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## Carinata Tolerance to Preemergence and Postemergence Herbicides

Ramon G. Leon, Jason A. Ferrell, and Michael J. Mulvaney\*

Carinata is a new biofuel crop that was recently introduced in the southeastern USA as a winter crop. This crop is competitive after canopy closure, but there is a need for weed control options at earlier growth stages. Field experiments were conducted from 2014 to 2016 to determine the safety of several PRE and POST herbicides in carinata. Pendimethalin at 1080 g ai ha<sup>-1</sup> applied preplant incorporated (PPI) and PRE caused no carinata injury, or plant density and yield reductions. S-metolachlor was also safe at 694, 1070, 1390, and 2780 g ai ha<sup>-1</sup> applied at PRE, 3 d after planting (DAP) and at the 2- to 6-leaf stage. Flumioxazin at 72 g ai ha<sup>-1</sup> applied PRE was highly injurious on carinata preventing its establishment. Among the POST herbicides evaluated, clopyralid at 210 g ae ha<sup>-1</sup> and clethodim at 136 g ai ha<sup>-1</sup> caused minor injury to carinata but did not reduce yield compared to the nontreated control. Acifluorfen at 420 g ai ha<sup>-1</sup>, bentazon at 840 g ai ha<sup>-1</sup>, and carfentrazone at 18 g ai ha<sup>-1</sup> applied POST to carinata caused 75 to 100% injury. Under stressful conditions (i.e. high summer temperatures) all POST herbicides caused more injury than under more favorable conditions for growth in Florida (i.e. winter). The present study identified pendimethalin, S-metolachlor, clopyralid and clethodim as potential herbicides for weed control in carinata, and flumioxazin, acifluorfen, bentazon, and carfentrazone as herbicides that can be used to control volunteer carinata plants in rotational crops.

**Nomenclature:** Acifluorfen; bentazon; carfentrazone; flumioxazin; pendimethalin; S-metolachlor; carinata, *Brassica carinata* A. Braun.

**Key words:** Crop establishment, crop rotation, integrated weed management, volunteer crop control.

Carinata, also known as Ethiopian mustard, is an oilseed crop that is currently being developed for biofuel production (Bouaid et al. 2009; Cardone et al. 2003). Among the advantages that this crop has for large-scale biofuel production is that seed shattering is considerably lower than that of other crops such as spring and winter canola (*Brassica napus* L.) (Choudhary et al. 2000; Prakash and Chopra 1988), it can be grown and harvested using regular agricultural equipment (e.g., grain drills and small grain combines), and there are cultivars that can grow in either temperate or subtropical conditions (D. Males, personal communication), giving this crop a potentially large range of production opportunities. Another important trait is its high oil content (>40%) with particularly high levels of erucic and linoleic acids with little saturated fatty acid content (Cardone et al. 2003; Seepaul et al. 2016).

The process of refining carinata into biodiesel and bio-jetfuel is simple and cheap because erucic and linoleic acids yield the desired longer hydrocarbon chains than other fatty acids, reducing the need for additional synthetic chemical reactions.

Most row-crop farms in northwest Florida grow cotton–cotton–peanut summer rotation with either fallow or cover crops of oats or wheat during the winter (Katsvairo et al. 2006; Leon et al. 2015). Recently, growers have been interested in growing carinata as a winter crop because of industry demand. Because carinata is a relatively new crop in the southeastern United States, it is necessary to identify tools and develop strategies to manage weeds effectively with minimal crop injury. Furthermore, if carinata is introduced into existing rotations, it is important to make sure that volunteer plants will not interfere with rotational crops. Thus, it is also

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valuable to develop methods to control volunteer carinata plants when needed. Unfortunately, there is little information available about herbicide safety on carinata (Gasol et al. 2007; Rana 2006). The objective of the present study was to assess the safety of PPI, PRE, and POST herbicides on carinata seedling establishment and plant growth and development.

## Materials and Methods

Field experiments were conducted between 2014 and 2016 at the West Florida Research and Education Center in Jay, FL (30.78°N, 87.14°W). The soil was a Tifton sandy loam (fine-loamy, kaolinitic, thermic Plinthic Kandiudult) with 2.3% organic matter and pH 6.0. All experiments were conducted under conventional tillage (two disk passes and a rotavator pass before planting). The experimental area was maintained weed-free for the duration of the experiment to test crop injury and not weed interference. Plots were six rows wide and 4.6 m long with 36-cm row spacing and had a 1.5-m buffer area between plots. Carinata 'Avanza' (Agrisoma, Inc., Saskatoon, Canada) was planted at 5.6 kg ha<sup>-1</sup> with a vegetable planter (Jang Seeder JPH-U, Jang Automation Co. Ltd., Chungcheongbuk-do, South Korea). All experiments were arranged as randomized complete block designs, had four replications, and were conducted at least twice.

**Winter Experiment.** An experiment was conducted to evaluate carinata herbicide tolerance during the winter, which is the most favorable time to grow this crop in Florida (Seepaul et al. 2016). The crop was planted the first week of November in both 2014 and 2015, and was fertilized with 20 kg N ha<sup>-1</sup>

at planting and 40 kg N ha<sup>-1</sup> at bolting. The treatments were single or sequential applications of PRE herbicides including pendimethalin, S-metolachlor, and flumioxazin, and POST herbicides including clopyralid, acifluorfen, bentazon, carfentrazone, and clethodim, with single rates as specified in Table 1. A nontreated control was included for comparison. PPI treatments were applied the day prior to planting and were incorporated with a field cultivator. PRE treatments were applied immediately after planting and were incorporated with 12 mm of irrigation the same day. This irrigation was applied across all treatments after planting. POST treatments were applied when carinata plants reached the three- to six-leaf stage. Herbicides were applied with a tractor-mounted sprayer calibrated to deliver a spraying volume of 187 L ha<sup>-1</sup> at 28 psi with 11003 flat-fan nozzles (TeeJet Technologies, Glendale Heights, IL).

**Summer Experiment.** The above described experiment was repeated during summer with the goal of assessing carinata tolerance to the same herbicide treatments applied in a high temperature environment. The crop was planted the first week of June of 2014, 2015, and 2016, and crop management and treatment application procedures were as described for the winter experiment.

**S-Metolachlor Tolerance Experiment.** In order to characterize in more detail carinata tolerance to S-metolachlor, an experiment was conducted evaluating S-metolachlor at 694, 1,070, 1,390, and 2,780 g ai ha<sup>-1</sup> applied PRE, 3 d after planting, and POST at the three- to six-leaf stage. A nontreated control was included. Carinata was planted the first week of November 2015, and repeated in June 2016.

Table 1. Herbicide information and rates for treatments used in winter and summer experiments.

Treatment	Rate	Product	Concentration	Manufacturer
	g ai or ae ha <sup>-1</sup>			
Pendimethalin	1,080	Prowl® H <sub>2</sub> O	456 g ai L <sup>-1</sup>	BASF, Research Triangle Park, NC
S-metolachlor	1,420	DualMagnum®	915 g ai L <sup>-1</sup>	Syngenta Crop Protection, Greensboro, NC
Flumioxazin	72	Valor® SX	51% ai (w/w)	Valent USA Corp., Walnut Creek, CA
Clopyralid	210	Stinger®	360 g ae L <sup>-1</sup>	Dow AgroSciences LLC, Indianapolis, IN
Acifluorfen <sup>a</sup>	420	Ultra Blazer®	240 g ai L <sup>-1</sup>	United Phosphorus Inc., King of Prussia, PA
Bentazon <sup>a</sup>	840	Basagran®	480 g ae L <sup>-1</sup>	Arysta LifeScience North America, Cary, NC
Carfentrazone <sup>b</sup>	18	Aim® EC	240 g ai L <sup>-1</sup>	FMC Corp., Philadelphia, PA
Clethodim <sup>b</sup>	136	SelectMax®	116 g ai L <sup>-1</sup>	Valent USA Corp., Walnut Creek, CA

<sup>a</sup> Applied with crop oil concentrate at 0.5% (v/v) (Agri-dex®, Helena Holding Company, Collierville, TN).

<sup>b</sup> Applied with nonionic surfactant at 0.25% (v/v) (Induce®, Helena Holding Company, Collierville, TN).

Table 2. Temperature (mean, minimum, and maximum) and rainfall during winter (November to May) and summer (April to August) experiments in Jay, FL.

Season	T <sub>mean</sub>	T <sub>max</sub>	T <sub>min</sub>	Total rainfall mm
	C			
Summer 2014	26	35	17	146
Winter 2014/2015	15	28	-1	116
Summer 2015	26	36	17	140
Winter 2015/2016	16	28	1	156
Summer 2016	26	36	18	150

**Data Collection and Analysis.** Four weeks after POST treatments (WAT), plant density was determined by counting carinata plants in a 0.5-m row length in the middle two rows of each plot. Herbicide injury (e.g., chlorosis, necrosis, stunting) of carinata was visually estimated using a 0% to 100% rating scale in which 0% indicated no symptoms and 100% indicated crop death. For experiments conducted during winter, grain was harvested 120 to 150 days after planting when grain moisture content reached 8% to 12%. Yield was determined by harvesting the four middle rows of each plot. Yield was adjusted to 10% moisture content. No yield data was collected for summer experiments because the crop was too stressed due to high temperatures (Table 2), and no seed set was observed.

Results were analyzed by ANOVA using PROC GLIMMIX with SAS (Statistical Analysis Systems, version 9.4, Cary, NC 27513). For the winter and summer experiments the model included herbicide,

year, and their interaction as fixed effects, while block was considered a random effect. For the S-metolachlor study, the model included application timing, rate, year and their interactions as fixed effects, and block was considered a random effect. Treatments with all replications having values of 0% or 100% were not included in the ANOVA. Means separations were done using Tukey's HSD test ( $\alpha = 0.05$ ), and 95% confidence intervals were used to determine differences with treatments having means of either 0% or 100%.

## Results and Discussion

For all experiments treatment by year interactions were significant ( $P < 0.0001$ ), so data were analyzed and are presented by year.

**Winter Experiment.** Pendimethalin PRE reduced carinata plant density in 2015 but not in 2016. Although some variation was observed across years, pendimethalin PPI and S-metolachlor PRE did not reduce plant density when compared with the non-treated control. These herbicides caused no visible injury or yield reductions (Table 3). Carinata can compensate for reductions (40% to 75%) in plant density by increasing plant size and branch production, thus preventing yield reductions (Angrej et al. 2002; D. Wright personal communication). Among PRE herbicides, only flumioxazin consistently reduced carinata plant density in both years (Table 3). Carinata was highly sensitive to flumioxazin,

Table 3. Carinata plant density, injury, and yield in response to herbicide treatments in Jay, FL, during the winters of 2014/2015 and 2015/2016.<sup>a</sup>

Treatment	Rate	Plant density 4 WAT		Injury 4 WAT		Injury 8 WAT		Yield	
		2014/2015	2015/2016	2014/2015	2015/2016	2014/2015	2015/2016	2014/2015	2015/2016
	g ai ha <sup>-1</sup>	no. m <sup>-1</sup>		%				kg ha <sup>-1</sup>	
Nontreated		33 Ab <sup>b</sup>	42 a	0 b	0 d	0 b	0 c	215 abc	930 abc
Pendimethalin PPI	1,080	36 a	43 a	0 b	0 d	0 b	0 c	234 abc	1,475 a
Pendimethalin PRE	1,080	5 cd	43 a	3 b	5 d	0 b	0 c	233 abc	1,478 a
S-metolachlor PRE	1,420	22 abc	50 a	6 b	0 d	0 b	0 c	344 a	1,186 ab
Flumioxazin PRE	72	1 d	0 b	100 a	100 a	100 a	100 a	0 d	0 c
Pendimethalin PRE fb clopyralid POST	1,080 fb 210	30 ab	32 a	11 b	47 b	0 b	15 c	121 bcd	1,222 ab
Pendimethalin PRE fb acifluorfen POST	1,080 fb 420	36 a	0 b	96 a	98 a	100 a	82 ab	0 d	32 c
Pendimethalin PRE fb bentazon POST	1,080 fb 840	30 ab	2 b	94 a	100 a	100 a	100 a	0 d	511 bc
Pendimethalin PRE fb carfentrazone POST	1,080 fb 18	33 a	2 b	89 a	100 a	100 a	100 a	0 d	456 bc
Pendimethalin PRE fb clethodim POST	1,080 fb 136	27 ab	41 a	5 b	11 cd	0 b	0 c	184 abcd	1,204 ab
S-metolachlor PRE fb clopyralid POST	1,420 fb 210	23 abc	39 a	18 b	36 bc	0 b	0 c	227 abc	1,094 ab
S-metolachlor PRE fb acifluorfen POST	1,420 fb 420	22 abc	0 b	100 a	100 a	100 a	100 a	0 d	0 c
S-metolachlor PRE fb bentazon POST	1,420 fb 840	14 bcd	2 b	100 a	87 a	100 a	67 b	0 d	620 abc
S-metolachlor PRE fb carfentrazone POST	1,420 fb 18	22 abc	3 b	5 b	100 a	0 b	100 a	241 abc	657 abc
S-metolachlor PRE fb clethodim POST	1,420 fb 136	18 abcd	42 a	6 b	9 cd	0 b	0 c	270 ab	1,331 ab

<sup>a</sup> Abbreviation: WAT, weeks after treatment; fb, followed by; PPI, preplant incorporated.

<sup>b</sup> Treatments with the same letter within a column were not different based on Tukey's HSD ( $\alpha = 0.05$ ).

and plots treated with this herbicide had very few to no plants. Flumioxazin has been reported to effectively control other *Brassica* species such as wild mustard [*Brassica kaber* (DC.) L.C. Wheeler] and canola (*Brassica napus* L.). Also, because of its sensitivity to this herbicide, rotational intervals for canola vary from 6 to 12 months (Anonymous 2010). Therefore, flumioxazin can be an effective tool to control volunteer carinata in corn (*Zea mays* L.), cotton (*Gossypium hirsutum* L.), peanut (*Arachis hypogaea* L.), and soybean [*Glycine max* (L.) Merr.] (Anonymous 2010), which are common rotational crops in northwestern Florida.

In 2014/2015, very few differences in plant density were observed among treatments with POST applications. Conversely, dramatic differences were present in 2015/2016 (Table 3). The reason for this is not clear, but in 2014/2015, during the 2 wk after POST applications, temperatures were at least 4 C lower than they were in 2015/2016 (data not shown). It is possible that herbicide action was slower with plants less affected. However, considering both plant density and injury results, a clearer pattern of the impact that POST treatments had on carinata can be observed. For example, clopyralid did not reduce plant density and only caused intermediate injury rating from 11% to 18% in 2014 and from 36% to 47% in 2014/2015 4 WAT. Despite these levels of injury, plants had recovered by 8 WAT and no yield reductions were found when compared with the nontreated control. Yield in 2014/2015 was considerably lower than it was in 2015/2016 because in February 2015 a frost caused partial flower abortion (Table 3). Clethodim did not reduce plant density or yield and caused less than 11% injury in both years. This was expected because clethodim is a grass herbicide (Shaner 2014). Therefore, clopyralid and clethodim could complement PRE herbicides by providing a broadleaf- and a grass-weed control option for carinata growers, respectively.

Unlike clethodim and clopyralid, acifluorfen, bentazon, and carfentrazone effectively terminated the carinata crop, causing 87% to 100% 4 WAT and 67% to 100% 8 WAT in both years (Table 3). In 2014, most of the acifluorfen, bentazon, or carfentrazone treatments produced no yield. In 2015, despite the high level of injury observed 4 WAT, a few plants survived and recovered by 8 WAT, but yield was less than 50% that of the highest yielding plots. Carfentrazone has provided >90%

control of wild mustard (Durgan et al. 1997) and it is highly injurious to canola at very low rates (<5 g ai ha<sup>-1</sup>; Légère et al. 2006). Similarly, acifluorfen and bentazon are used to control wild mustard (Anonymous 2012, 2013). Thus, based on their activity on other Brassicaceae species and the present results, acifluorfen, bentazon, and carfentrazone could be used to control volunteer carinata POST in rotational crops.

**Summer Experiment.** Temperatures during summer were on average more than 10 C higher than they were during winter (Table 2), creating stressful conditions for the growth of carinata, which is adapted to cooler temperatures (Cardone et al. 2003; Seepaul et al. 2016). More variability in carinata plant density was observed under this more challenging environment, and some treatments such as bentazon reduced densities when compared to the nontreated control during 2016 (Table 4). However, only flumioxazin treatments reduced plant densities in each of the three years. Under high temperatures, pendimethalin and S-metolachlor did not affect plant density. Higher summer temperatures increased carinata injury by POST treatments compared with values reported for the winter experiments (Table 3). Even treatments that exhibited limited injury during the winter, such as clopyralid and clethodim, caused 75% and 69% injury 4 WAT in the 2014 summer experiment and 40% and 22% in the 2015 summer experiment. However, plant injury decreased to 0% to 28% 8 WAT despite high summer temperatures (Table 4). It is not clear why clethodim, a grass herbicide, caused carinata plant injury, but it may be related to the heat stress the crop was experiencing. Acetyl coA-carboxylase-inhibiting herbicides have been reported to cause damage through oxidative stress regulatory mechanisms even in broadleaf species. For example, Luo et al. (2004) reported that fluazifop-butyl caused membrane peroxidation in the broadleaf species bristly starbur (*Acanthospermum hispidum* DC.). In the present study, acifluorfen and carfentrazone caused 100% injury in two out of three years, and bentazon caused 100% injury in each of three years of the study. This experiment demonstrated that PRE herbicides identified as safe when evaluated under adequate growth conditions in the winter, such as pendimethalin and S-metolachlor, were also safe under summer growing season conditions.

Table 4. Carinata plant density and injury in response to herbicide treatments in Jay, FL, during the summers 2014, 2015, and 2016.<sup>a</sup>

Treatment	Rate	Plant density 4 WAT			Injury 4 WAT			Injury 8 WAT		
		2014	2015	2016	2014	2015	2016	2014	2015	2016
	g ai ha <sup>-1</sup>	no. m <sup>-1</sup>			%					
Nontreated		13 abc <sup>b</sup>	51 a	33 a	0 c	0 c	0 d	0 c	0 b	0 c
Pendimethalin PPI	1,080	10 abc	52 a	31 a	0 c	0 c	0 d	0 c	0 b	0 c
Pendimethalin PRE	1,080	9 abc	31 ab	21 ab	0 c	0 c	0 d	0 c	0 b	0 c
S-metolachlor PRE	1,420	6 bc	39 a	22 ab	0 c	0 c	32 cd	0 c	0 b	0 c
Flumioxazin PRE	72	0 c	7 b	4 c	100 a	100 a	94 a	100 a	100 a	100 a
Pendimethalin PRE fb clopyralid POST	1,080 fb 210	10 abc	44 a	25 ab	75 b	40 b	0 d	28 b	16 b	0 c
Pendimethalin PRE fb acifluorfen POST	1,080 fb 420	14 abc	43 a	29 a	100 a	100 a	75 abc	100 a	100 a	100 a
Pendimethalin PRE fb bentazon POST	1,080 fb 840	15 ab	52 a	33 a	100 a	100 a	100 d	100 a	100 a	100 a
Pendimethalin PRE fb carfentrazone POST	1,080 fb 18	NE <sup>b</sup>	48 a	37 a	100 a	100 a	41 bcd	100 a	100 a	22 bc
Pendimethalin PRE fb clethodim POST	1,080 fb 136	20 ab	52 a	25 ab	5 c	17 c	0 d	0 c	0 b	0 c
S-metolachlor PRE fb clopyralid POST	1,420 fb 210	8 abc	33 a	20 ab	88 ab	36 b	4 d	100 a	0 b	0 c
S-metolachlor PRE fb acifluorfen POST	1,420 fb 420	11 abc	30 a	19 ab	100 a	100 a	100 a	100 a	100 a	100 a
S-metolachlor PRE fb bentazon POST	1,420 fb 840	7 abc	36 a	18 b	100 a	100 a	100 a	100 a	100 a	100 a
S-metolachlor PRE fb carfentrazone POST	1,420 fb 18	22 a	43 a	29 a	100 a	100 a	38 cd	100 a	100 a	86 a
S-metolachlor PRE fb clethodim POST	1,420 fb 136	7 abc	30 a	36 a	69 b	22 bc	12 d	11 c	0 b	0 c

<sup>a</sup> Abbreviations: fb, followed by; NE, not evaluated; WAT, weeks after treatment; PPI, preplant incorporated.

<sup>b</sup> Treatments with the same letter within a column were not different based on Tukey's HSD ( $\alpha=0.05$ ).

**S-Metolachlor Tolerance Experiment.** The previous two experiments indicated that S-metolachlor can be safely used for PRE control in carinata at a commonly recommended label rate (Anonymous 2011; Tables 3 and 4). In order to identify application strategies and to better characterize S-metolachlor safety on carinata, we evaluated different rates and application timings for this herbicide (Table 5). No differences were observed on carinata plant density or yield based on application timing and rate. There was an interaction

between application timing, rate, and year ( $P < 0.0001$ ) for carinata injury, but the only difference observed was that plots treated with 2,780 g ai ha<sup>-1</sup> (twice the recommended label rate) exhibited 68% injury 6 wk after initial treatment during the 2016 summer trial. This same treatment did not differ from the nontreated in the 2015/2016 winter season (Table 5), suggesting that under adequate growing conditions for carinata, S-metolachlor is safe in this crop. Anecdotal information from growers growing carinata in Florida suggested that

Table 5. Carinata plant density and injury in response to S-metolachlor treatments applied at different timings in Jay, FL, during the winter 2015/2016 and the summer 2016.<sup>a</sup>

Timing	Rate	Plant density		Injury (6 WAIT)		Yield <sup>b</sup>
		2015/2016	2016	2015/2016	2016	2015/2016
	g ai ha <sup>-1</sup>	no. m <sup>-1</sup>		%		kg ha <sup>-1</sup>
Nontreated	0	37 abc <sup>c</sup>	42 a	0 a	0 b	1204 a
Preemergence	694	47 a	46 a	8 a	0 b	1058 a
	1,070	38 ab	42 a	12 a	0 b	1131 a
	1,390	35 ab	48 a	10 a	0 b	1149 a
	2,780	25 b	48 a	8 a	25 ab	1313 a
	3 DAP	694	33 ab	38 a	0 a	0 b
2- to 6-leaf	1,070	45 ab	49 a	9 a	0 b	1459 a
	1,390	35 ab	39 a	19 a	34 ab	1222 a
	2,780	34 ab	35 a	22 a	68 a	1258 a
	694	50 a	47 a	4 a	38 ab	1350 a
2- to 6-leaf	1,070	40 ab	44 a	26 a	21 ab	1204 a
	1,390	43 ab	43 a	10 a	24 ab	1167 a
	2,780	48 a	49 a	25 a	32 ab	857 a

<sup>a</sup> Abbreviations: DAP, days after planting; WAIT, weeks after initial treatment.

<sup>b</sup> No yield data was collected in the summer 2016 due to crop mortality caused by high temperatures.

<sup>c</sup> Treatments with the same letter within a column were not different based on Tukey's HSD ( $\alpha=0.05$ ).

the risk of *S*-metolachlor injury might be higher in soils with coarser textures (i.e., sandy soils; Wright, personal communication) than the soil in the present study (i.e., sandy loam). Delaying PRE applications a few days after planting might reduce injury risk in sandy soils as reported by Sperry et al. (2016) on sesame (*Sesamum indicum* L.).

The present study identified two herbicides (pendimethalin and *S*-metolachlor) that can be safely used in *carinata* for PRE control, and a broadleaf and a grass herbicide for POST control (clopyralid and clethodim, respectively). Additionally, we identified a PRE herbicide (flumioxazin) and three POST herbicides (acifluorfen, bentazon, and carfentrazone) that can be used to effectively control volunteer *carinata* in rotational crops. Despite these findings, screenings of a larger number of herbicides and rates under different soil types and environmental conditions is needed. Additional research should investigate the development of cultural and mechanical approaches needed to provide growers with more weed control options for *carinata* production. This is particularly important to control closely related species such as wild radish (*Raphanus raphanistrum* L.) and to avoid carry-over issues with rotational summer crops.

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