

Determination of Allelopathic Effects of Residues and Aqueous Extract Two Wheat Cultivars on (*Hordeum bulbosum* L.) and (*Malva parviflora* L.).

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Abstract

The present study was conducted to evaluate the allelopathic effects of wheat residues and their aqueous extracts on the germination, seedling emergence, and dry weight of *Hordeum bulbosum* (wild barley) and *Malva parviflora* (mallow). The results revealed that the aqueous extracts of wheat residues exhibited significant inhibitory effects, particularly those of the Jihan cultivar. At a concentration of 20% of the shoot residues, Jihan extracts completely inhibited (100%) the growth of *M. parviflora*. Residues also suppressed seedling emergence by up to 42.8% in *M. parviflora*, which proved to be more sensitive than *H. bulbosum* (14.8% inhibition). In terms of biomass reduction, the dry weight of *H. bulbosum* decreased by 45.7% with residues of the Abaa-99 cultivar, while *M. parviflora* recorded the highest reduction (81.6%) under Jihan residues. Both species also exhibited varying degrees of inhibition in plumule and radicle elongation depending on the cultivar, with Jihan shoot residues exerting the strongest suppressive effects across most treatments.

Keywords: wheat (*Triticum aestivum*), cultivars, allelopathic effect, phenolic compounds, Weed management, *Hordeum bulbosum*, *Malva parviflora*

I. Introduction

The allelopathic effect arising from wheat residues is recognized as inhibitory towards certain plants and weeds, while simultaneously exhibiting stimulatory potential for the growth of other plants or subsequent crops within the same field. This phenomenon occurs through the release of biochemical compounds into the external environment, influencing neighboring plants or soil microorganisms (Aslam et al., 2017). Within this context, allelopathy is defined as a biological process whereby living organisms including plants and microorganisms such as fungi and bacteria release biochemical compounds into their surrounding environment. These compounds exert influence on various physiological factors, including germination, establishment, growth, and reproduction (Andrew, Storkey, & Sparkes, 2015; Hussain & Reigosa, 2021)



Weeds have caused significant damage to agricultural crops, with field losses reaching 5% of total production in developed countries, 10% in less developed countries, and up to 25% in developing countries. As the threat of weeds has increased, the need for chemical pesticides has also risen, leading to a sharp increase in the use of herbicides in recent decades. It has been found that 37% of chemical pesticides are herbicides and 24% are insecticides. This excessive use has led to the recording of 332 herbicide-resistant biotypes, including 189 plant species (113 broad-leaved and 76 grasses) in more than 300,000 agricultural fields worldwide. (Fishel, 2007). Despite the widespread use of chemical herbicides, their environmental impacts, rising costs, and the emergence of resistant weed species necessitate alternative, sustainable approaches for weed management. (Parven et al., 2024).

The allelopathic potential has been demonstrated in several crop plants (wheat, rye, maize), as they have been found capable of synthesizing and releasing low molecular weight compounds into their surrounding environment, which affect the growth and development of weeds. Wheat is considered one of the most important cereal food crops with high allelopathic content against weeds, as studies have shown the presence of allelochemicals from root exudates and straw residues. Among the most important compounds identified include hydroxamic acids, polyphenols and flavonoids, several phenolic compounds and flavonoids, which have also been detected in different wheat varieties. (Osterholz et al., 2020; Kashif et al., 2015; Fragasso et al., 2013).

This study is based on the hypothesis that wheat residues, through their allelopathic potential, can significantly inhibit the germination and growth of specific weed species, offering a natural method **for weed suppression**.

The objectives of this study are:

- Assess the allelopathic effects of wheat residues (root and shoot) incorporated into soil on the growth of (*Hordeum bulbesum*) and (*Malva parviflora*) in plastic pots under a shade net.
- Investigate the inhibitory effects of aqueous extracts of wheat residues on seed germination of (*H. bulbesum*) and (*M. parviflora*) using in vitro bioassays in Petri dishes.
- Conduct a comparative study on the impact of varying residue concentrations on embryonic root and shoot growth in both target species to identify effective thresholds for potential integration into integrated weeds management programs.

II. Materials and Methods

Plant materials

One of the agricultural fields in Wasit province was prepared for the 2024 growing season. The field was subdivided into two plots and sown with two wheat cultivars (Jihan and Abaa-99). After two months of growth from germination, the aboveground shoot and belowground root plant parts were harvested. The samples were carefully washed to remove adhering soil particles, oven-dried, ground to a fine powder, and subsequently prepared for extraction procedures.



Additionally, seeds of the weed species (*Hordeum bulbesum*) and (*Malva parviflora*) were obtained from agricultural fields in proximity to Wasit University.

Preparation of aqueous extracts

Respective grinded plant parts were weighed Three different weights (5, 10, 20 g) were each separately mixed with 100 mL of distilled water and soaked for 24 h at room temperature. The mixtures were then blended using an electric blender for 5 minutes, the mixture was then filtered using Whatman No. 42 filter paper to be used fresh in the bioassay, this procedure yielded three different extract concentration 5%, 10%, 20%.

Shade-house experiment

The wheat shoot and root residues were ground and mixed with soil at two concentrations (3 and 6 g) of shoot and root residues separately. The mixtures were placed in plastic pots containing 1200 g of soil. Control treatments were also used by adding peat moss at the same concentrations as the residues to ensure equal amounts of added material.

Germination tests/bioassay

Seeds were sown in the pots at a rate of 20 seeds per species, with three replicates for each treatment. The pots were kept in a shade house and watered as needed with equal amounts of running water. Ten days after sowing, the percentage emergence of weed seedlings was recorded. Additionally, the emerged weed seedlings were thinned to five seedlings per pot and left to grow for 67 days. After this period, whole plants were carefully removed, washed, and dried. The dry weights (mg) of the shoot and root parts were then measured.

For the extract experiments, 20 weed seeds were placed individually in Petri dishes lined with filter paper. Three replicates were used for each concentration (5%, 10%, 20%). 10 mL of the extract were added to each dish, and rehydration was performed as needed using the respective extracts. After 7 days, the germination percentage was recorded. (Ghaleb et al. 2022)

Radicle and plumule lengths(cm) were recorded 15 days after the standard germination period, five seedlings were randomly selected from each dish. The radicle was separated at its point of attachment to the seed, and the plumule was separated from the hypocotyl. Their lengths were measured using a ruler (AOSA, 1983; Association of Official Seed Analysts).

Statistical analysis

were collected, summarized, analyzed and presented using statistical package for social sciences (SPSS) version 26 and Microsoft Office Excel 2010. Numeric data were presented as mean, standard deviation after performance of Kolmogorov-Smirnov normality test and making decision about normally and non-normally distributed variables. One way anova test was used to study difference in mean among more than two groups provided that the variable is normally distributed. The level of significance was considered at P-value of less 0.05 and highly significant level at 0.01 or less (Daniel, 2018).



III. Results and Discussion

Table 1 presents the allelopathic effects of wheat residues on the dry weight of (*Hordeum bulbesum*). The effects of both shoot and root residues from the two wheat cultivars varied considerably. Root residues of the cultivar Abaa-99 at a concentration of 6 g exhibited the strongest inhibitory effect on the dry weight of the aboveground parts, with an inhibition percentage of 45.7%, while shoot residues of the cultivar Jihan resulted in 39.7% inhibition. The inhibitory effects of the other treatments ranged between 30.4% and 13.9%.

Notably, shoot residues of Abaa-99 at 3 g demonstrated a stimulatory effect, increasing the dry weight by 12.8% compared to the control. Regarding root dry weight, residues of Abaa-99 produced the highest stimulatory effect, increasing the dry weight of wild barley roots by 37.1%, with all these effects being statistically significant. In contrast, the remaining treatments of the cultivar Jihan exerted inhibitory effects ranging from 13.5% to 38.5%, whereas the lowest inhibitory effect for Abaa-99 root residues was 26.5%.

Table 1. Allelopathic effect of residue wheat cultivars on growth of (*Hordeum bulbesum*) weed.

Treatments	Dry weight (mg)					
	Shoots		Roots		Whole plant	
	3g	6g	3g	6g	3g	6g
Jihan shoot	789.7	800.7	442.3	478.0	1232.0	1278.7
Jihan root	817.3	924.7	348.7	461.7	1166.0	1386.4
Abaa-99 shoot	861.7	1126.3	438.0	934.7	1299.7	2061.0
Abaa-99 root	1144.7	721.3	486.7	406.7	1631.4	1128.0
Control	993.0	1328.0	566.3	553.8	1559.3	1869.3
LSD ≤ 0.05	Residue shoot	Residue	Residue shoot	Residue	Residue shoot	Residue
	root		root		root	
Concentrations	170	102.7	80.8	111.3	163.5	170.2
Cultivars	164	123.2	96.3	119.2	198.8	202.1
Interaction	201	151.4	112.3	145.5	223.1	218.8

Table 2 presents the effects of wheat residues on *Malva parviflora* (mallow). The results show that most treatments significantly affected the dry weight. Shoot residues of the cultivar Jihan at a concentration of 6 g caused the highest reduction in shoot dry weight, with an inhibition percentage of 81.6%, while the inhibitory effects of the other treatments on the aboveground parts ranged between 44.7% and 63.1%.



Regarding root dry weight, shoot residues of Jihan again exhibited the strongest inhibitory effect, reaching 83.6% inhibition. In the case of Abaa-99, the effects were relatively uniform, with an average inhibition of 53.8% compared to the control.

Table 2. Allelopathic effect of residue wheat cultivars on growth of (*Malva parviflora*) weed.

Treatments			Dry weight (mg)			
	Shoots		Roots		Whole plant	
	3g	6g	3g	6g	3g	6g
Jihan shoot	146.7	150.0	28.3	31.3	175.0	181.3
Jihan root	220.7	456.3	53.0	95.0	273.7	551.3
Abaa-99 shoot	219.0	133.0	54.0	87.3	273.0	220.3
Abaa-99 root	2403	252.0	54.0	74.3	294.3	326.3
Control	594.0	815.3	118.0	192.7	712.0	1008.0
LSD \leq 0.05	Residue shoot	Residue	Residue shoot	Residue	Residue shoot	Residue
	root		root		root	
Concentrations	96.3	112.3	30.3	28.4	155.1	130.3
Cultivars	116.4	134.4	52.3	42.9	188.3	174.8
Interaction	163.2	160.8	71.7	61.5	231.3	194.3

Table 3 shows that all treatments exhibited highly significant effects, as the aqueous extracts of wheat residues inhibited seed germination of both *Hordeum bulbesum* and *Malva parviflora*.

For wild barley, the strongest inhibitory effect was recorded for the shoot residues of the cultivar Jihan at a concentration of 20%, resulting in an 85.2% reduction in germination. This inhibition decreased to 56.7% at 10%. The remaining treatments of Jihan ranged between 27.3% and 23.1%.

For the cultivar Abaa-99, the shoot extract at 20% reduced germination by 56.8%, while the 10% concentration reduced it by 46.3%. The other treatments of this cultivar showed inhibition values ranging from 35.7% to 20.1% compared to the control.

In *M. parviflora*, significant inhibition was also observed for most treatments, except for the 5% concentration of Abaa-99. The strongest inhibition was achieved by the foliar residues of Jihan at 20%, with 100% inhibition (0% germination). At 10%, the inhibition percentage decreased to 83.2%. Other Jihan treatments showed inhibition values between 81.8% and 53.9%.

For Abaa-99, the highest effect was observed at 20% with 58.3% inhibition, whereas the lowest effect was recorded for the foliar and root extracts at 5%, with inhibition values of 3.4% and 5.1%, respectively. The other extracts showed relatively similar inhibition levels of around 45.2% compared to the control.



Treatments	<i>Hordeum bulbesum</i>				<i>Malva parviflora</i>			
	5%	10%	20%	Mean	5%	10%	20%	Mean
Jihan shoot	73.5	41.5	13.5	42.5	23.5	10.0	0.0	11.1
Jihan root	69.30	70.0	71.5	66.5	28.6	13.5	10.0	17.3
Abaa-99 shoot	61.5	51.5	41.5	51.5	58.5	35.5	30.0	41.1
Abaa-99 root	70.0	68.5	76.5	71.5	51.5	38.5	25.0	38.5
Control	95.0	95.0	95.0	95.0	60.0	60.0	60.0	60.0

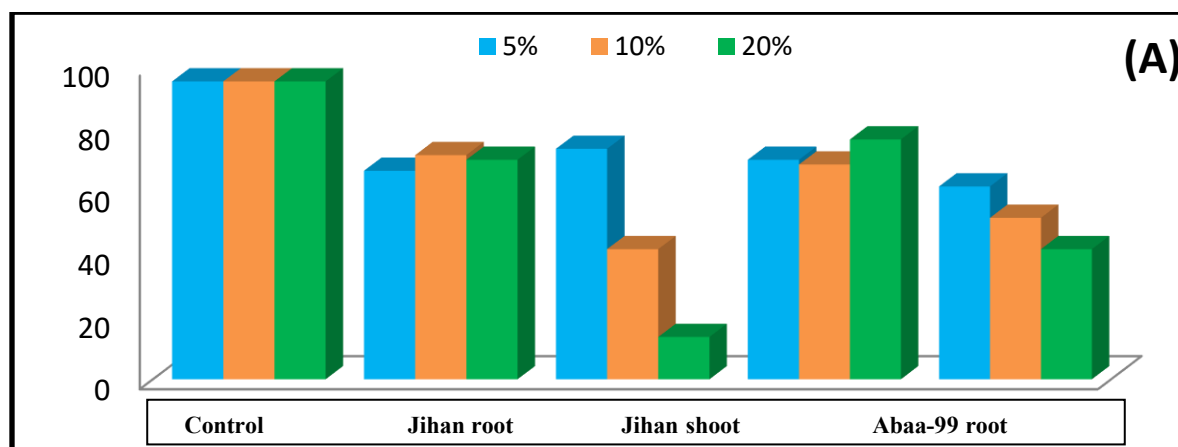
LSD ≤ 0.05

Concentrations 7.78 11.23

Cultivars 8.41 14.33

Interaction 11.82 17.21

Table 3: Allelopathic effect of (*Triticum aestivum* L.) aqueous extract on germination percentage of test weeds on Petri dishes.



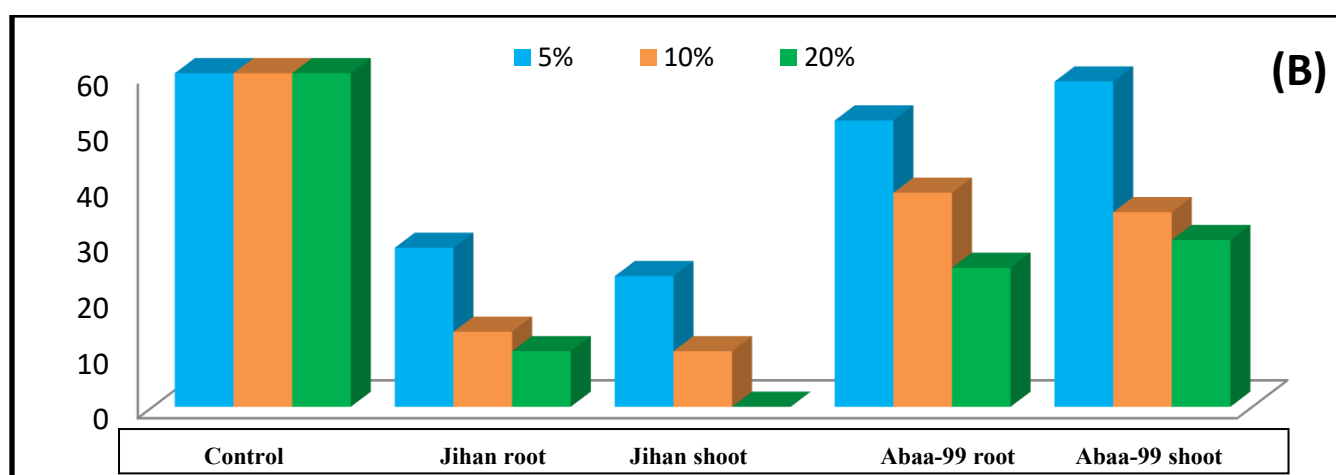


Figure 1. Allelopathic potential of (*Triticum aestivum* L.) extract against test species :(A) germination percentage of (*Hordeum bulbesum*) and (B) germination percentage of (*Malva parviflora*)

Table 4 presents the allelopathic effects of wheat residues on the emergence percentage of *Hordeum bulbesum* and *Malva parviflora*.

For *H. bulbesum*, most treatments had no significant effect. The shoot residues of both cultivars showed inhibition levels of 14.8%, except for the foliar residues of Abaa-99 at 6 g, which exhibited a weak inhibition of 2.4%. In the root residues, Abaa-99 at 6 g showed a stimulatory effect of 11.2%, consistent with the increase in dry weight observed in the previous table, while the 3 g treatment was equivalent to the control. The root residues of Jihan produced similar inhibitory effects at both concentrations, ranging from 18.1% to 22.3%.

For *M. parviflora*, the foliar residues of Jihan caused a 42.8% inhibition at 6 g, whereas the 3 g treatment resulted in a much lower inhibition of 11.7%. In Abaa-99, the 3 g foliar residues enhanced emergence by 16.3%, while the higher concentration was comparable to the control. Root residues of both cultivars produced relatively similar inhibitory effects, ranging from 39.2% to 40.1%, compared to the control.

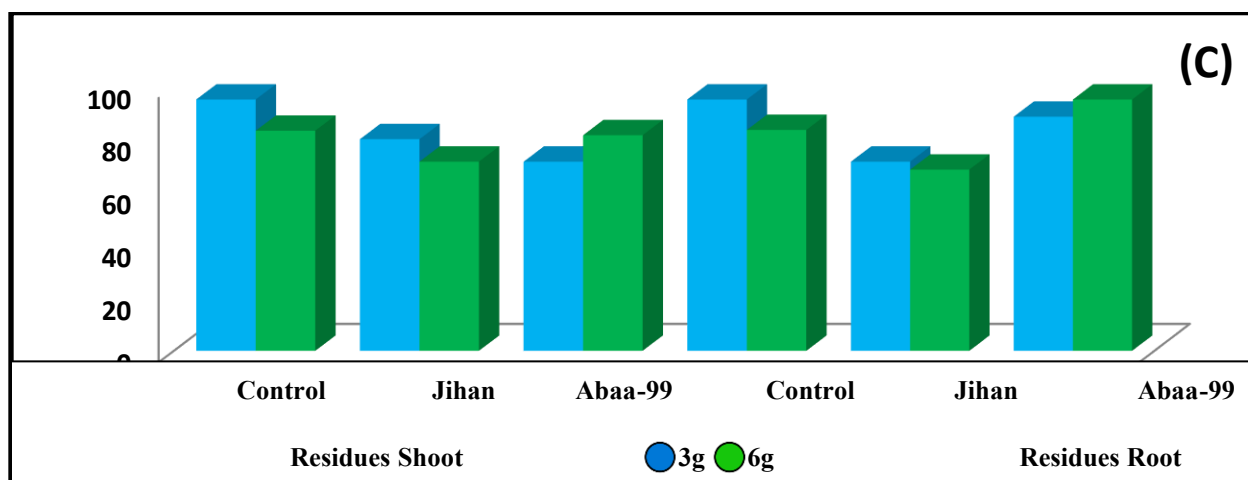
Treatments

	<i>(Hordeum bulbesum)</i>				<i>(Malva parviflora)</i>			
	Residues Shoot		Residues Root		Residues Shoot		Residues Root	
	3g	6g	3g	6g	3g	6g	3g	6g
Jihan	80.0	71.5	71.5	68.5	45.0	40.0	30.0	41.5
Abaa-99	71.5	81.5	88.5	95.0	61.3	46.5	33.5	35.0
Control	94.0	83.2	94.0	83.2	51.5	70.0	51.5	70.0

LSD ≤ 0.05

Concentrations	8.03	5.91	11.12	10.33
Cultivars	10.3	9.17	7.12	13.21
Interaction	13.22	11.81	13.39	16.20

Table 4: Allelopathic effect of wheat (*Triticum aestivum* L.) on germination percentage of test species on soil .



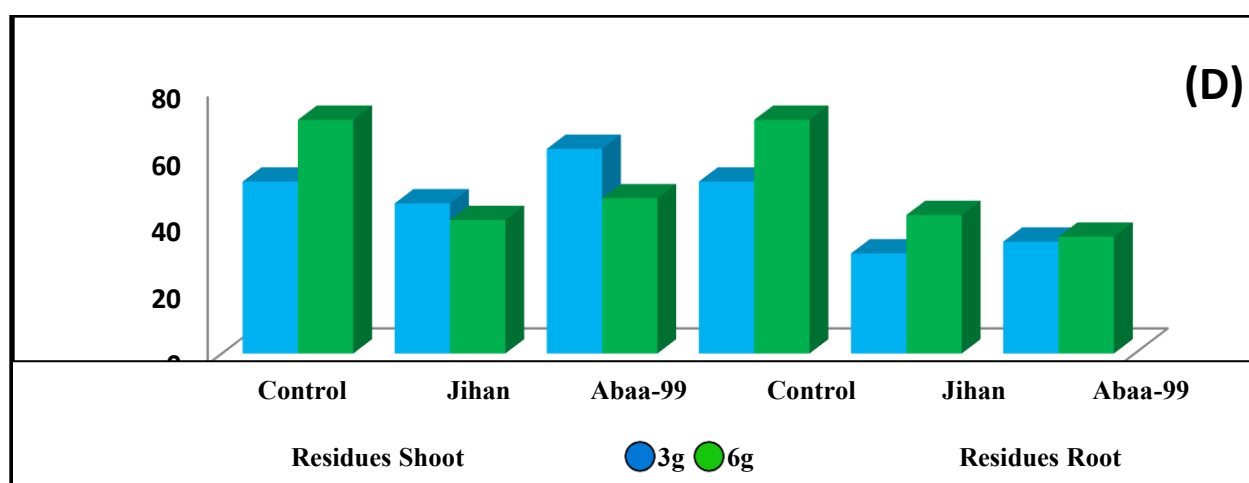


Figure 2: Allelopathic potential of (*Triticum aestivum* L.) residues (3 g ,6g) against test species : (C) germination percentage of (*Hordeum bulbosum*) and (D) germination percentage of (*Malva parviflora*)

In **Table 5**, the data clearly demonstrate that all wheat extracts induced highly significant allelopathic suppression of plumule elongation in both *Hordeum bulbosum* and *Malva parviflora*.

In *H. bulbosum*, the shoot extract of cultivar Jihan exerted the most pronounced inhibitory effect, reducing plumule length by 92.2% at the 20% concentration and 62.06% at 10%. The remaining 'Jihan' treatments caused moderate inhibition, ranging from 17.2% to 45.6% relative to the control. Conversely, cultivar Abaa-99 exhibited comparatively milder allelopathic activity, with its highest reduction (57.3%) recorded for the shoot extract at 20%, which decreased to 39.6% at 10%. Other concentrations produced inhibition levels between 7.8% and 36.2%.

In *M. parviflora*, the shoot extract of Jihan completely suppressed plumule elongation at the 20% concentration, matching its total inhibition of germination percentage, while the 10% and 5% treatments reduced plumule length by 51.3% and 36.5%, respectively. The root extracts of Jihan showed intermediate activity, with reductions ranging from 29.2% to 41.4%. For cultivar Abaa-99, the strongest inhibitory effect on *M. parviflora* was observed for the shoot extract at 20% (58.5% reduction), while the remaining treatments varied between 9.6% and 45.1% inhibition.

Treatments	<i>Hordeum bulbosum</i>				<i>Malva parviflora</i>			
	5%	10%	20%	Mean	5%	10%	20%	Mean
Jihan shoot	6.25	4.38	0.91	3.85	2.65	2.03	0	1.56
Jihan root	9.63	7.78	6.31	7.91	2.89	2.96	2.39	2.75
Abaa-99 shoot	9.64	6.98	4.93	7.18	3.72	3.04	1.79	2.85
Abaa-99 root	10.73	8.69	7.42	8.95	3.08	2.92	2.23	2.74



Control	11.62	11.62	11.62	11.62	4.13	4.13	4.13	4.13
LSD ≤ 0.05								
Concentrations			1.26				0.60	
Cultivars			1.47				0.45	
Interaction			2.01				0.88	

Table 5: Allelopathic effect of (*Triticum aestivum* L.) aqueous extract on plumule length (cm) of test species in Petri dishes lined with filter paper.

In **Table 6**, the effects of wheat extracts from the cultivars Jihan and Abaa-99 on radicle length of *Hordeum bulbosum* and *Malva parviflora* are presented. Significant inhibitory effects were recorded for both species.

For *H. bulbosum*, the shoot extract of Jihan at 20% concentration produced the highest reduction (72.1%), while at 10% and 5% the reductions decreased to 52.5% and 44.3%, respectively. The root extracts at 10% and 5% exhibited similar inhibition levels (31.9%), which increased to 46.3% at 20%. In contrast, Abaa-99 exerted comparatively weaker allelopathic activity, with the highest inhibition (58.1%) observed for the 20% shoot extract, decreasing to 48.4% at 10% and 21.6% at 5%. The root extracts of Abaa-99 displayed a non-linear pattern, showing 31.3% inhibition at 5%, which dropped sharply to 9.2% at 10% possibly reflecting an equilibrium between active compounds before slightly increasing to 10.3% at 20%, suggesting a reactivation of allelopathic constituents at higher concentration.

In *M. parviflora*, all treatments caused highly significant inhibition, consistent with the patterns observed for plumule length and germination percentage. The shoot extract of Jihan at 20% completely suppressed radicle growth (100% reduction), while the 10% and 5% concentrations reduced growth by 66.2% and 56.1%, respectively. Root extracts of Jihan inhibited radicle length by 62.8% at 20%, 49.6% at 10%, and 37.6% at 5%. For Abaa-99 the foliar extract at 20% reduced radicle length by 63.1%, whereas the strongest effect overall for this cultivar was observed for the 20% root extract (64.6% reduction). The remaining concentrations produced inhibition ranging from 9.1% to 40.2% relative to the control.

Treatments	<i>Hordeum bulbosum</i>				<i>Malva parviflora</i>			
	5%	10%	20%	Mean	5%	10%	20%	Mean
Jihan shoot	5.39	4.60	2.68	4.22	2.21	1.72	0	1.31
Jihan root	6.81	6.39	5.25	6.15	3.12	2.52	1.86	2.50
Abaa-99 shoot	7.61	4.92	4.06	5.53	3.33	2.91	1.87	2.71
Abaa-99 root	6.66	8.87	8.76	8.10	4.55	3.20	1.77	3.17
Control	9.76	9.76	9.76	9.76	5.02	5.02	5.02	5.02

LSD ≤ 0.05

Concentrations

1.24

0.81



Cultivars	1.81	0.93
Interaction	1.52	1.21

Table 6: Allelopathic effect of (*Triticum aestivum* L.) aqueous extract on radical length (cm) of test species in Petri dishes lined with filter paper.

Table 7 presents the analysis of wheat extracts using HPLC, where eight compounds were identified. Six of them were phenolic compounds, namely: gallic acid, Syringic acid, hydroxybenzoic acid, p-coumaric acid, ferulic acid, and vanillic acid, while two were flavonoids, namely: rutin and kaempferol. The results revealed that the highest concentration among the phenolic compounds was recorded for gallic acid in the vegetative extract of the cultivar Jihan, reaching 88.91 ppm. In contrast, the highest concentration among all extracts was observed for the flavonoid rutin, with a value of 90.25 ppm. Several studies have demonstrated that these compounds possess strong allelopathic activity against weeds and microorganisms. (Li et al. 2024; Alghamdi et al., 2022; Canarini et al.2019; Ahmed et al. (2021).

Allelochemicals	Concentration (ppm) wheat cultivars			
	Jihan shoot	Jihan root	Abaa-99 shoot	Abaa-99 root
Gallic acid	88.9	84.23	74.58	74.12
Syringic acid	83.6	50.11	70.33	40.6
Hydrobenzoic acid	53.23	71.45	42.65	60.22
p-coumaric acid	62.58	65.41	50.22	54.12
Ferulic acid	80.4	42.66	70.32	30.24
Rutin	60.22	90.25	50.56	75.66
Vanillic acid	74.89	55.08	67.11	46.25
Kaempferol	70.22	80.11	60.32	70.25
Total	573.84	539.30	486.09	451.46

Table 7: High-Performance Liquid Chromatography (HPLC) Analysis of Alcoholic Extract Concentrations from Wheat Residues.

IV. Discussion

The allelopathic effects of wheat residues (roots and shoot) on accompanying weeds have been studied, and the results showed a clear variation between the two cultivars. While the Jihan cultivar exhibited a high allelopathic potential, the Abaa-99 cultivar showed a noticeably lower inhibitory effect. This variation is attributed to differences in the chemical composition of root exudates and the content of inhibitory compounds, particularly phenolic acids.



The literature confirms that wheat possesses a tangible allelopathic capacity, with modern cultivars differing in the levels of this trait. Several allelopathic compounds have been identified in wheat, including phenolic acids, hydroxamic acids, and short-chain fatty acids, which play a key role in inhibiting weed growth. (Ahmed et al. 2021 ; El-Shora et al., 2022).

The findings of this study indicate that a principal mechanism of weed suppression by these compounds involves their direct detrimental effect on ion absorption, culminating in ionic imbalance within plant cells and a consequent disruption of essential metabolic functions (Altameme et al., 2015 ; Hierro & Callaway, 2021). At the physiological level, treatments utilizing wheat extracts and residues induced significant reductions in weed seed germination rates and notable decreases in biomass, as quantified by dry weight measurements. A pronounced inhibition in the growth of both coleoptiles and radicles was also evident, suggesting a direct influence of phenolic compounds on the processes of cell division and elongation, particularly during the early growth stages that are most susceptible to chemical interference. These results are consistent with the established role of allelochemicals, which are known to contain endogenous growth regulators that directly modulate auxin (IAA) concentrations in plant tissues, leading to the suppression of germination and inhibition of seedling development (Singh et al., 1999; Hanwen Wu, 2003). corroborating these findings, wheat residues have been documented to severely affect weed germination, in certain instances achieving complete (100%) inhibition, alongside significant reductions in coleoptile and radicle length (Altameme et al., 2015). Collectively, these results substantiate the hypothesis that wheat residues, particularly from the Jihan cultivar, can be an effective tool for sustainable weed management by harnessing their inherent allelopathic properties. Further research is warranted to isolate and precisely identify the active compounds involved and to elucidate their specific molecular and physiological modes of action, thereby facilitating the development of novel, environmentally benign natural herbicides (Li et al., 2010; Sarbout et al., 2024).

V. Conclusions

The germination percentage, plumule and radicle lengths, as well as dry weight of the tested seedlings of weeds (*Hordeum bulbesum*) and (*Malva parviflora*). were significantly reduced, particularly in response to the aqueous extracts of the shoot residues of the Jihan wheat cultivar. The inhibitory effect increased proportionally with the concentration of the plant material used, and these findings are consistent with those reported by other researchers. The study revealed that wheat residues contain polar, water-soluble chemical compounds that significantly affected weed growth. Based on the current findings, it can be concluded that certain toxic allelochemicals are present in the examined plant parts, and are responsible for the inhibition of weed growth and development. Future studies are recommended to isolate and identify these compounds, which could potentially be utilized in the development of natural herbicides.

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