

**MORNINGGLORY INTERFERENCE WITH TRIPLOID WATERMELON
AND CUCURBIT RESPONSE TO FOMESAFEN**

by

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I dedicate this thesis to my family in Nicaragua, my visiting scholar and master's degree advisors, and my friends at Purdue. My mom, Auxiliadora Cordonero; dad, Jean Paul Arana; and sister, María Arana; supported me in all my decisions and encouraged me to keep fighting whenever I felt defeated. Dr. Jim Camberato and Dr. Bob Nielsen trusted me and provided enormous feedback to come to this step. My main advisor, Dr. Stephen L. Meyers, guided me patiently through this learning process and always believed in me. I appreciate everything Dr. Meyers taught and continues to teach me. Dr. Wenjing Guan and Dr. Bill Johnson helped me every step of the way with their extensive knowledge and amazing vibes. My friends at Purdue kept me sane and became my family in Indiana. I will always remember this part of my life with joy because of them.

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ABSTRACT

Indiana is among the top 10 cucurbit-producing states. Growers identified interference from troublesome weeds such as morningglory spp. (*Ipomoea* spp.) and a lack of registered herbicides as barriers to production. Thus, experiments were developed to determine (1) the interference of *Ipomoea* spp. with triploid watermelon, (2) the tolerance of pumpkin to fomesafen herbicide applied preemergence, and (3) the tolerance of plasticulture summer squash and watermelon to fomesafen herbicide applied pre-transplanting. 1) An additive design study was performed in 2020 at the Meigs Horticulture Research Farms (MEIGS) - Lafayette, IN and the Southwest Purdue Agricultural Center (SWPAC) - Vincennes, IN, to evaluate the interference of *Ipomoea* spp. with watermelon. The presence of *Ipomoea* spp. densities increasing from 3 to 24 per 27 m² increased watermelon yield loss from 58 to 99%, reduced watermelon fruit number 49 to 98%, reduced watermelon fruit weight 17 to 45%, and reduced watermelon aboveground biomass 83 to 94%. The most likely reason for watermelon yield loss was interference with photosynthesis and consequently less dry matter being partitioned into fruit development. Yield loss was attributed to fewer fruit and the reduced weight of each fruit. 2) Dose-response trials were performed in 2020 at SWPAC and in 2021 at SWPAC and the Pinney Purdue Agricultural Center (PPAC) - Wanatah, IN, to evaluate the tolerance of pumpkin to the herbicide fomesafen applied preemergence. Increasing the fomesafen rate from 280 to 1,120 g ha⁻¹ decreased emergence from 85 to 25% of the non-treated control at SWPAC in 2020, but only from 99 to 74% at both locations in 2021. The severe impact on emergence at SWPAC in 2020 was attributed to the herbicide moving into the crop's root zone due to excessive rainfall. Fomesafen is highly mobile under water-saturated soil conditions, especially in soils with low organic matter content, high pH, and a high proportion of sand content. Injury included brown and white spots and chlorosis due to the herbicide splashing from the soil surface onto the leaves and included stunting, but injury was transient. As the fomesafen rate increased from 280 to 1,120 g ha⁻¹, the predicted marketable orange pumpkin yield (kg per plot) decreased from 95 to 24% of the non-treated control at SWPAC in 2020 and 98 to 74% at PPAC in 2021. The predicted marketable orange pumpkin number decreased from 94 to 21% at SWPAC in 2020 and 98 to 74% at PPAC in 2021. Fomesafen rate did not affect marketable orange pumpkin yield and fruit number at SWPAC in 2021 and did not affect individual marketable orange pumpkin weight at any location-year. Overall, the fomesafen rate of 280 g ha⁻¹

is safe for use in the pumpkin cultivars 'Bayhorse Gold' and 'Carbonado Gold' within one day after planting, but there is a risk of increased crop injury with increasing rainfall. 3) Dose-response trials were performed in 2020 and 2021 at MEIGS, PPAC, and SWPAC to evaluate the tolerance of summer squash and watermelon to fomesafen applied pre-transplanting. Fomesafen rates increased from 262 to 1,048 g ai ha⁻¹ in 2020 at both locations, and from 280 to 1,120 g ai ha⁻¹ in 2021 at MEIGS did not affect summer squash yield. However, in 2021 at PPAC, rates from 280 to 1,120 g ha⁻¹ delayed harvest and decreased predicted marketable yield from 95 to 61% of the 0 g ha⁻¹ non-treated control. Fomesafen rates increased from 210 to 840 g ai ha⁻¹ did not affect marketable watermelon yield and fruit number. Crops' safety was attributed to rain washing off most of the herbicide from the plastic before transplanting or no excessive rain after transplanting. At PPAC in 2021, summer squash injury was attributed to excessive cumulative rain shortly after transplanting and no rain before transplanting. Overall, the 1x rates used for each trial are safe for use 1 d before planting summer squash and 6 to 7 d before transplanting watermelon. Rainfall before transplanting may be necessary to reduce the risk of the herbicide moving into the crop's root zone through the punched hole.

CHAPTER 1. INTERFERENCE OF MORNINGGLORIES (IPOMOEA SPP.) WITH 'FASCINATION' TRIPLOID WATERMELON

1.1 Abstract

Morningglories (*Ipomoea* spp.) are among the most troublesome weeds in cucurbits in the United States; however, we know little about *Ipomoea* spp. interference with horticulture crops. Two additive design field studies were performed in 2020 at two locations in Indiana to investigate the interference of *Ipomoea hederacea* and *I. lacunosa* with triploid watermelon. Watermelon density was kept constant, and *Ipomoea* spp. density was varied (0, 3, 6, 12, 18, and 24 per 27 m²). Immediately after transplanting watermelon, two-week-old *Ipomoea* spp. seedlings were transplanted into the same watermelon planting holes. Plots were harvested once a week for four weeks and each fruit was classified as marketable (≥ 4 kg) or non-marketable (<4 kg). One week after the final harvest, aboveground biomass samples were collected from 1 m² per plot and oven-dried to obtain watermelon and *Ipomoea* spp. dry weight. Seed capsules and the number of seeds in 15 capsules were counted from the biomass sample to estimate seed production. *Ipomoea* spp. densities increasing from 3 to 24 per 27 m² increased watermelon yield loss from 58 to 99%, reduced watermelon fruit number 49 to 98%, reduced watermelon fruit weight 17 to 45%, and reduced watermelon aboveground biomass 83 to 94%. *Ipomoea* spp. seed production ranged from 549 to 7,746 seeds m⁻², greatly increasing the weed seed bank. *Ipomoea* spp. hindered harvest due to their vines wrapping around watermelon fruits. The most likely reason for watermelon yield loss was interference with photosynthesis and consequently less dry matter being partitioned into fruit development. Yield loss was attributed to fewer fruit and the weight of each fruit.

Keywords

Additive design, morning-glory, morning glory, seedless watermelon, yield loss

1.2 Introduction

Watermelon [*Citrullus lanatus* (Thunb.) Matsum. & Nakai] production in the United States (US) averaged 1.7 billion kg between 2015 and 2019 (Kramer et al. 2020), placing it in the world's top 10 watermelon producing countries (FAO 2022). In Indiana, 2,469 ha were harvested in 2019, valued at \$35 million (USDA-NASS 2019). Watermelon is usually transplanted into raised-beds

covered with plastic polyethylene mulch, with a between-row distance of 1.8 to 3.7 m and in-row spacing of 90 to 180 cm (Egel 2020). Watermelon vines start covering between-row areas (row middles) at 3 wk after transplanting (WAP) and fully cover the row middles by 7 WAP (Andino and Motsenbocker 2004). Thus, watermelons are particularly vulnerable to weed competition because of the wide row spacing required for vine growth and their slow initial growth. The high temperatures necessary for watermelon production enable summer annual weeds to establish as well.

Adkins et al. (2010) reported that 'Super Crisp' triploid watermelon fields must be kept weed-free for 3.6 wk to limit yield losses to 10%. To avoid yield losses above 5%, Bertucci et al. (2019b) reported that 'Exclamation', 'E-Carnivor', and 'E-Kazako' triploid watermelon must be kept weed-free for 2.3, 1.9 and 2.6 WAP, respectively. Weed management strategies for watermelon production involve preemergence (PRE) and postemergence (POST) herbicide applications, raised beds with drip irrigation and plastic mulch, in-season cultivation, hand-hoeing, and hand-weeding. Ideally, watermelon fields should be kept weed-free throughout the growing season or at least during the critical weed-free period. Unfortunately, weed escapes do occur.

Collectively, the summer annual morningglories (*Ipomoea* spp. L.) were ranked as the fourth most troublesome weeds in the US and Canada in cucurbits (Van Wychen 2019). The most relevant *Ipomoea* spp. to weed science in Indiana are ivyleaf morningglory [*Ipomoea hederacea* (L.) Jacq.], entireleaf morningglory (*I. hederacea* var *integriuscula* A. Gray), pitted morningglory (*I. lacunosa* L.), and tall morningglory [*I. purpurea* (L.) Roth].

Ipomoea spp. compete for resources and climb and twine around crops, affecting harvest efficiency and yield. They find, climb and twine neighboring plants using several mechanisms, including phototropism, circumnutation, and shade-avoidance reactions. *Ipomoea* spp. grow towards other plants due to phototropism, most likely because other plants reflect solar radiation. Their vining habit and circumnutation allow them to twine around these plants (Price and Wilcut 2007). Vines are considered 'structural parasites' because they lean on other plants for support, which can cause structural damage (Paul and Yavitt 2011). Finally, because of shade-avoidance reactions, *Ipomoea* spp. grow over other plants. *Ipomoea* spp. are highly competitive organisms because they increase their biomass and thus their seed production the closer they are to other plants (Price and Wilcut 2007). As a result, affected plants grow under stress and often die. Thus,

the presence of *Ipomoea* spp. negatively affects yield and harvest and increases the weed seedbank in the soil, intensifying weed competition in subsequent years.

We know little about the interference of *Ipomoea* spp. with horticulture crops. However, *Ipomoea* spp. interference with soybean [*Glycine max* (L.) Merr.] and cotton (*Gossypium hirsutum* L.) have been studied. In soybean, full-season competition of a single *I. hederacea* plant 15 cm⁻¹ reduced yield 13 to 36% (Cordes and Bauman 1984), and one *I. purpurea* plant m⁻² reduced yield by 26% (Pagnoncelli et al. 2017). In cotton, one *I. hederacea* var *integriscula* plant 10 m⁻¹ reduced yield 3 to 7% (Wood et al. 1999), and one *I. hederacea* plant 2 m⁻¹ reduced yield 11% (Keeley et al. 1986). We hypothesized that the biology of *Ipomoea* spp. allows them to be competitive with susceptible crops such as watermelon and predicted that as *Ipomoea* spp. density increases, watermelon production would decrease. Thus, we established additive design studies to determine the influence of season-long *Ipomoea* spp. interference on plasticulture triploid watermelon.

1.3 Materials and Methods

Two additive design studies in which crop density was kept constant, and *Ipomoea* spp. density was varied were performed in 2020 at the Southwest Purdue Agricultural Center (SWPAC), Vincennes, IN, USA (38.73°N, 87.48°W) and the Meigs Horticulture Research Farm (MEIGS), Lafayette, IN, USA (40.28°N, 86.88°W). Soil types were Lomax loam (coarse-loamy, mixed, superactive, mesic Cumulic Hapludolls) with 1.5% organic matter (OM) and pH 6.6 at SWPAC, and Drummer silty clay loam (fine-silty, mixed, superactive, mesic Typic Endoquolls) with 4.5% OM and pH 6.5 at MEIGS. 'Fascination' triploid watermelon and 'Wingman' pollinizer watermelon seeds were planted on April 20, 2020, in a SWPAC greenhouse into 50-cell black seedling flats containing a peat-based potting media (Metro-Mix 360; SunGro Horticulture, Agawam, MA).

Fields were prepared with tillage prior to the formation of raised beds on April 21 at SWPAC and on April 28, 2020, at MEIGS. Each raised bed was covered with black polyethylene mulch and contained a drip tape placed in the middle, near the soil surface for irrigation. Plots were 27 m² and consisted of three rows, each 4.9 m long, with a between-row spacing of 1.8 m. Crop fertilization, irrigation, and diseases and insects management followed the recommendations of (Egel 2020). To manage weeds in row middles after bed formation and before transplanting, 1.1 kg ai ha⁻¹ S-metolachlor (Dual Magnum®, Syngenta Crop Protection, LLC, Greensboro, NC) at SWPAC, and a tank mix of 40 g ha⁻¹ halosulfuron-methyl (Sandea®, Canyon Group LLC C/O

Gowan Company, Yuma, AZ) and 1.4 kg ha⁻¹ ethalfluralin plus 421 g ha⁻¹ clomazone (Strategy®, Loveland Products, Inc. Greeley, CO) at MEIGS were broadcast applied. When needed, all other weeds not part of the experiment were removed from all plots, either by hand or with hoes or cultivators.

Two wk prior to the watermelon transplanting date at each location *Ipomoea* spp. seeds were planted into 72-cell trays containing a peat-based potting media (Berger BM2 Seed Germination Mix; Hummert International, Earth City, MO) at the Purdue University Horticulture Greenhouses, West Lafayette, IN. *Ipomoea* spp. seeds (Azlin Seed Service, Leland, MS) contained a mixture of predominantly *I. hederacea* and *I. hederacea* var *integriuscula*, but also included *I. lacunosa*. Crowley and Buchanan (1978) reported that the three species did not differ with respect to their effect on cotton yield. For this reason, no effort was made to select a single species for this research, and hereinafter, these three species are collectively referred as *Ipomoea* spp. At planting time, *Ipomoea* spp. plugs contained a single plant at a two true leaves stage and averaged 10 to 15 cm tall. Plugs were used to ensure that the intended *Ipomoea* spp. densities were achieved.

Transplanting occurred on May 21, 2020, at SWPAC and June 5, 2020, at MEIGS. Transplanting holes were punched in the plastic mulch with a manual hole punch at SWPAC and a water wheel transplanter at MEIGS. Four triploid watermelon seedlings were transplanted 1.2 m apart in each row resulting in 12 triploid watermelon plants per 27 m². After that, two pollinizer watermelon seedlings were planted per row, which resulted in a 1:2 pollinizer-to-triploid watermelon ratio per plot. Immediately after transplanting watermelons, *Ipomoea* spp. plugs were planted into triploid watermelon transplanting holes to achieve densities of 0, 3, 6, 12, 18, and 24 *Ipomoea* spp. per 27 m². Only one *Ipomoea* spp. plug per hole was planted for the densities of 3, 6, and 12 *Ipomoea* spp. per 27 m²; one or two plugs per hole were intercalated for the density of 18 *Ipomoea* spp. per 27 m² and two plugs per hole were transplanted for the density of 24 *Ipomoea* spp. per 27 m². We included the 0 *Ipomoea* spp. per 27 m² density as the non-treated control (Figure 1.1). The experiment design was a randomized complete block with four replications.

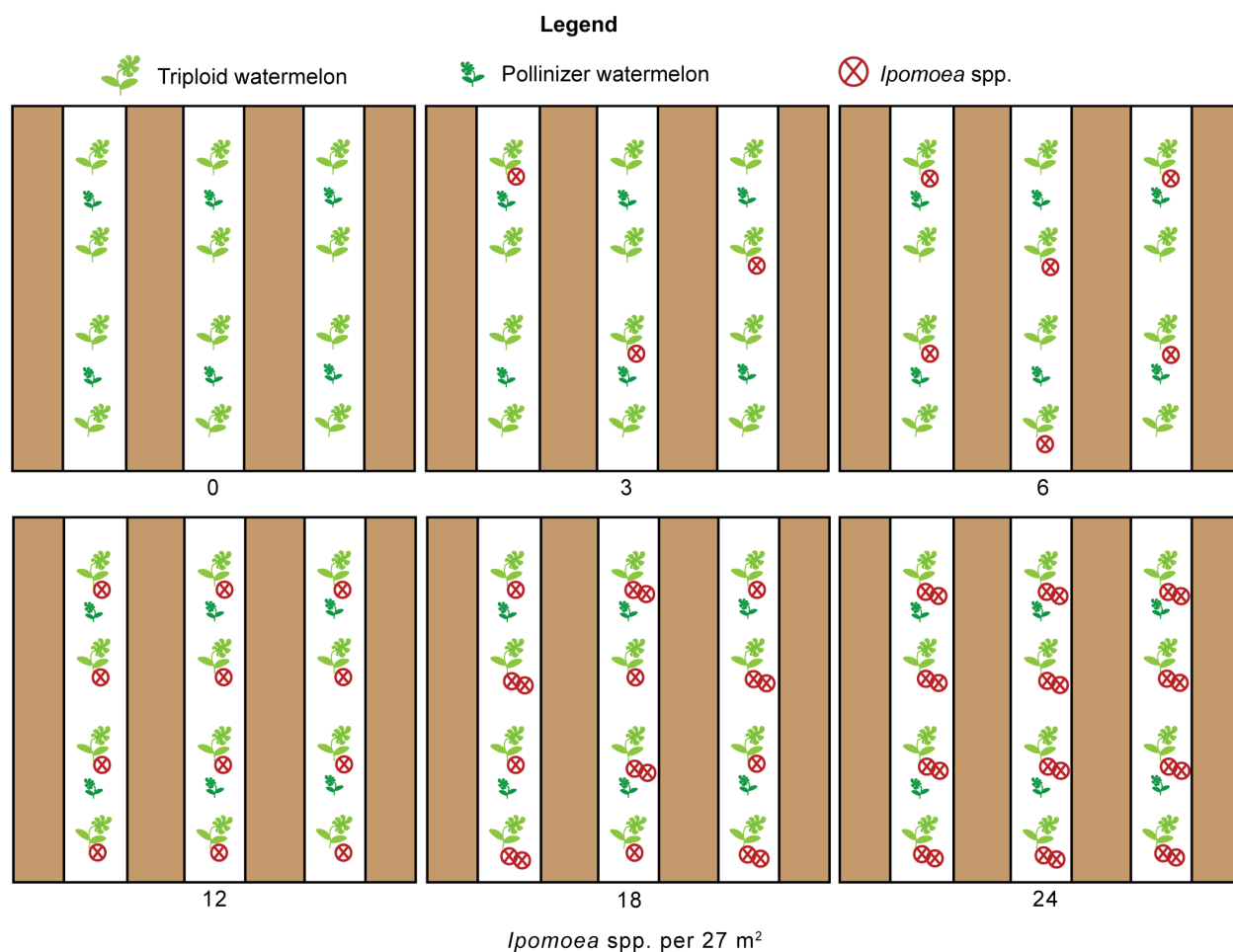


Figure 1.1. Additive design plot layout. Constant density of 12 'Fascination' triploid watermelon and six 'Wingman' pollinizer plants, and varied densities of 0, 3, 6, 12, 18, and 24 *Ipomoea* spp. per 27 m².

The length of the longest vine for all triploid watermelon and *Ipomoea* spp. plants in each plot were measured with a ruler from the soil surface in the planting hole to the vine growing point at 1, 2, and 4 WAP. After that, it was impractical to measure vine length because the watermelon and *Ipomoea* spp. were intertwined. The *Ipomoea* spp. percent canopy cover was visually estimated at 6 and 8 WAP. Watermelon fruits were harvested once wk⁻¹ for 4 wk, beginning July 22 at SWPAC and August 19 at MEIGS. Fruits were picked when the tendril that developed from the same node as the fruit peduncle was necrotic and the ground spot was yellow. The weight of each fruit was recorded and classified as marketable (≥ 4 kg) or non-marketable (<4 kg). Total marketable yield and fruit number were calculated as the sum of marketable watermelon yield pooled across all four harvests.

One wk after the last harvest, watermelon and *Ipomoea* spp. aboveground biomass was cut and collected using manual hedge shears from inside a 1 m² quadrat in the middle row of each plot. All biomass within a quadrat was placed inside a 114 L paper yard waste bag in the field to record total fresh weight. In the laboratory, the watermelon and *Ipomoea* spp. biomass were separated. Watermelon biomass was oven-dried at 60°C for 24 hours and the *Ipomoea* spp. biomass for 7 d to get aboveground dry biomass. After recording *Ipomoea* spp. dry weight, *Ipomoea* spp. seed capsules were separated and counted from the dried samples. Seed number from a subsample of 15 capsules was recorded and used to determine total seeds m⁻².

Marketable yield and fruit number, average individual fruit weight (including marketable and non-marketable fruits), and watermelon aboveground dry biomass were converted to a percent reduction of the non-treated control values using Equation 1.1:

$$\text{Percent reduction} = \frac{M - B}{M} \times 100 \quad [1.1]$$

where M is the mean value of the non-treated control treatments average for each location and B is the variable value of each data point for each location.

R software (RStudio ®, PBC, Boston, MA) was used to analyze our data. Data was evaluated as a linear model and subjected to an ANOVA to determine if statistically significant interactions ($P \leq 0.05$) existed between *Ipomoea* spp. density and location for each response variable. Response variables were watermelon vine length, *Ipomoea* spp. canopy cover percent at 6 and 8 WAP, marketable yield loss, marketable fruit number reduction, average individual fruit weight reduction and watermelon aboveground dry biomass reduction.

R code from Oliveira et al. (2018) was used to graph the results, using the *nls* (nonlinear least squares) function from the *nlstools* library to fit the rectangular hyperbola model (Cousens 1985) using Equation 1.2:

$$\text{Yield loss} = \frac{I * x}{1 + (\frac{I}{A}) * x} \quad [1.2]$$

where x represents *Ipomoea* spp. density in plants per 27 m², I represents yield loss per unit weed density as x approaches zero and A represent yield loss as x approaches infinity.

Data from the non-treated control were excluded from the seed production ANOVA due to zero variance. Seed production without the non-treated control data were then subjected Tukey's

Honest Significant Difference (HSD) test to separate mean seed production at a $P \leq 0.05$ significance level if density was statistically significant.

1.4 Results and Discussion

1.4.1 Watermelon Vine Length and *Ipomoea* spp. Canopy Cover

Watermelon vine length was not affected by the presence of *Ipomoea* spp. at 1, 2, or 4 WAP at either location (data not shown). However, by 6 and 8 WAP, percent *Ipomoea* spp. canopy cover was affected by *Ipomoea* spp. density. We combined *Ipomoea* spp. canopy cover data for both locations due to a non-significant treatment-by-location interaction. As the density of *Ipomoea* spp. increased from 3 to 24 per 27 m², predicted *Ipomoea* spp. percent canopy cover increased from 34 to 89% at 6 WAP and 52 to 100% at 8 WAP (Figures 1.2 and 1.3).

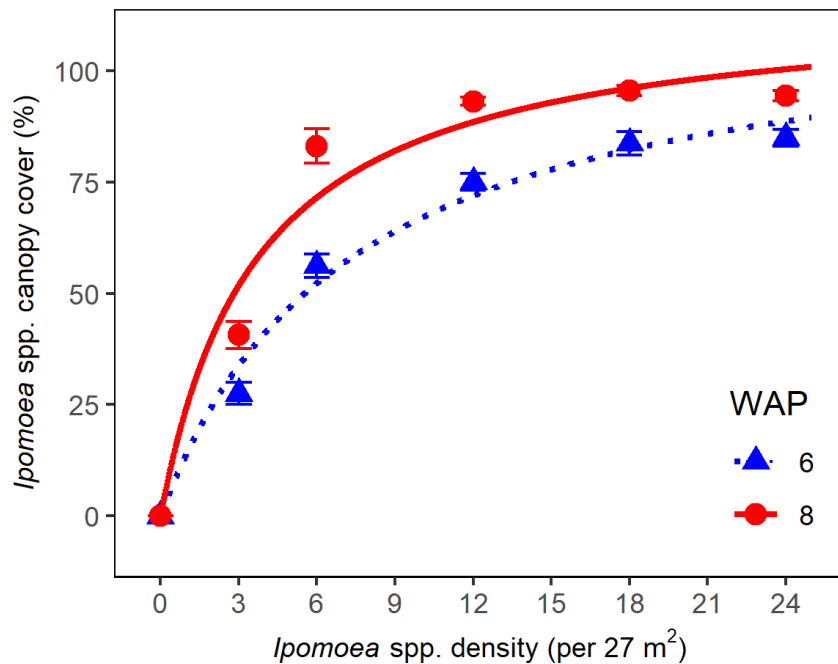


Figure 1.2. Relationship between *Ipomoea* spp. density and *Ipomoea* spp. canopy cover percent at six and eight wk after transplanting (WAP) described with a rectangular hyperbola. The model is $y = (I * x) \div (1 + (I * x/A))$; where $I=15.97$ and $A=115.33$ at 6 WAP, and $I=31.29$ and $A=115.86$ at 8 WAP. Data points represent the observed mean data with their standard error bars, and the solid and dashed lines represent the predicted values based on the model for each WAP. Data were pooled across two locations in 2020; the Southwest Purdue Agricultural Center and the Meigs Horticulture Farm.

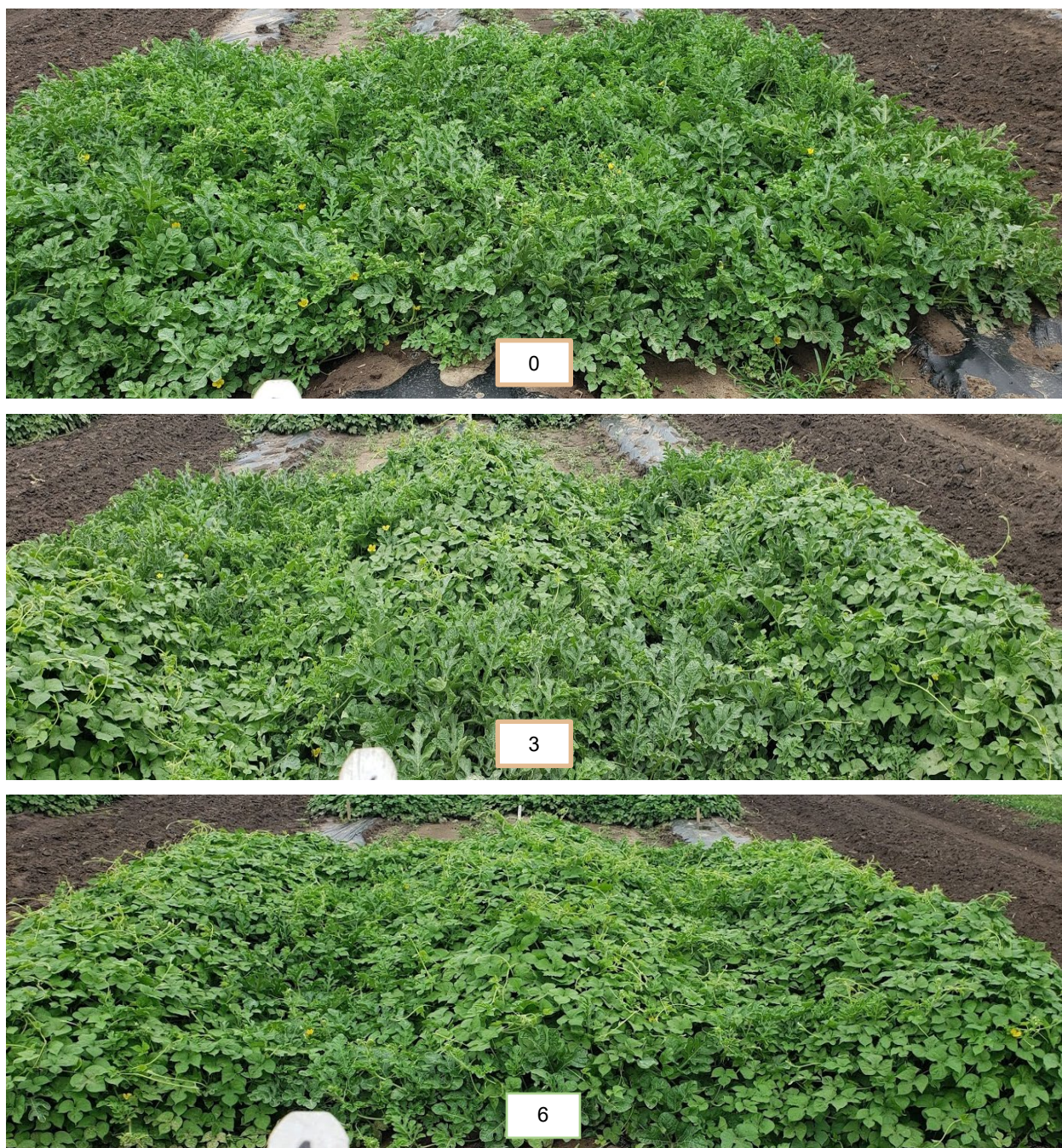
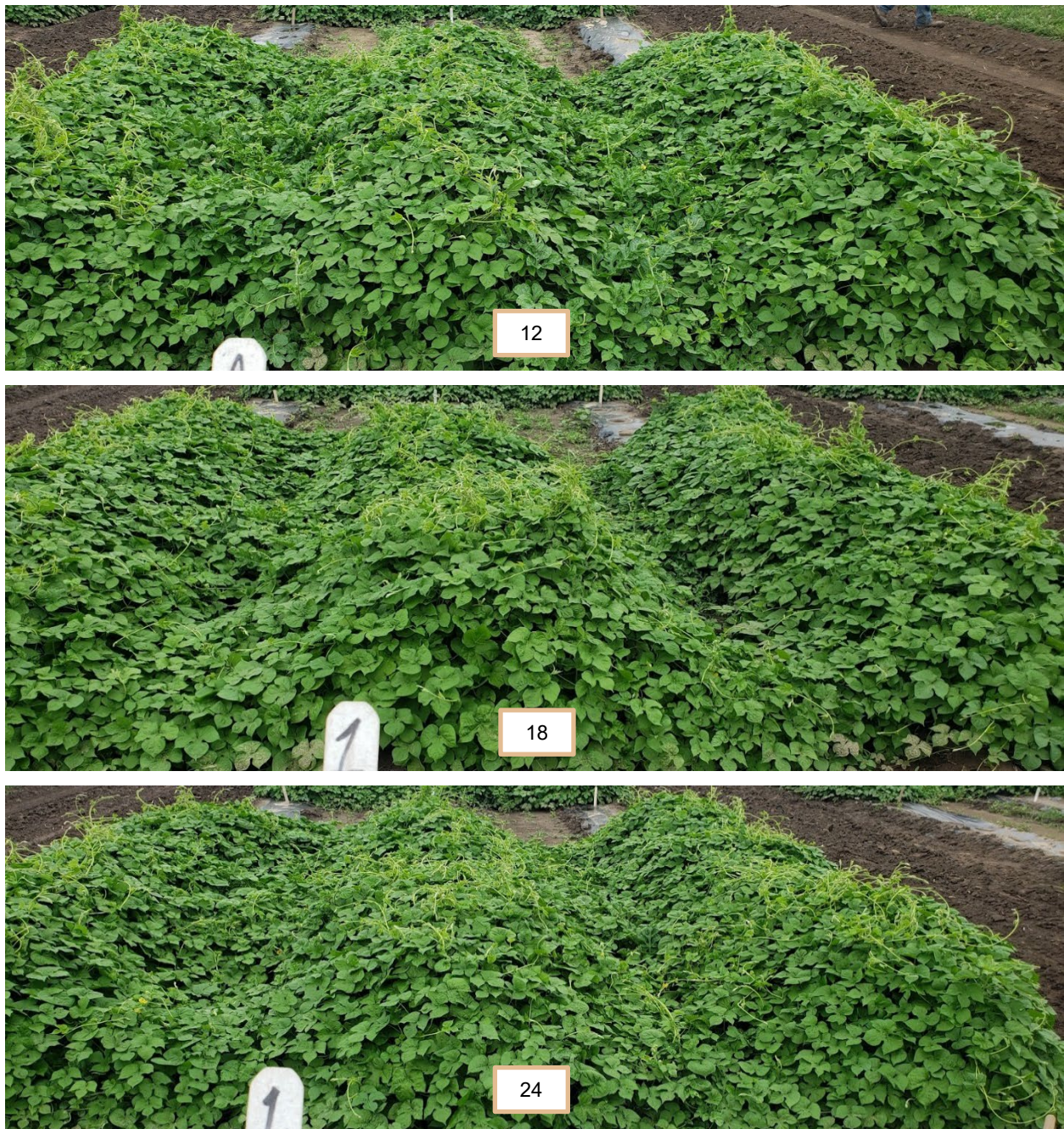


Figure 1.3. Plot canopy cover at 0, 3, 6, 12, 18 and 24 *Ipomoea* spp. per 27 m² 8 wk after transplanting at the Southwest Purdue Agricultural Center in 2020.

Figure 1.3. continued



1.4.2 Harvest Interference

The presence of *Ipomoea* spp. hindered the harvest process because watermelon fruits were hidden and wrapped around *Ipomoea* spp. vines (Figure 1.4). Although harvesting efficiency was not measured, we presume that *Ipomoea* spp. slowed the harvesting because multiple *Ipomoea* spp.

vines had to be removed or cut to harvest the fruits. The impact of *Ipomoea* spp. interference on harvesting efficiency is well-documented in other crops.



Figure 1.4. *Ipomoea* spp. vines wrapped around a watermelon fruit at harvest at the Meigs Horticulture Research Farm in 2020.

Schutte (2017) reported that the presence of *I. purpurea* slowed the manual harvesting of chile pepper (*Capsicum annuum* L). He also stated that professional chile pepper harvesters typically would avoid weedy areas, but if they did harvest weedy patches, it would extend harvesting time. Wood et al. (1999) judged it impossible to mechanically harvest cotton without damaging the equipment at some locations at *I. hederacea* densities of 10 and 12 weed 10 m⁻¹. Ellis et al. (1998) reported that the combine speed to harvest soybean was slowed slightly with *I. hederacea* densities of 0.25, 0.5, 1, and 2 plants row m⁻¹. Wilson and Cole (1966) reported that the

presence of *I. hederacea* and *I. purpurea* caused severe soybean lodging and decreased harvest availability. A trial to determine watermelon manual harvest efficiency in the presence of various weeds, including *Ipomoea* spp., could be beneficial to corroborate our results.

1.4.3 Marketable Watermelon Yield and Fruit Number

Marketable yield loss and fruit number reduction data were combined across locations due to a non-significant treatment-by-location interaction. Marketable yield and fruit number of the non-treated control were 187 kg and 26.3 fruit per 27 m². As *Ipomoea* spp. density increased from 3 to 24 per 27 m², predicted marketable watermelon yield loss increased from 58 to 99% (Figure 1.5) and predicted marketable fruit number reduction increased from 49 to 98% (Figure 1.6). These models followed almost an identical path as the model for the canopy cover percent at 8 WAP (Figure 1.2), suggesting that *Ipomoea* spp. canopy cover at 8 WAP was an indicator of yield loss.

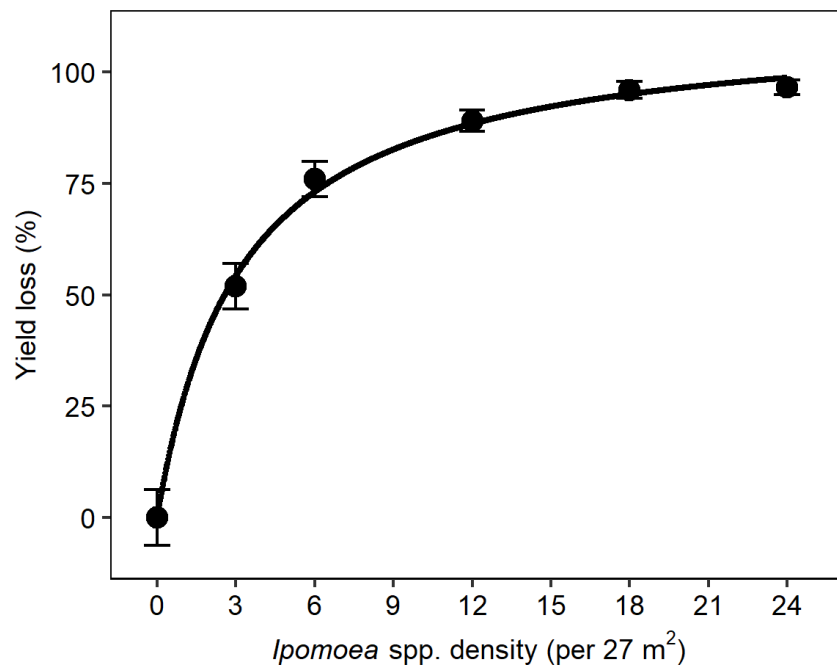


Figure 1.5. Relationship between *Ipomoea* spp. density and watermelon marketable yield loss described with a rectangular hyperbola. The model is $y = (I * x) \div (1 + (I * x/A))$ where $I = 40.80$ and $A = 109.89$. Data points represent the observed mean data with their standard error bars, and the solid line represents the predicted values based on the model. Fruit was classified as marketable if ≥ 4 kg. Data were pooled across two locations in 2020; the Southwest Purdue Agricultural Center and the Meigs Horticulture Farm.

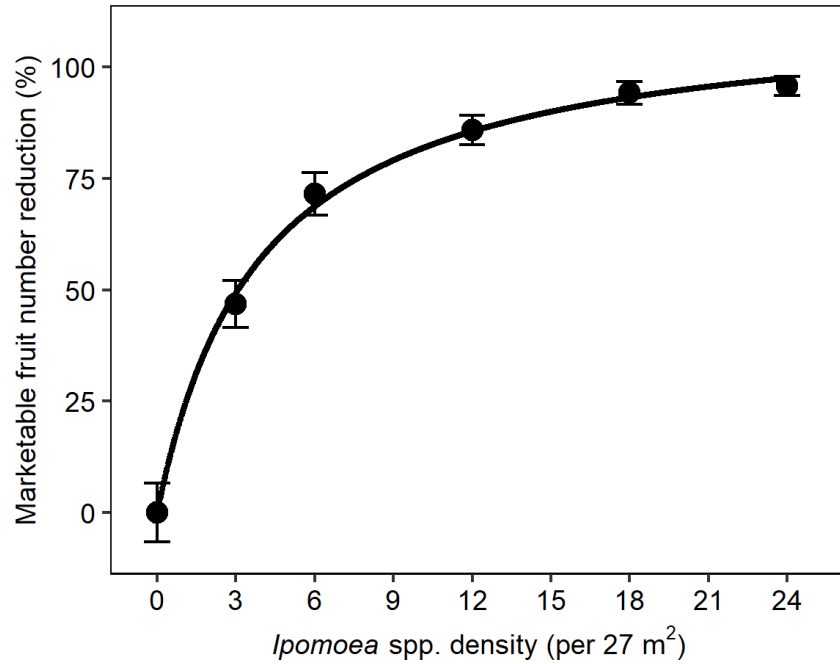


Figure 1.6. Relationship between *Ipomoea* spp. density and watermelon marketable fruit number reduction described with a rectangular hyperbola. The model is $y = (I * x) \div (1 + (I * x/A))$ where $I = 29.16$ and $A = 113.29$. Data points represent the observed mean data with their standard error bars, and the solid line represents the predicted values based on the model. Fruit was classified as marketable if ≥ 4 kg. Data were pooled across two locations in 2020; the Southwest Purdue Agricultural Center and the Meigs Horticulture Farm.

1.4.4 Average Individual Fruit Weight

Individual fruit weight reduction data were combined across locations due to a non-significant treatment-by-location interaction. The mean fruit weight of the non-treated was 7.4 kg per fruit. As *Ipomoea* spp. density increased from 3 to 24 per 27 m², predicted individual fruit weight reduction increased from 17 to 45% (Figure 1.7).

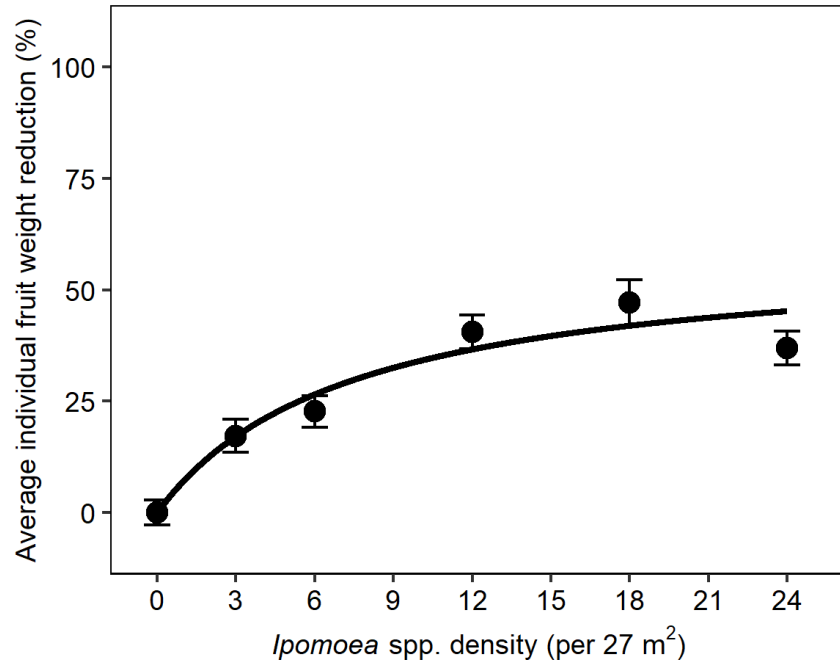


Figure 1.7. Relationship between *Ipomoea* spp. density and watermelon average individual fruit weight reduction (marketable and non-marketable fruits) described with a rectangular hyperbola. The model is $y = (I * x) \div (1 + (I * x/A))$ where $I = 8.05$ and $A = 58.97$. Data points represent the observed mean data with their standard error bars, and the solid line represents the predicted values based on the model. Fruit was classified as marketable if ≥ 4 kg and non-marketable if < 4 kg. Data were pooled across two locations in 2020; the Southwest Purdue Agricultural Center and the Meigs Horticulture Farm.

This study confirmed that watermelons are poor weed competitors, and our results are consistent with the severe watermelon yield loss caused by other weeds. Season-long American black nightshade (*Solanum americanum* Mill), at a density of 2 plants m⁻², reduced watermelon yield around 54 to 58% (Adkins et al. 2010; Gilbert et al. 2008). A 10% watermelon yield loss was observed with only two yellow nutsedges (*Cyperus esculentus* L.) m⁻² permitted to grow season-long (Buker III et al. 2003). Season-long interference of six smooth amaranths (*Amaranthus hybridus* L.) m⁻¹ reduced watermelon yield by around 60% (Terry et al. 1997), and one Palmer amaranth (*Amaranthus palmeri* S. Watson) per planting hole reduced the yield of three watermelon varieties from 45 to 75% (Bertucci et al. 2019a).

Adkins et al. (2010), Buker III et al. (2003), and Gilbert et al. (2008) reported yield loss due to fruit number reduction but not smaller fruit size. In the current study, we found that fruit weight reduction impacted yield loss. However, at low *Ipomoea* spp. densities, yield reduction was mainly due to reduced fruit number rather than fruit size. The physical pressure that *Ipomoea* spp.

vines put on the watermelon fruits likely contributed to the size reduction. While a physical force is not likely to occur on *C. esculentus* and *S. americanum* because these grow vertically and are vineless. Although, if vertically growing weeds are present at high densities, they may also reduce watermelon fruit size and individual watermelon fruit development. For instance, *Amaranthus* spp. allowed to grow season-long reduced individual watermelon fruit mass by 37% at a density of six *A. hybridus* m⁻¹ (Terry et al. 1997) and by 9% at four *A. palmeri* per planting hole (Bertucci et al. 2019a).

1.4.5 Watermelon Aboveground Dry Biomass

Watermelon aboveground dry biomass reduction data were combined across locations due to a non-significant treatment-by-location interaction. Mean watermelon aboveground biomass dry weight of the non-treated was 292 g m⁻². Predicted watermelon biomass reduction increased from 83 to 94% as the *Ipomoea* spp. density increased from 3 to 24 per 27 m² (Figure 1.8).

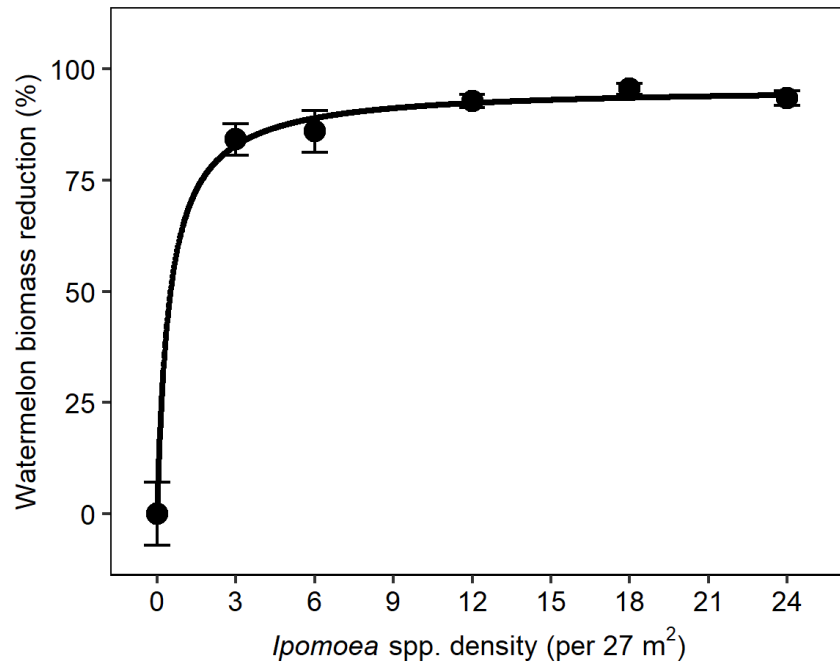


Figure 1.8. Relationship between *Ipomoea* spp. density and watermelon biomass reduction in 1 m² described with a rectangular hyperbola. The model is $y = (I * x) \div (1 + (I * x/A))$ where $I = 202.96$ and $A = 95.94$. Data points represent the observed mean data with their standard error bars and the solid line represent the predicted values based on the model. Watermelon biomass data were collected from 1 m² per plot and oven-dried at 60°C to obtain dry weight. Data were pooled across two locations in 2020; the Southwest Purdue Agricultural Center and the Meigs Horticulture Farm.

Watermelon biomass reduction has not been reported in the studies mentioned previously, but we saw that *Ipomoea* spp. significantly affect watermelon aboveground dry biomass. Dry matter partitioning into the harvestable organs contributes to the crop's yield, and leaves are the primary source of dry matter. In vegetable crops in which harvest is performed over an extended period, a balance between dry matter partitioning into the fruits and other vegetative organs is essential (Marcelis et al. 1998). Because *Ipomoea* spp. outgrew and covered the watermelon plants, watermelon plants could not photosynthesize to create dry matter. Consequently, we hypothesize that biomass reduction most likely is the primary reason for yield loss and the reduction in fruit number and fruit weight reduction in the presence of season-long *Ipomoea* spp.

1.4.6 *Ipomoea* spp. Seed production

Ipomoea spp. seed production data were analyzed by location. Pooled across all *Ipomoea* spp densities, seed production was greater at MEIGS (6,703 seeds m⁻²) than at SWPAC (1,007 seeds m⁻²) (data not shown). The effect of *Ipomoea* spp. density was significant at SWPAC but not at MEIGS. *Ipomoea* spp. seed production ranged from 549 to 1,956 seeds m⁻² at SWPAC and from 5,016 to 7,746 seeds m⁻² at MEIGS (Table 1). Unexpectedly, at SWPAC, the lowest density of three *Ipomoea* spp. per 27 m² had a significantly higher seed production (1,956 seeds m⁻²) than the two highest densities of 18 and 24 *Ipomoea* spp. per 27 m² (549 and 555 seeds m⁻², respectively).

Table 1.1. *Ipomoea* spp. seed production with standard error (SE) at the Southwest Purdue Agricultural Center (SWPAC) and the Meigs Horticulture Farm (MEIGS) in 2020

Density	<i>Ipomoea</i> spp. seed production ^a	
	SWPAC	MEIGS
<i>Ipomoea</i> spp. per 27 m ²	seeds m ⁻²	
3	1956 (205) a ^b	6659 (2617)
6	1432 (404) ab	7213 (2067)
12	1045 (303) ab	5016 (1081)
18	549 (62) b	6880 (748)
24	555 (200) b	7746 (1302)

^aSeed production per 1 m² was obtained by counting the total seed capsules in 1 m², counting the number of seeds in 15 capsules, and then extrapolating the total number of seeds in that 1 m².

^bMeans separation applying Tukey's HSD at a $\alpha = 0.05$ significance level. Means that do not share a common letter are significantly different.

Seed production at MEIGS is comparable to the values reported by Crowley and Buchanan (1982) and Gomes et al. (1978): 5,000 and 14,600 seeds per one *I. hederacea* var *integriscula*, 10,000 and 15,200 seed per one *I. lacunosa*, and 6,000 and 5,800 seed per one *I. hederacea*, respectively. However, we hypothesize that seed production may have been affected by intraspecific competition at both locations, but SWPAC was more affected than MEIGS. Colom and Baucom (2020) reported that intraspecific competition of two *I. hederacea* reduced seed production by around 35% with respect to a single *I. hederacea*. Another possible reason for reduced seed production at SWPAC compared to MEIGS would be the effect of environmental factors not measured.

1.5 Conclusions

Overall, this study demonstrated that watermelon production is significantly affected by the presence of *Ipomoea* spp. that are permitted to grow season-long. If *Ipomoea* spp. escape the initial weed control practices, and grow all season, they will hinder harvest and reduce yield, fruit number and size, and biomass because of its propensity for climbing, vining, and twining. *Ipomoea* spp. densities increasing from 3 to 24 per 27 m² increased watermelon yield loss from 58 to 99%,

watermelon fruit number reduction from 49 to 98%, watermelon fruit weight reduction from 17 to 45%, and watermelon aboveground biomass reduction from 83 to 94%.

Despite no *Ipomoea* spp. density affecting watermelon vine length at early stages, by 6 and 8 WAP, *Ipomoea* spp. outgrew the watermelon, and the canopy cover of *Ipomoea* spp. was prominent as the *Ipomoea* spp. density increased. Because of this, we presumed that the most likely reason for watermelon yield loss was due to interference with photosynthesis and consequently less dry matter being partitioned into fruit development. Moreover, fruit weight was possibly also affected by the physical force imposed by the *Ipomoea* spp. vines, so yield loss was attributed to fewer fruits and the weight of each fruit. We also demonstrated that *Ipomoea* spp. seed production increases the weed seedbank in the soil immensely, reinforcing the importance of post-harvest weed control.

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CHAPTER 2. DOSE-RESPONSE OF TWO JACK O'LANTERN PUMPKIN CULTIVARS TO FOMESAFEN APPLIED PREEMERGENCE

2.1 Abstract

Three dose-response trials were performed between 2020 and 2021 at two Indiana locations: the Southwest Purdue Agricultural Center (SWPAC) and the Pinney Purdue Agricultural Center (PPAC), to determine the tolerance of two Jack O'Lantern pumpkin cultivars to fomesafen applied preemergence. The experiment was a split-plot arrangement in which the main plot was the fomesafen rate (0, 280, 560, 840, and 1,220 g ai ha⁻¹), and the subplot was the pumpkin cultivar ('Bayhorse Gold' and 'Carbonado Gold'). As the fomesafen rate increased from 280 to 1,120 g ha⁻¹, the predicted pumpkin emergence decreased from 85 to 25% of the non-treated control at SWPAC-2020, but only from 99 to 74% at both locations in 2021. The severe impact on emergence at SPWAC-2020 was attributed to rainfall. Visible injury included brown and white spots and chlorosis due to the herbicide splashing from the soil surface onto the leaves and included stunting, but injury was transient. As the fomesafen rate increased from 280 to 1,120 g ha⁻¹, the predicted marketable orange pumpkin yield decreased from 95 to 24% of the non-treated control at SWPAC-2020 and 98 to 74% at PPAC-2021. Similarly, the predicted marketable orange pumpkin fruit number decreased from 94 to 21% at SWPAC-2020 and 98 to 74% at PPAC-2021. Fomesafen rate did not affect marketable orange pumpkin yield and fruit number at SWPAC-2021 and marketable orange pumpkin fruit weight at any location year. Overall, the fomesafen rate of 280 g ha⁻¹ was safe for use preemergence in the pumpkin cultivars 'Bayhorse Gold' and 'Carbonado Gold' within one day after planting, but there is a risk of increased crop injury with increasing rainfall.

Nomenclature

Fomesafen; pumpkin 'Bayhorse Gold' and 'Carbonado Gold', *Cucurbita pepo* L.

Keywords

Dose-response; emergence; injury; PPO; yield

2.2 Introduction

The United States (US) is ranked fifth among pumpkin, squash, and gourds-producing countries (FAO 2022). In 2020, pumpkin production in the US totaled \$194 million, with Indiana ranked fifth among the top pumpkin-producing states in the US, with 2428 ha valued at ~ \$16 million (USDA NASS 2021). Although production practices can vary widely, pumpkins are usually direct-seeded into bare ground rows placed 1.2 to 1.8 m apart. Pumpkins can be bushy or vining. In-row seed spacing is determined based on this distinction and ranges from 46 to 240 cm (Phillips 2021). The wide row and plant spacing required for this crop's growth allows weeds to establish easily.

Weeds can reduce pumpkin yield by as much as 67% (Walters and Young 2010). Common, difficult-to-control weeds in Midwestern cucurbit production are Eastern black nightshade (*Solanum ptychanthum* Dunal), marestalk (*Erigeron canadensis* L.), morningglory spp. (*Ipomoea* spp. L.), pigweeds (*Amaranthus* spp. L.), giant ragweed (*Ambrosia trifida* L.), wild buckwheat [*Fallopia convolvulus* (L.) Á.Löve], Canada thistle [*Cirsium arvense* (L.) Scop.], dandelion (*Taraxacum officinale* F.H. Wigg.), field bindweed (*Convolvulus arvensis* L.), Johnsongrass [*Sorghum halepense* (L.) Pers.], and yellow nutsedge (*Cyperus esculentus* L.) (IPMdata 2005). Weed management in conventional pumpkin production generally includes chemical control. Preemergence (PRE) herbicides combined with shielded postemergence (POST) row middle application of nonselective herbicides are those most often used by pumpkin producers (Phillips 2021).

Herbicides can significantly reduce production costs by helping farmers overcome labor scarcity and elevated costs associated with other weed management practices. It has been estimated that crop production would decrease by 20% in the US without herbicides (Gianessi and Reigner 2007). Farmers can only use state-registered herbicides for tolerant crops. In Indiana, few herbicides have been registered for use in pumpkin production (Phillips 2021), so farmers have to rely on the same herbicides year after year, which can contribute to herbicide resistance (Evans et al. 2016; Gressel 1991).

Fomesafen is a Group 14 herbicide that inhibits protoporphyrinogen oxidase (PPO). It is registered with 24C Special Local Need labels for use PRE after pumpkin seeding but before crop emergence in Illinois, Kansas, Michigan, Minnesota, and Ohio, where it successfully controls several of the problematic weeds found in pumpkin production. However, it is not registered in

Indiana (Phillips 2021). Farmers have noted this inconsistency in extension meetings and want fomesafen to be registered in Indiana as well. In-state tolerance data is desirable for a herbicide to be registered with a 24C label. A crop is considered tolerant when the applied herbicide does not cause any toxicity (Pitty 1995) or when it shows some injury but completely recovers by the end of its growing cycle (Seefeldt et al. 1995).

Dose-response studies can be used to derive a model from the biological effect of a herbicide, or multiple herbicides on a crop, or multiple crops (Streibig 1980). Dose-response curves are often sigmoidal and constrained by an upper and a lower limit. The upper and lower limits are defined by the response from non-treated plants (control) and the highest dose applied (Knezevic et al. 2007). Our objective was to fit fomesafen dose-response curves to evaluate the biological response of two pumpkin cultivars. With this, we can determine possible outcomes regarding crop tolerance at other fomesafen rates within the range of rates used in our study.

2.3 Materials and Methods

Fomesafen dose-response field trials were conducted in 2020 and 2021 at the Southwest Purdue Agricultural Center (SWPAC), Vincennes, IN, US (38.73°N, 87.48°W) and in 2021 at the Pinney Purdue Agricultural Center (PPAC), Wanatah, IN, US (41.44°N, 86.93°W). At SWPAC, soil types were a Conotton gravelly loam (loamy-skeletal, mixed, active, mesic Typic Hapludalfs) with 0.8% organic matter (OM) and pH 6.6 in 2020, and a mixture of Lomax loam (coarse-loamy, mixed, superactive, mesic Cumulic Hapludalfs) and Lyles sandy loam (coarse-loamy, mixed superactive, mesic Typic Endoaquolls) with 0.9% OM and pH 6.4 in 2021. At PPAC, the soil type was a mixture of Tracy sandy loam (coarse-loamy, mixed, active, mesic Ultic Hapludalfs) and Bourbon sandy loam (coarse-loamy, mixed, active, mesic Aquultic Hapludalfs) with 1.7% OM and pH 6.8.

Fields were prepared with tillage prior to the formation of raised beds. Raised beds with subsurface drip tape were prepared on June 17, 2020, and June 15, 2021, at SWPAC and June 2, 2021, at PPAC. The experimental design was a randomized complete block design with a split-plot treatment arrangement and four replications. The main plots consisted of the fomesafen rate and the subplots of the pumpkin cultivar randomly placed within each main plot. Subplots were 27 m² and contained three 4.9 m long rows, 1.8 m apart. Fomesafen (Reflex®, Syngenta Crop Protection, LLC, Greensboro, NC) rates were 0, 280, 560, 840, and 1,120 g ai ha⁻¹, where 0 g ai

ha⁻¹ was the non-treated control. Pumpkin cultivars were 'Bayhorse Gold' and 'Carbonado Gold' (Rupp Seeds, Inc. Wauseon, OH). Crop fertilization, irrigation, and diseases and insect management followed recommendations by Phillips (2021).

Two pumpkin seeds were hand-planted into the same hole 1.2 m apart in-row on June 18, 2020, and June 16, 2021, at SWPAC, and on June 2, 2021, at PPAC, and thinned to one plant per hole 2 to 4 wk after planting. Fomesafen was broadcast-applied on top of the bed and respective row middles (Figure 2.1A) within 1 d of planting. To help manage weeds, 1,070 g ai ha⁻¹ *S*-metolachlor (Dual Magnum®, Syngenta Crop Protection, LLC, Greensboro, NC) was broadcast-applied in a separate application across all plots within 1 d after applying fomesafen. At SWPAC, both herbicides were applied using a tractor-mounted PTO-driven Hypro 7560 C roller pump with four TeeJet XR 8003 VS nozzles (Spraying Systems Co., Wheaton, IL) calibrated to deliver 187 L ha⁻¹ at 207 kPa. At PPAC, fomesafen was applied using a CO₂-pressurized backpack sprayer equipped with four TeeJet XR 11004 VS nozzles calibrated to deliver 187 L ha⁻¹ at 165 kPa and *S*-metolachlor was applied using PTO-driven Hypro model 6500 C roller pump with four TeeJet XR 8003 VS nozzles calibrated to deliver 187 L ha⁻¹ at 138 pKa.

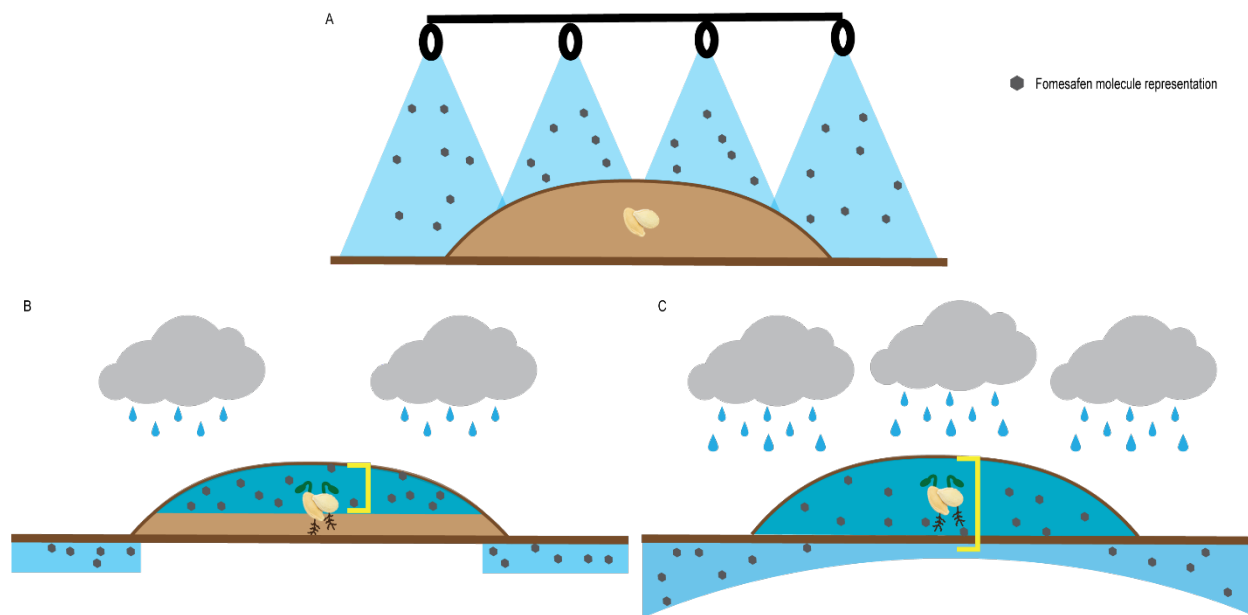


Figure 2.1. Preemergence herbicide application (A) and the effect of rain on the herbicide zone distribution (B and C). (B) Illustrates a scenario where low to moderate rain shortly after planting moves the herbicide to the weeds' grow zone but not the crop's root zone. (C) Illustrates a scenario where excessive rain shortly after planting moves the herbicide to the crop's root zone, increasing the risk of crop uptake.

Data collection included counting the number of emerged pumpkin plants 2 wk after treatment (WAT). Visual pumpkin injury was rated using a scale of 0 (no injury) to 100% (crop death) at 2, 4, 6 and 8 WAT. Weed control was rated 4 WAT on a scale of 0 (no control) to 100% (complete control) relative to the 0 g ha⁻¹ fomesafen treatment. After the 4 WAT weed control rating, weeds were removed either by hand or with hoes or cultivators to maintain plots weed-free and avoid yield loss due to weed interference. Pumpkin harvest was performed on September 12, 2020 [86 days after planting (DAP)] and September 17, 2021 (93 DAP) at SWPAC, and on September 1, 2021 (91 DAP) at PPAC. All fruits were harvested from each plot, individually weighed, and the color of each fruit was recorded. A fruit was classified as marketable if it weighed ≥ 1.5 kg. Marketable fruits were categorized as orange ($\geq 50\%$ of the surface area was orange), green ($< 50\%$ of the surface area was orange), and immature (green, tender rind). Individual marketable fruit weight average was calculated by dividing the marketable yield by the marketable fruit number of each category.

Emergence and marketable orange pumpkin yield and fruit number data were converted to a percent of the non-treated control using Equation 2.1:

$$\text{Percent control} = \frac{B}{M} \times 100 \quad [2.1]$$

where M was the average of the non-treated control variable value pooled across the four repetitions within a location-year for each pumpkin cultivar and B was the variable value of each data point for each location-year.

Data were subjected to statistical analysis using R software (RStudio®, PBC, Boston, MA). Data were first analyzed for each location-year as a linear model and subjected to ANOVA to determine if the models were statistically significant for each trial. If models were significant, data were combined across all three or only two location-years to check if the normality of the data was affected and determine if statistically interactions ($P \leq 0.05$) existed between fomesafen rate, pumpkin cultivar, and location-year for each response variable. If data normality was affected or interactions between the explanatory variables existed, data are presented separately. Response variables were emergence as a percent of the non-treated control, visual pumpkin injury at 2, 4, and 6 WAT, weed control 4 WAT, marketable orange pumpkin yield and fruit number as a percent of the non-treated control, and marketable pumpkin yield (kg 27 m⁻²), fruit number and average individual fruit weight (kg fruit⁻¹) for the green and immature fruits. Visual pumpkin injury and weed control data were arcsin-squareroot transformed for analysis and are presented as back-

transformed data. Data from the non-treated check were excluded from the visual pumpkin injury and weed control data analysis due to zero variance.

Significant response variables' models were then subjected to non-linear regression analyses using the package *drc* in R software and fit to either a three-parameter log-logistic model using Equation 2.2:

$$y = \frac{d}{1 + \text{Exp} [b(\log x - \log e)]} \quad [2.2]$$

where y is the predicted response variable value, d is the upper limit, b is the growth rate, e is the inflection point, and x is the fomesafen rate in g ai ha⁻¹, or a three-parameter logistic model using Equation 2.3:

$$y = \frac{d}{1 + \text{Exp} [b (x - e)]} \quad [2.3]$$

where y is the predicted response variable value, d is the upper limit, b is the relative slope, e is the inflection point, and x is the fomesafen rate in g ai ha⁻¹. Non-linear models fit were analyzed with a lack-of-fit test, where a $P > 0.05$ indicates that the non-linear model provides adequate description of the data. If data did not fit a model, a Tukey's Honestly Significant Difference (HSD) means separation test was performed at a $P \leq 0.05$ significance level.

2.4 Results and Discussion

2.4.1 Pumpkin Emergence

The SWPAC-2020 emergence data as a percent of the non-treated control was separated from the 2021 data due to significant fomesafen rate-by-location-year interaction ($F_{8, 89}=7.32$, $P=1.98 \times 10^{-7}$) when pooled across all three location-years. However, emergence data from both locations in 2021 were pooled. Data were pooled across cultivars due to no significant fomesafen rate-by-cultivar interactions. A three-parameter log-logistic model (Equation 2.2) was fit to the SWPAC-2020 and the pooled 2021 data. At SWPAC-2020, as fomesafen rate increased from 280 to 1,120 g ha⁻¹, predicted emergence decreased from 85 to 25% of the non-treated control at SWPAC-2020 (Figures 2.2 and 2.3), but only from 99 to 74% in 2021 at both locations (Figure 2.2).

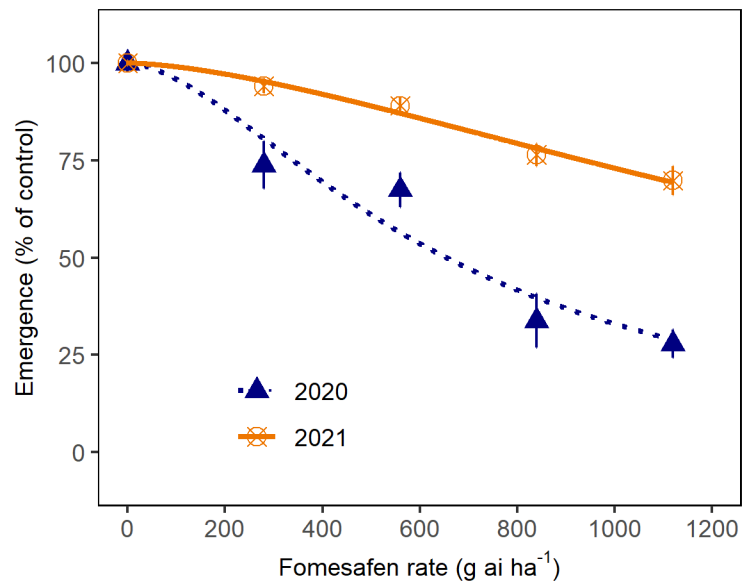


Figure 2.2. Effect of fomesafen rate on Jack O'Lantern pumpkin emergence as a percent of the non-treated control pooled across cultivars at the Southwest Purdue Agricultural Center (SWPAC) in 2020 and across cultivars and locations [SWPAC and the Pinney Purdue Agricultural Center] in 2021 described with a three-parameter log-logistic model $[d/(1 + \text{Exp}[b(\log x - \log e)])]$. Parameters for 2020: $b = 2$, $d = 100$, and $e = 654$; lack-of-fit $P=0.056$. Parameters for 2021: $b = 2$, $d = 100$, and $e = 1875$; lack-of-fit $P=0.710$.

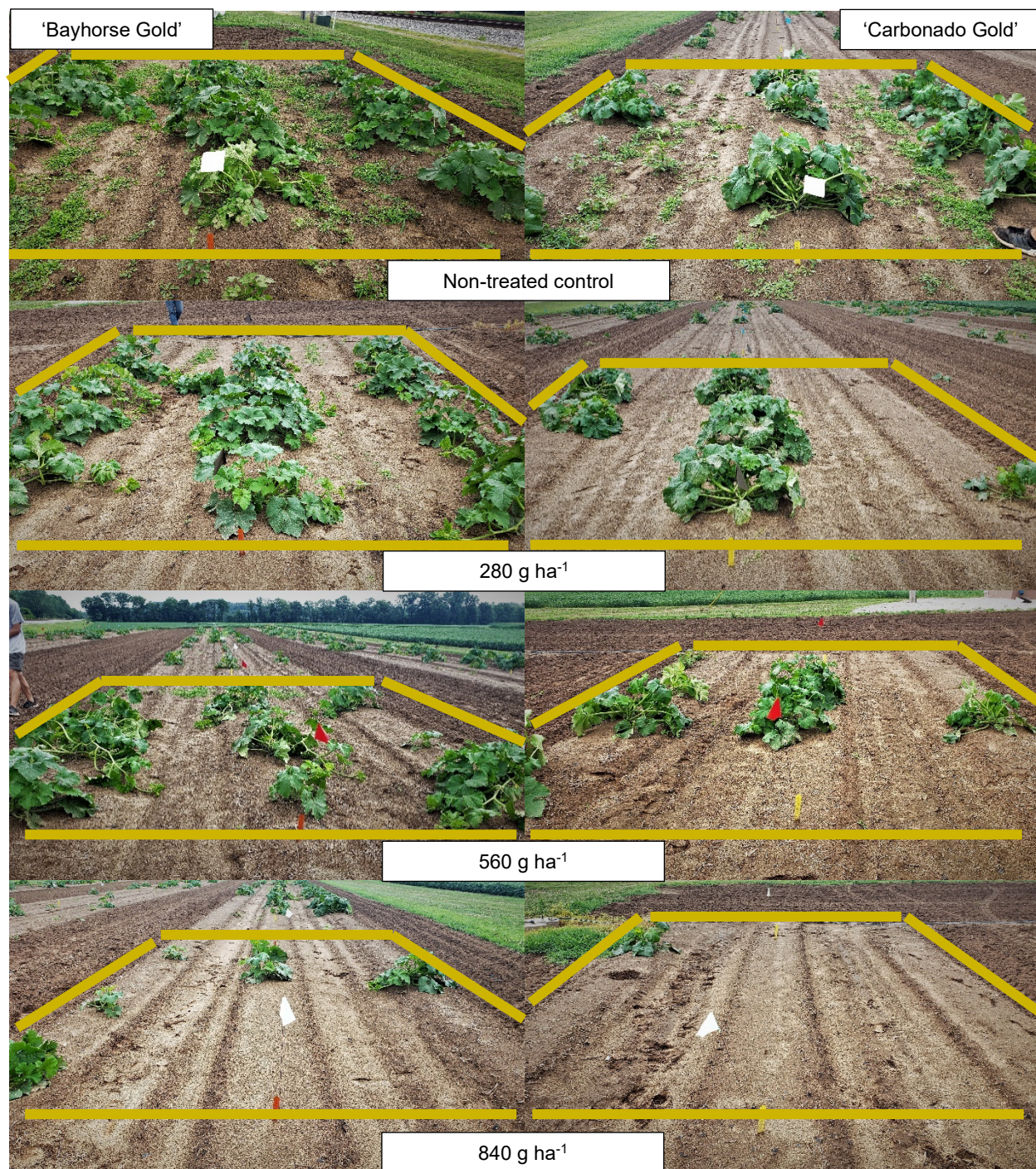
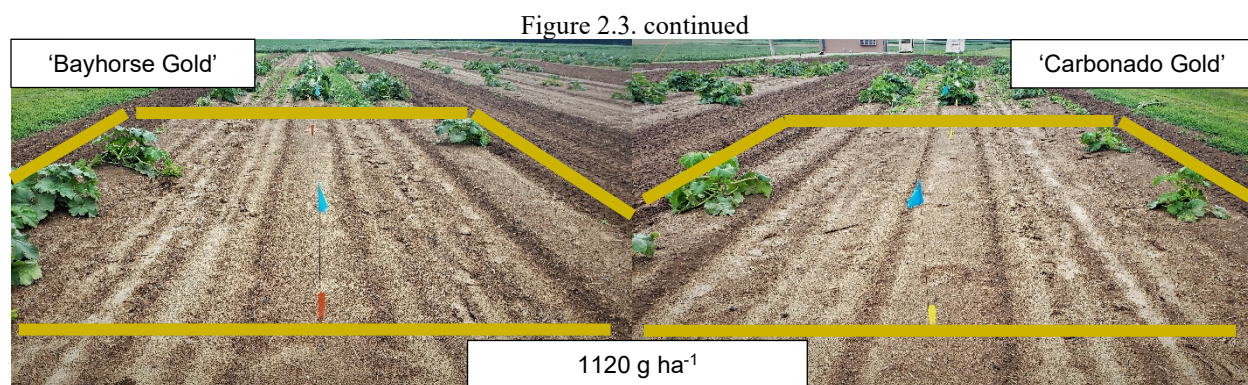


Figure 2.3. Effect of fomesafen rate on Jack O' Lantern pumpkin emergence and weed control 4 wk after treatment for two cultivars ('Bayhorse Gold' and 'Carbonado Gold') at the Southwest Purdue Agricultural Center in 2020.



We attributed the reduction in emergence to excessive rainfall. Cumulative rainfall within 2 WAT in 2020 was 120 mm, but in 2021 it rained only 44 mm at SWPAC and 17 mm at PPAC (Table 2.1). Soil-applied herbicide uptake happens mainly in the root for dicotyledonous plants via diffusion, interception, or mass flow. Herbicide uptake via mass flow, the process where the herbicide moves due to the hydrostatic gradient, accounts for the majority of the herbicide uptake (Menendez et al. 2014). Rain is necessary to incorporate PRE herbicides into the soil profile (Figure 2.1B). However, excessive rain moves the herbicide deeper in the soil profile into the crop's root zone (Figure 2.1C), enhancing the hydrostatic gradient, thus increasing herbicide absorption. Fomesafen is highly mobile under water-saturated soil conditions, especially in soils with low OM content, high pH, and a high proportion of sand content (Guo et al. 2003; Li et al. 2019; Weber et al. 1993; Weber et al. 2004). Low soil OM content and the excessive rain through the first 2 wk at SWPAC-2020 increased fomesafen available for uptake in the crop root zone in an early, vulnerable stage, thus reducing emergence.

Table 2.1. Biweekly rainfall accumulation for the first eight wk after fomesafen application at the Southwest Purdue Agricultural Center (SWPAC) in 2020 and 2021 and the Pinney Purdue Agricultural Center (PPAC) in 2021.

Location-Year	Planting date	Date of the first rain	Cumulative rainfall ^a			
			0 to 2 WAT	2 to 4 WAT	4 to 6 WAT	6 to 8 WAT
-----mm-----						
SWPAC-2020	18-Jun	21-Jun	120.40	23.37	69.60	116.59
SWPAC-2021	16-Jun	19-Jun	43.69	134.87	13.21	90.68
PPAC-2021	2-Jun	7-Jun	16.76	157.23	69.09	68.58

^aData from the Midwest Regional Climate Center, West Lafayette, IN.

Peachey et al. (2012) reported that fomesafen at 560 g ha⁻¹ reduced the emergence of 'Eureka' cucumber (*Cucumis sativus* L.), 'Golden Delicious' Hubbard squash (*Cucurbita maxima* Duchesne), 'Dickinson' pumpkin and 'Ultra' butternut winter squash (*Cucurbita moschata* Duchesne ex. Poir.), and 'Elite' zucchini, 'Yellow Crookneck' summer squash, and 'Small Sugar' pumpkin (*C. pepo*) on average from 2.8 to 2.1 plant m⁻¹ (25% reduction). However, the pumpkin cultivar 'Small Sugar' emergence was not affected by the fomesafen rate of 280 g ha⁻¹ and was reduced only by 8% at the fomesafen rate of 560 g ha⁻¹. Our results differ from their result because we found a somewhat wide range of emergence reduction (5 to 21%) even at the lowest fomesafen rate of 280 g ha⁻¹, presumably because of the soils' OM content. Peachey et al. (2012) reported OM content $\geq 2.1\%$ for all soil types, which could have increased fomesafen sorption to the soil. Also, other environmental conditions like rainfall must be taken into consideration. Similar to our results, Ferebee (2018) reported that fomesafen at 280 g ha⁻¹ reduced the plant stand of 'Kratos' (*C. moschata*) and 'Cougar' (*C. pepo*) pumpkin by 20% and 63 to 75%, respectively. These trials were also conducted in soils with low OM content (1%). Likewise, they attributed the reduction in plant stand to rainfall (7 to 26 mm) shortly after planting (2 to 3 DAP).

2.4.2 Pumpkin Injury

We observed necrosis (Figure 2.4A), small white and brown spots (Figure 2.4B&C), chlorosis (Figure 2.4D&E), and stunting injury (Figure 2.5). Injury data were analyzed separately by location-year due to a significant fomesafen rate-by-location-year interaction. Injury was pooled across both cultivars in each location-year due to a non-significant fomesafen rate-by-cultivar interaction. Injury data 4 WAT at SWPAC-2020 and PPAC-2021 were fit a three-parameter logistic model (Equation 2.3; Figure 2.6). All other injury data were subjected to a Tukey's HSD mean comparison test (Table 2.2). At 2 WAT, as the fomesafen rate increased from 280 to 1,120 g ha⁻¹, injury increased from 6 to 28% at SWPAC-2021 and 5 to 36% at PPAC-2021 (Table 2.2). At 4 WAT, predicted injury increased from 7 to 26% (SWPAC-2020) and 4 to 50% (PPAC-2021) (Figure 2.6), and observed injury data at SWPAC-2021 increased from 0 to 13% (Table 2.2). At 6 WAT, injury ranged from 1 to 11% at SWPAC-2020 and 2021 and from 1 to 21% at PPAC-2021 (Table 2.2). Injury 8 WAT increased from 0 to 4% at SWPAC-2021 and 5 to 33% at PPAC-2021 (Table 2.2). With the exception of PPAC-2021, injury decreased from 2 to 8 WAT.

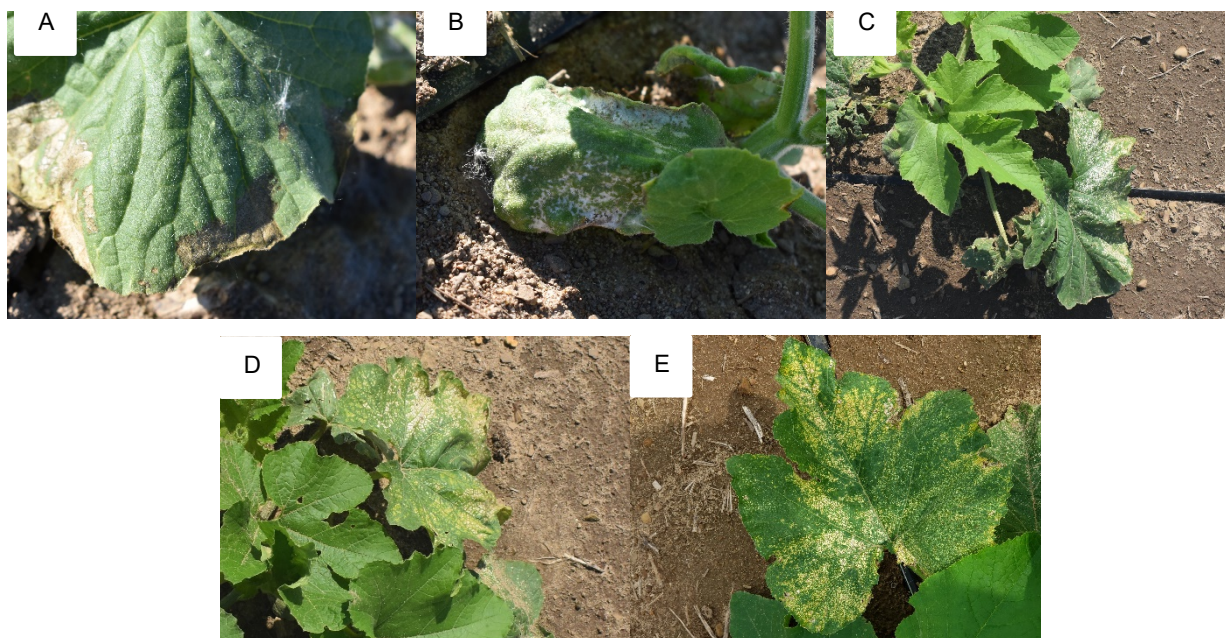


Figure 2.4. Jack O'Lantern pumpkin injury symptoms at a fomesafen rate of $1,120 \text{ g ha}^{-1}$. Necrosis 2 wk after treatment (WAT) (A), small white and brown spots at 2 (B) and 4 WAT (C), and chlorosis at 4 (D) and 6 WAT (E) at the Pinney Purdue Agricultural Center in 2021.



Figure 2.5. Non-treated control (0 g ha^{-1}) vs. highest fomesafen rate ($1,120 \text{ g ha}^{-1}$) treatment to represent Jack O'Lantern pumpkin stunting at 6 wk after transplanting at the Southwest Purdue Agricultural Center in 2021.

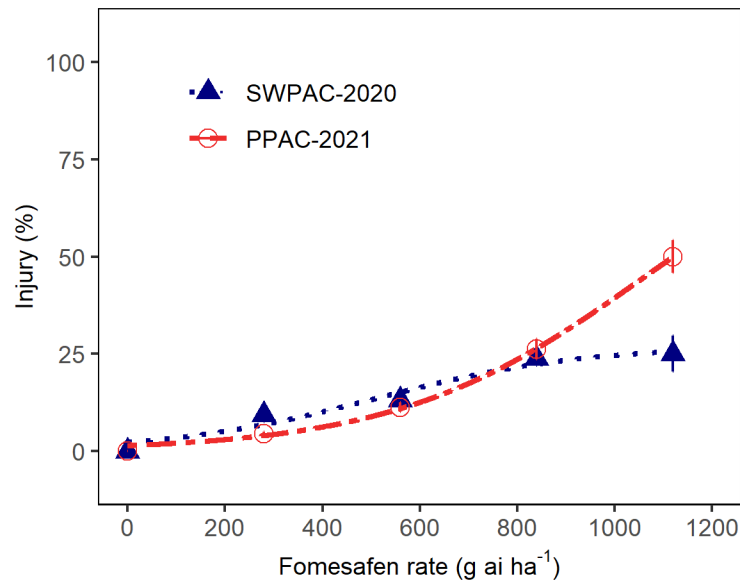


Figure 2.6. Effect of fomesafen rate on Jack O'Lantern pumpkin injury at 4 wk after treatment at the Southwest Purdue Agricultural Center in 2020 (SWPAC-2020) and the Pinney Purdue Agricultural Center in 2021 (PPAC-2021), described with a three-parameter logistic model $[d/(1 + \text{Exp}[b(x - e)])]$. Parameters for SWPAC-2020: $b=-0.005$, $d=27$, and $e=509$; lack-of-fit $P=0.275$. Parameters for PPAC-2021: $b=-0.004$, $d=89$, and $e=1060$; lack-of-fit $P=0.819$.

Table 2.2. Jack O'Lantern pumpkin injury and standard error (SE) at 2, 4, 6, and 8 wk after treatment (WAT) with fomesafen at the Southwest Purdue Agricultural Center (SWPAC) in 2020 and 2021 and the Pinney Purdue Agricultural Center (PPAC) in 2021 pooled across pumpkin cultivars 'Bayhorse Gold' and 'Carbonado Gold'.

Rate	Pumpkin Injury ^a							
	2 WAT		4 WAT ^b	6 WAT			8 WAT	
	SWPAC-2021	PPAC-2021	SWPAC-2021	SWPAC-2020	SWPAC-2021	PPAC-2021	SWPAC-2021	PPAC-2021
g ha ⁻¹	-----%							
280	6 (2) a ^c	5 (1) a	0 (0) a	1 (1) a	1 (1)	1 (1) a	0 (0) a	5 (2) a
560	9 (1) ab	14 (2) b	2 (1) ab	1 (1) a	5 (2)	4 (2) a	1 (1) a	6 (3) a
840	15 (3) bc	28 (4) c	6 (2) b	11 (1) b	1 (1)	13 (3) b	0 (0) a	16 (3) ab
1120	28 (5) c	36 (4) c	13 (2) c	7 (3) ab	11 (4)	21 (3) b	4 (1) b	33 (5) b

^aInjury was arcsin transformed for analysis and back-transformed for the table.

^bData for SWPAC-2020 and PPAC-2021 at 4 WAT were fit a three-parameter logistic model (Figure 2.6).

^cMeans separation using Tukey's HSD test $P \leq 0.05$. Means followed by the same letter are not significantly different. Lack of letters indicates that the F statistic was not significant at a $\alpha=0.05$.

Heavy rainfall events increase the chance of injury due to the splashing of fomesafen from the soil onto the leaves (Peachey et al. 2012). This could explain the necrosis and chlorosis scattered patterns on cotyledons and leaves laying close to the ground. Injury 8 WAT was mainly stunting. Lingenfelter and VanGessel (2016) also mentioned stunting up to 8 WAT with fomesafen applied at 175 and 350 g ha⁻¹ to five pumpkin cultivars (*C. pepo* and *C. maxima*). As mentioned before, fomesafen persistence in the soil varies with OM, sand content, and pH. Because of this, fomesafen half-live values in diverse soil types ranged variably from 4 to 66 d (Li et al. 2019; Mueller et al. 2014). Pumpkin injury inconsistency across trials was possibly due to its variable persistence depending on soil characteristics and other environmental factors such as microbial degradation (Feng et al. 2012; Mielke et al. 2022) and rainfall pattern. PPAC-2021 had the most prolonged injury (Table 2.2), likely because there could have been more available herbicide due to less leaching. Herbicide was probably less likely to leach at PPAC-2021 due to a higher OM content (1.7%) than the other two location-year and less rainfall within the first 8 WAT (Table 2.1).

2.4.3 Weed Control

Weed control data were analyzed separately by location and year. Increasing fomesafen rates did not affect weed control at SWPAC-2021 ($F_{7,21} = 2.01$, $P=0.102$). Relative to the 0 g ha⁻¹ fomesafen rate treatment that only received *S*-metolachlor, weed control was above 90% for all fomesafen rates in all three location-years 4 WAT (Table 2.3 and Figure 2.3). Fomesafen controlled carpetweed (*Mollugo verticillata* L.), common ragweed (*Ambrosia artemisiifolia* L.), morningglory spp., pigweeds, and prickly sida (*Sida spinosa* L.), and grass species at SWPAC-2020; carpetweed, common purslane (*Portulaca oleracea* L.), and grass species at SWPAC-2021; and carpetweed, common lambsquarters (*Chenopodium album* L.), giant ragweed, morningglory spp., velvetleaf (*Abutilon theophrasti* Medik.), volunteer soybean [*Glycine max* (L.) Merr.] and grass species at PPAC-2021. Weed control during the first 4 wk after emergence is ideal in pumpkin production due to its critical weed-free period of 4 to 6 wk (Dittmar and Boyd 2019; eOrganic 2015). Because plots were maintained weed-free after 4 WAT, we cannot determine from this study how fomesafen rate-related weed control would have impacted crop growth and yield.

Table 2.3. Effect of fomesafen rate on weed control and standard error (SE) 4 wk after treatment at the Southwest Purdue Agricultural Center (SWPAC) in 2020 and 2021 and the Pinney Purdue Agricultural Center (PPAC) in 2021.

Rate	Weed control ^a		
	SWPAC-2020	SWPAC-2021	PPAC 2021
g ha ⁻¹	-----%-----		
280	95 (1) c ^b	90 (3.3)	92 (2.1) b
560	98 (0.8) bc	98 (1.1)	94 (1.7) b
840	99 (0.2) ab	97 (1.4)	99 (0.6) a
1120	100 (0) a	99 (0.8)	99 (0.6) a

^aWeed control was arcsin transformed for analysis and back-transformed for the table.

^bMeans separation using Tukey's HSD test $P \leq 0.05$. Means followed by the same letter are not significantly different. Lack of letters indicates that the F statistic was not significant at a $\alpha=0.05$.

2.4.4 Pumpkin Yield

Due to a significant fomesafen rate-by-location-year interaction, marketable orange pumpkin yield ($F_{8,88} = 4.78$, $P = 6.67 \times 10^{-5}$) and fruit number ($F_{8,89} = 5.32$, $P = 1.81 \times 10^{-5}$) as a percent of the non-treated control data were analyzed separately by location-year. There were no differences in yield, nor fruit number among treatments at SWPAC-2021, where the average marketable orange pumpkin yield was 109 kg 27 m⁻² and fruit number was 16 fruits 27 m⁻² pooled across all treatments (data not shown). Marketable orange pumpkin yield data as a percent of the non-treated control at SWPAC-2020 and PPAC-2021 fit a three-parameter log-logistic model (Equation 2.2, Figure 2.7A). As the fomesafen rate increased from 280 to 1,120 g ha⁻¹, marketable orange pumpkin yield decreased from 95 to 24% of the non-treated control (102 kg 27 m⁻²) at SWPAC-2020 and 99 to 66% of the non-treated control (119 kg 27 m⁻²) at PPAC-2021 (Figure 7A). Marketable orange pumpkin fruit number as a percent of the non-treated control fit a three-parameter log-logistic model at SWPAC-2020 (Equation 2.2) and a three-parameter logistic model at PPAC-2021 (Equation 2.3) (Figure 2.7B). As the fomesafen rate increased from 280 to 1,120 g ha⁻¹, the marketable orange pumpkin fruit number decreased from 94 to 21% of the non-treated control (15 fruits 27 m⁻²) at SWPAC-2020 and 98 to 74% of the non-treated control (17 fruits 27 m⁻²) at PPAC-2021 (Figure 2.7B).

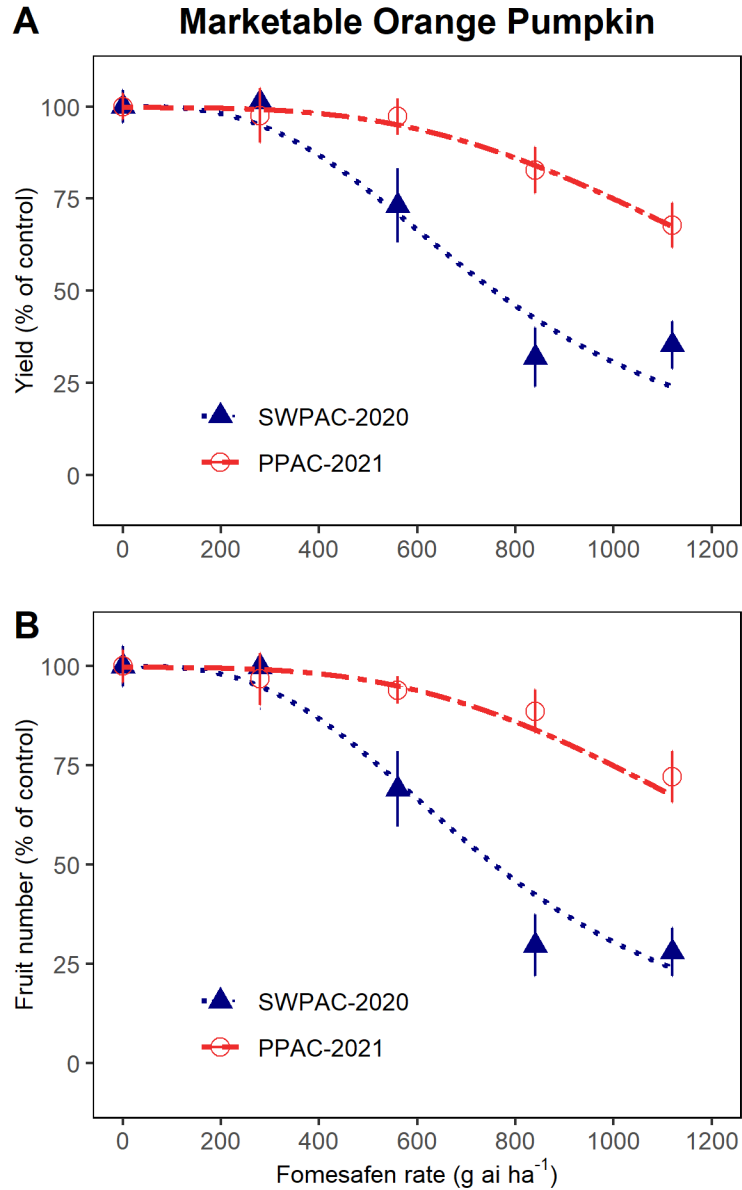


Figure 2.7. Effect of fomesafen rate on marketable Jack O'Lantern pumpkin yield (A) and fruit number (B) as a percent of the non-treated control at the Southwest Purdue Agricultural Center in 2020 (SWPAC-2020) and at the Pinney Purdue Agricultural Center in 2021 (PPAC-2021). Marketable pumpkin yield at both location-year and fruit number at SWPAC-2020 described with a three-parameter log-logistic model $[d/(1 + \text{Exp}[b(\log x - \log e)])]$. Parameters for (A) SWPAC-2020: $b = 3, d = 100, \text{ and } e = 757$; lack-of-fit $P=0.241$. Parameters for (A) PPAC-2021: $b = 3, d = 100, \text{ and } e = 1402$; lack-of-fit $P=0.869$. Parameters for (B) SWPAC-2020: $b = 3, d = 100, \text{ and } e = 713$; lack-of-fit $P=0.500$. Fruit number at PPAC-2021 described with a three-parameter logistic model $[d/(1 + \text{Exp}[b(x - e)])]$. Parameters for (B) PPAC-2021: $b = 0.004, d = 99, \text{ and } e = 1387$; lack-of-fit $P=0.930$.

Fomesafen rate did not significantly influence the individual marketable orange pumpkin fruit weight nor the marketable green and immature pumpkin yield, fruit number, and individual fruit weight (data not shown).

Although predicted pumpkin marketable orange pumpkin yield and fruit number decreased as the fomesafen rate increased from 280 to 1,120 g ha⁻¹, the values for the lowest fomesafen rate used were not statistically different from the non-treated control. These results confirm the results of Lingenfelter and VanGessel (2016) and Peachey et al. (2012), who reported that pumpkin yield was not affected by fomesafen rates of 175 and 350, and 280 and 560 g ha⁻¹, respectively. Lingenfelter and VanGessel (2016) noted no effect on individual fruit weight as well. Because the individual marketable orange pumpkin fruit weight average was not affected by any fomesafen rate, marketable yield loss at high fomesafen rates was attributed only to the reduced plant stand.

2.5 Conclusions

Overall, the recommended, labeled fomesafen rate for use PRE in other Midwestern states of 280 g ha⁻¹ was safe for use PRE in Jack O'Lantern pumpkin cultivars 'Bayhorse Gold' and 'Carbonado Gold' at SWPAC and PPAC. Despite the impact on emergence at SWPAC-2020, the pumpkins recovered and predicted yield loss was only 5%, and visible injury was less than 7% in all the ratings at all locations. Also, adding this fomesafen rate to a blanket application of S-metolachlor improved weed control (>90% compared to the non-treated control). However, in soils with a low OM content and a high portion of sand, heavy rainfall events shortly after planting are expected to move the herbicide to the crop root zone and affect emergence. Consequently, it is necessary to plan its application carefully. If emergence reduction happens, significant yield loss due to reduced plant stand is expected only at fomesafen rates higher than 280 g ha⁻¹.

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CHAPTER 3. DOSE-RESPONSE OF PLASTICULTURE SUMMER SQUASH AND TRIPLOID WATERMELON TO FOMESAFEN APPLIED PRE-TRANSPLANTING

3.1 Abstract

Dose-response trials to determine the tolerance of summer squash and watermelon to fomesafen applied pre-planting over-the-top of plastic mulch were performed between 2020 and 2021 at three Indiana locations: the Meigs Horticulture Research Farm (MEIGS), the Pinney Purdue Agricultural Center (PPAC), and the Southwest Purdue Agricultural Center (SWPAC). The experiments had a split-plot arrangement in which the main plot was either of the five herbicide rates, and the subplot was either of the two cultivars. Summer squash injury included necrotic leaf margin, chlorosis, brown and white spots, and stunting. Injuries except stunting were attributed to the herbicide splashing from the plastic or soil onto the leaves. Fomesafen rates increased from 262 to 1,048 g ai ha⁻¹ in 2020 at both locations, and from 280 to 1,120 g ai ha⁻¹ in 2021 at MEIGS did not affect summer squash yield. However, in 2021 at PPAC, rates from 280 to 1,120 g ha⁻¹ delayed summer squash harvest and decreased marketable yield from 95 to 61% of the 0 g ha⁻¹ non-treated control. Watermelon injury included bronzing, also attributed to splashing, and stunting. Fomesafen rates from 210 to 840 g ai ha⁻¹ did not affect marketable watermelon yield and fruit number. Crops' safety was attributed to rain washing off most of the herbicide from the plastic before transplanting or no excessive rain shortly after transplanting. At PPAC in 2021, summer squash's severe impact was attributed to no rain before transplanting and excessive cumulative rain after transplanting. Overall, the 1x rates used for each trial are safe for use 1 d before planting summer squash and 6 to 7 d before transplanting watermelon. Rainfall before transplanting may be necessary to reduce the risk of the herbicide moving into the crop's root zone through the punched hole.

Nomenclature

Fomesafen; summer squash, *Cucurbita pepo* L.; watermelon, *Citrullus lanatus*, (Thunb.) Matsum & Nakai

Keywords

Dose-response; soil-residual; injury; weed control; yield

3.2 Introduction

Summer squash and watermelon are high-value cucurbits crops produced in the United States (US). In 2020, production of squash in the US totaled 345 million kg on 18 thousand harvested hectares with a value of \$218 million. Watermelon production in the US totaled 1.7 billion kg on 39 thousand harvested hectares valued at \$575 million. Midwestern states are among the top cucurbit-producing states. Michigan ranked first among the top squash-producing states and Indiana ranked fifth among the top watermelon-producing states (USDA-NASS 2021).

Summer squash and watermelon are usually transplanted into raised-beds covered with plastic polyethylene mulch. Row spacing ranges from 1.2 to 1.8 m for summer squash and 1.8 to 3.7 m for watermelon. In-row spacing ranges from 46 to 61 cm for summer squash, and 90 to 180 cm for watermelon (Phillips 2021). Plastic mulch successfully aids with in-row weed control (Bonanno 1996; Skidmore et al. 2019). However, row-middle weeds must be controlled using other strategies. Plasticulture summer squash marketable yield was reduced by 11 and 19% in 2013 and 2014, respectively, and average muskmelon (*Cucumis melo* L.) individual fruit weight was reduced from 2 to 1.7 kg when no in-row weed control strategy (row cover) was applied (Tillman et al. 2015a, 2015b). Weeds also interfere with harvesting these manually-harvested crops, exposing laborers to allergens (Gadermaier et al. 2004; Piotrowska-Weryszko et al. 2021), increasing accidents (de Oliveira Procópio et al. 2015), or complicating the harvesting process.

Several technologies can be used to control row-middle weeds, including plant-based mulches and cultivators. However, they are usually cost-ineffective or labor-intensive (i.e. moving vines before cultivating) for vegetable growers to implement (Peruzzi et al. 2017; Wilhoit et al. 2012). Therefore, herbicides are generally integrated with plasticulture for row-middle weed management. Farmers have widely accepted and adopted herbicide use due to lower production costs and higher yields (Gianessi and Reigner 2007). Soil-residual herbicides, which remain adsorbed to soil particles for moderate to long time, are encouraged because they can delay herbicide resistance (Busi et al. 2020). However, by law, farmers can only use state-registered herbicides for crops that have tolerance to the specific herbicide registered for use on it.

In Indiana, only a few herbicides are registered for preemergence (PRE) use in summer squash and watermelon, including Groups 3 (ethalfluralin and trifluralin), 13 (clomazone) and 15 (*S*-metolachlor) and bensulide (unknown mode of action). Watermelon farmers in Indiana can also use Groups 2 (halosulfuron and imazosulfuron), 3 [dimethyl tetrachloroterephthalate (DCPA) and pendimethalin], 5 (terbacil) and 14 (flumioxazin) (Phillips 2021). Due to the low number of preemergence herbicide groups available for use in these vegetable crops, farmers have to rely on the same herbicides each year or on postemergence (POST) applications, which considerably contributes to the increase in selection pressure for herbicide-resistant weed populations (Evans et al. 2016). If more soil-residual herbicide groups are registered for use for each crop, farmers can integrate soil-residual herbicide mixtures to delay herbicide resistance (Beckie and Reboud 2009; Busi et al. 2020)

Fomesafen, a protoporphyrinogen oxidase inhibitor herbicide (Group 14), is registered for use PRE in cucurbits in some Midwestern states but not in Indiana. It is registered for use in squash production in Illinois, Kansas, Michigan, Minnesota, and Ohio at rates from 140 to 280 g ai ha⁻¹ and in watermelon production in Kansas and Missouri at rates from 175 to 280 g ai ha⁻¹. In Indiana, there is no Group 14 PRE herbicide registered for use in squash. Flumioxazin, a Group 14 herbicide, is registered for as PRE in watermelon and cantaloupe in Indiana with a 24c Special Local Needs label. However, flumioxazin broadcast applied over-the-top of plastic can cause watermelon yield loss (Meyers et al. 2021), probably because it slowly dissipates from plastic, increasing the chance of the herbicide contacting the crop and causing damage (Grey et al. 2009). Specialty crop farmers in Indiana prefer to spray over-the-top of plastic due to lack of hooded spray equipment. To support the registration of fomesafen for use in summer squash and watermelon through a 24C label, it is advisable to have in-state crop tolerance data. A tolerant crop would not exhibit toxicity symptoms, or develop symptoms but recover afterward (Pitty 1995; Seefeldt et al. 1995). Our objective was to evaluate the biological effect of several rates of fomesafen on two summer squash and watermelon cultivars grown in plasticulture.

3.3 Materials and Methods

3.3.1 Summer Squash

In 2020 and 2021, four summer squash dose-response to fomesafen trials were conducted at Meigs Horticulture Research Farm (MEIGS), Lafayette, IN, USA (40.28°N, 86.88°W) and the Pinney Purdue Agricultural Center (PPAC), Wanatah, IN, USA (41.44°N, 86.93°W). At MEIGS, the soil types were a Drummer silty clay loam (fine-silty, mixed, superactive, mesic Typic Endoaquolls) with 2.8% organic matter (OM) and pH 6.9 in 2020 and a mixture of Toronto and Millbrook silt loam (fine-silty, mixed, superactive, mesic Udollic Epiaqualfs) with 2.1% OM and pH 6 in 2021. At PPAC, the soil type was a Tracy sandy loam (coarse-loamy, mixed, active, mesic Ultic Hapludalfs) with 1.4% OM and pH 6.4 in 2020 and 1.6% OM and pH 6.9 in 2021.

Fields were prepared with tillage before the formation of raised beds. Raised beds with subsurface drip tape covered with black polyethylene plastic mulch were prepared on April 28 (MEIGS) and June 1 (PPAC) in 2020 and on May 24 (MEIGS) and June 2 (PPAC) in 2021. Experimental units consisted of a single row, 4.9 m long. Crop fertilization, irrigation and diseases, and insect management followed Phillip's (2021) recommendations. To help manage weeds, S-metolachlor (Dual Magnum®, Syngenta Crop Protection, LLC, Greensboro, NC) was broadcast applied across all plots before or the same day of treatment application at 1.1 and 1.8 kg ai ha⁻¹ at MEIGS, and at 1.6 and 1.1 at PPAC in 2020 and 2021, respectively.

The experiment design was a split-plot with four replications. The main plots consisted of fomesafen rate, and subplots of summer squash cultivar that were randomly placed within each main plot. Fomesafen rates were 0, 262, 524, 786, and 1048 g ai ha⁻¹ in 2020, and 0, 280, 560, 840, and 1120 g ai ha⁻¹ in 2021, where 0 g ai ha⁻¹ was the non-treated control, and 262 and 280 g ai ha⁻¹ were the 1x labeled rate in other Midwestern states for squash production. Summer squash cultivars were 'Blonde Beauty' yellow straightneck squash and either 'Spineless Beauty' (2020) or 'Liberty' (2021) zucchini. Seeds for each cultivar (Rupp Seeds, Inc. Wauseon, OH) were planted into 72-cell trays containing the peat-based Metro-Mix®360 Professional Growing Mix (Sun Gro Horticulture, Agawam, MA) at MEIGS shade house on April 13, 2020 and on April 21, 2021, for the MEIGS trials, and containing Berger BM2 Seed Germination Mix (Hummert International, Earth City, MO) at the Purdue University Horticulture Greenhouses, West Lafayette, IN on June 5, 2020, and June 3, 2021 for the PPAC trials.

Fomesafen (Reflex®, Syngenta Crop Protection, LLC, Greensboro, NC) was broadcast-applied over-the-top of plastic and respective row middles on May 26 on both years at MEIGS, and June 22, 2020, and June 23, 2021, at PPAC with an output of 187 L ha⁻¹. Fomesafen was applied using a CO₂-pressurized backpack sprayer equipped with four TeeJet XR 11003 VS nozzles (Spraying Systems Co., Wheaton, IL) at 200 kPa at MEIGS in 2020 and with four TeeJet XR 11004 VS nozzles at PPAC at 165 kPa (2020) and at 159 kPa (2021). At MEIGS in 2021, fomesafen was applied using a tractor-mounted, compressed air sprayer with four TeeJet XR 8003 VS nozzles at 276 kPa.

One day after spraying fomesafen, planting holes on the plastic were made with a water wheel transplanter at MEIGS and with a manual hole punch at PPAC in both years. Seedlings were hand-transplanted 1.2 m apart, totaling eight plants per subplot. Weeds were removed as required either by hand or with hoes to maintain plots weed-free and avoid yield loss due to weed interference.

Data collection included visual crop injury using a scale of 0 (no injury) to 100% (crop death) compared to the non-treated control and plant stand at 2 and 4 wk after transplanting (WAP). Harvest was initiated on June 23, 2020 and July 2, 2021 at MEIGS, and July 23, 2020 and July 21, 2021 at PPAC. The six plants in the middle of each subplot were harvested twice a wk for 4 wk (8 harvests total). All fruits ≥ 8 cm long were harvested and graded into mature (darker green/yellow, thickened skin), immature (lighter green/yellow, thin skin), and cull (misshapen and rotten). The number of fruits per category was counted and weighed together. Total marketable yield was calculated by adding the total weight of each of the eight harvests pooled across mature and immature fruits.

Total marketable yield was converted to a percent the non-treated control using Equation 3.1:

$$\text{Percent control} = \frac{B}{M} \times 100 \quad [3.1]$$

where M was the average of the non-treated control variable value of the four repetitions for each summer squash cultivar and B was the variable value of each rate by cultivar treatment data point.

Data were subjected to statistical analysis using R software (RStudio®, PBC, Boston, MA). Data were first analyzed for each location-year with a linear model and subjected to ANOVA to determine if the models were statistically significant for each trial. If models were significant, data were combined across locations for each year to check if the normality of the data was affected

and to determine if statistically interactions ($P \leq 0.05$) existed between fomesafen rate, summer squash cultivar, and location for each response variable. If the data's normality was affected or statistically significant interactions between the explanatory variables existed, data are presented separately. Response variables were visual summer squash injury at 2 and 4 WAP, fruit number per harvest, total marketable yield as a percent of the non-treated control, and cull fruits number. Visual injury data were arcsin-squareroot transformed for analysis and are presented as back-transformed data. The visual injury data analysis did not include data from the non-treated control due to zero variance.

All data were subjected to a Tukey's HSD means separation test performed at a 0.05 significance level. Total marketable yield data that showed a response to fomesafen were fit a three-parameter log-logistic model using Equation 3.2:

$$3P \log - \logistic = \frac{d}{1 + \text{Exp} [b(\log x - \log e)]} \quad [3.2]$$

where d is the upper limit, b is the growth rate, e is the inflection point, and x is the fomesafen rate in g ai ha⁻¹. The fit of each non-linear model was analyzed with a lack-of-fit test, where a $P > 0.05$ indicates that the non-linear model provides an adequate description of the data.

3.3.2 Watermelon

Watermelon fomesafen dose-response trials were conducted at the Southwest Purdue Agricultural Center (SWPAC), Vincennes, IN, USA (38.73°N, 87.48°W) and at MEIGS in 2021. At SWPAC, the soil type was a mixture of Lomax loam (coarse-loamy, mixed, superactive, mesic Cumulic Hapludolls) and Lyles sandy loam (coarse-loamy, mixed, superactive, mesic Typic Endoaquolls) with 1.5% OM and pH 6.8 in 2021. At MEIGS, the soil type was a Drummer silty clay loam (fine-silty, mixed, superactive, mesic Typic Endoaquolls) with 2.1% OM and pH 6 in 2021.

'Exclamation' and 'Fascination' triploid watermelon seeds and 'Wingman' pollinizer watermelon seeds were planted on April 19, 2021, in a SWPAC greenhouse into 50-cell black seedling flats containing a peat-based potting media (Metro-Mix 360; Sungro Horticulture, Agawam, MA).

Fields were prepared with tillage before the formation of raised beds. Raised beds with subsurface drip tape covered with black polyethylene plastic mulch were prepared on April 23,

2021, at SWPAC, and on April 28, 2021, at MEIGS. Experimental units consisted of a 27 m² plot containing three 4.9 m long rows, 1.8 m apart at SWPAC, and two 7.4 m long rows, 1.8 m apart at MEIGS. Crop fertilization, irrigation and diseases, and insect management followed Phillips (2021) recommendations. To help manage weeds, *S*-metolachlor at 1.1 kg ha⁻¹ was broadcast-applied across all plots at SWPAC on May 14, 2021, and a tank mix of 40 g ha⁻¹ halosulfuron-methyl (Sandeo®, Canyon Group LLC C/O Gowan Company, Yuma, AZ) and 1.4 kg ha⁻¹ ethalfluralin plus 420 g ha⁻¹ clomazone (Strategy®, Loveland Products, Inc. Greeley, CO) at MEIGS on May 13, 2021.

The experiment design was a split-plot with four replications. Main plots consisted of fomesafen rate and subplots of triploid watermelon cultivars randomly placed within each main plot. Fomesafen rates were 0, 210, 420, 630, and 840 g ha⁻¹, where 0 g ai ha⁻¹ was the non-treated control and 210 g ai ha⁻¹ was the 1x labeled rate in other Midwestern states for watermelon production. At SWPAC, fomesafen was applied using a tractor-mounted PTO-driven Hypro 7560 C roller pump sprayer with four TeeJet XR 8003 VS nozzles calibrated to deliver 187 L ha⁻¹ at 207 kPa on May 13, 2021. At MEIGS, fomesafen was applied using a tractor-mounted, compressed air sprayer with four TeeJet XR 8003 VS nozzles calibrated to deliver 187 L ha⁻¹ at 276 kPa on May 26, 2021.

Triploid watermelon seedlings were hand-transplanted immediately after punching holes in the plastic with a water wheel-transplanter on May 20, 2021, at SWPAC [7 d after treatment (DAT)] and June 1, 2021 at MEIGS (6 DAT). Triploid watermelon seedlings were transplanted 1.2 m apart in each row resulting in 12 triploid watermelon plants per subplot. After that, we planted two pollinizer watermelon seedlings per row of each subplot, resulting in a 1:2 pollinizer-to-triploid watermelon ratio.

Data collection included visual crop injury on a scale of 0 to 100% at 2, 4, and 6 WAP. Weed control was rated 4 WAP on a scale of 0 (no control) to 100% (complete control) relative to the 0 g ha⁻¹ fomesafen rate (non-treated control). After the 4 WAP weed control rating, weeds were removed either by hand or with hoes or cultivators to maintain plots weed-free and avoid yield loss due to weed interference. Watermelon fruits were harvested once wk⁻¹ for 4 wk, beginning July 28, 2021, at SWPAC and August 11, 2021, at MEIGS. We picked fruits when the tendril that developed from the same node as the fruit peduncle was necrotic and the ground spot was yellow.

The weight of each fruit was recorded and classified as marketable (≥ 4 kg) or non-marketable (<4 kg). Total marketable yield and fruit number were calculated as the sum of all four harvests.

Data were subjected to statistical analysis using R software. Data were first analyzed for location with a linear model and subjected to ANOVA to determine if the models were statistically significant for each trial. If models were significant, data were combined across locations to check if the normality of the data was affected and determine if statistically interactions ($P \leq 0.05$) existed between fomesafen rate, watermelon cultivar, and location for each response variable. If the data's normality was affected or statistically significant interactions between the explanatory variables existed, data are presented separately. Response variables were visual watermelon injury at 2, 4, and 6 WAP, weed control at 4 WAP, and total marketable yield and fruit number. Visual injury and weed control data were arcsin-squareroot transformed for analysis and are presented as back-transformed data. Data from the non-treated control were not included in the visual injury and weed control data analysis due to zero variance. Finally, all data were subjected to a Tukey's HSD means separation test was performed at a 0.05 significance level.

3.4 Results and Discussion

3.4.1 Summer Squash

Injury

Summer squash injury included necrotic margins, chlorosis, brown to white spots, and stunting (Figures 3.1 and 3.2). Due to a lack of fomesafen rate-by-cultivar interaction, injury was analyzed across cultivars within each location-year. With the exception of PPAC 2020 which had no visible crop injury, summer squash injury increased with increasing fomesafen rate at 2 WAP and ranged from 8 to 18% at MEIGS 2020, 3 to 19% at MEIGS 2021, and 5 to 28% at PPAC 2021 (Table 3.1). By 4 WAP, there was no visible crop injury at MEIGS 2020. Meaningful injury at PPAC 2020 was present only at the highest fomesafen rate (14% with 1,048 g ha⁻¹ fomesafen), and injury trends at MEIGS 2021 and PPAC 2021 were similar to observations made 2 WAP. Overall, injury from the lowest fomesafen rates used (262 and 280 g ha⁻¹) was minimal ($\leq 9\%$) at 2 and 4 WAP. In 2021, plant stand at 2 and 4 WAP was not significantly affected by fomesafen

rate in any trial. Plants per plot averaged 7.9 at 2 WAP and 7.7 at MEIGS and 7.6 at 2 WAP, and 7.3 at 4 WAP at PPAC (data not shown).

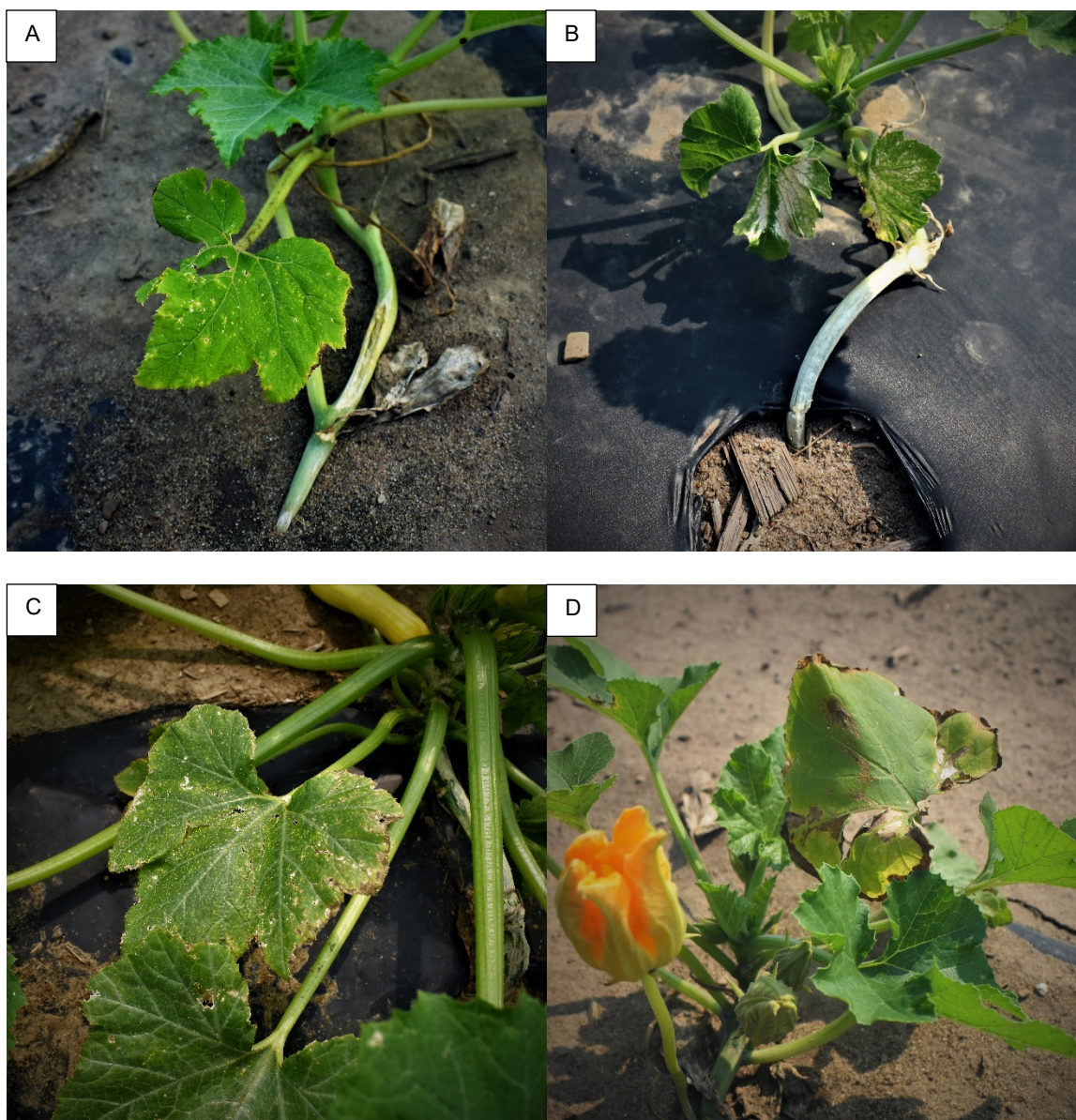


Figure 3.1. Summer squash injury symptoms 2 and 4 wk after transplanting (WAP) at the Pinney Purdue Agricultural Center in 2021. A) 'Blonde Beauty' yellow squash leaf chlorosis, necrotic leaf margins and white spots on the stem at a fomesafen rate of 280 g ai ha⁻¹ and B) 'Liberty' zucchini white spots on leaf and stem at a fomesafen rate of 560 g ai ha⁻¹, 2 WAP. C) 'Blonde Beauty' yellow squash leaf chlorosis and brown to white spots, and necrotic leaf margins and D) 'Liberty' zucchini necrotic leaf margins and brown to white spots on leaves at at a fomesafen rate of 280 g ai ha⁻¹, 4 WAP.



Figure 3.2. Summer squash stunting at 4 wk after transplanting at the Pinney Purdue Agricultural Center (PPAC) in 2021. A) 'Liberty' zucchini and B) 'Blonde Beauty' yellow squash non-treated control (0 g ha⁻¹) vs. highest fomesafen rate (1120 g ha⁻¹).

Table 3.1. Summer squash injury with standard error (SE) at increasing fomesafen rates at the Meigs Horticulture Research Farm (MEIGS) and the Pinney Purdue Agricultural Center (PPAC) in 2020 and 2021 at 2 and 4 wk after transplanting (WAP) pooled across summer squash cultivars 'Blonde Beauty' yellow straightneck squash and 'Spineless Beauty' (2020) or 'Liberty' (2021) zucchini.

Rate	Summer squash injury ^a			
	2 WAP		4 WAP	
	MEIGS	PPAC	MEIGS	PPAC
g ai ha ⁻¹	-----%			
<i>2020</i>				
262	8 (1) a ^b	0	0	0 (0) a
524	16 (2) b	0	0	0 (0) a
786	18 (2) b	0	0	1 (1) a
1048	18 (2) b	0	0	14 (2) b
<i>2021</i>				
280	3 (1) a	5 (1) a	6 (3)	9 (2) a
560	9 (2) b	7 (1) a	16 (5)	9 (3) a
840	13 (2) bc	23 (5) b	17 (4)	24 (4) b
1120	19 (2) c	28 (3) b	15 (2)	31 (4) b

^aInjury were arcsin transformed for analysis and back-transformed for the table

^bMeans separation using Tukey's HSD test $P \leq 0.05$

We attributed the reduced summer squash injury at MEIGS in both years and at PPAC in 2020 to rainfall events before transplanting and during the growing season (Figure 3.3).). A rainfall event happened after spraying fomesafen but before transplanting the summer squash seedlings at PPAC 2020 and at MEIGS 2021, potentially moving some of the herbicide from the plastic to the row-middles. Injury at both location-years was likely a function of rainfall amount prior to transplanting. At PPAC 2020, there was minimal injury, probably because the total rain before transplanting (34 mm) removed most of the fomesafen off the plastic except for the highest rate. At MEIGS 2021, the rainfall was less than 9 mm. Although this rainfall likely removed some of the fomesafen residue from the plastic mulch, it did not remove as much as the 34 mm at PPAC 2020. Injury at 2 and 4 WAP was attributed to the residual herbicide splashing from the plastic mulch or from soil particles on the plastic mulch or near the soil surface onto the leaves close to the ground. At MEIGS 2020, it did not rain before transplanting, but the cumulative rain following transplanting was 10 mm over the next 2 wk. With so little rain the herbicide likely did not move down the soil profile into the crop's root zone, but it probably splashed from the plastic onto the leaves.

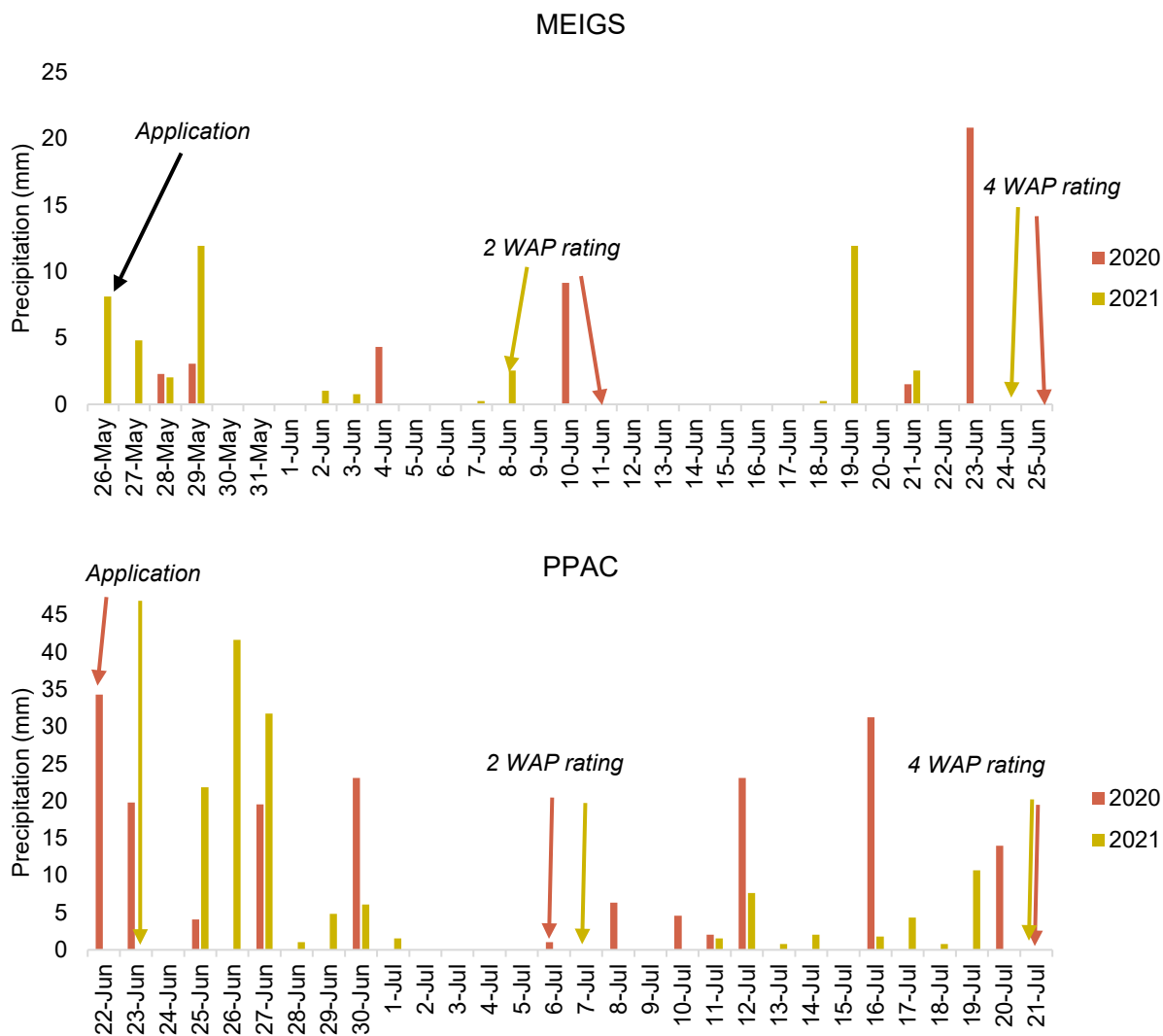


Figure 3.3. Precipitation at fomesafen application date and overtime and indication of summer squash injury rating dates at 2 and 4 wk after transplanting (WAP) at Meigs Horticulture Research Farm (MEIGS) and the Pinney Purdue Agricultural Center (PPAC). Summer squash transplanting was performed 1 d after application at all location-years.

Dissimilar to the other location-years, at PPAC in 2021, we saw increased injury, possibly because it did not rain before transplanting, and from 2 to 8 d after transplanting, it rained a total of 109 mm. Therefore, the herbicide was not washed off the plastic before transplanting and moved into the crop's root zone with the rain, increasing injury. In addition, at PPAC, we often saw the beds covered with soil (Figure 3.2), which was probably moved by the wind. Thus, because it rained regularly, it is likely that fomesafen splashed from the soil onto the leaves with the rain.

Similar to our results, Reed et al. (2018) reported 3% injury in hybrid 'Sunburst' yellow scallop squash (*C. pepo*), 2 wk after treatment (WAT) when using fomesafen at 420 g ha⁻¹ under

various plastic mulches. Peachey et al. (2012) reported that fomesafen at 280 g ha⁻¹ did not affect the emergence of direct-seeded 'Tigress' and 'Elite' zucchini and 'Yellow Crookneck' summer squash (*C. pepo*) and caused 0, 30, and 30% injury 2 WAT and 0, 33 and 16% injury 4 WAT, respectively. Reed et al. (2018) and Peachey et al. (2012) reported that injury was transient.

Yield

Yield data were analyzed separately by location-year because the effect of fomesafen rate was insignificant, except at PPAC 2021. Data were pooled across cultivars due to a lack of fomesafen-by-cultivar interaction at PPAC 2021. Fomesafen delayed harvest at PPAC in 2021. On the first ($F_{9,30} = 5.09$, $P = 0.0003$) and second ($F_{9,30} = 4.95$, $P = 0.0004$) harvests there was a significant fruit number decrease (Table 3.2). Harvestable fruits were only developed at the 0, 280, and 560 g ha⁻¹ rates on the first harvest. All rates differed from the non-treated control. The average fruit number of the non-treated control was 5 per six plants and only 2 per six plants for the 280 and 560 g ha⁻¹ rates. Harvestable fruits developed in all the treatments on the second harvest, where only the 840 and 1120 g ha⁻¹ rates differed from the non-treated control. The average fruit number of the non-treated control was 7 per six plants, and were 3 and 2 per six plants for the 840 and 1120 g ha⁻¹ rates, respectively. Accordingly, marketable yield loss was significant and fit to a three-parameter log-logistic model (Equation 3.2). The total marketable yield of the non-treated control at PPAC in 2021 averaged 20 kg per six plants. As the fomesafen rate increased from 280 to 1120 g ha⁻¹, the predicted marketable yield decreased from 95 to 60% compared to the non-treated control (Figure 3.4). Fomesafen did not significantly affect the marketable yield at the other location-years.

Table 3.2. Summer squash fruit number for the first two harvests with standard error (SE) at increasing fomesafen rates at the Pinney Purdue Agricultural Center in 2021 pooled across summer squash cultivars 'Blonde Beauty' yellow straightneck squash and 'Liberty' zucchini.

Rate	Fruit number	
	Harvest 1	Harvest 2
g ai ha ⁻¹	-----%-----	
0	5 (1) a ^a	8 (1) a
280	2 (1) b	5 (1) ab
560	2 (1) b	7 (1) a
840	0 (0) b	3 (1) b
1120	0 (0) b	2 (1) b

^aMeans separation using Tukey's HSD test $P \leq 0.05$

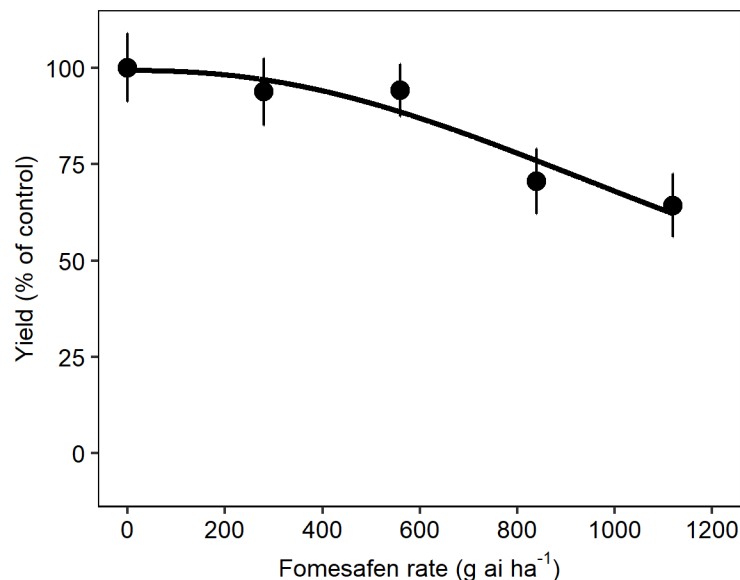


Figure 3.4. Effect of fomesafen rate on summer squash marketable yield as a percent of the non-treated control at the Pinney Purdue Agricultural Center in 2021, described with a three-parameter log-logistic model [$d/(1 + \text{Exp}[b(\log x - \log e)])$]. Parameters for $b = 2$, $d = 99$, and $e = 1402$; lack-of-fit $P=0.582$.

At MEIGS in 2020, there was a significant effect of cultivar across all treatments, where marketable yield averaged 13 and 18 kg per six plants for 'Blonde Beauty' and 'Spineless Beauty', respectively. Marketable yield pooled across cultivars and rates averaged 24 kg per six plants at PPAC in 2020 and 27 kg per six plants at MEIGS in 2021. Fomesafen rate did not increase the number of cull fruits (data not shown). Similar to our results, Peachey et al. (2012) and Reed et al. (2018) reported no significant summer squash yield loss when 280 g ha⁻¹ of fomesafen were

applied preemergence over-the-top bare ground, and 420 g ha⁻¹ pre-planting under plastic mulch, respectively.

3.4.2 Watermelon

Injury

Watermelon injury included bronzing (Figure 3.5) and stunting. Due to a lack of fomesafen rate-by-cultivar interaction, injury was analyzed across cultivars (Table 3.3). At 2 WAP, as the fomesafen rate increased from 210 to 840 g ha⁻¹, injury increased from 5 to 17% at MEIGS and 2 to 10% at SWPAC. At 4 WAP, injury ranged from 3 to 6%, but did not differ by fomesafen rate at SWPAC and increased from 2 to 13% at MEIGS. Injury at SWPAC decreased between 2 and 4 WAP, while injury at MEIGS did not decline between 2 and 4 WAP. At 6 WAP we did not see more injury. Overall, injury from the 210 g ha⁻¹ fomesafen rate was minimal ($\leq 5\%$) at 2 and 4 WAP.



Figure 3.5. Bronzing symptom on A) 'Exclamation' and B) 'Fascination' watermelon cultivars at a fomesafen rate of 560 g ai ha⁻¹, 2 wk after transplanting at the Southwest Purdue Agricultural Center in 2021.

Table 3.3. Watermelon injury with standard error (SE) at increasing fomesafen rates at the Meigs Horticulture Research Farm (MEIGS) and the Southwest Purdue Agricultural Center (SWPAC) in 2021 at 2 and 4 wk after transplanting (WAP) pooled across watermelon cultivars 'Exclamation' and 'Fascination'.

Rate	Watermelon injury ^a			
	2 WAP		4 WAP	
	SWPAC	MEIGS	SWPAC	MEIGS
g ai ha ⁻¹	-----%-----			
210	5 (1) a ^b	2 (1) a	3 (1)	2 (1) a
420	8 (1) ab	4 (1) b	4 (2)	5 (1) ab
630	11 (1) bc	7 (1) bc	6 (2)	8 (1) bc
840	17 (1) c	10 (1) c	5 (2)	13 (2) c

^aInjury data were arcsin transformed for analysis and back-transformed for the table.

^bMeans separation using Tukey's HSD test $P \leq 0.05$

Cumulative rain before transplanting was 5 mm at SWPAC and 27 mm at MEIGS. At SWPAC, the 6 d following transplanting, it did not rain; thus, the chances of the herbicide entering through the planting hole and reaching the crop's root zone was minimal. After that, it rained 53 mm over 6 d before the 2 WAP rating. At MEIGS, it rained 5 mm over 4 d before the 2 WAP injury rating and 95 mm over 5 d before the 4 WAP. We presume that rain caused fomesafen to splash from the plastic onto the leaves resulting in the injury symptoms observed.

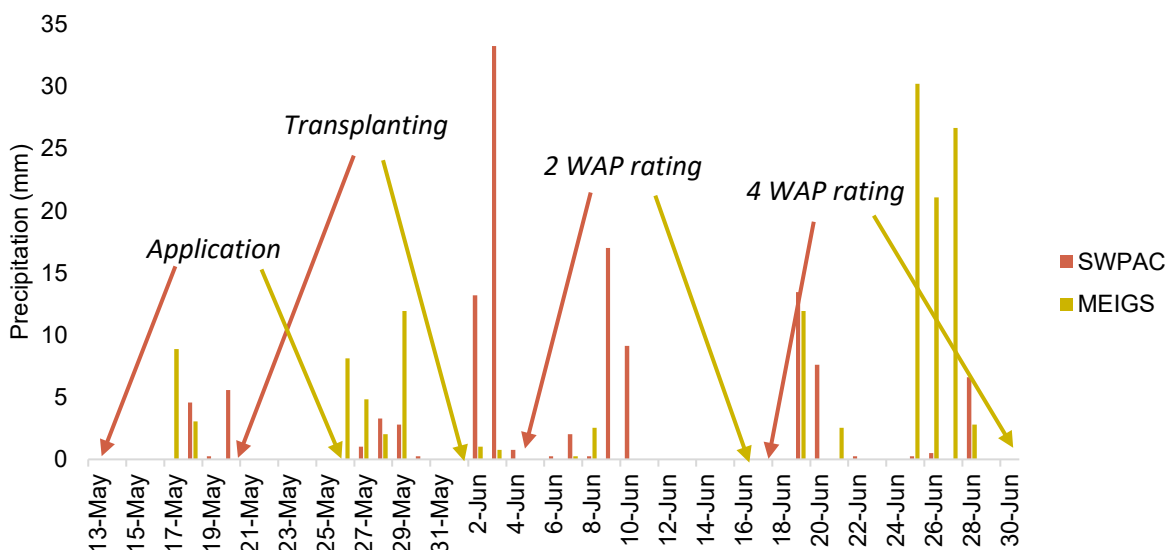


Figure 3.6. Precipitation between fomesafen application and watermelon transplanting dates, and overtime to indicate the date of watermelon injury ratings at 2 and 4 wk after transplanting (WAP) at the Southwest Purdue Agricultural Center (SWPAC) and Meigs Horticulture Research Farm (MEIGS).

Likewise, Johnson and Talbert (1993) reported 11% injury 3 wk after seeding watermelon into bare ground soils immediately or 1 wk after incorporating fomesafen at 280 g ha⁻¹. Bertucci et al. (2018) reported <2% injury symptoms at 3 WAT when 175 g ha⁻¹ of fomesafen were applied under the plastic 1 d before transplanting triploid watermelon.

Weed Control

Due to a significant difference across locations, watermelon weed control data was analyzed by location. At 4 WAP, as the fomesafen rate increased from 210 to 840 g ha⁻¹, weed control on the watermelon trials increased from 76 to 91% at SWPAC and 96 to 100% at MEIGS (Table 3.4) relative to the 0 g ha⁻¹ fomesafen rate treatment that only received *S*-metolachlor at SWPAC or a mix of halosulfuron, ethalfluralin and clomazone at MEIGS. At SWPAC, fomesafen fully controlled carpetweed (*Mollugo verticillata* L.) and morningglory spp. (*Ipomoea* spp. L.) and partially controlled common lambsquarters (*Chenopodium album* L.), pigweeds (*Amaranthus* spp. L.), dandelion (*Taraxacum officinale* F. H. Wigg.). At MEIGS, fomesafen controlled carpetweed, common purslane (*Portulaca oleracea* L), Eastern black nightshade (*Solanum ptychanthum* Dunal), giant ragweed (*Ambrosia trifida* L.), morningglory spp., velvetleaf (*Abutilon theophrasti* Medik.), and grass species. The increased weed control at MEIGS was most likely because we sprayed four herbicide groups (Groups 2, 3, 13, and 14) rather than only two (Groups 14 and 15) like at SWPAC. This demonstrates the importance of soil residual herbicide mixtures, which aid in delaying herbicide resistance (Beckie and Reboud 2009; Busi et al. 2020)

Table 3.4. Weed control with standard error (SE) at increasing fomesafen rates at the Meigs Horticulture Research Farm (MEIGS) and the Southwest Purdue Agricultural Center (SWPAC) in 2021 at 4 wk after transplanting pooled across watermelon cultivars 'Exclamation' and 'Fascination'.

Rate	Weed control ^a	
	SWPAC	MEIGS
g ai ha ⁻¹	-----%	-----
210	76 (5) b ^b	96 (3)
420	86 (4) ab	98 (2)
630	91 (1) a	100 (0)
840	91 (2) a	100 (0)

^aWeed control data were arcsin transformed for analysis and back-transformed for the table.

^bMeans separation using Tukey's HSD test $P \leq 0.05$

Yield

Watermelon yield was not significantly affected by any fomesafen rate. Yield averaged 258 kg 27 m⁻² at MEIGS and 166 kg 27 m⁻² at SWPAC, and fruit number averaged 42 and 27, respectively. Bertucci et al. (2018), who applied fomesafen under the plastic 1 d before transplanting at 175 g ha⁻¹, reported no triploid watermelon yield or fruit number losses.

Although the studies compared differ from ours regarding the herbicide application (over-the-top of bare ground and incorporated vs. over-the-top of plastic) and planting (seeds vs. seedlings); the results reported by others support our results as we saw only minor damage in summer squash and watermelon at the lowest rates we used, and injury was transient. We presume that plasticulture may reduce the risk of injury due to less direct contact of the herbicide with the crops' roots and leaves if rain washes off the herbicide from the plastic to the middle-rows.

Currently, there is no evidence quantifying fomesafen dissipation from plastic over time. Other herbicides wholly wash off the plastic with rain, such as 2,4-D, glyphosate, and paraquat (Culpepper et al. 2009; Grey et al. 2009; Hand et al. 2021), or bind to the plastic but wash off over time, such as flumioxazin and halosulfuron (Grey et al. 2009, 2018; Randell et al. 2019), or irreversibly bind to the plastic, like carfentrazone (Culpepper et al. 2009; Grey et al. 2009). We hypothesize that fomesafen washes-off plastic rapidly, but more water must be necessary as the herbicide concentration increases. We assumed this because the fomesafen molecule used is a sodium salt, which is highly soluble in water (600,000 mg/L at 25°C) (Shaner 2014), explaining its movement with rainwater. Experiments to determine the behavior of fomesafen on plastic and

other mulches are recommended. Moreover, fomesafen could have also dissipated from the plastic due to photodecomposition. Fomesafen decomposes rapidly under relatively low sunlight conditions (Shaner 2014).

3.5 Conclusions

Fomesafen caused necrosis, chlorosis, brown to white spots and stunting on summer squash, and bronzing and stunting on watermelon. Fomesafen rates from 280 to 1120 g ha⁻¹ delayed harvest and decreased marketable yield from 95 to 60% of the non-treated control at PPAC in 2021. Fomesafen did not cause marketable yield loss at any of the other summer squash trials and the watermelon trials. Presumably, the rain before transplanting washed off the herbicide from the plastic, reducing the risk of the herbicide reaching the crops' root zone after transplanting. At PPAC in 2021, it did not rain before transplanting and 1 d after transplanting it rained a total of 109 mm over the next 7 d, increasing the movement of fomesafen into the planting hole.

Overall, crop safety was excellent for fomesafen broadcasted over-the-top of the plastic 1 d before transplanting summer squash at 262 and 280 g ha⁻¹ and 6 to 7 d before transplanting triploid watermelon at a rate of 210 g ha⁻¹ at MEIGS, PPAC and SWPAC. Fomesafen applied at these rates caused minimal injury, and the crops recovered over time. Also, these rates did not significantly affect summer squash or triploid watermelon yield and increased weed control. Rainfall before transplanting may be necessary to wash off the herbicide from the plastic mulch to reduce the risk of the herbicide entering through the planting hole and reaching the crops' root zone with excessive rain.

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