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ARTIFICIAL DEFOLIATION ON *Eucalyptus benthammii* AND *Eucalyptus dunnii* FOR SIMULATION OF LEAF-CUTTING ANT ATTACKS

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Resumo

Desfolha artificial em Eucalyptus benthammii e Eucalyptus dunnii para simulação de ataques de formigas cortadeiras. Os efeitos de diferentes níveis de desfolha causados por formigas cortadeiras ainda não foram avaliados em plantas de *Eucalyptus benthammii* Maiden et Cambage e *Eucalyptus dunnii* Maiden. O objetivo deste trabalho foi avaliar as perdas causadas por diferentes níveis de desfolha artificial em *E. benthammii* e *E. dunnii* aos 30 dias após o plantio, simulando o ataque de *Acromyrmex*. Foram avaliados quatro tratamentos: 0: controle, 1: 50%, 2: 75% e 3, 100% de desfolha. A altura e o diâmetro foram registrados aos 6, 12, 24, 36 e 72 meses. Aos seis meses, a altura e o diâmetro de *E. bentammii* submetidos a 100% de desfolha diferiram significativamente das plantas controle, no entanto, não foram observadas diferenças significativas na altura e diâmetro após 12 meses. Em *E. dunnii*, 100% de desfolha resultou em mortalidade significativa, e as plantas que sobreviveram a este nível de desfolha tiveram perdas significativas em seu desenvolvimento. No caso de ataque de *Acromyrmex* aos 30 dias após o plantio, sugere-se o replantio de *E. dunnii* 100% desfolhados para evitar perdas no final do ciclo florestal. Para *E. benthammii*, sugere-se avaliar o vigor das mudas 100% desfolhadas, pois elas podem se recuperar ao longo do tempo. Apenas 100% de desfolha pode levar a perdas no crescimento das plantas de eucalipto. Porém, estudos regionalizados são necessários para avaliar as perdas que desfolhas intensas podem causar em cada situação e em cada espécie florestal, pois algumas espécies, como *E. benthammii*, podem se recuperar ao longo do tempo.
Palavras-chave: Acromyrmex; danos; praga florestal; replantio.

Abstract

The effects of different defoliation levels caused by leaf-cutting ants have not yet been evaluated in *Eucalyptus benthammii* Maiden et Cambage and *Eucalyptus dunnii* Maiden plants. The aim of this study was to evaluate the losses caused by different defoliation levels on *E. benthammii* and *E. dunnii* plants artificially defoliated 30 days after planting, simulating *Acromyrmex* attacks. Four treatments were evaluated: 0: control, 1: 50%, 2: 75%, and 3, 100% defoliation. Plant height and diameter were recorded at 6, 12, 24, 36 and 72 months after planting. At six months, height and diameter of *E. bentammii* plants subjected to 100% defoliation differed significantly from control plants, however no significant differences in plant height and diameter were seen after 12 months. In *E. dunnii*, 100% defoliation resulted in significant plant mortality, and plants that survive this level of defoliation had significant losses in their development. In case of *Acromyrmex* attack on eucalyptus plants at 30 days after planting, it is suggested to replant *E. dunnii* 100% defoliated to avoid losses in the end of the forest cycle. In *E. benthammii* plantations, it is suggested to evaluate the vigor of the 100% defoliated seedlings, as they can recover over time. Only 100% defoliation can lead to losses in *Eucalyptus* plant growth. However, regionalized studies are necessary to assess the losses that intense defoliation can cause in each situation and in each plant species, as some species, such as *E. benthammii*, can recover along time.
Keywords: Acromyrmex; damage; forest pest; replanting.

INTRODUCTION

Leaf-cutting ants of the genera *Atta* and *Acromyrmex* rank among the insects of tremendous economic importance in the Neotropical region, as they defoliate a wide variety of plant species and use the vegetal material to cultivate the symbiotic fungus, their main food source (HÖLLDOBLER; WILSON, 2011). Leaf-cutting ants can cause total defoliation of both seedlings and adult plants. However, plant age can influence the vulnerability to damage caused by ants. Damage is greater in young plants, and attacks on recently-planted seedlings can be irreversible, due to the fragility of the seedlings (DELLA LUCIA, 2011).

Leaf-cutting ants are considered the main pest in eucalyptus plantations. In *Eucalyptus grandis* W. Hill ex Maiden, losses in height and diameter are proportional to the intensity of defoliation, and one year after defoliation at 30 days of planting, there are significant losses in the development of plants submitted to defoliation levels greater than 75 % (REIS FILHO *et al.*, 2011). When defoliation occurs at six months of planting, only 100% defoliation causes significant damage to plant growth (OLIVEIRA *et al.*, 2014). Greater damage, however, is seen after consecutive defoliation over the first year of *E. grandis* plantation (MATRANGOLO *et al.*, 2010).

Artificial defoliation is a valuable tool in measuring the damage of the different intensities of defoliation caused by insects. The reduction in plant growth can be assessed by comparing defoliated and non-defoliated plants when grown under the same conditions (KULMAN, 1971). Several studies have used artificial defoliation to simulate leaf-cutting ant attacks on pine and eucalyptus plants (RIBEIRO; WOESSNER, 1980; REIS FILHO *et al.*, 2011; MATRANGOLO *et al.*, 2010; NICKELE *et al.*, 2012; OLIVEIRA *et al.*, 2014). However, the effects of different levels of defoliation caused by leaf-cutting ants have not yet been evaluated for *Eucalyptus benthammii* Maiden et Cambage and *Eucalyptus dunnii* Maiden, both of economic interest. These species are commonly planted in southern Brazil, as they are tolerant to frost, which is very common in the winter of this region (PALUDZYSZYN FILHO *et al.*, 2006; GONÇALVES *et al.*, 2013).

In the southern Brazil, there is a dominance of leaf-cutting ants of the *Acromyrmex* genus in forest plantations (NICKELE *et al.*, 2009; KRÜGER *et al.*, 2010; UKAN *et al.*, 2010). Attacks of these ants on eucalyptus plants are significant only in the first months after planting, being more intense during the first 30 days (BARBOSA *et al.*, 2021). Leaf-cutting ants can cause three levels of defoliation in recently-planted *Eucalyptus*: level 1: 50%; level 2: 75%; and level 3: 100% defoliation (REIS FILHO *et al.*, 2011). Our hypothesis is that not all defoliation levels cause damage on *E. benthammii* and *E. dunnii*, as already observed in other species. Thus, the aim of this study was to evaluate the losses caused by different defoliation levels on *E. benthammii* and *E. dunnii* plants artificially defoliated 30 days after planting, simulating *Acromyrmex* attacks.

MATERIAL AND METHODS

The study was carried out in an *E. benthammii* plantation located in the municipality of Mafra (26°12'30.1"S 49°55'19.0"W), and in an *E. dunnii* plantation located in the municipality of Três Barras (26°08'29.1"S 50°09'47.3"W), both in the state of Santa Catarina, Brazil. *Eucalyptus benthammii* was planted in October 2016 and *E. dunnii* was planted in January 2017, at a spacing of 2.5 x 2.5 m.

Four treatments were evaluated: 0: control (without defoliation), 1: 50% defoliation, 2: 75% defoliation, and 3, 100% defoliation. Plants were defoliated using pruning shears 30 days after planting. The experimental design was completely randomized and artificial defoliation was performed on 20 plants for each defoliation level. Twenty plants were also selected as controls (level 0). Each plant was numbered and identified according to treatments. Plants were equally spaced 12.5 × 5 m from each other, interspersed with non-defoliated plants. Leaf-cutting ant control was performed in the experimental area to prevent later damage by ants.

The initial plant height was recorded before performing the artificial defoliation at 30 days after planting. Subsequently, plant height and diameter were recorded at six, 12, 24, 36 and 72 months after planting. Plant height was recorded with a Vertex hypsometer, and diameter at 1.30 m was recorded with a Mantax Blue 650mm calliper.

Shapiro–Wilks test was performed to test the normality of data. Analysis of variance was performed, followed by Tukey's post-hoc test (5% probability) to compare the growth of the defoliated plants at different defoliation levels. Statistical analyses were performed using the program R 3.5.1 (2018).

RESULTS

There was no difference in the initial height of *E. benthammii* plants ($F_{3,76}$ =0.41, P=0.74, Figure 1). At six months, 100% of defoliation caused 20% of plant mortality. In the control and 50% of defoliation, there was 5% of plant mortality. After this period, there was no plant mortality. In this period, only 100% of defoliation caused losses in growth in height and diameter (height: $F_{3,68}$ = 3.94, P=0.01, Figures 1; diameter: $F_{3,68}$ = 4.15, P=0.009, Figure 2). However, plants remade their foliage along time and there was the recovery of these plants, not differing anymore from control plants at 12 (height: $F_{3,68}$ = 0.64, P=0.57; diameter: $F_{3,68}$ = 2.23, P=0.09), 24 (height: $F_{3,68}$ = 0.76, P=0.51; diameter: $F_{3,68}$ = 2.29, P=0.08), 36 (height: $F_{3,68}$ = 1.52, P=0.21; diameter: $F_{3,68}$ = 1.15, P=0.33), and 72 (height: $F_{3,68}$ = 1.26, P=0.31; diameter: $F_{3,68}$ = 0.87, P=0.47) months after planting.

In *E. dunnii* plants there was also no difference in the initial height ($F_{3,76}$ =1.97, P=0.12, Figure 3). In the control and 75% of defoliation treatments there was 5% of plant mortality, and 10% of plant mortality in the 50% defoliation treatment. There was 95% of plant mortality in the 100% defoliation treatment, leaving only one surviving plant and this treatment was excluded from statistical analysis. At six months after planting, it was not possible to measure the diameter at 1.30 m, as plants were still very low. For height, plants subjected to 75% defoliation differed significantly from control ($F_{2,51}$ =3.49, P=0.03, Figure 3). At 12 months after planting, 75% defoliation caused losses in growth in height and diameter (height: $F_{2,51}$ =3.35, P=0.04, Figure 3; diameter: $F_{2,51}$ =4.14, P=0.02, Figures 4). However, from 24 months, there was the recovery in plant height and diameter, and 75% defoliation did not differ from control plants at 24 (height: $F_{2,51}$ = 0.42, P=0.65; diameter: $F_{2,51}$ = 1.32, P=0.28), 36 (height: $F_{2,51}$ = 1.38, P=0.25; diameter: $F_{2,51}$ = 1.24, P=0.29), and 72 (height: $F_{2,51}$= 0.97, P=0.39; diameter: $F_{2,51}$= 2.33, P=0.11) months after planting. Only the surviving plant with 100% defoliation have the growth in height and diameter reduced.

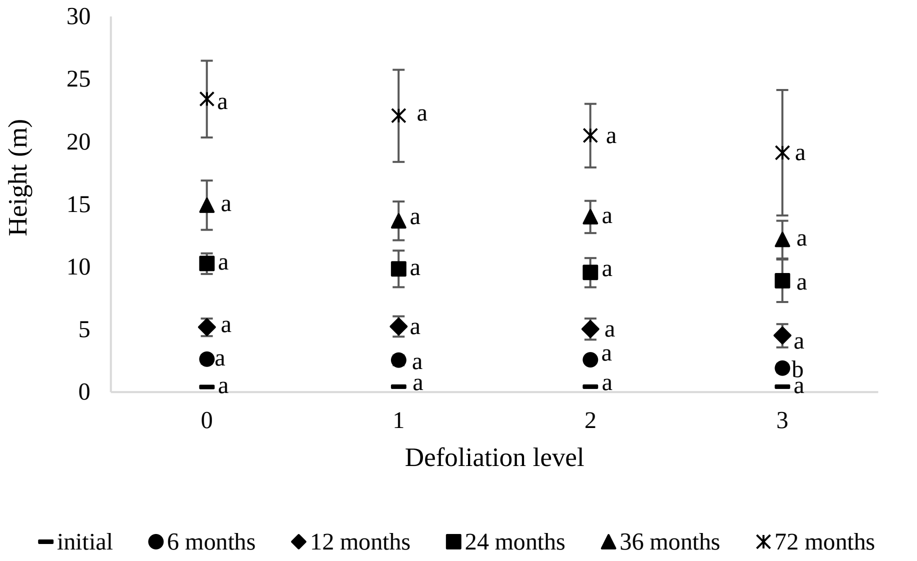

Figure 1. Height (mean ± confidence interval (CI)) along time of *Eucalyptus benthamii* plants artificially defoliated at 30 days after planting for simulation of *Acromyrmex* attack. Different letters indicate significant differences among treatments according to Tukey's test, at 5% probability. Defoliation level: 0: Control, 1: 50%, 2: 75%, 3, 100% defoliation.

Figura 1. Altura (média ± intervalo de confiança (IC)) ao longo do tempo de plantas de *Eucalyptus benthamii* desfolhadas artificialmente aos 30 dias após o plantio para simulação do ataque de *Acromyrmex*. Letras diferentes indicam diferenças significativas entre os tratamentos de acordo com o teste de Tukey's a 5% de probabilidade. Níveis de desfolha: 0: Controle, 1: 50%, 2: 75%, 3, 100% de desfolha.

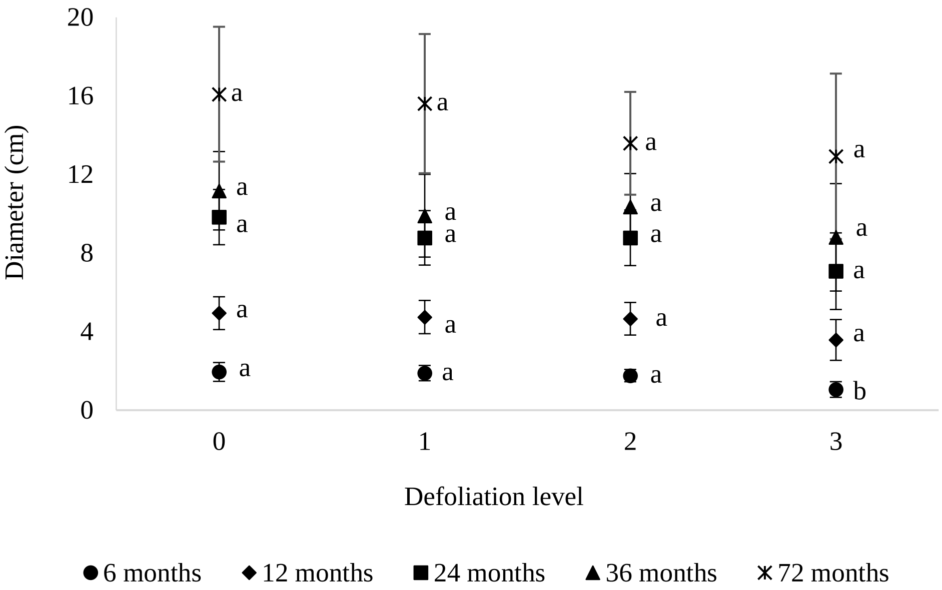

Figure 2. Diameter (mean ± CI) along time of *Eucalyptus benthamii* plants artificially defoliated at 30 days after planting for simulation of *Acromyrmex* attack. Different letters indicate significant differences among treatments according to Tukey's test, at 5%probability. Defoliation level: 0: Control, 1: 50%, 2: 75%, 3, 100% defoliation.

Figura 2. Diâmetro (média ± IC) ao longo do tempo de plantas de *Eucalyptus benthamii* desfolhadas artificialmente aos 30 dias após o plantio para simulação do ataque de *Acromyrmex*. Letras diferentes indicam diferenças significativas entre os tratamentos de acordo com o teste de Tukey's a 5% de probabilidade. Níveis de desfolha: 0: Controle, 1: 50%, 2: 75%, 3, 100% de desfolha.

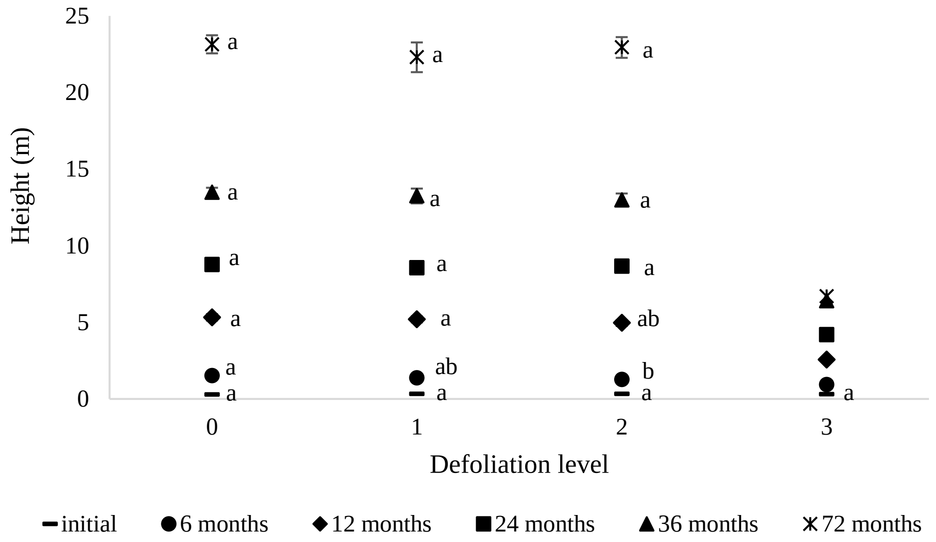

Figure 3. Height (mean ± CI) along time of *Eucalyptus dunnii* plants artificially defoliated at 30 days after planting for simulation of *Acromyrmex* attack. Different letters indicate significant differences among treatments according to Tukey's test, at 5% probability. Defoliation level: 0: Control, 1: 50%, 2: 75%, 3, 100% defoliation.

Figura 3. Altura (média ± IC) ao longo do tempo de plantas de *Eucalyptus dunnii* desfolhadas artificialmente aos 30 dias após o plantio para simulação do ataque de *Acromyrmex*. Letras diferentes indicam diferenças significativas entre os tratamentos de acordo com o teste de Tukey's a 5% de probabilidade. Níveis de desfolha: 0: Controle, 1: 50%, 2: 75%, 3, 100% de desfolha.

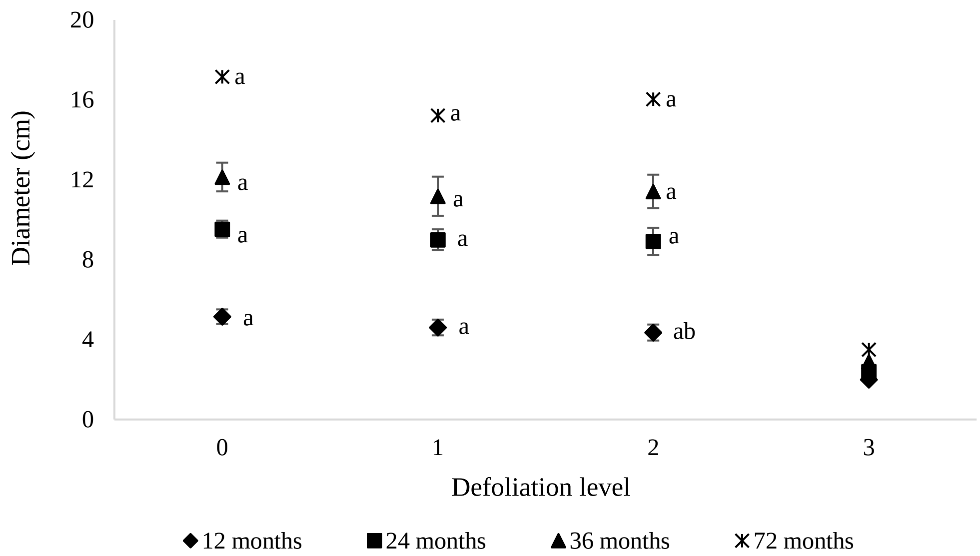

Figure 4. Diameter (mean ± CI) along time of *Eucalyptus dunnii* plants artificially defoliated at 30 days after planting for simulation of leaf-cutting ant attack. Different letters indicate significant differences among treatments according to Tukey's test, at 5% probability. Defoliation level: 0: Control, 1: 50%, 2: 75%, 3, 100% defoliation.

Figura 4. Diâmetro (média ± IC) ao longo do tempo de plantas de *Eucalyptus dunnii* desfolhadas artificialmente aos 30 dias após o plantio para simulação do ataque de *Acromyrmex*. Letras diferentes indicam diferenças significativas entre os tratamentos de acordo com o teste de Tukey's a 5% de probabilidade. Níveis de desfolha: 0: Controle, 1: 50%, 2: 75%, 3, 100% de desfolha.

DISCUSSION

Only 100% defoliation led to losses in *E. bentammii* plant growth soon after defoliation. The reduction of the photosynthetic area promotes a physiological breakdown of defoliated plants, decreasing carbohydrate production which leads to nutrient shortage. Immediately after defoliation, photoassimilates are allocated for the emission of new leaves, causing growth loss (FREITAS; BERTI FILHO, 1994). However, *E. bentammii* plants recovered after twelve months, with no difference in growth in height and diameter in relation to plants without defoliation. Studies carried out with other eucalyptus plants have shown significant losses in the development of plants submitted to 100% defoliation. In *E. grandis*, a single defoliation of 100% at 56 days after planting, results in difference in plant growth at the end of the cycle (92 months), with a reduction of 18.9% in diameter and 12.0% in height, in relation to the treatment without defoliation (MATRANGOLO *et al.*, 2010). Another study found that 100% defoliation in *Eucalyptus urophylla* S.T. Blake led to 42% reduction of volume at 4.6 years (ZANÚNCIO *et al.*, 1999). In the present study, 100% defoliation in *E. dunnii* resulted in significant plant mortality, and the plant that survived this defoliation level had significant reduction in its development. Thus, severe defoliation can cause significant losses in the growth of eucalyptus plants (FREITAS; BERTI FILHO, 1994).

However, secondary agents, such as the vigor of the plant before defoliation, plant age, site, soil moisture, spacing, stress hydric and climatic conditions can influence the survival of plants (CAFFARINI *et al.*, 2014; OLIVEIRA *et al.*, 2014). *Eucalyptus benthammi* plants were much taller (40 cm, on average), and therefore more vigorous than *E. dunnii* plants (27 cm, on average) at 30 days after planting. Therefore, regionalized studies are necessary to assess the losses that defoliation can cause in each situation and in each plant species, as well as to carry out the monitoring of defoliated plants during a long time after defoliation, preferably during the entire productive cycle of the forest (CANTARELLI *et al.*, 2008; OLIVEIRA *et al.*, 2014). In this study, the evaluation of plant growth until 72 months after planting proved that plants could recover over time, showing consistent and more accurate results, as it was evaluated almost until the end of the cycle.

A decline in productivity and, consequently, in revenues, can be great according to the intensity of defoliation, which can make economically unfeasible to maintain areas with total defoliation (MATRANGOLO *et al.*, 2010). Then, leaf-cutting ant control is fundamental to avoid attacks of these insects in forest plantations (ZANETTI *et al.*, 2014). In the present study, *E. benthammi* plants was more vigorous and recovered within twelve months when was 100% defoliated at 30 days after planting. However, the same did not occurred in *E. dunnii*, which suffered with this defoliation level. In case of leaf-cutting ant attack on plants at 30 days after planting, it is suggested to replant *E. dunnii* 100% defoliated to avoid losses in the end of the forest cycle.

Also, regionalized studies are necessary to assess the losses that defoliation can cause in each situation and in each plant species, as some species, such as *E. benthammii* 100% defoliated at 30 days after planting, can recover along time. For *E. benthammii* it is suggested to evaluate the vigor of the 100% defoliated seedlings, as they can recover over time.

CONCLUSION

- Only 100% defoliation can lead to losses in *E. dunnii* plant growth.
- *Eucalyptus benthammii* 100% defoliated at 30 days after planting, can recover along time.

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