

# Soybean Response to Weed Residues in the Soil

Dwayne D. Joseph<sup>1</sup>, Michael W. Marshall<sup>2\*</sup> , Matthew Cutulle<sup>3</sup>

<sup>1</sup>Department of Plant and Environmental Sciences, Clemson University, Clemson, SC, USA

<sup>2</sup>Edisto Research and Education Center, Clemson University, Blackville, SC, USA

<sup>3</sup>Coastal Research and Education Center, Clemson University, Charleston, SC, USA

Email: \*marsha3@clemson.edu

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## Abstract

Soybean production systems that return plant residues to the soil surface are gaining in popularity. As these practices become more widespread, more crop and weed residues are being introduced into the upper soil profile. Greenhouse studies were conducted to determine the effects of varying concentrations of Palmer amaranth and pitted morningglory plant residues (above-ground portion of the plant) on soybean production. The study was arranged in a completely randomized experimental design with five treatments and five replications. Palmer amaranth and pitted morningglory residues were incorporated into soil at 20,000, 40,000, 80,000 and 160,000 ppm. Inert plastic residue at the same residue levels was included as a check. Soybean dry weight, leaf area and leaf tissue nutrient content were recorded during the study. A decrease in soybean dry weight and leaf area was observed as Palmer amaranth residue in the soil increased. Palmer amaranth residues of 160,000 ppm and 80,000 ppm in the soil significantly reduced soybean dry weight by 69% and 59%, respectively, and soybean leaf area by 60% and 57%, respectively. In contrast, pitted morningglory and inert plastic residues had no observable effect on soybean growth and development. This study demonstrated Palmer amaranth residues in the soil impacted early season soybean growth and development.

## Keywords

Reduced Tillage, Plant Residues, Allelopathy, *Glycine max* L.

## 1. Introduction

Field preparation at the beginning of the growing season or after harvest varies depending on whether a producer chooses conservative or aggressive methods of soil preparation [1]. Soil preparation may be achieved through conventional til-

lage or ploughing or by more conservative methods like strip tilling. Disc harrowing followed by strip tilling to create crop rows is sometimes paired together. In soybean fields, shallow tilling is practiced for weed control in spring planted fields. Recently, reduced-till and no-till soybean production has become more popular as producers seek to reduce input costs. Reduced and no-till enhance soil properties [2]; however, they leave residue (above-ground portion of the plants) which is not incorporated into the upper soil profile [3]. In many instances, these are viewed as good methods of weed control because most of all plants are killed during these procedures, especially when using a disc harrow. This may generally be a good practice that promotes a healthy soil by recycling the nutrients present in the plant residues; however, in some instances, increased residues from plants known to have inhibitory or allelopathic effects may lead to reduced yield, seed germination and overall crop health [4]-[6]. Although not common in an agricultural setting and, specifically in agronomic crop production, researchers have reported inhibition of crop growth after specific plant residues were incorporated into soil [7].

In South Carolina and most of the southern US, Palmer amaranth (*Amaranthus palmeri* L. S. Wats) is the most common and troublesome weed in row-crop production [8]. Its ability to germinate and grow throughout the growing season often makes it the first significant weeds to emerge and the last weed present at harvest. Palmer amaranth is responsible for yield losses in soybean [9], and its presence at harvest often hinders harvest efficiency [9]-[11]. Palmer amaranth's growth habit and its resilience in drought-prone, low nutrient soils increase its competitiveness with crops [12]. Currently, resistance to multiple herbicide modes-of-action has aided Palmer amaranth biotypes in their spread as they become increasingly difficult to control chemically [8]. Since herbicide resistant Palmer amaranth has become a problem in the Southern US, research studies have intensified on developing effective management strategies including alternative herbicides and other cultural practices for this weed in agronomic crops [12]-[18].

A previous study conducted in Wisconsin found that redroot pigweed (*Amaranthus retroflexus* L.), a relative of Palmer amaranth, reduced soybean yield when its residue was incorporated into soil before planting [4]. Other studies have shown that Palmer amaranth soil incorporated residues have an inhibitory effect on cabbage and carrot seedlings [6]. A possible conclusion from these studies may be that Palmer amaranth is using some unknown chemical to help it thrive in crop production fields at the expense of other plants. That claim may not be made before investigating what is going on in this weed-crop competition dynamic. As stated earlier, in many instances, the introduction of plant residues to soil should promote the healthy growth of plants. This can be attributed to the introduction of organic matter and nutrients found in the plant residue, similar to the effect of composting. If the opposite occurs and plant growth and seed germination are suppressed, there might be potential phytotoxic chemicals being

released by the plant residues either from the roots or shoot that negates the positive effects of the presence of organic matter and nutrients that are being introduced into the soil that result in the promotion of plant growth.

There are some plants that possess the ability to produce and release allelopathic chemicals in their immediate vicinity that inhibit plant growth and or seed germination of other plants [19]-[21]. When there is a perceived threat, plants may also induce the production of allelochemicals, in other instances, this defense mechanism is constitutive, and those chemicals are produced regardless of a perceived threat or competition from other plants. In some cases, the allelochemicals are only effective on non-kin species [22] [23] to prevent the competition from other plants in their habitat. These allelochemicals are released in several different ways; from exudates produced in the plant roots, volatilization, decomposition of residues and from shoot allelochemicals that are released to soil through leaf litter [24]-[26]. The main purpose of these defenses is to give the plant an added advantage when trying to grow and thrive in the presence of other plants.

Studies on the ability of Palmer amaranth to inhibit plant growth are limited. Phenolic acids have been referred to as putative allelochemical [27] and their production by Palmer amaranth may suggest the use of allelopathy; however, allelopathy is often the result of several compounds working together [28] and their discovery and isolation are beyond the scope of this study. Therefore, the objective of this study was to determine the effect of varying levels of Palmer amaranth residue on the growth and development of soybean plants.

## 2. Materials and Methods

Greenhouse studies were conducted at the Biosystems Research Complex in Clemson University, Clemson, SC, USA. Palmer amaranth (AMAPA) and pitted morningglory (*Ipomoea lacunosa* L.) [IPOLA] plants grown for eight weeks in individual plastic pots using a commercial mineral soil mix at the following conditions with a photoperiod of 18/6 hour day/night cycle using supplemental overhead lights and temperature regime of 25°C/20°C day/night. Plants were harvested at the soil surface level and the shoot material was oven dried for 72 hours at 50°C. The dried plants were then ground to pass through a 2-mm sieve using a Wiley mill (Thomas Scientific, Swedesboro, NJ). The resulting ground plant residue were then stored in zip-lock bags and refrigerated at 10 C for subsequent use.

The study was arranged in a completely randomized experimental design that consisted of 5 treatments and 5 replications, and the experiment was performed twice. The 5 treatments consisted of varying weights of plant residue material incorporated into soil. The control treatments contained no plant residue (0 ppm), the 20,000 ppm treatment contained 12 g of plant residue material, the 40,000 ppm treatment contained 24 g of plant material, the 80,000 ppm treatment contained 48 g of plant residue material and the 160,000 ppm treatment

contained 96 g of plant residue material. Each treatment was thoroughly incorporated into 600 g of potting soil mix and then placed in a plastic pot. The nutrient content of the AMAPA and IPOLA residue was not collected before the initiation of the study due to the potential variation between residue samples collected. The 25 plastic pots were then sown with three soybean seeds each (to maximize the probability of seed germination). When all the soybean seedlings reached the unifoliolate growth stage, the first emerging seedling was kept for the remainder of the study and the others were removed. The above procedure was repeated using pitted morningglory residues as a weed comparison and an inert shredded plastic material as a control material to the effects of the AMAPA residues in the soil. Pitted morningglory was selected because of its ability to produce high amounts of residue during the growing season and return to the soil and its prevalence in southern US cropping systems.

All plants ( $N = 75$ ) were grown for eight weeks. Plants were watered when needed using an overhead sprinkler system and subjected to a 16-hour daylight period. After seven weeks, leaf area measurements were taken for each plant using a LI-COR LI-3100C leaf area meter (LI-COR Biosciences Inc, Lincoln, NE, USA). The aboveground portion of each plant were clipped, individually bagged, and then oven dried for 72 hours at 50°C. After drying, the dry weight of each plant was recorded, and each sample was sent to the Clemson University Agricultural Services Lab for plant tissue nutrient analysis.

Data collected were analyzed using one-way ANOVA in JMP Pro 12.2 (SAS Institute Inc, Cary, NC). Treatment means were separated using Fisher's Protected LSD at the  $P = 0.05$  level.

### 3. Results

#### 3.1. AMAPA Residue

In the AMAPA residue trials, there was an overall decrease in soybean dry weight and leaf area as AMAPA residue concentration in the soil increased. When the data was subjected to a one-way ANOVA, there was a significant effect of AMAPA residue in the soil on soybean dry weight ( $P = 0.0058$ ). The mean soybean dry weight steadily decreased as the AMAPA residue in the soil increased. This resulted in the treatments with lower concentrations of AMAPA residue (20,000 ppm and 40,000 ppm) being significantly different to the treatments with higher AMAPA residue concentrations (80,000 ppm and 160,000 ppm). AMAPA residues of 160,000 ppm in the soil reduced soybean dry weight by 69% (**Table 1**). Similarly, AMAPA residues of 80,000 ppm in the soil reduced soybean dry weight by 58%. A significant drop in soybean dry weight (50%) was also observed when AMAPA residue concentration in the soil was increased from 20,000 ppm to 80,000 ppm. The lower AMAPA residue concentrations (20,000 ppm and 40,000 ppm) showed no significant differences when compared to the control but as the concentrations increased exponentially, significance was observed at the higher concentrations (80,000 ppm and 160,000 ppm).

**Table 1.** Soybean dry weight and leaf area response to various levels of Palmer amaranth (AMAPA), pitted morningglory (IPOLA), and plastic residues in the soil.

Residue <sup>2</sup>	AMAPA		IPOLA		Plastic	
	Dry Weight	Leaf Area	Dry Weight	Leaf Area	Dry Weight	Leaf Area
	g	cm <sup>2</sup>	g	cm <sup>2</sup>	g	cm <sup>2</sup>
0	8.7 a	1112.7 a	9.6 a	1271.8 a	9.6 a	1207.8 a
20000	7.3 a	1012.2 a	8.5 a	1206.3 a	9.6 a	1167.0 a
40000	6.5 a	934.2 a	6.5 a	984.9 a	9.3 a	1167.2 a
80000	3.7 b	483.9 b	6.0 a	1026.6 a	9.9 a	1189.8 a
160000	2.7 b	440.8 b	5.6 a	945.0 a	9.5 a	1227.2 a

<sup>1</sup>Means followed by the same letter are not significantly different according to Fishers Protected LSD at 5%; <sup>2</sup>Residue level units are given in ppm.

Similar to the dry weight data, soybean leaf area decreased as the AMAPA residue concentration in the soil increased, resulting in a significant difference among treatment means when the data was subjected to a one-way ANOVA ( $P = 0.0155$ ). There were greater differences in treatment means as the AMAPA residue concentration in the soil increased, as was observed with the dry weight data. The greatest decrease in soybean leaf area was observed between the highest AMAPA residue concentration treatment (160,000 ppm) and the control treatment, where the soybean leaf area decreased by 60% from a mean of 1112.7 cm<sup>2</sup> to 440.8 cm<sup>2</sup>. A 57% decrease in leaf area was observed between the control (0 ppm) and the 80,000 ppm AMAPA residue treatment. Increases in AMAPA residue in the lower concentration treatments (20,000 ppm and 40,000 ppm) did not result in large decreases in soybean leaf area; an 8% decrease compared to a 48% decrease exhibited when comparing the 40,000 ppm and 80,000 ppm concentration treatments. In general, soybean leaf area treatment differences mirrored the results of the soybean dry weight data.

The pattern observed with the nutrient concentration within soybean plant tissue went contrary to what was expected. As the AMAPA residue concentrations in the soil increased, the expected outcome was the macronutrient levels would become more deficient; however, the opposite occurred. Phosphorous leaf tissue concentrations showed significant differences among treatments; the treatments containing a higher concentration of AMAPA residues in the soil (80,000 ppm and 160,000 ppm) had greater levels of phosphorous in their tissues (**Table 2**). The treatments containing a lower concentration of AMAPA residue in the soil (20,000 ppm and 40,000 ppm) showed significantly lower phosphorous levels in their tissue compared to the other two treatments. There was a 547% increase in the levels of soybean plant tissue phosphorous from the control treatment to the highest residue concentration treatment (**Table 2**). Potassium leaf tissue concentrations showed significant differences among treatment means. As the AMAPA residue concentration in the soil increased, the potassium concentration in soybean plant tissue also increased (**Table 2**). The two

treatments containing the higher concentration of AMAPA residue in the soil had potassium levels in the plant that were significantly higher than the lower two concentrations. The nitrogen concentration in soybean leaf tissue showed no significant differences in treatment means.

**Table 2.** Macronutrient concentrations in soybean tissue as affected by various levels of Palmer amaranth (AMAPA) residues in the soil.

AMAPA Residue <sup>2</sup>	Soybean Tissue Macronutrients <sup>1</sup>		
	N mg·g <sup>-1</sup>	P mg·g <sup>-1</sup>	K mg·g <sup>-1</sup>
0	23.0 a	2.9 b	20.3 b
20000	31.3 a	3.7 b	23.3 b
40000	31.1 a	5.0 b	22.3b
80000	30.3 a	18.6 a	34.6 a
160000	25.5 a	18.4 a	41.5a

<sup>1</sup>Means followed by the same letter are not significantly different according to Fishers Protected LSD at 5%; <sup>2</sup>Residue level units are given in ppm.

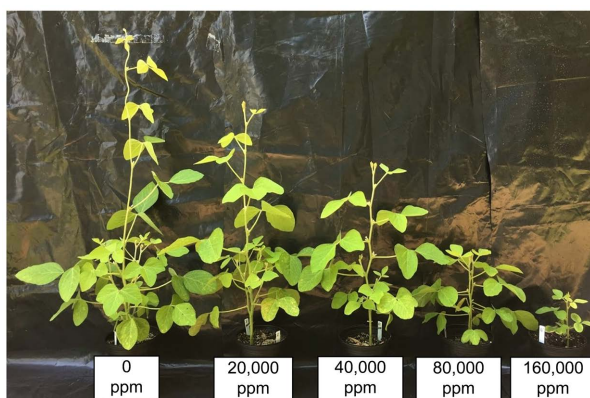
### 3.2. IPOLA and Plastic Residue

Visually, there were slight decreases in soybean dry weights as the IPOLA residue concentrations in the soil increased; however, when data was subjected to a one-way ANOVA, there were no significant differences observed [ $P = 0.1668$ ] (**Table 2**). No significant differences ( $P = 0.6675$ ) were observed with soybean leaf area data when subjected to a one-way ANOVA. Plastic residue (shavings) when added to the soil in the identical treatment concentrations as the previous two plant residues, produced no significant differences among treatments for soybean dry weight ( $P = 0.9769$ ) or leaf area ( $P = 0.9867$ ) (**Table 1**).

The experiment demonstrated that as the concentration of AMAPA residue levels increased in the soil, the growth rate and overall plant fitness of soybean seedlings decreased (**Figure 1**). The same was not observed with the incorporation of IPOLA residues into the soil. With everything else remaining constant (soil in pots, water applied per pot and light) we can deduce that the AMAPA residue concentration increase is actively inhibiting the growth of soybean seedlings. From daily observations of individual soybean plants, a case can be made that the suppression of soybean growth may be attributed primarily to slow germination along with slow leaf and stem growth (data not reported).

The soybean seedlings in the control treatment needed about 3 - 4 days for them to reach 100% VE (emergence) growth stage. Within 7 days, 100% of those seedlings were at the VC (cotyledons) growth stage. As the AMAPA residue concentration increased in the soil, slight delays were observed in the respective treatments in reaching those growth stage milestones. There were negligible differences in the growth stages observed between the 20,000 ppm and 40,000 ppm

AMAPA residue treatments, with about a 1-day delay occurring between them and the control. However, major differences were observed with the 80,000 ppm and 160,000 ppm AMAPA residue concentration treatments; after 7 days the 80,000 ppm treatment had 80% of the seedlings at VE and 20% of the seedlings were not emerged. In the 160,000 ppm treatment, after 7 days, 20% of the seedlings were at VC and 80% were not emerged.



**Figure 1.** Soybean plant growth as affected by varying levels of Palmer amaranth residue in the soil.

After 10 days, the 0 ppm and 20,000 ppm AMAPA residue treatments were all at the unifoliate leaf growth stage and appeared healthy with slight variations in height, the 40,000 ppm AMAPA residue treatment showed slightly more pronounced differences in height and appearance compared to the 0 ppm and 20,000 ppm AMAPA residue treatments. After 10 days, the 80,000 ppm AMAPA residue treatment had seedlings that were all at VC and the 160,000 ppm AMAPA residue treatment samples had 40% at VC, 20% at VE and 40% were yet to emerge. Identical concentrations of IPOLA and plastic residues did not significantly delay soybean emergence. By observing the emergence and the time it took for the soybean seedlings to reach different growth milestones, the AMAPA residue present in the soil delayed soybean germination and inhibited seedling growth as its concentration increases (**Figure 1**).

#### 4. Discussion

Phenolic acids and associated compounds derived through the shikimic acid pathway are the most common type of growth inhibitors produced in living plants or released by the decomposition of plant parts by microbial action or leaching [29]. In this study, the observed delay in soybean germination followed the results found by [30]. They reported that the compound hexanal inhibited soybean germination and subsequent growth. Hexanal is biosynthesized in plants by the action of lipoxygenase and hydroperoxide lyase on linoleic acid in plants [31]. Various other phenolic compounds including caffeic, t-cinnamic, p-coumaric, ferulic, gallic and vanillic acid, at certain concentrations, have all

been reported as growth inhibitors in soybean [29]. Extensive research hasn't been done on the phenolic compounds present in AMAPA; therefore, definitive answers regarding the specific compounds present in AMAPA tissue residues do not exist, but these phenolic compounds present in AMAPA may be the primary drivers behind the observed inhibition of soybean germination and subsequent soybean growth.

The leaf area accumulation of a plant is directly related to its leaf cell expansion ability [32], this measurement declined as Palmer amaranth residue increased in the soil. Various stress factors and conditions may contribute to the inhibition of leaf cell production and expansion, including water and salt stress [32] among others. In this experiment, water levels in all the plants were kept at a sufficient level; however, the uptake of water by those plants was a parameter not measured. The increased levels of AMAPA residue in the soil may have trapped the water in the soil and reduced the available water for the soybean seedlings during leaf growth and expansion. The complexity of the various pathways and circumstances that led to reduced leaf area are vast; however, the underlying issue is one that was initiated by the potential presence of phenolic acids or other allelochemicals present in the AMAPA residues in the soil. In cucumber, phenolic acids inhibited leaf expansion [33]. A reduction in the hydraulic conductivity of cucumber seedlings resulted in reduced water uptake and decreased water potential [34] which are the primary effect of phenolic acids.

## 5. Conclusion

The vast scope of isolating, extracting, and determining the specific phenolic compounds present in AMAPA plant residues is beyond this study. When pure compounds from fresh AMAPA residue are introduced into the soil, microorganisms facilitate their decay and metabolize these compounds resulting in a vast array of new substances, some can be allelopathic. Further studies must be performed to determine what specific compounds or pathways in soil lead to the inhibition of soybean growth by AMAPA residues. This study highlighted the potential presence of inhibitory substances in AMAPA plant residues that soybean producers should be aware of when planning their production practices. The evidence from this study demonstrates that AMAPA residues introduced to soils can have an inhibitory effect on seedling soybean growth and development. Occasional tillage or vertical mixing of the soil could reduce the effects of these AMAPA residues in the upper portion of the profile.

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## Conflicts of Interest

The authors declare no conflicts of interest regarding the publication of this paper.

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