




Biometric and morphological adjustments of *Eugenia dysenterica* DC. seedlings in response to increased soil water deficit

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ABSTRACT. Water stress can affect all aspects of plant growth and development, compromising its productive potential. The cultivation of fruit species native to the Cerrado and the study of their behavior under conditions of low water availability are necessary activities, in view of their socioeconomic and environmental potential. This study was conducted to evaluate the effect of water deficit on growth and biomass production of the cagaita tree (*Eugenia dysenterica* DC.), a species endemic to the Cerrado (Brazilian Savannah). The experiment was performed in a greenhouse for 120 days in a randomized block design (RBD), with a double factorial arrangement (2x5), corresponding to two cagaita mother plants and five water conditions. We evaluated the growth and leaf, stem and root functional characteristics. The most severe water deficit conditions reduced shoot growth, number of leaves, production of biomass and dry matter and increased the specific and per plant mass root length of cagaita plants. Deficit extension had negative effects on its growth and development. There was a statistical difference between mother plants, with mother plant 2 being more resistant to drought. The application of a severe or longer water deficit affected shoot growth and reduced the appearance of new leaves. The stress caused by water deficiency allowed an increase in root functional characteristics, supporting the hypothesis that plants under adverse conditions focus on higher biomass production and convert a higher amount of dry matter into the roots. Leaf area did not show to be a functional characteristic, explaining the stress effects in *E. dysenterica* DC plants.

Keywords: cerrado biome; dry matter; leaf area; root/shoot ratio; soil moisture.

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Introduction

Water stress affects several aspects of plant growth and development and has an influence on elongation and cell differentiation, reduction in cellular turgor and leaf area and on production and translocation of photoassimilates to new growth areas (Farooqi et al., 2020). These effects are observed with the occurrence of water stress, due to loss of cell turgor and dehydration, resulting from the impossibility of cells to perform their normal functions (Rahdari & Hoseini, 2012). The final response of plants to stress is manifested through the reduction in production and allocation of dry matter, especially in the shoot, by influencing CO₂ and O₂ exchange and carbon balance (Quintero-Vallejo, Pena-Claros, Bongers, Toledo, & Poorter, 2015; Li, Kristiansen, Rosenqvist, & Liu, 2019).

Several regions of the planet are currently prone to climate change, highlighted mainly in tropical ecosystems such as the Cerrado (Brazilian Savannah) (Jia et al., 2019), considered a world's biodiversity hotspot (Ballesteros-Mejia, Lima, & Collevatti, 2018). These climate changes have caused a reduction in rainfall levels and increased evapotranspiration (Collins et al., 2013; Lindner et al., 2014), heat waves and drought episodes (Li et al., 2019). Physiologically, most fruit species are classified as C3 plants, with high photosynthetic rates and high transpiration (Aref et al., 2014; Taiz, Zeiger, Møller, & Murphy, 2017). Therefore, the reduction in water availability causing water stress can directly affect its development and production.

Water stress can be defined as the effect of the deficit of water in the soil for the plants, characterized by a condition of physiological adversity in which the plant does not express its genetic potential, reducing crop yield (Taiz et al., 2017). The degree of water deficiency of a plant can be assessed by means of leaf variables, such as leaf water potential, as well as the measurement of transpiration and photosynthetic rates. Thus, characterizing the effect of water deficit in the soil and the consequent stress caused on the plants becomes increasingly necessary to indicate more tolerant species/genotypes with productive potential for regions strongly affected by this abiotic stress.

In early stages, plant growth is associated with increases in the allocation of stem and leaf biomass, specific leaf area and leaf area ratio, which are characteristics that improve light capture and carbon gain (Quintero-Vallejo et al., 2015). However, for fruit species of the Cerrado, there is a greater investment in the root system throughout their lifecycle with larger soil exploration. This depends on the water storage capacity in the soil and how much the species can extract from the root zone without significantly affecting the transpiratory flow (Aref et al., 2014).

Cerrado fruit plants can have a great economic potential in the food sector through fresh consumption or processed products such as juice pulp, ice cream, jams, jellies, liqueurs and wine, in addition to their medical use, since they are also a source of vitamin C, A and folates (Guedes et al., 2017; Silva et al., 2017; Mancianti & Ebani, 2020). Among the various plants native to the Cerrado, which have potential for traditional agricultural production, it is possible to highlight the cagaiteira, *Eugenia dysenterica* DC. – Myrtaceae. This species has value for fruit production, ornamentation, honey production and other activities such as cork extraction, small civil construction, charcoal manufacture, use of its bark in tanneries and medicinal use (Sousa, Camilo, & Vera, 2018).

The cagaiteira is recognized for its rusticity and ability to withstand long periods of drought (Oliveira et al., 2019), presenting little production alternation. Furthermore, it has great ability to survive in environments with low soil fertility, that is, poor in organic matter, with high acidity and in the presence of toxic aluminum (Rodrigues et al., 2019). Such attributes give this species a great environmental socioeconomic potential, being useful for reforestation and recovery of degraded areas (Pellizzaro et al., 2017; Oliveira et al., 2019), extractivism and commercial production in Savannah / Cerrado areas (Araujo, Neri-Numa, Paulo Farias, Cunha, & Pastore, 2019).

Due to anthropic pressure, many species native to the Cerrado are at risk of extinction. However, despite the fact that cagaiteira seeds have satisfactory germination, their initial development is quite slow, making seedling production in nurseries an activity still incipient in the country (Souza, Naves, Carneiro, Leandro, & Borges, 2002). Moreover, their survival in the field is poorly studied. Such facts, allied to the climate changes predicted for the century, arises the need and the production of seedlings with capacity to survive in environments with stress.

By stimulating development under different water conditions, the plants of *E. dysenterica* DC. develop better in conditions without water restriction in the soil? Evaluating such changes in plant characteristics associated with soil water availability, provides a unique opportunity to understand the responses of this species to possible short or prolonged droughts in nature, because of its perpetuation in the Cerrado biome. Given the above, the hypotheses for this study are: i) plants grown without water limitation, at pot capacity (PC), with no stress would have increased shoot growth compared to plants conducted in conditions with some level of water deficit; ii) limiting soil water conditions are capable of leading to harmful morphological changes in the early stages of growth of cagaita plants, despite their recognized resistance to drought, and iii) plants cultivated under limiting water conditions would have higher biomass allocation to the roots, greater root/shoot *ratio* (R/S) and would produce roots with a greater specific root length and per plant mass to improve the absorption of water and nutrients.

In this context, the objective of this study was to evaluate how different soil water availability conditions affect growth, biomass allocation and morphological characteristics of this Cerrado species and determine its response to the soil water condition.

Material and methods

Study site and experimental design

The experiment was conducted in the central region of the state of Minas Gerais, Brazil, at 19°28' S, 44°15' W and 732 m altitude, in pots within a greenhouse with controlled temperature and relative

humidity, during the hottest hours of the day with a variation of $25 \pm 2^\circ\text{C}$ and $65 \pm 5\%$, respectively. There was no irrigation system installed. The experimental design was in randomized blocks (RBD), with a 2×5 factorial arrangement, corresponding to two mother plants and five soil water content condition treatments. Control (without deficit), semi-moderate deficit, moderate deficit, severe deficit and permanent wilting point (PWP), according to the nomenclature proposed by Nascimento, Nogueira, Silva, and Silva (2011), were maintained at 100% of pot capacity (PC), 85% PC, 70% PC, 55% PC, and PWP (51% PC), respectively. There were four replicates for each treatment. A fixed interval of 15% of the PC from one water treatment to another was established, assuming that the lowest assigned value (55% PC) was higher than the water content at the permanent wilting point (PWP), which was previously known through the determination of the soil water retention curve (SWRC) in a previous study (Campos et al., 2019). Each plot consisted of four polyethylene pots filled with 3.5 kg of soil and containing one cagaita tree plant per pot, totaling 40 experimental plots and 160 pots (sample units).

Mother plants and collection site

Ripe fruits from two *E. dysenterica* DC. mother plants, from the same study location, were collected in the fruiting period (October), manually pulped and their seeds were washed in water and placed to dry in the shade. The plants were obtained through heterogeneous seeds, from sowing in 290 cm^3 -tubes containing peat substrate and maintained with intermittent irrigation in a seedling nursery.

Soil and greenhouse experiment

Emergence occurred 42 days after sowing (42nd DAS). Two months after seedling emergence (60th DAE), they were transplanted to plastic pots, with four base holes, each containing 3.5 kg of Dystrophic Red Yellow Latosol (Embrapa, 2013) or dystrophic Haplustox (Soil Survey Staff, 2014). This soil is representative of the Cerrado region and was collected at a depth of 0-20 cm, proceeding from the study location (Table 1).

Table 1. Fertility analysis of soil collected in the Cerrado area, in the central region of Minas Gerais State - Brazil, used in the pot experiment under greenhouse conditions.

	pH	P	K	S	Ca/Mg	Ca	Mg	Al	H+Al
H ₂ O	CaCl ₂	-----	mg dm ⁻³	-----	ratio	-----	cmol _c dm ⁻³	-----	-----
5.7	5.2	2.9	43.7	2.9	11	2.2	0.2	0.1	4.6
B	Zn	Fe	Mn	Cu	Total CEC *	SOM *	TOC *	V *	M *
	-----	mg dm ⁻³	-----	-----	cmol _c dm ⁻³	dag kg ⁻¹	-----	%	-----
0.1	0.9	62.9	37.1	0.9	7.1	1.4	0.8	35	4

*Total CEC: Total Cation Exchange Capacity; SOM: Soil Organic Matter; TOC: Total Organic Carbon; V: Base Saturation; m: Aluminum saturation.

After transplanting, magnesium was corrected in the soil, with magnesium sulfate application (MgSO_4), providing $48.6\text{ mg of Mg dm}^{-3}$ soil in order to reduce the Ca/Mg ratio of the soil, which was beyond acceptable limits. In addition, two cover fertilizations were applied at 125 and 175 DAT (days after transplanting), using Castellane and Araújo (1995) nutrient solution. The pH was monitored at application time with a portable pH meter and adjusted to 5.5 ± 0.5 using NaOH or HCl 0.1 mol L^{-1} solution. Electrical conductivity of the nutrient solution was stabilized with a portable conductivity meter, keeping it under 2.4 dS m^{-1} , as indicated by Távara, Ferreira, and Hernandez (2001) for Myrtaceae plants; the water used as a solvent was under 0.3 dS m^{-1} .

Determination of water conditions

In a previous study (Campos et al., 2019), the SWRC was determined in the laboratory, according to Klute (1986), and it was modeled using the Van Genuchten (1980) model, with Mualem restriction [$m = 1 (1/n)$], through the RETC software (Van Genuchten, Leij, & Yates 1991). The inflection point (IP) of the SWRC was used as the water content ($\omega - \text{g g}^{-1}$) at field capacity (FCIP), determined according to Dexter (2004) and Silva, Silva, Oliveira, Ferreira, and Serafim (2014), besides the matric potential (Ψ_{mIP}). Furthermore, from the SWRC, the water content at field capacity was estimated at -6 kPa (FC_6). From the water content of the PWP, we calculated the available water capacity (AWC) at 6 kPa (AWC_6) and at IP (AWC_{IP}) (Table 2).

After 90 days of acclimatization (150th DAE) and maintenance of the plants under PC, water treatments were differentiated in the greenhouse with controlled environment and without an installed irrigation system, starting the experimental period that ran for 120 days (from the 150th to the 270th DAE).

Table 2. Physical analysis of soil collected in the Cerrado area, in the central region of Minas Gerais State - Brazil, used in the pot experiment under greenhouse conditions.

Bulk Density g cm ⁻³	TP m ³ m ⁻³	Granulometric Composition			Soil Textural Classification ¹		
		Sand	Silt	Clay	Very clayey		
1.03	0.677	140	160	700			
Initial Soil Moisture	PC ⁴	FC ₆	FC _{IP} ³	PWP ⁵	Ψ _{mIP} ²	AWC ₆	AWC _{IP} ³
		ω (g g ⁻¹) ^{2,3}			kPa	m ³ m ⁻³	
0.324	0.51	0.465	0.515	0.258	3.8	0.207	0.257

*TP: Porosity; ω: Water Content (mass of water per mass of dry soil); PC: Pot Capacity; FC₆: Field Capacity estimated at -6 kPa; FC_{IP}: Field Capacity estimated at the Inflection Point of SWRC; PWP: Permanent Wilting Point estimated at -1500 kPa; Ψ_{mIP}: Matrix Potential at the Inflection Point of SWRC; AWC₆: Available Water Capacity between FC₆ and PWP; AWC_{IP}: Available Water Capacity between FC_{IP} and PWP.¹(Embrapa, 2013); ²(Campos et al., 2015); ³(Dexter, 2004; Silva et al., 2014); ⁴(Souza et al., 2000); ⁵(Klute, 1986).

Pot capacity (PC) is a direct method of determining field capacity (FC) for experiments with potted plants, representing a soil water condition in which there are no limitations of water availability for the plant, proposed by Souza et al. (2000). This method is adopted as being the water content retained by the soil after saturation and consequent gravity action, until the cessation of drainage. For PC determination, four pots were used, considering the following equation: $WM_{PC} = DM \times (1 + \omega_{PC})$, where, WM_{PC} is the wet mass in the PC obtained by the average of the weighing of the four pots, in kg, according to Souza, Oliveira, Silva, and Amorim Neto (2000); DM is the dry mass, in kg, estimated for the pots (N = 160) and ω_{PC} is the soil moisture in the PC in g g⁻¹, obtained in the laboratory by the drying oven standard gravimetric method (Embrapa, 2011). Comparisons of FC and PC estimation for our soil shows high correlation between them. More detail can be found in Campos et al. (2019).

After estimating the dry mass, the wet mass of each pot (WM) in each treatment was defined by the % of the ω_{PC} assigned to each treatment (100%, 85%, 70%, 55%, and PWP). In this case, as DM was constant, when ω_{PC} varied in a respective treatment, WM also varied for the same treatment. For the determination of ω_{PC} , 24 samples of 50 g of soil were collected from the four pots used to obtain the PC and, from each pot, three surface samples and three samples in the deepest region were removed. Subsequently, 10 g were weighed from each sample and dried in a drying and sterilizing oven at 105°C, until constant weight (Embrapa, 2011), obtaining an average value of 0.510 g g⁻¹ (standard deviation: 3.653). To define the initial soil moisture of the experiment, five samples were used and the same method was applied (Embrapa, 2011), obtaining a mean value of 32.42% or 0.324 g g⁻¹ (standard deviation: 0.5358) (Table 2).

As one of the treatments, the water condition under PWP was adopted to evaluate plant response when exposed to a severe drought condition. Soil moisture under the PWP was estimated in the laboratory using Richards' extractor, using samples collected from the same soil of the experiment, through water retained in the water potential (Ψ) of -1.5 MPa (Klute, 1986). The samples were then dried at 105°C in a drying oven to quantify the ω, associated with Ψ established for the PWP. The value of ω under the PWP was determined as 0.258 g g⁻¹, corresponding to 51% of PC (Table 2).

Irrigation management

Water treatments were maintained by weighing the pots on alternate days and replacing the volume of water lost by evapotranspiration, until reaching the WM corresponding to each water condition, using a Welmy precision scale with 15 kg capacity and a graduated 0.5-L beaker. In this methodology, the weight gain of the plants over time after transplanting is disregarded (Souza et al., 2000).

Biometric evaluations

Plant growth was evaluated throughout the experimental period, where shoot height (SH), in cm, and stem diameter (lap region) (SD), in mm, were measured each month using, respectively, a graduated ruler and a 150-mm digital caliper (Lee Tools 684132). The ratio between SH and SD (SH/SD) and the number of fully expanded leaves (NL) of each plant were determined. The SH and SD assessment was based on an indelible mark made on the stem base. The first biometric evaluation was performed on the first day after differentiation of water treatments, followed by evaluations every 30 days, totaling five evaluations for the Season factor.

Phytotechnical evaluations

At the end of the experimental period, the plants were collected and separated into leaves, stem and root. The green mass (GM) in g, was measured. The *E. dysenterica* DC. seedling leaf areas (LA) were estimated according to the methodology proposed by Oga and Fonseca (1994) through a leaf area meter (CID Inc., CI-202). The LA value was considered in m² from two methodologies studied. The amount of dry matter (DM) in kg was determined after drying in an oven with air circulation and renewal (65°C) for 72 hours, as well as the percentage of green mass reduction to dry matter (GMredDM).

From DM and LA values and total dry matter (TDM), leaf dry matter (LDM), stem dry matter (SDM) and root dry matter (RDM) in g, the biomass allocation in leaves (LBA= LDM/TDM), stems (SBA=SDM/TDM) and roots (RBA=RDM/TDM), respectively, in g g⁻¹; leaf area ratio (LAR=LA/TDM), in m² kg⁻¹ of DM; specific leaf area (SLA=LA/LDM), in m² kg⁻¹ of DM and the root/shoot ratio (R/S=RDM/(LDM+SDM)) were calculated (Benincasa 1988).

We also calculated the dry matter content in leaves, stems and roots (LDMC, SDMC and RDMC, respectively in g g⁻¹) by the ratio between DM and GM of each plant section; the specific root length (SRL in m kg⁻¹), by the ratio between root length of the main root (RL, in m) and RDM and the root length per plant mass (RLPM, in m kg⁻¹), as RL over TDM (Quintero-Vallejo et al., 2015).

Statistical analysis

The information about growth and biomass allocation were subjected to the *a priori* test of Shapiro-Wilk (normality test with 5% significance) for further analysis of variance (ANOVA) in order to verify the effect of treatments and finally, the means were compared by testing *a posteriori* with Scott-Knott, at 5% probability, using the R software, version 3.2.1, ExpDes package (Ferreira, Cavalcanti, & Nogueira, 2014). For the growth variables, a triple factorial (2 x 5 x 5) was adopted, where the factor Mother Plant had two levels; and the factors Water Condition and Season had both five levels, corresponding to each date of biometric evaluation performed during the 120 days of experiment. As for variables of biomass allocation, dry matter production, leaf area and root length, a double factorial arrangement (2 x 5) was used with the factors Mother Plant and Water Condition, respectively, due to their acquisition having been at the end of the experiment, thus excluding the Season factor analysis.

Results

Plant Growth Evaluation

Statistical analyses showed that there was no significant interaction between the three factors (Mother Plant, Season and Water Condition), and there was no interaction between Mother Plant x Season for SH, only checking the interactions between Water Condition x Season (Table 3) and Water Condition x Mother Plant (Table 4), for the same variable. For SH, a significant difference was observed from the 3rd Season, showing that the effect of water deficit for the PWP only occurred after the 60th day after the differentiation (DAD) of water treatments (Table 3).

The best shoot growth response of plants was the control (100% PC), which occurred in the seasons during 90 and 120 DAD, corroborating hypothesis one in this study. The plants under optimal growth conditions (100% PC) reached 0.091 m, on average, of SH, at 270 DAE or the last DAD (120th day). It was also noted that the water condition of 55% PC provided a limiting result for shoot growth in the last Season (120th day), similar to that observed in PMP, for the same season (Table 3).

These results demonstrate that *E. dysenterica* DC. plants show sensitivity to the most severe deficit conditions at an early growth stage, and that a drought of approximately 60 days is sufficient to cause changes in growth in height. However, the most important result is that cagaita plants continue to grow over time, even under more severe water deficit situations (55% PC and PWP) (Table 3).

Mother plant 1 had, on average, the greatest height when subjected to both water conditions with increased amounts of water in the soil. However, mother plant 2 did not present this result, with the highest SH averages at 100% and 70% PC, respectively (Table 4).

Regarding stem diameter (SD), there was only interaction for Water Condition x Mother Plant (p-value < 0.001). Similar SD values for all water conditions were obtained, except for the PWP, mother plant 2. Mother plant 1 presented statistical differences as early as the second best water condition, only 15% below the PC (Table 4).

Table 3. Average shoot height (SH), in cm, number of leaves (NL) and ratio between SH and SD (SH/SD) of different *Eugenia dysenterica* DC. plants grown under different water conditions for 120 days).

Season	Water Condition				
	100% PC	85% PC	70% PC	55% PC	51% PC (PWP)
SH – cm					
0	4.98 dA	5.32 dA	5.26 cA	5.03 cA	4.80 bA
30	5.69 cA	5.59 dA	5.92 bA	5.58 bA	5.22 bA
60	5.85 cA	6.03 cA	6.14 bA	5.91 aA	5.31 bB
90	7.39 bA	6.82 bB	6.58 aB	6.10 aC	5.55 aD
120	9.09 aB	8.26 aB	7.12 aC	6.51 aD	5.97 aD
NL					
0	3.56 bA	3.90 bA	3.78 aA	3.93 aA	3.87 aA
30	3.81 bA	3.90 bA	3.87 aA	4.25 aA	4.03 aA
60	3.94 bA	4.09 bA	4.21 aA	4.22 aA	2.64 bB
90	4.62 bA	4.59 bA	4.33 aA	4.12 aA	2.33 bB
120	5.78 aA	5.40 aA	4.28 aB	3.90 aB	2.17 bC
SH/SD m m ⁻¹					
0	35.66 cA	39.62 bA	39.83 aA	38.92 aA	37.68 aA
30	40.16 bA	40.66 bA	44.10 aA	42.54 aA	39.71 aA
60	39.63 bA	42.38 bA	44.25 aA	41.24 aA	39.18 aA
90	43.84 bA	41.52 bB	44.78 aA	38.52 aB	39.03 aB
120	52.65 aA	47.31 aB	45.42 aB	41.67 aC	38.69 aC

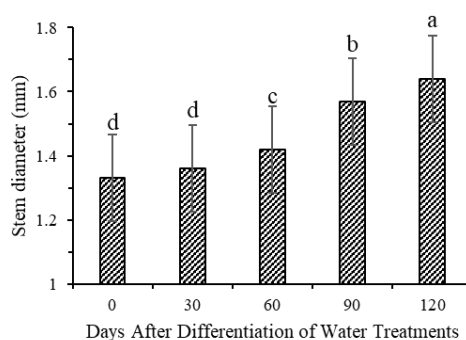
Means followed by the same lowercase letter in the column and uppercase letter in the row do not differ for each variable by the Scott-Knott test, $p < 0.05$

Table 4. Average shoot height (SH), in m, stem diameter (SD) in mm, ratio between SH and SD (SH/SD), in m m⁻¹, number of leaves (NL) and stem dry matter content (SDMC), in g g⁻¹, of different *Eugenia dysenterica* DC. plants grown under different water conditions for 120 days.

Mother Plant	Water Condition				
	100% PC	85% PC	70% PC	55% PC	51% PC (PWP)
SH – cm					
1	6.61 aA	6.95 aA	5.96 bB	5.69 aB	5.23 aC
2	6.60 aA	5.86 bB	6.45 aA	5.97 aB	5.51 aC
SD – mm					
1	1.66 aA	1.56 aB	1.39 aC	1.42 bC	1.36 aC
2	1.46 bA	1.46 bA	1.47 aA	1.54 aA	1.37 aB
SH/SD					
1	39.9 bB	44.6 aA	42.9 aA	40.5 aB	38.6 aB
2	44.9 aA	39.9 bB	44.4 aA	39.4 aB	40.3 aB
NL					
1	4.54 aA	4.62 aA	3.55 bB	3.75 bB	3.31 aB
2	4.15 aA	4.13 aA	4.64 aA	4.42 aA	2.71 bB
SDMC – g g ⁻¹					
1	0.544 aA	0.640 aA	0.777 aA	0.679 aA	0.569 bA
2	0.600 aB	0.556 aB	0.599 aB	0.579 bA	0.940 aA

Means followed by the same lowercase letter in the column and uppercase letter in the row do not differ for each variable by the Scott-Knott test, $p < 0.05$

As there was no interaction between Water Condition x Season, the effect of Season (p -value < 0.001) was analyzed separately and it was observed that SD increased over the evaluated periods, with statistical differences from the 60th DAD, showing that its initial secondary growth is slow. On average, the increase in SD for 120 days of maintenance in the water treatment represented 0.31 mm or a gain of 23.19% (Figure 1).

**Figure 1.** Average stem diameter (SD), in mm, of *Eugenia dysenterica* DC. plants grown under different water conditions for 120 days.

For SH/SD, as with SH, there was no significant interaction between the three factors and no interaction occurred between Mother Plant x Season. Only interactions between Water Condition x Season and Water Condition x Mother Plant (Tables 3 and 4) were verified.

There were higher SH/SD ratio averages for the last season (120 DAD) and for the control (100% PC), reflecting the key role of water for the investment in shoot height and, at the same time, stabilizing stem diameter thickness (Table 3).

In relation to the interaction between Water Condition x Mother Plant, mother plant 2 was superior only in the condition with the highest hydration level (100% PC) (Table 4), a fact that can be understood as a drought strategy, that is, a greater vegetative investment and primary growth only when under optimal water availability conditions. Restriction conditions of the water supply are expected to lower growth in SH (primary growth) and stabilization of SD (secondary growth), as occurred in plant 2 at 85% PC (Table 4).

The variable number of leaves (NL) showed interaction only between Water Conditions x Season and Water Conditions x Mother Plant (p -value < 0.001). Water stress affected the issuance of new leaves in 3.6 and 2.3 leaves, respectively, when compared to the PWP at 100% PC, at 120 and 90 DAD. It was demonstrated that water deficit induced a reduced emission of new leaves in *E. dysenterica* DC. seedlings after four months of assays, already showing statistical differences under 70% PC (Table 3).

In this study, we observed a progressive increase in leaf abscission in some pots, reaching 100% (M. Campos, personal observation) for plants subjected to the most intense water treatments (55% PC and PWP). Under the water condition with the highest water restriction (PWP), there was a reduction in the number of leaves (NL) from 30 to 60 DAD and, under other water conditions, there were no verified reductions in this variable over the evaluation period (Table 3).

Plant 2 was more resistant to leaf abscission in relation to Plant 1, once the plants reduced their number of leaves only in the PWP. In Plant 1, there was a reduction in number of leaves of 1.07 leaves on average, under 70% PC (Table 4); thus, deficit level influences how certain mother plants resist to leaf abscission.

Biomass production and plant response to drought

No significant interaction was verified between Water Condition x Mother Plant for biomass production, dry matter partitioning, leaf growth and root growth; the results were presented by factor (Table 5 and 6). The SDMC was the only variable of dry matter partitioning that had interaction between the two factors (Table 4). There were statistical differences for the Water Condition factor for the variables GM, TDM, LDM, SDM, RDM, LBA, SBA, and RBA (Table 5).

We verified a higher GM for seedlings grown under water conditions with higher soil water levels (100 and 85% PC), as well as for the water condition of 55% PC. The reduction in GM, comparing the treatments 100% PC and PWP, represented, on average, more than 50% biomass loss. The plants were very sensitive, as to biomass production and dry matter under the PWP, and the worst results were obtained for GM, TDM and LDM, with mass losses above 50%, when compared to the control (Table 5).

Severe water stress, characterized by the water condition of 51% PC (PWP), provided less LBA and increased SBA and RBA of *E. dysenterica* DC plants (Table 5). This result corroborates hypothesis 3 of this study. Regarding Mother Plant, there were significant differences only for LDM and LBA and, in both cases, mother plant 1 was higher than 2 (Table 5). This increased investment in leaves may not be an interesting strategy for a water stress condition. However, it may be physiologically beneficial under optimum water conditions, since there are more photosynthetic apparatus, thus resulting in a higher gain in plant growth. Therefore, future studies are necessary to investigate this trend.

SDMC showed interaction between the two factors and a higher accumulation in mother plant 2 in relation to mother plant 1, when compared under the most unfavorable water condition, the PWP. The higher investment in the stem for this situation could be assigned as a response to water deficit, but it is not possible to say that this result represents a characteristic that confers drought resistance. Mother plant 1 showed no statistical difference between the water conditions, while Plant 2 obtained higher SDMC values under the PWP (Table 4).

There was a significant difference for Water Condition in LAR, RL, SRL and RLPM. For Mother Plant, there was no statistical difference between the averages for any variables (Table 6). The LAR was significantly higher at the PWP due to the reduction in TDM conversion observed in plants grown under this condition, with only 0.595 g of TDM (Table 5), since the LA was statistically similar for all water conditions (Table 6). These results demonstrated that the LA was not an interesting functional characteristic to explain the effect of water deficit in *E. dysenterica* DC. seedlings.

Table 5. Average biomass production and dry matter partition of different *Eugenia dysenterica* DC. plants grown under different soil water availability conditions after 120 days.

Water Condition	GMredDM (%)*	Green Mass (g)	Dry matter				R/S*	Biomass Allocation			Dry Matter Content	
			----- (g) -----					----- (g g ⁻¹) -----			--- (g g ⁻¹) ---	
			Total	Leaf	Stem	Root		Leaf	Stem	Root	Leaf	Root
100% PC	50.3 a*	2.568 a	1.248 a	0.310 a	0.126 a	0.813 a	1.865 a	0.248 a	0.101 b	0.651 b	0.633 a	0.475 a
85% PC	51.5 a	2.768 a	1.272 a	0.307 a	0.119 a	0.847 a	1.990 a	0.241 a	0.093 b	0.666 b	0.516 a	0.470 a
70% PC	46.7 a	1.838 b	0.934 b	0.182 b	0.097 a	0.656 b	2.356 a	0.194 a	0.104 b	0.702 a	0.503 a	0.511 a
55% PC	49.9 a	2.289 a	1.096 b	0.280 a	0.110 a	0.706 b	1.809 a	0.256 a	0.100 b	0.644 b	0.419 a	0.485 a
PWP *	49.1 a	1.213 c	0.595 c	0.112 b	0.069 b	0.415 c	2.302 a	0.188 b	0.115 a	0.697 a	0.342 a	0.476 a
Mother Plant												
1	49.4 a	2.236 a	1.082 a	0.268 a	0.106 a	0.708 a	1.893 a	0.213 a	0.113 a	0.674 a	0.492 a	0.484 a
2	49.6 a	2.033 a	0.976 a	0.208 b	0.102 a	0.666 a	2.150 a	0.177 b	0.132 a	0.699 a	0.472 a	0.483 a
CV (%)	10.3	25.3	22.9	37.3	20	22.2	62.6	23.5	28.6	6.69	38.8	9.75

^{*}Means followed by the same letter in the column do not differ by the Scott-Knott test, $P < 0.05$; The effect of each factor was analyzed separately; R/S: root/shoot ratio; GMredDM: percentage of green mass reduction to dry matter; PWP: Permanent Wilting Point estimated at -1500 kPa or 51% PC.

The RL was increased under 85 and 55% PC. However, a higher average of this variable under the greatest stress condition (PWP) was expected, since this was statistically similar to the average of 100 and 70% PC. Regarding SRL and RLPM, the expected under hypothesis 3 of this study was verified; values statistically higher for PWP compared to other water conditions (Table 6).

Table 6. Averages of the variables leaf area (LA); leaf area ratio (LAR); specific leaf area (SLA); root length (RL); specific root length (SRL) and root length per plant mass (RLPM) of different *Eugenia dysenterica* DC. plants grown under different soil water availability conditions after 120 days.

Water Condition	LA	LAR	SLA	RL	SRL	RLPM
	m ²	m ² kg ⁻¹ of DM	m ² kg ⁻¹ of DM	m	m kg ⁻¹ of DM	m kg ⁻¹ of DM
100% PC	0.00069 a	0.554 b	2.23 a	0.537 b	741 b	472 c
85% PC	0.00068 a	0.536 b	2.22 a	0.669 a	834 b	548 c
70% PC	0.00067 a	0.715 b	3.67 a	0.503 b	892 b	636 b
55% PC	0.00069 a	0.632 b	2.47 a	0.711 a	1064 a	706 b
51% - PWP	0.00058 a	0.972 a	5.17 a	0.427 b	1229 a	905 a
Mother Plant						
1	0.00069 a	0.637 a	2.569 a	0.591 a	958 a	646 a
2	0.00064 a	0.651 a	3.055 a	0.548 a	946 a	660 a
CV (%)	21.4	51.7	53.5	26.6	21.6	23

Means followed by the same lowercase letter in the column do not by the Scott-Knott test, $p < 0.05$.

Discussion

Cagaita plants adapted their biometric and morphological characteristics in response to increased water deficit in the soil. However, this behavior did not occur in any variables analyzed. For the variables that did not show significant differences, the effect seems to have been determined by the intrinsic characteristics of this species. This lack of significant differences for some variables seems to have been determined by the intrinsic characteristics of this species. It was noticeable that the most severe drought in the soil of the experiment pots caused limiting results to the shoot growth of *E. dysenterica* DC., in its early stages, corroborating hypothesis 2 of this study (very limiting soil water conditions are capable of leading to harmful morphological changes in the early growth stages of cagaita plants, despite their recognized resistance to drought), elucidating the dependence of this species, under conditions of natural environment, regarding rainfall in the rainy season.

Cagaita seedlings are found around the mother plants in March, with natural seed germination between December and January, shortly after fruit fall, showing that there is no natural impediment to germination, but the germinative power cannot last beyond three months under low soil water content conditions (Martinotto et al., 2007; Pellizzaro et al., 2017). Therefore, in the years with periods of short summers in full rainy season in the Cerrado, especially with an incidence during the months of natural germination of *E. dysenterica* DC., there will not be propagation of this species, reducing its chance to perpetuate in the natural

environment (Bucci et al., 2008). Furthermore, if short summers are between March and April, the end of the rainy season, there will hardly be seedling growth (Franco et al., 2005), explaining the importance of this study.

Plant growth evaluation

The slow initial growth of the plants in this study may be associated with several factors, such as characteristics intrinsic to the species itself (Franco et al., 2005) and it may also have occurred due to the container and size of the seeds used (Nietsche et al., 2004); the water condition applied to some plants (Trouet, Mukelabai, Verheyden, & Beeckman, 2012) or the difficulty in assimilating nutrients in the soil, despite topdressing fertilization being carried out with nutrient solution at 125 and 175 DAT.

In a similar study, Nascimento et al. (2011) found similar results for SD, where the SD of jatobá (*Hymenaea courbaril* L.) plants under water deficit was evaluated weekly, with a statistical difference from the 49th DAD. However, when comparing both species typical of the Cerrado regarding radial growth, *E. dysenterica* DC. proved to be more resistant to deficit than *H. courbaril* L., once it presented a statistical difference only after 60 DAD.

In this study, there were statistical differences between the two groups of mother plants evaluated, which were obtained sexually using seeds with size heterogeneity. The mother plants from seeds of different sizes present a better initial growth when the seeds have the greatest volume (Nietsche et al., 2004). The maximum SD value found for both plants at 120 DAD was 1.66 mm (Table 4), as expected for this species at this age (270 DAE), similarly to the values obtained by Mota et al. (2018).

The tree species under growth tend to increase vertically and stabilize radially; thus, a temporal increase in the SH/SD ratio is expected when there is better nutritional status in the plant (Trouet et al., 2012; Marcati et al., 2016; Mota et al., 2018). According to Fonseca, Valéri, Miglioranza, Fonseca, and Couto (2002), in a study on shading with *Trema micrantha* L. seedlings, a pioneer species that have fast growth and is unsuitable to humid environments, showed a significant linear increase in the ratio, explained by the increase in shoot height and reduction in lap diameter (Marcati et al., 2016).

In order to obtain a quality seedling standard, the values for this ratio must be less than 8 cm of shoot per mm of lap diameter, or less than 80 mm mm⁻¹ (Carneiro, 1995). However, this ratio must be used in conjunction with other parameters in the determination of the best quality standard of seedlings suitable for transplanting. The results of the SH/SD ratio, for this study, were within the recommended values for quality seedling standards (Carneiro, 1995), regardless of Water Condition, Mother Plant and/or Season analyzed (Table 3).

One of the most striking effects of water deficiency on plants is assigned to a reduction in leaf development, with less emission of new leaves, a reduction or stabilization in leaf area and increase in the abscission of old leaves (Franco et al., 2005; Bucci et al., 2008; Carnicer et al., 2011; Ogaya, Barbeta, Başnou, & Peñuelas, 2015). Not only does water restriction limit the size, but also the number of leaves, as the number and the growth rate of the branches are reduced (Marcati et al., 2016; Taiz et al., 2017). In addition, water availability in the soil is one of the factors that control exchange activity in plants and regulate their growth (Lara, Silva, Nogueira, & Marcati, 2017).

There was a reduction in the NL under the most intense water conditions and an increase for conditions with the lowest water restrictions (Table 3). The same behavior of *E. dysenterica* DC. seedlings in the north of Minas Gerais was observed by Nietsche et al. (2004). Other authors, such as Nascimento et al. (2011), in jatobá seedlings; Figueirôa, Barbosa, and Simabukuro (2004), in *Myracrodruon urundeuva* young plants; Carvalho, Santana, Pereira, Pereira, and Queiroz (2004), in eggplant and Tognon, Petry, and Cuquel (2012), in *Ipomoea cairica* L. SWEET plants have found similar results with several species submitted to water deficit.

Eugenia dysenterica DC. is classified as a deciduous species, with old leaf abscission in the dry season and renewal of foliage at the end of the dry season and the beginning of the rainy season (Brasil, 2008). However, when in early growth stages, the NL was evaluated, a reduction over time was considered as a kind of resistance strategy. Therefore, for the particular situation of this experiment with plants between five and nine months old, still unsuited to final transplanting, the higher leaf abscission was an immediate response to the most severe drought, a way to withstand water deficit. By observing the phenological behavior of *E. dysenterica* DC. trees under natural conditions of Cerrado and deciduous conditions during the dry season, this was seen as a resistance strategy. It has an effect similar to the behavior observed in early growth stages, when the plant gives up a greater photosynthetic apparatus for a carbon gain (Quintero-Vallejo et al., 2015) by development of the root system.

Biomass production and plant response to drought

Similarly to the biometric evaluations, the different soil water levels were representative to affect the total GM production, besides shoot and root dry matter (SDM+LDM); PWP conditioned the worst results for these variables.

Almeida, Soares, Castro, Vieira, and Gajego (2005) obtained values similar to those of this study, using plants with unrestricted water conditions, corroborating higher dry matter production by the plants submitted to 85 and 100% PC. The reduction in DM production is more visible in plants whose exposure to stress is more prolonged (Almeida, Tezara, & Herrera, 2016).

Biomass fractions of plant sections describe how they allocate biomass for light interception in tissues, in the case of leaves, or how they allocate biomass to capture nutrients, in the tissues by the roots (Markesteijn & Poorter, 2009). Nascimento et al. (2011) found no significant differences for allocation in the three sections (leaves, stem and root) of jatobá seedlings submitted to 100, 75, 50 and 25% PC. Sánchez-Costa, Poyatos, and Sabaté (2015) pointed out that one of the possible explanations for such similarity in the biomass allocation of some forest species occurs due to the low photosynthetic and transpiration rates presented in studies about growth of tropical and Mediterranean tree species, respectively, submitted to water deficit.

In this study with *E. dysenterica* DC., there was greater mass accumulation in the roots and stem, at the PWP (Table 5). This can be understood as a strategy of this plant to not invest in leaves when water supply is lower and there is a decrease in transpiration (Vieira, Graças Silva, Moro, & Laura, 2017), as well as a response to a certain adverse situation, as occurs at the PWP, concentrating the accumulation of reserves in the roots and stem. This fact allows to infer a probable response to drought and/or short summers that occur in the natural environment of Cerrado for young *E. dysenterica* DC. plants (Callado, Roig, Tomazello-Filho, & Barros, 2013; Vieira et al., 2017). These results confirm that the PWP, an unfavorable water condition for plant growth, should not be taken as a lower soil water limit and, therefore, there is a soil water content above this value, under which the plant is no longer able to supply its evaporative demand.

Regarding the R/S ratio, there were no significant differences between the treatments applied (Table 5). One of the reasons for this behavior may be related to the balance or slowness in the growth between root and shoot of Cerrado tree species (Callado et al., 2013; Marcati et al., 2016; Vieira et al., 2017). There is no need to invest exclusively in the deepening roots, a fact corroborated by Nascimento et al. (2011), studying *H. courbaril* L. seedlings submitted to different soil water availabilities.

The SDMC indicates the tenacity of the plant tissue, which can be understood as a good indicator of plant tissue longevity (Markesteijn & Poorter, 2009). The higher SDMC accumulation can be considered as a behavior of tolerance to adverse conditions (Sánchez-Costa et al., 2015), as was observed at the PWP for mother plant 2 in this study. Thus, it is possible to obtain seedlings of this mother plant with greater durability, at the tissue level, than those of mother plant 1.

The variables SLA and LAR indicate how plants efficiently invest in light interception (Markesteijn & Poorter, 2009). Higher LBA, in combination with higher SLA and LAR values, may increase the light capturing capacity of the leaf surface and lead to biomass growth (Westoby, Falster, Moles, Vesk, & Wright, 2002). Larcher (2006) complements that LA is related to leaf development and, consequently, the area of light capture, the photosynthetic process and the gas exchange are directly influenced by it. The plant LA may be related to relative humidity, being higher in humid environments and lower in arid environments (Figueirôa et al., 2004). Many plant species tend to decrease LA, an important xeromorphic characteristic against excessive water loss (Markesteijn & Poorter, 2009; Vieira et al., 2017).

In this study, there was no alteration of the LA regarding deficit (Table 6), a fact that may be related to the anatomical characteristics found in *E. dysenterica* DC. The presence of stomata on the abaxial face and a thick cuticle on the adaxial face were also observed for other species of the genus *Eugenia* (Esposito-Polesi, Rodrigues, & Almeida, 2011). These characteristics allow a protection for the plant against high temperatures, resulting in lower respiratory rates and, consequently, a water loss economy, as there is no need to reduce the LA.

The lower values observed for RL under 100% PC and at the PWP can be explained by the lower and higher water limitation for the plants, respectively; there is no need for *E. dysenterica* DC. plants to invest much in the roots and, therefore, to convert photoassimilates into shoot biomass, once the deficit was sufficiently intense to interrupt root growth (Table 6). This reflected in a lower investment in RDM and in the plant as a whole, in all the plants that were submitted to the conditions with less soil water availability, since the PWP condition is considered agronomically restrictive. Therefore, when analyzing RL separately, we have no support to assert that water availability influences it, and a broader approach is necessary with the inclusion of RDM and TDM variables.

SRL and RLPM measurements indicate how the biomass allocated to the root system can improve nutrient capture by increasing the absorption surface (Markesteijn & Poorter, 2009; Quintero-Vallejo et al., 2015). Therefore, higher mean values for these variables are expected in the most limiting water conditions (Table 5), with another response from cagaita plants to the low soil water availability, as the only alternative to seek some source of mineral nutrition or even water. Therefore, for large-scale commercial cultivation, the cagaiteira may have the potential to be irrigated with positive economic results, which is suggested as research in future studies.

Conclusion

Cagaita plants adjusted their biometric and morphological characteristics in response to increased soil water deficit. Our results suggest that the more severe and prolonged the drought, the more the shoot growth will be affected with the reduced appearance of new leaves. The stress caused by water deficiency led to an increase in root functional characteristics, supporting our hypothesis that plants under adverse conditions focus on higher biomass production and convert a greater amount of RDM. Contrary to our expectations, *Eugenia dysenterica* DC. plants have no modification of LA under water deficit situations and this variable, separately, was not ideal to explain the water stress.

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