# BIOMASS OF TWO Eucalyptus CLONES (E. grandis × E. urophylla) IRRIGATED WITH SALINE WATER

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ABSTRACT – Using saline water for irrigation relies on strategies that include selecting salt-tolerant cultivars and leaching salts away from zones close to the roots. A greenhouse experiment was carried out to assess early growth and biomass production of two *Eucalyptus* clones (*E. grandis* × *E. urophylla*), CO 865 and CO 1407, irrigated with saline water and under different leaching fractions. Treatments were laid out in a randomized block design and arranged in a  $4 \times 2 + 2$  factorial scheme: four leaching fractions (3, 10, 20, and 30 % of crop water demand for plants irrigated with saline water), two *Eucalyptus* clones (VCC 865 and CO 1407), and two additional treatments, one for each clone, conventionally irrigated with fresh water. The treatments were replicated four times. Measurements were made at 114 days after transplanting. Soil salinity decreased with increasing leaching fraction where VCC 865 was grown; however, leaf dry weight production was lower in treatments irrigated with saline water. Compared to fresh water-irrigated plants, irrigation with saline water resulted in lower: canopy diameter, leaf number, dry leaf mass, dry root mass, aerial parts dry mass, aerial part/ root ratio, and total plant dry weight. Overall, the VCC 865 *Eucalyptus* clone performed better under saline irrigation than CO 1407.

Keywords: Electrical conductivity; Eucalyptus spp; Tolerance.

# BIOMASSA DE DOIS CLONES DE Eucalyptus (E. grandis × E. urophylla) IRRIGADOS COM ÁGUA SALINA

RESUMO – O uso de água salina para irrigação depende de estratégias que incluem a seleção de cultivares tolerantes ao sal e a lixiviação de sais das zonas próximas às raízes. Um experimento em casa de vegetação foi realizado para avaliar o crescimento inicial e a produção de biomassa de dois clones de **Eucalyptus** (CO 865 e CO 1407), provenientes do cruzamento entre o **E. grandis** × **E. urophylla**, irrigados com água salina e sob diferentes frações de lixiviação. Os tratamentos foram organizados em blocos casualizados, com quatro repetições e arranjados em esquema fatorial  $4 \times 2 + 2$ , sendo quatro frações de lixiviação (3, 10, 20 e 30 % da demanda hídrica da cultura para plantas irrigadas com água salina), dois clones de **Eucalyptus** (VCC 865 e CO 1407), e dois tratamentos adicionais, um para cada clone, irrigados convencionalmente com água doce. As avaliações foram realizadas aos 114 dias após o transplante. A salinidade do solo diminuiu com o aumento da fração de lixiviação onde o clone VCC 865 foi cultivado, no entanto, a produção de massa seca das folhas foi menor nos tratamentos irrigados com água salina. Em comparação com as plantas irrigadas com água doce,



Revista Árvore 2022;46:e4612 http://dx.doi.org/10.1590/1806-908820220000012 a irrigação com água salina resultou em menores: diâmetro da copa, número de folhas, massa seca de folhas, massa seca de parte aérea, relação parte aérea/raiz e peso seco total da planta. No geral, o clone de **Eucalyptus** VCC 865 teve melhor desempenho sob irrigação salina do que o CO 1407.

Palavras-Chave: Condutividade Elétrica; Eucalyptus spp; Tolerância.

### **1. INTRODUCTION**

Farming faces a major challenge to meet the needs of a growing global population, expected to reach 9.8 billion by 2050 (FAO, 2017; Calicioglu et al., 2019). Major obstacles to accomplishing this goal are water scarcity and salinity (Deinlein et al., 2014; Gorji et al., 2020). High soil salinity affects over 20 % of arable lands around the world and this percentage is rising (Gupta and Huang, 2014). Soil salinity constrains plant production, especially in arid and semi-arid regions (Ashraf and Wu, 1994; Lopes and Klar, 2009; Leksungnoen and Andrivas, 2019; Yang et al., 2020) where evapotranspiration rates always exceed rainfall over a crop year (Hanin et al., 2016). Improper practices, such as overusing fertilizers and irrigation with saline water, may further increase soil salinity.

High salt levels in the soil not only negatively affect its fertility but also its physical properties (Mohamed et al., 2018). As infiltration and storage of water in the soil decrease, runoff and erosion increase (Daliakopoulos et al., 2016), which also has a negative impact on soil biodiversity (Singh, 2016). Therefore, soil salinity is an important abiotic stress that reduce plant growth and yield (Yasar et al., 2016; Afridi et al., 2019). In the soil, sodium chloride (NaCl) is a common salt that adversely affects plant growth through water stress and excessive uptake of Na<sup>+</sup> and Cl<sup>-</sup> (Tester and Devenport, 2003; Isayenkov and Maathuis, 2019). These ions also disturb the ionic balance within plant tissues, resulting in lower nutrient uptake; however, plant responses to salinity stress vary with plant genotype, as some species are tolerant to salinity and others are highly sensitive to it (Hanin et al., 2016).

With 5.6 million ha cultivated with *Eucalyptus*, Brazil is a major producer of pulp and wood (ABAF, 2015). Soil salinity affects many trees stand in Brazil, thus the identification of *Eucalyptus* species that are tolerant to high-salinity conditions is highly important because the *Eucalyptus* response to salinity varies across species or even across individuals of the same species (Daas-Ghrib et al., 2011; Bush et al., 2013). *E. camaldulensis* (Marcar, 1993; Rawat and Banerjee, 1998; Su et al., 2005) and *E. tereticornis* (Marcar, 1993; Sun and Dickinson, 1995; Tomar et al., 2003) have been reported to be tolerant to salinity, but there is little information on salinity stress in *E. pellita* (Mendonça et al., 2007). Specific adaptive traits of *Eucalyptus* species are important in breeding programs aimed at producing hybrids with desirable traits. For example, by crossing *E. grandis* with *E. camaldulensis*, breeders could produce a hybrid that grows fast and yields high quality wood, both traits inherited from *E. grandis*, and is tolerant to low water and nutrient supply, a trait inherited from *E. camaldulensis* (Pereira et al., 2019).

By reviewing the literature, little information about soil salinity-reducing techniques and/or irrigation management lacks for *Eucalyptus* species under saline water irrigation, as well as the response of different clones of the same *Eucalyptus* species under irrigation with saline water. We hypothesized that: (*i*) the early response of a *Eucalyptus* species to saline water irrigation depends on the clone; and (*ii*) leaching fractions alleviates salt-induced stress. The objectives were to evaluate vegetative growth and biomass production of two *Eucalyptus* clones, VCC 865 and CO 1407 (*E. grandis* × *E. urophylla*), irrigated with saline water and to identify the most suitable leaching fraction to offset salinity effects on the plants.

#### 2. MATERIALS AND METHODS

A greenhouse experiment was carried out at the Universidade Estadual do Sudoeste da Bahia, located in southwestern Bahia State, Brazil (14°51'LS, 40°50'LW, and 876 m altitude). The experiment was conducted using 50 L plastic containers, which served as miniature drainage lysimeters. The drainage system consisted of a hose measuring 40 cm in length and 16 mm in diameter attached to the bottom of the mini lysimeter. The other end of the hose was attached to a 2-L plastic bottle for collecting the drained water.

A eucalypt plant was grown in each mini lysimeter, which represented an experimental unit.

The mini lysimeters were filled with Yellow Latosol (Oxysol in USDA classification) containing 255 g kg<sup>-1</sup> coarse sand, 175 g kg<sup>-1</sup> fine sand, 30 g kg<sup>-1</sup> silt, and 540 g kg<sup>-1</sup> clay. Prior to liming, soil tests had revealed the following: pH in water, 4.5; P, 1 mg dm<sup>-3</sup> (Mehlich-1); K<sup>+</sup>, 0.13 Cmol<sub>2</sub> dm<sup>-3</sup>; Ca<sup>2+</sup>, 0.6 Cmol<sub>2</sub> dm<sup>-3</sup>; Mg<sup>2+</sup>, 0.6 Cmol<sub>c</sub> dm<sup>-3</sup>; Al<sup>3+</sup>, 0.5 Cmol<sub>c</sub> dm<sup>-3</sup>; H<sup>+</sup>, 2.2 Cmol<sub>2</sub> dm<sup>-3</sup>; Na, 0.0 Cmol<sub>2</sub> dm<sup>-3</sup>; Sum-ofbases, 1.3 Cmol<sub>a</sub> dm<sup>-3</sup>; ECEC, 1.8 Cmolc dm<sup>-3</sup>; and CEC (cation exchange capacity), 4.0 Cmol<sub>a</sub> dm<sup>-3</sup>. Soil acidity was corrected by applying dolomite (70 % ECCE) at a concentration of 0.77 g.L<sup>-1</sup> to raise the base saturation to 60 %. Before filling the containers, cattle manure was added to sieved soil at a ratio of 5:1 (soil: manure) to improve the soil structure. Chemical analysis of manure [(total contents, determined in the acid extract (nitric acid with perchloric acid) (N -Kjeldahl Method) (Organic carbon - Walkley Method - Black)], revealed the following: pH (H<sub>2</sub>O) 8.55; N, 1.26 %; P, 0.30 %; K, 2.08 %; Ca, 0.39 %; Mg, 0.34 %; S, 0.17 %; Na, 0.11 %; organic carbon, 19.34 %; Zn, 82.9 mg dm<sup>-3</sup>; Fe, 13640.5 mg dm<sup>-3</sup>; Mn, 209.0 mg dm-3; Cu, 11.7 mg dm-3; and B, 17.1 mg dm-3.

Two *Eucalyptus* clones, VCC 865 and CO 1407 (*E. europhylla* × *E. grandis*), were irrigated with saline water using different leaching fractions (LF). The experimental design was randomized blocks with treatments arranged in a  $4 \times 2 + 2$  factorial scheme with four replicates: four leaching fractions (3, 10, 20, and 30 % of the crop demand with saline water), two clones (VCC 865 and CO 1407), and two additional treatments, one for each clone, conventionally irrigated with fresh water. Saline water had electrical conductivity (EC) of 2.5 dS m<sup>-1</sup> and fresh water, 0.31 dS m<sup>-1</sup>.

Saline water was prepared based on an ionic ratio of 3Na to 2Ca. This is the most common ratio in saline waters in northeastern Brazil (Medeiros, 1992). The characteristics of the irrigation water were: Fresh water (pH = 7.53, EC = 0.31 dS m<sup>-1</sup>, Na<sup>+</sup> = 2.20 meq L<sup>-1</sup>, Ca<sup>2+</sup> = 0.25 meq L<sup>-1</sup>, Mg<sup>2+</sup> = 0.25 meq L<sup>-1</sup>, K<sup>+</sup> = 0.01 meq L<sup>-1</sup>, RAS = 4.57 mmol<sub>c</sub> L<sup>-1</sup>, Residual free chloride = 0.07 mg L<sup>-1</sup>) and Saline Water (pH = 7.7, EC = 2.50 dS m<sup>-1</sup>, Na<sup>+</sup> = 8.31 meq L<sup>-1</sup>, Ca<sup>2+</sup> = 3.20 meq L<sup>-1</sup>, Mg<sup>2+</sup> = 2.80 meq L<sup>-1</sup>, K<sup>+</sup> = 1.15 meq L<sup>-1</sup>, RAS = 9.20 mmol<sub>c</sub> L<sup>-1</sup>, Residual free chloride = 0.08 mg L<sup>-1</sup>)

Ninety-day-old nursery-grown *Eucalyptus* seedlings were transplanted to mini lysimeters. All seedlings were irrigated with fresh water for ten days, so that they could establish evenly. Then, saline water treatments were applied.

Soil water balance was calculated to determine the crop evapotranspiration (ETc) (Equation 1).

$$ETc = \frac{I + P - D}{S}$$
 Eq.1

where:

ETc = crop evapotranspiration (mm d<sup>-1</sup>); I = amount of irrigation water applied (L); P = precipitation (L); D = drainage water (L) and S = Surface area (container opening) (m<sup>2</sup>).

The amount of applied irrigation water for each treatment was calculated using Equation 2.

$$Wi = \frac{ETc}{1 - LF}$$
where:  
Eq.2

Wi= irrigation water applied, mm; ETc = crop evapotranspiration, mm and LF = leaching fraction, decimal.

At 114 days after the onset of treatments, the following measurements were made: plant height (PH), stem diameter (SD), canopy diameter (CD), leaf number (LN), absolute growth rate (AGR), relative growth rate (RGR), leaf dry weight (LDW), stem dry weight (SDW), aerial part dry weight (APDW), root system dry weight (RSDW), ratio of APDW to RSDW (APDW/RSDW), total plant dry weight (TPDW), electrical conductivity of drainage water (EC<sub>dw</sub>), and electrical conductivity of saturated soil-paste extract (EC<sub>e</sub>).

AGR and RGR were calculated using Equations 3 and 4, respectively (Cairo et al., 2008).

$$AGR = \frac{Hf - Hi}{t}$$

$$RGR = (\frac{Hf - Hi}{Hi})/t$$
Eq.4

where,

AGR = absolute growth rate; RGR = relative growth rate; Hf = final height; Hi = initial height and t = time.

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Electrical conductivity of the drainage water  $(EC_{dw})$  was measured using a conductivity meter (Micronal, model B330) with which readings were taken from the drainage water. Electrical conductivity of the of saturated soil-paste extract  $(EC_e)$  was determined using the standard method, which consisted of separating the extract from the paste using a Büchnere funnel paper filter coupled to a Kitasato flask and a suction pump. Measurements were made in accordance with standards provided by the US Salinity Laboratory Staff (Richards, 1954). The saturated paste was obtained using 300 g of airdried soil to which water was added up to saturation. This was done at both the beginning and the end of the experiment.

To assess the effect of saline water on plant growth, fresh water-irrigated plants were compared to saline water-irrigated ones. Factorial analysis was carried out to determine the responses of saline waterirrigated clones to leaching fractions. Evaluations took place 114 days after transplanting. The collected data were tested by analysis of variance and F-test ( $p \le 0.05$ ) and the means were compared using Tukey's test. Regression analysis was conducted to fit models to the collected data as a function of leaching fractions. The models were chosen considering the significance of the coefficient of determination. The data were further studied by Pearson's correlation ( $p \le 0.05$ ).

#### **3. RESULTS**

# 3.1 Fresh water irrigation: VCC 865 vs CO 1407 and Fresh water irrigated clones vs saline water irrigated clones

Plant height (PH), leaf dry weight (LDW), stem dry weight (SDW), aerial part dry weight (APDW), APDW/ root system dry weight (RSDW), total plant dry weight (TPDW), absolute growth rate (AGR), canopy diameter (CD), leaf number (LN), stem diameter (SD),  $EC_{dw}$ , and  $EC_e$  were statistically similar between the clones irrigated with fresh water. RSDW of CO 1407 was greater than that of VCC 865 by 15.6 % (Figure 1A). Plant height, SD, AGR, SDW, and APDW/RSDW were not affected by saline water.

Irrigation with saline water (EC 2.5 dS  $m^{-1}$ ) raised the soil salinity to 3.22 dS  $m^{-1}$  (Figure 2B). This may have led to reductions in CD, LN, LDW, APDW,

RSDW and TPDW that were from 12, 56, 37, 46, 27 and 38 %, respectively (Figure 1C-H).

# 3.2 Saline water irrigation: clone VCC 865 vs clone CO 1407

Stem diameter, RGR, and RSDW showed no differences across saline water irrigated clones. Conversely, PH, CD, AGR, LDW, SDW, APDW, APDW/RSDW, and TPDW were affected by saline water irrigation; the clone VCC 865 outperformed CO 1407 by 11, 16, 20, 37, 29, 37, 24, and 40 %, respectively (Figure 2).

# **3.3** Leaching fractions (LF) and Interaction between leaching fractions (LF) and clones (C)

Leaching fractions had no influence on PH, SD, CD, AGR, RGR, SDW, APDW, RSDW, and TPDW ( $p\leq0.05$ ). Leaf dry weight (LDW) and APDW/RSDW ratio decreased linearly with increasing LFs, reducing by 38 % and 21 %, respectively (Figure 3A and B).

The interaction between LF and C affected  $EC_{dw}$ and  $EC_e$ . A quadratic model was fitted to the response of  $EC_{dw}$  to LF for both clones. In VCC 865, the increasing curve reaches the maximum value at 13.2 % LF, which corresponded to an EC of 2.4 dS m<sup>-1</sup>. In CO 147, however, the curve was decreasing, and the minimum value was at 23.3 % LF, corresponding to 1.6 dS m<sup>-1</sup> (Figure 3C and D). A decreasing straightline function was fitted to  $EC_e$  values as a function of LF for VCC 865 (Figure 3E). As for CO 1407, although  $EC_e$  values decrease as LF increases, no model tested could be fitted to the data. By comparing the  $EC_e$  values between clones, the difference was significant only at 30 % LF (Figure 3F).

# **3.4** Correlation analysis of biomass and salinity variables in each clone

Studying how salinity correlates with plant growth biomass parameters revealed a total of 19 linear correlations for clone VCC 865, 14 of which were positive and 5 were negative, and a total of 18 linear correlations for clone CO 1407, all of which were positive (Figures 4A and 4B).

Canopy diameter appears to correlate strongly with biomass parameters in VCC 865 clone. For clone CO 1407, canopy diameter only correlated with the aerial part (p <0.05), having no relationship with the total plant dry weight (p> 0.05).



**gure 1** – Root system dry weight (RSDW) in different eucalyptus clones irrigated with fresh water – (A); electrical co

Figure 1 – Root system dry weight (RSDW) in different eucalyptus clones irrigated with fresh water – (A); electrical conductivity of soil saturated paste extract – EC<sub>2</sub> - (B); canopy diameter – CD (C); leaf number – LN (D), leaf dry weight – LDW (E); aerial part dry weight – APDW (F); root system dry weight - RSDW (G); and total plant dry weight – TPDW (H), in eucalypt plants. Different letters for the same parameter show significant different at 0.05 by F test. Error bars represent the mean's standard error.
 Figura 1 – Peso seco do sistema radicular (RSDW) em differentes clones de eucalipto irrigados com água doce – (A); condutividade

Figura 1 – Peso seco do sistema radicular (RŠDW) em diferentes clones de eucalipto irrigados com água doce – (A); condutividade elétrica do extrato de pasta saturada do solo (EC) - (B); diâmetro do dossel – CD (C); número de folhas – LN (D), peso seco de folhas – LDW (E); peso seco da parte aérea – APDW (F); peso seco do sistema radicular - RSDW (G) e peso seco total da planta – TPDW (H), em plantas de eucalipto. Letras diferentes para o mesmo parâmetro mostram diferenças significativas em 0,05 pelo teste F. As barras de erro representam o erro padrão da média.

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Figure 2 – Plant height – PH (A); canopy diameter – CD (B); absolute growth rate – AGR (C); leaf dry matter – LDM (D); stem dry matter (E); aerial part dry matter – APDW (F); ratio of aerial part dry weight to root system dry weight-APDW/RSDW (G); and total plant dry weight – TPDW (H), in *Eucalyptus* plants. Different letters, between the same parameter, differ significantly from each other at 0.05 by F test. Error bars represent the mean's standard error.

Figura 2 – Altura da planta – PH (A); diâmetro do dossel – CD (B); taxa de crescimento absoluta– AGR (C); peso seco foliar – MLD (D); peso seco do caule (E); peso seco da parte aérea – APDW (F); relação peso seco da parte aérea e peso seco do sistema radicular - APDW/RSDW (G); e peso seco total da planta – TPDW (H), em plantas de eucalipto. Letras diferentes, para o mesmo parâmetro, diferem significativamente umas das outras em pelo teste F a 5% de probabilidade de erro. As barras de erro representam o erro padrão da média.



- **Figure 3** Leaf dry weight LDW (A) and ratio of aerial part dry weight to root system dry weight APDW/RSDW (B), as a function of leaching fractions in eucalypt plants.  $EC_{dw}$  estimates as a function of leaching fractions (C); mean  $EC_{dw}$  values as a function of leaching fractions in the water saline irrigated clones (2.5 dS m<sup>-1</sup>) (D); EC estimates as a function of leaching (E); and mean EC values as a function of leaching fractions in the water saline irrigated clones (2.5 dS m<sup>-1</sup>) (F). \*\*Significant ( $p \le 0.01$ ). Different letters, for the same parameter at each leaching fraction, are significant by Tukey's HSD test ( $p \le 0.05$ ). Error bars represent standard error.
- **Figura 3** Peso seco da folha LDW (A) e razão da peso seco da parte aérea e peso seco do sistema radicular APDW/RSDW (B), em função das frações de lixiviação em plantas de eucalipto. Estimativas de  $EC_{dw}$  em função das frações de lixiviação (C); valores médios de  $EC_{dw}$  em função das frações de lixiviação (C); valores médios de  $EC_{dw}$  em função das frações de lixiviação (C); valores médios de  $EC_{dw}$  em função das frações de lixiviação (C); valores médios de  $EC_{dw}$  em função das frações de lixiviação (C); valores médios de  $EC_{dw}$  em função das frações de lixiviação (C); valores médios de  $EC_{dw}$  em função das frações de lixiviação (C); valores médios de  $EC_{dw}$  em função da lixiviação (E); e valores médios de  $EC_{dw}$  em função das frações de lixiviação nos clones irrigados com água salina (2,5 dS m<sup>-1</sup>) (D); Estimativas de  $EC_{dw}$  em função da lixiviação (E); e valores médios de  $EC_{dw}$  em função das frações de lixiviação nos clones irrigados com água salina (2,5 dS m<sup>-1</sup>) (F). \*\*Significativo ( $p \le 0,01$ ). Letras diferentes, para o mesmo parâmetro em cada fração de lixiviação, são significativas pelo teste de Tukey ( $p \le 0,05$ ). As barras de erro representam o erro padrão.



Figure 4 – Pearson's correlation matrix including all data collected from clones VCC 865 (A) and CO 1407 (B). Correlations were validated by the t test at a significance level of 0.05. Squares with an × mean that the correlation was not significant (p > 0.05).
 Figura 4 – Matriz de correlação de Pearson incluindo todos os dados coletados dos clones VCC 865 (A) e CO 1407 (B). As correlações foram validadas pelo teste t a 5 % de probabilidade de erro. Quadrados com × significam que a correlação não foi significativa (p > 0,05).

Stem diameter and LF correlated negatively with  $EC_{dw}$  and  $EC_{e}$  for VCC 865. However, these variables did not correlate for CO 1407.

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 $EC_{e}$  correlated positively with  $EC_{dw}$  in the mini lysimeters cultivated with VCC 865.

No correlation was observed between  $EC_{dw}$  and  $EC_{a}$  for clone CO 1407.

#### 4. DISCUSSION

The greater root system development in CO 1407 might relate to a characteristic intrinsic to the clone. In comparing physiological traits of both clones, Silva et al. (2020) reported that leaf area, photosynthetic pigments, and photosynthesis rates were higher in CO 1407 than in VCC 865.

Osmotic adjustment is the main process by which plants alleviate the salt-induced stress imposed by saline irrigation water (EC 2.5 dS  $m^{-1}$ ) (Lopes and Klar, 2009). Both clones were negatively affected to salinity, which might be associated with the exposure time of plants to saline condition — which lasted 114 days. Conversely, no negative effects were reported for

*E. urophylla*, but the plants were exposed to salinity conditions for only 30 days (Souza et al., 2015).

Elevated soil salinity because of using saline irrigation water negatively affects plant growth by reducing plants' water uptake (decreased osmotic potential) and damaging metabolic and photosynthetic processes due to higher uptake of Na<sup>+</sup> and Cl<sup>-</sup> (Mäser et al., 2002; José et al., 2016; Isayenkov and Maathuis, 2019). Particularly, NaCl can negatively impact hormone synthesis and translocation from roots to aerial parts, resulting in decreased leaf area and dry matter (Ferreira et al., 2001). Plant growth may be further affected by the reduction in cell division and elongation due to changes in plant cell wall extensibility (Munns, 2002). In E. urograndis NaCl levels above 4.5 dS m<sup>-1</sup> reduced dry matter production in both shoot and roots (Lopes and Klar, 2009). Similarly, Lacerda (2016) reported decreases in dry matter production in leaves and roots of Eucalyptus clones (AEC 144, AEC 1528, VCC 361, and VCC 865). In evaluating responses of Eucalyptus genotypes (E. camaldulensi Dehnh: '169'; E. grandis Hill ex Maiden  $\times$  *E. urophylla* S. T. Blake: '5E'; and E. globules Labill: 'Anselmo' and 'Odiel') to salinity stress, Sixto et al. (2016) found out that the clones showed a high survival rate even under moderate (50 mM) and severe (125 mM) saline conditions.

The greater plant growth of VCC 865 when irrigated with saline water suggests a better phenotypic adaptation to salinity stress. The wide range of phenotypic responses depends on plant genotype; while some genotypes are tolerant to salinity, others are highly sensitive to it (Hanin et al., 2016). High Na<sup>+</sup> levels in the soil may impair K<sup>+</sup> uptake by roots; however, the soil concentration at which Na<sup>+</sup> has a significant effect on K<sup>+</sup> uptake depends on crop species (Ferreira et al., 2001) or genotype, as observed in this study. The higher APDW/RSDW in VCC 865 confirms the better adaptation of the genotype to salinity. Lacerda (2016) also reported high APDW/ RSDW ratio for the clone VCC 865.

The decrease in LDW as a result of increasing LF might be due to the fact that some nutrients are more leachable, e.g., K<sup>+</sup> (Possen et al., 2014). The ratio of APDW to RSDW tends to 1.0 with increasing leaching fraction; values above 1.0 mean that partitioning of nutrients and photoassimilate is imbalanced, leading to reduced plant growth and water uptake (Alfaro et al., 2004; Caldeira et al., 2013). When using a lower leaching fraction, elevated Na<sup>+</sup> levels in the soil might have induced root damage (Ferreira, 2001), so the growth of aerial parts occurs to the detriment of roots.

In CO 1407,  $\mathrm{EC}_{\mathrm{dw}}$  was lower at 10 and 30 % LF. CO 1407 might have accumulated or taken up higher amounts of salt, which explains the lower growth rate of the clone when irrigated with saline water. When comparing EC<sub>dw</sub> values of the two clones irrigated with saline water, drainage water collected from mini lysimeters containing VCC 865 clone revealed that less salt was leached (lower  $EC_{dw}$ ) at 3 % LF, but more salt was leached at 20 % LF. In average, EC values increased from 0.2 dS m<sup>-1</sup>, before applying saline treatments, to 3.2 dS m<sup>-1</sup>, at the end of the experiment. In the mini lysimeters cultivated with VCC 865, the highest EC values were recorded when using the lowest LF. Similar results were reported for beet and peanut plants (Ferreira et al., 2006; Santos et al., 2012). This shows that using leaching is an efficient way of alleviating the impact of excessive salt in the irrigation water on plant growth. However, this positive response was observed only in the clone VCC 865.

Canopy diameter has a direct relationship with the accumulation of dry weight in the whole plant because the canopy consisted of leaves, where photosynthesis occurs.

Stem diameter and LF correlated negatively with  $EC_{dw}$  and  $EC_{e}$  for VCC865, i.e., increases in the number of leaves and stem diameter contributed to reductions in the electrical conductivity of the soil and the drained water. However, these variables did not correlate for CO 1407. This might be explained by the influence of saline irrigation on vegetative development of the clone (CO 1407), as shown for the different characteristics in Figure 2. For VCC 865, although salinity affects the accumulation of biomass, this effect is not as pronounced as in CO 1407.

Moreover,  $EC_e$  correlated positively with  $EC_{dw}$  in the mini lysimeters cultivated with VCC 865, that is, despite the increase in soil salinity by saline irrigation (Figure 1B), the largest leaching fractions promoted the reduction of salts in the soil and in the drained water (Figure 3C and 3E). On the other hand, no correlation was observed between  $EC_{dw}$  and  $EC_e$  for clone CO 1407, probably due to the uptake of salts by the clone.

### **5. CONCLUSIONS**

Only root dry weight differed between the clones when irrigated with fresh water, with clone CO 1407 outperforming clone VCC 865. However, when irrigating with saline water, both clones had their biomass negatively affected. The negative effects were greater in clone CO 1407, which had lower means for biomass.

By using saline irrigation water, soil salinity increases, even when using leaching fraction. Increases in soil salinity are different for each clone. The *Eucalyptus* clones perform similarly when irrigated with fresh water. However, if saline water is to be used, the results suggest using VCC 865 rather than CO 1407.

## AUTHOR CONTRIBUTIONS

All authors made essential contributions to the elaboration of this workThe manuscript is part of the master's thesis of the author OLIVEIRA FS. CASTRO FILHO MN was responsible for and the analysis the writing of the manuscript. OLIVEIRA FS e ALVES RO was responsible for conducting the experiment. Da SILVA BL assisted in the writing and translation of the manuscript. TAGLIAFERRE C was the supervisor teacher professor of the study and contributed to the supervision of the methodology and correction of the work as a whole. De PAULA A and BARROS FM assisted in supervision and review of the work as a whole.

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