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Article

Development of Different Grass Species with the Use of the Growth Regulator Trinexapac-Ethyl

Desenvolvimento de Diferentes Espécies de Gramas com o Uso do Regulador de Crescimento Trinexapac-Ethyl

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ABSTRACT - The use of synthetic compounds to retard the growth of lawns aims at reducing the number of operations for their maintenance. Thus, the main goal of the present study was to evaluate the effect of different doses of trinexapac-ethyl on the development and rooting of five different grass species. Bahiagrass, St. Augustine grass, Japanese lawn grass, Bermuda grass and, broadleaf carpetgrass were grown in pots of 20 L. The experimental design was completely randomized, with four replications. Trinexapac-ethyl (g ha^{-1}) was applied at doses of 113+113, 226+113, 226+226, 452+113, 452+226, 452+452 in a sequential spraying, and also at single doses of 678 and 904. The second application was carried out 14 days after the first. On the day prior to applications all species were cut at 3 cm height. The phytotoxicity evaluations were performed weekly, and evaluations on plant height, number and height of inflorescences, biomass of aerial parts and roots were carried out 35 days after the second application. The trinexapac-ethyl application was effective in inhibiting the development of the Japanese lawn grass, Bermuda grass and broadleaf carpetgrass when applied in sequential doses higher than 226 g ha^{-1}, or single doses of 678 or 904 g ha^{-1} and, the visual appearance of lawns was not affected. In the case of St. Augustine grass and Bahiagrass species, the growth regulator did not promoted reduction in the growth of both grass species.

Keywords: *Paspalum notatum, Stenotaphrum secundatum*, *Zoysia japonica*, *Cynodon dactylon*, *Axonopus compressus*, plant growth regulator.

RESUMO - *A utilização de compostos sintéticos para retardar o crescimento de gramados tem como principal objetivo a diminuição do número de operações para a sua manutenção. Este estudo teve a finalidade de verificar o efeito de diferentes doses de trinexapac-ethyl, aplicadas sequencialmente, sobre o desenvolvimento e enraizamento de cinco espécies de gramas. As gramas batatais, santo agostinho, esmeralda, bermuda tifton 419 e são carlos, foram plantadas em vasos de 20 L. O delineamento experimental utilizado foi o inteiramente casualizado com quatro repetições, onde foram aplicadas as doses do trinexapac-ethyl em g* ha^{-1}*: 113+113, 226+113, 226+226, 452+113, 452+226 e 452+452 em pulverização sequencial e doses únicas de 678 e 904. Utilizou-se um intervalo de 14 dias entre as duas aplicações. Um dia antes de cada aplicação, todas as espécies foram cortadas a 3 cm de altura. As avaliações de fitointoxicação foram feitas semanalmente, e as avaliações de altura de plantas, número e altura de hastes florais e de biomassa da parte aérea e das raízes foram realizadas aos 35 dias após a segunda aplicação. O trinexapac-ethyl mostrou-se efetivo na inibição do desenvolvimento das gramas esmeralda, tifton 419 e são carlos quando aplicado em doses sequenciais superiores a 226 g* ha^{-1}*, ou em doses únicas de 678 ou 904 g* ha^{-1}*, e a aparência dos gramados*

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não foi alterada. Quanto às espécies de grama santo agostinho e batatais, o regulador de crescimento não proporcionou redução no desenvolvimento delas.

Palavras-chave: *Paspalum notatum*, *Stenotaphrum secundatum*, *Zoysia japonica*, *Cynodon dactylon*, *Axonopus compressus*, regulador vegetal.

INTRODUCTION

Controlling plant growth with the use of exogenous chemicals has intrigued researchers around the world since the mid-1940s, when the first compound commercially used for that purpose appeared: maleic hydrazide. Later, with the advent of chlorine choline chloride (CCC), the potential for controlling plant growth has gained in strength, and thus this compound became widely used during the 1960s and 1970s (Cooke, 1987).

Particularly in grasses, the use of compounds to retard plant growth was first reported also in the mid-1940s, and ethephon was the least phytotoxic among commercially available products at that time (Freeborg and Daniel, 1981). Subsequently, other products have also been used for this purpose, mainly maleic hydrazide, chlorflurenol and mefluidide. Among these three compounds, mefluidide gained special prominence by inhibiting plant growth for long periods at low doses and because it was less phytotoxic to roots (Watschke et al., 1977).

The use of the term “plant hormone” does not refer to nutrients, but to naturally occurring organic compounds produced in plants and which, at low concentrations, promote, inhibit or modify morphological and physiological processes. Growth regulators are synthesized substances that, when exogenously applied, trigger some actions that are similar to those of the known groups of plant hormones (Arteca, 1995). In the case of grasses, these synthetic compounds can be used with the main objective of obtaining individuals with all the characteristics that are common to the species, but in reduced size. Therefore, the ideal growth regulator for grasses is one that decreases plant height without reducing density or causing visible damage to the plants, such as necrotic spots of phytotoxicity, discoloration or thinning, maintaining a high quality of the treated area (Velini, 2003).

Following on from this, the use of growth regulators in grasses also aims at decreasing the number of cutting operations performed during the spring and summer growth period, which would indirectly act as a potential component for labor, fuel and equipment cost reduction. In addition, its use also eliminates the need of certain operations, such as raking, ridging, shiping, transport and disposal of crop residues resulting from trimming, which must be removed from lawns (Davis and Curry, 1991).

The use of growth regulators to promote the development of the root system would be another potential application objective and, consequently, improving the quality of the seedlings. Once again, this can make possible the production and commercialization of species whose carpet production is considered problematic due to the weakness of the root system, for example the broadleaf carpetgrass. The growth regulator has the task of driving photoassimilates to the root system, promoting root development (Steffens et al., 1985; Richie et al., 2001; Beasley et al, 2005). This increase in the root system probably can be obtained through the use of growth regulators that inhibit the formation of gibberellins, such as trinexapac-ethyl, which only inhibits cell elongation without interfering with the mitotic multiplication process (Marchi et al., 2013a).

The present study aimed at analyzing the effect of different doses of trinexapac-ethyl, sequentially applied, on the growth of aerial parts and roots of different grass species.

MATERIALS AND METHODS

The experiments, one for each grass species, were carried out in pots with a capacity of 20 L and kept in the full sun, with water available when necessary. The pots were filled with Red-Yellow Oxisol (Embrapa, 2013), whose physical characteristics are: clay (18,9%), silt (3,6%) and sand (77,5%), and the soil physical-chemical analysis revealed the follwing results: pH ($CaCl_2$) = 5,9; P_{resina} = 12 mgdm^{-3}; Ca = 27 mmol$_c$ dm^{-3}; Mg = 13 mmol$_c$ dm^{-3}; K = 9,6 mmol$_c$ dm^{-3};

Al = 0,3 $mmol_c$ dm^{-3}; H+Al = 32,9 $mmol_c$ dm^{-3}; CTC = 82,9 $mmol_c$ dm^{-3}; V = 60%; Fe_2O_3 = 176 g kg^{-1}. The major species studied were the following: bahiagrass (*Paspalum notatum*), St. Augustine grass (*Stenotaphrum secundatum*), Japanese lawngrass (*Zoysia japonica*), Bermuda grass tifton 419 (*Cynodon dactylon* x *C. transvaalensis*) and broadleaf carpetgrass (*Axonopus compressus*). St. Augustine, Japanese lawn, Bermuda grass and broadleaf carpet grass species were planted using plug-type seedlings, with three units per pot. The "plugs" are small-sized seedlings which have been germinated and grown in trays filled with substrate. The bahiagrass was planted using a set of plates directly obtained from the field, in the amount of one plate per pot. All grass species had good vegetative vigor, i.e., good development and establishment; one day before the first and second applications of the plant regulator, a standardization cut was made at 3 cm height.

The treatments were arranged in a completely randomized design with four replicates, in which the plant regulator trinexapac-ethyl (MODDUS) was applied in two sequential doses, within a 14 day interval between applications (Table 1). The use of two sequential doses in a 14 day interval aimed at optimizing the action of the growth regulator in the five species studied, as proposed by Marchi et al. (2013b).

The application time was determined when the plants had not yet fully filled the pot surface and, consequently, the roots had not yet fully filled the soil volume contained in the experimental unit. This procedure had the objective of avoiding intraspecific competition for space.

The application was made using a backpack sprayer equipped with a bar containing two flat spray nozzles TP 80.02 VS, spaced 0,50 cm apart. Todo o conjunto foi mantido à pressão constante de 200 kPa, The whole set was maintained at a constant pressure of 200 kPa, which provived a spray mix volume of 200 L ha^{-1}.

Visual evaluations of phytotoxicity were performed using a scoring range, where '0' corresponded to no injuries shown by plants and '100' to plant death (SBCPD, 1995). Within 35 days after the second application (DASA), plant height and the number and height of floral stems emitted when they occurred, as well as the mass of the aerial part and the root system were evaluated. The aerial part was cut using pruning shears, with the cuts made at ground level; afterwards, the entire root system was obtained by washing the substrate contained in the pots in running water on a 20-mesh sieve. The collected materials were placed individually into paper bags and kept in a forced-air circulation oven at 65°C until they reach a constant dry mass.

Table 1 - Trinexapac-ethyl doses used in the first and second applications on the different grass species evaluated. Botucatu/SP, 2013

Treatment	Dose (g i.a. ha^{-1})		Total
	1st application(1)	2nd application(1)	
trinexapac-ethyl	113	113	226
trinexapac-ethyl	226	113	339
trinexapac-ethyl	226	226	452
trinexapac-ethyl	452	113	565
trinexapac-ethyl	452	226	678
trinexapac-ethyl	452	452	904
trinexapac-ethyl	678	0	678
trinexapac-ethyl	904	0	904
Control	0	0	0

(1) Dose applied 14 days after the first spraying.

The results related to plant height, number and height of inflorescences and dry biomass of aerial parts and root system were subjected to analysis of variance by the F test, and their means were compared by Tukey's test (p>0.05), using the SISVAR 5.1 statistical software (Ferreira, 2011).

RESULTS AND DISCUSSION

The doses of trinexapac-ethyl that were applied did not trigger noticeable symptoms of injuries to the different species of grass evaluated, and thus their appearance was not altered. These results corroborate those found by Marchi et al. (2013b) and McCullough et al. (2007). JHowever, Marchi et al. (2015) observed high levels of phytotoxicity with the single applications of trinexapac-ethyl, at seven days after applications (AAD) in St. Augustine grass; at 56 DAA, when the 452, 678 e 904 g ha^{-1} doses of the growth regulator were applied, producing slight phytoxicity symptoms.

Marchi et al. (2014) also observed mild injury symptoms in Bermuda grass when trinexapac-ethyl was applied in sequential doses (113 + 113 g ha^{-1}) up to the 14 DASA (days after the second application), but no effects were observed from 21 DASA.

For St. Augustine grass (Table 2) no significant differences between the growth regulator doses tested were observed on the average height of the plants at 35 DASA. In a similar study with trinexapac-ethyl application for the same species, Marchi et al. (2015) observed a reduction of up to 61% at 28 DAA, which does not corroborate the results found by the present research; as for the accumulation of dry mass in the aerial part and in the roots of the St. Augustine grass, the application of the different doses of the growth regulator did not produce any effects. Regarding the aesthetics of the grass, the reduction in inflorescence populations or their absence improves the appearance of the landscape for a long period, besides also reducing the need for lawn mowing. St. Augustine grass, in this study, did not flourish (Table 2).

Table 2 **-** Mean values of plant height and dry biomass of the aerial part and roots of St. Augustine grass, at 35 days after the second application of trinexapac-ethyl. Botucatu/SP 2013

trinexapac-ethyl	Height	Floral stems	Dry Mass (g)	
(g ha^{-1})	(cm)	Number	Aerial part	Root
113+113[(1)]	5.87	0	20.53	105.43
226+113[(1)]	5.62	0	20.39	102.97
226+226[(1)]	5.00	0	19.77	103.21
452+113[(1)]	5.37	0	17.43	91.28
452+226[(1)]	5.37	0	18.69	79.89
452+452[(1)]	5.37	0	14.70	70.45
678	5.37	0	18.10	107.10
904	5.00	0	16.31	107.96
Control	6.05	0	22.52	109.39
$F_{treatment}$	0.89^{ns}	-	1.47^{ns}	0.87^{ns}
LSD	1.78	-	9.39	70.91
VC (%)	13.7	-	21.1	30.5

Means followed by the same letter in the column are not significantly different from each other by Tukey's test (p<0,05). [ns] not significant. [(1)] Dose applied 14 days after the first spraying .

As for the Japanese lawn grass, it was observed that all the doses of trinexapac-ethyl that were applied reduced the average height of the plants (Table 3). One should stress out the greatest reduction of approximately 53%, in the treatment with the sequential application of the 452 + 452 g ha^{-1} dose of the growth regulator. Marchi et al. (2013b) found in the field a reduction of approximately 73% at the height of the Japanese lawn grass at 28 DASA, confirming the results found in this study. Likewise, Ervin et al. (2002) and Qian and Engelke (1999) verified that the application of trinexapac-ethyl in *Zoysia japonica* and *Z. matrella* reduced plant height, which improved lawn quality.

Table 3 shows that the trinexapac-ethyl growth regulator, regardless of the dose and number of applications, significantly reduced the number and height of flower stems of the Japanese lawn grass; these reductions were less accentuated by the application of the 113 + 113 g ha^{-1} dose for the number of stems and 113 + 113 e 226 + 113 g ha^{-1} for the height of these stems. Some studies have shown that, with the application of trinexapac-ethyl, there may be a decrease or delay in the emission of inflorescences, maintaining the aesthetics of the lawn for a longer time (Johnson, 1990; Fry, 1991; Costa et al., 2009; Mittlesteadt et al., 2009; Marchi et al., 2013b).

Type II growth regulators, which is the case of trinexapac-ethyl, are not efficient in suppressing the emission of floral stems because they interfere with the biosynthesis of gibberellins, thereby reducing cell elongation (Marchi et al., 2013a; Marchi et al., 2014); however, in this study, with Japanese lawn grass, significant effects resulting from its application were observed at high doses.

Regarding dry biomass accumulation in the aerial part of the Japanese lawn grass, a significant reduction was observed with all treatments, particularly in comparison with the

Tabela 3 - Mean values of plant height and dry biomass of the aerial part and roots of the Japanese lawn grass, at 35 days after the second application of trinexapac-ethyl. Botucatu/SP 2013

trinexapac-ethyl ($g\ ha^{-1}$)	Height (cm)	Floral stems		Dry mass (g)	
		Number	Height (cm)	Aerial part	Root
113+113[(1)]	5.25 b	8.00 ab	7.25 ab	21.47 ab	94.21
226+113[(1)]	5.25 b	6.50 bc	7.12 ab	20.01 b	93.73
226+226[(1)]	5.12 bc	4.25 bcd	6.13 b	20.11 b	90.11
452+113[(1)]	4.50 bc	3.25 cd	2.37 c	18.95 b	84.40
452+226[(1)]	4.50 bc	2.50 d	2.00 c	18.06 b	72.37
452+452[(1)]	3.75 c	1.25 d	1.88 c	17.89 b	63.44
678	4.25 bc	3.75 cd	3.87 bc	19.60 b	91.23
904	4.13 bc	3.25 cd	2.25 c	18.89 b	87.12
Control	8.00 a	10.50 a	10.37 a	32.80 a	104.86
$F_{treatment}$	16.30**	13.69**	15.93**	3.23*	0.82^{ns}
LSD	1.47	3.80	3.62	12.21	64.96
VC (%)	12.5	31.2	31.7	24.6	31.4

Means followed by the same letter in the column are not significantly different from each other by Tukey's test ($p<0,05$). ^{ns} not significant; **Significant ($p<0,01$); * Significant ($p<0,05$). [(1)] Dose applied 14 days after the first spraying .

treatment without the application of the growth regulator, except with the dose of 113 + 113 ha^{-1} (Table 3). The average reduction of dry biomass in the aerial part, regardless of dose, was approximately 41%. A decreased number of cutting operations was observed in the plants that received a sequential application of trinexapac-ethyl at the doses of 425 + 425 $g\ ha^{-1}$ (Table 3), but similar to the other doses. This growth regulator promotes inhibition of gibberellin biosynthesis, as it diminishes the levels of active gibberellins, especially GA1, thus reducing cell elongation rather than cell division (Ervin and Koski, 2001; Mittlesteadt, 2009).

The periodic application of trinexapac-ethyl in *Poa pratensis* grass can reduce cuttings by up to 41% per year and thus improve the quality of lawns during the application period, as well as reducing the incidence of fungal diseases, such as *Sclerotinia homeocarpa* (Lickfeldt et al., 2001).

However, trinexapac-ethyl did not influence the accumulation of dry biomass in Japanese lawn grass roots (Table 3), as it was reported in the aerial part; probably due to the low activity of trinexapac-ethyl in these tissues up to 35 DASA (Richie et al., 2001).

It is observed in Table 4 that the application of trinexapac-ethyl reduced the average height of Bermuda grass (*Cynodon dactylon*), which reached values close to 50%, and all doses were effective and similar. These results corroborate those found by McCullough et al. (2007), Costa et al. (2009) and Maciel et al. (2011). Regarding the number of flower stems, reductions were observed from the dose of 226 + 226 $g\ ha^{-1}$, but were more drastic at the dose of 452 + 452 ha^{-1} or at higher doses. The lowest numbers of flower stems emitted were found in the treatments in which the application of 452 + 226 and 452 + 452 $g\ ha^{-1}$ and those in which a single dose of 678 and 904 $g\ ha^{-1}$ was applied, that is to say, a reduction of 52%, 70%, 69% and 73%, respectively (Table 4).

After three sequential applications with seven-day intervals between them, McCullough et al. (2007) observed a reduction in flower stems of approximately 80% in Bermuda grass (*Cynodon dactylon x C. transvaalensis*). In order for a growth regulator to be commercially acceptable, it needs to provide a 70% reduction of flower stem numbers (Johnson, 1994), as it was observed with some doses in this study.

All doses of the growth regulator significantly reduced the height of the floral stems. The shortest inflorescence height was observed in the treatment in which there were two applications of 452 $g\ ha^{-1}$ of trinexapac-ethyl, with a 92% reduction in comparison to the treatment without the application of this hebicide (Table 4).

It is observed that the accumulation of dry biomass in the roots and aerial part of Bermuda grass was also affected by the application of trinexapac-ethyl. The average reduction achieved by

Tabela 4 **-** Mean values of plant height and dry biomass of the aerial part and roots of the Bermuda grass, at 35 days after the second application of trinexapac-ethyl. Botucatu/SP 2013

trinexapac-ethyl ($g\ ha^{-1}$)	Height (cm)	Floral stems		Dry mass (g)	
		Number	Height (cm)	Aerial part	Root
113+113(1)	3.87 b	24.75 a	5.87 b	35.44 ab	44.17 ab
226+113(1)	3.62 b	21.25 ab	4.88 bc	26.39 abc	38.24 abc
226+226(1)	3.50 b	17.50 bc	4.87 bc	24.10 bc	37.48 abc
452+113(1)	3.37 b	15.25 bc	3.25 cd	23.28 bc	34.88 abc
452+226(1)	3.28 b	13.25 cd	2.25 de	23.47 bc	31.76 bcd
452+452(1)	3.23 b	8.25 d	0.88 e	20.76 c	21.06 d
678	3.19 b	8.50 d	2.25 de	24.16 bc	34.48 bc
904	3.10 b	7.25 d	1.63 de	22.04 bc	28.41 cd
Control	6.13 a	27.25 a	11.50 a	40.57 a	47.15 a
$F_{treatment}$	8.71**	28.54**	45.49**	5.02**	9.09**
LSD	1.67	6.56	2.28	14.27	12.43
VC (%)	19.8	17.3	23.0	22.5	14.8

Means followed by the same letter in the column are not significantly different from each other by Tukey's test ($p<0,05$). ns not significant; ** Significant ($p<0,01$). (1) Dose applied 14 days after the first spraying .

the application of growth regulator when statistical difference occurred was 45% in relation to the treatment with no application. The greatest reduction was observed in the treatments in which a dose of 452 $g\ ha^{-1}$ of trinexapac-ethyl or higher was applied. Fagerness and Yelverton (2000) also found that, when the application of trinexapac-ethyl was carried out in a sequential manner, reductions in the dry biomass of the aerial part were approximately 40%.

In relation to dry biomass of Bermuda grass roots, there was a reduction in accumulation with doses equal to or greater than 678 $g\ ha^{-1}$, divided or not. The highest reduction occurred when two doses of 425 $g\ ha^{-1}$ of the growth regulator were applied, with a 54% reduction (Table 4). These reductions were higher than those found in the literature (Fagerness and Yelverton, 2001; Beasley et al., 2005; McCullough et al., 2007; Maciel et al., 2011).

Concerning broadleaf carpetgrass, it was noticed that the application of growth regulator suppressed its development at all doses of trinexapac-ethyl tested; the grass height reduction was approximately 50% when applying the sequential doses of 452 $g\ ha^{-1}$ of the growth regulator (Table 5). It should be emphasized that there was no inflorescence emission during the experimental period.

Tabela 5 **-** Mean values of plant height and dry biomass of the aerial part and roots of the Broadleaf carpetgrass and Bahiagrass, at 35 days after the second application of trinexapac-ethyl. Botucatu/SP 2013

trinexapac-ethyl ($g\ ha^{-1}$)	Broadleaf carpetgrass			Bahiagrass		
	Height	Dry mass (g)		Height	Dry mass (g)	
	(cm)	Aerial part	Root	(cm)	Aerial part	Root
113+113(1)	5.75 b	23.91	81.22 ab	11.37	24.62	34.15
226+113(1)	5.50 bc	21.11	79.73 ab	9.88	24.10	30.98
226+226(1)	5.25 bc	21.31	76.62 ab	9.62	23.84	27.16
452+113(1)	4.86 bc	20.77	74.33 ab	9.75	21.46	27.44
452+226(1)	4.25 bc	17.41	67.33 b	8.00	21.07	26.54
452+452(1)	3.88 c	17.63	63.49 b	7.88	19.01	25.66
678	5.25 bc	19.83	73.70 ab	9.13	21.04	29.24
904	5.13 bc	17.35	61.27 b	7.75	19.19	26.87
Control	7.88 a	24.67	94.36 a	11.13	26.10	35.65
$F_{treatment}$	10.82**	0.90^{ns}	4.47**	1.15^{ns}	0.48^{ns}	2.70^{ns}
LSD	1.64	13.51	22.78	5.97	17.04	10.33
VC (%)	12.9	27.8	12.8	26.7	32.1	14.8

Means followed by the same letter in the column are not significantly different from each other by Tukey's test ($p<0.05$). ns not significant; **Significant ($p<0.01$). (1) Dose applied 14 days after the first spraying.

Trinexapac-ethyl did not influence the accumulation of dry biomass of the aerial part, but the application of the growth regulator reduced the accumulation of dry mass in the roots at the doses of 452 + 226, 452 + 452 and 904 g ha^{-1} (Table 5). According to Beasley et al. (2005), the application of the growth regulator in *Poa pratensis* grass resulted in height reduction, but there was an increase in the number of tillers, increasing dry biomass. However, in the present study, no effect was observed on the dry biomass of the aerial part.

Reductions in dry biomass of the roots can be directly correlated with a restriction in the development of the aerial part of grasses in general; in this research, the effect of trinexapac-ethyl on plant height was strongly noticeable (Table 5).

Moreover, Table 5 shows that there were no differences in the variables plant height, aerial part biomass and root biomass for bahiagrass. This shows that, under the experimental conditions described, the application of the growth regulator did not influence the development of this grass species. These data are not in line with those found by Freitas et al. (2002), who observed a significant decrease in bahiagrass height with a higher dose of trinexapac-ethyl.

In experimental conditions, the application of the trinexapac-ethyl growth regulator was effective in inhibiting the development of the Japanese lawn grass, Bermuda grass and broadleaf carpetgrass when applied in sequential doses higher than 226 g ha^{-1}, or in single doses of 678 and 904 g ha^{-1}, which could contribute to a good grass management of those species without affecting their aesthetics. In the case of St. Augustine grass and bahiagrass, the growth regulator did not provide reductions in their development under the experimental conditions used in the present study.

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