

Investigation of Palmer Amaranth (*Amaranthus palmeri*) Resistance to Glufosinate in Mississippi

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Abstract

One of the most critical issues for weed scientists today is the management of herbicide-resistant weeds. Herbicide-resistant Palmer amaranth [*Amaranthus palmeri* (S.) Watson] is the most problematic pest problem for corn (*Zea mays* L.), cotton (*Gossypium hirsutum* L.), and soybean (*Glycine max* (L.) Merr.) producers in Mississippi. Greenhouse research was conducted at the Delta Research and Extension Center from 2019 to 2021 to investigate Palmer amaranth accessions potentially resistant to glufosinate and its distribution in Mississippi. In the 2019 screening, 20 out of 54 Palmer amaranth populations tested had individuals exhibiting 2 to 20% survival from a 1X rate (0.68 kg ai ha⁻¹ as recommended rate) of glufosinate. Bolivar-3 (9% survival), Coahoma-1 (11% survival), Holmes-3 (11% survival), Issaquena-2 (13% survival), Tunica-4 (17% survival), and Yazoo-3 (20% survival) accessions had the highest rate of survival. Twelve of those accessions were selected for a dose-response study in August 2020. Palmer amaranth plants that survived a 1X rate of glufosinate (suspected glufosinate-resistant Palmer amaranth) in 2019 were transplanted to larger pots and grown for seed production in 2020. In 2020, seedheads from female Palmer amaranth were harvested, threshed, and cleaned to obtain progeny, which were then stored until further evaluation. Only one Palmer amaranth plant (F1 progeny) from Quitman-1, Tunica-4, and Washington-5 survived glufosinate (1X) in 2020. No Palmer amaranth plants survived 2X and 4X rates of glufosinate in 2020. In the 2021 screening, four out of five original Palmer amaranth accessions selected (seed from 2017) survived, except for Issaquena-2 accession. Palmer amaranth individuals from accession Coahoma-1, Holmes-3, Tunica-4, and Yazoo-3 exhibited 3.7, 3.2, 1, and 1% survival, respectively, from a 1X rate of glufosinate. No F1 Palmer amaranth survivors were identified from any of those accessions evaluated. At this time, we cannot say with certainty that these survivors are resistant to glufosinate. Additional research (including physiological and molecular studies) needs to be conducted to confirm whether offspring of these survivors have developed resistance to glufosinate.

Keywords: population, herbicide, progeny, weed

1. Introduction

Weeds compete with crops such as corn (*Zea mays* L.), wheat (*Triticum aestivum* L.), grain sorghum [*Sorghum bicolor* (L.) Moench], cotton (*Gossypium hirsutum* L.), soybean [*Glycine max* (L.) Merr], and peanut (*Arachis hypogaea* L.) for limited resources and can reduce the quantity and quality of crop yield (Swanton et al., 2015). Therefore, successful weed management programs are crucial in row crop production systems to mitigate yield loss. Herbicides serve as the foundation for complete weed control programs in agriculture due to application feasibility and overall effectiveness on troublesome weeds (Norsworthy et al., 2012; Priess et al., 2022). However, the overreliance and subsequent repetitive use of the same herbicide or different herbicides with the same mode of action can lead to selection for herbicide-resistant weeds (Perez-Jones et al., 2005; Powles et al., 1997; Simarmata et al., 2005); hence, one of the major concerns for weed scientists today is the successful management of herbicide-resistant weeds (Norsworthy et al., 2012).

Palmer amaranth is among the top five most common and troublesome weeds in Mississippi row crops (such as corn, grain sorghum, cotton, soybean, peanut, and many other crops) (Riar et al., 2013). Palmer amaranth exhibits a unique ability to quickly develop resistance to herbicides (Norsworthy et al., 2016); therefore, effective chemical control options for the weed are limited. Additionally, Palmer amaranth can accrue a large quantity of biomass that interferes with crop growth considerably (Bararpour et al., 1991; Bell et al., 2015; Jha &

Norsworthy, 2009). As a result, Palmer amaranth has the potential to reduce soybean and furrow-irrigated rice yields by 68% and 64%, respectively, at densities of 10 plants m^{-2} (King et al., 2025; Klingaman & Oliver, 1994). In cotton, one Palmer amaranth plant per 10 m of row was reported to cause 11.5% yield loss (Rowland et al., 1999). In addition to its inherent competitive attributes, Palmer amaranth has evolved resistance to eight unique herbicide sites of action (SOA) (Heap, 2025). Considering Palmer amaranth has developed resistance to many previously effective herbicides (Ward et al., 2013), glufosinate serves a key herbicide in weed management programs within the LibertyLink® (BASF, Florham Park, NJ, USA), Enlist™ (Corteva Agriscience, Indianapolis, IN, USA), and XtendFlex® (Bayer CropScience, Pittsburgh, PA, USA) systems, in which crops contain the glufosinate resistance trait.

Glufosinate is a nonselective herbicide that controls broadleaf (dicotyledonous) and some grass (monocotyledonous) weed species. With the commercial launch of glufosinate-resistant soybean and cotton in the United States, the annual use of glufosinate has increased more than 100-fold from 2007 to 2019 (USDA-NASS 2021). Sequential applications of glufosinate beginning at the V2-V3 growth stage of soybean followed by (fb) V4-V6 reduced glyphosate-resistant Palmer amaranth seed production 99% (Bararpour et al., 2020). The previously mentioned research highlights the value of glufosinate in preventing weed (glyphosate-resistant Palmer amaranth and other weeds) seed deposition to the soil seedbank, which will help slow the development of herbicide-resistant weeds while preserving available herbicide technologies (Bararpour et al., 2020). Palmer amaranth is one of the most troublesome and problematic weeds in many agronomic crops such as corn, soybean, cotton, peanuts and more throughout the southern United States (Van Wychen, 2022, 2023). Additionally, Palmer amaranth has the ability to develop resistance to many herbicides. Priess et al. (2022) reported three distinct accessions of Palmer amaranth (A2019, A2020, and B2020) for which glufosinate failed to provide appreciable control and were likely to harbor traits that allow for multiple herbicide resistance. Among those Palmer amaranth accessions selected, accession A2020 and B2020 were confirmed to be resistant to glufosinate (Priess et al., 2022).

Glufosinate translocation (to tissues below the treated) leaf was greater in the resistant Palmer amaranth compared with the susceptible accessions (Carvalho-Moore et al., 2022). These findings indicate that gene amplification and increased expression of the chloroplastic glutamine synthetase enzyme are the mechanisms glufosinate resistance in Palmer amaranth. Therefore, the suspected glufosinate resistance in the investigated Palmer amaranth accessions from Arkansas is likely a result of increased chloroplastic glutamine synthetase copy number and overexpression. No alterations were observed in the cytosolic glutamine synthetase isoform (Carvalho-Moore et al., 2022).

Managing herbicide-resistant weeds is one of the major concerns for weed scientists today. Herbicide-resistant Palmer amaranth is the greatest pest problem for corn, cotton, peanuts, and soybean producers in Mississippi (Riar et al., 2013). As of today, glufosinate-resistant Palmer amaranth has not been reported in Mississippi. Therefore, the objective of this research was to investigate Palmer Amaranth resistance to glufosinate (Group 10: glutamine synthetase inhibitors) in Mississippi.

2. Materials and Methods

2.1 2019—The Original 2017 Population

In 2019, greenhouse research was conducted at the Delta Research and Extension Center in Stoneville, Mississippi, to investigate possible Palmer amaranth resistance to glufosinate and its distribution in Mississippi. In 2017, seeds from 54 Palmer amaranth accessions were collected from across Mississippi (Table 1). Female Palmer amaranth seedheads were threshed and sifted to obtain clean seed. Offspring were then planted on September 12, 2019, in a tray (51 by 25 by 5 cm). This emerged on September 15, 2019. On September 27, 2019, Palmer amaranth plants were sprayed at the two- to four-leaf stage (3.8 to 5 cm in height) with glufosinate at 0.68 kg ai ha^{-1} (1X rate). Palmer amaranth injury/control was visibly scored on a scale of 0 to 100% (0 being no injury/control and 100 being complete plant death), using the corresponding nontreated check for each population as reference. The percentage of Palmer amaranth that survived from glufosinate application was calculated based on the number of plants sprayed. Glufosinate was sprayed on 52 (out of 54) viable Palmer amaranth populations. For each accession evaluated, more than 100 plants were sprayed across four replications. The percentage of Palmer amaranth survival was also determined following the applications of glufosinate by visible observation ($< 70\%$ control). The distribution of Palmer amaranth that survived the applications of glufosinate has been identified four-weeks after herbicide application. Surviving Palmer amaranth (Figure 1) plants were initially transplanted into pots (10 cm diameter by 9 cm height) (Figure 2) and then later moved to larger pots (15 cm diameter by 17 cm height) to promote weed growth and development, as well as seed

production. Transplanted Palmer amaranth plants were maintained in the greenhouse for a few months (until flowering) to obtain additional offspring from female plants. The seedheads from female Palmer amaranth plants were processed using the same method as previously described to obtain clean seed (F1 progeny).

2.2 2020—The F1 Progeny (Suspected Glufosinate-Resistant Palmer Amaranth)

In 2020, seedheads of female Palmer amaranth plants from the 2019 screen were harvested and processed (threshed and sifted). A limited number of seeds were obtained and stored for dose response evaluation. Suspected glufosinate-resistant Palmer amaranth population number 2, 3, 5, 12, 18, 24, 40, 42, 43, 47, 50, and 53 (exhibited survivors following glufosinate treatment) were selected (Table 1) for a dose response evaluation utilizing a 1/2X, 1X, 2X, and 4X rate of glufosinate. Progeny (F1) of the suspected glufosinate-resistant Palmer amaranth were planted in a tray on August 29, 2020, and emerged on August 31, 2020. On September 15, 2020, Palmer amaranth seedlings were treated with a 1/2X, 1X, 2X, and 4X rate of glufosinate once the plant reached the two- to four-leaf growth stage (3.8 to 5 cm in height). Visible estimations for Palmer amaranth control were recorded as previously described. Palmer amaranth accessions exhibiting survivors to different rates of glufosinate were recorded. At this point (four-weeks after glufosinate application), the level (1/2 X, 1X, 2X, 4X) of Palmer amaranth (2019 survivor's progeny—F1) resistance to glufosinate identified.

2.3 2021—

In August 2021, five Palmer amaranth accessions from the original samples, which included accessions 5, 12, 18, 42, and 53, along with their progeny (F1), were selected (with the highest survival rate) for an additional glufosinate screening to verify these accessions were resistant to glufosinate. The study was conducted in the greenhouse that temperatures were 32, 25, 22, 25, and 27 °C and relative humidities were 55, 45, 45, 40, and 44% in September, October, November, December, and January, respectively. Seeds from all Palmer amaranth accessions were planted in a tray on September 22, emerged on September 25, and were then treated with glufosinate at 0.68 kg ha⁻¹ on October 13, 2021, once individual plants reached the two- to four-leaf growth stage (2.5 to 5 cm in height). The percentages of Palmer amaranth plants that survived 1X rate of glufosinate were calculated based on the number of plants sprayed utilizing the following equation:

$$\text{Survived Palmer amaranth \%} = a/b \times 100 \quad (1)$$

where, a = total number of Palmer amaranth that survived glufosinate treatment; b = total number of Palmer amaranth present before the glufosinate application.

3. Results

3.1 2019

In the 2019 screening, some Palmer amaranth accessions of the 54 evaluated exhibited survivors (ranging from 2 to 20 survivors) following 1X rate (0.68 kg ai ha⁻¹) of glufosinate (Table 1). Palmer amaranth population numbers 2, 3, 5, 12, 18, 24, 40, 42, 43, 47, 50, and 53 had some level of survivors (Table 1). The Palmer amaranth accessions from Bolivar-3, Coahoma-1, Holmes-3, Issaquena-2, Tunica-4, and Yazoo-3 had 9, 11, 11, 13, 17, and 20% survival from 1X rate of glufosinate application (Table 1).

Table 1. Fifty-four Palmer amaranth accessions collected from various counties in Mississippi, the total number of Palmer amaranth plants before treatment with glufosinate, the total number of Palmer amaranth plants that survived treatment with glufosinate at 0.68 kg ai ha⁻¹, and the percentage of Palmer amaranth survivors from each accession

Population No.	County	Plants (#)	Survived (#)	Survival (%)
1	Bolivar-1	171	0	0
2	Bolivar-2	228	10	4.4
3	Bolivar-3	159	14	8.8
4	Bolivar-4	235	10	4.3
5	Coahoma-1	245	28	11.4
6	Coahoma-2	123	0	0
7	Coahoma-3	172	10	5.8
8	Desoto-1	142	0	0
9	Desoto-2	178	1	0.6
10	Holmes-1	114	0	0

11	Holmes-2	92	0	0
12	Holmes-3	225	26	11.6
13	Humphrey-1	75	0	0
14	Humphrey-2	147	0	0
15	Humphrey-3	145	0	0
16	Humphrey-4	208	5	2.4
17	Issaquena-1	135	0	0
18	Issaquena-2	162	22	13.6
19	Issaquena-3	112	2	1.8
20	Issaquena-4	145	2	1.4
21	Leflore-1	109	4	3.7
22	Leflore-2	129	1	0.8
23	Leflore-3	102	0	0
24	Quitman-1	125	5	4
25	Quitman-2	145	5	3.4
26	Quitman-3	142	0	0
27	Quitman-4	82	0	0
28	Sharkey-1	142	8	5.6
29	Sharkey-2	96	0	0
30	Sharkey-3	199	0	0
31	Sharkey-4	174	8	4.6
32	Sunflower-1			
33	Sunflower-2	110	0	0
34	Sunflower-3	225	1	0.4
35	Sunflower-4	132	0	0
36	Tallahatchie-1	159	0	0
37	Tallahatchie-2	102	0	0
38	Tallahatchie-3	151	4	2.6
39	Tunica-1	152	0	0
40	Tunica-2	167	12	7.2
41	Tunica-3	118	0	0
42	Tunica-4	157	28	17.8
43	Warren-1	215	18	8.4
44	Warren-2	121	0	0
45	Warren-3	124	0	0
46	Washington-1			
47	Washington-2	162	3	1.9
48	Washington-3	142	0	0
49	Washington-4	135	4	3
50	Washington-5	109	3	2.8
51	Yazoo-1	38	0	0
52	Yazoo-2	120	0	0
53	Yazoo-3	157	32	20.4
54	Yazoo-4	47	0	0

Note. * Red color means no emergence.

3.2 2020

Palmer amaranth accessions that survived from 1X rate of glufosinate application in 2019 (2019 survivor's progeny—F1) (Table 2a), were sprayed at 1/2 X, 1X, 2X, 4X rates to identify the level of glufosinate-resistant Palmer amaranth. Only one Palmer amaranth plant (F1 progeny) from Quitman-1 (#24), Tunica-4 (#42), and Washington-5 (#50) survived a 1X rate (0.68 kg ai/ha) of glufosinate (Table 2b). No Palmer amaranth plants

survived glufosinate at 2X and 4X rates. Based on these results, it is not definitive whether these survivors are resistant due to survivors Palmer amaranth produced susceptible offspring.

Table 2a. Total number of F1 progeny of Palmer amaranth from different Counties of Mississippi before glufosinate application in 2020

Glufosinate rate	F1 (progeny) Palmer amaranth population #											
	2	3	5	12	18	24	40	42	43	47	50	53
kg ai ha ⁻¹	# of Palmer amaranth survivors											
0.34 (1/2X)		29	11	15	27				61			10
0.68 (1X)		15	7	48	25				62			6
0.68 (1X)	2	12	12	46	32	53	12	3	56	47	51	11
1.36 (2X)	3	13	10	88	22	46	7	11	58	54	59	11
2.72 (4X)		19	11	68	7				69			15

Table 2b. Number of F1 progeny of Palmer amaranth survivors from different Counties of Mississippi 4-weeks after glufosinate application in 2020

Glufosinate rate	F1 (progeny) Palmer amaranth population #											
	2	3	5	12	18	24	40	42	43	47	50	53
kg ai ha ⁻¹	# of Palmer amaranth survivors											
0.34 (1/2X)		3	0	3	1				6			1
0.68 (1X)		0	0	0	0				0			0
0.68 (1X)	0	0	0	0	0	1	0	1	0	0	1	0
1.36 (2X)	0	0	0	0	0	0	0	0	0	0	0	0
2.72 (4X)		0	0	0	0				0			0



Figure 1. Palmer amaranth accession from Tunica-4 (population #42) with 17% survivors from 1X rate of glufosinate application (0.68 kg ai ha⁻¹)—2019



Figure 2. Palmer amaranth populations survived from glufosinate application were transplanted to small pots for establishment and later transplanted to a bigger pot for seed production (F1) for future dose response testing in 2020

3.3 2021

In 2021 screening, four out of five Palmer amaranth accessions [except Issaquena-2 (no survivor)] from the original populations (seed collected in 2017) had survival from 1X rate of glufosinate (Table 3a). Palmer amaranth accessions Coahoma-1, Holmes-3, Tunica-4, and Yazoo-3 exhibited 3.7, 3.2, 1, and 1% survival following glufosinate application at 1X rate, respectively. None of the F1 Palmer amaranth progeny survivors were observed from any of the previously mentioned accessions (Table 3b).

Table 3a. Palmer amaranth original population (2017) (2021 screening) from Coahoma-1 (#5), Holmes-3 (#12), Issaquena-2 (#18), Tunica-4 (#42), and Yazoo-3 Counties (#53) populations (suspected glufosinate-resistant) responses (survivor) to glufosinate (Liberty) application at 0.68 kg ai ha⁻¹

Population #	County	Total plant	# Survived	% Survivor
5	Coahoma-1	190	7	3.7
12	Holmes-3	310	10	3.2
18	Issaquena-2	210	0	0
42	Tunica-4	490	2	1
53	Yazoo-3	185	2	1

Table 3b. Palmer amaranth F1 progeny population (2021 screening) from Coahoma-1 (#5), Holmes-3 (#12), Issaquena-2 (#18), Tunica-4 (#42), and Yazoo-3 Counties (#53) populations (suspected glufosinate-resistant) responses (survivor) to glufosinate (Liberty) application at 0.68 kg ai ha⁻¹

Population #	County	Total plant	# Survived	% Survivor
5	Coahoma-1	12	0	0
12	Holmes-3	120	0	0
18	Issaquena-2	24	0	0
42	Tunica-4	5	0	0
53	Yazoo-3	8	0	0

4. Discussion

Palmer amaranth is a major problem in most row crop production systems in Mississippi and has developed resistance to many herbicides with different modes of action. In 2022, glufosinate-resistant Palmer amaranth was reported in Arkansas (Priess et al., 2022). However, as of today, no reports of Palmer amaranth being resistant to glufosinate exist in Mississippi. In 2019 screening, there was 2 to 20% Palmer amaranth survival of the original populations (seeds collected in 2017) from the application of glufosinate application at 0.68 kg ai ha⁻¹. But from the progeny of those survivors, only one Palmer amaranth plant (F1 progeny) from Quitman-1, Tunica-4, and Washington-5 survived at 1X rate of glufosinate in 2020. In 2021 screening, there was only 1 to 3.7% survival of the original Palmer amaranth populations from Coahoma-1 (11% survival in 2019), Holmes-3 (12% survival in 2019), Tunica-4 (18%, survival in 2019), and Yazoo-3 (20% survival in 2019) and no survivor from the F1 progeny from the application of glufosinate at 1X rate. Based on these results, it remains unclear whether these survivors from the original Palmer amaranth populations are resistant to glufosinate due to those Palmer amaranth plants that survived producing susceptible offspring (F1). Sabba et al. (2003) also found F1 progeny that were susceptible to clopyralid and picloram herbicides, suggesting that the resistance trait was inherently recessive and that it was of nuclear origin. However, we can say that the agricultural fields from which these samples were collected have difficult-to-control Palmer amaranth populations. These fields are identified, and growers will be advised to manage these fields intensively to control Palmer amaranth escaping herbicide applications. Therefore, those suspected glufosinate-resistant Palmer amaranth fields need to be monitored and seeds to be collected for further investigation and testing. Glufosinate is the key herbicide in soybean, cotton, and corn in weed management programs especially for weeds that are resistant to glyphosate herbicide. The monitoring of fields suspected to contain glufosinate-resistant Palmer amaranth is necessary to detect any resistance issue and prevent the spread of resistance traits to other fields in Mississippi. An efficient weed management program should focus on eliminating crop-weed interference and sustaining this result for as long as possible by preventing the occurrence of herbicide-resistant weeds and reducing the soil seedbank (Bararpour et al., 2020, 2022). Sequential applications of glufosinate beginning at the V2-V3 growth stage of soybean followed by (fb) V4-V6 reduced glyphosate-resistant Palmer amaranth seed production 99% (Bararpour et al., 2020). Using preemergence residual herbicide, such as boundary (*S*-metolachlor + metribuzin) in soybean and cotolan (fluometuron) or brake (fluridone) in cotton, will be one strategy to control Palmer amaranth in many cropping systems. The results of this research will help to combat the suspected glufosinate-resistant Palmer amaranth in order to identify solutions for current weed problems and to communicate the results to stakeholders, but also to preserve the existing and new technologies for as long as possible.

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Authors Contributions

Dr. Taghi Bararpour designed, conducted, collected data, wrote and revised the manuscript. Dr. Jason Bond and Mr. Tanner King edited and revised the manuscript in its entirety.

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Competing Interests

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