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Allelopathic Effect of Three Wild Species on Seed Germination and Seedling Growth of *Vicia faba*, *Hordeum vulgare* and *Triticum aestivum*

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Author's contribution

The sole author designed, analyzed and interpreted and prepared the manuscript.

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ABSTRACT

Many wild and cultivated grasses, exhibit allelopathy to exclude the associated species by reducing their regeneration. This process involving secondary metabolites produced by plant influence the growth and development of agricultural and biological system. This research was to determine the allelopathic potential of aqueous extracts of different wild species (*Calligonum comosum* L, Her, *Rhanterium epapposum* Oliv and *Rhazya stricta* Decne) on seed germination and seedling growth of three crops (*Vicia faba*, *Hordeum vulgare* and *Triticum aestivum*). Effect of water extracts from dry aboveground plant biomass in concentration of 10% (100 g/L) was examined under laboratory conditions in petri dishes. Germination indicators (Germination Percentage (GP) and germination start) were evaluated. The results showed that all the extracts significantly decreased germination percentage, plumule and radicle length of seedlings. However, leaf extracts of *R. stricta Decne* had greater inhibitory potential and reduced seedling growth. The significant allelopathic effect remained up to 10 days. The extract had strong inhibitory effect to root elongation of seedling of plants. Differences in sensitivity between plant cultivars were documented. Based on the study

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results, stem, leaves and flowers residues of (*C. comosum* L, Her, *R. epapposum* and *R. stricta* Decne) showed a negative allelopathic effects on plant growth and should be eliminated from the field.

Keywords: Allopathic; wild plants; Vicia faba; Hordeum vulgare; Triticum aestivum.

1. INTRODUCTION

Allelopathy is the interactions among plants, bacteria, fungi and algae with the living organisms in a certain environment. Interaction that are mediated by production of secondary metabolites release by plants or microorganisms into the environment, are stimulatory or inhibitory and produce biological changes [1,2]. Allelochemicals usually are called secondary plant product or waste products of the main metabolic pathways in plants. These chemicals are the major factor in regulating the structure of plant communities [3,4]. Allelochemicals interference must be species-specific to explain why nonindigenous species dominate an invaded community while they normally do not reach high dominance in their native community. Most research on allelopathy has focused on the effect of allelochemicals as weed control [5-9]. Identification of allelopathic potential of wild plants on growth and yield of crops leads to better understanding of their negative impact on plants which will help to control invasive plants. However, [10] reported that the difficult to find noticeable effect in the established communities because of possible adaptation of co-existing species to allelochimicals released by competitors. *Calligonum comosum* L, Her, *Rhanterium epapposum* Oliv and *Rhazya stricta* Decne are a wild plants, which naturally grows on the plains and hilly areas of Saudi Arabia and neighboring countries.

Calligonum comosum L, Her, locally known as (Alarta) belongs to the botanical family Polygonaceae (Fig. 1). It is a plant of tropical and subtropical regions and is wide spread in Saudi Arabia and Arabian Peninsula. Several studies have confirmed the anti-bacterial activity and benefits of *C1. comosum* leaves in the treatment of certain skin diseases [11,12,13].

Rhanterium epapposum Oliv, locally known as (Alarfaj) belongs to the botanical family Asteraceae (Fig. 2). It is a C_3 desert shrub that can form monotonous stands covering vast areas of north-eastern Arabia [14]. It grows as a perennial woody shrub approximately 80 cm high with many stems branching, small and narrow leaves and straw- yellow flowers. It is grow mainly in deep sandy soil [15].

Fig. 1. Alarta (*Calligonum comosum L,* Her)

Fig. 2. Alarfaj (*Rhanterium epapposum* Oliv)

Rhazya stricta Decne, locally known as (Alharmal) belongs to the botanical family Apocynaceae (Fig. 3). It is a desert evergreen perennial sub-shrub. It is poisonous, thus not browsed by livestock. It is a valuable medicinal plant as it contains several chemical compounds and has ethno-medicinal uses world-wide. Despite its medicinal importance, *R. stricta* is a troublesome plant as it invades deteriorated rangelands. *R. stricta* species have harmful effects on crops including reduced seed germination [16,17,18].

Fig. 3. Alharmal (*Rhazya stricta* Decne*)*

The aim of this study was to determine the possible allelopathic effects of commonly distributed wild species in Saudi Arabia (*C. comosum* L, Her, *R. epapposum* Oliv and *R. stricta* Decne) on germination and seedling growth of *Vicia faba*, *Hordeum vulgare* and *Triticum aestivum*.

2. MATERIALS AND METHODS

2.1 Collection of Plant Material and Preparation of Extracts

The profile of three wild plants used in this study is shown in (Table 1). Fresh plant material used in this study were: *C. comosum* L, Her, *R. epapposum* Olive and *R. stricta* Decne collected from the north-east of Riyadh city, in "Dahna" area in Saudi Arabia. The fresh leaves and roots were cut into small pieces, washed in distilled water and air-dried in 75% shade at room temperature. After drying, the plant materials were ground to powder and extracted with hot water, 10 g of the powdered sample was suspended in 100 ml of distilled hot water. The samples were then shaken and allowed to settle at room temperature for 24 hrs in hot water. The solutions were filtered thought a double layers of muslin cloth followed by a Whatman No. 1 filter paper, the pH values were adjusted to 7 with 1.0 M HCl, these were kept in refrigerator at 4 C^{o} until further use [19].

The osmotic concentrations of bioassay solutions were less than 0.1 Mpa and hence not considered a factor affecting germination [20]. Seeds of *Vicia faba*, *Hordeum vulgare* and *Triticum aestivum* were collected from the different localities of study area, sterilized by 0.3% sodium hypochlorite, rinsed by distilled water and shade dried again on the filter paper in the laboratory at room temperature for 7 days according to [21].

2.2 Preliminary Phytochemical Screening

The plant materials were screened for the presence of different classes of secondary metabolites including alkaloids, flavonoids, phenols, saponins and tannins using previously described methods [22].

2.3 Germination and Growth Bioassays

Two layers of Whatman No. 1 filter paper were placed in 90 mm diameter glass petri dishes. In each petri 20 seeds were placed and 10 mm of each plant extract added. A check treatment was assigned with distilled water and let at room temperature. Starting from the first day after experiment set on, germinated seeds were counted and removed daily. A seed with 0.5 cm of radical was considered germinated. The plume and radicle lengths of seedlings were measured on 10 day after treatment (DAT).

Experiment designed was Randomized Complete Block design with four replicates and experiment repeated twice and the percentage of germination was calculated.

2.4 Germination and Growth Indicators

Percentage of Germination (PG) and germination start compounds (GS) were calculated according to the following extract formulas:

1) Percentage of germination = 100(n/N).
2) Germination start = The long time between seed.
3) Where N is the number of seeds germinated on day i and T is the number of days from sowing.

Table 1. Used part of the selected plants

Botanical name	Family	Local name	Part used	Extract dry weight (g ml^{-1})
*Calligonum comosum L'*Her	Polygonaceae	Alarta	Stem	10
Rhanterium epapposum Oliv	Asteraceae	Alarfaj	Flower	10
Rhazya stricta Decne	Apocynaceae	Alharmal	Leaves	10

3. RESULTS AND DISCUSSION

3.1 Phytochemical Analysis

Freshly prepared extracts were subjected to preliminary phytochemical screening for various constituents. The results (Table 2) revealed the presence of phenols, polyphenols and alkaloids in *C. comosum*, *R. epapposum* and *R. stricta* Decne, Tannis and saponins were also detected in the extracts except in the extract of R. *epapposum*. The chemical compounds such as Tannins, Trepenoids, Phenylpropans, Acetogenins, Terpenoids, Steroids, Alkaloids, may play an important role in plant-plant interferences [1, 23, 24].

3.2 Effect of Aqueous Extracts of Wild Plants on Germination of *Vicia faba, Hordeum vulgare and Triticum aestivum* Growth

The allelopathic effect of aqueous extracts of *C. comosum*, *R. epapposum* and *R. stricta* Decne on the germination percentage of *Vicia faba, Hordeum vulgare and Triticum aestivum* after 5 days after treatments (DAT) are shown in Table 3. After 5 days of treatment, significant germination reduction was observed with all extracts ($P \leq 0.05$). The *R. stricta* Decne extracts exhibited higher negative effective on seed germination of 22% and 30% for seed of *Vicia faba and Triticum aestivum*, except *Hordeum vulgare,* there was complete failure of seed germination of *R. stricta* Decne aqueous extract. All aqueous extracts of wild plants significantly reduced seed germination compared with distill water control treatments (Table 3).

The inhibition of germination was found strong in extract of *R. stricta* Decne, was the most potent inhibitor of seed germination; in agreements with [18] found that, allelopathic effects of *R. stricta* Decne aqueous leachate from leaves, stems and follicles reduced the germination of *A. leucoclada*, *L. scindicus* and *S. villosa.*

The probable reason for *R. stricta* Decne potent allelopathic activity may be due to the presence of many bioactive secondary metabolites in this plant, particularly phenolics and alkaloids (Table 2). According to [25,17] the extract of *R. stricta* Decne plant contain alkaloids, that may be cause significant reductions in the germination of the seed of many plants [16]. In addition, [26] demonstrated that leaf aqueous extracts of *R. stricta* Decne plant exhibited significant inhibitory effects on germination of lettuce seeds.

Moreover, allolopathic potential could be attributed to the bioactive compounds such as phenolic and flavonoid contents [27,28,29]. From preliminary screening it was found that leaf extract of *R. stricta* Decne had the strongest allelopathic effect on seed germination compared to the stem extract of *C. comosum* and flower extract of *R. epapposum*. The results are agreed with [30] they reported that the most inhibitory effect of allelopathic plants produced by leaf extract. Moreover, [31] found that high degree of inhibition occurred with leaves extracts at the highest concentrations in all tested crop plants. In addition, [32] also reported that the inhibitory allelopathic potential of leaf extract was more powerful than of other vegetative parts. Furthermore, negative impact of allelopathic compounds from the aqueous extract of *C. comosum* stem was predictable, reduction of the low germination percentage and the decreased of Plumule and radical length detected when compared with the control (Tables 3, 4 and 5). [33] reported that the genus *Calligonum* revealed the presence of new lipoxygenase flavonoids, alkaloids, proteins, tanins, steroids, phenols and terpenoids [34]. Moreover, [35], reported that the *Calligonum polygonoides* extract at concentrations (75 g/L) inhibited seed germination of *Echinochloa crus-galli* by about 60.42%, while it inhibited the root and shoot growth by about 99.12% and 67.10%, respectively.

In this study, the indirect association between lower seed germination and allelopathic inhibition may have resulted to inhibition of water uptake

Table 2. Phytochemical composition of selected plant species

Treatment	Phenols	Polyphenols	Tannins	Saponins	Alkaloids	Flavonoids
Calligonum comosum L, Her	+	+	+	+	+	+
Rhanterium epapposum Oliv	+	+	-	-	+	+
Rhazya stricta Decne	+	+	+	+	+	+

(+): Present; (-): Absent

Table 3. Effect of different plant extracts on germination percentage after 5 days

Treatment	*Vicia faba*	*Hordeum vulgare*	*Triticum aestivum*
*Calligonum comosum L'*Her	47±0.35	60±0.40	70±0.60
Rhanterium epapposum Oliv	40±0.65	50±0.25	60±0.59
Rhazya stricta Decne	22±0.58	NG	30±0.33
Control	90±0.45	95±0.32	85±0.51
$LSD_{0.05}$	3.3	4.1	3.7

**: mean value ± standard error. Different superscript letters indicate values significantly lower than the respective control (P ≤ 0.05). NG= No Germination*

Table 4. Effect of different plant extracts on the plumule length (cm) after 10 days

Treatment	*Vicia faba*	*Hordeum vulgare*	*Triticum aestivum*
*Calligonum comosum L'*Her	0.36±0.02	0.08±0.40	1.68±0.03
Rhanterium epapposum Oliv	3.55±0.53	0.51±0.06	2.03±0.08
Rhazya stricta Decne	0.24±0.49	NG	0.33±0.63
Control	5.24±0.40	8.03±0.14	5.25±0.06
$LSD_{0.05}$	0.3	0.1	0.2

**: mean value ± standard error. different superscript letters indicate values significantly lower than the respective control (P ≤ 0.05). NG= No Germination*

Table 5. Effect of different plant extracts on the radical length (cm) after 10 days

Treatment	*Vicia faba*	*Hordeum vulgare*	*Triticum aestivum*
Calligonum comosum L, Her	2.93±0.12	3.14±0.17	2.56±0.85
Rhanterium epapposum Oliv	1.45±0.32	1.53±0.00	1.25±0.02
Rhazya stricta Decne	0.14±0.19	NG	0.06±0.25
Control	4.54±0.53	10.66±0.03	5.76±0.44
$LSD_{0.05}$	0.2	0.1	0.2

**: mean value ± standard error. Different superscript letters indicate values significantly lower than the respective control (P ≤ 0.05). NG= No Germination*

and enzyme activity. Although enzyme activity was not analyzed in this study, this observation is in accordance to the study of [36] reported that, increasing the concentration of aqueous leaf extracts significantly inhibited the water uptake by germination of *Rhanterium epapposum* seeds.

Moreover, [1] reported that many enzymatic functions important to plants are inhibited by the presence of allelochemicals, such as protease which plays an important role in the hydrolysis of proteins during germination. Usually, the allelo-chemicals inhibit seed germination by blocking hydrolysis of nutrients reserve and cell division and cause significant reductions in the growth of plumule and radical of various crops [37,38].

3.3 Effect of Aqueous Extracts of Wild Plants on Length of Plumule and Radicle of *Vicia faba, Hordeum vulgare and Triticum aestivum* Growth

The allelopathic effect of wild plants extracts on plumule growth of *Vicia faba*, *Hordeum vulgare* and *Triticum aestivum* are shown in (Table 4). The leaves extracts of *R. stricta* Decne were the most effective extract as it inhibit plumule length of seedling was about 0.24 and 0.33 cm for plumule of *Vicia faba and Triticum aestivum*, and the *Hordeum vulgare,* there was failure of seed germination of *R. stricta* Decne aqueous extract. Generally, all extracts of the tested wild plants significantly reduced the plumule length at depended manner. The allelopathic activities of water extracts from tested wild plant on radical growth of *Vicia faba*, *Hordeum vulgare* and *Triticum aestivum* seedling are shown in (Table 5). The extracts of all wild plants showed inhibition of radical length. The extract of *R. stricta* Decne was the most effective as the radicle length was 0.14 and 0.06 cm for radicale of *Vicia faba and Triticum aestivum*, and the *Hordeum vulgare,* there was failure of seed germination of *R. stricta* Decne aqueous extract. Plumule and radical length was significantly reduced by plants extracts when compared with control treatments (Tables 4 and 5). The radicle length was relatively more sensitive to autotoxic allelochemicals from extracts than plumule length. These results are in agreement with [36] reported that water extracts of allelopathic plants were more pronounced effects on radicle growth

than Plumule growth. Because it is likely that roots are the first to absorb the allele-chemicals or autotoxic-compounds from the environment.

4. CONCLUSION

The results concluded that the wild plants plays an important role in the formation of its natural habitats as it contains the allele-chemical compounds that enable the plant to compete with other species. Moreover, it may change communities when recycled as a green manure in the soil for increasing organic matter in agro-ecosystems, where it inhibits crop growth and production. Moreover, the present results indicated that higher concentrations of extracts of tested species could be used for weed control. Since the experiment was conducted only in Petri dishes, further studies in pots or under field conditions are necessary to fully determine plants allelopathic potential effects. Also needed to determine the influence of seasonal and cultivar variations, and to identify the active compounds of autotoxicity and allelopathy effects.

COMPETING INTERESTS

Author has declared that no competing interests exist.

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