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CROP PRODUCTION

Manihot leaf abscission induced by ethrel

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ABSTRACT. This study aimed to assess the impact of ethrel spraying on cassava leaf abscission at different stages of cultivar development. Two experiments were performed using randomized block designs, with 15 total treatments (three repetitions of five conditions) arranged according to the factorial scheme 3 x 5. Experiment 1 (ethrel application in the vegetative development period) consisted of three seasons (November 2013 and 2014, January and February 2014) and four concentrations of ethrel (1,500, 3,000, 4,500, and 6,000 ppm) and a control (no application); Experiment 2 (application in pre-harvest period) consisted of three periods of ethrel spraying (20, 40, and 60 days before harvest), four ethrel concentrations (1,500, 3,000, 4,500, and 6,000 ppm) and a control (without ethrel). The characteristics evaluated included the number of fallen leaves at 20 and 70 days after application, total leaf dry mass and foliar degreening. The effects of different concentrations and times of application of ethrel were significant during the first seven days. Ethrel application during the second period of vegetative development did not affect the number of fallen leaves over a period of 70 days.

Keywords: *Manihotesculenta*Crantz; leaf fall; plant growth regulator.

Abscisão foliar em plantas de mandioca induzida por ethrel

RESUMO. Este trabalho teve por objetivo avaliar o impacto da aplicação de ethrel na abscisão foliar em plantas de mandioca em diferentes estágios de desenvolvimento da cultura. Foram conduzidos dois experimentos utilizando o delineamento em blocos casualizados, com quinze tratamentos, três repetições, arranjados segundo o esquema fatorial 3x5. O experimento 1(aplicação de ethrel no desenvolvimento vegetativo) consistiu de: três épocas de aplicação (novembro/2013, janeiro/2014 e fevereiro/2014) e quatro concentrações de ethrel (1.500, 3.000, 4.500 e 6.000 ppm) e uma testemunha (sem aplicação); o experimento 2 (aplicação de ethrel em pré colheita) foi constituído por três períodos de aplicação (20, 40 e 60 dias antes da colheita), quatro concentrações de ethrel (1.500, 3.000, 4.500 e 6.000 ppm) e uma testemunha (sem aplicação). As características avaliadas foram a abscisão foliar aos sete, vinte e setenta dias após aplicação, massa seca foliar total, índice de velocidade de abscisão e desverdecimento foliar. O ethrel induz elevação da velocidade de abscisão de folhas, independente do estágio de desenvolvimento da cultura da mandioca. O efeito das diferentes concentrações e épocas de aplicação de ethrel é expressivo durante os primeiros sete dias. A aplicação de ethrel durante o segundo período de desenvolvimento vegetativo não afeta o número de folhas que sofrem abscisão durante um período de 70 dias.

Palavras-chave: *Manihot esculenta* Crantz; queda de folhas; regulador de crescimento.

Introduction

Leaf abscission is an important avoidance mechanism during water stress periods, which have become increasingly frequent and prolonged. The importance of this phenomenon is significant in cassava cultures, whose physiological resting period occurs at least once during cultivation (Liao et al., 2016). According to Nuwamanya et al. (2015), most varieties of cassava that are sensitive to leaf abscission, induced by factors such as water restriction and high temperature and associated with the precocity of foliar recomposing of a canopy after stress, have a higher rate of harvest.

Our understanding of the cassava leaf abscission process has a broad and well-grounded knowledge base, and there has been strong interest in topics related to inducers of this phenomenon, such as water stress (Zhang et al., 2010; Duque & Setter, 2013), shortened photoperiod (Fagundes et al., 2009), and low (Schons, Streck, Kraulich, Pinheiro, & Zanon, 2007) and high temperatures (Akparobi, Tobih, Togun, Ekanayake, & Oyetunji, 2001).

In defining the occurrence of several environmental factors, the localization and planting system are crucial to the understanding of the interaction between natural leaf abscission and periods of vegetative development and physiological rest in plants. The Conquista Plateau, Southwest of Bahia State, is characterized by an altitude of 900 to 1000 m, an average annual precipitation of approximately 700 mm, and mild conditioning temperatures for cassava cultivation as compared to other regions. According to Lopes, Viana, Matsomoto, Cardoso Júnior, and São José (2010) the specific climate of this region results in a vegetative development period interspersed by a physiological rest period in each year of cassava cultivation, characterized by the occurrence of leaf abscission induced by water restriction and decreasing photoperiods and air temperatures. As the period between planting and root harvest lasts approximately 18 months, the cultivation cycle consists of at least of two periods of vegetative development and two periods of physiological rest.

As the tuberization process occurs simultaneously with cassava development, organs from the shoot and root compete for the photoassimilate partition (Mitprasat, Roytrakul, Jiemsup, Boonseng, & Yokthongwattana, 2011). Theplant shoot management approach may affect the photoassimilate distribution, highlighting the importance of this intervention. Hence, when root ontogeny is determined under favorable environmental conditions, a slight reduction of the excessive vegetative vigor of shoots through the application of growth retardants, such as onium and triazoles, could extend to root growth (Gomathinayagam, Jaleel, Laksmanan, & Panneerselvam, 2007; Medina, Burgos, Difranco, Mroginski, & Cenóz, 2012). However, if shoot growth restriction is intense, through drastic pruning for example, the breaking of vegetative bud dormancy could promotes foliar development, and thus restrict root production and quality (Oliveira et al., 2010; Andrade, Viana, Cardoso, Matsumoto, & Novaes, 2011).

During the final period of culture, reduction in shoot vigor is beneficial, primarily viathe slowing of root post-harvest deterioration (Burgos, Cenóz, López, & Rodríguez, 2005; Salcedo & Siritunga, 2011), which has a strong relationship between root sugars and starch (Oirschot et al., 2000) and a reduction in scopolet in levels (Salcedo & Siritunga, 2011). For cassava culture, intense defoliation, caused by 600 and 800 ppm ethylene treatments 60 days before the anticipated harvest date, were previously shown to elevate root quality and yield (Song, Qin, Feng, Li, & Zheng, 2012).

The defoliation induced by ethrel (ethylene-releasing compound) treatment has been utilized in many crops, such as sweet potatoes (Wang, Ramón, Main, & Shankle, 2013; Clark et al., 2013), vines (Costa, Scarpare Filho, & Fidelibus, 2015; Souza et al., 2015) and citrus (Torregrosa, Porás, & Martin, 2010), due to the efficiency and speed of the process, resulting in higher quality and yield. Leaf abscission, as a senescence process, has a climacteric respiration pattern similar to fruits; however, the leaf sensitivity to ethylene is lower in comparison to fruits (Gan, 2010).

The possible management of leaf abscission intensity by developmental stage and calibration of ethrel concentration application is an important strategy that requires further development, because the knowledge of leaf abscission induced by growth regulators remains in its infancy for cassava crops.

As described above, this work was developed with the aim of evaluating the impact of ethrel spraying in cassava leaf abscission when applied in the vegetative development and pre-harvest periods.

Material and method

This study was performed via two experiments in the municipality of Vitória da Conquista, Southwest of Bahia State, Brazil (14°51' S; 40°50' W), at 941 m altitude, from January 2013 to July 2015, in a dystrophic yellow latosol. Chemical characteristics of 0-20 soil layer sin Experiment 1 included: pH (water) = 6.4; Al^{3+}(cmol $_c$ dm^{-3}) = 0.00; H^{+}+ Al^{3+}(cmol $_c$ dm^{-3}) = 2.0; Ca^{2+}+ Mg^{2+}(cmol $_c$ dm^{-3}) = 4.00; Ca^{2+}(cmol $_c$ dm^{-3}) = 2.90; K^{+}(cmol $_c$ dm^{-3}) = 0.51 and P (mg dm^{-3}) = 20.0; in Experiment 2, soil characteristics included: pH (water) = 5.4; Al^{3+}(cmol$_c$$dm^{-3}$) = 0.10; H^{+}+ Al^{3+}(cmol $_c$ dm^{-3}) =1.8; Ca^{2+}+ Mg^{2+}(cmol $_c$ dm^{-3}) = 2.4; Ca^{2+}(cmol $_c$ dm^{-3}) = 1.6; K^{+}(cmol $_c$ dm^{-3}) = 0.26 and P (mg dm^{-3}) = 10.0

According to Köppen, the climate of this area is classified as Cwa (highland tropical), with 717 mm of annual average precipitation concentrated in November to March, and an annual median temperature of 19.6°C, with a maximum and minimum average temperature of 23.5°C and 15.1°C, respectively (SEI, 2010). Median, maximum and minimum monthly average temperatures and accumulated rainfall for the duration of the experiments (Figure 1) were recorded.

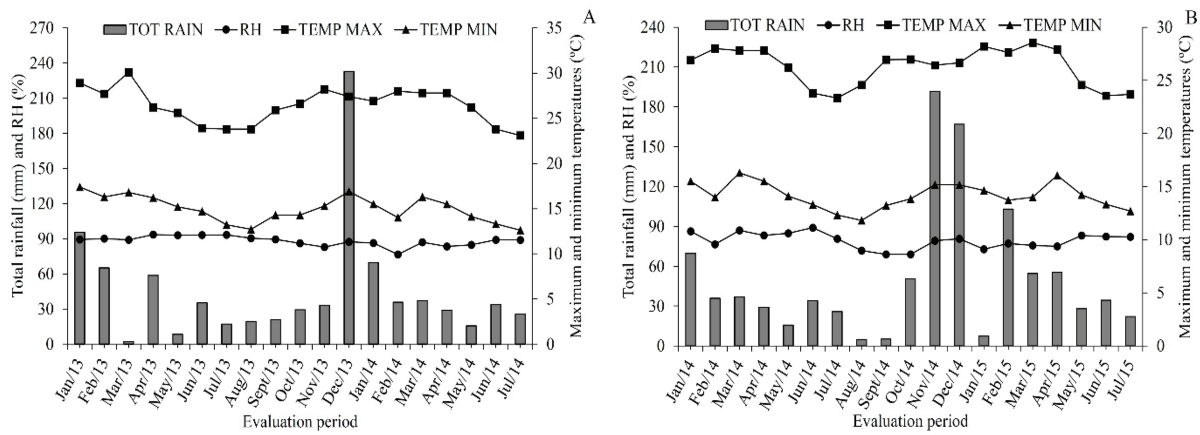

Figure 1. Precipitation (mm), atmospheric relative humidity (%), and maximum and minimum monthly average temperatures (°C) of the periods January2013 – July 2014 (A) and January2014 – July2015 (B).

Fonte: NationalInstituteofMetereology – INMET/ Vitória da Conquista, Bahia State, Brazil (2017).

For Experiment 1, according to the soil characteristics, liming and fertilizing practices were not required, and we performed only cover fertilization 60 days after vegetative sprouting with urea (40 kg ha^{-1} of N). For experiment 2, liming was not necessary, only fertilization at the initial planting with simple superphosphate (40 kg ha^{-1} of P2O5) and cover fertilization, as in Experiment 1.

In the soil, rows were securely created with a plow with a groove scarifier attached to a tractor, spaced 1 m apart, for the initial planting of Experiment 1 in January 2013 and Experiment 2 in January 2014. The stem cuttings utilized in the initial planting were obtained from healthy plants of the Caitité variety from the cassava germplasm collection of Universidade Estadual do Sudoeste da Bahia (UESB), whose roots are designated for industrialization. The initial planting was performed soon after the stem cuttings were obtained, and they were distributed every 0.60 m within each furrow. The cuttings were obtained from the middle-third part of stem and were approximately 20 cm long and 2 to 3 cm in diameter, with straight cuts on both ends and eight vegetative dormant buds.

In Experiment 1, plants were sprayed with soluble concentrated ethrel (240 g L^{-1} of 2-cloroetil phosphonic acid) at the second vegetative development period that preceded harvesting, in November 2013, January 2014, and February 2014 [at 300, 345, and 390 days after emergence (DAE), respectively]. In Experiment 2, plants were sprayed with ethrel during the second physiological rest period, at 480, 500, and 520 DAE [at 20 (05/26/2015), 40 (06/15/2015), and 60 days (07/06/15) prior to harvesting]. The spraying was performed at the end of the evening with a costal CO_2 pressurized sprayer (2 kgf cm^{-2}) coupled to a bar containing two flat jet tip nozzles (110.02 VS), with syrup consumption equivalent to 200 L ha^{-1}. The root harvest for Experiment 1 was performed in July2014, and for Experiment 2 in July 2015, 18 months after planting, with the aid of a anual practices.

Experiments 1 and 2 were conducted in randomized blocks with fifteen total treatments (three replicates per treatment), arranged according to a factorial design of 3 x 5. Experiment 1 included three developmental stages (November 2013, January 2014, and February 2014, during the second stage of vegetative development) and tested four ethrel concentrations (1,500, 3,000, 4,500, and 6,000 ppm) with a control (no ethrel spraying). Experiment 2 included a combination of three spraying dates (20, 40, and 60 days before harvesting, at the end of the second physiological rest period) and the same four ethrel concentrations as used in experiment 1. For both experiments, each experimental plot (27. $0m^2$), was characterized by four rows of plants, with 60 plants (of which 26 were considered useful), in a 15.6 m^2 area.

We evaluated the number of fallen leaves, designated in this study as foliar abscission (FA), during the period 7 to 70 days after treatment (Experiment1) and 7 to 20 days after treatment (Experiment 2), the total leaf dry mass (TLDM), the foliar degreening and abscission velocity index (AVI) (Experiment 2).

To delimit the soil area forfallen leaf sampling inside each useful parcel, four wooden beams were used to post a polyethylene screen 1.20 m tall, spanning an area of 3.8 m^2, containing six plants. We determined an abscission speed index (ASI) as the sum of the daily number of fallen leaves collected within the sampling soil area during the evaluation period after ethrel application. After harvest, leaves were carried to an air circulating oven and maintained at 65°Cuntil they reached a constant weight, which was determined to be the dry weight mass.

The ASI was calculated from an adaptation of the equation described by Maguirre (1962) for quantitative studies of germination speed index. The index was obtained from the sum of the number of leaves thatreached abscission in a single day divided by the number of the day when abscission occurred relative to the application of ethrel, during a 70-day period for Experiment 1 and during a 20-day period for Experiment 2, as described below:

$$\mathrm{ASI} = \frac{\text{Number of fallen leaves at first day after treatment}}{\text{Day of first count}} + \frac{\text{Number of fallen leaves at day n after treatment}}{\text{Day of final count (n)}}$$

Leaf degreeningwas calculated from SPAD index readings using four fully expanded leaves in the middle-third part of two plants in each plot, as measured via a portable chlorophyllmeter (SPAD 502, Minolta, Japan). For the calculation of leaf degreening, we used the following equation: 100-[(SPAD index reading at last evaluation before leaf fall x 100) / SPAD index reading after ethrel application]. This term is usually utilized for color alterations related to chlorophyll degradation in leaf senescence and fruit ripening period (Lira et al., 2014).

Statistical analysis was performed using SAEG software (System for Statistical Analysis and Genetics), version 9.1, by variance analysis at 5% probability. The means were classified by Tukey's test at 5% probability, and the relationships between dependent variables and ethrel concentrations in polynomial models were defined by regression variance analysis at 5% probability. The selected mathematical functions used to express the independent variable effects on independent variables obeyed the greater coefficient of determination and demonstrated superior biological behavior for the feature.

Result and discussion

In Experiment 1, performed during the second vegetative stage, ethrel spraying affected all traits evaluated. With the exception of AF70, ethrel affected almost all evaluations, with interactions between time of application and ethrel concentration observed only for leaf abscission at 7 days post-spraying (LA7) and abscission speed index (ASI) (Table 1). In Experiment 2, performed during the second physiological rest stage, there was an interaction of factors evaluated for the sum of the fallen leaves at 7 (LA7) and 20 days (LA20) after treatment and for the abscission speed index (ASI). All features were affected by ethrel concentration and, with the exception of DF, the application date also affected the majority of the parameters evaluated (Table 1).

Table 1. Summary of variance analysis and coefficient of variation (CV) of fallen leaf number at 7 (LA7), 20 (LA20), and 70 days (LA70) after treatment, leaf degreening (LD), total leaf dry mass (LDM) and abscission speed index (ASI) for the Caitité variety cassava at different times of application (T) and ethrel concentrations (C).

		Mean Square ($x10^3$)									
		Experiment 1					Experiment 2				
Parameter	DF	LA7	LA2070	LD	LDM	ASI	LA7	LA70	LD	LDM	ASI
T	2	679.7**	587.2**	2.8**	524.5**	247.9**	301.7**	59.1**	17.4	223.1**	32.7**
C	4	246.5**	56.0	1.5**	8.5	46.9**	214.9**	45.1**	202.3*	52.5**	11.2**
T*C	8	22.0*	11.7	0.2	11.4	8.6**	46.6**	8.8**	20.2	6.6	4.0**
Block	2	23.6	54.9	0.1	1.6	3.7	9.1	1.8	203.4*	4.6	0.4
Residue	28	9.3	24.0	0.2	5.8	2.2	8.4	2.1	64.2	4.4	0.5
CV (%)		22.7	22.7	27.9	10.3	27.7	26.0	23.2	29.2	13.0	31.0

*Statistically significant at 5% probability, by F test.

In January, major leaf abscission was verified 7 days after treatment (Figure 2A). However, for controlsand plants treated with 3,000 ppmethrel, there were no differences between January and February. Increasing ethrel concentrations induced a significant increase in the number of fallen leaves between January and February ethrel applications.

It is generally agreed that leaf physiological maturity occurs only when the maximum leaf expansion isreach. Due to waterrestriction, which occurred in November 2013, maintenance of leaves in the juvenile state was related to a higher capacity for leaf retention. According to Helal, Eisa, and Attia (2013), the water availability limits the leaf cellular elongation, induces biochemical changes and reduces the ethylene sensitivity. Young leaves are characterized by an intense auxin gradient from leaf blade to stem, which reduces abscission zone cell sensitivity, inhibiting leaf shedding (Scarpella, Barkoulas, & Tsiantis, 2010).

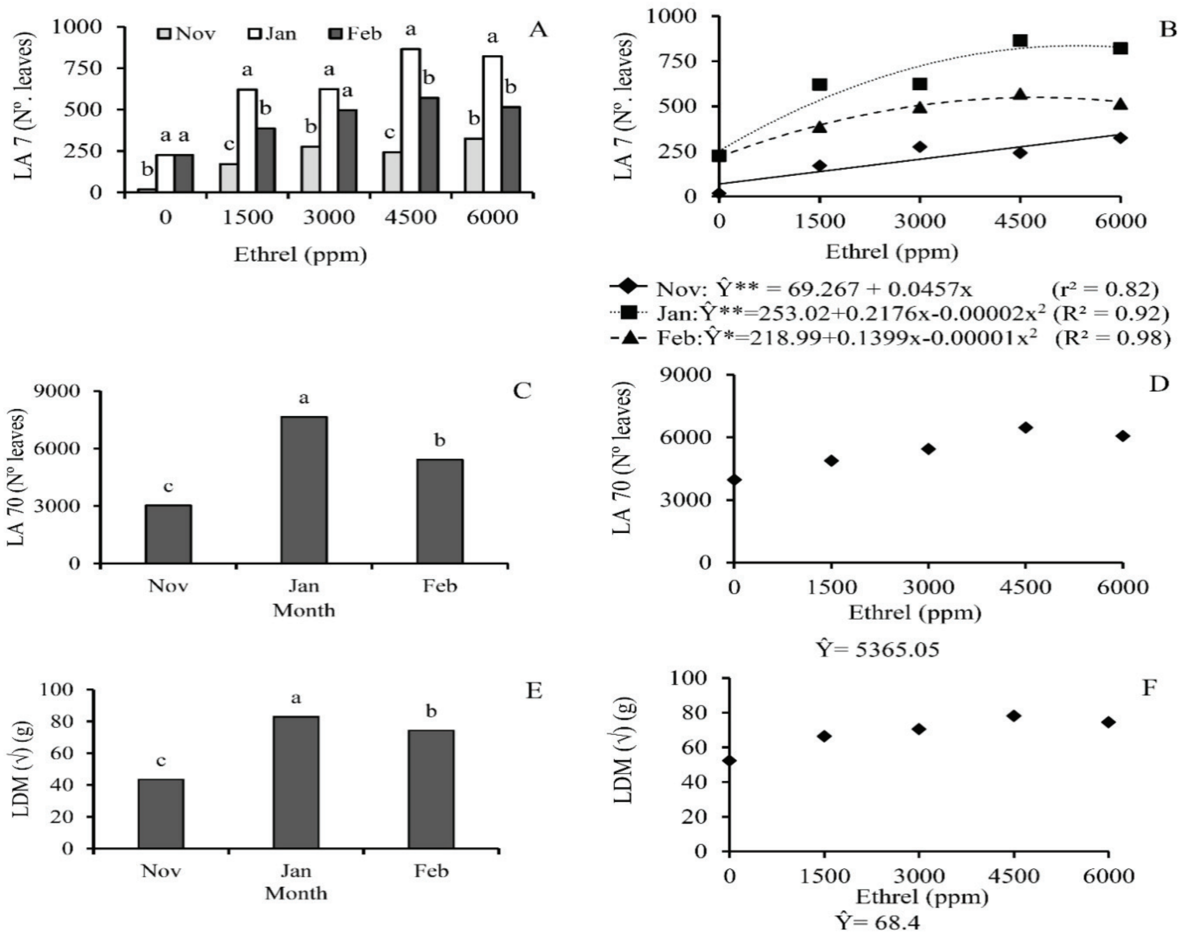

Figure 2. Number of fallen leaves in the soil at 7 (LA7) and 70 days (LA70), and total leaf dry mass (LDM) according to application dates (A, C, and D) and ethrel concentrations (B, E, and F) in two culture cycles of Caitité variety cassava plants. Means followed by the same letter do not differ from each other by the Tukey's test($p \leq 0.05$). **and*, significant difference by regression variance analysis at $p \leq 0.01\%$ and $p \leq 0.05$, respectively.

It is possible that the rainfall and temperature increases in December 2013 and January 2014 (Figure 1A) favored the elongation of young leaves in January 2014 and February 2014, representing an intense resource drain responsible for redistributing growth factors, such as nutrients and photoassimilates from source organs such as the mature leaves and roots. This mobilization may have resulted in changes in plant metabolism through the activation of molecular markers in the abscission zone, which promoted leaf abscission in the cassava. Another consideration is the natural occurrence of senescence. Abscission can be regarded as a final result of the leaf senescence process, occurring as the organs advance in age (Meyer et al., 2014).

When LA7 was evaluated in relation to ethrel concentrations (Figure 2B), we observed a linear increase in the effect of the treatment in November 2013, as well as an exponential increase in effect in January 2014 and February 2014.The major angular coefficient and the maximum number of fallen leaves had a minor concentration in the last two months of ethrel application (January 2014 at 5,438 ppm and February 2014 at 4,662 ppm), and were associated with intense vegetative development in these periods. This vegetative vigor was associated with water availability and favorable temperatures, resulting in major leaf elongation and an increased capacity for response to ethrel.

A difference was verified between periods of ethrel application, with the highest abscission number (FA70) and total leaf dry mass (LDM) of fallen leaves occurring in plants treated in January 2014 (Figure 2C and D). Increasing rainfall occurring during this period was associated with greater vegetative vigor, which favored ethylene effects, resulting in higher leaf abscission. The ethylene response requires that the organs are responsive to phytohormones, as well as a minimum concentration sufficient to bind to the receptor (Agarwal, Choudhary, Singh, & Arora, 2012).

For LA70 and LDM, there were no differences between ethrel concentrations (Figure 2E and F). Climacteric respiration is a well-known phenomenon in fruit maturation; however, it can also be observed in leaf blades as part of an abscission response pattern induced by ethylene self-catalysis (Gan, 2010). 70 days after regulator spraying, it was observed that the regulator interfered only in leaf abscission speed and did not change the total number of fallen leaves (Gan, 2010). In the present study, the elevation of treatment concentrations with leaf abscission was verified in climacteric fruits. In fruits characterized by climacteric respiration, the response to increasing levels of ethylene anticipates the rise in respiration, and had no effect on the intensity of this process (Mohamed-Nour& Abu-Goukh, 2010).

At different stages of application in the two experiments analyzed, we observed an increasing number of fallen leaves in relation to ethrel concentration. In the first experiment, for the relationship between ASI70 and regulator concentrations, we utilized a linear model for the three seasons of application (Figure 3B). In Experiment 2, when we evaluated the ASI20,we noted that the regulator induced values that were always superior to control plants (Figure 3D), best described byan increasing linear model for treatment at the 60-day application period before harvest, and a quadratic model for ethrel application in the periods 20- and 40-days prior to harvest.

In Experiment 1, the highest abscission speed index (ASI70) was observed for cassava plants treated with ethrel in January 2014 in comparison to the other dates (Figure 3A). Although it was observed that IVA70 increased for all application dates, the higher total values and greatest increases were observed for plants treated with ethrel in January 2014.

In Experiment 2, differences were observed for concentrations above 3,000 ppm of ethrel, with faster falling of leaves observed when the regulator was sprayed 20 days before root harvest (Figure 3C). A similar relationship was also noted between IVA20 and ethrel concentrations applied 20 days before harvest (Figure 3D). As mentioned earlier, this could possibly be associated with the cassava leaf physiological maturity. According to Edelman and Jones (2014), the leaf developmental stage influences sensitivity to ethylene.

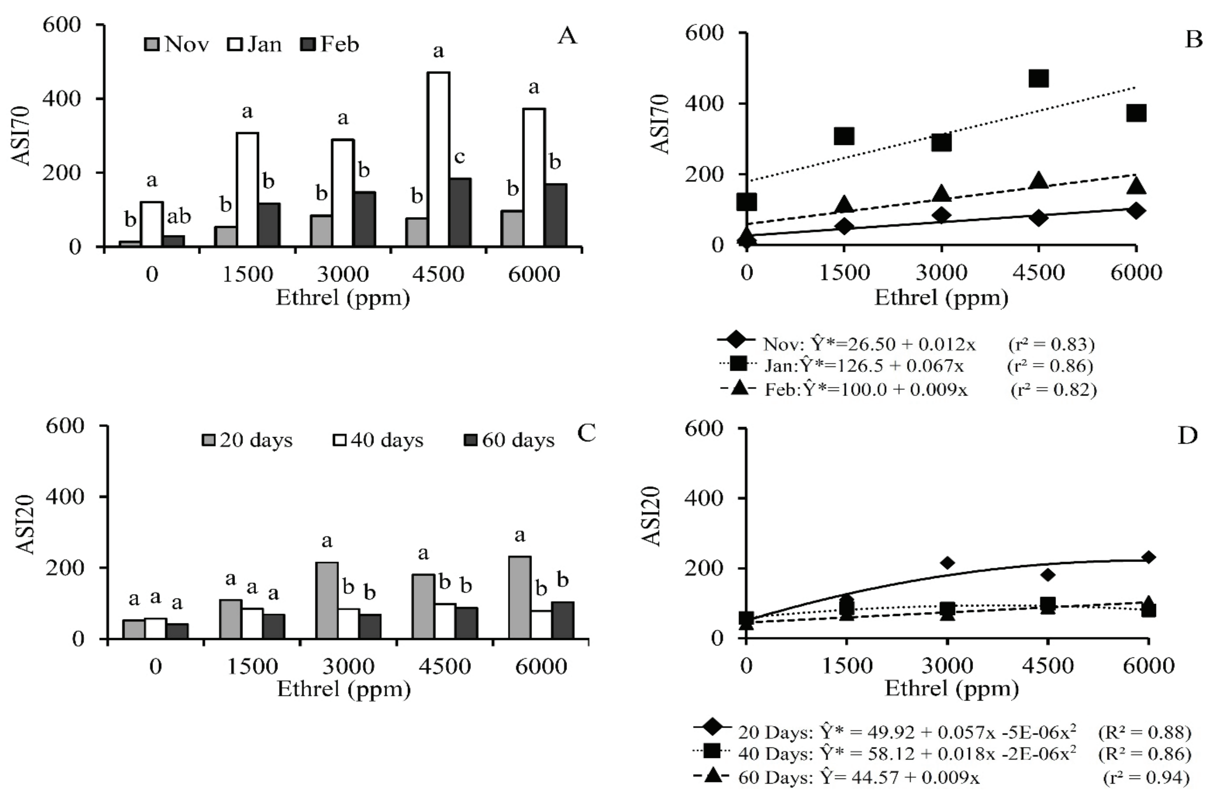

Figure 3. Abscission speed index at 70 days (ASI70) and at 20 days (AVI20) after ethrel spraying relative to application time (A and C) and ethrel concentration (B and D) during the two cultivation cycles of cassava plants of the Caitité variety. For each ethrel concentration, means followed by the same letter do not differ from each other by the Tukey's test ($p \leq 0,05$). ★★and★, significant by regression variance analysis at $p \leq 0.01\%$ and $p \leq 0.05$, respectively.

For the two evaluation dates, although we observed an increase in the number of fallen leaves, the maintenance of a relatively standard level was verified among the treatments. Therefore, seven days after ethrel application could be considered a standard time for leaf abscission that could be utilized in future studies, optimizing the analysis and evaluation process.

When regulator spraying was performed during the pre-harvest period, the last physiological resting period of the cassava plants, differences between application dates were analyzed only for 3,000, 4,500, and 6,000 ppm of ethrel for leaf abscission numbers at 7 (LA7) and 20 (LA20) days after treatment. We observed a greater accumulation of fallen leaves when the regulator was applied during the closer period of harvesting (20 days), which was characterized by the lowest temperature (Figure 4A and B).Greater leaf dry mass weight was also observed for plants that underwent ethrel treatment 20 days before harvest (Figure 4E).

Leaf abscission is a process affected by several environmental factors, such as air temperature, water availability, pathogens, nutrition and mechanical injuries (Alves & May-De-Mio, 2008; Silva, Lilagres, Silva, Nick, & Castro, 2011). Minimum temperatures below 13 to 15°Chave been reported as leaf abscission induction factors in cassava plants (Singh, Khurana, Chakrabarti, Mukherjee, & Nedunchezhiyan, 2013; Moreira et al., 2014; Soares et al., 2015). The leaf abscission process overlaps with leaf emission as the temperature decreases, which can result in complete leaf abscission of the canopy. During this study, we recorded a minimum temperature of 12.3°C and a rainfall of 25.0 mm at 20 days before the harvest (Figure 1B). Thus, the greater number and weight of leaf dry mass observed in plants that underwent ethrel treatment 20 days before the harvest date may have been related to the response to favorable conditions for the leaf abscission process, mediated via growth hormones, mainly auxin, ethylene, abscisic acid and gibberellin; these can activate abscission zone enzymes in different organs and are highly correlated with changes in environmental conditions, such as temperature and water availability (Botton et al., 2011).

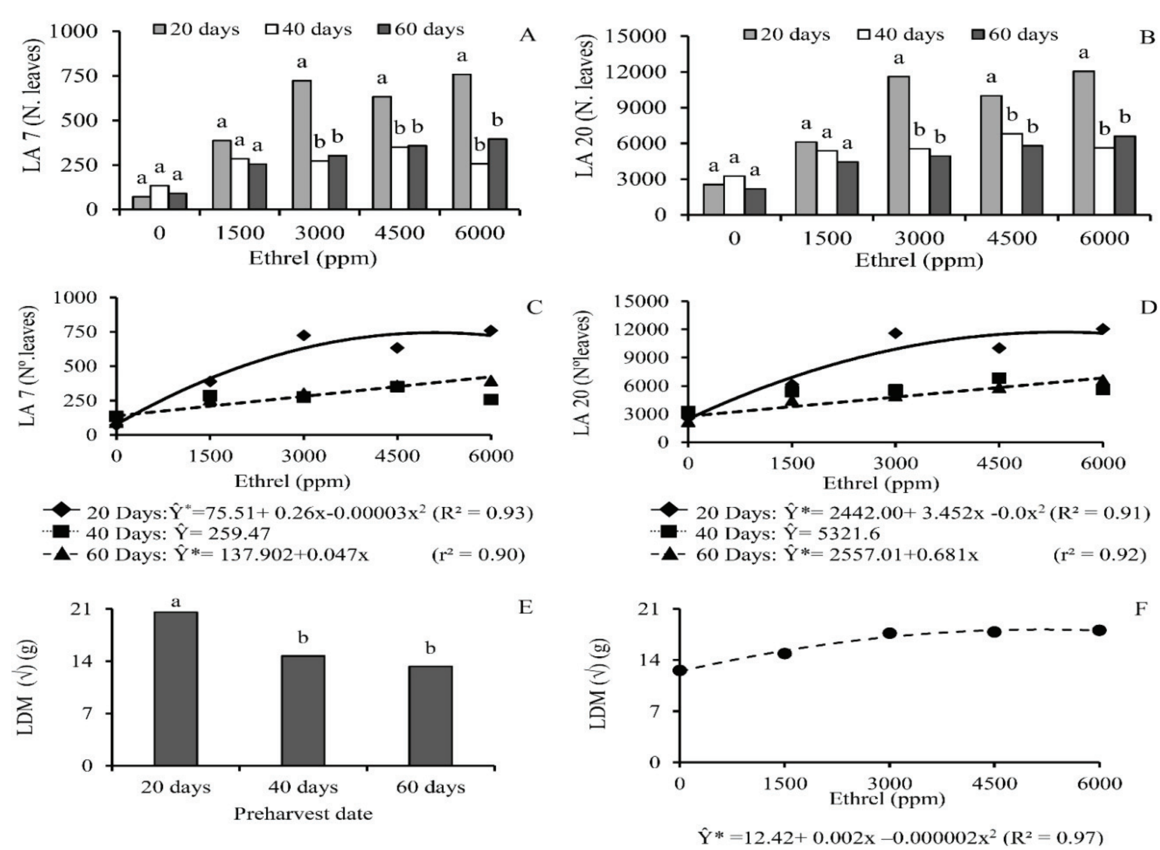

Figure 4. Fallen leaf number at 7 (LA7) and 20 days after treatment (LA20), and total leaf dry mass 20 days after treatment in relation to the date of application and ethrel concentration (C. D and F) during the pre-harvesting period in two culture cycles of the Caitité variety cassava plants. Means followed by the same letter do not differ from each other by the Tukey's test($p \leq 0.05$). **and*, significant by regression variance analysis at $p \leq 0.01\%$ and $p \leq 0.05$, respectively.

For fallen leaf number and dry mass evaluated at 7 days prior to harvesting and at harvesting of plants that were treated at 20 days before the harvest date in relation to ethrel concentration, we found that the effects could be represented by a quadratic model, with maximum points at 4,365, 3,452, and 5,000 ppm, respectively (Figure4C, D, and F). A linear increasing effect was detected for the relationship between FA7 and FA20 and ethrel concentration in plants treated at 60 days before harvesting, with maximum points at 2,910 and 4,054ppm, respectively. There were no models that defined a relationship between FA7 and FA20 and ethrel treatment at 40 days before harvesting. Despite the quantitative differences between the evaluation dates (seven and 20 days after treatment), the tendency towards a relationship between leaf abscission and ethrel concentration was similar.

The largest amplitude observed for the intensity of leaf falling in the period closer to harvest (20 days) was associated with low water availability and minimum temperature decreases, which occurred in the experimental period. According to Duque and Setter (2013), cassava plants subjected to at least one week of water stress showed a higher index of foliar abscission in comparison to control plants. The decrease in minimum temperature alters the longevity pattern, intensifying the senescence and abscission processes and restricting the leaf blade expansion (Brown, Cavagnaro,Gleadow,& Miller, 2016; Singh et al., 2013). In this way, weather conditions were likely a determining factor to the increase in number of fallen leaves of these plants.

The highest values in leaf degreening (LD) were observed when ethrel was sprayed in January and February2014 in comparison to November 2013, during the vegetative development of the cassava (Figure 5A). The increasing water availability and temperature, which occurred in the previous month (December, 2013), likely contributed to stimulating growth, resulting in increases in the degreening index. Simão et al. (2013) demonstrated in the Mocotó, UFLA, bread of China and gold in the Valley varieties of cassava that there were higher SPAD index values in the vegetative development period, decreasing at the end of the cultivation cycle. Environmental conditions, such as water restriction and low temperatures, determined a strong induction of leaf abscission, resulting in fallen leaves with reduced chlorosis.

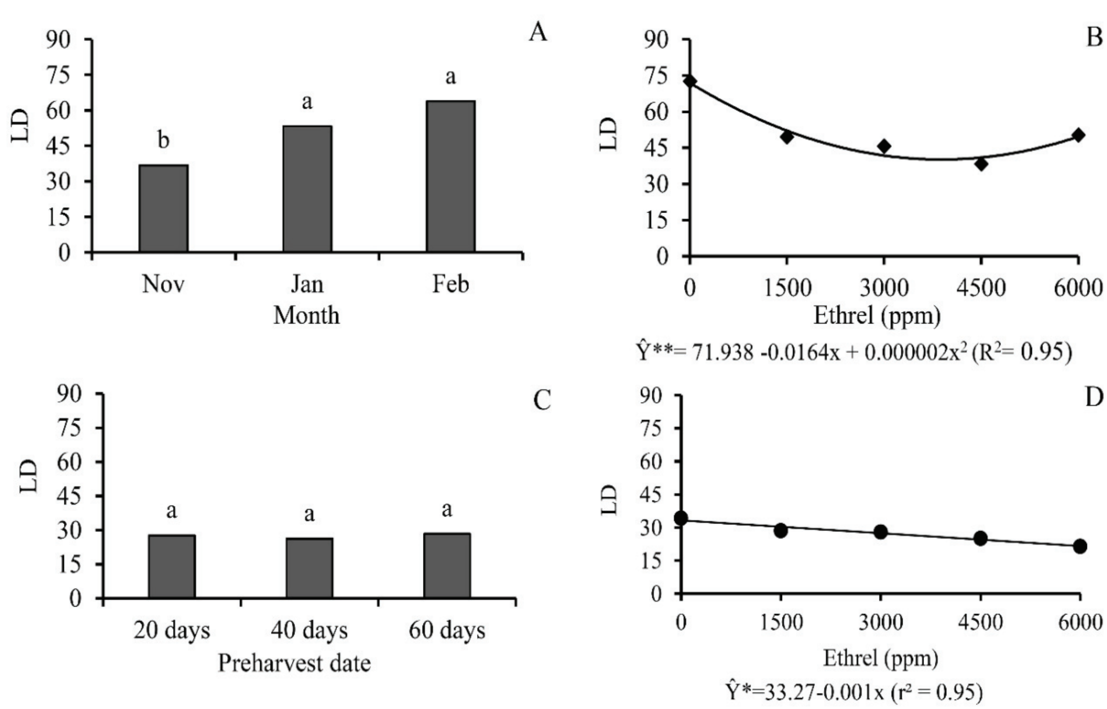

Figure 5. Leaf degreening (LD) in relation to application times and ethrel concentration during the vegetative development period (A and B) and preharvest period (C and D) of two culture cycles of cassava plants of the Caitité variety. Means followed by the same letter do not differ from each other by the Tukey's test($p \leq 0.05$). **and*, significant by regression variance analysis at $p \leq 0.01\%$ and $p \leq 0.05$, respectively.

For evaluating the DF in relation to ethrel concentrations, we determined a second-order polynomial model for treatment occurring in the vegetative development period of the cassava (Figure 5B), with an observed reduction in chlorosis in the range of 0 to 4,100 ppm of ethrel. For concentrations greater than 4,100 ppm, up to 6,000 ppm, there was an increase in LD; however, these values were still at a lower level in comparison to controls.

A similar effect was observed when ethrel was applied in the preharvest period (Figure 5D). In this stage, in contrast to the vegetative development period, the relationship between LD and concentration of the regulator was characterized by a continuous decrease for all concentrations of the regulator. LD reduction as a function of ethrel concentration was associated with cytokinin biosynthesis, which possibly delayed chlorophyll catabolism. In beans, it has been observed that ethrel application slows chlorosis, resulting in top whiteness, in a SPAD index through 125 days of cultivation (Campos, Ono, & Rodrigues, 2010). According to the same author, ethrel application reduced apical dominance, causing a greater number of branches; this is a strong indication that ethylene can induce the availability of cytokinins.

There were no differences in DF values of the cassava leaves among periods of ethrel application during the preharvest (Figure 5C). In this stage, leaf chlorosis is intense due to water restriction, as well as decreasing temperatures and photoperiods, leading to a smaller LD index.

Conclusion

Ethrel induction of leaf abscission depends on the developmental stage of cassava plants in which it is applied.

Concentration and application date of ethrelis related to the number of fallen leaves during the first seven days.

When ethrelis applied in the second period of vegetative development, there is no variation in the number of fallen leaves 70 days after treatment in relation to the regulator concentration.

Leaf degreening is affected by ethrel application date as a function of decreasing water availability and lower temperatures.

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