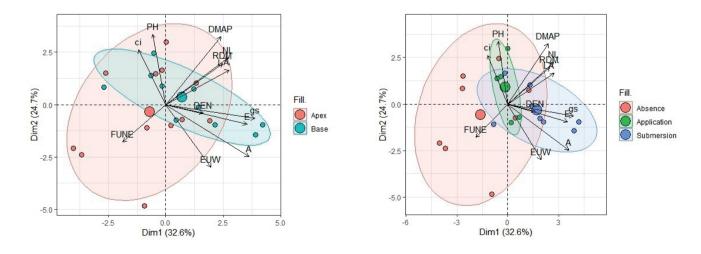


Figure 3. Biplot graph of analysis of significant principal components of BRS Capiaçú, showing the correlations between the positions of the buds in the culm and the ethephon application methods.

A significant difference was found for application of ethephon on gs, where conductance increased by 25% when ethephon was used (Table 2). Stomatal conductance was correlated with leaf transpiration (Figure 4).

No statistical differences were observed for ci, UEA (Table 2), stomatal density and stomatal functionality in BRS Capiaçú (Table 3).

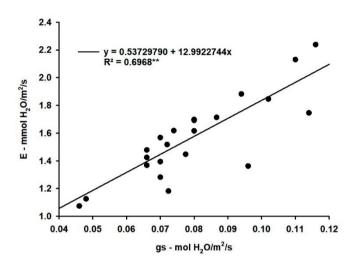


Figure 4. Linear regressions between stomatal conductance (gs) with transpiration (E) of BRS Capiaçú shoots.

No statistical difference was found for PH from buds in different positions in the culm and application of ethephon. NL was statistically different with shoots from buds from the base of the culm showing 14% more leaves than shoots from apex buds. Treatment with ethephon resulted in 33.23% more leaves (Table 4).

Table 3. Mean values of stomatal density (DEN-number of stomata/mm²) and stomatal functionality (FUNE) of BRS Capiaçú shoots.

Position	DEN	FUNE	
Apex	60.96a	1.60a	
Base	64.03a	1.53a	
LSD	9.05	0.21	
p-value	0.4866ns	0.5147ns	
Application			
Absence	57.55a	1.62a	
Immersion	62.85a	1.52a	
On the bud	67.09a	1.55a	
LSD	13.46	0.31	
P-value	0.2219ns	0.6950ns	
P-value P x A	0.1516ns	0.0947ns	
CV%	16.88	15.68	
OM	62.50	1.56	

OM=Overall mean; CV=Coefficient of variation; LSD=Least significant difference; **=significant at the 1% probability level (P<0.01); *=significant at 5% probability level (P<0.05). Means followed by the same letter in the row are not statistically different.

Table 4. Mean values of plant height (PH), leaf number (NL), leaf area (LA), dry mass of the aerial part (DMAP) and root dry mass (RDM) of BRS Capiaçú shoots.

Position	PH (cm)	NL	LA (cm2)	DMAP(g)	RDM (g)
Apex	77.00a	28.91b	547.57a	5.39b	2.85a
Base	80.08a	33.75a	529.64a	7.34a	3.28a
MSD	6.45	4.29	116.01	0.92	0.85
p-value	0.3290ns	0.0294*	0.7492ns	0.0003**	0.3112ns
Application					
Absence	80.87a	23.87b	389.68b	5.52b	2.17b
Immersion	75.25a	35.75a	635.19a	6.63ab	3.93a
On the bud	79.50a	34.37a	590.95a	6.96a	3.10ab
MSD	9.60	6.38	172.60	1.37	1.26
P-value	0.3201ns	0.0003**	0.0043**	0.0380*	0.0088**
P-value P x A	0.3201ns	0.1829ns	0.2775ns	0.0046**	0.0079**
CV%	9.58	15.96	25.11	16.87	32.40
OM	78.54	31.33	538.60	6.37	3.07

OM=overall mean; CV=Coefficient of variation; MSD=Minimum Significant Difference. **=significant at the level of 1% of probability (P<0.01); *=significant at 5% probability level (P<0.05). Means followed by the same letter are not statistically different.

A significant effect was found for mode of application of ethephon on leaf area (LA), where the immersion of the bud in the solution provided a greater leaf area of approximately 39% in the shoot than those from buds without ethephon (Table 4). Ethephon treatment resulted in more and larger eaves, supporting photosynthetic rate and carbon dioxide assimilation (Table 2) which was reflected in the correlation with the increase in the dry mass of the plant (Figure 1).

There was a positive correlation between number of leaves and dry mass of the aerial part of the shoot (Figure 5).

Ethephon increased dry matter with buds from the base of the culm treated with ethephon producing approximately 56% more dry matter than the buds at the apex without ethephon (Table 5).

Visual differences in shoot development were noticeable after treatment with ethephon (Figure 6).

A significant interaction was observed between root dry mass and ethephon treatment (Table 4), where the shoots from buds at the base of the culm immersed in ethephon solution, showed 77% higher root mass when compared to shoots from buds at the apex without ethephon (Table 6).

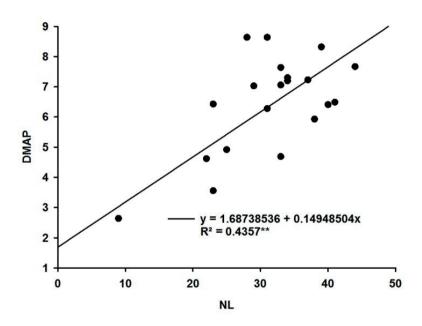


Figure 5. Significant linear regressions between number of leaves (NL) and dry mass of the aerial part (DMAP) of BRS Capiaçú shoots.

Table 5. Average values (g) of the interaction among bud position on the culm and ephethon treatment on dry mass of the aerial part (DMAP) of BRS Capiaçú shoots.

	Absence	Immersion	On the bud
Apex	3.35bB	6.20aA	6.62aA
Base	7.05aA	7.68aA	7.30aA
	MSDCol: 1.59	MSDLin: 1.94	

Means followed by the same lowercase letter in the column do not differ statistically. Means followed by the same capital letter in the row do not differ statistically. MSD=Minimum Significant Difference.

Table 6. Average values (g) of the interaction among bud position on the culm and ephethon treatment on root dry mass (RDM) from BRS Capiaçú shoots.

	Absence	Immersion	On the bud
Apex	0.95bB	3.78aA	3.55aA
Base	3.40aA	4.07aA	2.66aA
	MSDCol: 1.47	MSDLin: 1.79	

Means followed by the same lowercase letter in the column do not differ statistically. Means followed by the same capital letter in the row do not differ statistically. MSD=Minimum Significant Difference.

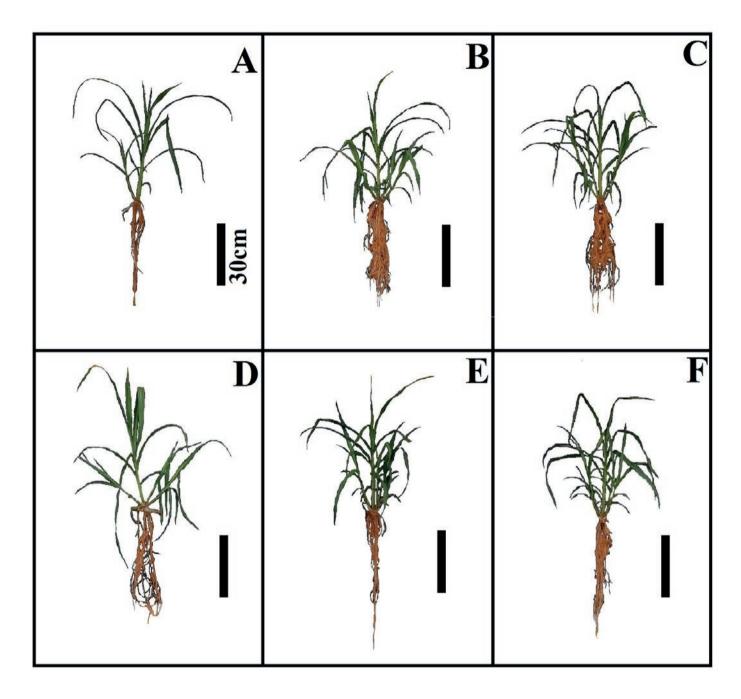


Figure 6. BRS Capiaçú shoots after planting the shoots with buds originating from different positions on the culm and with ethephon application after 30 days growth. A=Bud from the apex of the culm without ethephon; B=Bud from the apex of the culm immersed in ethephon for 1 min; C=Bud from the apex of the culm with application on the culm; D=Bud from the base of the culm without ethephon; E=Bud from the base of the culm immersed in ethephon for 1 min; F= Bud from the base of the culm with application on the culm.

Discussion

This research demonstrates that ethephon can be used to increase shoot development from buds in BRS Capiacú. Ethephon is low cost, easy to apply and can be applied to buds through soaking or buds on culms on the mother plants or buds on culms in the planting furrow.

The use of a phytoregulator may be reflected in CO, assimilation rates, particularly when it releases ethylene during degradation and metabolism, because ethylene acts directly on the stomatal opening and closing mechanisms and guarantees greater gas exchange (Gao Yang et al. 2022; Yao Guang-Qian et al. 2020).

Buds from the base of the culm usually have a higher concentration of sucrose while buds from the apex of the culm have a higher concentration of glucose, therefore implying a greater expenditure of energy for the reduction of sucrose into glucose in buds from the base of the culm, which may have been reflected in transpiration by the leaves (Aude 1993).

Treatment method also affected the results, possibly because the immersion of the buds in the solution may have provided a greater contact surface with ethephon, which was reflected in increased release of ethylene in the bud and increased foliar transpiration in BRS Capiaçú (Lisboa et al. 2019a; Pérez-Pérez et al. 2020).

The stomatal conductance (gs) results confirm that after the degradation of ethephon and ethylene release, ethylene started to influence the stomatal opening, and provided greater stomatal conductance (Lisboa et al. 2019b; Yao Guang-Qian et al. 2020). The concentration of abscisic acid and ethylene hormones within the xylem vessels are the factors that most influence stomatal opening and closing (Pérez-Pérez et al. 2020).

Differences for plant height (PH) can be explained by ethylene supporting better development of size and number of leaves, producing similar results to the higher number of leaves observed when ethephon was applied exogenously in sugarcane (Lisboa et al. 2019b). The increase in the ethylene concentration in the leaves also accelerates its senescence (Peerzada and Igbal 2021). Further work is needed to determine the appropriate concentrations of ephethon to increase plant growth without causing senescence in BRS Capiaçú.

Increased growth with application of ephethon in buds at the base of the culm in BRS Capiacú was not expected because buds at the base of the culm have higher amounts of lignin (Li Quing et al. 2022). Increased lignin and availability of glucose for respiration in the buds at the base of the culm may make them slower at sprouting. Ethylene may help the process of degradation of sucrose into glucose (Accácio et al. 2021) and subsequent development of the aerial part. Translocation of nutrients inside the plant supports shoot and root development in the initial phase (Schenato et al. 2007). Nutrient availability can be increased through better root development, allowing access to nutrients at a greater soil depth and increased water absorption. This may explain why buds from the base of the culm showed increased root growth with application of ethephon.

Conclusions

The position of the buds on the culm and the application of ethephon on buds positively influenced gaseous exchange via stomata and shoot and root growth. Buds immersed for 1 min in a solution of 0.650 L ethephon/ha showed a higher, CO₂ assimilation rate, transpiration and stomatal conductance. Based on these results, it is recommended to use buds originating from the base of the BRS Capiaçú culm treated with ephethon for the propagation of the crop.

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(Note of the editors: All hyperlinks were verified 17 September 2025).

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