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Different proportion of root cutting and shoot pruning influence the growth of citronella plants

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Abstract. Environment concern, sustainable products demand, and natural components conscious are currently global movement factors. Related to the global movement factors, citronella grass (*Cymbopogon nardus* L.) is being widely used in folk medicine, and has insect repellent activity, fungal and bactericidal action. Its essential oil has high content of citronellal, citronellol, geraniol. The essential oil is mostly extracted from leaves which turns this plant with high commercial demand. However, to obtain the best therapeutic quality and productivity of medicinal plants, which culminates in greater quantity and quality of the active compounds, the proper management of the crop is fundamental, as several factors can interfere during its growth and development. Thus, we analyzed the growth of citronella plants submitted to different levels of shoot and root cuts. Five different proportions of root pruning (0, 25, 50, 75, 100%), after 145 days of seedling planting and four cuttings in the shoots: blunt; a cut at 145 DAP (days after planting) along with the root cut; a cut at 228 DAP; and cuts at 145 and 228 DAP (two cuts). Four harvesting for dry matter accumulation and photoassimilate partition data were performed. The treatment with 100% root cut, but without leaf cut, increased the total dry mass accumulation of the plant in relation to the other treatments, for the last analysis period, demonstrating a recovery. Thus, the application of two leaf cuts or no leaf cutting within the 100% root cut treatment for leaf dry mass accumulation is more effective when compared to the blunt root treatment.

Key words: pruning, citronella growth, *Cymbopogon nardus* L., medicinal plant, photoassimilates, stress root.

INTRODUCTION

Concern for the environment and the quest to consume sustainable products with the conscious use of natural components is a growing movement worldwide (Chen et al., 2019; Xiong et al., 2019; Hatanaka, 2020; Yang et al., 2020). The research extends to several areas such as: medicines, cosmetics and food (De Hooge et al., 2018; Kozlowska et al., 2019). In Brazil, the juice and tea market grow 15% a year. Brazil has also stood out in the market for products for hygiene, cosmetics and perfumes and has grown over 20% in exports over the last years (Corrêa Junior & Scheffer, 2013).

Thus, the species known as citronella grass (*Cymbopogon nardus* L.), is a plant of great interest as a raw material for natural products production (Kačániová et al., 2017; Silva & Ricci-Júnior, 2020). Citronella is a perennial species that fully develops in warm environments with fertile and well-drained soils, in addition to a large amount of biomass production, similarly to plants with C_4 metabolism (Sivashanmugam et al., 2009; Faria et al., 2018; Zhang et al., 2019).

The essential oil (EO) produced on leaves of citronella grass has several properties, such as repellent effect, especially for mosquitoes, and on negatively growth of fungi and bacteria (Solomon et al., 2012; Capoci et al., 2015; Hernandez-Lambraño et al., 2015), besides presenting aromatic characteristics (Gupta et al., 2018). Citronella leaves can also be used as a sedative and soothing in Brazilian folk medicine and in Indonesia as a soothing and digestive tea (Castro et al., 2007; Castro et al., 2010).

The leaves are the most economically valuable part of citronella grass, where EO is extracted. The EO is rich in citronellal aldehyde and contains high content of geraniol, citronellol and esters (Barbosa et al., 2008; Oliveira et al., 2010). The potential of EO has been reported to control pathogenic bacteria and fungi (Silva et al., 2010; Demuner et al., 2011; Nascimento et al., 2011).

The value of medicinal, aromatic or flavoring plants is determined by the production of chemical compounds, which are called active ingredients. In general, the active ingredients are mainly produced in leaves, flowers, roots or barks, depending on the plant species (Corrêa Junior & Scheffer, 2013). Moreover, the biosynthesis of chemical compounds vary according to several factors, ranging from changes in crop management to environmental factors such as light, temperature, soil state and photoperiod which can directly influence in growth and development, as well as yield (Peixoto et al., 2011; Pavarini et al., 2012; Nobre et al., 2013). The productivity is even reported as being widely affected according to weather conditions in the trial years (Plūduma-Pauniņa et al., 2019).

Thus, it is necessary to consider the optimal environmental conditions for each species, aiming high quantity and quality of chemical compounds, especially plants with high production of EO and can be highly impacted due to environmental changes that interfere on active compounds production (Souza et al., 2011).

Besides that, cutting processes mainly to obtain material for compound extractions can also be a stressful factor. Therefore, it is important to understand how the pruning of the shoot along with cuts in the root system during the citronella cultivation can influence biomass production, which is the main important raw material for EO extractions. In addition to the stress caused by pruning and cutting to the plants, it also can provide a better crop yield such as for example a better root and shoot development with high quantity and quality of EO (Kaczorowska–Dolowy et al., 2019).

Several studies have reported that pruning levels can influence plant metabolism: (i). pruning intensity influences growth, flower, and fruit development (Toit et al., 2020); (ii). early or late pruning influences EO yield; (Thakur et al., 2019); (iii). the number of pruned branches has a positive correlation with the growth attributes (Kathiresan et al., 2019); (iv). Partial root pruning lead to high plasticity of plants when the biomass of source organs is changed (Fanello et al., 2020); (v). the metabolite profile of shoots changes significantly after pruning (Arkorful et al., 2020).

Thus, to test how plants change the shoot growth when the roots are reduced (cut roots) by influencing the source-sink relationship during the critical period when leaves are removed from plants. Besides that, the requirement to comply with well-defined quality standards in medicinal and aromatic plants requires the elaboration of specific production strategies (Máthé & Franz, 1999). Therefore, the aim of this study was to analyze the biomass partitioning of citronella plants submitted different proportion of root cutting and shoot pruning.

MATERIALS AND METHODS

The experiment was carried out between March 2015 and May 2016 (22º54'00" S, 43º08'00" W and 8 m altitude). The annual average temperature was 23 ºC, with Aw climate, according to the Köppen classification. Citronella grass (*Cymbopogon nardus* (L.) Rendle) seedlings were planted in 8 L-polyethylene pots containing sandy soil, using a spacing of 40 cm between rows and between plants. Plants were watered to keep at field capacity. The plants were cultivated for five months until the beginning of the treatments, where they have adequate leaf area for the first pruning. For this, the experiment was conducted in a fully randomized design, with five different proportions of root cutting (0, 25, 50, 75 and 100%) and distinct shoot pruning after 145 days of seedling planting. The first pruning of all leaves was performed on the same day as the root cutting. The leaf pruning was composed of four treatments: blunt; pruning at 145 DAP (days after planting) along with the root cutting; pruning at 228 DAP; and pruning at 145 and 228 DAP (two pruning).

The roots were cut into a container with water and kept immersed in water for 24 h (at room temperature) to avoid cavitation in the vascular system. Root cutting treatments were through transverse cuts in the root system. After 24 h, the plants were replanted in 8 L-pots containing the same soil as above described. Throughout the experiment, the plants were fertilized (six and twelve months after seedling planting) using 300 g of Bokashi® compost plus 13 g of NPK (nitrogen: 0.4%; phosphorus: 14% and potassium: 0.8%) per pot.

To obtain the data of dry matter accumulation and photoassimilate partitioning, successive harvesting were performed, starting on the day of seedling planting, totaling four harvesting and in each harvesting the dry mass of the plant organs (leaf, stem and roots) was determined. The dry mas was getting by drying the fresh mass samples in an oven dried with forced air circulation at 65 °C until reach constant mass (± 72 h). The dry matter mass was calculated by the difference in fresh and dry mass for each analysis period.

The data was compared by its respective standard deviation with split plots. Treatment a: five different proportions of root cuts and treatment b: four different types of leaf cuts, with five samples per treatment.

RESULTS AND DISCUSSION

The growth analyzes show that when the roots are cut, but without pruning the leaves, there is an increase of the root dry mass in relation to other treatments. Leaf cutting alone showed the highest dry mass production for the leaves, and the treatments that remained at levels of non-leaf pruning were 50 and 75% root cutting.

Thus, the results of the evolution of citronella dry matter in internal comparisons (leaf pruning with different root cuts) (Fig. 1), with the first and second analysis being identical in all results, as the plants with no cutting and pruning. After 228 DAP, there was alterations, by being more relevant in the last analysis, since plants contained the treatments in the third and fourth analysis. After 145th day, changes were observed, with emphasis on the last analysis, since all plants had undergone some treatment.

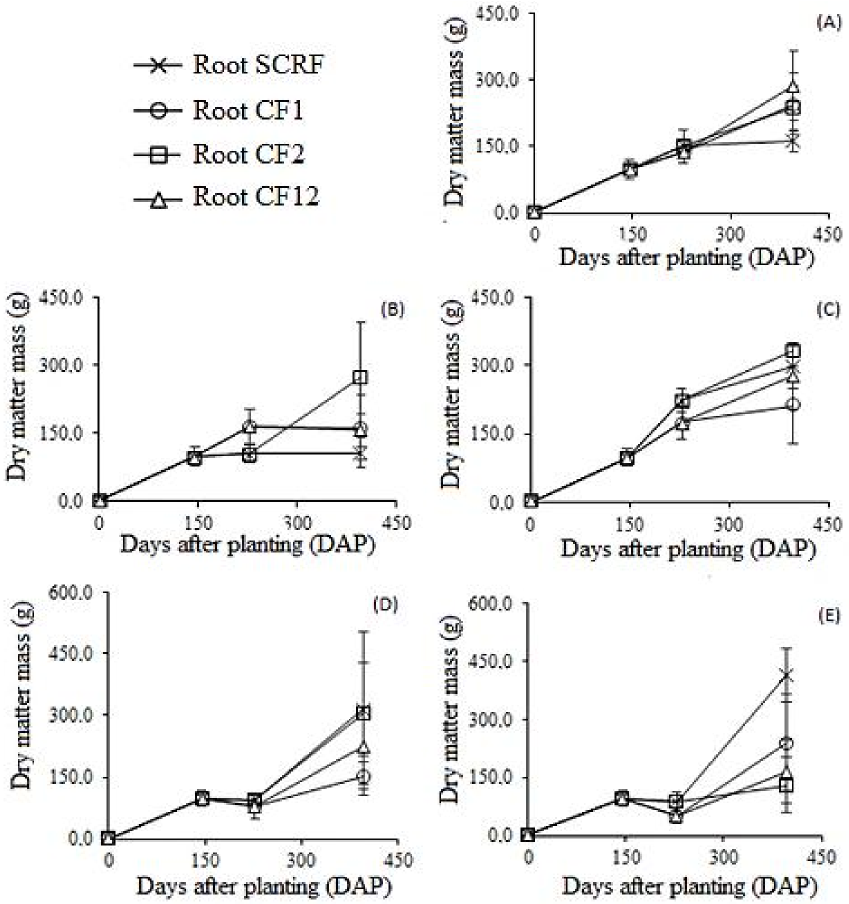

Figure 1. Evolution of citronella plant root dry matter accumulation in leaf cut treatments: (A) blunt root; (B) 25% root cut; (C) 50% root cut; (D) 75% root cut; (E) 100% root cut. (SCRF: no leaf pruning; CF1: leaf pruning at 145 DAP; CF2: leaf pruning at 228 DAP; CF12: leaf pruning at 145 and 228 DAP). Bar indicates Standard Deviation.

The treatment with no cutting on root (Fig. 1, A), which involved two leaf pruning at 145 and 228 DAP showed the highest root dry matter accumulation in the last analysis period. According to this treatment, it was observed that in the final analysis period the treatments with cutting at 145 and 228 DAP presented very similar values in their respective dry matter accumulation curves.

The treatment with 100% root cutting (Fig. 1, E) and no leaves pruning increased the root dry mass in relation to the other treatments, showing a 74.11% increase compared to the 25% root cutting treatment (Fig. 1, B) for the same treatment of leaves in the last analysis period. This suggests that even under stress, the 100% root cutting plants had a higher root dry mass accumulation, as compared to the control (Fig. 1, A).

When the total dry mass accumulation of citronella was analyzed in the different root cutting treatments in relation to the four leaf pruning treatments, the treatment with 100% root cutting only and without leaf pruning showed a total of 838.67 g, as previously published data from this experiment (Daflon et al., 2019). This was the treatment that showed the largest increase in total mass in relation to the control, especially in the last analysis, where the root mass had a major contribution in the total mass of these plants, with an increase of 270 g, and soon after treatment with 50% of the root cut.

The redirection of photoassimilates to biomass production at the affected site (roots) is probably due to the fact that this energy is essential for the formation of new roots at this site, and most experiments involving root cutting demonstrate the reorientation of the roots assimilated toward the cut roots (Fanello et al., 2020). The same authors state that in cuttings performed on soybean roots in different proportions also promoted a greater accumulation of root biomass in pruned plants, causing a large increase in root respiration (increased alternative oxidase activity) and nitrate absorption, as well as also of water consumption towards the end of growth, as the pruned plants age.

Studies indicate that biomass accumulation by the plant and its productivity can be strongly affected by environmental conditions, among other factors (Lucas et al., 2013; Poorter et al., 2016; Xiao et al., 2016; Plūduma-Pauniņa et al., 2019; Yin et al., 2019), even if crop and weed mismanagement occurs, this can affect yield, which reflects the quality of the final product (Glatkova & Pacanoski, 2019).

In the treatments that suffered root cuts and a leaf pruning: i: 25% (Fig. 1, B), ii: 50% (Fig. 1, C) iii: 75% (Fig. 1, D) demonstrated higher dry mass accumulation compared to other treatments, including control (Fig. 1, A). However, the 75% treatment (Fig. 1, D) achieved a 36.19% reduction compared to the 100% treatment (Fig. 1, E) which showed higher root dry mass in relation to this leaf pruning.

Several factors can affect the growth and development of the crops, as well as the production of secondary metabolites, not only the injury caused by the application of pruning, but the characteristics of the soil, the temperature, among other environmental factors (Pavarini et al., 2012). However, such oscillations may be due to the fact that in some situations plants may show growth stimulus, including the production of secondary metabolites, in response to stress, in order to help organisms to establish adaptive responses, presenting the phenomena hormesia, which can contribute to improving the performance of plants when they are less aggressive to the plant (Tavares et al., 2015; Vargas-Hernandez et al., 2017; Hussain et al., 2019).

Regarding the data related to stem dry matter accumulation, under the action of treatments with pruned leaves in relation to the root cuts are presented in Fig. 2. The treatment that presented higher accumulation of stem dry mass in relation to the pruning of the leaves. The leaves did not contain root cuts, but with two leaf cuts (145 and 228 DAP) (Fig. 2, A), for the last period analyzed. Fanello et al. (2020) found the opposite in soybean plants subjected to longitudinal cutting in the roots of 15, 25 and 50%, where the plants whose roots were pruned showed large increases in dry matter in all vegetative organs compared with the plants intact.

When comparing the two leaf pruning with the different root cuts, the treatment with the lowest accumulation was the treatment of 75% of the root cut (Fig. 2, D) and this reduction compared to the control was 38.25%. Thus, it can be analyzed that if there is no damage to the root system, the greatest accumulation of dry mass is in the stems, but when it contains two pruning in the leaves, because it can present energetic reserves for the plant, when in the absence of the photosynthetically active part (leaves), as several studies indicate that different parts of plants, such as stem and root, have an average accumulation of different nutrients, being able to present considerable individual development, despite these differences in allocated photoassimilates (Poorter et al., 2012; Rocha et al., 2014; Souza et al., 2018; Xu et al., 2018).

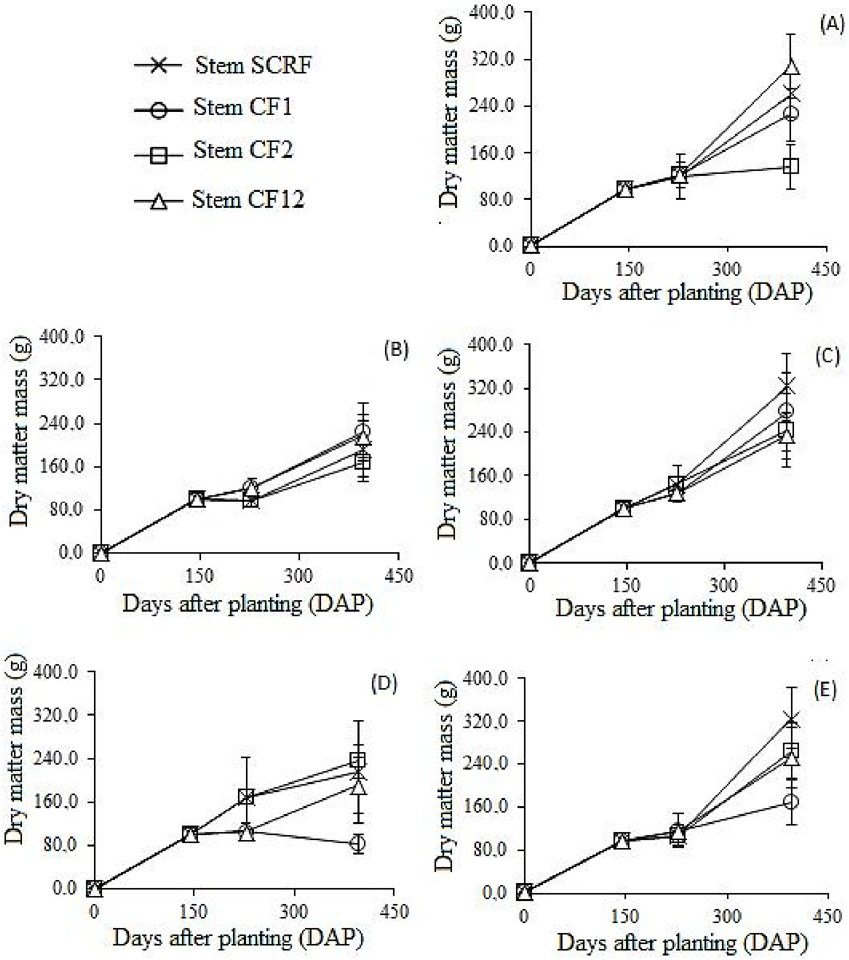

Figure 2. Evolution of citronella plant stem dry matter accumulation in leaf pruning treatments: (A) blunt root; (B) 25% root cut; (C) 50% root cut; (D) 75% root cut; (E) 100% root cut. (SCRF: no leaf pruning; CF1: leaf pruning at 145 DAP; CF2: leaf pruning at 228 DAP; CF12: leaf pruning at 145 and 228 DAP). Bar indicates Standard Deviation.

Depending on the species, up to 50% of the photoassimilates are consumed by the roots and the photoassimilate fraction used for root respiration increases with plant age as they are spent on maintenance respiration when root growth decreases (Lambers et al., 2002; Fanello et al., 2020). This energy expenditure in the primary metabolism directly influences the production of the compounds of the secondary metabolism, such as essential oils, can provide an imbalance in the metabolism (Li et al., 2020).

When leaf pruning was analyzed only at 145 DAP in relation to the different root cuts, the treatment with the highest stem dry mass accumulation was 50% of the root cut (Fig. 2, C), followed by the blunt root treatment (Fig. 2, A), 25% (Fig. 2, B), 100% (Fig. 2, E), and to a lesser extent the treatment of 75% (Fig. 2, D), as it presented the greatest alterations between the values of stem dry matter accumulation, among their different types of leaf stress. Treatment involving leaf cutting at 145 DAP along with 75% root cutting (Fig. 2, D) decreased from the third analysis and continued this way until the last analysis period.

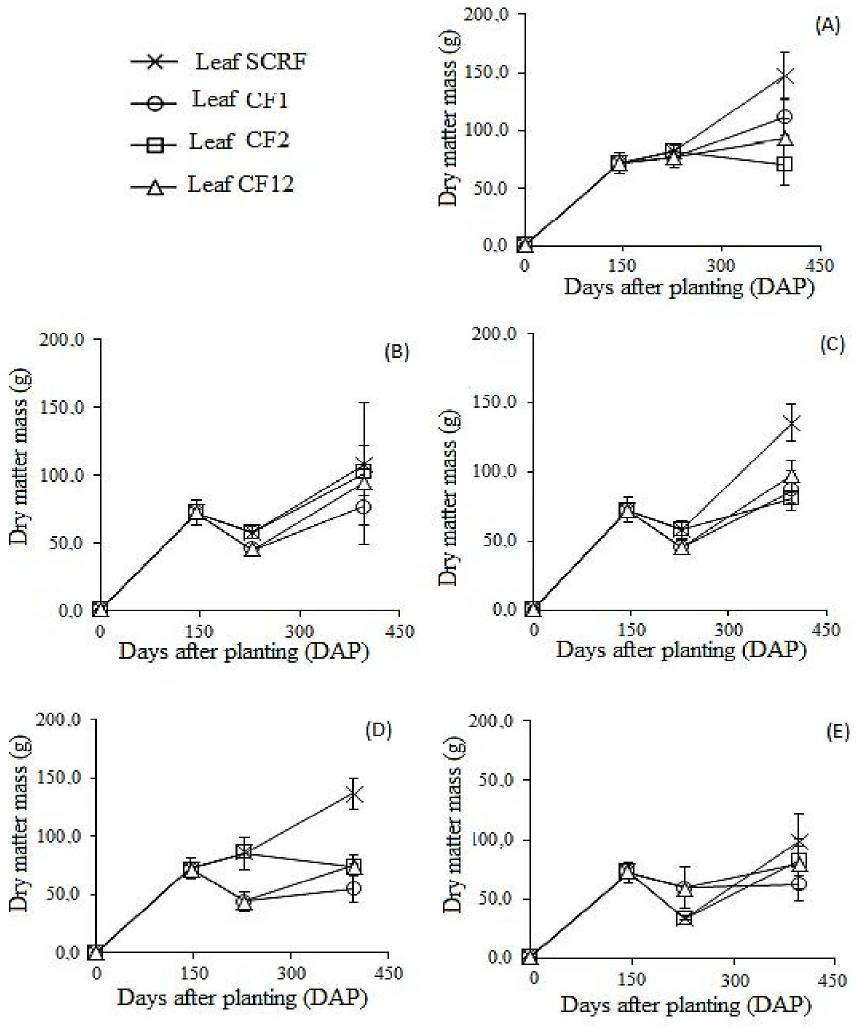

Figure 3. Evolution of leaf dry matter accumulation of citronella plant in leaf pruning treatments: (A) without root cutting; (B) 25% root cut; (C) 50% root cut; (D) 75% root cut; (E) 100% root cut. (SCRF: no leaf pruning; CF1: leaf pruning at 145 DAP; CF2: leaf pruning at 228 DAP; CF12: leaf pruning at 145 and 228 DAP). Bar indicates Standard Deviation.

Regarding the results of the evolution of leaf dry matter accumulation, for treatments of leaf pruning in relation to root cuts (Fig. 3), it was observed that the treatments in which the values were at control levels (Fig. 3, A) were 50% (Fig. 3, C) and 75% (Fig. 3, D), data from the analysis of 228 days after planting in relation to the treatment without pruning in the leaves, thus evidencing that the maintenance of both organs (root and leaves) influences the overall biomass of the plant.

When analysing the dry mass of the leaves and making two pruning in the leaves (145 and 228 DAP), these plants show a reduction, mainly among the analyzes, as can be seen mainly from the treatments with 100% root cut (Fig. 3, E) and 25% (Fig. 3, B). It was observed that the treatment that accumulated more dry matter in the leaf region was the one that did not have thinning neither in the root nor in the leaves, because it presented the maintenance of the two organs responsible for water and nutrient uptake and energy generation in the plant, respectively, which allowed its full growth and development, maintaining the photosynthetic activity for the production of organic molecules, being later distributed through the plant and transformed into biomass (Poorter et al., 2012; Sims et al., 2012; Reich et al., 2014; Li et al., 2019).

However, it is noteworthy that the plants that were not stressed with pruning in the shoots, these also have leaves that were senescent or entering senescence, all being accounted for dry mass, because they are retained in the plants, however, in other treatments presented leaves in senescence, due to the fact that the pruning was performed and these removed together with the other leaves at the time of pruning.

After pruning, many plants present an accelerated metabolic activity of the cells adjacent to the lesion, as in the case of citronella, which showed a rapid growth and increase of biomass after cutting, both in the root and shoots. was evidenced by the agility and ability to acclimate to these new situations imposed by the treatments, the same found by Fanello et al. (2020), where the soybean plants that suffered root cutting were able to fill the pods while regenerated their root system, well in the critical period of grain filling, without loss of yield, thus demonstrating the root plasticity in pruning situations.

Many agricultural crops use pruning, which may even be periodic to increase yield, among others (Albarracín et al., 2019; Gutierrez-Coarite et al., 2018; Fanello et al., 2020), experimental conditions, even the citronella undergoing drastic pruning, such as the complete removal of the root system, they survived and still showed greater biomass gain at the end, thus highlighting a positive reaction to its high metabolic adjustment capacity, including its relationship drain source.

Thus, the applicability of this form of cutting in the roots, can be carried out both in smaller crops, in pots, as well as in crops grown in beds, which facilitates the management of this crop and contributes, depending on the intensity of pruning, to a greater biomass production. However, there is a need for future work to also seek to relate this type of pruning in different environmental and cultivation conditions, in addition to relating to the production and yield of essential oil.

CONCLUSIONS

Citronella plants containing root cutting show compensatory growth in relation to root thinning, and the increase of root biomass gradually depends on the increase of thinning level, the same not being verified for stem and leaf biomass, which varied according to the levels of shoot pruning.

The treatment with 100% cut in the root, but without pruning in the leaves, increased the accumulation of total dry mass of the plant in relation to the other treatments, for the last period of analysis, showing a recovery.

The application of two pruning in the leaves or no pruning the leaves, within the treatment with 100% of the cut in the root for the accumulation of dry mass of the leaf is more effective when compared to the treatment without cutting in the roots.

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