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IN VITRO ALLELOPATHIC POTENTIAL OF *Photinia × fraseri* EXTRACTS ON THE SEED GERMINATION OF SELECTED CROP AND WEED SPECIES

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Abstract

In sustainable agriculture, allelopathy emerges as a promising strategy for eco-friendly weed management. This study provides a preliminary in vitro assessment of the allelopathic effects of aqueous flower and leaf extracts of *Photinia* × *fraseri* on the seed germination of selected crop (wheat, corn, lentil, lettuce) and weed (radish, purslane) species. The chemical composition of the extracts was analyzed using GC-MS, and germination assays were conducted at five concentrations: 1%, 25%, 50%, 75%, and 100%, corresponding to 0.5, 12.5, 25, 37.5, and 50 mg^{ml-1}, respectively. The results showed a concentration-dependent inhibition of germination in all tested species, with flower extracts exhibiting higher allelopathic effects than leaf extracts. Lettuce and radish seeds were the most sensitive, while corn and wheat were the least affected. Given that wild radish is a common agricultural weed, flower extract concentrations of 50% and above may represent potential candidates for bioherbicide development. These findings are an initial step toward understanding the allelopathic potential of *P. × fraseri*, and further in vivo pot and/or field studies are recommended to validate its practical applicability.

Keywords: Bioherbicide. Flower extract. Germination assay. Leaf extract. Red Robin.

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1. Introduction

Ensuring access to an adequate and secure food supply is recognized as a vital component for supporting human life and well-being in modern days. However, due to rapid population growth, access to adequate and safe food decreases daily. Furthermore, it is essential to limit product waste while enhancing crop production and quality to meet nutritional demands. Weeds continually compete with crops, resulting in significant yield and quality losses (Mushtag and Siddiqui 2018; Chauhan 2020). Allelopathy, an ecological

phenomenon and a subdiscipline of chemical ecology (Cheng and Cheng 2015), has gained attention as a beneficial strategy, especially for weed control, in sustainable agriculture (Jabran et al. 2015).

Synthetic pesticides pose a significant threat to the environment and human health, and contribute to higher agricultural production costs. Conversely, exploring and using allelopathic plants and allelochemicals represent a promising alternative. These natural compounds, particularly those with potent herbicidal effects, are considerably relevant for sustainable agriculture. Unlike synthetic pesticides, allelochemicals have the advantage of being easily degraded in nature, thereby minimizing environmental harm. Moreover, their application may reduce agricultural production costs (Hasan et al. 2021; Budak and Işık 2022).

The genus *Photinia* (Rosaceae) belongs to the group of perennial large shrubs and small trees. It comprises around 60 known species, with 43 distributed in China, including endemic species, as well as in North America, Mexico, Europe, and parts of Asia. *Photinia x fraseri*, commonly known as "Red Robin," is an evergreen ornamental plant. This variety results from the hybridization between *P. glabra* (Thunb.) Maxim and *P. serrulata* (Desf.) Kalkman (Çetiner and Zencirkıran 2020), which demonstrated bioactive properties (Appiah et al. 2015); thus, *P. × fraseri* may also harbor allelochemicals of agricultural significance.

As the importance of effectively utilizing allelopathy in agriculture becomes more acknowledged, research in the field has expanded, leading to the discovery of new and more potent allelopathic plants and allelochemicals (Khamare et al. 2022; Kostina-Bednarz et al. 2023). To the best of our knowledge, no previous study has evaluated the allelopathic properties of *P. × fraseri* extracts. In this context, the present study aims to fill this gap by investigating the effects of aqueous flower and leaf extracts on the germination of selected crop and weed species under *in vitro* conditions.

2. Material and Methods

Plant material and preparation of extracts

The flowers and leaves of *P. x fraseri* were collected in May 2023 from the landscaping areas of the central campus of the Tekirdağ Namık Kemal University, Türkiye. After washing the flowers and leaves (mixed in equal amounts with red leaves just below the flowers and green leaves below them) in tap water, the plant material was left to dry for approximately 15-20 days in a well-ventilated room at an average temperature of 22-24°C and relative humidity of around 50-60%. Following the drying process, the leaves were manually crushed into powder with an average particle size of approximately 1-2 mm. From this material, 20 g was soaked in 200 ml of distilled water at room temperature in a dark environment for 24 hours. This mixture was first filtered through cheesecloth, then filtered twice through No. 1 Whatman filter paper (Yaraş and Yılmazer 2024). The filtrate was stored in the refrigerator at +4°C. An additional 200 ml of distilled water was added to the residue, and the same process was repeated to obtain a second filtrate. The two filtrates were combined and stored in the refrigerator as a stock extract (5%), corresponding to a concentration of 50 mg ml^{-1}. For clarity and consistency, all extract concentrations throughout the manuscript are expressed as 100% (50 mg ml^{-1}), 75% (37.5 mg ml^{-1}), 50% (25 mg ml^{-1}), 25% (12.5 mg ml^{-1}), and 1% (0.5 mg ml^{-1}).

In vitro germination assays

This study used seeds of wheat (*Triticum* L. sp.), lettuce (*Lactuca sativa* L.), lentil (*Lens* Mill. sp.), corn (*Zea mays* L.), purslane (*Portulaca oleracea* L.), and radish (*Raphanus sativus* L.) plants, as these crops are widely cultivated across various agricultural landscapes and play a significant role in human nutrition. Seeds with cracks, deformities, or unusual color were sorted out through visual inspection. Empty and underdeveloped seeds were identified by floating them in a basin filled with tap water. The selected intact seeds underwent surface sterilization for two minutes in a 10:1 solution of distilled water and commercial bleach. After sterilization, the seeds were rinsed three times with distilled water. Petri dishes, drying (filter) paper, glassware, and forceps used in germination experiments were wrapped in aluminum foil and sterilized in an autoclave at 121°C for 15 minutes (Ertem and Adak 2022). Seeds were germinated between two sheets

of drying paper in 12 cm diameter Petri dishes. Depending on the size, 40 wheat, lentil, and corn seeds, 60 radish seeds, 150 lettuce seeds, and 200 purslane seeds were uniformly placed with forceps in each Petri dish. Before sowing the seeds in Petri dishes, filter paper placed at the bottom of the Petri dish was moistened with 3 ml of extract (100%, 75%, 50%, 25%, and 1%) for experimental seeds and 3 ml of distilled water for control seeds. After placing the seeds, they were covered with drying paper. Then, 9 ml of the respective extract concentration was added to each experimental group, and the same amount of distilled water was added to the control group. The Petri dishes were sealed and maintained in an incubator at 22-24°C for 72 hours. Seeds with radicle length greater than 2 mm at the end of the germination period were considered germinated (Kaçal et al. 2020). Each germination assay was repeated independently three times on separate days using freshly prepared extract solutions and seeds from the same seed lot, representing biological replicates.

Germination rate and effect compared to the control (%)

The germination rate (%) was calculated by multiplying the ratio of the number of germinated seeds to the total number of seeds at the end of the germination period by 100 (Ertem and Adak 2022). The results of the three repetitions were given as mean ± standard error. The effect (%) of different concentrations of the extract compared to the control was calculated using the following formula (Yaraş and Yılmazer 2024):

Effect (%) compared to the control = (Germination rate of the extract concentration - Germination rate of the control) x 100 / Germination rate of the control

Gas chromatography-mass spectrometry (GC-MS) analysis

The GC-MS analysis employed a QP2010-Ultra coupled with an AOC-6000 Plus Multifunctional Autosampler (Shimadzu), following the methods outlined in previous studies (Jie et al. 2007; Kasapoğlu Uludamar 2024). An Rxi-5ms capillary column (30 m, 0.25 mm ID, 0.25 µm film thickness, Restek) was used. Helium served as the carrier gas at a flow rate of 1.0 ml/min. The temperature program consisted of increasing the oven temperature from 50 to 250°C at a rate of 3°C/min, with a final hold time of 10 minutes. The injector and interface temperatures were set at 220 and 250°C, respectively. Compound identification was based on spectral matching with the NIST 2A, WILEY 7, NIST 147, and NIST 107 libraries. No authentic chemical standards were used for confirmation; thus, compound identification remains tentative and based on library similarity indices.

Statistical analysis

The statistical analysis was performed in GraphPad Prism software (GraphPad Prism Version 9, San Diego, CA, USA). After one-way analysis of variance (ANOVA), Tukey's multiple comparison test was conducted. A value of $p < 0.05$ was statistically significant.

3. Results

The germination of all tested species decreased progressively with increasing concentrations of *P. × fraseri* flower and leaf extracts, indicating an evident concentration-dependent inhibitory effect (Figures 1 and 2).

Lettuce and radish seeds were consistently the most sensitive to both extracts, showing significant reductions even at lower concentrations. Conversely, wheat and corn exhibited higher tolerance, with notable inhibition only at the highest extract levels. Flower extracts demonstrated higher allelopathic activity than leaf extracts across all plant species. Although the low-concentration leaf extract (1%) slightly promoted corn germination, this effect was not statistically significant. Tables 1 and 2 detail the germination rates and inhibitory percentages.

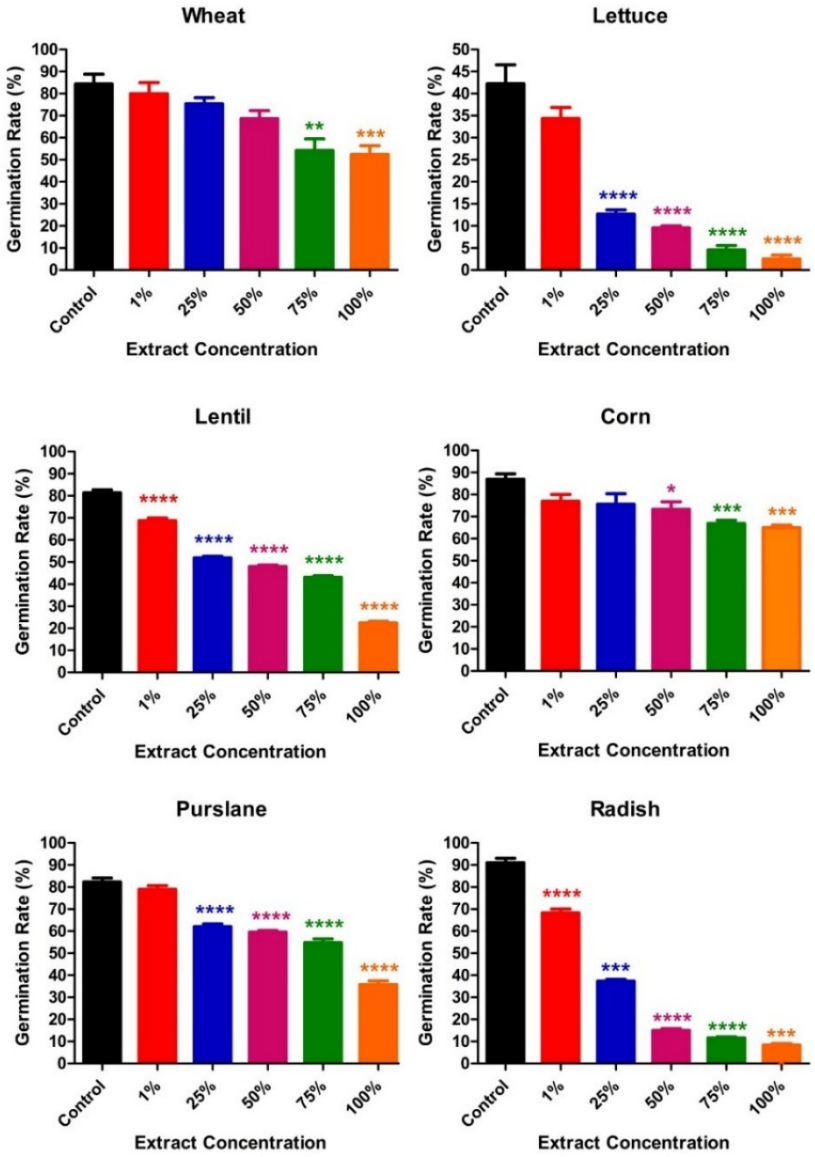

Figure 1. The effect of 1%, 25%, 50%, 75%, and 100% concentrations of *Photinia x fraseri* aqueous flower extract on the germination of wheat, corn, lentil, lettuce, radish, and purslane seeds. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; **** $p < 0.0001$; statistical significance compared to the control group.

Table 1. The effect of different concentrations of *P. x fraseri* aqueous flower extract compared to the control.

Test Plants	Germination Rates (%)*					
	Control	Extract Concentrations				
		1%	25%	50%	75%	100%
Wheat	84.41	79.94	75.40	68.66	54.21	52.46
The effect compared to the control		-5.30	-10.67	-18.66	-35.78	-37.85
Corn	86.94	76.96	75.63	73.31	66.88	64.97
The effect compared to the control		-11.48	-13.01	-15.68	-23.07	-25.27
Lentil	81.33	68.67	51.87	47.94	43.09	22.44
The effect compared to the control		-15.57	-36.22	-41.06	-47.02	-72.41
Lettuce	42.22	34.36	12.71	9.56	4.55	2.50
The effect compared to the control		-18.62	-69.90	-77.36	-89.22	-94.08
Radish	91.03	68.31	37.25	14.93	11.62	8.40
The effect compared to the control		-24.96	-59.08	-83.60	-87.24	-90.77
Purslane	82.27	78.99	62.00	59.52	54.78	35.81
The effect compared to the control		-3.99	-24.64	-27.65	-33.41	-56.47

*Germination rates are given as "Mean." Concerning the effect compared to the control, negative (-) values indicate the inhibition of germination.

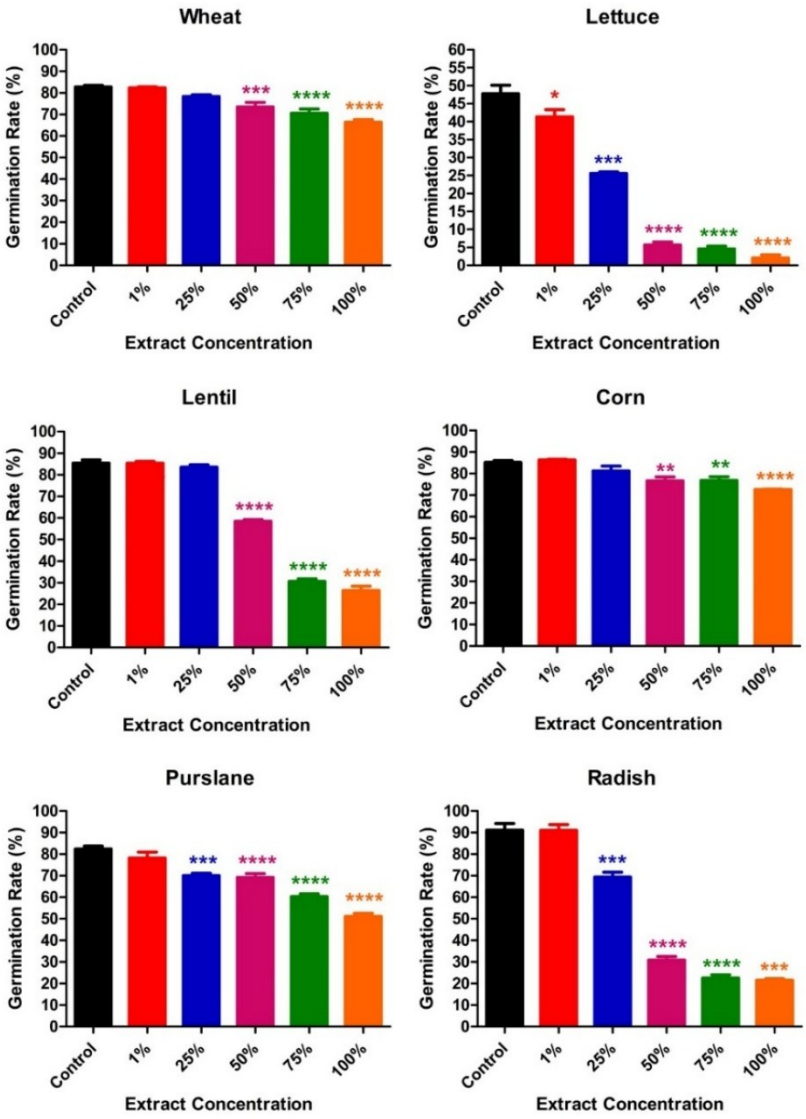

Figure 2. The effect of 1%, 25%, 50%, 75%, and 100% concentrations of *Photinia x fraseri* aqueous leaf extract on the germination of wheat, corn, lentil, lettuce, radish, and purslane seeds. * $p < 0.05$; ** $p < 0.01$; *** $p < 0.001$; **** $p < 0.0001$; statistical significance compared to the control group.

Table 2. The effect of different concentrations of *P. x fraseri* aqueous leaf extract compared to the control.

Test Plants	Germination Rates (%)*					
	Control	Extract Concentrations				
		1%	25%	50%	75%	100%
Wheat	82.72	82.25	78.26	73.42	70.54	66.38
The effect compared to the control		-0.57	-5.39	-11.24	-14.72	-19.75
Corn	85.11	85.31	81.21	76.62	76.80	72.67
The effect compared to the control		+0.24	-4.58	-9.98	-9.76	-14.62
Lentil	85.37	85.31	83.48	58.41	30.61	26.45
The effect compared to the control		-0.07	-2.21	-31.58	-64.14	-69.02
Lettuce	47.78	41.31	25.58	5.66	4.57	2.06
The effect compared to the control		-13,54	-46.46	-88.15	-90.44	-95.69
Radish	91.11	91.02	69.34	30.86	22.50	21.50
The effect compared to the control		-0.10	-23.89	-66.13	-75.31	-76.40
Purslane	82.34	78.22	70.06	69.24	60.28	51.16
The effect compared to the control		-5.00	-14.91	-15.91	-26.79	-37.87

*Germination rates are given as "Mean." Concerning the effect compared to the control, positive (+) values indicate the promotion of germination, while negative (-) values indicate the inhibition of germination.

The GC-MS analysis of leaf and flower extracts identified 31 compounds in each extract, as listed in Table 3, representing 97.84% and 99.35% of the total compounds in leaf and flower extracts, respectively. A small portion of the total compounds was not identified, accounting for 2.16% in the leaf extract and 0.65% in the flower extract. Benzaldehyde was the dominant compound in both extracts, comprising 48.73% of the leaf extract and 69.51% of the flower extract. That was followed by Phenol, 2,2'-methylenebis[6-(1,1-dimethylethyl)-4-methyl-, which constituted 10.18% of the leaf extract and 10.21% of the flower extract. The remaining compounds had a weak representation in both extracts, each comprising less than 10%.

Table 3. Main compounds in *P. x fraseri* aqueous leaf and flower extracts.

Leaf		Flower	
Compound	**Area (%)**	**Compound**	**Area (%)**
Benzaldehyde	48.73	Benzaldehyde	69.51
Phenol, 2,2'-methylenebis[6-(1,1-dimethylethyl)-4-methyl-	10.18	Phenol, 2,2'-methylenebis[6-(1,1-dimethylethyl)-4-methyl-	10.21
7,9-Di-tert-butyl-1-oxaspiro(4,5)deca-6,9-diene-2,8-dione	4.62	Pentadecane	1.98
Pentadecane (CAS) n-Pentadecane	4.15	Acetic acid, octadecyl ester	1.73
Phenol, 2,4-bis(1,1-dimethylethyl)- (CAS) 2,4-Di-tert-butylphenol	3.89	Hexadecane	1.70
Heneicosane	3.63	Tetradecane (CAS) n-Tetradecane	1.65
Tetradecane (CAS) n-Tetradecane	2.73	Heneicosane	1.15
Dodecane, 4,6-dimethyl-	2.26	Phenol, 2,4-bis(1,1-dimethylethyl)- (CAS) 2,4-Di-tert-butylphenol	0.94
Hexadecane	2.21	Heptadecane (CAS) n-Heptadecane	0.77
Acetic acid, octadecyl ester	2.14	d-Glucitol, 2,5-anhydro-1-O-octyl-	0.74
Hexadecane, 2,6,10,14-tetramethyl- (CAS) Phytane	2.01	Dodecane, 4,6-dimethyl-	0.64
Eicosane	1.05	N ,N-dimethyl palmitamine	0.62
2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl- (CAS) dihydro	1.04	Hexadecane, 2,6,10,14-tetramethyl- (CAS) Phytane	0.60
Dodecane	1.01	7,9-Di-tert-butyl-1-oxaspiro(4,5)deca-6,9-diene-2,8-dione	0.59
Tetradecane, 5-methyl-	0.77	1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester (CAS) Isobutyl phthalate	0.55
Heptadecane	0.76	Nonane, 5-methyl-5-propyl- (CAS)	0.54
Docosane (CAS) n-Docosane	0.75	Decane, 1,1'-oxybis-	0.53
Heptadecane	0.75	Heptadecane, 2,6,10,15-tetramethyl-	0.48
Eicosane, 2,4-dimethyl-	0.67	2-Docosanone, 4,21,21-trimethyl-, L-(-)-	0.45
Octadecane	0.62	N ,N-dimethyl palmitamine	0.41
1,2-Benzenedicarboxylic acid, bis(2-methylpropyl) ester	0.54	Hexadecane, 1-iodo-	0.37
1,3-Dioxolane, 4-ethyl-5-octyl-2,2-bis(trifluoromethyl)-, cis- (CAS) CIS-2,2-BI	0.51	Decane, 3-cyclohexyl-	0.36
1,4-Methanoazulen-9-ol, decahydro-1,5,5,8a-tetramethyl-, [1R-(1.alpha.,3a.beta, 4alpha,8Abeta,9S*)]-	0.49	Nonanal (CAS) n-Nonanal	0.35
8-Methyl-6-nonenamide	0.47	Butyl Hydroxy Toluene	0.33
3-Ethyl-3-methylheptane	0.42	Decane, 3,8-dimethyl- (CAS) 3,8-Dimethyldecane	0.33
Docosane	0.30	Tetratriacontane (CAS) n-Tetratriacontane	0.33
2-Propen-1-one, 1,3-diphenyl-	0.27	Heptadecane	0.31
Pentadecane	0.27	Heptadecane, 1-bromo-	0.31
2H-Benzazepine-2-carboxylic acid, 1,3,4,5-tetrahydro-5-methyl-1-oxo-3-spiroc	0.22	10-Methylnonadecane	0.30
Spiro(adamantane)-2,2'-(1,3-dithiolane), 4-methanesulfonate	0.20	Hexane, 2,2,3,3-tetramethyl- (CAS) 2,2,3,3-Tetramethylhexane	0.29
4-(4-Bromobutyl)-2,2,6-trimethyl[1,3]dioxane	0.18	Undecane, 3,8-dimethyl-	0.28
Total	97.84	Total	99.35

4. Discussion

In sustainable agriculture, allelopathy (a beneficial strategy especially for weed control) draws attention to ensuring a sufficient and safe food supply for the growing human population. Increasing studies have focused particularly on plant-plant, weed-plant, and weed-weed interactions (Ain et al. 2023; Xu et al. 2023). This study described the potential allelopathic effects of *P. x fraseri* flower and leaf extracts on the germination of wheat, corn, lentil, and lettuce seeds, as well as radish and purslane weeds, selected as target weed species. No study to date has been conducted on the allelopathic property of *P. x fraseri*. Only one investigation has analyzed the allelopathic effect of the *P. glabra* leaf extract on seedling development in lettuce, inhibiting it by 90-100% (Appiah et al. 2015). Considering that *P. x fraseri* is a hybrid from the crossbreeding between *P. glabra* and *P. serrulata*, this finding confirms the allelopathic property of *P. x fraseri*.

Experimental design and study rationale

Laboratory experiments play a significant role in determining the allelopathic properties of a plant. The most frequent laboratory experiments are seed germination and seedling development tests (Janusauskaite 2023). These experiments frequently employ the Petri dish method - an *in vitro* germination technique. Seed germination tests often express the results as germination rates (Lotina-Hennsen et al. 2006). The present study conducted seed germination experiments using the Petri dish method to determine the allelopathic properties, and the results were given as germination rates.

Selecting target species from monocotyledonous and dicotyledonous plants to determine the potential selectivity of allelochemicals is one of the most significant aspects of allelopathic studies (Lotina-Hennsen et al. 2006). Therefore, this study selected monocotyledonous (wheat and corn) and dicotyledonous (lettuce, lentil, radish, and purslane) species as test plants. Lettuce (*Lactuca sativa* L.) is the preferred model plant in allelopathy studies, widely used as a test plant due to its rapid germination and high sensitivity (Lotina-Hennsen et al. 2006); therefore, our study also included lettuce among the test plants.

Considering that many allelochemicals are hydrophilic (Turk et al. 2003; Chou 2006), they are fully or partially soluble in water (Soltys et al. 2013; Singh et al. 2021). Consequently, in nature, they are usually released from the aboveground parts of plants through dew, rain, or fog droplets and washed into the soil (Wetzel and Howe 1999). They may exhibit effects in this form or acquire true allelopathic properties after being altered by soil microorganisms (Zhu et al. 2021). Therefore, this study employed aqueous extracts to simulate dissolved/washed allelochemicals in nature.

Selection of radish and purslane as test weed species

The present study selected radish and purslane as test plants within the scope of weeds. Wild radish (*Raphanus raphanistrum* ssp. *raphanistrum* L.; synonym *R. sativus* L.) is one of the four most widespread weed species that significantly harm agricultural fields worldwide (Vercellino et al. 2021). Purslane ranks ninth among the most invasive weeds globally due to its high ability to propagate through vegetative reproduction by forming lateral roots from stem parts and producing numerous seeds within six weeks after germination (a single plant can produce from 10,000 to 242,000 seeds) (El-Shora et al. 2022). Our study included commercial seeds of radish and purslane, sold as cultivated plants. However, the germination success of seeds from wild plants in nature is weaker than that of commercial seeds of the same cultivated plants (Fernández et al. 2008). Therefore, the allelopathic effect in our experiments might be even higher on wild radish and purslane.

Dose-response relationship and species sensitivity

The effect of allelochemicals varies considerably depending on the solvent used for extract preparation (Williamson and Richardson 1988) and concentration (dose) (Belz et al. 2005), and the test plant

species (Prinsloo and Du Plooy 2018; Oraon and Mondal 2021). Most studies to date have shown that increasing extract concentrations significantly reduce seed germination and seedling development in test plants, and may even lead to complete inhibition (Tefera 2002; Synowiec and Nowicka-Połeć 2016; Prinsloo and Du Plooy 2018; Bari et al. 2019). Consistent with the findings of these studies, our investigation showed that, as the concentration of *P. x fraseri* flower and leaf extracts increased, the germination rates of all test plants decreased. The allelopathic effect of *P. x fraseri* flower extract on germination rates depending on the concentration was as follows: lettuce (18.62-94.08%), radish (24.96-90.77%), lentil (15.57-72.41%), purslane (3.99-56.47%), wheat (5.30-37.85%), and corn (11.48-25.27%). The leaf extract results were lettuce (13.54-95.69%), radish (0.10-76.40%), lentil (0.07-69.02%), purslane (5.00-37.87%), wheat (0.57-19.75%), and corn (4.58-14.62%). Particularly at 100% concentration, the flower extract inhibited radish seed germination by around 90%, while the leaf extract at the same concentration inhibited it by approximately 76%. As seen in our study, responses to the same extract at the same concentration vary depending on the plant species. Differences in seed structure and coat permeability are the most significant factors contributing to this phenomenon (Bashar et al. 2022).

Allelochemicals may also promote the germination and growth of the same or other species when used at different concentrations (Lovett et al. 1989; Narwal 1994). Various studies have shown that, particularly, low-concentration aqueous extracts stimulate the germination and growth of several crops, increasing their productivity (Oudhia et al. 1998; Tefera 2002; Cheema et al. 2013). Although our investigation found that a 1% concentration of leaf extract promoted the germination of corn seeds, this increase was not statistically significant.

Species-specific tolerance and corn resistance

Among the tested species, corn exhibited the highest tolerance to the allelopathic extracts. This species-specific resistance may be attributed to morphological and physiological characteristics such as larger seed size, thicker seed coat, reduced permeability to water-soluble allelochemicals, and inherent metabolic defenses. Larger seeds, particularly, tend to be more resilient due to their higher carbohydrate reserves, which support early seedling development and enhance stress tolerance. Supporting this, Motalebnejad et al. (2023) reported that smaller-seeded and early-emerging species are often more vulnerable to allelopathic inhibition, primarily because of their limited carbohydrate storage.

Comparison between flower and leaf extracts

All parts of plants, including roots, stems, leaves, flowers, rhizomes, fruits, seeds, and pollen, contain allelochemicals (Bashar et al. 2022), but the effect of extracts from different parts of the same plant may vary (Janusauskaite 2023). For instance, the aqueous stem extract of sunflower exhibits high allelopathic activity in wheat, the aqueous leaf extract shows lower activity, and the root extract shows very little allelopathic activity (Bashir et al. 2017). Similarly, the *Parthenium hysterophorus* L. leaf methanol extract presents a higher allelopathic effect on the germination and seedling development of *Vigna subterranea* (L.) Verdc., *Raphanus sativus*, *Cucurbita maxima* Lam., *Cucumis sativus* L., *Solanum lycopersicum* L. (*Lycopersicon esculentum* Mill.), *Capsicum frutescens* L., *Zea mays* L., *Abelmoschus esculentus* (L.) Moench, *Daucus carota* L., *Digitaria sanguinalis* (L.) Scop., and *Eleusine indica* (L.) Gaertner than stem and flower methanol extracts (Bashar et al. 2022). Our study also observed that the allelopathic effect of the flower extract was higher than that of the leaf extract.

Identified compounds and possible synergistic effects

P. glabra leaves contain two biphenyl (phenylbenzene) compounds (2'-methoxyaucuparin and 4'-methoxyaucuparin) (Appiah et al. 2015), while its fruits contain anthocyanins (Ishikura 1975). The most abundant compound found in *P. serrulata* flowers is benzoyl aldehyde (benzaldehyde) (Wei et al. 2013; Shu-feng and Qiao-yun 2019). Furthermore, various terpenes have been isolated from *P. serrulata* leaves (Jie et al. 2007). Song et al. (2021) determined that *P. x fraseri* leaves contain catechin and epicatechin. All these

compounds in *Photinia* species belong to various classes of allelochemicals (Cheng and Cheng 2015; Denaxa et al. 2022). Similarly, benzaldehyde was the main compound in leaf and flower extracts in our study, with a higher concentration in the latter, followed by Phenol, 2,2'-methylenebis[6-(1,1-dimethylethyl)-4-methyl- in both extracts. Benzaldehyde inhibits cell division and elongation in seedling roots (Choi et al. 2016), while catechins significantly inhibit the root growth of Bambusa and Koeleria seedlings (Inderjit et al. 2008). Phenolic compounds are universally distributed in plants and might inhibit plant root elongation and cell division, change cell ultrastructure, and interfere with the regular growth and development of the whole plant (Li et al. 2010). The phytotoxic effects found in this study are likely due to these bioactive compounds, although the precise mode of action requires elucidation.

It is also plausible that the inhibitory activity is not solely due to one key compound but rather to synergistic interactions among multiple allelochemicals. Complex mixtures of phenolics, aldehydes, and terpenoids may enhance each other's phytotoxic effects, as demonstrated in studies on plant-derived bioherbicides (Chou 2006). The GC-MS analysis revealed several minor compounds that may contribute to or amplify the activity of dominant compounds such as benzaldehyde.

Practical implications for weed control and study limitations

Although the *in vitro* findings of this study highlight the promising allelopathic potential of *P. × fraseri* extracts, several limitations should be acknowledged. Laboratory-based germination assays are inherently simplified systems that do not fully capture the complexity of natural agroecosystems. Therefore, further pot and field experiments are necessary to assess the stability and bioavailability of allelochemicals in soil, their interactions with soil microbiota, and the selectivity of their effects on target weed species versus crops. Additionally, potential phytotoxic impacts on non-target organisms require analyses to ensure the environmental safety and sustainability of using such extracts in real-world conditions.

One limitation of the current study is the absence of a positive control treatment using a commercial synthetic herbicide. The inclusion of such a benchmark would have allowed a more direct comparison of the efficacy of *P. × fraseri* extracts with conventional herbicidal standards. Future studies should incorporate positive controls to better contextualize the inhibitory strength of plant-derived allelochemicals within existing weed management frameworks.

5. Conclusions

This study provides the first empirical evidence of the allelopathic potential of *P. × fraseri*, particularly its floral extract, in inhibiting the germination of selected crop and weed species under *in vitro* conditions. The concentration-dependent phytotoxicity combined with the identification of known allelochemicals such as benzaldehyde suggests the potential of this ornamental plant as a source of natural herbicidal agents. Radish seeds were the most sensitive among the tested species. Thus, the potential use of high-concentration *P. x fraseri* flower extracts as an effective bioherbicide should be evaluated for combating wild radish, which grows as a weed in agricultural fields. However, to validate the practical applicability and environmental safety of these extracts, further *in vivo* assessments, including pot and field trials under real agronomic conditions, are essential.

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