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Allelopathic Potential of *Solanum forskalii* Dunal: A Tropical Ruderal Weed

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Abstract: Effect of *Solanum forskalii* Dunal on seed germination and early seedling growth of three test plants namely mustard, wheat and corn was examined. Aqueous extract of *S. forskalii* at different concentrations (25, 50, 75 and 100% stock solution) inhibited the germination, root and shoot growth of all the three test species. Germination and growth were reduced by the shoot extract in the order: mustard > corn > wheat. Decaying shoot of *S. forskalii* in sandy loam at 5, 10 and 20 g/kg soil substantially inhibited germination and seedling growth of bulrush millet (*Pennisetum americanum*) at all the dosages. Bioassay of the ether extract of *S. forskalii* revealed four zones of inhibition at Rf values 0.1-0.2, 0.2-0.3, 0.8-0.9 and 0.9-1.0. Chromatography for the phenolics revealed the presence of nine phenolic compounds: salicylic acid, vanillic acid, syringic acid, catechol, gentisic acid, 4-methylresorcinol, protocatechuic acid, pyrogallol and an unknown.

Key words: Allelopathy, *Solanum forskalii*, syringic acid, vanillic acid, salicylic acid

Introduction

A number of investigations on the distribution of vegetation in natural, semi-natural and managed floristic communities have shown that allelopathy is an important determinant in these plant associations (Rice, 1984) and plays an eminent role in the intra specific and interspecific competition. The plants may have stimulatory or inhibitory effect on germination and growth. Various workers have reported allelopathic potential of weeds (Einhellig and Rasmussen, 1973; Rasmussen and Einhellig, 1975; Ashraf and Sen, 1978; Shaukat *et al.*, 1985; Ahmed and Wardle, 1994; Burhan and Shaukat, 1999; Rebaz *et al.*, 2001; Shaukat and Siddiqui, 2002).

Chemicals released from the plants are responsible for replacing susceptible species with the invading allelopathic species during succession (Al-Saadawi and Rice, 1982; Rice, 1995). Putnam and Weston (1986) listed 90 weed species while Narwal (1994) listed 129 weed species having allelopathic potential. Several workers have shown that allelopathy may play an important part in weed-weed interaction (Wilson and Rice, 1968; Rasmussen and Rice, 1971; Newman and Rovira, 1975) and weed-crop interaction (Colton and Einhellig, 1980). Extensive studies regarding allelopathic potential of weeds on crops are conducted all over the world by different workers (Casado, 1995; Inderjit *et al.*, 1996; Lydon *et al.*, 1997; Rajbanshi and Inubushi, 1997; Peres *et al.*, 1998; Ito *et al.*, 1998; Al-Humaid and Warrag, 1998).

The secondary plant compounds including alkaloids, terpenoids, flavonoids, steroids, tannins and phenolic compounds have inhibitory effects on crops (Whittaker and Feeny, 1971). Phenolic compounds often constitute the principal allelopathic agents in weeds and other allelopathic plants (Inderjit, 1998; Ferreira *et al.*, 1998; Wang *et al.*, 1998). *Solanum forskalii* a perennial ruderal weed, grows in almost pure stands in waste grounds, vacant lots and abandoned fields (rarely in cultivated fields) in Karachi and its suburbs. It is suspected that allelopathy could be involved in the suppression of the neighboring plant species. So, experiments were designed to test the allelopathic potential of *S. forskalii*. Our objectives were: 1) to evaluate the effect of aqueous extract of *S. forskalii* on three crop species *in vitro*, 2) to study the phytotoxicity of decaying *S. forskalii* in soil, and 3) to identify the phenolic principles of *S. forskalii*.

Materials and Methods

Effect of aqueous extract of *Solanum forskalii* on germination and seedling growth of three test species: *Solanum forskalii* was collected from a waste ground in Malir, Karachi and its shoot extract was studied. The plant material was air-dried under shade and chopped into small pieces. Extract of *S. forskalii* was prepared by soaking 10 g plant material in 100 ml of distilled water for 24 h to obtain stock solution. Using stock solution (100%), three other concentrations were prepared i.e., 25, 50 and 75%. Effect of various concentrations was tested against *Brassica campestris* L. (mustard), *Triticum aestivum* L. (wheat) and *Zea mays* L. (corn). Crop seeds were first surface sterilized by 0.3% calcium hypochlorite for five min. and then placed on 9 cm diam., sterile

petri dishes on Whatman No.1 filter paper. Each plate received 5 ml of the extract. For controls, distilled water was used. Germination counts were made daily and shoot and root length of the seedlings were recorded after 72 h.

Effect of decaying *Solanum forskalii* on germination and seedling growth of *Pennisetum americanum*: Dried powdered material of *S. forskalii* was mixed thoroughly with loamy sand (76.1% sand, 15.3% silt and 8.6% clay) at 5, 10 and 20 g/kg of soil. Pots were watered once and soil was left for biodegradation. After one week, 10 seeds of bulrush millet (*P. americanum*) were sown in each pot. Controls and treatments were replicated thrice and pots were randomized on the greenhouse bench. Daily rate of emergence was recorded while shoot and root lengths were measured after four days.

Bioassay: Ten gram air-dried shoots of *S. forskalii* were blended in 200 ml distilled water. The centrifuged homogenate was adjusted to pH 3 with 0.5 N H₂SO₄ extracted thrice with peroxidase-free ether and evaporated to dryness using argon gas. Two ml of 80% ethanol was added to the dried material and was streaked on Whatman No.1 filter paper. Duplicate 10 cm wide chromatograms were developed by descending chromatography in isopropanol:ammonia:water (10:1:1, v/v/v). When the solvent had moved 30 cm, the chromatograms were dried and 10 equal width strips were cut and assayed for growth regulators using wheat coleoptile straight growth test of Nitsch and Nitsch (1956). Five mm segments of 3 day old dark grown wheat were excised and floated in distilled water for 1 h. Ten coleoptile segments were placed in between two strips of the same Rf value, and kept in 11.5 cm diam., petri dishes over two layers of tissue paper moistened with 4 ml of 0.02 M citrate phosphate buffer (pH 4.8). After 48 h of growth in dark, the length of coleoptile segments was measured.

Chromatography: Ether extract of *S. forskalii* was evaporated to dryness, dissolved in 2 ml of 80% ethanol and used for loading on silica gel F₂₅₄ thin layer chromatographic plate. The chromatogram was developed in acetic acid-chloroform 1:9 v/v by ascending chromatography using reference phenolic compounds. Phenolic compounds were detected using ferric chloride-ferric cyanide reagent and UV light (Harborne, 1973).

Statistical analysis: A factorial analysis of variance (FANOVA) was performed, after arcsine transformation of the percentage germination data (Sokal and Rohlf, 1995). As a follow up of FANOVA, a least significant difference (LSD) test and Duncan's multiple range test were performed at p=0.05. Computer programs for the analyses were developed in FORTRAN-77.

Results

Effect of aqueous extract on germination and seedling growth of the test species: Germination of all the three test species was inhibited by various concentrations of the extract (p < 0.05) over

Table 1: Effect of aqueous shoot extract of *Solanum forskalii* on shoot and root growth (cm) of *Brassica campestris*, *Triticum aestivum* and *Zea mays*

Test species	Concentrations (% stock solution)									
	0		25		50		75		100	
	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot	Root	Shoot
<i>B. campestris</i>	3.6 ± 0.3	5.4 ± 0.5	3.0 ± 0.3	2.3 ± 0.4	1.8 ± 0.3	2.0 ± 0.4	0.3 ± 0.2	0.4 ± 0.1	0 ± 0	0 ± 0
<i>T. aestivum</i>	4.8 ± 0.6	7.1 ± 0.4	3.2 ± 0.3	6.1 ± 0.4	2.4 ± 0.4	3.1 ± 0.2	0.8 ± 0.2	1.2 ± 0.5	0.2 ± 0	0.3 ± 0.1
<i>Zea mays</i>	4.6 ± 0.7	7.2 ± 0.5	3.5 ± 0.4	4.1 ± 0.3	2.8 ± 0.4	2.7 ± 0.3	0.5 ± 0.2	0.8 ± 0.2	0 ± 0	0 ± 0

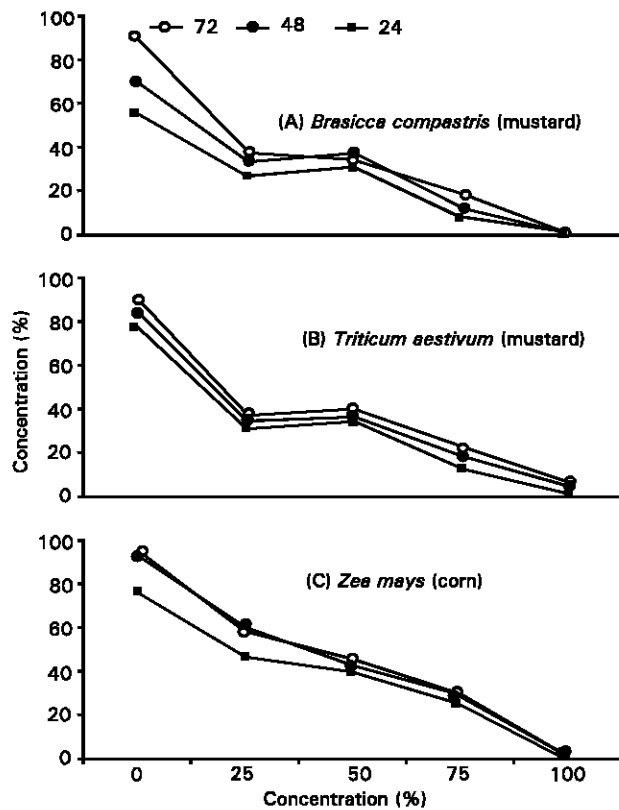


Fig. 1: Effect of various concentration of the aqueous shoot extract of *Solanum forskalii* on percentage germination of the three test species. (LSD_{0.05} Test species = 9; Concentration = 12; Days = 9)

Table 2: Phytotoxic effects of decomposing *Solanum forskalii* on germination and root and shoot growth of bulrush millet (*Pennisetum americanum*)

Concentration g/400 g soil	Germination (%)	Root length (cm)	Shoot length (cm)
0	90	11.2 ± 2.5	13.5 ± 1.3
5	90	8.6 ± 1.9	9.2 ± 2.3
10	75	6.2 ± 1.3	8.8 ± 1.3
15	55	4.4 ± 1.5	4.2 ± 0.8

Table 3: Rf-values (x 100) of phenolic principles in ether fraction of aqueous extract of *Solanum forskalii* and their reaction to a developing reagent and UV light

Compounds	Rf	Ferric chloride-ferric cyanide	UV light
Unknown	96.34	Purple	Blue
Salicylic acid	90.85	Purple	Blue
Vanillic acid	84.14	Purple	Light blue
Syringic acid	79.87	Purple	Blue
Catechol	37.80	Magenta	Dull blue
Gentisic acid	33.50	Purple	Blue
4-methyl resorcinol	28.6	Bluish pink	Blue
Protocatechuic acid	18.2	Brownish purple	Blue
Pyrogallol	7.3	Brownish purple	Bluish-brown

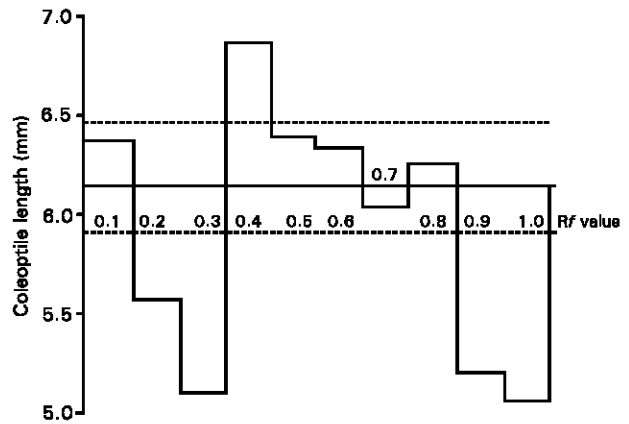


Fig. 2: Results of wheat coleoptile bioassay of the ether extract of *Solanum forskalii* showing inhibitors and promoters at different Rf values. Dotted lines represents 95% confidence interval

the controls (Fig. 1A-C). The inhibitory effect increased with the increase in concentration. Different species were affected to a different extent; the degree of inhibition varied in the order: mustard > corn > wheat. Similarly, seedling growth of the test species was also adversely affected and growth reduction was greater at higher concentrations. Root and shoot growth of mustard was inhibited to a greater degree compared with wheat and corn (Table 1). Corn (*Zea mays*) was least affected. Generally, root growth was reduced to a greater degree than the shoot growth.

Phytotoxicity of decaying *S. forskalii*: Germination of bulrush millet was significantly ($p < 0.001$) reduced at all the dosages of decaying *S. forskalii* (Table 2). Germination percentage declined sharply with the increase in concentration. Likewise, both root and shoot growth were significantly ($p < 0.001$) suppressed at all the concentrations of the decaying shoot material compared to the controls. This effect was generally more pronounced at higher concentrations.

Wheat coleoptile bioassay: Wheat coleoptile bioassay revealed four inhibitors at Rf-values of 0.1-0.2, 0.2-0.3, 0.8-0.9 and 0.9-1.0, while one significant promoter was detected at Rf-values of 0.4-0.5 (Fig. 2).

Chromatographic study: Chromatograms sprayed with ferric chloride-ferricyanide reagent examined under UV light, revealed nine spots which were matched with the standard phenolic compounds and identified as salicylic acid, vanillic acid, syringic acid, catechol, gentisic acid, 4-methyl resorcinol, protocatechuic acid, pyrogallol and an unknown (Table 3).

Discussion

This study provides evidence on the allelopathic potential of *Solanum forskalii*. Seed germination and seedling growth of mustard, corn and wheat were inhibited by aqueous extract of *S. forskalii*. Extracts of a number of weed species have been shown

to be inhibitory to germination of crop seeds (Shaukat *et al.*, 1985; Rafique and Hafeez, 1994; Casado, 1995; Hofmann *et al.*, 1996; Demchuk and Yurchak 1996; Lydon *et al.*, 1997). To the best of our knowledge there are no previous report on the allelopathic potential of *S. forskalii*. However, Sharma and Sen (1971) reported the presence of some phytotoxic substances in the fruit pulp of *S. surattense* that inhibited the seed germination of *Pennisetum americanum* and *Sesamum indicum*. The aqueous extract of *S. forskalii* exhibited differential inhibitory effect on germination and early seedling growth of the three test species presumably because the different phenolic compounds (and other inhibitors) might have differential effect due to morphological and physiological differences among the species.

The germination of bulrush millet was reduced in soil incorporated with decaying *S. forskalii*. Wilson and Rice (1968) have reported both stimulatory and inhibitory effect on various crop species with decaying sunflower leaves. In the present study, maximum reduction was observed in soil incorporated with 20-g shoot material. This may presumably be due to the release of phytotoxins from the decaying *S. forskalii* that remain active and stable for considerable duration in soil. Similarly, Shaukat *et al.*, (1985) reported adverse effects of decaying *Citrullus colocynthis* while Burhan and Shaukat (1999) reported inhibitory effects of decaying *Argemone mexicana* on bulrush millet.

It is possible that phytotoxins released from *S. forskalii* may accumulate in soil in biologically significant amounts and thereby play a key role as a habitat variable, exerting a causative influence on growth and development of other neighboring plants. The wheat coleoptile bioassay of *S. forskalii* revealed four inhibitory zones. These presumably represented phenolic compounds. Paper chromatography revealed the presence of nine phenolic compounds including salicylic acid, syringic acid, catechol, vanillic acid, gentisic acid, 4-methyl resorcinol, protocatechuic acid, pyrogallol and an unknown compound. Only the phenolic inhibitors were investigated. However, phytotoxic chemicals other than phenolic compounds might be present and could constitute important allelopathic agents. The toxic nature of the phenolic compounds have been reported by several workers (Stowe *et al.*, 1987; Blum, 1996; Inderjit, 1998).

It is concluded from our results that *S. forskalii* is considerably phytotoxic and being a dominant species in some of the disturbed communities in vacant lots of cities and in the suburbs accumulates phytotoxins over a period of several years and eventually suppresses other species in the community that are eliminated, thereby greatly reducing species diversity leading to pure or almost pure populations. Thus, this species poses problem in maintaining biodiversity of ruderal habitats.

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