

“Evaluation of allelopathic potential of Weedy Species of *Vicia* on Gram (*Cicer Arietenum*)”

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Abstract

This research was conducted to evaluate the allelopathic effects of aqueous extracts of shoot and root of weedy species of *Vicia* (*Vicia hirsuta* and *Vicia sativa*) on seed germination, seedling length and dry weight of Gram (*Cicer arietinum*). Root and shoot aqueous extracts of *Vicia hirsuta* and *Vicia sativa* at 5%, 10%, 15% and 20% concentrations were applied to determine their effect on seed germination and seedling growth of test plants under laboratory conditions. The aqueous extracts reduced all the test parameters in 7 days old Gram plants. Results showed that the allelopathic activity of weedy species of *Vicia* inhibited both the germination and seedling growth of Gram. The degree of inhibition in terms of both germination and seedling growth was directly proportional to the concentration of aqueous extracts. It was observed that they extracts from shoots had a more inhibitory effect than root extract. This result confirmed the better growth of these weeds during initial stage of growth in rabi fields.

Keywords: *Allelopathy ; Aqueous extracts; Gram; Inhibition; Vicia hirsuta ;Vicia sativa.*

Introduction

The word allelopathy was first used by Hans Molisch in 1937 when describing the beneficial and deleterious chemical interactions of plants and microorganisms (Willis,1985). It is any direct or indirect effect of one plant on another mediated through the production of chemical compounds that escape into the environment (Rice 1974). The effect may be beneficial or harmful on both the crop and weed species by the release of chemicals from plant parts by leaching, root exudation, volatilization, residue decomposition, and other processes in both natural and agricultural systems (Ferguson and Rathinasabapathi, 2003). It is essentially a chemical process used by plants to keep other plants out of their space.

Allelopathic plants prevent other plants from using the available resources and thus influence the evolution and distribution of other species. Allelochemicals (inhibitors) are produced by plants as end products, by-products and metabolites and are contained in the stem, leaves, roots, flowers, inflorescence, fruits and seeds of the plants (Sisodia and Siddiqui, 2010). The commonly cited effects of allelopathy includes reduced seed germination and seedling growth, however, it may affect cell division, pollen germination, photosynthesis and various enzyme functions.

The present study deals with the interaction between weeds and cultivated plants which is simultaneous and/or subsequent with direct or indirect impact of one plant species on another, through synthesis of different chemical compounds – allelochemicals (Kadioglu et al., 2005; Verma and Rao, 2006; Aleksieva and Serafimov, 2008). Number of studies were conducted (Vasilakoglou et al., 2006; Ashrafi et al., 2007 and Koloren, 2007) to determine the allelopathic interference between weeds and cultivated plants, (Adetayo et al., 2005; Kayode and Ayeni, 2009).

The genus *Vicia L.* (Leguminosae, Fabaceae) comprises about 180 species extensively distributed in the temperate zone of both southern and northern hemisphere (Hanet and Mettin 1989). This is an important genus and several of its species are grown in India and abroad because of its high protein content and are used as pulses, vegetables, and as fodder. Chemical analysis of *Vicia* species shows that it contains NaHCO_3 , Na_2CO_3 , Na_3PO_4 , protein carbohydrates and fats (Al-Nouri and Siddiqui, 1981). Of the many species of *Vicia*, *Vicia hirsuta* and *Vicia sativa* are the most common and dominant weed species associated with rabi crops such as wheat and Gram. It has been observed that these weeds are present in abundance in rabi crop fields and cause considerable damage to the crop yields. Due to their competitive nature they reduce the availability of nutrients to crops thereby reducing productivity. Since, not much research has been done in this area with specific reference to the effects of these weeds over rabi crops, this study was undertaken with main objective to examine, evaluate, and observe the allelopathic potential of two weedy species of *Vicia* on Gram which is a major constituent of rabi crops in India. This study particularly observes the heterotoxicity of *Vicia hirsuta* and *vicia sativa* on Gram plants.

MATERIALS AND METHODS

This study was carried out in the Dept. of Botany, R. D and D. J. College. The plants of *Vicia hirsuta* and *Vicia sativa* at their mature stage were collected from around R. D. and D. J. College campus (24° 30' N, 86° 30' E and 45 m above sea level).

Preparation of aqueous extract

The weedy species of *Vicia*, namely *Vicia hirsuta* and *Vicia sativa* were uprooted at their mature stage from around R. D. and D. J. College campus during February and March 2007 for conducting experiments. The plants were separated into shoot and root respectively. The respective organs were chopped into small pieces and 10 gm of each organ was crushed separately. The crushed material was then mixed with 100 ml of distilled water and grinded in a mixer to prepare extracts. The mixture was stored for 72 hrs at room temperature and then filtered through Whatman (No.1) filter paper. The filtrate was brought to 100 ml with addition of water to make the stock solution. From this stock solution the desired concentration (5% , 10 % , 15 % and 20 %) respectively were prepared by further dilution with distilled water as adopted from Singh et al. (1989) .

Bioassays and experimental design

A number of 25 seeds of Gram were placed in a sterile glass petridish (10 cm dia.) lined with single layer of filter paper. The seeds surface was sterilized by dipping in 0:1 (w/v) Hgcl₂ and rinsed many times in distilled water before use. 10 ml solution of aqueous extract was added per petridish and seeds were allowed to germinate at room temperature (20⁰ ± 2⁰ C) for 7 days. Each variant was laid out in three replications. Distilled water was used as a control. The samples were

opened periodically for aeration. Gram seedlings were harvested for measurement after 7 days of growth and following characteristics were determined: percentage of seed germination (%); linear growth of radicle and plumule (cm); dry weight of radicle and plumule (mg).

Statistical analysis

The experiment was established in a factorial design, completely randomized with 3 replications. Seed germination and linear growth of radicle and plumule of Gram were measured after 7 days. On account of very low dry weight /Gram seedling, 10 seedlings were dried together instead to constant dry weight at 80⁰ C for 72 hr in a hot air oven, weighed and mean dry weight per seedling were calculated. Mean dry weight was used for computing standard deviation. The data were subjected to students' t' test to see significance (Konis, 1940).

RESULTS

The aqueous extracts from shoot and root of *Vicia* spp. showed an inhibitory effect on all tests parameters viz. seed germination, seedling length and seedling dry weight of Gram (table 1, 2; figures 1-3). However, the inhibition values calculated for seeds indicated that the inhibition was found to be increasing progressively as the concentrations of different aqueous extracts of *Vicia* spp. enhanced (Rahman and Jabeen, 2005b). Among shoot and root extracts, shoot extracts was found to be more inhibitory in both species of *Vicia*.

Evaluation of potential allelopathic activity

i. Seed germination

It was evident that with increase of extract Concentration, the percentage of germinated seed decreased disproportionately in all test plants, as compared to the control variant, the differences being statistically significantly smaller (Table 1, 2; Fig. 1).

The inhibition rate (IR) on the seed germination of the tested Gram seeds for the aqueous extracts from shoot and root of *Vicia hirsuta* varied from 11% to 43% and 16% to 39% respectively (Fig 1). The inhibition rate (IR) on the seed germination of the tested Gram seeds for the aqueous extracts from shoot and root of *Vicia sativa* varied from 11% to 46% and 14% to 37% respectively (Fig. 1). Effect of different aqueous extract concentration of root of *Vicia hirsuta* on Gram seedling germination demonstrated that inhibition was more (39%) as compared to *Vicia sativa* (37%) at the highest concentration level of 20%. Contrary to this it was observed that shoot extracts of *Vicia sativa* was more inhibitory (46%) as compared to *Vicia hirsuta* (43%) at the highest level of concentration (20%) over control. The inhibition of seed germination of gram seedlings in root aqueous extracts of *Vicia hirsuta* at the lowest concentrations (5%) over the control was 16% as compared to 18% of *Vicia sativa*. In shoot aqueous extract the inhibition of Gram seedling germination was recorded to be equal (11%) at lowest concentration level (5%).

ii. Seedling length

In case of *Vicia hirsuta* extracts, at 5% concentration of shoot extract in Gram the length of radicle and plumule was measure to be 3.80 cm and 3.28 cm respectively which reduced at 20% concentration to 3.21 cm and 2.78 cm ,respectively . Observation with root extract of *Vicia hirsuta* demonstrated a gradual decline in the length of plumule as the concentration increased onwards (Table 1; Fig 2). The radicle length varied from 3.8 cm to 3.21 cm at 5% and 20% concentration level respectively and the plumule length varied from 3.31 cm to 2.99 cm at 5% to 20 % concentration respectively.

Experiments with *Vicia sativa* extracts resulted in the same inhibitory effects as seen in *Vicia hirsuta* (Table 2; Fig. 2). At 5% conc. of shoot extracts, the length of radicle and plumule measured 5.84 cm and 4.3 cm respectively whereas at 20% conc. it was 5.01 cm and 4.10 cm respectively. At 5% conc. of root extracts, the length of radicle and plumule measured 5.25 cm and 3.37 cm respectively whereas at 20% conc. it was 4.81 cm and 2.85 cm respectively.

iii. Seedling dry weight

Dry matter weight of radicle declined from 1.31 mg (5% conc.) to 1.71 mg (20% conc.) in case of shoot extracts while the plumule dry matter weight showed variation from 1.20 mg (5% conc.) to 0.80 mg (20% conc.). The effect of root extract showed decline in radicle dry matter weight as 1.32 mg (5% conc.) to 1.25mg (20% conc.) and in plumule dry matter weight as 1.19 mg (5% conc.) to 0.86mg (20% conc.) (Table 1; Fig 3).

Similar inhibitory effects were observed in case of dry matter production with *vicia sativa* shoot and root extracts treated seedlings of Gram (Table 2; Fig 3). Dry matter of radicle declined from 1.54 mg (5% conc.) to 1.32 mg (20% conc.) in case of shoot extracts while the plumule dry matter showed variation from 0.71 mg (5% conc.) to 0.60 mg (20% conc.). The effect of root extract showed decline in radicle dry weight as 1.41 mg (5% conc.) to 1.02mg (20% conc.) and in plumule dry weight as 1.12 mg (5% conc.) to 0.69 mg (20% conc.).

DISCUSSION

From the studies on the effect of aqueous extracts of shoot and root of *Vicia hirsuta* and *Vicia sativa* on seed germination, seedling length and dry matter production of Gram it appeared that inhibition in all the test parameters was recorded in all the concentrations used in the present bioassay.

Both the aqueous extracts appeared to be inhibitory towards Gram seedling germination (Table 1,2; Fig.1) however, the root extract of *Vicia sativa* appeared to be more inhibitory in Gram than *Vicia hirsuta* thereby indicating the presence of toxin in it. At higher concentration of weed extracts the radicle growth was found to be more sensitive than plumule, as also reported by Bhowmick and Doll (1982) and Gogoi et al. (2005). With increase in concentration of extract a greater reduction in seed germination is indicative of the presence of germination inhibitors in aqueous extracts which is in conformity with other researchers who worked with different plants such as: *Cyperus rotundus* (Quayyum et al., 2000), *Cardaria draba* (Kiemnec and McInnis, 2002), *Brassica nigra* (Tawaha and Turk, 2003), *Raphanus raphanistrum* (Norsworthy, 2003).

All concentrations of extract inhibited the seedling length of Gram (1,2; Fig 2). Progressive inhibition was observed in the linear growth of radicle and plumule with proportionate increase in concentration of the extract. Here the inhibitory

effect of shoot extract was more as compared to the root extract. Such observation was suggestive of the presence of inhibitory substance in higher quantity in shoot extract of *Vicia hirsuta* and *Vicia sativa* as compared to the root extract. The observed different phytotoxicity of *Vicia* species might be attributed to the presence of different amount of toxic or allelochemic substances in different parts that leach out under natural conditions. This might be due to differential behavior of the allelochemic present in extract of weeds. Foliar leachates have been regarded to be most phytotoxic in nature probably owing to their proportionately greater biomass and with greater metabolic activity or production of more metabolites (Xuan et al., 2004). The growth of Gram seedling was adversely affected more by *Vicia sativa* than *Vicia hirsuta* ($V.sativa > V.hirsuta$).

The extracts appeared to be inhibitory towards the production of dry weight in radicle and plumule of gram plants (Table 1,2;Fig.3) which were similar to inhibition in seedling length. Reduction in dry weight of seedlings (radicle and plumule) of Gram with the increase in concentration the indicated the presence of allelochemicals in *vicia* spp. that are responsible for reduction in plant growth by reducing the dry matter content. This observation confirms the adverse effects of *vicia* spp. extracts at biosynthetic level.

From the above discussion it can be concluded that differential behavior of extracts from the different organs of the same plant indicates the quantitative variations in the content of the active chemicals and their variable influence on growth of different organs. Thus, *Vicia* spp. behaved as potent phytotoxic agents to inhibit the growth of Gram plants in natural population which can be attributed as one of the major reasons for better growth of these weeds during initial growth period with rabi crops. Table -1 Effects of Aqueous Extract of *Vicia hirsuta* shoot and root on seed germination (%), seedling length (cm) and dry weight (mg) of radicle and plumule of Gram after 7 days (mean \pm S. D.).

Plant extract	Aqueous Extract Concentration (%)	Germination (%)	Seedling Length (cm)		Dry weight(mg)	
			Radicle	Plumule	Radicle	Plumule
Control	0	98	5.55 \pm 1.04	3.61 \pm 0.81	1.39 \pm 0.20	1.23 \pm 0.02
Shoot	5	87	5.01 \pm 1.14	3.28 \pm 0.54	1.31 \pm 0.14	1.20 \pm 0.01
	10	73	4.36 \pm 0.55	3.22 \pm 0.58	1.29 \pm 0.13	1.00 \pm 0.01
	15	64	4.10 \pm 0.81	3.11 \pm 0.76	1.20 \pm 0.12	0.98 \pm 0.02
	20	56	4.01 \pm 0.44	2.78 \pm 0.50	1.17 \pm 0.09	0.80 \pm 0.01

	Mean	70	4.37 ± 0.73	3.09 ± 0.59	1.24 ± 0.12	0.99 ± 0.01
Root	5	82	3.80 ± 1.07	3.31 ± 0.68	1.32 ± 0.19	1.19 ± 0.04
	10	73	3.78 ± 1.01	3.24 ± 1.05	1.29 ± 0.15	0.98 ± 0.03
	15	71	3.74 ± 1.09	3.12 ± 1.02	1.28 ± 0.15	0.92 ± 0.03
	20	60	3.21 ± 0.62	2.99 ± 0.89	1.25 ± 0.10	0.86 ± 0.04
	Mean	71.5	3.63 ± 0.94	3.16 ± 0.91	1.28 ± 0.14	0.98 ± 0.03

All differences significant at 1% level.

Table - 2 . Effects of Aqueous Extract of Vicia sativa shoot and root on seed germination (%), seedling length (cm) and dry weight (mg) of radicle and plumule of Gram after 7 days (mean ± S. D.).

Plant extract	Aqueous Extract Concentration (%)	Germination (%)	Seedling Length (cm)		Dry weight(mg)	
			Radicle	Plumule	Radicle	Plumule
Control	0	95	9.05 ± 1.58	5.36 ± 0.80	1.27 ± 0.61	1.18 ± 0.14
Shoot	5	85	5.84 ± 1.59	4.53 ± 0.65	1.23 ± 0.31	1.12 ± 0.14
	10	71	5.06 ± 0.68	4.25 ± 0.48	1.17 ± 0.64	0.97 ± 0.14
	15	60	5.03 ± 1.50	4.18 ± 0.95	1.16 ± 0.67	0.93 ± 0.16
	20	51	5.01 ± 1.26	4.10 ± 1.04	1.15 ± 0.40	0.70 ± 0.10
	Mean	66.75	5.23 ± 1.25	4.26 ± 0.86	1.18 ± 0.55	0.93 ± 0.13
Root	5	82	5.25 ± 0.71	3.37 ± 0.64	1.23 ± 1.40	1.12 ± 0.33
	10	76	4.99 ± 1.26	3.24 ± 0.86	1.19 ± 0.94	0.94 ± 0.33
	15	71	4.91 ± 0.35	3.09 ± 0.56	1.18 ± 0.10	0.89 ± 0.24
	20	60	4.81 ± 0.98	2.85 ± 0.51	1.14 ± 0.38	0.69 ± 0.14
	Mean	72.25	4.99 ± 0.82	3.13 ± 0.64	1.18 ± 0.71	0.91 ± 0.26

All differences significant at 1% level.

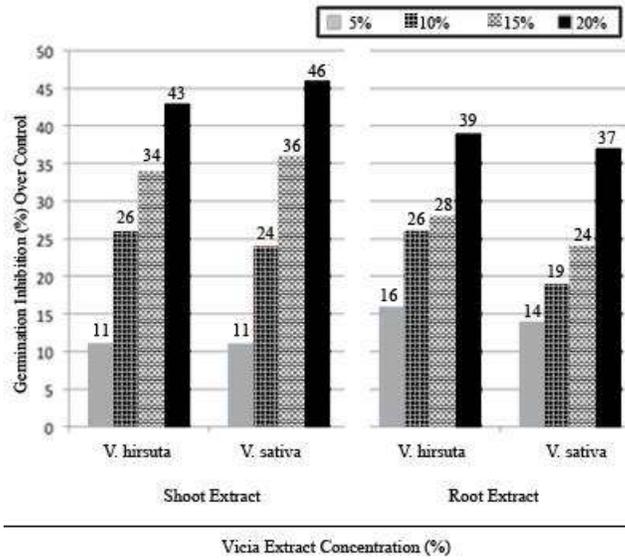


Fig.1 - Effect of Shoot and Root Aqueous Extract of Vicia Spp. on Germination of Gram

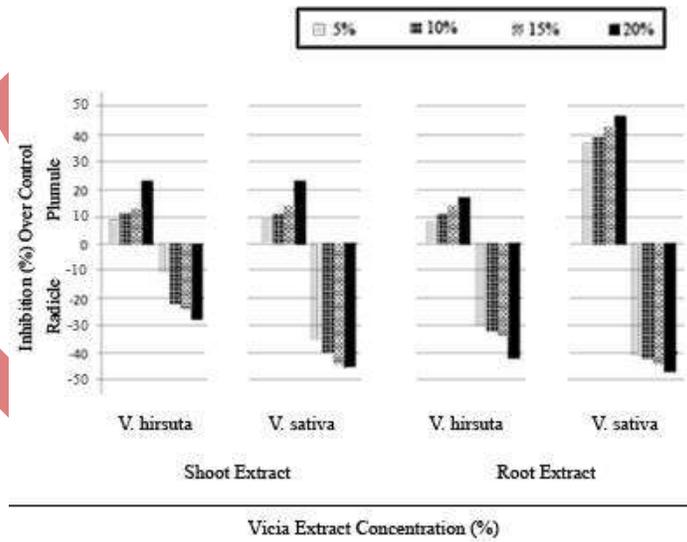


Fig.2 - Inhibitory Effect of Shoot and Root of Vicia Spp. Aqueous Extract on Seedling Length of Gram

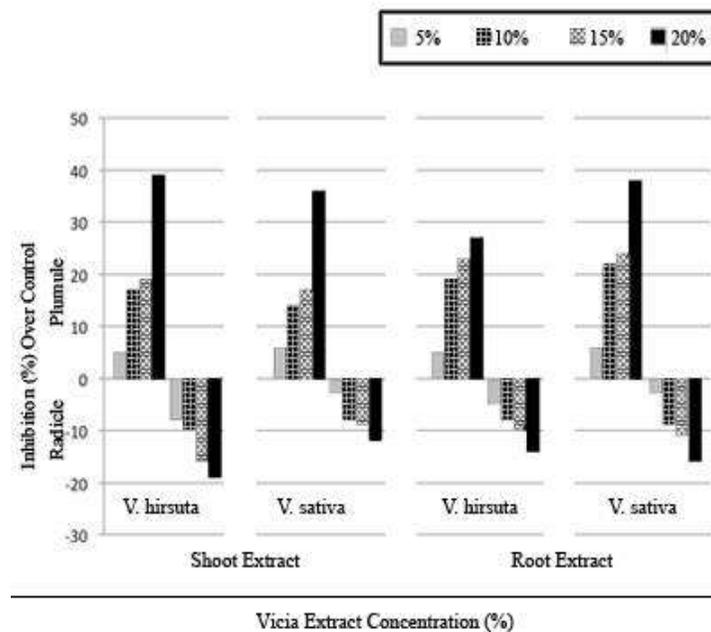


Fig.3 - Inhibitory Effect of Shoot and Root of Vicia Spp. on Dry Weight of Radicle and Plumule of Gram

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