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Allelopathic and Antifungal Potential of *Lantana camara* Root Leachates in Soil

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Abstract: The allelopathic and antifungal potential of the root leachates of *Lantana camara* L., a tropical weed was evaluated under glasshouse conditions. Soil drench with full strength concentration caused marked suppression of two root-infecting fungi including *Fusarium solani* and *Rhizoctonia solani*. Whereas, a high concentration of the root leachate caused marked inhibition of germination and growth of mungbean, a low concentration (1:2 dilution) enhanced plant growth. When soil was added with urea as an extra source of nitrogen (N) to overcome the phytotoxic symptoms, the toxic effect was not alleviated, suggesting that allelopathic effect due to the root leachate of this weed was not associated with N depletion in the soil. When soil was applied with urea, previously amended with root leachate of *L. camara*, the efficacy in the control of two soil-borne root-infecting fungi was increased.

Key words: *Lantana camara*, *Fusarium solani*, *Rhizoctonia solani*, root leachates

Introduction

Although the word 'allelopathy' is derived from the two words 'allelon' (of one another) and 'pathos' (feeling), the term refers to effects that are both detrimental and beneficial among the interacting organisms (Rice, 1984). These effects have been observed in all classes of plants and also extend to microorganisms. The formal definition of allelopathy is "any direct or indirect harmful or beneficial effect by one plant (including microorganisms) on another via production of chemical compounds that escape into environment" (Birkett *et al.*, 2001). Toxic compounds emitted from an allelopathic plant not only interfere with the normal growth of the associated crop but also have detrimental influence on associated microorganisms especially those that are known to cause root diseases (Narwal, 1984; Shaukat *et al.*, 2001).

Certain allelochemicals from plants have been used as pesticides (Grainge and Ahmed, 1988; Shaukat and Siddiqui, 2001a). Some have also served as sources of new chemistry for production of synthetic pesticides. The pyrethroids, a group of insecticidal metabolites from *Chrysanthemum cinerariaefolium* and related species, are used as insecticides in their naturally occurring form and have also served as models for the production of synthetic insecticides such as permethrin, pyridin, and resmethen (Duke *et al.*, 1988). Likewise, phenolic acids including *p*-hydroxybenzoic acid, *p*-coumaric acid and caffeic acid from plants are known to suppress root-infecting fungi and root-knot nematode (Shaukat *et al.*, 2001; Shaukat and Siddiqui, 2001).

Rice (1984) and Inderjit and Dakshini (1994) found that root leachates of several plant species are known to influence growth and establishment of other plant species. *Lantana camara* L., is a perennial weed of semiarid regions of Indo-Pak subcontinent. This weed is often grown for the purpose of hedgerows and sometimes occurs as an escape. In our previous studies, leaf extract and decomposed leaves of *L. camara* not only inhibited germination but also caused marked suppression of several root-infecting fungi (Shaukat *et al.*, 2001) and a plant-parasitic nematode (Ali *et al.*, 2002). The present paper evaluates the potential impact of root leachates of *L. camara* on I) germination and growth of mungbean, and ii) control of two root-infecting fungi including *Fusarium solani* and *Rhizoctonia solani*.

Materials and Methods

Roots of *L. Camara,* collected from the shrubs grown at Karachi University campus during April, 2001 were carefully washed to remove soil and dried for 96 h at room temperature. Root leachate was prepared by soaking 15 g of roots in 220 ml distilled water for 72 h. followed by filtration using Whatman No. 42 filter paper. The filtrate was identified as full strength (FS) root leachate. The FS root leachate was further diluted with appropriate amounts of distilled water to obtain final concentrations of 1:2 and 1:4 (v/v) root leachate to distilled water. The leachate was stored at 6°C in refrigerator prior to use.

The soil used for the experiment was obtained from the experimental field of the Department of Botany, University of Karachi. The soil (sandy-loam, pH-8.1, moisture holding capacity 40%) was passed through 2-mm sieve to discard non-soil particles. The soil was naturally infested with 3000-cfu g^{-1} of soil of mixed population of *Fusarium* spp., (*F. oxysporum* and *F. solani*) as estimated by soil dilution technique (Nash and Snyder, 1962) and 5-7% colonization of *Rhizoctonia solani* on sorghum seeds used as baits (Wilhelm, 1955). Eight mungbean *Vigna radiata* (L.) Wilczek seeds were sown in 8 cm-diam., plastic pots containing 350 g sandy-loam soil. Before planting, the soil was drenched with 50 ml FS, 1:2 dilution, or 1:4 dilution of root leachate. The different root leachate to soil ratios were selected to simulate the field situations. The soil drenched with 50 ml sterile distilled water served as the control. Treatments were replicated four times and arranged in a randomized complete block design. After one week, the percentage of seed germination was recorded.

The experiment was terminated 45 days after seedling emergence and growth-parameters (plant height and fresh weight of shoot) were recorded. To determine the incidence of root-infecting fungi, 5 mm long root pieces from each plant were surface sterilized with 1% $Ca(OCl)_2$ and plated onto PDA plates containing penicillin (100,000 units/l) and streptomycin sulphate (0.2 g/l.). After incubation for one week at 28°C, the incidence of root-infecting fungi was recorded as follows:

$$\text{Infection \%} = \frac{\text{no. of plants infected by the fungus}}{\text{total no. of plants x 100}}$$

One of the concern regarding allelopathy is that the addition of plant debris or leachates results in a temporary depletion of nitrogen (Harper, 1997) and thus any growth response after addition of plant debris or leachates may be due to nitrogen in the soil and not to organic molecules (Inderjit and Foy, 1999). Another experiment was designed to understand the effect of N fertilization on the allelopathic potential of soil amended with different dilutions of *L. camara* root leachates. The experiment was a 2 x 4 factorial. The factors included 2 levels of N (0 and 0.18g urea/kg soil) and 4 levels of root leachates of *L. camara* (0, FS, 1:2 dilution and 1:4 dilution). The soil samples, 350 g each, were amended with 25 ml FS, 1:2 dilution, or 1:4 dilution of root leachates of *L. camara* and 0.18g/kg of urea (prepared in 25 ml sterile distilled water) as N fertilization. Soil samples amended with only 0.18 g/kg of urea were used as controls. All amendments were replicated thrice. Plant growth and disease parameters were recorded as outlined above.

The data were subjected to analysis of variance (ANOVA) or factorial analysis of variance (FANOVA) followed by a least significant difference (LSD) test in accordance with Sokal and Rohlf (1995).

Results

Soil amended with the full strength of root leachate significantly ($P < 0.05$) suppressed the root infection caused by *Fusarium solani* and *Rhizoctonia solani* (Table 1). The suppressive effect was 44% and 51% respectively against *F. solani* and *R. solani* over the untreated controls. The germination percentage of mungbean was significantly ($P < 0.05$) reduced compared with controls by the root leachate, and greater reduction occurred at higher concentrations. Both plant height and fresh weight of shoot were significantly ($p < 0.05$) retarded over the controls in soils receiving full strength of leachate and its 1:2 dilution but were markedly enhanced when treated with 1:4 dilution.

To investigate if allelopathic activity of *L. camara* root leachate is modified after the addition of N fertilization, soils were amended with different dilutions of *L. camara* root leachate (FS, 1:2 or 1:4) plus N fertilization (0.18g/kg of urea). In general, soils treated with urea significantly suppressed *F. solani* and *R. solani* over the controls (Table 2). The inhibitory effect was even more pronounced when soil was treated with both full strength of root leachate and urea (69% and 59% reduction respectively, against *F. solani* and *R. solani*). Similarly, addition of urea significantly increased germination percentage of mungbean seeds over the controls. At high concentration of root leachate without urea markedly inhibited seed germination. Plant height and fresh weight of shoot were significantly decreased when full strength and 1:2 dilution of root leachate was applied in the soil. Addition of urea did not significantly overcome the inhibitory effect of root leachate of *L. camara*. However, when soil was amended with 1:4 dilution of root leachate with N, there was a slight increase in plant height and fresh weight of shoot over the untreated controls or in soil without urea.

Discussion

The results obtained here clearly demonstrate that root leachate of *L. camara* has the potential to control soilborne root-infecting fungi including *F. solani* and *R. solani.* Although, satisfactory control of the root infecting fungi is achieved here, little is known about the mode-of-action involved in suppression of the two fungi. Phenolic compounds or some other chemicals exuded from the roots of *L. camara* were probably responsible for the suppression of the root infecting fungi. In our previous study, it was observed that application of decomposed leaf materials of *L. camara* in the soil resulted in marked changes in the fungal community structure which in turn caused significant suppression of *Meloidogyne javanica*, the root-knot nematode (Shaukat and Siddiqui, 2001b). Both *F. solani* and *R. solani* are the important constraints of most of the agricultural fields of Pakistan causing root-rot and damping-off diseases thereby reducing crop yield. Application of chemical fungicides is a routine practice to ensure good emergence of the seedling and to protect the roots from invading the pathogens at later growth stages. According to the Montreal treaty, up to the year 2005, most of the chemical pesticides will be banned because of their potential toxicity to human beings and other organisms. Agricultural managers are always looking for some safer strategies to control pests and pathogens. Toxicants of plant origin or decomposed debris and root leachates seem to provide

Table 1: Effect of various concentrations of root leachates of *Lantana camara* on *Fusarium solani* and *Rhizoctonia solani* and growth of mungbean

Soil amended with root leachate[a]	Infection %		Germination percentage	Plant height (cm)	Shoot weight (g)
	Fusarium solani	*Rhizoctonia solani*			
Control	100	78	91	17.8	0.64
FS	56	38	53	13.8	0.41
1:2 dilution	75	56	75	17.1	0.56
1:4 dilution	75	63	81	19.6	0.73
$LSD_{0.05}$	29	23	17	1.7	0.19

[a]Roots of *L. camara* (15 g) were soaked in 220 ml of distilled water for 72 h followed by filtration. The filtrate is identified as full–strength root leachate and was further diluted with appropriate amount of distilled water to give final dilutions of 1:2 (v/v) and 1:4 (v/v).

Table 2: Effect of various concentrations of root leachates of *Lantana camara* and urea as N fertilizer on *Fusarium solani* and *Rhizoctonia solani* and growth of mungbean

Soil amended with root leachate	Infection %		Germination percentage	Plant height (cm)	Shoot weight (g)
	Fusarium solani	*Rhizoctonia solani*			
0 g/kg urea					
Control	91	56	91	18.5	0.52
Full strength	50	25	53	13.9	0.39
1:2 dilution	50	38	75	16.8	0.48
1:4 dilution	75	44	91	19.5	0.61
0.18 g/kg urea					
Control	81	44	100	19.3	0.57
Full strength	25	18	75	15.2	0.48
1:2 dilution	56	25	81	16.7	0.59
1:4 dilution	63	38	91	20.3	0.82
$LSD_{0.05}$ Treatments	27	19	13	2.08	0.19
Urea	11	9	7	0.97	0.08

alternative means to synthetic pesticides.

Whereas low concentration of the leachates enhanced the plant growth, high dosages of root leachate inhibited germination and growth of mungbean. From a practical standpoint, if the leachate of this weed is to be exploited in practical agriculture, its appropriate quantity (concentration) should be determined first under laboratory conditions (Duke and Lydon, 1987). The results of the present study to some extent provide the knowledge regarding its concentration under glasshouse conditions. However, the concentrations required under field conditions have yet to be determined. It is generally believed that the phytotoxicity of the allelochemicals is due to nitrogen depletion and could be overcome by the addition of access nitrogen in the soil. However, in the present study the additionally applied nitrogen in the form of urea did not alleviate the phytotoxic action of the root leachate of *L. camara* on mungbean. This clearly suggests that factors other than N depletion were involved in the inhibition of crop growth. In contrast to our study, Inderjit and Dakshini (1999) found that the allelopathic effect by root leachates of *Verbesina encelioides* was alleviated to some extent by the addition of N in the soil.

Since a variety of weeds grow along with the crop, which interfere with crop growth, the phytotoxicity of *L. camara* could be exploited for weed control. Phytotoxic compound(s) from plants and microorganisms represent a wide range of chemistries and mechanisms of action that have potential in the design and development of new herbicides (Lydon and Duke, 1988). Although several natural products of higher plants have been patented as herbicides, none have so far been developed commercially (Heisey, 1990; Duke and Lyden, 1987). Such natural products or derivatives therefore, could have a number of advantages over the current synthetic pesticides including reduced environmental persistence and accumulation, greater target selectivity, and enhanced activity. Natural products are not necessarily safer than synthetic ones (Ames and Gold, 1989) and must be carefully evaluated before adoption. The new tools of molecular biology and biotechnology are making natural products more attractive alternatives in herbicide and fungicide discovery programmes.

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