

**SOIL AMENDMENT WITH *Aerva javanica* (Burm. f.) Juss. ex Schult.
IN THE CONTROL OF ROOT ROT
FUNGI OF COWPEA (*Vigna unguiculata* (L.) Walp.)
AND MUNG BEAN [*Vigna radiata* (L.)]**

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Abstract

Root rot fungi cause severe losses of crop plants, so the present work was carried out to determine the effect of *Aerva javanica* parts powder on root infecting fungi of mung bean (*Vigna radiata* (L.) and cowpea (*Vigna unguiculata* (L.) Walp.). *A. javanica* parts (stem, leaves and flower) were used as soil amendments at 0.1, 1 and 5% to check the effectiveness on growth parameters. All the plant parts showed a significant reduction in root rot fungi like *Fusarium* spp., *Rhizoctonia solani* Kuhn, and *Macrophomina phaseolina* (Tassi) Goid. It was noted that germination percentage, fresh weight, leaf area and number of nodules were significantly higher and the inhibitory effect on root rot fungi increased when the soil was amended with *A. javanica* leaves at 1%. Thus, among all the treatments, *A. javanica* leaves at 1% were found to be the most effective against root rot fungi.

Key words: *Aerva javanica*, root rot fungi, mung bean, cowpea

INTRODUCTION

Biological treatments provide longer protection to the crop compared to fungicidal seed treatment. In addition, they offer benefits not obtainable with fungicidal seed protectants, especially the ability to colonize and protect the seed and germinating seedling (Mukhopadhyay, 1994). Root-rot diseases caused by soil-borne fungi are the most important diseases of many crops. Several fungi have been recorded as causal pathogens of root-rot and wilt diseases like *Rhizoctonia solani* and *Fusarium solani* (Abdallah, 1969; Abou-zeid et al. 1990; Abou-zeid et al. 1997). *Fusarium* wilt disease caused by pathogenic fungi, particularly the fungus *Fusarium oxysporum*, can cause severe losses in a wide variety of crop

plants (Larkin and Fravel, 1998). Schroth and Cook (1963) tested three bean varieties for variation in susceptibility to pre-emergence damping off caused by *Rhizoctonia solani*, *Fusarium solani*, and *Pythium* spp. as well as for the amount of seed exudation and they suggested that exudates influence the incidence of pre-emergence damping-off by providing fungi with nutritive substances necessary for germination and growth in soil. Gliotoxin is an immuno-suppressive cytotoxin produced by pathogenic fungal species (Grovel et al. 2006). *Macrophomina phaseolina* is the causal agent of charcoal root rot, a pathogen affecting agricultural and forest crops (Shaner et al. 1999), with more than 500 susceptible hosts (Wyllie et al. 1984).

Organic amendment is an important method for control of plant diseases. Organic amendments are generally used for the improvement of crop plants and increasing agricultural productivity. Various organic amendments have a suppressive effect on plant parasitic fungi and nematodes (Alam, 1990). The method involves the amendment of different plant parts for the control of fungal and nematode diseases (Mital and Gowsami, 2001). Of the organic substrates, neem cakes have shown promising results in the control of root infecting fungi (Alam, 1990; Abid et al. 1992). Organic amendments are generally used for improving crops, increasing agricultural productivity and suppressing soil-borne diseases (Stone et al. 2003). In a wide variety of organic matters that have been tested as organic amendments for managing plant pathogens, there are oil seed cakes which decreased the population of soilborne pathogens (Sharma et al. 1995). Dawar et al. (2007) reported that sea weeds

(*Melanothanus afaqhusainii*, *Padina tetrastromatica*, *Cytoconium purpureum* and *Hypnea valentiae*) used alone or in combination with some bacteria (*Rhizobium meliloti*, *Pseudomonas aeruginosa*, *Bacillus subtilis*) significantly suppressed root-infecting fungi on mash bean and sunflower.

Aerva javanica grows under a wide range of climatic and edaphic conditions in its natural habitat. It commonly grows in the Karachi University campus. Shariif et al. (2011) isolated six natural products from the whole plant of *Aerva javanica*. Isoquercetrin (1), 5-methylmellein (2), 2-hydroxy-3-O- β -primeveroside naphthalene-1,4-dione (3), Apigenin 7-O-glucuronide (4), Kaempferol-3-O- β -D-glucopyranosyl-(1 \rightarrow 2)- α -L-rhamno-pyranoside-7-O- α -L-rhamnopyranoside (5), 7-(1-hydroxyethyl)-2-(2"-hydroxyethyl)-3,4-dihydrobenzopyran (6) the first time from *Aerva javanica*. Structural evidence was found by the extensive use of chemical and spectral studies. Different crude extracts (*n*-hexane, chloroform, ethyl acetate, methanol and water) and all the known isolated compounds were tested for their antimicrobial activity and they displayed inhibitory activity. Nabeel et al (1990) introduced a new flavonol, isorhamnetin 3-O- β -[4-*p*-coumaroyl- α -rhamnosyl(1 \rightarrow 6)galactoside], from *Aerva javanica* along with its unacylated derivative, its kaempferol analogue and various common kaempferol, quercetin and isorhamnetin glycosides. Shariif et al. (2011) introduced Isoquercetrin, 5-methylmellein, apigenin 7-O-glucuronide from *Aerva javanica* var. *javanica*.

Mansoor et al. (2011) found that some species, including *Abutilon* spp., *Aerva javanica*, *Capparis decidua*, *Cleome brachycarpa*, *Crotalaria burhia*, *Dipterygium glaucum*, *Gisekia pharnacioides* and *Suaeda fruticosa*, could be used against intestinal worms due to their vermifugal and anthelmintic properties or against bacteria and other microorganisms due to their antimicrobial activity.

Chemical control of plant diseases has proved very effective, but the majority of these chemicals are highly expensive and exhibit lethal effects. The present study reports the effect of soil amendment with *Aerva javanica* in the control of root infection caused by *Fusarium* spp., *R. solani* and *M. phaseolina* in mung bean and cowpea crops.

MATERIALS AND METHODS

Aerva javanica plant parts (stem, leaves and flower) were collected from the University of Karachi campus, air dried, ground in an electric blender, and stored in airtight bottles for further studies. The soil used was obtained from an experimental plot of the Department of Botany, University of Karachi. The sandy loam soil (containing sand, silt, clay in the proportions of 70,

11 and 10%) had the following properties: pH ranged from 7.1-9.65, moisture holding capacity (MHC) of 49% (Keen and Raczowski, 1922), total nitrogen 0.077-0.099% (Mackenzie and Wallace, 1954), 3-7 sclerotia/g of *M. phaseolina* g⁻¹ as found by wet sieving technique (Sheikh and Ghaffar, 1975), 5-20% of *R. solani* on sorghum seeds used as baits (Wilhelm, 1955), and *Fusarium* spp. 2000 cfu g⁻¹ as assessed by soil dilution technique (Nash and Synder, 1962). The soil was amended with *Aerva javanica* leaf, stem and flower powder at 0.1, 1 and 5% w/w and surface-sterilized seeds were sown in 8 cm diam. plastic pots, each containing 300 g of soil and watered regularly to maintain sufficient moisture required for the growth of plants. The pots were kept in a screen house in randomized complete block design with three replicates per treatment. Non-amended soil served as the control. The growth parameters such as shoot and root length and weight, leaf area, and number of nodules were recorded after 30 days of seed germination. To determine the incidence of root rot fungi, one cm long root pieces, after washing them in running tap water, were surface sterilized with 1% Ca(OCl)₂ and transferred on PDA plates supplemented with penicillin at 200 mg and streptomycin at 200 mg/liter, with 5 pieces per plate. Petri dishes were incubated at room temperature and after one week infection of roots by root-infecting fungi was recorded.

The data were subjected to analysis of variance (ANOVA) followed by the least significant difference (LSD) test at P = 0.05 and Duncan's multiple range test to compare treatment means (Sokal and Rohlf, 1995).

RESULTS

In the case of cowpea, when the soil was amended with *A. javanica* stem, leaf and flower powder at 0.1, 1 and 5% w/w, there was 100% germination observed when the soil was amended with *A. javanica* stem and leaf powder at 0.1 and 1% w/w (P<0.001), as compared to leaf and flower powder at 5%. The soil amended with *A. javanica* leaves at 1% significantly enhanced shoot length and weight, in contrast to the soil amended with *A. javanica* stem and flower powder at 0.1 and 5%. Root length and root weight significantly (P<0.05) increased when *A. javanica* leaves at 0.1% were applied to the soil, as compared to stem, leaf and flower powder at 1 and 5% w/w. Leaf area was maximum when the soil was amended with *A. javanica* leaf powder at 1%. Number of nodules was high when *A. javanica* leaf powder was applied to the soil at 1%, in contrast to stem and flower powder at 0.1 and 5%. Thus, we observed that *A. javanica* parts at 1% significantly enhanced all the growth parameters (Table 1).

Table 1
Effect of soil amendment with *Aerva* stem, leaf and flower powder on growth parameters.

COWPEA							
Treatment	Germination %	Shoot length (cm)	Shoot weight (gm)	Root length (cm)	Root weight (gm)	Leaf area (cm)	Number of nodules
Control	60.66±1.0	10.00±0.9	2.00±0.2	5.00±0.1	0.68±0.1	9.330±0.5	2±0.0
0.1%stem	100.0±0.0	11.32±0.5	2.49±0.4	6.66±1.7	0.84±0.2	14.91±1.9	5±1.5
1%stem	100.0±0.0	11.10±0.9	2.24±0.2	5.44±0.3	0.82±0.0	14.87±0.3	5±0.6
5%stem	100.0±0.0	12.05±0.7	2.07±0.2	6.00±0.8	0.90±0.4	14.31±2.5	5±0.3
0.1%leaves	86.66±1.1	11.11±1.0	2.18±0.1	6.66±1.4	0.99±0.3	11.68±2.2	6±0.3
1%leaves	100.0±0.0	12.21±1.7	2.99±0.7	5.96±0.6	1.00±0.1	13.85±1.4	6±1.3
5%leaves	6.666±1.0	9.000±5.1	2.08±1.3	5.00±4.6	0.24±0.4	13.00±7.7	3±1.7
0.1%flower	100.0±0.0	10.83±2.6	2.03±0.3	6.13±0.6	0.71±0.3	11.72±0.6	7±0.6
1%flower	100.0±0.0	11.63±1.2	2.29±0.3	5.55±1.7	1.12±0.5	13.57±4.2	5±1.1
5%flower	6.666±1.1	10.00±5.7	1.96±1.1	1.66±2.8	0.27±0.4	9.290±5.3	3±1.7
LSD=0.05	21.99	5.318	1.40	3.00	0.54	4.712	3.00
MUNG BEAN							
Treatments	Germination %	Shoot length (cm)	Shoot weight (gm)	Root length (cm)	Root weight (gm)	Leaf area (cm)	Number of nodules
Control	80.0±1.0	9.000±0.6	0.60±0.1	4.00±1.0	0.19±0.1	4.00±1.4	2±0.1
0.1%stem	100±0.0	13.32±1.5	0.63±0.0	6.33±0.5	0.26±0.0	5.26±0.8	5±0.9
1%stem	100±1.1	12.66±1.8	0.72±0.2	6.66±1.2	0.36±0.0	7.04±2.2	5±0.1
5%stem	13.3±1.5	9.555±8.0	0.61±0.4	4.00±3.2	0.22±0.1	3.91±3.6	4±3.1
0.1%leaves	100±0.0	13.44±1.1	0.85±0.0	5.61±1.3	0.28±0.0	7.06±1.5	6±0.7
1%leaves	100±0.0	13.44±0.8	0.72±0.1	7.44±0.0	0.27±0.0	6.13±1.1	5±0.3
5%leaves	33.3±2.3	10.77±1.9	0.52±0.4	3.44±0.5	0.27±0.1	5.00±2.7	4±0.5
0.1%flower	100±2.0	12.60±2.7	0.74±0.2	5.00±3.4	0.33±0.0	6.08±1.3	7±2.8
1%flower	100±2.0	11.16±1.0	0.77±0.0	7.36±1.5	0.32±0.0	6.66±1.0	5±1.2
5%flower	33.3±2.3	11.16±0.5	0.84±0.2	5.66±0.7	0.40±0.0	5.32±1.3	3±1.3
LSD=0.05	26.16	5.65	0.40	3.06	0.19	3.18	2.68

There was a significant reduction in root-infecting fungi such as *R. solani*, *M. phaseolina* and *Fusarium* spp. *Fusarium* spp. was significantly suppressed when the soil was amended with *A. javanica* stem powder at 5% ($P<0.001$). *R. solani* infection was significantly reduced by 6.66% from 100% when the soil was amended with *A. javanica* leaf powder at 1% ($P<0.001$). Infection of *M. phaseolina* was highly reduced when *A. javanica* leaf powder was applied to the soil at 1% ($P<0.001$) (Table 2).

Germination percentage of mung bean seeds significantly increased when soil was amended with *A. javanica* stem, leaf and flower powder at 1 % w/w ($P<0.001$). Fresh weight and length of plants were maximum when *A. javanica* leaf powder mix was added to the soil at 1% w/w. Root length and weight were high when *A. javanica* leaf powder mix was applied to the soil at at 1% w/w. Leaf area of plants was greater

when the soil was amended with *A. javanica* leaves at 0.1% w/w. Number of nodules per plant was maximum when flower powder was applied to the soil at 0.1% w/w. Mung bean plants had the highest height and weight when *A. javanica* leaf powder was added to the soil at 0.1 and 1 % w/w, in contrast to *A. javanica* stem and leaf powder at 5% w/w (Table 1).

Root-infecting fungi, such as *R. solani*, *M. phaseolina* and *Fusarium* spp., were significantly reduced when *A. javanica* powder was applied to the soil at 0.1, 1 and 5% w/w. *Fusarium* spp. infection was significantly reduced when the soil was amended with *A. javanica* leaf powder at 1% w/w ($P<0.001$). Infection of *R. solani* decreased when *A. javanica* leaf powder was applied to the soil soil at 1% w/w ($P<0.001$). There was significant suppression of *M. phaseolina* when *A. javanica* leaf powder was added to the soil at 1% w/w ($P<0.001$) (Table 2).

Table 2
Effect of soil amendment with *Aerva*
stem, leaf and flower in the control of root rot fungi of mung bean and cowpea.

COWPEA			
Treatment	<i>Fusarium</i> spp.	<i>R. solani</i>	<i>M. phaseolina</i>
Control	100.0±1.0	100.0±0.0	77.66±4.0
0.1%stem	28.66±1.0	42.00±3.4	26.33±6.5
1%stem	19.88±1.3	52.66±1.1	24.44±7.6
5%stem	11.11±1.0	15.55±7.7	11.10±7.7
0.1%leaves	35.32±1.3	15.22±9.8	15.22±9.8
1%leaves	16.00±1.9	6.666±1.1	6.666±1.1
5%leaves	22.22±3.8	13.10±1.1	11.11±3.8
0.1%flower	30.66±1.3	8.777±3.6	17.55±9.8
1%flower	31.11±0.0	10.99±3.7	15.55±3.8
5%flower	44.44±7.6	40.90±7.6	22.20±3.8
LSD=0.05	20.19	16.96	13.61
MUNG BEAN			
Treatments	<i>Fusarium</i> spp.	<i>R. solani</i>	<i>M. phaseolina</i>
Control	100.0±1.0	100.0±1.1	100.0±0.9
0.1%stem	35.11±1.1	55.00±2.5	37.55±7.3
1%stem	30.88±4.2	46.00±1.1	30.88±1.6
5%stem	19.77±6.3	24.00±1.0	15.33±9.8
0.1%leaves	53.11±1.7	44.00±3.4	15.55±3.8
1%leaves	11.11±1.5	13.00±7.0	13.33±6.6
5%leaves	24.22±1.8	33.11±2.4	13.10±1.6
0.1%flower	17.77±3.8	31.11±1.0	37.33±2.5
1%flower	19.77±6.3	33.22±1.9	19.99±1.7
5%flower	37.33±1.2	33.11±1.3	35.44±2.0
LSD=0.05	19.216	23.065	22.066

DISCUSSION

The present observations showed that soil amendment with *A. javanica* enhanced the plant growth and reduced the infection by pathogenic fungi like *Fusarium* spp., *R. solani* and *M. phaseolina*. Infection by *M. phaseolina* was significantly reduced on mung bean and cowpea when the plant powder was used at 0.1, 1 and 5% w/w. Similarly, *Eucalyptus* sp. leaf, stem, bark and fruit powder has the potential to reduce infection caused by root-infecting fungi, viz. *Fusarium* sp., *R. solani* and *M. phaseolina*. (Dawar et al. 2007). Neem cake has shown promising results in the control of root-infecting fungi (Alam, 1990; Abid et al. 1992). Tariq et al. (2006) used different parts of *Avicennia marina*, viz. leaves, stems and pneumatophores, for the control of root-infecting fungi. *Eucalyptus* essential oil is considered to have marked antiseptic activity against infectious bacteria, viruses and fungi (Inouye et al. 2001). Zainab et al. (2009) reported that seed powder of *Azadirachta in-*

dica, *Adenanthera pavonina*, *Leucaena leucocephala*, and *Eucalyptus* spp. control root rot diseases and the application of *A. pavonina*, *L. leucocephala* and *Eucalyptus* spp. at 0.1 and 1% w/w showed significant control of root rot fungi.

M. phaseolina, which produces charcoal rot disease in plants, is reported to be suppressed by *Avicennia marina* plant parts like leaves, stem and pneumatophore when soil is amended at 5% w/w in the case of okra and mash bean plants (Tariq et al. 2006, 2008). Suratuzza man (1995) observed an excellent inhibitory effect of *Allium sativum* and *Zingiber officinale* on soybean seeds to control seed-borne fungi, namely *Colletotrichum dematium* var. *truncatum*, *Macrophomina phaseolina* and *Colletotrichum kikuchii*. Soil amendments with biochar provide high surface area and the porosity of biochar enables it to adsorb or retain nutrients and water, and they also provide a habitat for beneficial microorganisms to flourish (Glaser et al. 2002; Lehmann and Rondon, 2006; Warnock et al. 2007).

The valuable medicinal properties of different plants are due to the presence of several constituents i.e. saponins, tannins, alkaloids, alkenyl phenols, glycoalkaloids, flavonoids, sesquiterpenes lactones, terpenoids and phorbol esters (Tiwari and Singh, 2004). Natural chemicals and their use for plant protection is one of the focuses of the research all over the world (Kiran et al. 2006).

CONCLUSION

Soil amendment with *A. javanica* needs to be introduced into fields on a larger scale. It can therefore be suggested that such soil amendments could represent an environmentally friendly strategy for controlling soilborne pathogens as a substitute for chemical fungicides.

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Wzbogacanie gleby poprzez zastosowanie *Aerva javanica* (Burm. f.) Juss. ex Schult. w zwalczaniu grzybów powodujących zgniliznę korzeni wspanięgi chińskiej (*Vigna unguiculata* (L.) Walp.) oraz fasoli mung (*Vigna radiata* (L.)

Streszczenie

Grzyby powodujące zgniliznę korzeni przynoszą poważne straty roślin uprawnych, dlatego przeprowadzono badania w celu określenia wpływu sproszkowanych z części roślin *Aerva javanica* na grzyby porażające korzenie fasoli mung (*Vigna radiata* (L.) Walp.) oraz wspanięgi chińskiej (*Vigna unguiculata* (L.) Walp.). Części roślin *A. javanica* (łodyga, liście oraz kwiaty) zastosowano jako dodatki wzbogacające glebę w proporcjach 0,1, 1 oraz 5% w celu sprawdzenia ich efektywności w odniesieniu do parametrów wzrostu roślin. Wszystkie części roślin wykazywały znacznie zmniejszenie obecności grzybów wywołujących zgniliznę korzeni, takich jak *Fusarium* spp., *Rhizoctonia solani* Kuhn i *Macrophomina phaseolina* (Tassi) Goid. Zanotowano, że procent kiełkowania, świeża masa, powierzchnia liści oraz liczba brodawek były istotnie wyższe, jak również wzrosło działanie hamujące na grzyby powodujące zgniliznę, kiedy gleba została wzbogacona proszkiem z liści *A. javanica* w proporcji 1%. Stwierdzono, że spośród wszystkich kombinacji doświadczalnych liście *A. javanica* zastosowane na poziomie 1% były najbardziej skuteczne w zwalczaniu grzybów wywołujących zgniliznę korzeni.