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Short Title: Hexazinone rainfall

Sporobolus indicus var. pyramidalis **Management in Response to Hexazinone Rates, Rainfall, and Application Timing in Florida Pasture Systems**

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Abstract

Rainfall is the main driving factor for soil-active herbicides, influencing their incorporation, leaching and absorption. Studies were conducted to determine the effects of simulated rainfall and hexazinone application rates on *Sporobolus indicus var. pyramidalis* control, and the impacts of application timing and rates on *Sporobolus indicus var. pyramidalis* in the field. Greenhouse experiments were established in Florida between 2017 and 2018, comprising hexazinone application rates of 0.56 and 1.12 kg ai ha^{-1}, and seven simulated rainfall accumulation volumes (0, 6, 12, 25, 50, 100 and 200 mm), distributed in a completely randomized design with four replicates and a non-treated control. Field experiments were conducted in a split-plot arrangement, where main plots were application timings at one-week intervals, subplots were two hexazinone application rates (0.56 and 1.12 kg ha^{-1}), and a non-treated control, distributed in a randomized complete block design with four replicates. In the greenhouse experiment, 49 and 92 mm were required to obtain 50% visual control and 35 and 82 mm to reduce biomass by 50% for hexazinone rates of 0.56 and 1.12 kg ai ha^{-1}, respectively. Field experiments showed that hexazinone peak efficacy was from mid-June to mid-August when applications were followed by 10 to 75 mm of rainfall during the first 7 DAT. The recommended rate of hexazinone at 1.12 kg ai ha^{-1} should be applied as it has an extended window of optimum application timing.

Keywords: hexazinone efficacy, simulated rainfall accumulation, density, and biomass reduction.

Management implications

Hexazinone at 1.12 kg ai ha^{-1} provided a greater application window, and increased *S. indicus var. pyramidalis* control and consistency compared to 0.56 kg ai ha^{-1} in the greenhouse and field experiments. The herbicide provided maximum efficacy from mid-June to mid-August when applications were followed by 10 to 75 mm of rainfall within the first 7 DAT. Therefore, as suggested by earlier studies, hexazinone should be applied at this rate during this application timeframe in Florida to maximize efficacy on *Sporobolus* species . Additionally, ranchers may consider applying hexazinone when the rainfall predicted for the next few days is favorable for *S. indicus var. pyramidalis* management. Although natural rainfall occurrences cannot be controlled, herbicide applications should be planned to maximize the likelihood of success. Furthermore, hexazinone use for *S. indicus var. pyramidalis* management should be part of an integrated management program to achieve maximum and sustainable control.

Introduction

Florida's sub-tropical climate, together with its approximately 1,300 mm of annual average rainfall, potentially allows high levels of forage production throughout the year, making forages an important source of nutrients in cow-calf operations (Mossler 2008). Forage systems in Florida are composed of warm-season perennial grasses including bahiagrass (*Paspalum notatum* Fluggé), bermudagrass (*Cynodon dactylon* (L.) Pers.), stargrass (*Cynodon nlemfuensis* Vanderyst), and limpograss (*Hemarthria altissima* (Poiret) Stapf & C.E. Hubbard), with bahiagrass being the primary forage utilized for year-round grazing (Chambliss and Sollenberger 1991). The popularity of bahiagrass among cow-calf producers is due to its tolerance to low soil pH, overgrazing, pesticide applications, as well as low fertilization requirements (Chambliss and Sollenberger 1991). Although bahiagrass will grow year-round in the southern portions of the Florida peninsula, shortened daylengths during the winter season (October through March) reduces bahiagrass growth significantly, which typically results in overgrazing. Although bahiagrass will persist when overgrazed, underfertilized, and without proper attention to soil pH, it is likely also more subject to increased weed infestations.

Weed interference can negatively impact forage production (de Marchi et al. 2022; Jakelaitis et al. 2010; Arnold and Santelmann 1970) and utilization (Herbin et al. 2021; Sather et al. 2013). Among all weeds present in Florida's grasslands, the weedy *Sporobolus* grasses small smutgrass [*Sporobolus indicus* (L.) R. Br.] and giant smutgrass [*Sporobolus indicus* (L.)

R. Br. *var. pyramidalis* (P. Beauv.) Veldkamp] have been persistent invaders in established bahiagrass (*Paspalum notatum* Fluggé) pastures (Rana et al. 2015). This is due to low grazing preference of weedy *Sporobolous* grasses by beef cattle, which results in overgrazing and loss of the desirable forage species over time (Wilder et al. 2008). *Sporobolus indicus var. pyramidalis* also invades other habitats such as natural areas, roadsides, and other disturbed open areas, influencing their biodiversity by competing with native plants for growth resources (Mooney and Cleland 2001). Additionally, *Sporobolus indicus var. pyramidalis* invasion in natural areas alters the soil, affecting infiltration, stabilization and carbon sequestration, devaluing these areas for native species (Rai and Sigh 2020). Furthermore, *Sporobolus indicus var. pyramidalis* can diminish the recreational value of many roadsides and natural areas, reducing their aesthetic worth. Hence, the management of weedy *Sporobolus* species is an important and necessary practice.

Sporobolus indicus var pyramidalis is a perennial warm-season grass with an average bunch size of 30–45 cm diameter . The grass weed has a seed head with panicle branches directed upwards and produced seeds from July to October, and sometimes spring, with more than 45,000 seeds per plant. It is dormant in the winter season, and the seeds remain viable in the soil for more than two years. *Sporobolus indicus var pyramidalis* grows in a wide range of environments, tolerates poor soil conditions, and rapidly germinates at 20 – 35 °C (Shay et al. 2022). The seeds are smaller which allows for easier distribution mainly by animals, wind, and water.

The efficacy of many different active ingredients has been investigated over the past 50 years to control weedy *Sporobolus* (Johnson 1975; Mislevy and Currey 1980; Nishimoto and Murdoch 1994; Rana et al. 2015; Smith et al. 1974). Hexazinone is currently the only selective option available for use in *P. notatum* and bermudagrass pastures that has shown to effectively control *Sporobolus* species (Ferrell et al. 2006; Mislevy et al. 2002). It is a herbicide with apoplastic mobility and limited translocation through the phloem when absorbed by leaves (Shaner 2014). Application of hexazinone is recommended during the summer at a rate of 1.12 kg ai ha^{-1} for weedy *Sporobolus* species management when rainfall is sufficient for incorporation and uptake from the soil solution (Ferrell et al. 2006; Mislevy et al. 2002). Although hexazinone is the only current selective option for managing *Sporobolus* species in

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Florida pastures, there are differences in forage tolerance due to environmental conditions before and after application as well as differences in species and/or cultivars (Sellers et al. 2008; Wilder et al. 2008).

Similar to differences among forage species and/or cultivars, there are discrepancies in the literature concerning application timing and rate efficacy on weedy *Sporobolus* species control. For example, field experiments conducted by Brecke (1981) showed that *S. indicus* control with hexazinone at 1.7 kg ha^{-1} applied during the spring did not differ from the same treatment applied during the fall as both provided effective control. Conversely, lack of control for the standard recommendation of hexazinone at 1.12 kg ai ha^{-1} applied during mid-summer has also been observed in Florida. For example, Ferrell and Mullahey (2006) reported excellent *S. indicus var. pyramidalis* control with this treatment in 1998, but lack of *S. indicus var. pyramidalis* control for the same treatment applied in the following year.

Several different factors could be associated with the variable hexazinone efficacy, but rainfall patterns after hexazinone application are likely to play a significant role. Hexazinone is mainly absorbed by *Sporobolus* roots, thus, it must be present at lethal concentrations within the root zone of target plants to be effective. However, hexazinone is prone to leaching due to its high solubility (33,000 mg L^{-1}) and low coefficient of adsorption (Koc = 54 mL g^{-1}) (Felding 1992). Therefore, the potential for hexazinone to leach below the root zone is high, especially during the summer when rainfall is abundant in Florida. Additionally, Florida soils are predominantly sandy from Lake Okeechobee northward (Brown et al. 1990). Sandy surface textures and low OM content increase the likelihood of hexazinone leaching below the root zone.

Pesticide transport into the soil has been well documented and extensively modeled for several decades (Aslam et al. 2015; Monquero et al. 2008; Beulke et al. 2002; Edwards et al. 1992). It has been clearly described that excessive rainfall can leach soil-applied herbicides out of the root zone, resulting in inadequate control. Conversely, insufficient rainfall after application can result in the absence of incorporation and subsequent lack of herbicide efficacy (Landau et al. 2021; Savage and Barrentine 1969; Weise and Hudspeth 1968). Furthermore, failure to adequately incorporate soil-applied herbicides exposes them to degradation through certain environmental factors (e.g. volatilization, photolysis, surface runoff), which can also decrease efficacy (Kanissery et al. 2019; Knake et al. 1967; Savage and Barrentine 1969).

Although the effects of rainfall on hexazinone leaching have been extensively studied using soil columns (Feng et al. 1988; Monquero et al. 2008; Tonieto and Regitano 2014) and in environmental fate experiments (Neary et al. 1983), there is limited information available regarding the impacts of rainfall on hexazinone incorporation and herbicidal efficacy for *S. indicus var. pyramidalis* control in Florida grasslands. Hence, the objectives of this study were to determine: (i) the effects of simulated rainfall after application on hexazinone efficacy, (ii) the effects of application timing and hexazinone rate on *S. indicus var. pyramidalis* control in the field, and (iii) the influence of natural rainfall on hexazinone efficacy in the field.

Material and Methods

Greenhouse Experiment

Greenhouse experiments were conducted at the University of Florida Institute of Food and Agricultural Sciences, Range Cattle Research and Education Center (RCREC), near Ona, FL (27°39’84” N, 81°94’07” W) in 2017 and 2018. Field soil was collected at the research center on 26 March 2017 and sieved through a 5-mm screen to remove unwanted debris. The soil was an Ona fine sand (sandy, siliceous, hyperthermic Typic Alaquods) with a mean soil pH of 4.9 and 3.0% organic matter. *Sporobolus indicus var. pyramidalis* clumps were collected from a pasture near the research center (27°38’04” N, 81°94’33” W) on 25 May 2017 and 30 May 2018. Individual culms with intact roots were separated from the clumps and planted in 3 L plastic pots (16-cm diameter top, 12-cm diameter base, and 16-cm depth), filled with field-collected soil amended with 14-14-14 (N: P2O5: K2O) slow-release fertilizer (Osmocote Smart-Release Plant Food, Scotts-Sierra Horticultural Products Company, Marysville, OH 43040). Plants were grown in a greenhouse maintained at 30/24 °C day/night temperatures under a 14-h photoperiod using a combination of natural and supplemental lighting during the experimental period. Clear vinyl 10-cm deep by 14-cm diameter saucers were placed underneath each pot and plants were sub-irrigated as needed.

Treatments included a 2 × 7 factorial arrangement of two hexazinone (Velpar L, 240 g ai L^{-1}, DuPont, Wilmington, DE) rates of 0.56 and 1.12 kg ai ha^{-1}, and seven simulated

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rainfall accumulation volumes (0, 6, 12, 25, 50, 100 and 200 mm) distributed in a completely randomized design with four replicates. At the time of the experiment, Velpar L was produced by DuPont and ownership has changed to Novasource (Tessenderlo Kerley, Inc. Pheonix, AZ). A non-treated control was added for treatment comparisons. Non-treated controls were not subjected to simulated rainfall and were only sub-irrigated. Each pot was considered as the experimental unit. Before herbicide application, all pots were sub-irrigated to soil saturation and were allowed to drain for 48-h. Hexazinone treatments were applied using a CO2 pressurized backpack sprayer, equipped with a 1.5-m boom calibrated to deliver 187 L ha^{-1}. Herbicides were applied on 26 July 2017 and 20 July 2018 and plants were approximately 40-cm tall at the time of application, approximately two months after planting. A Tlaloc 3000 rainfall simulator at an intensity of 65 mm h^{-1} and under a pressure of 27.6 KPa was used to simulate incorporation with rainfall 2 h after hexazinone application. This rainfall simulator covers a 2.8 x 2.3 m^2 area with a central nozzle at 3 m above the plant canopy. Polyethylene tarps were used as a windscreen on all sides of the simulator to prevent uneven rainfall across the experimental units. Pots were allowed to drain for 3 h after the rainfall simulations before returning to the greenhouse. All pots were placed in plant irrigation saucers and sub-irrigated with 60 ml of water as needed after the rainfall treatments until the end of the experiments.

Sporobolus indicus var. pyramidalis control was determined qualitatively by visual estimates of control ranging from 0 (no control) to 100% (complete death) 30 d after treatment (DAT), and quantitatively by clipping the aboveground biomass 30 DAT at 7.5 cm above the soil surface. Biomass samples were dried at 60 °C for 72 hours, and dry weights were recorded. Aboveground biomass was expressed as percent reduction compared with the non-treated control for statistical analyses.

Statistical analyses were performed using the statistical software R

3.4.3 (R Core Team 2014). Normality, independence of errors, and homogeneity of variance were visually examined for all response variables, and no data transformation was deemed necessary. All response variables were analyzed by fitting mixed-effects models using the package "*nlme*" in R (Pinheiro et al. 2016). The model statement for all response variables included hexazinone rate, rainfall accumulation volume, and their interactions as fixed

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effects, whereas the experimental run was considered a random effect. The effects of simulated rainfall were modeled using nonlinear regression models. The effective rainfall accumulation volume needed to increase visual estimates of control and reduce biomass responses by 50% (ER50) for each hexazinone rate was derived from a four-parameter log-logistic regression model using the ED function under the '*drc*' package in the R statistical environment (Knezevic et al. 2007) (Equation 1):

$$Y = c + \{d - c / 1 + \exp[b(\log x - \log e)]\} \tag{1}$$

Where Y is the response variable (visual estimates of control or aboveground biomass percent of reduction), x is simulated rainfall accumulation volume (mm), b is the relative slope at the inflection point, d is the upper limit of the curve, c is the lower limit of the curve, and e is the inflection point (ER50) of the fitted line. Model selection was based on Akaike's information criterion (AIC) in the '*qpcR*' package of R (Ritz and Spiess 2008). Additionally, a lack-of-fit test at the 95% level ($P \leq 0.05$) comparing the nonlinear regression models to ANOVA was conducted to test the appropriateness of model fit (Ritz and Streibig 2005). Differences among parameter estimates were compared using SE, and t and F tests at the 5% significance level (Knezevic et al. 2007).

Field Experiment

Field experiments were conducted in a pasture with ≥90% *S. indicus var. pyramidalis* ground cover on private property near the University of Florida Institute of Food and Agricultural Sciences, Range Cattle Research and Education Center (RCREC)(27°37'99" N, 81°94'54" W) in 2017. The experiment was repeated in an adjacent location within the same pasture in 2018. The predominant soil was a Smyrna fine sand (sandy, siliceous, hyperthermic, Aeric Alaquods) with 1.7% organic matter. Soil pH prior to the initiation of the study was 5.0 and 4.7 in 2017 and 2018, respectively. A rainfall data logger (RainWise RainLog TM 2.0, RainWise Inc. Boothwyn, PA) was installed in the research area and rainfall data were recorded hourly throughout the experimental period. In addition, 20-yr average rainfall data from the weather station located at the research center are presented in Table 1.

The experiment was a split-plot design with four replications. Main plot (12 m by 15 m; 180 m^2) treatments included twenty-two weekly herbicide application timings beginning in the first week of May and ending in the last week of September. Subplot treatments included

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two rates of hexazinone at 0.56 and 1.12 kg ha^{-1} in plots measuring 6 m by 15 m (90 m^2). A non-treated control at each application timing was added for treatment comparisons. Herbicide treatments were applied with a tractor-mounted, compressed air broadcast sprayer equipped with a 3 m boom calibrated to deliver 233 L ha^{-1}.

Sporobolus indicus var. pyramidalis control was determined qualitatively as previously described in the greenhouse experiment at 35 DAT. Additionally, *S. indicus var. pyramidalis* plant density (number of live plants m^2) was assessed at the beginning of the following growing season after treatment application (7 May 2018 and 14 March 2019) by placing two 1.0-m^2 quadrats at two random locations per experimental unit. Plants were only considered dead when completely lacking any green tissue. The two counts were averaged to represent the mean plant density per plot. Plant density data were expressed as percent reduction for statistical analysis by comparing the density recorded in treated plots at the end of the experiments with the average number of live plants present in the twenty-two non-treated control plots (one non-treated plot per application timing) at the end of the experiments.

Statistical analyses were performed using the open-source statistical software R 3.4.3 (R Core Team 2014). Normality, independence of errors, and homogeneity of variance were visually examined for all response variables, and data were transformed when ANOVA assumptions were violated. Arcsine square root transformation was used on *S. indicus var. pyramidalis* visual estimates of control 35 DAT (%). Nonetheless, non-transformed means are presented. Data were subjected to ANOVA to test for year, application timing, hexazinone rate and the effects of their interactions. Treatments were considered different when $P \leq 0.05$ and interactions not discussed were not significant. Means were separated using Fisher’s LSD test at a 5% level of significance when appropriate.

In addition, field rainfall data were analyzed in three steps by fitting mixed-effects models using the package “*nlme*” in R (Pinheiro et al. 2016). Seven rainfall classes were designated to characterize the total amount of rainfall recorded during the first 7 DAT: 0 (0 to 9 mm); 1 (> 9 ≤ 25 mm); 2 (> 25 ≤ 50 mm); 3 (> 50 ≤ 75 mm); 4 (> 75 ≤ 100 mm); 5 (>100 ≤ 125 mm); and 6 (> 125 mm). The seven-day period was chosen because i) previous research suggested that rainfall 1 to 6 days after application of cinmethylin, which is also a soil-

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applied herbicide, resulted in optimal grass weed control (Wittsell et al.1983), and ii) to decrease the chances of confounding effects due to herbicide dissipation processes. Secondly, rainfall class and rainfall class by hexazinone rate interaction were considered covariates and were included in the model as fixed effects. Year was considered a random effect in the covariance model to ensure sufficient data points within each rainfall class. Finally, means were separated using Fisher's Protected LSD test at 5% level of significance when appropriate.

Results and Discussion

Greenhouse Experiment

There was a hexazinone rate × rainfall accumulation effect for visual estimates of *S. indicus var. pyramidalis* control (P = 0.0001) and aboveground biomass reduction at 30 DAT (P = 0.0034). Therefore, the effects of increasing rainfall accumulation were presented separately, for each hexazinone rate (Table 2; Figures 1 and 2). In addition, a lack-of-fit test at the 95% level was not significant for all curves, indicating that the regression models were appropriate (Ritz and Streibig 2005).

Increasing simulated rainfall accumulation significantly impacted hexazinone efficacy at both rates for visual control and biomass reduction (Figures 1 and 2). In general, control with hexazinone at 0.56 kg ai ha^{-1} did not decline until rainfall accumulation exceeded 25 mm, whereas control at 1.12 kg ai ha^{-1} did not decline until rainfall accumulation exceeded 50 mm. In addition, the effective rainfall accumulation volume needed to reduce visual estimates of control by 50% (ER50) was 49 mm for hexazinone at 0.56 kg ai ha^{-1}, and 92 mm for hexazinone at 1.12 kg ai ha^{-1} (Table 2). Similarly, the ER50 value estimates for biomass reduction were at least 2 times greater for hexazinone at 1.12 versus hexazinone at 0.56 kg ha^{-1} (82 mm vs 35 mm) (Table 2).

These data indicate a significant difference in the rainfall accumulation necessary following hexazinone application to activate hexazinone at 0.56 and 1.12 kg ai ha^{-1} efficacy for *S. indicus var. pyramidalis* control. This is consistent with other published studies that assessed the effects of simulated rainfall on herbicide efficacy. Negrisoli et al. (2011) reported that control of signalgrass [*Urochloa decumbens* (Stapf) R.D.] with a pre-mix of clomazone and hexazinone at 0.88 and 0.22 kg ai ha^{-1}, respectively, was enhanced when followed by 30

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mm of simulated rainfall compared to the treatments without rainfall after application. Furthermore, Tonieto and Regitano (2014) reported decreased hexazinone efficacy at 0.50 kg ha^{-1} on little bell [*Ipomoea grandifolia* (Dammer) O'Donell] when followed by 120 mm of simulated rainfall within the first 7 d after herbicide application. This same study also determined that wheat straw mulch reduced hexazinone leaching 7-fold, indicating that soil organic matter plays a key role in hexazinone leaching (Koskinen et al. 1996; Stone et al 1993). Given the fact that the soils utilized in this study and those in the majority of pastures in Florida are sandy with low organic matter, it is likely that hexazinone leaching below the root zone is a primary reason for reduced efficacy under high rainfall accumulations. The use of sub-irrigation in this experiment following simulated rainfall may have resulted in less overall leaching from the entire system during the length of the experiment. While it is possible that sub-irrigation kept hexazinone concentrations in some of the treatments longer than would be with surface watering, surface watering could have resulted in additional leaching, which would have removed the impact of simulated rainfall on the day of application.

Field Experiment

There was a year × application and timing × hexazinone rate effect for *S. indicus var. pyramidalis* visual control 35 DAT data (P = 0.0009) and density reduction (P = 0.0001). Therefore, data are presented per year (Tables 3 and 4). Additionally, characteristic symptoms of photosystem II-inhibiting herbicides were observed 35 DAT for all application timings and in both years, ranged from slight necrosis and chlorosis on leaves to complete desiccation.

In 2017, hexazinone at 1.12 kg ha^{-1} resulted in more consistent and greater control than hexazinone at 0.56 kg ha^{-1} (Table 3), as it provided above 90% control at five application timings (06/23; 06/30; 07/14; 07/21 and 07/28). In contrast, hexazinone at 0.56 kg ha^{-1} provided above 90% control at only one application timing (07/21; 94% control). Similar responses were recorded for *S. indicus var. pyramidalis* density reduction as hexazinone at 0.56 kg ha^{-1} provided more than 60% *S. indicus var. pyramidalis* reduction at only four application timings (06/23; 06/30; 07/21 and 07/28); whereas hexazinone at 1.12 kg ha^{-1} provided above 80% *S. indicus var. pyramidalis* reduction at seven application timings (06/23; 06/30; 07/14 to 08/11).

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Additionally, hexazinone applications at 1.12 kg ha^{-1} broadened the window of optimum application timing. For example, visual estimates of control were at least 80% twelve times following 1.12 kg ha^{-1} vs. nine times following 0.56 kg ha^{-1} hexazinone. Similarly, density reductions exceeded 70% ten times following 1.12 kg ha^{-1} vs only one time following 0.56 kg ha^{-1}.

In 2018, both rates provided similar visual estimates of control at 5 application times (06/22; 06/29; 08/03; 09/14 and 09/21), with greater control for 1.12 kg ha^{-1} hexazinone at all the other application timings (Table 4). Similarly, both rates provided similar decreases in *S. indicus var. pyramidalis* density only at 7 application times in 2018 (06/22; 06/29; 08/03; 09/14 and 09/21), with greater density reductions for 1.12 kg ha^{-1} hexazinone at all the other application timings (Table 4). Previous research has also reported greater and consistent *S. indicus var. pyramidalis* control with hexazinone at 1.12 kg ha^{-1} compared to 0.56 kg ha^{-1} in Florida (Wilder et al. 2011). Additionally, hexazinone at 0.56 kg ai ha^{-1} provided the greatest level of control (71–96%) and density reduction (60–79%) when applied from 3rd wk of June through 3rd wk of July, as well as during the first 3 wks of September. Similar to observations made in 2017, increasing the hexazinone rate to 1.12 kg ai ha^{-1} increased the window of optimum application timing. For hexazinone at 1.12 kg ai ha^{-1}, the greatest *S. indicus var. pyramidalis* control (76–97%) and density reduction (59–86%) were observed from the 2nd wk of June until the 2nd wk of August as well as during the first 3 wks of September. Hexazinone at 1.12 kg ai ha^{-1} extended the window of optimum application timing by approximately 30 days compared to when applying at 0.56 kg ai ha^{-1}.

Previous research has also investigated the effects of hexazinone rate and application timing on weedy *Sporobolus* species control. Mislevy et al. (1999) observed that *S. indicus* control in Florida was more effective when hexazinone was applied during mid-summer or fall compared with spring applications. Additionally, the authors stated that control with hexazinone at 0.56 kg ai ha^{-1} was variable, ranging from 65 to 89% during spring and 86 to 91% during midsummer. Conversely, Rana et al. (2015) found that a single hexazinone application at 0.56 kg ai ha^{-1} during mid-summer did not decrease *S. indicus var. pyramidalis*

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density compared with the non-treated control. Research conducted by Wilder et al. (2011) found that *S. indicus var. pyramidalis* control was above 90% twelve months after application when hexazinone rates were equal or greater than 1.12 kg ai ha^{-1}, but below 70% for hexazinone applied at 0.56 kg ai ha^{-1}. In addition, the authors observed that the variability in the level of control tended to decrease at hexazinone rates greater than 0.56 kg ai ha^{-1} while it increased at rates lower than 0.56 kg ai ha^{-1}. Moreover, Ferrell et al. (2006) reported excellent *S. indicus var. pyramidalis* control 365 DAT with hexazinone 1.12 kg ha^{-1} applied in July. However, significantly lower control for the same treatment was recorded the following year. Therefore, based on results from the literature, the control of weedy *Sporobolus* species with hexazinone can be variable, with effective and poor control recorded with both hexazinone rates and at different application timings. Nonetheless, hexazinone rates equal to 0.56 kg ai ha^{-1}, applied during spring or late fall may result in poor control. Results from our study support this statement as applications performed in May, the first half of June, and the second half of September showed lower effectiveness compared to the other application timings in both years.

Several different factors likely played a role in the variable hexazinone efficacy responses observed across application timings in both years. However, we hypothesize that the rainfall pattern after application is likely one of the main factors due to the fact that hexazinone is primarily absorbed through the root system, and much less so through the foliage (Shaner 2014). Rana et al. (2015) suggested that the amount of rainfall after application plays a critical role in hexazinone efficacy on *S. indicus var. pyramidalis*, with excessive or scarce rainfall often resulting in lack of control. Our greenhouse results also support this suggestion as hexazinone effectiveness was acceptable when followed by 12 to 25 mm of simulated rainfall for hexazinone at 0.56 kg ai ha^{-1}, and 12 to 50 mm of simulated rainfall for hexazinone at 1.12 kg ai ha^{-1} (Figures 1 and 2). Similarly, Nagy (2008) reported that at least 14 mm of rainfall is required during the first two weeks after application for the optimal activation of acetochlor, another soil-applied herbicide widely used for pre-emergence weed control in corn production systems. In addition, Smith et al. (2016) observed lack of palmer amaranth (*Amaranthus palmeri* S. Wats.) control with dicamba, a synthetic auxin herbicide that also has high solubility (4,500 mg L^{-1}) and low adsorption (Koc = 13.4 mL g^{-1}), when subjected to increasing irrigation volumes in field experiments. The authors attributed the lack of efficacy to the dynamic mobility of dicamba in the soil, suggesting that high volumes of irrigation leached dicamba beyond the seed germination zone.

Although it is widely accepted that scarce or excessive rainfall after application will impact soil-applied herbicide efficacy, studies determining this optimum rainfall range for *S. indicus var. pyramidalis* control with hexazinone have never been investigated in Florida. We attempted to accomplish this by recording the rainfall pattern hourly for two years in the field trials and recording the effect of hexazinone rate × rainfall class for control (P = 0.0028) and density reduction (P = 0.0189) (Table 5). We were then able to analyze the results by the rainfall class. Overall, greater control at 35 DAT was observed for both hexazinone rates ranging from 57 to 69% at 0.56 kg ha^{-1}, and from 81 to 90% at 1.12 kg ha^{-1} in simulated rainfall classes 1 (10 to 25 mm) to 3 (51 to 75 mm) (Table 5). Also, *S. indicus var. pyramidalis* control was the lowest in rainfall classes 5 (101 to 125 mm) and 6 (>125 mm), followed by the rainfall class 0 (0 to 9 mm), for both application rates (Table 5). Furthermore, a similar rainfall class effect was detected for *S. indicus var. pyramidalis* density reduction in the hexazinone 0.56 kg ha^{-1} treatment, as greater responses were recorded in rainfall classes 1 to 3 (54, 52, and 55% density reduction, respectively). However, the effects of rainfall class were not as pronounced for *S. indicus var. pyramidalis* density reduction in plots treated with hexazinone 1.12 kg ha^{-1}, as similar responses were recorded for rainfall classes 0, 1, 2, 3, and 5 (Table 5). Nonetheless, peak hexazinone efficacy for *S. indicus var. pyramidalis* control in the field studies appeared to have occurred when 10 to 75 mm of rainfall accumulated within 7 days of application, regardless of the hexazinone rate (Table 5).

Although the rainfall class analysis in the field indicated that there is a relationship between rainfall accumulation and hexazinone efficacy, there were application timings within the suggested optimum rainfall range that resulted in low hexazinone effectiveness (e.g. 05/19 and 09/01 in 2017; and 06/01, 06/15, 07/27 and 08/03 in 2018) as well as application timings outside the suggested optimum rainfall range that resulted in good hexazinone efficacy (e.g. 07/07 and 08/11 in 2017; and 06/30 in 2018). This was likely because several other factors may also contribute to the success of *S. indicus var. pyramidalis* management

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with hexazinone, including the timing and intensity of rainfall events within the first 7 d following application. Hexazinone at 1.12 kg ai ha^{-1} applied on 08/11/2017 was followed by daily rainfall amounts of 31, 0, 2, 16, 34, 7, and 13 mm during the first week after application (totaling 104 mm 7 DAT) and provided 91% reduction in density. Conversely, hexazinone at 1.12 kg ai ha^{-1} applied one week later (08/18/2017) and followed by daily rainfall amounts of 0, 2, 0, 0, 14, 6, and 61 mm during the first week after application (totaling 83 mm 7 DAT) provided only 49% reduction in density. Thus, the timing and intensity of the first rainfall events might significantly contribute to rainfall-hexazinone dynamics for *S. indicus var. pyramidalis* management. Our greenhouse results may corroborate this statement as rainfall was simulated the day of hexazinone application. Further research should be conducted to determine if delaying simulated rainfall impacts control under controlled conditions.

The impact of rainfall on hexazinone efficacy in most Florida soils will likely follow the results from this study. Research has shown that hexazinone leaching is higher in sandy than in clay soils (Cristina dos Reis et al. 2017; Dousset et al. 2004). However, a separate study determined that there were little differences in hexazinone leaching in clay and sandy soils, and leaching was correlated with organic matter content of the soil (Koskinen et al. 1996). Given this information, the relatively low organic matter content and high porosity of Florida's sandy soils will likely lead to rapid leaching of hexazinone below the root zone of *Sporobolus* plants.

Overall, rainfall within the first 7 DAT with hexazinone is important regarding *S. indicus var. pyramidalis* management in Florida pastures. While we recognize that natural rainfall cannot be controlled, this information is important for users of this herbicide to understand when applications will likely fail. Since hexazinone is one of the most expensive herbicides, it is important to understand how rainfall patterns impact *S. indicus var. pyramidalis* control. For instance, it is common for rainfall to be limited in the early spring (March through May) in southern Florida, but spring-time rainfall is common in the Florida panhandle. Additionally, as rainfall forecasting improves with new technology, pasture managers will be able to make application decisions that should provide more consistent *S. indicus var. pyramidalis* control.

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Conflict of interest

The authors declare no conflict of interest.

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Table 1. Average monthly rainfall (mm) recorded at the research sites and temperature (°C) recorded from the Florida Automated Weather Network weather station located at the Range Cattle Research and Education Center, near Ona, FL in 2017 and 2018 compared to the 20-year average.

	Rainfall			Temperature								
				2017			2018			20-yr average		
Month	2017	2018	20-yr avg	Avg	Min	Max	Avg	Min	Max	Avg	Min	max
	mm			C°								
May	64	354	96	25	8	35	24	13	32	24	12	34
June	419	130	207	26	20	35	26	20	34	26	19	35
July	197	128	172	27	22	35	27	21	35	26	20	35
August	272	173	228	27	20	35	27	20	35	27	20	35
September	304	135	181	26	21	34	24	22	35	26	19	34
October	60	41	51	23	6	33	25	10	33	23	10	33
Total	1,316	965	935	-	-	-	-	-	-	-	-	-

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Table 2. Log-logistic regression[a] parameter estimates (±standard error) and rainfall needed to achieve 50% visual plant damage and 50% dry biomass reduction of *S. indicus var. pyramidalis* in30 days after treatment (DAT) with hexazinone in whole plant experiments under greenhouse conditions in Ona, FL, in 2017 and 2018[b].

Response variables	Hexazinone rate	Parameter estimates			
		b	*c*	*d*	ER_{50}
	kg ai ha^{-1}				mm rainfall
Visual control (%)	0.56	14.41 (±11.87)	24.79 (±2.77)	65.20 (±1.96)	49.13 b (±3.96)
	1.12	5.42 (±2.92)	31.65 (±4.61)	90.88 (±1.95)	91.85 a (±6.37)
Biomass reduction at 30 DAT (%)	0.56	20.89 (±2.41)	36.11 (±2.25)	60.30 (±2.15)	34.92 b (±2.48)
	1.12	4.09 (±1.78)	31.67 (±4.88)	69.67 (±2.02)	81.65 a (±12.02)

[a] Log-logistic model: Y = c + {d – c / 1 + exp[b(log x – log e)]}, where Y is the response, x is the simulated rainfall volumes, b is the slope of the inflection point, c is the lower limit of the curve, d is the upper limit of the curve, and e is the inflection point of the fitted line (equivalent to the simulated rainfall volume (mm) to cause 50% decrease in hexazinone efficacy [ER_{50}]).

[b] ER_{50} estimates followed by the same letter within each species are not different according to t and F tests at the 5% significance level.

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Table 3. Rainfall received 7 days after treatment (DAT), *Sporobolus indicus var. pyramidalis* visual estimates of control 35 DAT, and density reduction the following year with at hexazinone rates applied in 2017 at twenty-two different timings at the Range Cattle Research and Education Center, near Ona, FL.

App. timing	App. date	7 DAT rainfall	Visual control[b]				Density reduction[b]			
			Hexazinone rate (kg ai ha^{-1})							
			0.56		1.12		0.56		1.12	
Week		mm	%							
1	05/05	2	11	A gh	15	A j	10	A i	11	A j
2	05/12	1	27	B ef	61	A g-i	33	A d-h	31	A i
3	05/19	30	11	B gh	62	A g-i	20	A g-i	33	A i
4	05/26	3	07	B gh	52	A hi	20	B g-i	57	A f-h
5	06/02	116	17	B fg	49	A i	21	B f-i	55	A gh
6	06/09	164	06	B h	60	A g-i	26	B e-i	59	A e-h
7	06/16	155	11	B gh	68	A f-h	43	B de	67	A d-g
8	06/23	67	85	B b	94	A ab	66	A a	82	A a-d
9	06/30	18	86	B b	96	A ab	62	B a-c	87	A a-c
10	07/07	95	39	B de	89	A bd	45	B cd	76	A a-e
11	07/14	49	84	B bc	95	A a-c	39	B d-f	91	A ab
12	07/21	37	94	A a	97	A a	63	B a-c	93	A a
13	07/28	66	82	B bc	96	A ab	77	A a	92	A a
14	08/04	23	87	A ab	86	A cd	50	B b-d	92	A a
15	08/11	104	71	A c	84	A de	49	B b-d	91	A ab
16	08/18	83	51	B d	85	A d	35	A d-g	49	A hi
17	08/25	158	19	B fg	51	A i	13	A i	11	A j
18	09/01	72	29	B ef	81	A d-f	16	B hi	55	A gh
19	09/08	231	55	A d	69	A e-h	20	A g-i	36	A i
20	09/15	0	89	A ab	76	B d-g	36	B d-g	72	A c-g
21	09/22	9	81	A bc	80	A d-f	13	B i	66	A d-h
22	09/29	30	85	A bc	80	A d-f	16	B hi	73	A b-f
ANOVA										
Timing		0.0002							0.0001	
Rate		0.0002							0.0002	
Timing × Rate		0.0021							0.0026	

[a]Effect of year × timing × rate was significant ($P < 0.05$), so results were analyzed and presented by year.

[b]Means within response variables and within rows followed by the same uppercase letter; and means within response variable and within columns followed by the same lowercase letter, are not significantly different according to Fisher's protected LSD test at $P \leq 0.05$.

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Table 4. *Sporobolus indicus var. pyramidalis* visual estimates of control 35 DAT and density reduction the following year with two hexazinone rates applied at twenty-two different timings at the Range Cattle Research and Education Center, near Ona, FL in 2018[a].

App. timing	App. date	07 DAT rainfall	Visual control[b]				Density reduction[b]			
			Hexazinone rate (kg ai ha^{-1})							
			0.56		1.12		0.56		1.12	
Week		mm	%							
1	05/04	34	17	B hi	49	A h	30	A ij	20	A h
2	05/11	151	25	B h	67	A fg	25	A jk	30	A gh
3	05/18	69	12	B i	57	A gh	13	A k	18	A h
4	05/25	102	14	B hi	69	A fg	26	B jk	48	A ef
5	06/01	21	50	B g	79	A d-f	45	A f-h	58	A de
6	06/08	38	80	B c-e	97	A a	48	B e-g	65	A b-d
7	06/15	13	69	B ef	96	A a	38	B g-j	59	A de
8	06/22	63	71	A d-f	79	A d-f	60	A b-e	65	A b-d
9	06/29	81	92	A ab	96	A a	78	A a	83	A a
10	07/06	9	76	B c-f	94	A ab	63	B b-d	86	A a
11	07/13	38	86	B a-c	95	A a	68	A a-c	80	A a
12	07/20	2	76	B c-f	91	A a-c	63	B b-d	78	A ab
13	07/27	28	47	B g	81	A c-e	44	B f-i	60	A de
14	08/03	37	72	A d-f	76	A ef	53	A d-f	60	A de
15	08/10	56	76	B c-f	96	A a	69	A a-c	83	A a
16	08/17	16	75	B c-f	89	A a-d	74	A ab	81	A a
17	08/24	81	40	B g	86	A a-e	31	B h-j	53	A d-f
18	08/31	39	75	B c-f	91	A a-c	55	A c-f	61	A c-e
19	09/07	36	81	B b-d	95	A a	65	A a-d	75	A a-c
20	09/14	15	96	A a	97	A a	79	A a	80	A a
21	09/21	50	94	A a	89	A a-d	79	A a	75	A a-c
22	09/28	14	66	B f	82	A b-e	36	A h-j	41	A fg
ANOVA										
Timing		0.0012					0.0002			
Rate		0.0002					0.0006			
Timing × Rate		0.0033					0.0435			

[a]Effect of year × timing × rate was significant ($P < 0.05$), so results were analyzed and presented by year.

[b]Means within response variables and within rows followed by the same uppercase letter; and means within response variable and within columns followed by the same lowercase letter, are not significantly different according to Fisher's protected LSD test at $P \leq 0.05$.

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Table 5. Rainfall class x hexazinone rate for visual control 35 DAT and density reduction of *S. indicus var. pyramidalis* on the following year at the IFAS- RCREC, near Ona, FL. Data represent the means across application timings and years (2017 and 2018)[a].

	Visual control		Density reduction	
	Hexazinone rate (kg ai ha^{-1})			
Rainfall class	0.56	1.12	0.56	1.12
	%			
0 (0 to 9 mm)	54 bc	67 bc	26 d	56 abc
1 (10 to 25 mm)	69 a	87 a	54 a	67 ab
2 (26 to 50 mm)	57 abc	81 ab	52 ab	60 abc
3 (51 to 75 mm)	65 ab	90 a	55 a	74 a
4 (76 to 100 mm)	44 cd	75 bc	22 d	49 c
5 (101 to 125 mm)	37 cd	59 c	31 bc	65 abc
6 (>125 mm)	30 d	61 bc	31 bc	51 bc

[a]Means within response variable, hexazinone rate and columns followed by the same lowercase letter are not significantly different according to Fisher's protected LSD test at $P \leq 0.05$.

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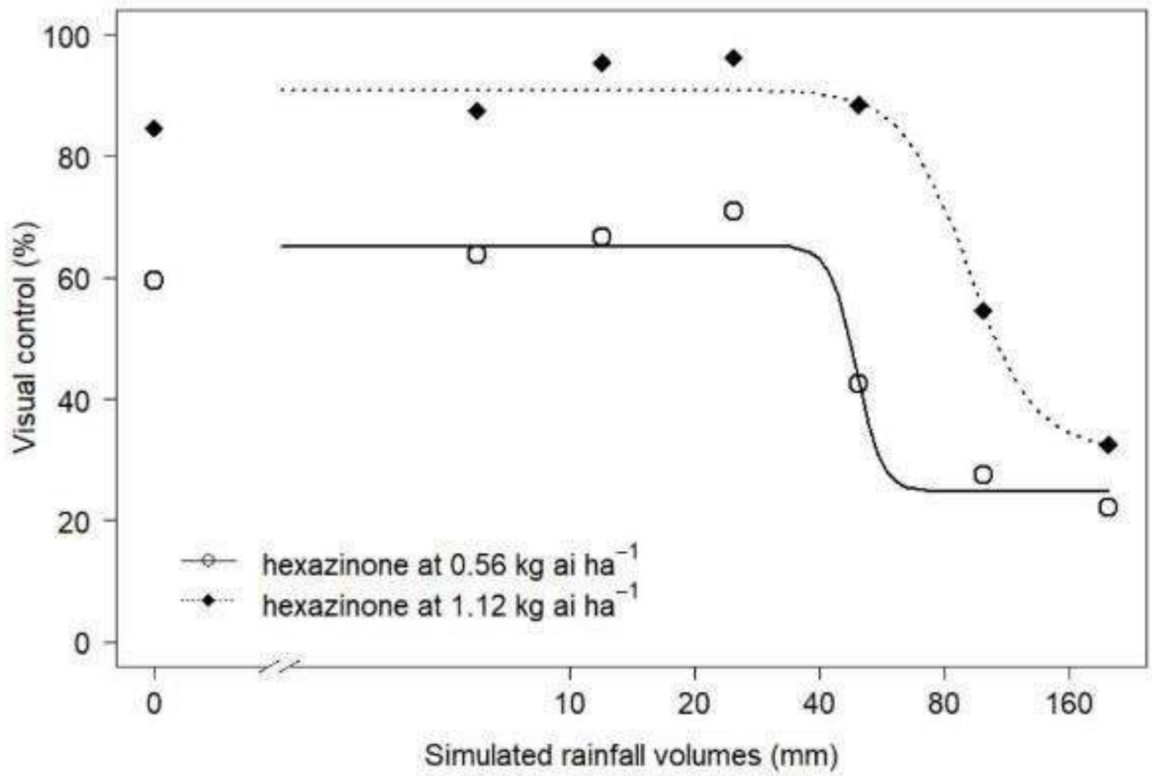

Figure 1. Visual estimates of control (%) (30 days after treatment) of *S. indicus var. pyramidalis* in response to two hexazinone rates and increasing volumes of simulated rainfall from whole plant studies conducted under greenhouse conditions in 2017 and 2018. Rainfall was simulated at 0, 6, 12, 25, 50, 100 and 200 mm. Solid and dashed lines represent predicted values. Data were fit to a four-parameter log-logistic regression model: Y = c + {d – c / 1 + exp[b(log x – log e)]}, where Y is the response, x is the simulated rainfall volumes, b is the slope of the inflection point, c is the lower limit of the curve, d is the upper limit of the curve, and e is the inflection point of the fitted line (equivalent to the simulated rainfall volume (mm) to cause 50% decrease in hexazinone activity [ER_{50}]).

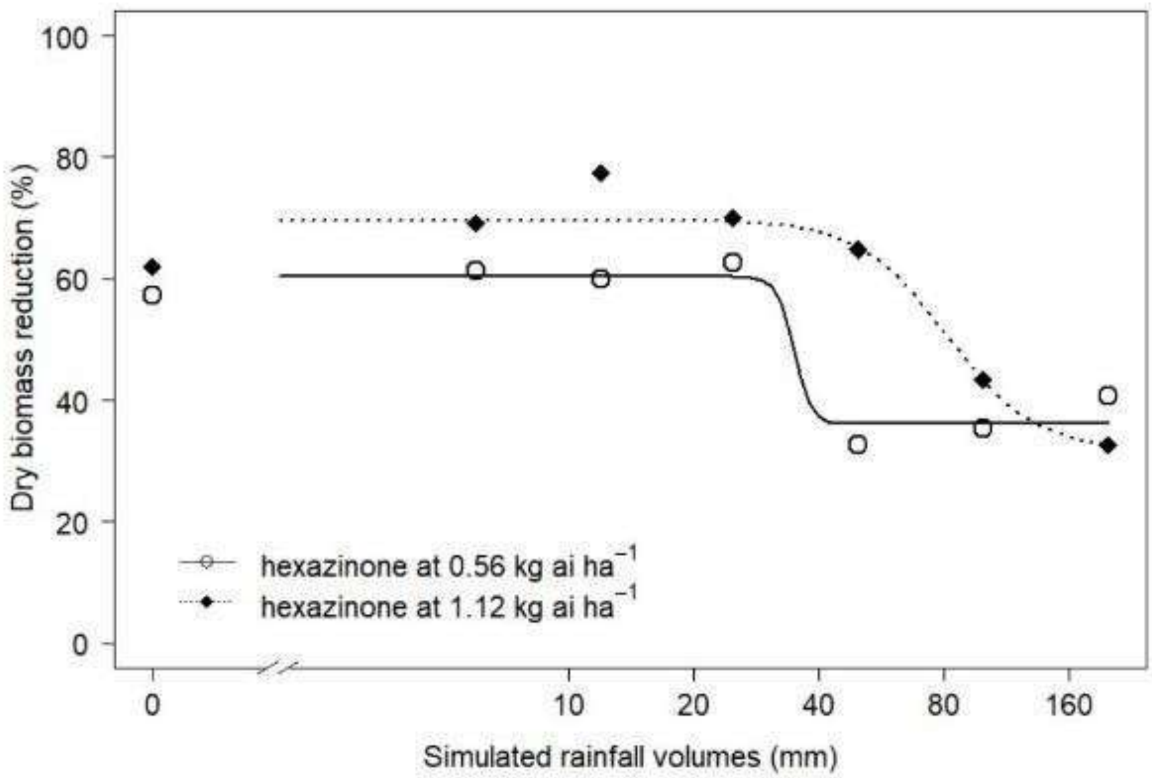

Figure 2. Dry aboveground biomass reduction (%) (30 d after treatment) of *S. indicus var. pyramidalis* in response to two hexazinone rates and increasing volumes of simulated rainfall from whole plant studies conducted under greenhouse conditions in 2017 and 2018. Rainfall was simulated at 0, 6, 12, 25, 50, 100, and 200 mm. Solid and dashed lines represent predicted values. Data were fit to a four-parameter log-logistic regression model: Y = c + {d – c / 1 + exp[b(log x – log e)]}, where Y is the response, x is the simulated rainfall volumes, b is the slope of the inflection point, c is the lower limit of the curve, d is the upper limit of the curve, and e is the inflection point of the fitted line (equivalent to the simulated rainfall volume (mm) to cause 50% decrease in hexazinone activity [ER_{50}]).

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