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Peach tree response to low dosages of dicamba as repeated application or with various spray nozzles

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Abstract

Two low-dose dicamba exposure trials were conducted on container-grown peach trees in Fayetteville, AR. Peach trees were 'July Prince' scions grafted onto 'Guardian' rootstock, were transplanted into 19-L containers, and received experimental dicamba treatments in each year. Container trials were initiated in 2020 and repeated on new trees in 2021. In the repeated application trial, dicamba was applied at 5.6 g as ha^{-1} (1/100X field rate) in five sequences: an untreated control receiving no herbicide, one treatment receiving only an initial application, and three treatments receiving an initial application plus sequential applications at the same rate occurring at 14 d, 28 d, and 14 d + 28 d after initial treatment (DAT). A separate trial assessed peach tree responses to dicamba applied at 11.2 g at ha^{-1} (1/50X field rate) using a selection of nozzles with differing droplet spectrum characteristics: Turbo TeeJet® induction nozzle TTI11002, air induction turbo TwinJet* nozzle AITTJ60-11002, air induction extendedrange (XR) TeeJet® nozzle AIXR11002, XR TeeJet® flat-fan nozzle XR11002, and XR TeeJet® flatfan nozzle XR1100067. Peach tree height, tree cross-sectional area, and leaf chlorophyll content were not reduced in response to any sequence of dicamba application or nozzle selection. Repeated applications of dicamba at a 1/100X rate did not increase peach injury after 28 DAT. By 84 DAT, no effect of nozzle type on peach tree injury was discernable, and all treatments caused below 4% injury. No dicamba or dicamba metabolites were observed in leaf samples collected at 14, 69, or 85 DAT from trees treated with XR1100067 or in untreated controls. While peach tree injury was observed throughout the experiment, dicamba residues were detected consistently only in 2020 from leaf samples of trees treated with dicamba at a 1/50X rate using TTI1102, AITTJ60-11002, AIXR11002, and XR11002 nozzles.

Introduction

The potential for dicamba exposure on crops has been well characterized, whether through herbicide drift, volatilization, or sprayer contamination (Behrens and Lueschen 1979; Egan et al. 2014; Inman et al. 2021). Specialty crops are especially impacted by off-target movement of dicamba due to their high value and the potential for dicamba residues to make crops like tobacco (Nicotiana tabacum L.) and tomato (Solanum lycopersicum L.) unmarketable (Inman et al. 2021; Meyers et al. 2022). The effects of dicamba exposure have been assessed extensively in annual specialty crop systems, including crops of the Brassicaceae, Convolvulaceae, Cucurbitaceae, Fabaceae, Ipomoaceae, and Solanaceae families (Culpepper et al. 2018; Hand et al. 2020; Inman et al. 2021; Mohseni-Moghadam and Doohan 2015; Shankle et al. 2021; Wasacz et al. 2022a). A consistent finding across crops is that exposure to reduced rates of dicamba causes visible crop injury, which may not impact yield. Similarly, the sensitivity of fruit trees and vines to synthetic auxins has been well documented (Bondada 2011). Several ornamental, fruit, and nut plant species exhibited sensitivity to driftable rates of dicamba in container studies (Dintelmann et al. 2020; Mohseni-Moghadam et al. 2016). Field studies of dicamba exposure in mature perennial fruit trees and vines have demonstrated crop injury to pecans [Carya illinoinensis (Wangenh.) K. Koch] and grapes (Vitis vinifera L.) (Bondada 2011; Dixon et al. 2021; Wells et al. 2019); however, as with annual cropping systems, visible crop injury symptoms did not always translate to yield reduction. Still, concern exists regarding residues in harvested portions of the plant and long-term effects on perennial systems.

Peach orchards may not generate revenue until 3 yr after orchard establishment (Singerman et al. 2021). Given the capital investment, maintenance costs, and delay in revenue generation, it may take 7 or 8 yr of production for a peach orchard to break even or become profitable



(Knudsen et al. 2016). Thus any crop injury or yield reduction due to dicamba exposure could threaten years of potential fruit production and disrupt the profitability of decades-long business investments. A further concern is the nature of potential dicamba exposures in sensitive specialty crops. Much of the dicamba exposure research has assessed the sensitivity of crop species to a single simulated particle drift event (Dintelmann et al. 2020; Shankle et al. 2021; Wasacz et al. 2022b). Some work has investigated simulated drift at various crop growth stages (Dittmar et al. 2016; Wasacz et al. 2022a), exposure from irrigation water (Willett et al. 2021), and exposure from residues on polyethylene mulch (Hand et al. 2021). However, little work assesses the effect of repeated exposure of specialty crops to dicamba.

There is cause for concern that the off-target movement of dicamba could negatively impact perennial fruit systems. This experiment was conducted to assess first-year peach tree growth and injury symptoms and to detect dicamba residues in response to (1) repeated exposures of driftable rates of dicamba and (2) differing droplet spectrums of dicamba applied with a selection of nozzle types.

Materials and Methods

Experiments were conducted at the Milo J. Shult Research and Extension Center in Fayetteville, AR (36.0987°N, 94.180°W), in 2020 and 2021. First-year, bare-root peach trees were ordered from a commercial nursery and stored at 4 C until transplanted into containers in the field. Peach trees were 'July Prince' scions grafted onto 'Guardian' rootstock and were transplanted into 19-L containers that were filled with a 2:1 (v:v) mixture of sand (<1% organic matter) to potting mix (Pro-Mix M Mychorrhizae, Premier Tech Horticulture, Quakertown, PA, USA). Container trials were conducted on a low-maintenance turf site with full sun, and landscape fabric was installed above the turf to prevent turf or weed encroachment around the experimental trees. Containers were irrigated via a drip irrigation system placed on shipping pallets to facilitate drainage. Fertility was added to each container individually by hand using a fixed volume (568 mL) of prepared solution of 3.0 g L⁻¹ of a 24-8-16 (N-P₂O₅-K₂O) soluble fertilizer (Sta-Green, Parker Fertilizer Company, Dallas, TX, USA) in water, based on recommendations for commercial production of young peach trees (Taylor 2012). According to recommendations for peaches per standard commercial practices, diseases and insects were prevented using registered products, when necessary (Blaauw et al. 2023).

Peach trees were transplanted into containers, ensuring graft unions were 6 to 10 cm above the substrate line, then placed in field sites on March 20, 2020, and March 30, 2021. The central leader of each tree was pruned to a height of approximately 1 m following transplanting, and branches were thinned to ensure that three to five healthy scaffold branches would develop over the growing season (Taylor 2012). Transplanting dates were selected to initiate trials as early in the season as possible without exposing trees to harmful freeze events. However, on April 21, 2021, a late-season freeze required all containers to be moved indoors overnight to prevent freeze injury to newly opening buds. Plants were returned to the field site within 36 h, and no symptoms of cold injury were observed as leaves expanded.

This work included two dicamba exposure trials: a repeated application trial with dicamba applied multiple times throughout the season and a trial in which dicamba was applied once using nozzles with different droplet spectrums. Dicamba applications used XtendiMax[®] with VaporGrip[®] (Bayer Crop Science, St. Louis, MO, USA) and were conducted approximately 1 km from the primary trial site in an isolated pasture. Containers were transported to the isolation site, sprayed with each respective dicamba treatment, and left in isolation for a minimum of 7 d if a rain event occurred or 10 d if no rain occurred. Following isolation, treated plants were moved back to the primary trial site. Treated plants were watered by hand at the same rate as plants left in the main trial area.

Within each trial, two peach trees in separate 19-L containers served as the experimental unit. Two containers of the same experimental unit were spaced 46 cm apart and a minimum of 51 cm from trees of neighboring experimental units. Peach containers were organized on pallets to ensure proper drainage from containers, and two experimental units (i.e., four containers) were organized onto each pallet. Pallets were arranged in 18-m rows on 3-m centers with 3-m in-row spacing. Each trial was fully contained within two rows of pallets. Figure 1 is an overhead image of the field site.

Repeated Application Trial

Dicamba treatments were applied in the repeated application trial with a CO₂ backpack sprayer fitted with four flat-fan 1100067 nozzles at a 51-cm spacing and calibrated to deliver 47 L ha⁻¹ at 276 kPa. During the dicamba application, peach trees were arranged in two rows 71 cm apart with 101 cm between containers within each row, and the boom was maintained 51 cm above the top of the peach tree canopy. Peach trees were treated with dicamba at 5.6 g ae ha⁻¹ (1/100X maximum registered field rate for soybean [*Glycine max* (L.) Merr.]; 1X = 560 g ae ha⁻¹) at each application after peach leaves had fully opened. The 1/100X rate represented a low but realistic rate of dicamba that may occur in field applications (Butts et al. 2022; Sousa Alves et al. 2017).

Treatment levels included an untreated control receiving no herbicide, one treatment receiving only the initial application, and three treatments receiving the initial application plus sequential applications at the same rate occurring 14 d, 28 d, and 14 d + 28 d after initial treatment (DAT), respectively. New peach trees were used in each year of this trial, and initial applications occurred on May 29, 2020, and June 14, 2021, respectively.

Nozzle Selection Study

For the varying droplet size and spray volume experiment, peaches were treated with dicamba at 11.2 g ae ha^{-1} (1/50X field rate) after peach leaves had fully opened. Herbicide applications occurred on May 29, 2020, and June 14, 2021, respectively. Droplet sizes were determined by nozzle type, which included Turbo TeeJet* induction (TTI11002), air induction turbo TwinJet* (AITTJ60-11002), TeeJet* air induction extended-range (AIXR11002), and TeeJet* extended-range flat-fan (XR11002) nozzles calibrated at 187 L ha^{-1} at 276 kPa and a TeeJet* extended-range flat-fan (XR1100067) nozzle calibrated at 47 L ha^{-1} at 276 kPa (Table 1).

Data Collection

For both trials, data were collected to characterize peach tree growth, peach tree injury, and dicamba and metabolite residues within peach leaves. Growth characteristics were assessed by measuring plant heights, tree cross-sectional area (TCSA), leaf chlorophyll content, and visible ratings of plant vigor. Plant heights

Table 1.	Sprayer	settings and	corresponding of	droplet	characteristics	for d	icamba	exposure stu	dies. ^{a,b,c}
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					Droplet size ^d		Velocity			
Nozzle type ^g	Spray volume	Dicamba rate	Spray classification ^e	Dv10	Dv50	Dv90	Driftable fines ^f	Max.	Average	
	L ha ⁻¹	g ae ha ⁻¹			μm		%	—— m	s ⁻¹	
TTI11002	187	11.2	extremely coarse	342 a	676 a	1,060 a	2.5	6.72 cd	2.10 b	
AITTJ60-11002	187	11.2	coarse	333 a	427 b	580 b	0.9	5.63 d	2.03 b	
AIXR11002	187	11.2	coarse	242 b	376 c	579 b	7.3	8.80 b	2.46 a	
XR11002	187	11.2	medium	166 c	221 d	317 c	31.7	10.44 a	1.97 b	
XR1100067	47	5.6	fine	92 d	159 e	224 d	79.1	7.32 c	1.21 c	
XR1100067	47	11.2	fine	92 d	158 e	224 d	79.1	7.20 c	1.20 c	
P-value				< 0.0001	< 0.0001	< 0.0001	_	0.0003	< 0.0001	

^aDroplets were characterized in a laboratory experiment in Lonoke, AR, using a track sprayer system and a VisiSize P15 Portable Particle/Droplet Image Analysis System. Each value represents the least squares mean output of three replications, each comprising a 2,500-droplet observation.

^bAbbreviations: AITTJ, air induction turbo TwinJet® nozzle; AIXR, air induction extended-range nozzle; TTI, Turbo TeeJet® induction nozzle; XR, extended-range nozzle.

Wears were separated using Tukey's honestly significant difference at a $\alpha = 0.05$ significance level, and means followed by the same letter are not significantly different. Means lacking letters indicate no significant treatment effect at the $\alpha = 0.05$ significance level.

^dDv10, Dv50, and Dv90 refer to the droplet diameter where 10%, 50%, and 90% of spray volume consisted of smaller diameters, respectively.

^eDetermined using ASABE S572.1.

^fDefined as the percentage of spray volume with droplet diameters < 200 μ m. Values were predicted from a Rosin–Rammler model $V(d) = 100 - 100 \times \exp(-(d/c)^m)$, where V is the cumulative percent volume of droplets with diameter lower than a certain value (d); c is the characteristic droplet diameter, defined as the diameter at which the cumulative volume fraction is 63.2%; and m is a constant indicating the uniformity of the distribution. Driftable fines are modeled from all observations combined across replications and are not suitable to analysis of variance.

^gSprayer operated at 276 kPa for all nozzle types.



Figure 1. Container-grown peach trees at the experiment site at the Milo J. Shult Research and Extension Center in Fayetteville, AR, in 2020.

and TCSA were recorded for each tree at 0 and 84 DAT. Plant heights were measured from the container substrate level to the highest growing point. Trunk diameter was recorded as the average of two perpendicular measurements 6.4 cm above the graft union using an electronic digital caliper (CID Bio-Science, Camas, WA, USA). Peach TCSA is a calculated response based on the formula for the area of a circle:

$$\text{TCSA} = \left[\left(\frac{d_1}{2} + \frac{d_2}{2} \right) / 2 \right]^2 \times \pi$$
 [1]

where d_1 and d_2 are the perpendicular trunk diameter measurements from each tree. Leaf chlorophyll content was recorded at 0, 28, 56, and 84 DAT on two representative leaves per tree using a handheld soil plant analysis development (SPAD) chlorphyll (SPAD-502Plus, Konica Minolta, Ramsey, NE, USA) and taking care that the receptor was centered on tissue outside of the leaf midvein. Plant vigor was assessed visually on a 0 to 9 scale (where 0 = total plant death and defoliation and 9 = a completely healthy tree with no indicators of stress or injury). Peach tree injury ratings were recorded on a 0 to 100 scale (where 0 = no injury and 100 = total plant death). Plant vigor and peach tree injury ratings occurred at 14, 28, 56, and 84 DAT.

Droplet size and velocity for all nozzles and dicamba rates were determined in a laboratory experiment conducted at the Lonoke Extension Center near Lonoke, AR. Herbicide solutions were prepared at their respective rates, and an operating pressure of 276 kPa was used in a CO₂-powered track spray chamber outfitted with a VisiSize P15 Portable Particle/Droplet Image Analysis System (Oxford Lasers, Imaging Division, Oxford, UK) as described in Kouame et al. (2023). An imaging system characterized the droplet size and velocity 51 cm from the nozzle tip to correspond with the boom height above peach canopies in field trials. Each replication comprised a 2,500-droplet sample. Three observations were recorded for each nozzle type, spray volume, and dicamba concentration treatment combination for a total of 7,500 individual droplets measured. Droplet velocities were characterized by the maximum and average recorded during each observation. Droplet sizes were characterized by Dv10, Dv50, and Dv90, which refer to the droplet diameter where 10%, 50%, and 90% of spray volume consisted of smaller diameters, respectively. The percentage of spray droplets 200 µm in diameter and smaller was determined using the Rosin-Rammler equation

$$V(d) = 100 - 100 \times \exp\left[-\left(\frac{d}{c}\right)^{\mathrm{m}}\right]$$
 [2]

where V is the cumulative percent volume of droplets with diameter lower than a certain value (d); c is the characteristic droplet diameter, defined as the diameter at which the cumulative volume fraction is 63.2%; and m is a constant indicating the uniformity of the distribution.

Dicamba residue analysis was conducted on detached leaves and submitted to the Mississippi State Chemical Lab at Mississippi State University for residue plant tissue analysis for dicamba, 5-hydroxy dicamba, and 3,6-dichlorosalicylic acid (DCSA). Fifteen leaves were removed by hand from each peach tree of each experimental unit, using a separate pair of nitrile gloves for every tree. Leaf samples were collected from 3 to 4 fully expanded leaves proximal from the growing point on 4 to 5 different peach scaffold branches until 15 leaves had been collected. Collected leaves from both trees in an experimental unit were bulked into a single 30-leaf sample and stored in a 0 C freezer before overnight shipping to the Mississippi State Chemical Lab for analysis. Initial leaf sampling occurred on June 12, 2020, and June 28, 2021, which coincided with a 14 DAT sampling date for each year. In the repeated application trial when dicamba exposures had occurred as late as 28 DAT, a second leaf sampling was collected each year, on August 6, 2020, and September 7, 2021, coinciding with a 69 DAT and 85 DAT sampling, respectively. Residue detection was conducted with liquid chromatography, using the Agilent Zorbax Eclipse XDB-C18 column (Agilent Technologies, Santa Clara, CA, USA) to separate the tested compounds and residues.

Statistical Analysis

The repeated application trial was a one-factor experiment with application timing as the main effect. The experiment was arranged in a randomized complete block design with four replications, repeated over 2 yr. Application timing was treated as a fixed effect, whereas year and block (nested within year) were treated as random effects. The nozzle selection trial was a onefactor experiment with nozzle type as the main effect. The experiment was arranged in a randomized complete block design with four replications, repeated over 2 yr. The main effect of nozzle type was treated as a fixed effect, whereas year and block (nested within year) were treated as random effects. Plant responses and dicamba residue testing were analyzed separately for each trial using the GLIMMIX procedure in SAS version 9.4 (SAS Institute Inc., Cary, NC, USA) to conduct analysis of variance (ANOVA) with a significance level of $\alpha = 0.05$. Tukey's honestly significant difference (HSD) was used as a post hoc multiple comparisons adjustment for all means separation.

Droplet velocities and sizes were analyzed across all nozzle types, carrier volumes, and dicamba rates from each trial. The droplet data were analyzed as a single-factor experiment with the combined sprayer settings as the main factor. Sprayer settings included nozzle type, spray volume, and dicamba rate for seven sprayer settings (Table 1). The sprayer setting was treated as a fixed effect with three 2,500-droplet replications arranged in a completely randomized design. Droplet velocities and sizes were analyzed using PROC GLIMMIX to conduct ANOVA with a significance level of $\alpha = 0.05$. Tukey's HSD was used as a post hoc multiple comparisons adjustment for all means separation. The percentage of driftable fines is not suitable to ANOVA and means separation because the modeled values from Equation 2 use all observations for each treatment to generate one predicted value. The predicted values are presented and discussed without further statistical testing.

Results and Discussion

Droplet Spectrum Analysis

Image analysis and characterization of droplet spectrums at each sprayer setting revealed discrepancies in droplet size and droplet velocity. The Dv10, Dv50, and Dv90 consistently identified the TTI11002 nozzle as producing the largest droplet diameter. The TTI11002 nozzle had a Dv50 of 676 μ m, classified as extremely coarse spray according to American Society of Agricultural and Biological Engineers (ASABE 2009) standards. The AITTJ60-11002 and AIXR11002 nozzles produced a Dv50 of 427 and 376 μ m, respectively, and both were classified as having a coarse spray according to ASABE standards. The XR11002 nozzles had a Dv50

		lorophyll ntent		TCSA			Height	
Herbicide timing	0 DAT	84 DAT	0 DAT	84 DAT	Δ	0 DAT	84 DAT	Δ
	SF	PAD		mm ²			cm	
0 DAT	35.5	42.5	122	254	131	124	149 a	24.8
0 DAT fb 14 DAT	34.2	40.0	132	254	122	125	139 b	14.2
0 DAT fb 14 DAT fb 28 DAT	34.1	39.9	131	270	139	123	142 ab	19.5
0 DAT fb 28 DAT	36.0	43.0	128	253	125	123	143 ab	19.6
Untreated	34.4	43.3	134	263	130	125	150 a	25.4
P-value	0.5040	0.0916	0.6834	0.6846	0.7725	0.9894	0.0363	0.1077

Table 2. Effect of individual and repeated applications of a 1/100X rate of dicamba at selected days after treatment on growth characteristics of container-grown peach trees at the Milo J. Shult Research and Extension Center in Fayetteville, AR, in 2020 and 2021.^{a,b,c}

^aDicamba at a 1/100X rate was equivalent to 5.6 g ae ha⁻¹. Initial applications (0 DAT) occurred when peach leaves had fully expanded, on May 29, 2020, and June 14, 2021. Δ represents the change in a measured value throughout the experiment and was calculated by subtracting the initial measurement (0 DAT) from the final measurement (84 DAT).

^bAbbreviations: DAT, days after initial treatment; fb, followed by; SPAD, soil plant analysis development value, an indirect measure of chlorophyll content; TCSA, tree cross-sectional area. ^cMeans were separated using Tukey's honestly significant difference at a α = 0.05 significance level, and means followed by the same letter are not significantly different. Means lacking letters indicate no significant treatment effect at the α = 0.05 significance level.

of 221 μ m, which was only 31% of the diameter of the Dv50 of the TTI11002 nozzle and was classified as a medium spray (ASABE 2009). Across both dicamba rates, the XR1100067 nozzle produced a spray classified as fine (ASABE 2009) and only 24% of the diameter of the Dv50 recorded for the TTI11002 nozzle. No differences in droplet diameter were detected between the XR1100067 nozzles when spraying dicamba at 5.6 and 11.2 g ae ha⁻¹, corresponding to the 1/100X and 1/50X rate, respectively. The present findings that the AITTJ, AIXR, and TTI nozzles produced the largest droplet diameters are consistent with previous research and are attributed to the air induction and preorifice technology associated with those nozzles (Creech et al. 2015).

The percentage of driftable fines (droplet diameters 200 μ m and smaller) for each nozzle setting was modeled using Equation 2 (Table 1). Overall, nozzle treatments that produced a spray with larger droplet diameters were observed to have lower percentages of driftable fines. XR1100067 nozzles produced the largest fraction of driftable fines, comprising 79.1% of the total spray volume, regardless of dicamba rate. For the XR11002 nozzle, which had a medium droplet spectrum (ASABE 2009), 31.7% of the total spray volume comprised driftable fines (Table 1). Nozzles classified as producing coarse or extremely coarse (ASABE 2009) droplet spectrums in this experiment exhibited the smallest percentages of driftable fines. For AIXR11002, TTI11002, and AITTJ60-11002 nozzles, driftable fines comprised 7.3%, 2.5%, and 0.9% of the total spray volume, respectively (Table 1).

Spray droplet velocities also differed by sprayer settings. The average velocity of droplets produced by the AIXR11002 nozzle was 2.46 m s⁻¹, which was the fastest observed at the 51-cm distance (Table 1). The next grouping included the TTI11002, AITTJ60-11002, and XR11002 nozzles, which produced average droplet velocities of 2.10, 2.03, and 1.97 m s⁻¹, respectively. The droplets from XR1100067 nozzles produced an average droplet velocity of 1.21 and 1.20 m s⁻¹ for the 1/100X and 1/50X dicamba rate, respectively. Thus the XR1100067 nozzle produced similar average velocities, regardless of dicamba rate. The XR1100067 nozzle exhibited the slowest average droplet velocity and was <50% of the average velocity of droplets produced by the AIXR11002 nozzle. The present characterizations are consistent with previous research that nozzle selection and operating pressure have the greatest effects on droplet diameter and velocity (Creech et al. 2015). Although the central question of this experiment was not focused on droplet spectrum analysis, characterization of spray patterns can help with predicting potential risks of off-target movement, particularly the fraction of driftable fines, and the present work can characterize the response of peach trees to exposure to dicamba for each of the droplet spectrums.

Peach Growth Responses

Peach tree growth responses were minimally affected by dicamba treatments in the repeated application and nozzle selection studies. In the repeated application study, leaf chlorophyll content at 84 DAT was unaffected by any exposure sequence (Table 2). No differences were observed in SPAD throughout the trial, including ratings at 28 and 56 DAT (data not shown). At 84 DAT, trees treated with dicamba on day 0 followed by (fb) 14 DAT were shorter than untreated trees and trees treated with dicamba on day 0 (Table 2). However, when corrected for initial plant heights, there were no differences in the change in plant heights throughout the study, from 0 to 84 DAT. Similarly, the calculated TCSA values and changes in TCSA were unaffected by any sequence of dicamba exposure.

In the nozzle selection study, leaf chlorophyll content did not change in response to nozzle selection (Table 3). Relative to the untreated trees, TCSA was reduced in plots treated with dicamba from AITTJ60-11002, AIXR11002, and XR1100067 nozzles at 84 DAT. When accounting for the change in TCSA over the 84-d trial, trees treated with dicamba from AITTJ60-11002 and AIXR11002 nozzles had the smallest increases in TCSA (Table 3). The apparent reduction in TCSA from treatment with the XR1100067 nozzle at the 84 DAT measurement was no longer observed when accounting for initial tree size (Table 3). Finally, plant heights at 84 DAT indicated a reduction in height when treated with dicamba using AITJ60-11002, TTI11002, or XR11002 nozzles; however, a lack of significance of a change in plant height from 0 to 84 DAT indicates no consequential effect on peach tree height in response to nozzle selection (Table 3).

Peach Tree Injury

In both trials, peach tree injury symptoms were characterized by varying degrees of leaf curling, leaf elongation, and deformation of the leaf surface on new leaves of peach trees. In the repeated application trial, peach tree injury varied throughout the growing season. This makes sense as new exposures to the 1/100X rate of dicamba (5.6 g ae ha⁻¹) occurred within the season, depending on

	Leaf chlorophyll content			TCSA			Height		
Nozzle type	0 DAT	84 DAT	0 DAT	84 DAT	Δ	0 DAT	84 DAT	Δ	
	SF	PAD		mm ²		. <u></u>	cm		
TTI11002	35.1	41.5	124	269 ab	145 a	129	147 bc	18.1	
AITTJ60-11002	35.7	42.1	115	232 c	117 c	124	146 bc	22.3	
AIXR11002	36.1	42.5	126	249 bc	123 bc	127	149 ab	21.8	
XR11002	33.8	43.7	126	263 ab	137 ab	119	140 c	20.5	
XR1100067	33.5	41.7	116	253 bc	137 ab	126	148 a-c	21.8	
Untreated	35.0	43.2	129	279 a	150 a	125	155 a	29.4	
P-value	0.2642	0.6006	0.2297	0.0020	0.0072	0.1111	0.0248	0.0797	

Table 3. Effect of nozzle selection on growth characteristics of container-grown peach trees exposed to dicamba at a 1/50X rate at selected days after treatment at the Milo J. Shult Research and Extension Center in Fayetteville, AR, in 2020 and 2021.^{a,b,c}

^aDicamba at a 1/50X rate was equivalent to 11.2 g ae ha⁻¹. Herbicide applications (0 DAT) occurred when peach leaves had fully expanded, on May 29, 2020, and June 14, 2021, respectively. ^bAbbreviations: AITTJ, air induction turbo TwinJet® nozzle; AIXR, air induction extended-range nozzle; DAT, days after initial treatment; fb, followed by; SPAD, soil plant analysis development value, an indirect measure of chlorophyll content; TCSA, tree cross-sectional area; TTI, Turbo TeeJet® induction nozzle; XR, extended-range nozzle.

^cMeans were separated using Tukey's honestly significant difference at a α = 0.05 significance level, and means followed by the same letter are not significantly different. Means lacking letters indicate no significant treatment effect at the α = 0.05 significance level.

Table 4. Injury response of container-grown peach trees treated with a 1/100X rate of dicamba as single or repeated applications at selected days after initial treatment.^{a,b,c}

Herbicide timing	14 DAT	28 DAT	56 DAT	84 DAT
		%	ó ————	
0 DAT	5.4	6.0 bc	4.3	1.7
0 DAT fb 14 DAT	5.8	8.1 ab	6.7	1.4
0 DAT fb 14 DAT fb 28 DAT	5.9	10.3 a	7.4	1.9
0 DAT fb 28 DAT	4.1	5.0 c	7.6	1.1
P-value	0.1447	0.0069	0.1975	0.7395

^aDicamba at a 1/100X rate was equivalent to 5.6 g ae ha⁻¹. Peach tree injury was visually assessed on a 0 to 100 scale (where 0 = no injury and 100 = total plant death) at 14, 28, 56, and 84 DAT. Experiments were conducted at the Milo J. Shult Research and Extension Center in Fayetteville, AR, in 2020 and 2021. Initial herbicide applications occurred when peach leaves had fully expanded on May 29, 2020, and June 14, 2021, respectively.

^bAbbreviations: DAT, days after initial treatment; fb, followed by.

^cMeans were separated using Tukey's honestly significant difference at a $\alpha = 0.05$ significance level, and means followed by the same letter are not significantly different. Means lacking letters indicate no significant treatment effect at the $\alpha = 0.05$ significance level.

Table 5. Injury response of container-grown peach trees treated with a 1/50X rate of dicamba applied with a selection of nozzles.^{a,b,c}

Nozzle type	14 DAT	28 DAT	56 DAT	84 DAT
		%	ó	
TTI11002	15.3 a	17.8 a	13.1 a	3.1
AITTJ60-11002	14.1 a	14.7 a	11.4 a	1.9
AIXR11002	16.9 a	16.6 a	13.5 a	3.3
XR11002	18.1 a	18.4 a	10.9 a	3.0
XR1100067	5.6 b	4.1 b	5.6 b	1.3
P-value	0.0017	<0.0001	0.0081	0.2468

^aDicamba at a 1/50X rate was equivalent to 11.2 g ae ha⁻¹. Peach tree injury was visually assessed on a 0 to 100 scale (where 0 = no injury and 100 = total plant death) at 14, 28, 56, and 84 DAT. Experiments were conducted at the Milo J. Shult Research and Extension Center in Fayetteville, AR, in 2020 and 2021. Herbicide applications occurred when peach leaves had fully expanded, on May 29, 2020, and June 14, 2021, respectively.

^bAbbreviations: AITTJ, air induction turbo TwinJet[®] nozzle; AIXR, air induction extendedrange nozzle; DAT, days after initial treatment; TTI, Turbo TeeJet[®] induction nozzle; XR, extended-range nozzle.

^cMeans were separated using Tukey's honestly significant difference at a $\alpha = 0.05$ significance level, and means followed by the same letter are not significantly different. Means lacking letters indicate no significant treatment effect at the $\alpha = 0.05$ significance level.

the treatment. Throughout the repeated application trial, no peach tree injury exceeded 11% (Table 4). At the 14 DAT rating, peach tree injury responses were 4% to 5% in all treatments (Table 4). It is worth noting that all treatments had received only the initial application at 14 DAT, so no differences were observed among treatments. At 28 DAT, peach tree injury was more pronounced in treatments sprayed with dicamba at 14 DAT (0 DAT fb 14 DAT and 0 DAT fb 14 DAT fb 28 DAT) and differed from the two treatments that received only the initial application at 0 DAT (Table 4). Past 28 DAT, no regime of single or repeated applications caused peach tree injury distinct from the other treatments. At 56 DAT, trees that received only the initial dicamba application had 4.3% injury, which was not different from trees receiving additional applications of dicamba. At 84 DAT, peach tree injury was minor at <2%, regardless of the number of exposures to dicamba. Repeated applications caused the most prominent injury levels at 28 DAT among plots that were treated both at 0 DAT and at 14 DAT (Table 4), indicating that repeated exposure to dicamba early in the season will cause ephemeral injury that will not be distinguishable by 84 DAT (Table 4).

In the nozzle selection study, the 1/50X rate of dicamba (11.2 g ae ha⁻¹) caused more prominent peach tree injury than was

assessed in the repeated application trial. Injury was as high as 18% at 28 DAT when trees were treated with dicamba using TTI11002 or XR11002 nozzles (Table 5). At 14, 28, and 56 DAT, dicamba applied with XR1100067 nozzles caused less injury than dicamba applied with TTI11002, AITTJ60-11002, AIXR11002, or XR11002 nozzles (Table 5). Though droplet spectrums differed based on nozzle selection (Table 1), it is likely that the disparity in injury was due to carrier volume, as XR1100067 nozzles delivered only 47 L ha⁻¹ of spray solution relative to other nozzles, which delivered 187 L ha⁻¹ (Table 1). Previous work has demonstrated that smaller droplets from XR nozzles had reduced coverage in soybean canopies relative to more coarse droplets from TTI or AIXR nozzles (Legleiter and Johnson 2016). Furthermore, Legleiter and Johnson found that decreasing carrier volume (from 140 to 94 L ha⁻¹) reduced coverage more substantially than the nozzle type. In the present study, XR1100067 nozzles exhibited a fine spray classification with 79% of the spray volume comprising driftable fines (Table 1), so it is likely that not all droplets were intercepted by the peach trees, which have a vertical canopy and were treated from a height of 51 cm. Across all rating dates, no

Table 6. Plant vigor response of container-grown peach trees treated with a 1/100X rate of dicamba as single or repeated applications at selected days after initial treatment.^{a,b,c}

Herbicide timing	0 DAT	14 DAT	28 DAT	56 DAT	84 DAT
		— Plant	vigor (0 to	9 scale) ^a -	
0 DAT	6.7	7.6	7.4 b	6.8	7.5
0 DAT fb 14 DAT	6.9	7.9	7.5 b	6.4	7.3
0 DAT fb 14 DAT fb 28 DAT	7.3	7.8	7.3 b	6.3	7.8
0 DAT fb 28 DAT	7.1	7.9	7.9 b	6.5	7.3
Untreated	7.3	7.8	8.5 a	7.3	7.9
P-value	0.2716	0.7835	0.0025	0.0528	0.7229

^aPlant vigor was visually assessed on a 0 to 9 scale (where 0 = total plant death and

defoliation and 9 = a completely healthy tree with no indicators of stress or injury). Dicamba at a 1/100X rate was equivalent to 5.6 g ae ha⁻¹. Experiments were conducted at the Milo J. Shult Research and Extension Center in Fayetteville, AR, in 2020 and 2021. Initial herbicide applications (0 DAT) occurred when peach leaves had fully expanded, on May 29, 2020, and June 14, 2021, respectively.

^bAbbreviations: DAT, days after initial treatment; fb, followed by.

^cMeans were separated using Tukey's honestly significant difference at a $\alpha = 0.05$ significance level, and means followed by the same letter are not significantly different. Means lacking letters indicate no significant treatment effect at the $\alpha = 0.05$ significance level.

differences in peach tree injury were detected among the nozzles that delivered dicamba in a 187 L ha⁻¹ volume: TTI11002, AITTJ60-11002, AIXR11002, and XR11002 (Table 5). By 84 DAT, tree injury was less than 4% for all nozzle selection treatments, and no difference in injury was observed among nozzle treatments (Table 5).

Observed peach tree injury levels in this experiment were divergent from other findings, likely owing to the reduced rates in the present trials relative to previous research: 1/50X for the nozzle selection trial and 1/100X for the repeated application trial. Dicamba at a 1/20X and a 1/2X rate in container-grown peaches caused 19% and 57% injury, respectively (Dintelmann et al. 2020). Similarly, reduced rates of dicamba may cause high levels of crop injury in grapevines, up to 36% and 29% in response to a 1/100X and 1/20X rate, respectively (Dintelmann et al. 2020; Mohseni-Moghadam et al. 2016). Thus there is evidence that exposure to higher rates of dicamba will result in more dramatic injury symptoms in peach trees. Furthermore, the results presented here do not reproduce a continuous exposure to volatile dicamba as measured in geographies where use of the herbicide is common during summer months (Zaccaro-Gruener et al. 2023).

Plant Vigor

Rather than assessing specific herbicide symptomology relative to an untreated control, plant vigor reflects peach canopy status, tree development, and overall growth. Because plant vigor is not recorded relative to the untreated control, plant vigor assessments allow for comparisons that include the untreated control for each trial. Across both trials, peach trees appeared healthy, with no leaf drop or dying scaffold branches, and no treatment was assessed below 6 on the 0 to 9 plant vigor scale at any rating date (Tables 6 and 7). In the repeated application trial, the effect of herbicide timing on plant vigor was significant only at 28 DAT, and the only observed difference was between the untreated peaches (8.5) and peaches treated with dicamba at least once (Table 6). It is worth noting that the difference in plant vigor was minor, even where it was statistically significant. At 28 DAT, the lowest observed plant vigor was 7.3, a 14% reduction relative to the untreated control, and

Table 7. Effect of nozzle selection on visual ratings of plant vigor of containergrown peach trees exposed to dicamba at a 1/50X rate at 0, 14, 28, 56, and 84 days after treatment.^{a,b,c}

Nozzle type	0 DAT	14 DAT	28 DAT	56 DAT	84 DAT
		Plar	nt vigor (0 to	9 scale)ª ——	
TTI11002	7.1	7.3	7.1 bc	6.8 bc	7.6
AITTJ60-11002	7.3	7.4	7.3 bc	6.7 c	7.0
AIXR11002	7.4	7.2	7.1 bc	6.3 c	6.8
XR11002	6.9	7.6	6.6 c	6.3 c	7.0
XR1100067	7.3	7.6	7.7 ab	7.3 a	7.4
Untreated	6.8	7.2	8.4 a	7.2 ab	8.1
P-value	0.2280	0.8180	0.0001	< 0.0001	0.0685

^aPlant vigor was visually assessed on a 0 to 9 scale (where 0 = total plant death and defoliation and 9 = a completely healthy tree with no indicators of stress or injury). Dicamba at a 1/50X rate was equivalent to 11.2 g ae ha⁻¹. Initial herbicide applications (0 DAT) occurred when peach leaves had fully expanded, on May 29, 2020, and June 14, 2021, respectively. ^bAbbreviations: AITTJ, air induction turbo TwinJet[®] nozzle; AIXR, air induction extended-range nozzle; DAT, days after initial treatment; TTI, Turbo TeeJet[®] induction nozzle; XR, extended-range nozzle.

^cMeans were separated using Tukey's honestly significant difference at a $\alpha = 0.05$ significance level, and means followed by the same letter are not significantly different. Means lacking letters indicate no significant treatment effect at the $\alpha = 0.05$ significance level.

was observed in response to dicamba applied at 0 DAT fb 14 DAT fb 28 DAT (Table 6). At 56 DAT and 84 DAT, no differences were detected in plant vigor, regardless of application timing.

In the nozzle selection study, no differences in plant vigor were observed in peach trees until 28 DAT (Table 7). At 28 DAT, untreated peaches and peaches treated with dicamba using XR1100067 nozzles were not statistically different (Table 7). Relative to the untreated control, a statistically significant reduction in plant vigor was observed in peach trees treated with dicamba using the TTI11002, AITTJ60-11002, AIXR11002, and XR11002 nozzles at 28 DAT (Table 7). At 56 DAT, the plant vigor response relative to the untreated controls remained similar to the 28 DAT rating, except for the TTI11002 nozzles, which were no longer different from untreated controls (Table 7). By 84 DAT, no differences in plant vigor were observed in response to nozzle selection.

Dicamba and Dicamba Metabolite Residues

In the repeated application trial, dicamba residues were detected in peach leaves at a very low frequency, even though the herbicide was directly applied to the trees. In 2020, dicamba residues were observed in only 4 of the 20 submitted samples from the 14 DAT sampling date, and no submitted samples from the 69 DAT sampling date had any detectable dicamba residues (data not shown). In 2021, no dicamba residues were observed in any submitted samples from the 14 or 85 DAT sampling date. Dicamba metabolites (5-hyroxy dicamba and DCSA) were not observed in any sample from the repeated application trial. No dicamba, 5hydroxy dicamba, or DCSA was detected in any leaf samples collected from untreated controls at any sampling date in the repeated application trial. These findings indicate that dicamba was absent or below detectable levels in most samples. It is possible that dicamba applied with the XR1100067 nozzles was not completely intercepted by the peach trees, considering the high percentage of driftable fines and the fine spray classification (Table 1). However, given that peach tree injury symptoms and plant vigor reductions were observed in response to dicamba exposure throughout the repeated application trial (Tables 4 and 6), peaches were more sensitive to dicamba than the analytical

Table 8. Effect of nozzle selection on dicamba residues detected in bulked leaf collected from container-grown peach trees treated with a 1/50X rate of dicamba in Fayetteville, AR, in 2020.^{a,b,c}

Nozzle type	Dicamba residue
	ppb
TTI11002	38.2 b
AITTJ60-11002	43.5 a
AIXR11002	51.5 a
XR11002	25.1 b
XR1100067	ND
Untreated	ND
P-value	0.0031

^aSamples were processed using liquid chromatography at the Mississippi State Chemical Lab at Mississippi State University. Dicamba at a 1/50X rate was equivalent to 11.2 g ae ha⁻¹. ^bAbbreviations: AITTJ, air induction turbo TwinJet[®] nozzle; AIXR, air induction extendedrange nozzle; ND, no detection of dicamba residues; TTI, Turbo TeeJet[®] induction nozzle; XR, extended-range nozzle.

^cMeans were separated using Tukey's honestly significant difference at a $\alpha = 0.05$ significance level, and means followed by the same letter are not significantly different. For analysis of variance, samples with no detectable dicamba were excluded from analysis rather than scored as zero values.

instruments at these sampling timings. This observation of symptomatic plants and no detectable levels of dicamba is consistent with other field observations. Soybean injury in response to florpyrauxifen-benzyl was detectable at a distance farther from a drift source than was detectable in spray deposits (Butts et al. 2022). In experiments with processing tomatoes, dicamba and dicamba residues from applications of 1/100X and 1/1,000X rates were detectable only in shoot tissues on the day of exposure (Meyers et al. 2022). However, the dicamba residues were more persistent in tomato fruit, indicating the potential for crop rejection at the market, based on agricultural regulations (U.S. Code of Federal Regulations 2022).

In the nozzle selection trial, dicamba residues were detected in 16 of 24 submitted leaf samples at 14 DAT in 2020. Of the eight samples with no dicamba residues, four came from the XR1100067 nozzle treatment and four were from untreated controls (Table 8). In 2020, peach leaves from the AITTJ60-11002 and AIXR11002 treatments were observed to have the highest concentrations of dicamba residues, with dicamba detected at 43.5 and 51.5 ppb, respectively (Table 8). In 2021, dicamba residues were detected in 10 of 24 samples; however, the irregular detection frequency led to unbalanced data unsuitable for ANOVA. A consistent finding in both years was that no dicamba residues were detected from leaf samples of peach trees treated with dicamba using the XR1100067 nozzles or of untreated peach trees (Table 8).

Low levels of peach tree injury were observed in response to both the 1/50X rate of dicamba and the 1/100X rate of dicamba; thus it is concluded that dicamba drift events pose a hazard to newly planted peach trees. Although it is intuitive that multiple exposures to dicamba could cause increased injury or reduced plant vigor, the observations in this trial did not appear additive in response to repeated applications. Thus there is no evidence that multiple dicamba exposures at a 1/100X rate increase peach tree injury. The composition of the droplet spectrum in a dicamba exposure event did not dramatically affect peach tree injury or growth characteristics. The droplet spectrum with the smallest Dv50, the XR1100067 nozzle, caused the lowest peach tree injury. However, the XR110067 nozzles used a lower carrier volume $(47 L ha^{-1})$ than the remaining treatments $(187 L ha^{-1})$, so it is not clear whether the carrier volume or droplet spectrum was the causal factor. Finally, given that injury symptoms and plant vigor reductions were observed in peach trees with no dicamba residues, peach trees are sensitive to dicamba at a level below the detection thresholds implemented in this trial.

Peach trees were grown in containers for these trials to ensure isolation during dicamba applications. Other works have similarly simulated herbicide drift and assessed injury response of container-grown perennial fruit, ornamental fruit, and nut crops (Dintelmann et al. 2020; Mohseni-Moghadam et al. 2016). Container production of perennial crops is a convenient way to assess foliar injury, and the observations from these trials are helpful in assessing peach injury responses to simulated dicamba exposure events. However, future work with field-grown trees over multiple seasons could further characterize the effect of dicamba exposure on this perennial fruit crop.

Practical Implications

The findings from this experiment demonstrate the potential harmful effects of a drift event that exposes young peach trees to dicamba at realistic, driftable rates (i.e., 1/50X and 1/100X field rates). Peach trees were more dramatically injured by a single exposure event at a 1/50X rate of dicamba than by multiple exposure events at a 1/100X rate, so a single exposure event was observed to cause more peach injury than multiple exposures at lower rates. Another important finding is the disparity between peach tree injury and dicamba residue detection. The lack of residue detection at later sampling dates (69 and 84 DAT) may not be surprising, but it is noteworthy that dicamba was not consistently detected in peach leaves from the 14 DAT sampling period. This implies that peach trees are sensitive to dicamba at a level below detection thresholds with certain lab instruments and that sampling may need to occur sooner than 14 DAT to detect the residues in the exposed plants.

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