



RESEARCH PAPER

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Assessment of allelopathic effects of some cruciferous plants on field dodder seed germination and eggplant seedling growth

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Abstract

This study was conducted in both field and greenhouse settings. The experiment featured five replications and followed a randomized plot design and was replicated twice in both 2020 and 2021. In the experiment, cruciferous plant extracts were applied at concentrations of 2%, 6%, and 10% in pots, each containing 20 field dodder seeds and a single eggplant seedling. The purpose of this study was to assess the allelopathic effects of these extracts on various parameters, including field dodder seed germination, field dodder fresh biomass, eggplant height, the total number of eggplant branches per plant, and the number of infected eggplant branches per plant. Additionally, the cruciferous plant samples underwent GC-MS analysis to determine their content of isothiocyanate compounds. The cruciferous plant extracts did not negatively affect eggplant height or the number of eggplant branches per plant. However, all the cruciferous plant extracts led to a reduction in the field dodder seed germination rate. The treatment involving turnip extract at a 10% concentration demonstrated the highest efficiency, resulting in a reduction of 55.72% in the field dodder seed germination rate. Moreover, it is noteworthy that the allelopathic effects of cruciferous plants increased in tandem with increasing extract concentrations. The results of the GC-MS analysis indicated that turnip exhibited the highest percentage of isothiocyanate compounds among the cruciferous plants, accounting for 56.6% of the total, whereas black radish exhibited the lowest percentage at 29.2%.

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Introduction

Cuscuta campestris Yunck., commonly known as field dodder, represents a parasitic plant categorized in the Convolvulaceae family (Yuncker, 1932). Field dodder, lacking chlorophyll, relies entirely on host plants for essential nutrients and water, presenting a significant obstacle to crop cultivation (Costea and Stevanovic, 2010). The adaptability of field dodder and its ability to infest a wide range of plant species highlight its considerable challenge as a weed in agricultural settings. This emphasizes the need for the formulation of effective management strategies to minimize its detrimental impact on crop production (Nadler-Hassar and Rubin, 2003; Ustuner, 2018).

Many studies have reported that the presence of field dodder in crops belonging to the Solanaceae family, such as eggplant, tomato, potato, and pepper, has led to yield reductions ranging from 50% to 90% (Nadler Hassar and Rubin, 2003; Lian *et al.*, 2006; Ustuner, 2020). Furthermore, previous research outcomes have documented diverse levels of yield decrease resulting from field dodder infestations, encompassing a 57% reduction in alfalfa (Dawson *et al.*, 1994), a 30% reduction in carrot (Konieczka *et al.*, 2009), and significant yield reductions of 86% in chickpeas and 87% in lentils (Mishra, 2009).

Numerous studies have recorded the potent allelopathic characteristics of cruciferous plants, specifically those belonging to the *Brassica* genus. These plants are recognized for releasing isothiocyanates, chemical compounds well-known for their toxicity to various soil-borne pathogens, nematodes, fungi, and weeds (Uremis *et al.*, 2009; Jabran *et al.*, 2015; Cipollini, 2016). Under controlled laboratory conditions, it was observed that the utilization of a 2% concentration of arugula extract led to a 70% decrease in the seed germination of *Sonchus oleracens* and a 62.5% reduction in *Sorghum halepense* seed germination (Shaker *et al.*, 2010). Additionally, it has been observed that the utilization of radish extracts successfully inhibited the germination of 11 weed species, including *Cuscuta* spp., *S. halepense*, and *Convolvulus arvensis* L. (Uygur *et al.*, 1991). In a particular study, the

allelopathic influence of five cruciferous plant species (white cabbage, red cabbage, broccoli, turnip, and arugula) was evaluated at concentrations of 2%, 5%, 10%, and 20% in relation to the germination of *S. halepense* seeds. The results demonstrated that arugula extract, notably at concentrations of 10% and 20%, entirely prevented seed germination (Elsekran *et al.*, 2023). The allelopathic impact of cruciferous plants within their biosphere is linked to isothiocyanate compounds, recognized as a significant hydrolysis outcome of glucosinolates (GLSs) (Wittstock and Halkier, 2002).

Glucosinolates (GLSs) are chemically inactive compounds subject to hydrolysis facilitated by the enzyme myrosinase, also known as beta-thioglucosidase. This enzymatic action results in the creation of biologically active chemical compounds. The specific hydrolysis products of GLSs exhibit variability contingent upon the prevailing degradation conditions (Bones and Rossiter, 1996). Enzymatic processes initiate the liberation of glucose and the formation of an unstable compound identified as aglucon-thiohydroxymate-O-sulfate. The conversion of aglucon into various classes of degradation compounds, such as isothiocyanates, thiocyanates, nitriles, epithionitriles, hydroxynitriles, oxazolidine-2-thiones, or indoles, is contingent upon factors such as the structural characteristics of the hydrolyzed glucosinolates (GLSs), pH levels, temperature, the presence of Fe²⁺ ions, and the presence of additional protein agents within the hydrolytic environment (Wittstock and Halkier, 2002). Isothiocyanate (ITC) compounds represent notable byproducts resulting from the hydrolysis of Glucosinolates (GSLs) and are recognized for their considerable biochemical reactivity. While ITCs exhibit diversity in both their chemical properties and taxonomic classification, they share a distinctive structural feature characterized by the presence of a carbon-nitrogen-sulfur double bond (Martins *et al.*, 2004).

The primary objective of this study was to assess the allelopathic effects of extracts derived from turnip, arugula, broccoli, and black radish.

Various parameters were evaluated, including field dodder seed germination, field dodder fresh biomass, eggplant height, the total number of eggplant branches per plant, and the number of infected eggplant branches per plant. Additionally, the cruciferous plant samples underwent GC-MS analysis to determine their isothiocyanate compounds content.

Materials and methods

The experiment was conducted in the trial area of the East Mediterranean Transitional Zone Agricultural Research Institute in Kahramanmaras and the Faculty of Agriculture greenhouses at Kahramanmaras Sutcu Imam University, Turkey in 2020 and 2021.

Preparation of plant materials

The experiment utilized field dodder seeds collected from infected eggplant fields in Kahramanmaras province in Turkey during the 2019 and 2020 seasons. On December 18th, 2019, and December 20th, 2020, broccoli seedlings were cultivated in the field. Turnip, arugula, and black radish seeds were sown on February 10th, 2020, and February 11th, 2021. The cruciferous plants were sampled during the head formation stage of broccoli and the flowering stage of turnip, arugula, and black radish by collecting the entire plants, including the roots, stem, leaves, and flowering and fruiting parts. These samples underwent cleaning and drying at room temperature ($25 \pm 2^\circ\text{C}$). Once the samples were completely dried, they were ground separately with a coffee grinder, and the powders were stored in plastic bags at room

temperature. Table 1 displays the varieties of cruciferous plants and eggplant used in the experiment.

Preparation of cruciferous plant extracts

To prepare the extracts, from each sample of cruciferous plant powders, weights of 20, 60, 100 g were taken, and each unit of weight was mixed with 1 L of distilled water to obtain concentrations of 2, 6, and 10%, respectively. The prepared extracts were placed in a shaker at 200 rpm and $25 \pm 2^\circ\text{C}$ for 8 hours. Then the mixtures were filtered using Whitman filter paper to separate all impurities.

Cruciferous plant samples were meticulously prepared for GC-MS analysis at the laboratory of the Plant Protection Department at Kahramanmaras Sutcu Imam University, following the methodology outlined by Vaughn and Berhow (2005). Initially, 10 grams of each plant powder were precisely weighed, and these powder samples underwent a 24-hour defatting process using hexane in a Soxhlet extractor. Following this, the plant powders were thoroughly dried in a fume hood. The defatted powders were then combined with 30 ml of 0.05M potassium phosphate buffer in separate flasks. To each mixture, 50 ml of dichloromethane was added, and the flasks were agitated at 25°C and 200 rpm for 8 hours. Subsequently, 10 g of NaCl and 10 g of Na_2SO_4 were introduced and homogeneously mixed. The resulting mixtures underwent filtration with additional dichloromethane. The resulting dichloromethane solutions were transferred to labeled ampoules according to the respective plants for GC-MS analysis.

Table 1. Species and varieties of cruciferous plants and eggplant used in the experiment

Plant species	Scientific name	Plant varieties
Turnip	<i>Brassica rapa</i> L. subsp. <i>rapa</i>	Kahramanmaras Salgam
Arugula	<i>Eruca vesicaria</i> subsp. <i>sativa</i> Mill.	Istanbul Rokasi
Broccoli	<i>Brassica oleracea</i> L. var. <i>italica</i>	Sakura F1
Black radish	<i>Raphanus sativus</i> L. var. <i>niger</i> J. Kern	Siyah Inci
Eggplant	<i>Solanum. melongena</i> L.var. <i>esculentum</i>	Adana eggplant

Experiment design and methodology

The experiment followed a randomized plot design with a total of five replications. The dormancy of field dodder seeds was broken by exposing seeds to concentrated sulfuric acid (98%) for 5 minutes then washing them with running water for 10 minutes

(Almhemed *et al.*, 2020). To implement the experiment, plastic pots with an outer diameter of 21 cm, height of 19 cm, and a base diameter of 12 cm for each one were used. For the pots soil, a mixture comprising equal parts 1:1:1 of soil, sand, and compost was prepared, with sterilization

accomplished using a 10% formaldehyde solution. Subsequently, the soil was allowed to undergo a ventilation period of 10 days before being placed into the pots. In each pot one eggplant seedling and 20 seeds of field dodder were populated. To each pot 300 ml of the prepared extracts were added adhering to the experimental design. For the control pots, 300 ml of distilled water was added instead. Throughout the greenhouse experiment, the maximum and minimum temperatures were recorded within the range of 18-33 °C, while the relative humidity was maintained at 70%. The experiment spanned duration of 45 days, during which regular water was added to the pots as required.

The effect of cruciferous plant extracts and concentrations on the field dodder seeds germination

Germination of field dodder seeds was monitored daily for two weeks, and the germination rate was calculated using the following formula:

$$\text{Germination rate (\%)} = (\text{Number of germinated seeds} / \text{Total number of seeds}) \times 100$$

To evaluate the efficiency of cruciferous plant extracts in reducing the germination rate of field dodder seeds, the following formula was used:

$$\text{Efficiency (\%)} = [1 - (G_x/G_o)] \times 100$$

Where, G_x: Germination percentage in x treatment, G_o: Germination percentage in the control plot.

The effect of cruciferous plant extracts and concentrations on the number of infected eggplant branches

Eggplant branches infected with field dodder were counted on the last day of the experiment and documented for all treatments and replicates.

The effect of cruciferous plant extracts and concentrations on the field dodder fresh biomass

After recording the number of infected eggplant branches, the field dodder plants were carefully collected from each pot and placed in bags marked with the name of each treatment and the replicate number. The bags were transported to the laboratory for weighing using a precision scale with a sensitivity of 0.01g.

To evaluate the efficiency of the extracts in reducing the fresh biomass weight of field dodder, the following formula was used:

$$\text{Efficiency (\%)} = [1 - (F_x/F_o)] \times 100$$

Where, F_x: Fresh biomass weight in x treatment, F_o: Fresh biomass weight in the control plot.

The effect of cruciferous plant extracts and concentrations on the eggplant height

To ensure accuracy in determining the effect of the extracts on eggplant height, eggplant seedlings of consistent lengths (16-17 cm) were used in the experiment. Plant height was calculated on the last day of the experiment using a 50 cm metal ruler.

The effect of cruciferous plant extracts and concentrations on the total number of eggplant branches

The number of branches per plant was determined at the end of the experiment, specifically 45 days after seedling cultivation.

Data analysis

The experiment was replicated in both 2020 and 2021, and an Independent T-test was employed to assess mean differences between the two years. The statistical analysis based on the T-test results supported the acceptance of the null hypothesis, indicating no significant differences in means between the two years. Consequently, the average of the data from both years was employed in the subsequent results discussion. The collected data underwent analysis of variance (ANOVA) using MSTAT-C software (Version 2.10). Post hoc analysis was conducted using the Least Significant Difference (LSD) test, with a significance level of 0.05, to facilitate the comparison of means.

Results

The effect of cruciferous plant extracts and concentrations on the field dodder seeds germination

Across all experimental treatments utilizing 2% concentrations of cruciferous plant extracts, a decrease in the germination percentage of field dodder seeds was observed in comparison to the control plot. The control plot exhibited a germination percentage of 35.0%, while the treatments involving 2% cruciferous plant extracts displayed germination

percentages ranging from 32.6% to 33.4%, with no statistically significant differences among them.

The treatment using turnip extract at a 6% concentration exhibited notable efficiency compared to other treatments. It resulted in a substantial reduction in the germination of field dodder seeds, with the lowest observed value at 26.3%. Nevertheless, when testing treatments involving 6% concentrations of arugula, broccoli, and black radish extracts, no statistically significant differences were observed in terms of field dodder seed germination percentages. These three treatments recorded germination percentages of 32.4%, 32.7%, and 32.9%, respectively.

In all treatments where cruciferous plant extracts were applied at a 10% concentration, a noticeable decrease in the germination percentage of field dodder seeds was observed when compared to the control plot. The treatment involving turnip extract at a 10% concentration displayed the most significant reduction in field dodder seed germination compared to other treatments. Subsequently, the treatment utilizing arugula extract at a 10% concentration also demonstrated a substantial decrease. The germination percentages for the various treatments, namely turnip, arugula, broccoli, and black radish at a 10% concentration, were 15.50%, 21.90%, 25.70%, and 31.40%, respectively (Table 2).

Table 2. Germination rate of field dodder seeds

Treatments	Germination rate		
	2%	6%	10%
Turnip	32.6 ^a	26.3 ^a	15.50 ^a
Arugula	32.7 ^a	32.4 ^b	21.90 ^b
Broccoli	33.2 ^a	32.7 ^b	25.70 ^c
Black radish	33.4 ^a	32.9 ^b	31.40 ^d
Ctrl	35.0 ^b	35.0 ^c	35.0 ^e
LSD 0.05	0.438	0.973	1.705

Values followed by the same letter(s) in the same column are not significantly different from each other at 0.05 level of probability

The treatments using cruciferous plant extracts at a 2% concentration exhibited no statistically significant differences among themselves in reducing the percentage of field dodder seed germination. These treatments yielded a modest reduction in germination percentage, with values ranging from 4.57% to 6.86%.

The treatment involving turnip extract at a concentration of 6% demonstrated the most substantial reduction in the percentage of field dodder seed germination, achieving a noteworthy decrease of 24.84%. This treatment significantly outperformed other treatments. In contrast, there were no statistically significant differences observed among the treatments utilizing 6% concentrations of arugula, broccoli, and black radish extracts with reductions of 7.42%, 6.54%, and 5.98%, respectively.

Each of the treatments utilizing cruciferous plant extracts at a 10% concentration yielded a reduction in the percentage of field dodder seed germination with varying levels of efficiency. The treatment using turnip extract at a 10% concentration exhibited a notably superior performance compared to the other treatments, resulting in a significant reduction of 55.72% in the percentage of field dodder seed germination. In contrast, treatments using arugula, broccoli, and black radish extracts at a 10% concentration led to reductions of 37.42%, 26.58%, and 10.28%, respectively (Table 3).

Table 3. Efficiency of treatments in reducing germination rate

Treatments	Efficiency (%)		
	2%	6%	10%
Turnip	6.86 ^a	24.84 ^a	55.72 ^a
Arugula	6.57 ^a	7.42 ^b	37.42 ^b
Broccoli	5.14 ^a	6.54 ^b	26.58 ^c
Black radish	4.57 ^a	5.98 ^b	10.28 ^d
Ctrl	0.00 ^b	0.00 ^c	0.00 ^e
LSD 0.05	1.172	2.681	4.877

Values followed by the same letter(s) in the same column are not significantly different from each other at 0.05 level of probability

The effect of cruciferous plant extracts and concentrations on the number of infected eggplant branches

The results indicated that there were no statistically significant differences in the number of infected branches per plant between the treatments using arugula, broccoli, and black radish extracts at a 2% concentration and the control plot. However, the treatment of turnip extract at a 2% concentration demonstrated superior efficacy in comparison to the control plot, with 2.2 infected branches per plant.

All treatments using cruciferous plant extracts at a 6% concentration outperformed the control plot in terms of reducing the number of infected branches per plant. Specifically, the treatment utilizing turnip extract at a 6% concentration significantly outperformed the treatments involving broccoli and black radish extracts at the same concentration. Conversely, no statistically significant disparities were observed between the treatment involving turnip extract and the treatment involving arugula extract at a 6% concentration. The numbers of infected branches per plant were 2.8, 1.5, 1.8, 2.1, and 2.3 for the Ctrl plot, and treatments of turnip, arugula, broccoli, and black radish extracts at a concentration of 6%, respectively.

Table 4. The effect of cruciferous plant extracts on the number of infected branches

Treatments	The number of infected eggplant branches per plant		
	2%	6%	10%
Turnip	2.2 ^a	1.5 ^a	0.9 ^a
Arugula	2.5 ^{ab}	1.8 ^{ab}	1.7 ^b
Broccoli	2.4 ^{ab}	2.1 ^{bc}	1.8 ^b
Black radish	2.4 ^{ab}	2.3 ^c	2.1 ^b
Ctrl	2.8 ^b	2.8 ^d	2.8 ^c
LSD 0.05	0.544	0.474	0.428

Values followed by the same letter(s) in the same column are not significantly different from each other at 0.05 level of probability

On the other hand, the treatment utilizing turnip extract at a 10% concentration outperformed other treatments, resulting in a reduction of the number of infected branches to 0.9 branches per plant. However, no statistically significant differences were detected among the treatments involving arugula, broccoli, and black radish extracts at a 10% concentration concerning the number of infected branches, which recorded values of 1.7, 1.8, and 2.1 branches per plant, respectively (Table 4).

The effect of cruciferous plant extracts and concentrations on the field dodder fresh biomass

No statistically significant differences were evident between the treatments employing turnip and arugula extracts at a 2% concentration in relation to the field dodder's fresh biomass. However, the treatment involving turnip extract at a 2% concentration

exhibited marked superiority over the treatments employing broccoli and black radish extracts at the same concentration. While there were no significant differences between the treatment using arugula extract and the treatment using broccoli extract at a 2% concentration, the former significantly outperformed the treatment involving black radish extract at a 2% concentration. In terms of field dodder fresh biomass, the recorded values were 155.1 g, 156.7 g, 159.1 g, and 161.5 g for the treatments involving turnip, arugula, broccoli, and black radish extracts at a 2% concentration, respectively.

The cruciferous plant extracts at a 6% concentration exhibited notable effectiveness in reducing the field dodder's fresh biomass when compared to the control plot. Furthermore, no statistically significant differences in field dodder fresh biomass were observed between the treatments employing turnip and arugula extracts at a 6% concentration. Notably, these two treatments also outperformed the treatments involving extracts of broccoli and black radish at a 6% concentration. The recorded field dodder fresh biomass for the treatments with extracts of turnip, arugula, broccoli, and black radish at a 6% concentration was 126.0 g, 125.8 g, 129.7 g, and 131.4 g, respectively.

Table 5. The effect of cruciferous plant extracts on the field dodder fresh biomass

Treatments	The field dodder fresh biomass (g)		
	2%	6%	10%
Turnip	155.1 ^a	126.0 ^a	98.4 ^a
Arugula	156.7 ^{ab}	125.8 ^a	116.8 ^b
Broccoli	159.1 ^{bc}	129.7 ^b	123.5 ^c
Black radish	161.5 ^c	131.4 ^b	130.2 ^d
Ctrl	167.0 ^d	167.0 ^c	167.0 ^e
LSD 0.05	2.954	3.297	5.513

Values followed by the same letter(s) in the same column are not significantly different from each other at 0.05 level of probability

All treatments yielded a reduction in the field dodder's fresh biomass when compared to the control plot. Among these treatments, the most effective in terms of reducing field dodder fresh biomass was the treatment employing turnip extract at a 10% concentration, resulting in a recorded value of 98.4 g. The treatments involving extracts of arugula, broccoli, and black radish at a 10% concentration exhibited

statistically significant differences among them, with field dodder fresh biomass values for these treatments measuring 116.8 g, 123.5 g, and 130.2 g, respectively (Table 5).

The results indicate that the treatments utilizing turnip and arugula extracts at a 2% concentration did not exhibit statistically significant differences in their efficiency in reducing the field dodder's fresh biomass. These treatments outperformed the treatment employing black radish extract at a 2% concentration, while no statistically significant differences were observed between the treatment involving arugula extract and the treatment involving broccoli extract at a 2% concentration. The treatments using turnip, arugula, broccoli, and black radish extracts at a 2% concentration led to reductions in field dodder fresh biomass of 7.1%, 6.2%, 4.7%, and 3.3%, respectively.

Table 6. The percent of field dodder fresh biomass reduction

Treatments	Efficiency of treatments in reducing fresh biomass of field dodder (%)		
	2%	6%	10%
Turnip	7.1 ^a	24.5 ^a	41.1 ^a
Arugula	6.2 ^{ab}	24.7 ^a	30.0 ^b
Broccoli	4.7 ^{bc}	22.3 ^b	26.0 ^c
Black radish	3.3 ^c	21.1 ^b	23.4 ^c
Ctrl	0.0 ^d	0.0 ^c	0.0 ^d
LSD 0.05	1.769	1.919	3.317

Values followed by the same letter(s) in the same column are not significantly different from each other at 0.05 level of probability.

Upon analysis of the acquired data, it was evident that the treatments utilizing turnip and arugula extracts at a 6% concentration displayed equivalent efficiency in reducing the field dodder fresh biomass, with no statistically significant differences observed between these treatments. Nevertheless, both of these treatments outperformed the treatments involving broccoli and black radish extracts at a 6% concentration. The overall reduction in the field dodder fresh biomass amounted to 24.5%, 24.7%, 22.3%, and 21.1% for the treatments employing extracts of turnip, arugula, broccoli, and black radish at a 6% concentration, respectively.

The treatment utilizing turnip extract at a 10% concentration exhibited superior performance in comparison to the treatments involving arugula, broccoli, and black radish extracts at the same concentration. The reduction in field dodder fresh biomass for these treatments measured 41.1%, 30.0%, 26.0%, and 23.4%, respectively (Table 6).

Table 7. The effect of cruciferous plant extracts on the eggplant height

Treatments	The eggplant height (cm)		
	2%	6%	10%
Turnip	24.5 ^a	24.8 ^a	25.7 ^a
Arugula	24.5 ^a	24.7 ^a	25.4 ^a
Broccoli	24.4 ^a	24.9 ^a	25.1 ^{ab}
Black radish	24.7 ^a	24.9 ^a	25.0 ^{ab}
Ctrl	24.4 ^a	24.4 ^a	24.4 ^b
LSD 0.05	0.800	0.830	0.884

Values followed by the same letter(s) in the same column are not significantly different from each other at 0.05 level of probability.

The effect of cruciferous plant extracts and concentrations on the eggplant height

Eggplant height was not affected by different treatments of cruciferous plant extracts when compared to control plots. Only when applying turnip extract at a concentration of 10% was there an increase in the eggplant height compared to the control plot, where the eggplant height in this treatment was 25.7 cm (Table 7).

Table 8. The effect of cruciferous plant extracts on the total number of branches

Treatments	The total number of branches per plant		
	2%	6%	10%
Turnip	2.8 ^a	2.8 ^a	2.8 ^a
Arugula	2.7 ^a	2.8 ^a	2.8 ^a
Broccoli	2.7 ^a	2.7 ^a	2.8 ^a
Black radish	2.5 ^a	2.6 ^a	2.6 ^a
Ctrl	2.8 ^a	2.8 ^a	2.8 ^a
LSD 0.05	0.662	0.544	0.454

Values followed by the same letter(s) in the same column are not significantly different from each other at 0.05 level of probability.

The effect of cruciferous plant extracts and concentrations on the total number of eggplant branches

The total number of eggplant branches per plant was not affected by different treatments of cruciferous plant extracts when compared to control plots,

ranging from 2.5 to 2.8 branches per plant, with no significant differences observed between all treatments at all concentrations and the control plots (Table 8).

Table 9. Types and percentage of ITC compounds in cruciferous plants based on the GC-MS analysis

Cruciferous plants	Isothiocyanate compounds	Percentage (%)
Turnip	Allyl ITC	17.24
	3-Butenyl ITC	10.09
	Cyclopentyl ITC	6.35
	Heptyl ITC	12.44
	Phenethyl ITC	5.96
	Octyl ITC	4.52
	Total	56.6
Arugula	benzyl ITC	7.12
	Phenethyl ITC	4.32
	sulforaphane	11.03
	1-Naphthyl ITC	13.76
Total	36.23	
Broccoli	Allyl ITC	1.24
	Raphasatin	7.14
	Iberin	5.82
	sulforaphane	16.27
	Total	30.47
Black radish	Allyl ITC	4.62
	Isobutyl ITC	10.28
	benzyl ITC	7.52
	Raphasatin	3.24
	Phenethyl ITC	3.54
Total	29.2	

Identification of isothiocyanate compounds (ITCs) in cruciferous plants

Turnip revealed the presence of six distinct isothiocyanate (ITC) compounds, namely Allyl ITC, 3-Butenyl ITC, Cyclopentyl ITC, Heptyl ITC, Phenethyl ITC, and Octyl ITC. Collectively, these ITC compounds constituted 56.6% of the total, with Allyl ITC as the most predominant, accounting for 17.24% of the total. Arugula, on the other hand, exhibited the presence of four different ITC compounds, which included Benzyl ITC, Phenethyl ITC, Sulforaphane, and 1-Naphthyl ITC. The cumulative ITC compounds represented 36.23% of the total, with 1-Naphthyl ITC being the most abundant at 13.76% of the total. Broccoli exhibited the presence of four distinct ITC compounds, encompassing Allyl ITC, Raphasatin, Iberin, and Sulforaphane. The total ITC compounds accounted for 30.47% of the total, with Sulforaphane as the primary ITC compound, contributing 16.27% of the total. Finally, black radish presented five ITC

compounds, including Allyl ITC, Isobutyl ITC, Benzyl ITC, Raphasatin, and Phenethyl ITC. The combined ITC compounds constituted 29.20% of the total, with Isobutyl ITC as the primary ITC compound, constituting 10.28% of the total (Table 9).

Discussion

Observations revealed that the utilization of cruciferous plant extracts in the experiment did not have a detrimental impact on the growth of eggplant seedlings or the quantity of eggplant branches. These results align with the research conducted by Elsekran and Ustuner (2022), in which they investigated the effects of four cruciferous plant extracts, comprising white cabbage, red cabbage, black radish, and cress, at concentrations of 2%, 5%, and 10%, on both tomato seedlings and johnsongrass in a greenhouse setting. Their study similarly indicated no adverse effects on the growth of tomato seedlings.

The study examined the impact of varying concentrations of cruciferous plant extracts on the fresh biomass weight of field dodder. When employing a 2% concentration, the treatments exhibited a modest effect in reducing the fresh biomass weight of field dodder. The treatment with turnip extract yielded the highest reduction efficiency (7.1%), while the treatment with black radish extract showed the lowest (3.3%). As the concentration increased, the efficiency of the treatments also increased, with the treatment using arugula extract at a 6% concentration achieving the maximum reduction efficiency (24.7%), and the treatment with black radish extract at a 6% concentration yielding the minimum reduction efficiency (21.1%). At a 10% concentration, the treatment with turnip extract recorded the highest reduction efficiency (41.1%), followed by the treatment with arugula extract at a 10% concentration, which achieved an efficiency of 30.0%. The observed decrease in the fresh biomass weight of field dodder resulting from the application of the investigated cruciferous plant extracts is primarily attributed to their allelopathic characteristics. These properties are primarily manifested through the effects of allelochemicals on field dodder seed germination, which subsequently

leads to a reduction in the number of parasitic plants, as well as their influence on the growth and development of field dodder, ultimately contributing to a decrease in fresh biomass weight. Numerous previous studies have also demonstrated that allelochemicals can induce alterations in both the micro and macro structure of cells (Wu *et al.*, 2004; Pawlowski *et al.*, 2012) and inhibit cell division and elongation (Ortega *et al.*, 1988; Hallak *et al.*, 1999), providing additional corroboration for this interpretation.

Turnip exhibited the highest content of ITC compounds, constituting 56.6% among the tested cruciferous plants. Six distinct compounds were identified in turnip, namely, Allyl ITC, 3-butenyl ITC, Cyclopentyl ITC, Heptyl ITC, Phenethyl ITC, and Octyl ITC. These findings align with previous studies that have also reported the presence of multiple ITC compounds in turnip. The distribution of these compounds varies across different plant parts, encompassing roots (N-butyl ITC, 3-butenyl ITC), leaves, and flowers (Benzyl ITC, Allyl ITC, 4-pentenyl ITC), and the entire plant (2-phenylethyl ITC), as documented by Petersen *et al.* (2001) and Paul *et al.* (2019). Furthermore, Vieites-Outes *et al.* (2016) identified eight distinct ITC compounds in turnip, providing further support for our results.

Arugula exhibited a collective ITC compound content of 36.23%, encompassing Benzyl ITC, Phenethyl ITC, Sulforaphane, and 1-naphthyl ITC. Previous research has substantiated the presence of Phenethyl ITC in arugula, stemming from the hydrolysis of Gluconasturtiin (Ioannides *et al.*, 2010). Additionally, several other studies have reported the identification of four ITC compounds in arugula, namely Phenethyl ITC, Allyl ITC, Iberin, and Sulforaphane (Hornalley, 2004; Villatoro-Pulido *et al.*, 2013; Rodrigues *et al.*, 2016), aligning partially with our findings.

Broccoli exhibited ITC compounds content of 30.47%, encompassing Allyl ITC, Raphasatin, Iberin, and Sulforaphane. These findings are partly in accordance with a previous study that reported the presence of three ITC compounds (Allyl ITC, Phenethyl ITC, Iberin) in

broccoli (Wang *et al.*, 2010). Other preceding research has also detected diverse ITC compounds in broccoli, including Sulforaphane, Iberin, Erucin, Allyl ITC, and Phenethyl ITC (Chiang *et al.*, 1998; Liang *et al.*, 2006; Lv *et al.*, 2021), providing partial corroboration for our results.

Black radish exhibited a 29.2% content of ITC compounds, which included Allyl ITC, Isobutyl ITC, Benzyl ITC, Raphasatin, and Phenethyl ITC. These findings corroborate the work of Elsekran and Ustuner (2022), who identified five ITC compounds in black radish (Tert-butyl ITC, 2-propenyl ITC, Benzyl ITC, 4-methylthio-3-butenyl ITC, 2-phenylethyl ITC) constituting 40.39% of the composition. Our study aligns partially with Uremis *et al.* (2009), who noted elevated levels of Benzyl ITC and Allyl ITC in black radish. Moreover, our findings are partially congruent with Castro-Torres *et al.* (2013), who reported the presence of two ITC compounds in black radish, Raphasatin, and Sulforaphane.

Conclusion

The findings revealed that all tested concentrations of cruciferous plant extracts led to a reduction in the germination percentage of field dodder seeds, with the extent of this effect amplifying in conjunction with rising extract concentrations. In terms of efficiency for diminishing field dodder seed germination, the cruciferous plants can be ranked from most effective to least effective as follows: turnip, arugula, broccoli, and black radish. However, it is noteworthy that the concentrations of the studied cruciferous plant extracts did not exert a significant influence on the eggplant height or the total number of branches they produced. Conversely, there was a noteworthy impact on the number of infected eggplant branches and the fresh biomass weight of the field dodder. Overall, these results underscore the potential of using allelopathic effects of cruciferous plants as an environmentally friendly alternative for controlling field dodder substitute to traditional herbicide-based control methods.

Recommendation(s)

Based on the results of this study, it is recommended to cultivate cruciferous plants in agricultural lands infested with field dodder. Cruciferous plants can also be useful in field dodder control if included in agricultural rotations. Incorporating these plants into the soil at certain stages of their life will help release ITC compounds, which have a major role in reducing the germination rate of field dodder seeds and thus reducing the damage caused by this parasitic plant to cultivated crops.

Competing interests

The authors declare that they have no competing interests.

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