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Allelopathic Potential of *Daphne mucronata* Royle on Seed Germination and Early Seedling Growth of *Convolvulus arvensis* and *Asphodelus tenuifolius* in Relation to Total Phenolic Content

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ABSTRACT

Weed-related crop losses and increasing herbicide resistance in Pakistan necessitate eco-friendly alternatives for sustainable weed management; however, the allelopathic potential of *Daphne mucronata* Royle, a medicinally important native shrub of Balochistan, remains scientifically unexplored. The present study investigated the allelopathic effects of *D. mucronata* against two problematic weeds, *Convolvulus arvensis* and *Asphodelus tenuifolius*. Plant specimens were collected from two ecologically distinct regions of Balochistan i.e. Hazarganji and Ziarat, and aqueous leaf extracts were prepared at concentrations of 0%, 8%, 16%, and 32% to evaluate their inhibitory effects on seed germination and early seedling growth under controlled laboratory conditions. The results demonstrated significant concentration-dependent suppression of germination percentage, root and shoot elongation, and fresh and dry biomass in both weed species. Extracts obtained from Ziarat exhibited comparatively stronger phytotoxic effects than those from Hazarganji, while *C. arvensis* showed greater sensitivity to the treatments in comparison to *A. tenuifolius*. In addition, total phenolic content (TPC) increased with increasing extract concentration, indicating a strong association between phenolic compounds and allelopathic activity. The findings suggest that *D. mucronata* contains potent allelochemicals with promising potential for the development of natural bio-herbicides and sustainable weed management strategies. However, further field-based investigations are necessary to validate these laboratory findings and to understand the physiological mechanisms underlying the observed phytotoxic effects.

KEYWORDS: Allelopathy, *Asphodelus tenuifolius*, Bio-herbicides, *Convolvulus arvensis*, *Daphne mucronata* Royle, Phytotoxicity, Seed Germination, Total Phenolic Content

INTRODUCTION

Plants continuously interact with their biotic and abiotic environment not only through competition for essential resources such as light, water, and nutrients, but also through the release of biologically active chemical compounds known as allelochemicals. These compounds can influence the growth, survival, and reproduction of neighboring plants, and such chemically mediated interactions are collectively termed allelopathy (Farooq *et al.*, 2011; Khamare *et al.*, 2022). Allelopathy plays a significant role in structuring plant communities and has gained considerable attention due to its ecological importance and potential applications in agriculture, particularly in the development of sustainable weed management strategies (Saeed *et al.*, 2018; Macias *et al.*, 2000).

Allelochemicals are released into the environment through multiple pathways, including leaching, root exudation, volatilization, and decomposition of plant residues (Farooq *et al.*, 2008; Farooq *et al.*, 2011; Jabran *et al.*, 2010). Once released, these compounds

can disrupt key physiological and biochemical processes in targeted plants, thereby affecting seed germination, seedling establishment, and overall growth (Einhelling, 2002). In agricultural systems, weeds remain one of the most serious constraints to crop productivity, as they compete aggressively for nutrients, water, light, and space, resulting in substantial yield losses. In Pakistan alone, weed infestation causes estimated annual losses of approximately 28 billion PKR in wheat fields (Ali *et al.*, 2017; Khan *et al.*, 2018).

In this context, allelopathy has emerged as a promising and eco-friendly approach to weed management, as plant-derived compounds are generally biodegradable and pose fewer risks to the environment compared to synthetic herbicides. The exploration of allelopathic potential in wild and medicinal plant species, particularly those native to Balochistan, offers valuable opportunities for developing natural bioherbicides and sustainable weed management strategies (Macias *et al.*, 2007; Saeed *et al.*

al., 2018; Khamare et al., 2022). Phenolic compounds, which are widely distributed in plant tissues, are recognized as key allelochemicals that inhibit weed germination and growth through disruption of cellular processes (Saeed et al., 2018; Swain & Hillis, 1959).

Daphne mucronata Royle, a wild shrub of the family *Thymelaeaceae*, is widely distributed in the northern regions of Pakistan and parts of Iran. It typically grows in mountainous areas and along riverbanks at elevations ranging from 800 to 3000 m. The plant is characterized by densely hairy young branches, alternate leaves, and clusters of whitish flowers (Zaidi et al., 2015). Traditionally, it has been used to treat a variety of ailments, including skin disorders, rheumatism, allergies, and ulcers (Rasool et al., 2009; Malik et al., 2019). Phytochemical studies have identified bioactive compounds such as coumarins and diterpenes, which are associated with its medicinal properties (Khodadadian et al., 2016). Despite its pharmacological importance, its allelopathic potential remains insufficiently explored.

Convolvulus arvensis L. (field bindweed) is a perennial weed belonging to the family *Convolvulaceae* and is regarded as one of the most persistent and difficult-to-control species. It reproduces both through seeds and vegetative propagation via an extensive rhizomatous root system, enabling rapid spread and regeneration from small root fragments (Weaver & Walker, 1982). Its creeping growth habit and competitive ability allow it to interfere significantly with crop growth, making its management challenging (Kaur & Kalia, 2012). Similarly, *Asphodelus tenuifolius*, a member of the family *Asphodelaceae*, is also a problematic weed widely distributed in agricultural fields. It is well adapted to diverse environmental conditions and competes effectively with crops during early growth stages, leading to reduced productivity (Hussain et al., 2017). Due to its sensitivity to environmental changes and rapid growth, it serves as a suitable test species for evaluating phytotoxic effects.

Although synthetic herbicides are commonly used to control these weeds, their excessive use has led to environmental pollution, health concerns, and the development of herbicide resistance (Khamare et al., 2022). These challenges necessitate the development of alternative, sustainable weed management strategies. Balochistan, particularly the Sibi and adjacent regions, are rich in wild medicinal flora; however, the allelopathic potential of many native species, including *D. mucronata*, remains largely unexplored. This gap limits the effective utilization of

indigenous plant resources for eco-friendly weed control.

Therefore, the present study was conducted to evaluate the allelopathic effects of aqueous leaf extracts of *D. mucronata* on the germination, seedling growth, and biomass of *C. arvensis* and *A. tenuifolius*. Additionally, the study compares extracts from two ecologically distinct regions, Hazarganji, Quetta and Ziarat. The findings are expected to contribute to the development of sustainable and environmentally safe weed management approaches.

MATERIALS AND METHODS

Study Area: The present study was conducted using plant material collected from two ecologically distinct regions of Balochistan, Pakistan, representing natural habitats of *D. mucronata* (Fig.1). The selection of these sites was based on their ecological significance, altitudinal variation, and natural abundance of the target species.

Site I: Hazarganji National Park: Hazarganji National Park is located at 30°07' N latitude and 66°58' E longitude, approximately 20 km southwest of Quetta (Fig.1). The area covers about 15,555 hectares and lies at an elevation ranging from 1,600 to 3,000 m above sea level. The vegetation of the park is dominated by juniper (*Juniperus* spp.), almond (*Prunus dulcis*), pistachio (*Pistacia* spp.), and ash (*Fraxinus* spp.), along with a variety of wild medicinal plants.

Site II: Ziarat: Ziarat District is situated at 30°22'51" N latitude and 67°43'37" E longitude, at an elevation of approximately 2,453 m above sea level, covering an area of 999.60 hectares (Fig.1). The region is globally recognized for hosting one of the world's largest juniper forests (*Juniperus excelsa*) and supports a diverse flora of ethnobotanically important medicinal plant species.

Plant material collection: The taxonomic identification of the research plant (*Daphne mucronata* Royle) and the target weed seeds (*Convolvulus arvensis* and *Asphodelus tenuifolius*) was conducted and authenticated by co-author and visiting lecturer, Mr. Abdul Rehman, Department of Botany at Mir Chakar Khan Rind University (MCKRU), Sibi. His field expertise and familiarity deposited in the departmental herbarium of Mir Chakar Khan with the local flora ensured the accuracy of the botanical identification. A representative voucher specimens were deposited departmental herbarium of (MCKRU), Sibi, for future reference: Voucher No: MCKRU-BOT-2026/28-A (Hazarganji National Park Quetta) and Voucher No: MCKRU-BOT-2026/28- B (ziarat). (Fig. 2)



Figure 1. *Daphne mucronata* Royle collection sites map

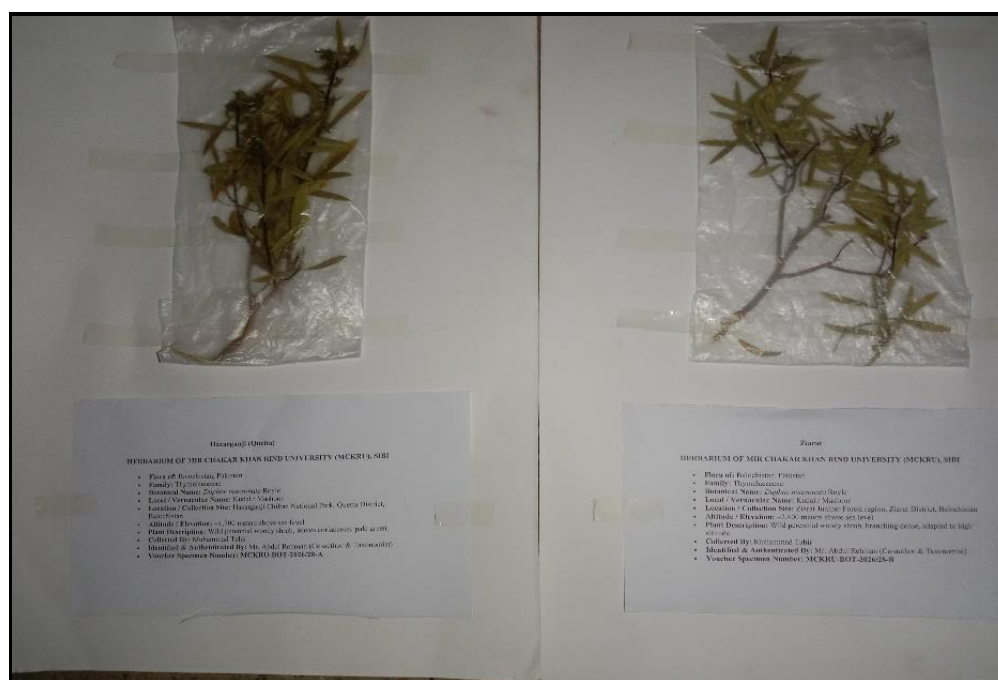


Figure 2. Voucher of *Daphne mucronata* Plant

Fresh and healthy leaves of *D. mucronata* were collected from both Hazarganji National Park and Ziarat District during April 2025 (Fig. 3). Care was taken to collect plant material from mature, disease-free individuals to ensure uniformity. The collected leaves were placed in sterile polyethylene bags, transported to the Botany lab MCKRU, Sibi and stored in refrigerator at 4 °C until further processing.

Seeds of the weed species *A. tenuifolius* (Fig. 4) and *C. arvensis* (Fig. 5) were collected from agricultural fields in the Dhadhar region of Balochistan during the same period. The seeds were air-dried, cleaned to remove debris, and stored in paper envelopes at room temperature further use. Additionally, a pre-germination viability test was performed, confirming a high seed viability rate of 80%.



Figure 3. *Daphne mucronata* Royle Plant



Figure 4. *Asphodelus tenuifolius* from Dhadhar city, Pakistan



Figure 5. *Convolvulus arvensis* from Dhadhar city, Pakistan

Morphological Characterization of *Daphne mucronata* Royle: To document phenotypic variation between populations from the two collection sites, selected morphological traits of *D. mucronata* were recorded. These included stem color and surface texture, leaf color, leaf length (cm), leaf width (cm), and shape, as well as floral characteristics such as flower length (cm) and number of flowers per inflorescence. Measurements were taken using a standard measuring scale, and observations were recorded to provide baseline information on morphological variability between the two ecological zones.

Preparation of Aqueous Leaf Extracts: Fresh, healthy leaves of *D. mucronata* were thoroughly washed with distilled water to remove dust and surface contaminants. The leaves were gently blotted dry and used immediately for extract preparation to preserve their natural phytochemical composition.

Aqueous extracts were prepared by weighing 8 g, 16 g, and 32 g of fresh leaf material separately and soaking each in 100 mL of double distilled water (DDW) to obtain concentrations of 8%, 16%, and 32% (w/v), respectively. Distilled water without plant material served as the control treatment (0%).

The mixtures were placed on an orbital shaker (inno-MC6micro) and agitated for 20 hours at room temperature ($25 \pm 1^\circ\text{C}$) to ensure proper extraction of water-soluble compounds, following the protocol described by Saeed *et al.* (2018) and Harborne (1984) with modification (fresh leaves used instead of dried leaves). After incubation, the extracts were filtered through four layers of cheesecloth to remove solid plant debris. The filtrates were then centrifuged at 5,000 rpm for 20 minutes to obtain clear supernatants. The supernatants were collected carefully and stored at 4°C until further experimental use. The pH of the extracts was monitored and maintained within the neutral range (6.8–7.4) to minimize any pH-related effects on seed germination, as recommended by Macias *et al.* (2000) and Saeed *et al.* (2018).

Phytotoxicity Bioassay: Seeds of *C. arvensis* and *A. tenuifolius* were surface-sterilized for 1–2 minutes using a 5% sodium hypochlorite aqueous solution (Oueslati, 2003; Saeed *et al.*, 2018) to prevent fungal contamination. The seeds were then rinsed thoroughly several times with distilled water and air-dried under sterile conditions. For the germination assay, sterilized 9 cm Petri dishes were lined with Whatman No. 1 filter paper. Ten seeds of each weed species were placed evenly in each Petri dish. Five milliliters of the respective extract concentration (0%, 8%, 16%, or 32%) were added to each dish, while distilled water was used for the control treatment (Saeed *et al.*, 2018). Each treatment was replicated three times in a

completely randomized design (CRD). The Petri dishes were sealed with parafilm to prevent moisture loss and incubated at $25 \pm 1^\circ\text{C}$ under a 16/8-hour light–dark photoperiod. Seed germination was recorded daily for seven days. After seven days, the final germination percentage (FGP%) was calculated using the formula of Bewley and Black (1986) as modified by Saeed *et al.* (2018): $\text{FGP}(\%) = (n/N) \times 100$ where n represents the number of germinated seeds and N represents the total number of seeds placed in each Petri dish. After seven days of incubation, seedling growth parameters were measured. Root length (RL) and shoot length (SL) were recorded in centimeters using a ruler with ± 0.1 cm precision. Fresh biomass was measured immediately after harvest, and dry biomass was determined after oven-drying seedlings to a constant weight.

Determination of Total Phenolic Content: Total phenolic content (TPC) of the aqueous extracts was determined using the Folin–Ciocalteu method as described by Saeed *et al.* (2018) and Swain & Hillis, (1959). Briefly, 1 mL of plant extract was mixed with 1 mL of Folin–Ciocalteu reagent and allowed to react for three minutes. Subsequently, 1 mL of 20% sodium carbonate (Na_2CO_3) solution was added, and the mixture was vortexed thoroughly. The reaction mixture was incubated in the dark for 30 minutes at room temperature. Absorbance was measured at 700 nm using a Shimadzu UV–visible spectrophotometer. Distilled water was used as a blank, and ferulic acid was employed as the standard. Total phenolic content was expressed as mg ferulic acid equivalents (FAE) per gram of extract.

Data Analysis: Data were collected from three replicates. Mean and standard deviation of average of three were taken for recording of results. Data were analyzed by using Microsoft Excel 2013.

RESULTS

Morphological Variation in *Daphne mucronata*: Morphological characteristics of *D. mucronata* collected from the two ecological zones, Hazarganji (HG) and Ziarat (ZI), showed slight but noticeable variation (Table 1). Stem color ranged from brown in HG samples to dark brown in ZI samples, while stem surface texture remained smooth across all specimens. Leaf color was uniformly green in plants from both locations. Internode length ranged from 0.7–0.9 cm in HG plants and 0.7–0.8 cm in ZI plants. Leaf length varied between 3.6–4.1 cm in HG and 2.4–4.3 cm in ZI populations, whereas leaf width ranged from 0.5–0.7 cm in HG and 0.4–0.6 cm in ZI samples. These observations indicate minor morphological variation between populations of two sites

Table 1. This table presents the quantitative data on morphological variations observed in plant samples of *D. mucronata* sourced from Hazarganji (Quetta) (HG) and Ziarat (ZI) locations, highlighting significant differences and trends in morphological responses.

S.No	Sample Code	Stem Color	Stem Surface	Internode(cm)	Leaf Color	Leaf Length(cm)	Leaf Width(cm)
1.	HG.01	Brown	Smooth	0.8	Green	3.6	0.7
2.	HG.02	Brown	Smooth	0.9	Green	4.1	0.5
3.	HG.03	Brown	Smooth	0.7	Green	3.8	0.6
4.	ZI.01	Dark brown	Smooth	0.7	Green	3.9	0.6
5.	ZI.02	Brown	Smooth	0.7	Green	2.4	0.4
6.	ZI.03	Dark brown	Smooth	0.8	Green	4.3	0.6

Effect of *Daphne mucronata* Extracts on Seed Germination %: Seed germination of both weed species declined steadily with increasing extract concentration (Fig. 6). Under control conditions (0%), germination remained high for both species, with HGAt showing 7.50 ± 0.71 seeds germinated and ZIAt showing 7.00 ± 0.00 seeds. As extract concentration increased to 32%, germination declined markedly. In *A. tenuifolius*, germination decreased to 4.50 ± 0.71 in HGAt and 3.50 ± 0.71 in ZIAt, indicating a stronger

inhibitory effect from the Ziarat extract. A similar trend was observed in *C. arvensis*. The ZICa treatment reduced germination from 7.50 ± 0.71 in the control to 4.00 ± 1.41 at 32% concentration. In general, Ziarat extracts (ZI) produced stronger inhibition than Hazarganji extracts (HG) for both weed species. Among the tested combinations, ZICa at 32% concentration exhibited the strongest inhibition, while the control treatments showed the highest germination rates

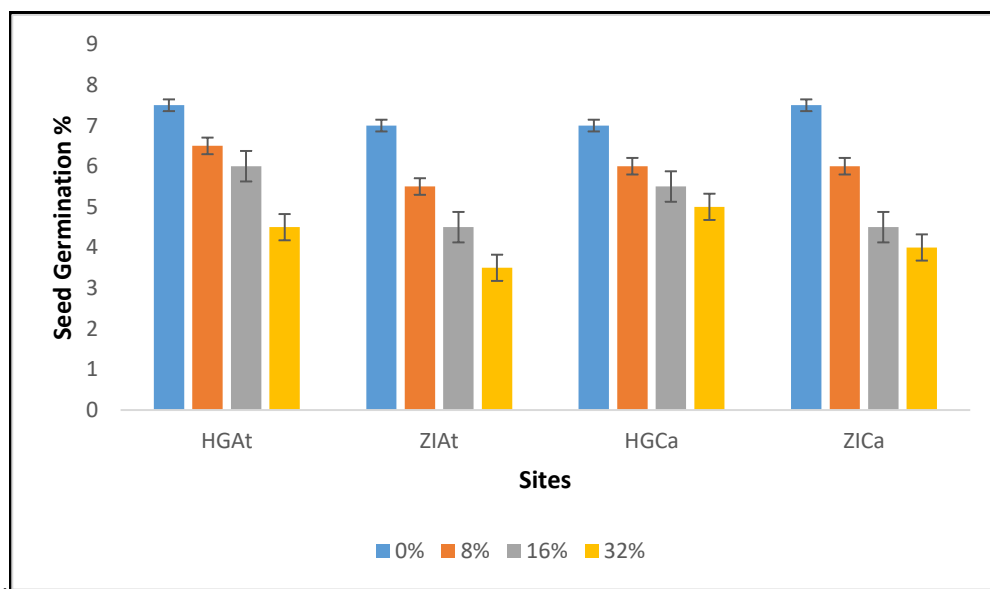


Figure 6. Allelopathic effect of *Daphne mucronata* extracts on the germination parameters of *A. tenuifolius* (HGAt: Hazarganji *A. tenuifolius*, ZIAt: Ziarat *A. tenuifolius*) and *C. arvensis* (HGCa: Hazarganji *C. arvensis*, ZICa: Ziarat *C. arvensis*) and all of the results are presented as mean $\pm$ SD. Mean values were derived from the performance of three replicates.

Effect on Shoot Length (cm): Shoot length (cm) of both weed species decreased significantly with increasing concentrations of *D. mucronata* extracts (Fig. 7). In *A. tenuifolius*, shoot length declined from 2.83 ± 0.09 cm (HGAt control) and 2.80 ± 0.08 cm (ZIAt control) to approximately 0.58 cm at the 32% concentration in ZIAt. A similar trend was observed

under both extract sources. In *C. arvensis*, ZICa treatment caused a reduction in shoot length from 3.28 ± 0.02 cm in the control to 0.28 ± 0.07 cm at the highest concentration. Overall, Ziarat-derived extracts exerted stronger inhibitory effects on shoot elongation than Hazarganji-derived extracts.

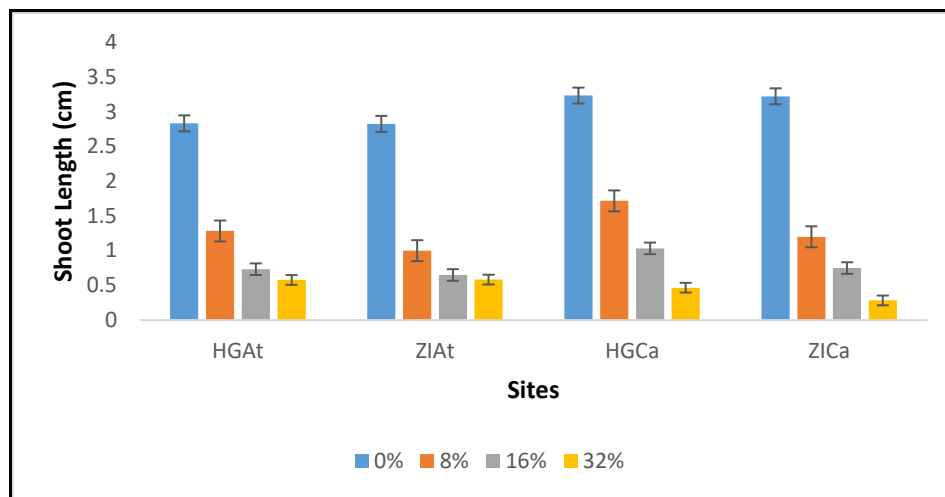


Figure 7. Allelopathic effect of *Daphne mucronata* extracts on the shoot length parameter of *A. tenuifolius* (HGAt: Hazarganji *A. tenuifolius*, ZIAt: Ziarat *A. tenuifolius*) and *C. arvensis* (HGCa: Hazarganji *C. arvensis*, ZICa: Ziarat *C. arvensis*) and all of the results are presented as mean $\pm$ SD. Mean values were derived from the performance of three replicates.

Effect on Root Length (cm): Root elongation was highly sensitive to the allelopathic extracts (Fig. 8). In *A. tenuifolius*, the control treatment showed a root length of 3.92 ± 0.02 cm, which decreased sharply with increasing extract concentration. At the 32% concentration, root length was reduced to 0.68 ± 0.07 cm in HGAt, demonstrating strong inhibition of root development. However, an even greater suppression

was observed in *C. arvensis* treated with Ziarat extracts. For ZICa, root length declined dramatically from 4.27 ± 0.09 cm in the control to only 0.42 ± 0.02 cm at the highest concentration, representing the most severe inhibition among all treatments. These results indicate that root growth was more sensitive to allelopathic stress than seed germination, particularly in *C. arvensis*.

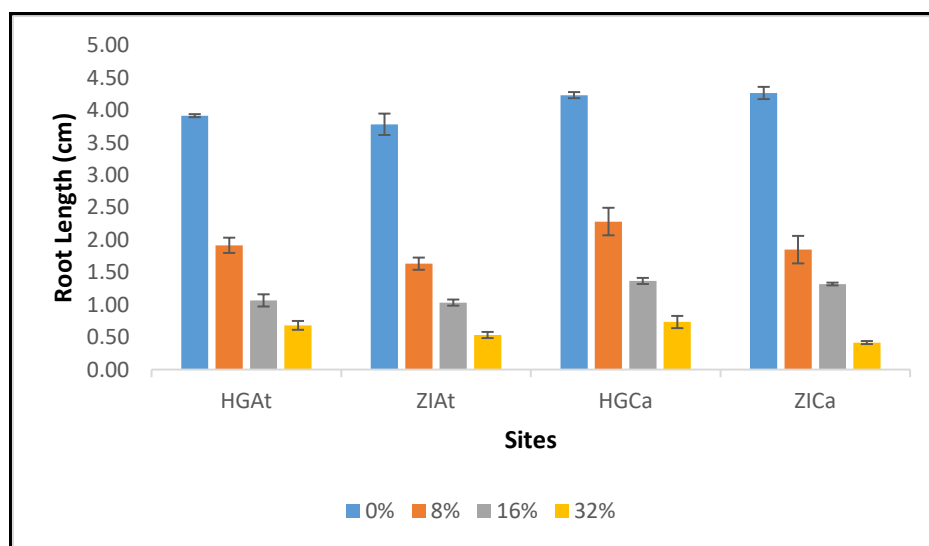


Figure 8. Allelopathic effect of *Daphne mucronata* extracts on the root length parameters of *A. tenuifolius* (HGAt: Hazarganji *A. tenuifolius*, ZIAt: Ziarat *A. tenuifolius*) and *C. arvensis* (HGCa: Hazarganji *C. arvensis*, ZICa: Ziarat *C. arvensis*) and all of the results are presented as mean $\pm$ SD. Mean values were derived from the performance of three replicates.

Effect on Fresh Shoot Weight (g): Fresh shoot biomass followed a similar decreasing pattern (Fig. 9). In *C. arvensis*, HGCa decreased from 0.240 ± 0.014 g in the control to 0.080 ± 0.014 g at the highest concentration, while ZICa reduced fresh shoot weight

further to 0.070 ± 0.014 g. For *A. tenuifolius*, the ZIAt treatment showed a decline from 0.210 ± 0.014 g to 0.060 ± 0.014 g at 32% concentration. Overall, Ziarat extracts produced slightly greater reductions in fresh biomass compared with Hazarganji extracts.

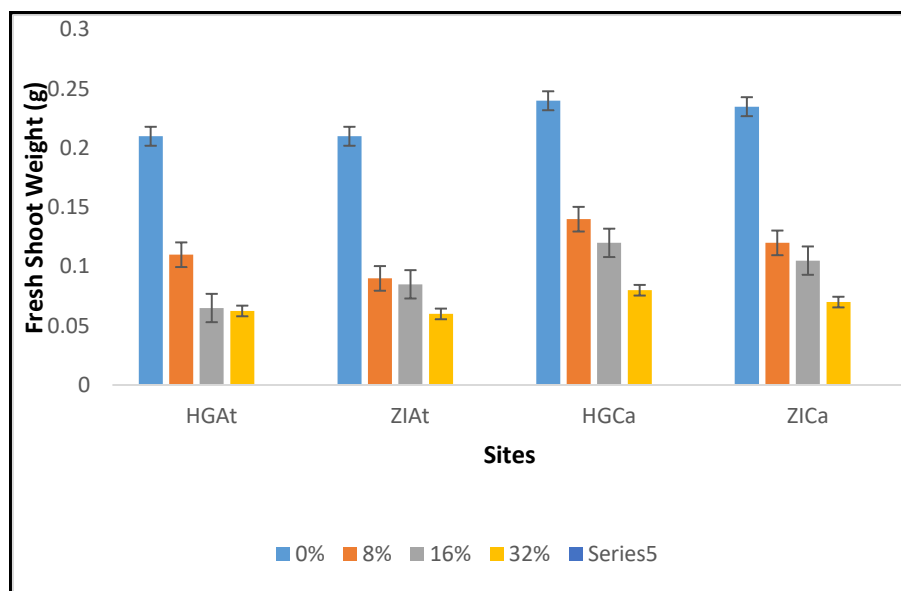


Figure 9. Allelopathic effect of *Daphne mucronata* extracts on the fresh shoot weight parameters of *A. tenuifolius* (HGAt: Hazarganji *A. tenuifolius*, ZIAt: Ziarat *A. tenuifolius*) and *C. arvensis* (HGCa: Hazarganji *C. arvensis*, ZICa: Ziarat *C. arvensis*) and all of the results are presented as mean $\pm$ SD. Mean values were derived from the performance of three replicates.

Effect on Fresh Root Weight (g): Fresh root biomass decreased substantially as extract concentration increased (Fig. 10). In *C. arvensis*, ZICa showed a reduction from 0.170 ± 0.014 g in the control to 0.035 ± 0.007 g at the 32% concentration. Similarly, in *A.*

tenuifolius, HGAt decreased from 0.105 ± 0.007 g to 0.045 ± 0.014 g over the same concentration range. These results indicate that root biomass was strongly suppressed by higher concentrations of the extract, especially those derived from Ziarat

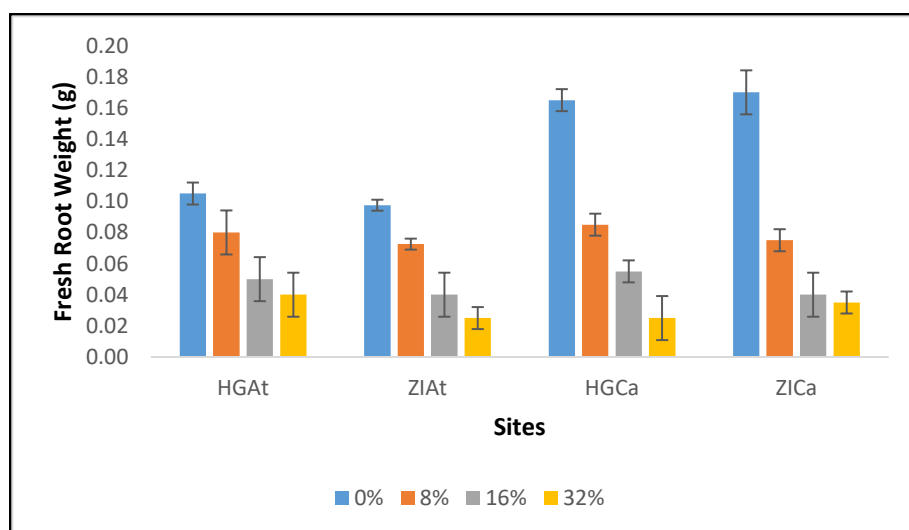


Figure 10. Allelopathic effect of *Daphne mucronata* extracts on the fresh root weight parameters of *A. tenuifolius* (HGAt: Hazarganji *A. tenuifolius*, ZIAt: Ziarat *A. tenuifolius*) and *C. arvensis* (HGCa: Hazarganji *C. arvensis*, ZICa: Ziarat *C. arvensis*) and all of the results are presented as mean $\pm$ SD. Mean values were derived from the performance of three replicates.

Effect on Shoot Dry Weight (g): Shoot dry weight was significantly suppressed with increasing extract concentration (Fig. 11). In *A. tenuifolius*, HGAt treatment reduced shoot dry weight from 0.0425 ± 0.0007 g in the control to 0.0085 ± 0.0007 g at the 32%

concentration. The strongest inhibition rate was observed in *C. arvensis* treated with ZICa, where shoot dry weight declined from 0.047 ± 0.002 g in the control to 0.0035 ± 0.0007 g at the highest concentration.

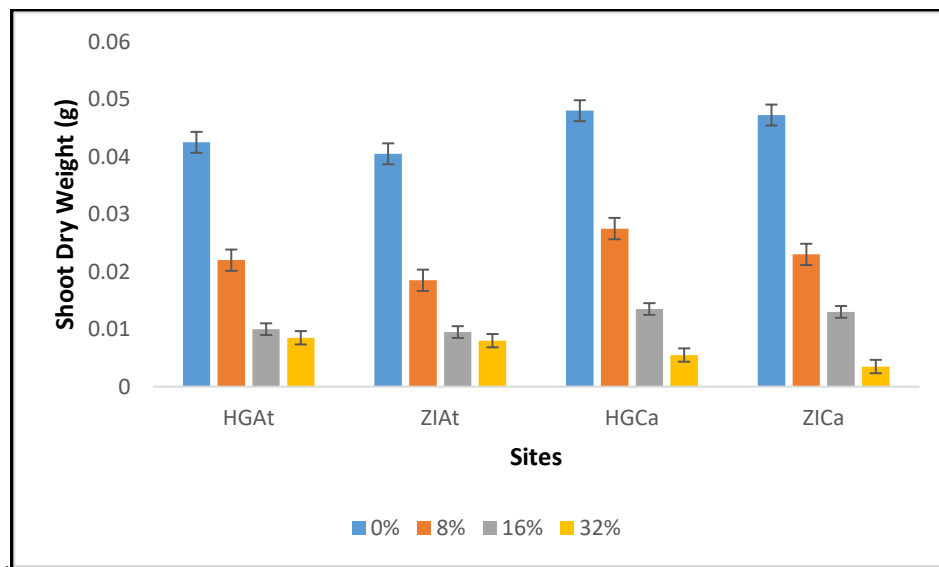


Figure 11. Allelopathic effect of *Daphne mucronata* extracts on the shoot dry weight parameters of *A. tenuifolius* (HGAt: Hazarganji *A. tenuifolius*, ZIAt: Ziarat *A. tenuifolius*) and *C. arvensis* (HGCa: Hazarganji *C. arvensis*, ZICa: Ziarat *C. arvensis*) and all of the results are presented as mean $\pm$ SD. Mean values were derived from the performance of three replicates.

Effect on Root Dry Weight (g): Root dry weight was markedly reduced by the aqueous extracts of *D. mucronata* (Fig. 12). In *A. tenuifolius*, HGAt and ZIAt treatments reduced root dry weight from 0.028 ± 0.002 g and 0.029 ± 0.001 g in the control to 0.0055 ± 0.0007

g at the 32% concentration in HGAt. Similarly, in *C. arvensis*, ZICa treatment reduced root dry weight from 0.036 ± 0.001 g in the control to 0.0045 ± 0.0007 g at the highest concentration.

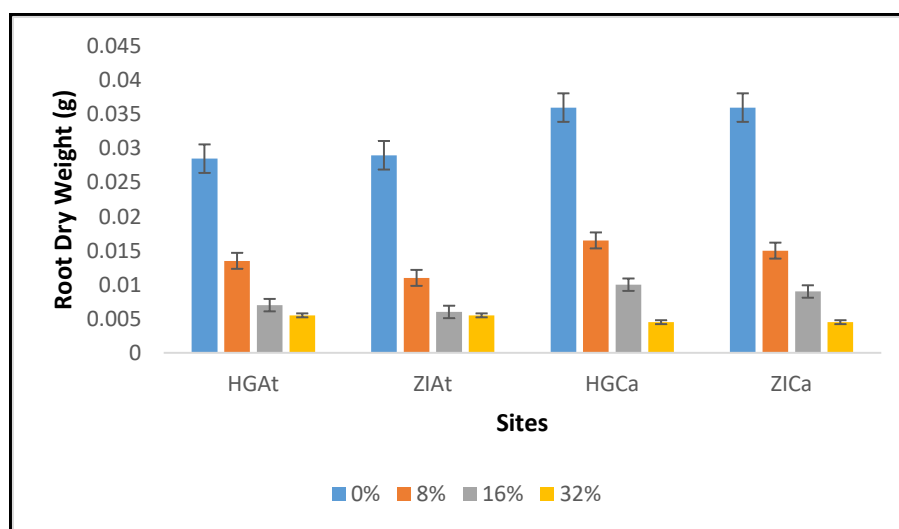


Figure 12. Allelopathic effect of *Daphne mucronata* extracts on the root dry weight parameters of *A. tenuifolius* (HGAt: Hazarganji *A. tenuifolius*, ZIAt: Ziarat *A. tenuifolius*) and *C. arvensis* (HGCa: Hazarganji *C. arvensis*, ZICa: Ziarat *C. arvensis*) and all of the results are presented as mean $\pm$ SD. Mean values were derived from the performance of three replicates.

Total Phenolic Content: Total phenolic content increased progressively with extract concentration, providing a biochemical basis for the observed allelopathic effects. (Table. 2) At 8% concentration, phenolic content was 0.17 mg FAE/g in HG and 0.19 mg FAE/g in ZI, indicating slightly higher accumulation in Ziarat samples. At 16%, this difference became more pronounced, with 0.25 mg FAE/g (HG) compared to 0.33

mg FAE/g (ZI). At the highest concentration (32%), phenolic content reached 0.35 mg FAE/g in HG and 0.38 mg FAE/g in ZI, representing the maximum values recorded. The increasing trend in phenolic content corresponds directly with the observed reduction in germination and growth parameters. The consistently higher values in Ziarat extracts suggest that greater phenolic accumulation enhances phytotoxicity, confirming their role as key allelochemicals

Table 2. Total phenolic content (mg FAE g⁻¹) of *D. mucronata* leaf aqueous extracts from Hazarganji (HG, Site 1) and Ziarat (ZI, Site 2) at three extract concentrations. Values are mean ± SD (n=3). Units: mg Ferulic Acid Equivalents per gram of extract (mg FAE g⁻¹).

Sites	8%	16%	32%
HG (Site 1)	0.17±0.03 mg FAE g ⁻¹	0.25±0.07 mg FAE g ⁻¹	0.35±0.12 mg FAE g ⁻¹
ZI (Site 2)	0.19±0.01 mg FAE g ⁻¹	0.33±0.17 mg FAE g ⁻¹	0.38±0.08 mg FAE g ⁻¹

DISCUSSION

Allelopathy is increasingly recognized as a sustainable approach to weed management, offering a viable alternative to synthetic herbicides. The present study demonstrated that aqueous leaf extracts of *D. mucronata* exert significant phytotoxic effects on seed germination, seedling growth, and biomass accumulation of *A. tenuifolius* and *C. arvensis*. The inhibitory effects were clearly concentration-dependent and varied between extracts obtained from different ecological zones, indicating that both extract concentration and geographical origin influence allelopathic activity.

The progressive reduction in seed germination with increasing extract concentration suggests that allelochemicals present in *D. mucronata* interfere with early physiological processes such as water uptake, enzyme activation, and hormonal regulation required for germination. This pattern aligns with established allelopathy literature, where bioactive secondary metabolites disrupt seed imbibition, radicle emergence, and subsequent seedling vigor by interfering with enzyme activity, hormone balance, and membrane integrity (Reigosa *et al.*, 2006; Macías *et al.*, 2020).

Root growth was found to be more sensitive than shoot growth in both weed species, indicating that roots are the primary targets of allelopathic interference. This higher sensitivity may be due to the direct and prolonged exposure of roots to allelochemicals, which can impair cell division, membrane integrity, and nutrient absorption. Comparable findings have been reported by Anwar *et al.* (2021), who observed stronger inhibition in root elongation than in shoot development.

The reduction in both fresh and dry biomass of root and shoot systems further confirms the strong inhibitory potential of *D. mucronata*. Biomass suppression reflects the cumulative effects of allelochemicals on physiological processes such as photosynthesis, respiration, and nutrient translocation. These results are consistent with previous studies reporting significant reductions in plant biomass due to allelopathic interference with metabolic and growth processes (Rahaman *et al.*, 2021).

Among the tested weed species, *C. arvensis* exhibited greater susceptibility compared to *A. tenuifolius*. This differential response may be attributed to species-specific physiological traits, seed coat permeability, or inherent tolerance mechanisms. Similar species-dependent variation in allelopathic sensitivity has been reported by Singh *et al.* (2005), indicating that the effectiveness of allelochemicals varies among target species.

A notable finding of this study was the stronger phytotoxic effect of extracts derived from Ziarat compared to those from Hazarganji. This variation may be associated with ecological factors such as altitude, temperature, soil conditions, and moisture availability, which influence the biosynthesis and accumulation of secondary metabolites in plants. Previous studies have shown that plants growing under environmental stress or high-altitude conditions often produce higher concentrations of bioactive compounds, resulting in enhanced allelopathic activity (Macias *et al.*, 2000).

The increase in total phenolic content with increasing extract concentration, particularly in Ziarat-derived samples, provides biochemical support for the observed phytotoxic effects. Phenolic compounds are well-known allelochemicals that inhibit germination and seedling growth through mechanisms such as oxidative stress induction, enzyme inhibition, and disruption of cellular signaling pathways. The positive relationship between phenolic content and phytotoxicity observed in this study is consistent with earlier findings (Anwar *et al.*, 2021). The comparatively higher phenolic content in Ziarat extracts suggests that these compounds play a major role in the allelopathic activity of *D. mucronata*.

Overall, the results confirm that *D. mucronata* possesses strong allelopathic potential, influenced by extract concentration, target weed species, and ecological origin. These findings contribute to the limited existing knowledge on this species and support its potential use in environmentally friendly weed management strategies.

CONCLUSION

The present study demonstrates that aqueous leaf extracts of *D. mucronata* exhibit significant allelopathic effects against *A. tenuifolius* and *C. arvensis*. All evaluated parameters, including seed germination, root and shoot length, and biomass production, were significantly reduced in a concentration-dependent manner.

Extracts collected from Ziarat showed consistently stronger phytotoxic effects than those from Hazarganji, highlighting the influence of ecological conditions on allelopathic potential. Root growth was more sensitive than shoot growth, and *C. arvensis* was more susceptible to the extracts than *A. tenuifolius*. The higher total phenolic content observed in Ziarat-derived extracts suggests that phenolic compounds are key contributors to the phytotoxic activity of *D. mucronata*.

Based on these findings, *D. mucronata* shows promising allelopathic activity under controlled

laboratory conditions and may warrant further investigation as a potential source of natural allelochemicals. However, these findings are preliminary and field-based validation, isolation of active compounds, and mechanistic studies are necessary before any practical application in sustainable weed management can be recommended.

AUTHORS' CONTRIBUTION

UA and MT conceived and designed experiment. MT performed the experiment. MT and AR collected the data. UA analyzed the data. UA and MT made the graphs and figures. MT and AR drafted the manuscript which was critically revised by UA and RA.

CONFLICT OF INTEREST

The All authors declare that there is no conflict of interest.

REFERENCE

- Anwar, S., et al. (2021). Bioherbicidal activity and metabolic profiling of potent allelopathic plant fractions against major weeds of wheat: Way forward to lower the risk of synthetic herbicides. *Frontiers in Plant Science*, **12**, 632390. <https://doi.org/10.3389/fpls.2021.709887>
- Ali, S., Tahir, M. N., Shah, G. A., Khan, M. A., & Mehmood, S. (2017). Dynamics of seed bank and weed flora in rain-fed wheat. *Pakistan Journal of Agricultural Sciences*, **54**(4), 907–917.
- Bewley, J.D. and M. Black. 1986. Seeds, physiology of development and germination, 2nd Ed. Plenum Press, New York.
- Einhellung, F.A.2002. The physiology of allelochemical action: clues and views. In M.J. Reigosa and N. Pedrol. (eds.) Allelopathy from Molecules to Ecosystem. Science Publishers, Enfield, New Hampshire, USA.
- Farooq, M., K. Jabran, H. Rehman and M. Hussain. 2008. Allelopathic effects of rice on seedling development in wheat, oat, barley and berseem. *Allelopath. J.*, **22**: 385-390.
- Farooq, M., K. Jabran, Z.A. Cheema, A. Wahid and H.M. Siddique. 2011. The role of allelopathy in agricultural pest management. *Pest Manage. Sci.*, **67**: 493-506.
- Hussain, M. I., & Reigosa, M. J. (2017). Evaluation of photosynthetic performance and carbon isotope discrimination in perennial ryegrass (*Lolium perenne* L.) under allelochemical stress. *Ecotoxicology*, **26**(5), 613–624.
- Harborne, J. B. (1984). *Phytochemical methods: A guide to modern techniques of plant analysis* (2nd ed.). Chapman & Hall
- Jabran, K., M. Farooq, M. Hussain, H. Rehman and M.A. Ali. 2010. Wild oat (*Avena fatua* L.) and canary grass (*Phalaris minor* Retz.) management through allelopathy. *J. Plant Prot.Res.*, **50**: 32-35.
- Kaur, M., & Kalia, A. N. (2012). *Convolvulus arvensis*: A useful weed. *International Journal of Pharmacy and Pharmaceutical Sciences*, **4**(1).
- Khamare, Y., Chen, S., & Liu, X. (2022). Allelopathy and its application as a weed management tool: A review. *Frontiers in Plant Science*, **13**, 1034649. <https://doi.org/10.3389/fpls.2022.1034649>
- Khan, S., Shinwari, M. I., Haq, A., Ali, K. W., Rana, T., Badshah, M., & Khan, S. A. (2018). Antifungal activity of methanolic plant extracts. *Pakistan Journal of Botany*, **50**(4), 1591–1598.
- Khodadadian, Z., Hassanpour-Ezatti, M., Mousavi, S. Z., & Asgarpanah, J. (2016). Analgesic and anti-inflammatory potential of aerial parts of *Daphne mucronata* Royle extract in mice: Opioid-independent action. *Asian Pacific Journal of Tropical Biomedicine*, **6**(3), 202–207. <https://doi.org/10.1016/j.apjtb.2015.12.004>
- Macias, F.A., D. Castellano and J.M.G Molinillo. 2000. Search for a standard phytotoxic bioassay for allelochemicals. Selection of target species. *J Agric. Food Chem.*, **48**:2512–2521.
- Macias, F. A., Molinillo, J. M., Varela, R. M., & Galindo, J. C. 2007. Allelopathy: A natural alternative for weed control. *Pest Management Science*, **63**(4), 327–348.
- Macias, F.A., Dur'an, A.G., Molinillo, J.M., 2020. Allelopathy: the chemical language of plants. *Prog. Chem. Org. Nat. Prod.* **112**, 1–84.
- Malik, K., Ahmad, M., Zafar, M., Ullah, R., Mahmood, H. M., Parveen, B., Rashid, N., Sultana, S., Shah, S. N., & Lubna. (2019). An ethnobotanical study of medicinal plants used to treat skin diseases in northern Pakistan. *BMC Complementary and Alternative Medicine*, **19**(1), 210.
- Oueslati, O. 2003. Allelopathy in two durum wheat (*Triticum durum* L.) varieties. *Agric. Ecosyst. Environ.*, **96**:161–163
- Rahaman, F., Juraimi, A. S., Raffi, M. Y., Uddin, K., Hassan, L., & Bashar, H. M. K. (2021). Allelopathic effect of selected rice (*Oryza sativa*) varieties against barnyard grass (*Echinochloa crus-galli*). *Plants*, **10**(10), 2017.
- Rasool, M. A., Imran, M., Nawaz, H., Malik, A., & Kazmi, S. U. (2009). Phytochemical studies on *Daphne mucronata*. *Journal of the Chemical Society of Pakistan*, **31**(5).
- Reigosa, M.J., Pedrol, N., Gonz'alez, L., 2006. Allelopathy: A Physiological Process with Ecological Implications. Published by Springer, Dordrecht, The Netherlands.
- Saeed, S., Ahmed, A., Shah, S. H., Begum, S., Zeeshan, M., & Khan, W. (2018). Phytotoxic potential of wild medicinal plants from different altitudinal gradients of Quetta Balochistan Pakistan on *Convolvulus arvensis* L. *Pakistan Journal of Weed Science Research*, **24**(4), 323–334.
- Singh, H. P., Batish, D. R., Shalinder, K., Kohli, R. K., & Dogra, K. S. (2005). Allelopathic interference of *Ageratum conyzoides* L. against some crop plants. In *Weeds management: Balancing people, planet, profit* (Proceedings of the 14th Australian Weeds Conference, Wagga Wagga, New South Wales, Australia, pp. 558)
- Swain, T., & Hillis, W. E. (1959). The phenolic constituents of *Prunus domestica* I: The quantitative analysis of phenolic constituents. *Journal of the Science of Food and Agriculture*, **10**, 63–66.
- Weaver, S. E., & Walker, R. R. (1982). The biology of Canadian weeds: *Convolvulus arvensis* L. *Canadian Journal of Plant Science*, **62**, 461–472. <https://doi.org/10.4141/cjps82-066>
- Zaidi, A., Bukhari, S. M., Khan, F. A., Noor, T., & Iqbal, N. (2015). Ethnobotanical, phytochemical, and pharmacological aspects of *Daphne mucronata* (Thymeleaceae). *Tropical Journal of Pharmaceutical Research*, **14**(8), 1517–1523.

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