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ALLELOCHEMICAL POTENTIAL OF SUNFLOWER (*HELIANTHUS ANNUUS L.*) IN WEEDS SUPPRESSION

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SUMMARY

The presented study transpired in 2015–2016 in the Wasit Province, Iraq, to investigate the allelopathic impact of two sunflower (*Helianthus annuus L.*) cultivars, namely, Asgaro and Flamme, on weed suppression. The study found that sunflower cultivars significantly reduced the number and biomass of companion weeds in sunflower fields, with a varying reduction depending on the cultivar genotype. The cultivar Asgaro exhibited the most pronounced allelopathic effects, resulting in a 68.6% reduction in total weed numbers and a 61.0% reduction in weed biomass compared with the control (no sunflower). However, the genotype Flamme had the minimum allelopathic effects. Specifically, genotype Flamme reduced weed population by 24% and weed biomass by 35.4% compared with the control. More analysis revealed that the root exudates of the Asgaro genotype were more efficient in suppressing weeds than the Flamme genotype. Substance analysis demonstrated that the Asgaro genotype had a significantly higher amount (0.403 mg/g) than the Flamme genotype (0.220 mg/g) of specific compounds and phenols. The study found that aggregate phenolic levels increased after 28 days, then declined by the end of the season, with chromogenic tests revealing several allelochemicals in the root exudates.

Keywords: Sunflower (*Helianthus annuus L.*), cultivars, allelopathic effect, phenolic compounds, weed flora and biomass

Key findings: Sunflower cultivars with allelopathic potential have significantly reduced the total population and biomass of companion weeds in the sunflower fields. However, the extent of reduction varied, depending on the genotype of the sunflower cultivar. This action refers to having different allelochemical compounds affecting weeds.

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INTRODUCTION

Weeds are a typical agronomic problem that may reduce crop profitability. Weeds compete with crops for moisture, nutrients, and light in field crops. As a result, weeds reduce either yield or quality. In maintaining productivity, it is critical to keep weeds under control. Herbicide use has increased due to widespread pesticide-resistant weeds (Shibayama, 2001). Agrochemical overuse has resulted in natural pollution, weed resistance, and human health issues. There must be viable weed management frameworks that lessen chemical uses, which are cheap and easy to use, and help protect biological systems (Sarbout *et al.*, 2023).

Chemical control of weeds using pesticides has become effective on weed crops, increasing crop productivity (Rice, 1984). The world may use about three million tons annually of pesticides in various agricultural systems to reduce the impact of weeds on productivity (Stephenson, 2000). However, their use is not scientific, and excessive research has become an issue of concern for individuals and society because these chemical compounds cause significant health and environmental damage. Therefore, the need to find new agricultural practices that would replace pesticides or legalize their use prevails. Using cropping rotations or competitive crops and high densities, in addition to some naturally produced compounds in weed management, can contribute in part to improving the agricultural ecosystem.

The phenomenon of allelopathy is one of the most promising alternatives researchers have focused on for managing weeds and improving crop growth (Chou, 1999; Hozayn *et al.*, 2011). Many researchers have found that allelochemicals released from some crop residues can ably inhibit the germination and growth of weeds under field conditions. Some have suggested possibly using them as environmentally friendly natural pesticides that apply directly in pest control or in developing new pesticides that are less harmful to the environment (Jamil, 2004).

The results studies reached were encouraging, as many researchers found that the aqueous extract of white corn, called Cheema, and its combination with sorgaab and sunflower extract, with doses lower than recommended for some pesticides, led to a significant reduction in weed density and dry weight, which exceeded pesticide effects and the extract alone (Omonov *et al.*, 2023). Such effects appeared in an increase in wheat, maize, cotton, and mung bean yields (Cheema *et al.*, 2000a, b; Khaliq *et al.*, 2002). Therefore, the idea occurred of using the allelopathic phenomenon with pesticides more practically and effectually via allelopathic crop residues left after harvest, mixing them with field soil (Cheema and Khaliq, 2000).

The results in this aspect were impressive, as a significant reduction in weed emergence and growth was visible along with an increase in barley and bean plant yields when treating with sunflower residues with doses lower than the recommended pesticides (Al-Temimi and Alsaadawi, 2010; Alsaadawi *et al.*, 1986). Moreover, adding plant residues to the soil improves its physical, chemical, and biological properties, resulting from increased organic matter (Alsaadawi and Dayan, 2009).

The introduction of sunflower genotypes for growing in Iraq ensued. According to early field assessments, buddy weed growth and population were adaptive across genotypes. Similarly, weeds that emerged in the field following the sunflower harvest showed fluctuating development and population diversity. It demonstrates that allelopathy controls weed formation and population suppression and that differences in allelopathic skills across cultivars may relate to cultivar allelopathic capabilities. The study team chose to test two sunflower genotypes for allelopathic inclination to regulate the growth and population of friendly weeds to find the most allelopathic genotypes and assess the influence of allelopathy on partner weed population and development.

MATERIALS AND METHODS

Experimental location and procedure

The experiments transpired in Wasit, Iraq's State Board of Agricultural Extension Research Station. The field experiment happened during the 2022–2023 season. The soil in the field was characteristic of a calcareous field topsoil. The text does not provide further details about the soil. An experiment proceeded on growing sunflower crops in a silty clay mixture soil, with some of its physical and chemical characteristics estimated in the laboratories of the Soil Department of the College of Agriculture, Wasit University. The natural carbon content, pH level, and electrical conductivity (EC) emerged as deterrents. The previous field history showed a heavy weed infestation. The soil at the experimental site was calcareous clay loam. Organic carbon, pH, and EC were 0.92%, 7.2, and 1.9 dS m⁻¹, respectively. Average annual rainfall is less than 50 mm, and day and night temperatures during the growing season were 30 °C–45 °C.

Identification of weeds

The presence of weeds around sunflowers reached exploring. Specialists from Wasit University's Herbarium and Wasit Province's State Board of Agriculture Extension helped identify them individually.

Companion weeds' effects of sunflower genotypes

The field experiment commenced in the Wasit region, with a location of 180 km south of Baghdad. The field plots were 2.5 m × 2.5 m in size. They were random selection in an overgrown, weedy region. Before planting, furrowing occurred on the plots to a depth of 30 cm using a spade. Applying nitrogen (N), phosphorus (P), and potassium (K) fertilizers proceeded in the field. The nitrogen fertilizer application, in the form of NPK (with 46% N content), was at a rate of 240 kg/ha. Half of the NPK fertilizer application occurred before planting, and the other half at two weeks after planting. Phosphorus treatment as triple

superphosphate (with 46% P₂O₅ content) was at 240 kg/ha. As with the NPK fertilizer, half of the phosphorus treatment ensued before planting and the other two weeks after planting (Cheema and Khaliq, 2000). Two sunflower genotypes, Asgaro and Flamme, continued to plant manually in lines. The spacing between seeds within a line was 25 cm, and the spacing between lines was 50 cm. The chosen field plots used a three-replication randomized complete block design (RCBD). The least significant difference (LSD_{0.05}) test proceeded to compare and separate the treatments. This design helps ensure proper statistical analysis by minimizing the effects of variation and providing reliable results. At the end of the yield development period, harvesting the plants of each sunflower genotype took place. During the summer of 2015, the collection of aboveground biomass of weeds present in the field happened. The weed biomass underwent air-drying for a few days and was then measured using modern technology, calculating the amount of each weed species.

The stair case approach to bioassay root exudates

The collected soil samples from each treatment attained air-drying. The dried samples sustained grinding into a fine powder using a mortar and pestle. Acquiring a known weight of the soil sample (approximately 5 g), continue to place it in a conical flask. The flask received 50 ml of a 1% aqueous solution of Folin-Denis reagent. The flask containing the soil sample and the Folin-Denis reagent underwent a water bath and heated at 80 °C for 30 min. During this time, the phenolic compounds present in the soil reacted with the Folin-Denis reagent to form a blue-colored complex. After the heating period, removing the flask from the water bath allowed it to cool at room temperature. Once cooled, transfer the flask contents into a volumetric flask received diluting with distilled water to a known volume (e.g., 100 ml). The diluted extract attained analysis using a spectrophotometer at a specific wavelength (usually around 760 nm). Measuring the absorbance of the extract ensured using a calibration curve to quantify the concentration

of phenolic compounds in the soil sample (Sarbout and Al-Sadawi, 2015).

Calculating the concentration of phenolic compounds in the soil sample depended on the absorbance reading and the calibration curve. The result's typical expression was milligrams of phenolic compounds per kilogram of soil (mg/kg). The procedure follows: The standard was ferulic acid phenol, an allelopathic compound found in sunflower plants (Haslam, 1988).

At one, four, and six weeks following planting, obtaining soil samples from plots with all treatments was at a depth of 30 cm (WAS). The soils were well mixed and dried for three days at room temperature. Separating 250-g dry soil samples received shaking at 200 rpm in 250 ml of distilled water for 24 h (Ben-Hammouda *et al.*, 1995). Under vacuum, soil suspensions' filtering used a Whatman No. 2 filter paper. Adding Folin-Denis (0.5 ml) and Na₂CO₃ (1 ml) to 1 ml of soil water extract continued to stand for 30 min. A spectrophotometer helped measure absorbance at 750 nm (Blum *et al.*, 1991). The identification of how much total phenolic content used a standard curve with different ferulic acid values. The study isolated, identified, and measured sunflower phytotoxins.

The experiment, laid out in a randomized complete block design (RCBD), had four replications. The collected data's analysis employed Fisher's analysis of variance technique. Applying the least significance difference (LSD) test was at the 0.05 probability level to compare treatment means (Steel and Torrie, 1980).

RESULTS

The study found that wide-leaf varieties comprised 60% of the weeds planted in the field, showing them as *Avena fatua*, *Lolium temulentum*, and *Phalaris minor*. *Beta vulgaris* and *Malva rotundifolia* were also visible in the grasses, including *Ducus carota*. The tested sunflower genotypes seemed to have a non-selective effect. Meanwhile, specific genotypes, such as Asagai, are more successful against

gasses and broad-leaf weeds, and others, such as Flamme, have little impact.

The findings also suggested that weed species reacted differently to the allelopathic capabilities of sunflower genotypes (Table 1). Genotype Asgaro showed an inhibitory effect on all weeds except *Beta vulgaris*. The Flamme genotype had a superior potential to lower the number of all test weeds, except for *Avena fatua*, *Plantago ovata*, and *Beta vulgaris*, acquiring only marginal impacts.

Allelopathic influence against companion weeds

Weed species present

Table 2 indicates the presence of weed species in the sunflower fields across all genotypes. The genotype named Asgaro demonstrated the highest efficiency in reducing the total number of weeds in the field, achieving a reduction of 68.6%. It means that compared with the control (no sunflower genotype), Asgaro significantly suppressed weed growth. On the other hand, the Flamme genotype showed a minimal reduction in the number of weeds, resulting in only a 24.0% loss versus the control. All sunflower test genotypes proved to decrease weed biomass compared with the control. The extent of the reduction varied depending on the genotype. The Asgai genotypes were the most effective in inhibiting weed biomass, reaching 61.0%. In contrast, the other genotypes exhibited lower weed biomass reduction than the control, ranging at 35.4% in Flamme.

Origin secretion of both uniformly held root and shoot development of *Echinochloa colonum* weed compared with the control (Table 2), with the Asagai cultivar having a 68.9% advantage over the Flamme cultivar in the concealment of the entire plant of the test weed (Table 3). The staircase research findings supported the field test results for the two cultivars' origin secretion. Within concealment of origin, shots, and whole plant removal, Asagai outperformed Flamme by 75.8% and 53.3%, respectively (Table 3). In root exudates, no effort occurred to isolate the allelochemicals.

Table 1. Allelopathic effect of sunflower genotype on growth of companion weed under field conditions.

Genotypes	Reduction % Inhibition control	Reduction % Inhibition
Asgaro	50.0	72.4
Flamme	22.4	40.3
Control	-	-

Table 2. Effect of root exudates of sunflower cultivars on growth of *Echinochloa colonum* weed.

Treatments	Dry weight (g)		
	Shoots	Roots	Whole plant
Asgaro	0.07	0.017	0.087
Flamme	0.15	0.070	0.22
Control	0.19	0.090	0.28
LSD _{0.05}	0.037	0.019	-

Table 3. Concentration of total phenolics released from root exudes of sunflower from different periods of decomposition.

Treatments	Total Phenol concentration	
	28 days	52 days
Weedy check (Control)	0.103	0.227
Asgaro	0.403	0.107
Flamme	0.220	0.150
LSD \leq 0.05	0.0869	0.0649

Table 4. Concentration of phytotoxins in the root exudes of test sunflower cultivars.

Phytotoxins	Concentration (ppm)					Total
	Sunflower genotypes					
	Protocatechuic	Vanillic	Syringic Acid	P-Coumaric acid	Ferulic	
Asgaro	127.4	111.2	123.7	109.6	138	609.9
Flamme	147.9	60.0	83.7	77.6	55	424.3

Nonsignificant differences in total phenolics appeared between any of the treatments, including the control (Table 4). The release of total phenolics started after two weeks of farming, peaking at four weeks, and steadily declined until reaching the lowest concentration after six weeks of decomposition. The content of total phenolics in the soil from sunflower root-affected plots was higher, and the concentration rose four weeks after cultivation. Weedy Check demonstrated lower total phenolic content at all breakdown periods.

HPLC tests revealed the presence of five phytotoxins in the soil root systems of the studied sunflower genotypes (Table 5). The scheduled maintenance of all isolated mixtures appeared to be altered, recognizing them as phenolic blends. The profile of each chemical differed depending on the test genotype. The following analysis showed the grouping of the separated mixes as protocatechuic, vanillic, syringic acid, p-coumaric, and ferulic, with the convergence appearing to be substantially higher in Asgaro genotypes than in Flamme genotypes.

DISCUSSION

Weed treatment has prevailed to be beneficial for allelopathy (Manish and Aakash, 2022). The study found that different sunflower genotypes exhibited varying inhibitory effect levels on thin and broadleaf weeds. Asagri genotypes showed a higher inhibitory effect, while Flamme genotypes had a lesser inhibitory effect. The researchers did not investigate the specific reasons for this difference. However, a reason is that the root exudates of sunflower genotypes contain a range of inhibitory compounds with different mechanisms of action. The study recommended probing the inhibitory effects of root exudate compounds alone or in combination to determine whether they have weed-specific effects in understanding further the allelopathic effects. It could help identify specific allelochemicals, such as phenolics, present in sunflower root exudates that contribute to their inhibitory effects on weed growth.

According to Tawifiq and Alsaadawi (2014), some allelochemicals of phenolics, such as neochlorogenic acid, 5-O-P-Coumaroyl quinic acid, chlorogenic acid, caffeoylquinic acid, and neochlorogenic acid acids, were naturally evident. Discoveries have shown that these phenolic acids impede several physiological activities, including respiration, photosynthesis, ion absorption, hormone generation, and cell division (Einhellig, 2004; Yang, 2004). The phenolic dynamic determination in soil confirmed the presence and release of phytotoxins from plant root secretions absorbed into the soil. The release of phenolics started after two weeks of agriculture, peaked at four weeks, remained at a higher concentration for four weeks, and then began to decline until reaching the lowest concentration after six weeks of decomposition (Sarbout *et al.*, 2023). It demonstrates that the principal mechanism of weed control is phenolics produced by sunflower roots in the soil. Accordingly, these results explain why weed development was so slow following cowpea sowing during the first two months. A study reported similar effects when spraying

sorghum residues on a massive bean crop (AL-Bedairy, 2011). No one tried to find and isolate phenolic acids in decomposed sunflower residues in soil.

Al-Temimi and Al-Asaadawi (2010) discovered and isolated phenolic acids, such as chlorogenic acid, isochlorogenic acid, caffeic acid, gallic acid, syringic acid, hydroxybenzoic acid, and p-coumaric acid. These phytotoxins have proven to inhibit a variety of metabolic processes, including chlorophyll biosynthesis (Alsaadawi *et al.*, 1986a; Weir *et al.*, 2004), ion uptake (Olmsted and Rice, 1970; Alsaadawi *et al.*, 1986; Majid *et al.*, 2022), and photosynthesis (Hejl *et al.*, 1993). Likewise, they inhibited plasma H⁺-ATPase activity, resulting in decreased ions and water absorption by guard cells of leaves, causing stomata to close (Hejl and Koster, 2004) and inhibition of photosystem II, which reduces the production of ATP and NADPH₂ required for CO₂ fixation in the dark (Barkosky *et al.*, 2000). Furthermore, phenolic acids have appeared to decrease mitochondrial numbers and modify the membranes surrounding nuclei, mitochondria, and dictyosomes (Lorber and Muller, 1976). According to the study data, one of the ways that sunflower residues inhibit the development of test weeds is by preventing ion absorption. In the soil root secretions of the sunflower genotypes under investigation, HPLC testing confirmed the presence of five phytotoxins.

CONCLUSIONS

The allelopathic potential of the sunflower genotypes under study varies, as does the quantity of aggregated phenolic phytotoxins. According to all accounts, the deposits were in charge of the allelopathic ability of the test genotypes.

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