

MANAGEMENT OF ROOT ROT FUNGI OF CROP PLANTS BY *MORINGA OLEIFERA* LAM.

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Abstract

The present study was conducted to evaluate the fungicidal effectiveness of *Moringa oleifera* Lam. plant parts (leaves, stem, fruit and seed) against root infecting fungi as well as improving the growth of cow pea and mash bean plants. *In vitro*, leaves extract used @ 75, 50 and 25% v/v concentrations showed significant inhibition in the growth of *Macrophomina phaseolina* (Tassi) Goid, *Rhizoctonia solani* (Kühn) and *Fusarium oxysporum* (Schlecht) in comparison with other parts extracts by using agar well and paper disc diffusion methods. Leaves extract and powder used as seed treatment, soil drenching and soil amendment with different concentrations showed a profound effect in controlling the colonization of root infecting fungi, and enhanced the weight and height of leguminous plants, whereas stem extract and powder improved plant growth and showed maximum inhibition of root rot fungi on tested crops under greenhouse condition.

Key words: *Moringa oleifera*, Concentrations, Root infecting fungi, Extract and powder.

Introduction

Plant pathogens recognized as the major threats to crop production (Fravel, 2005) especially root infecting fungi which include; *Macrophomina phaseolina*, *Rhizoctonia solani*, *Aphanomyces euteiches*, *Sclerotium rolfsii* and *Fusarium* spp. are reported to infect an extensive ubiquitous threat to all agricultural crops (Ghaffar, 1988; Abdel-Kader *et al.*, 2002; Infantin *et al.*, 2006). *F. solani* and *R. solani* causing root rot and damping off diseases on large variety of crop plants (Abu-Taleb *et al.*, 2011) occurs both in the field and greenhouse condition (Hartman & Fletcher, 1991). *M. phaseolina* found throughout the world (Hoes, 1985) causing damping off, charcoal rot, leaf and stem blight, wilt and dry rot diseases (Cowan, 1999). The principle of controlling plant disease is to get better growth of crop plants (Stephan *et al.*, 1988) which can be controlled by using fungicidal applications (Perez *et al.*, 2004) but fungicides are known to cause environmental hazards and human health risks (Mancini *et al.*, 2008). Recently, environment friendly methods are being made to control root rot fungi either by using plant drugs (Hanif & Dawar, 2015) or plant extracts (Rafi *et al.*, 2015) reported as antifungal activity and also improved crop growth and yield providing as a natural fertilizer (Raghavendra *et al.*, 2002; Sayeeda & Ahmad, 2005) due to the presence of several constituents which includes; tannins, alkenyl phenols, sesquiterpenes lactones, terpenoids, phorbol esters, saponins, flavonoids and glycoalkaloids (Tiwari & Singh, 2004). The plant extract significantly inhibited fungal pathogens and successfully devised as fungicides (Babu *et al.*, 2008).

Moringa oleifera (moringaceae) is best known for miracle/wonder tree (Fuglie, 1999) and widely distributed all over the world (Lockett *et al.*, 2000) is a fast-growing, drought resistant and are regarded as a nutritional powerhouse (Elevitch, 2011). This plant has been studied extensively due to its pharmacological activities (Dashputra *et al.*, 1977). It is rich in glucosinolates, isothiocyanates, carotenoids, kaempferol, zeatin, quercetin and many other phytochemicals and also an outstanding source of vitamin (A, B and C) calcium, iron, potassium, protein and the fruit

contain oil rich in β -carotene, sterols and lecithin (Anwar *et al.*, 2007). The oil comprises unusual fatty acids among naturally occurring plant growth stimulants having cytokinin compounds due to the presence of 4- α -L-rhamnopyranosyloxy benzyl isothiocyanate in the seeds of *M. oleifera*. After oil extraction, the remaining seed cake is used as a fertilizer, purify well water and to remove salt from sea water (Shaheen *et al.*, 1998). *M. oleifera* seeds are considered as a cheap bio-absorbent eliminating heavy metals (Sharma *et al.*, 2006). Leaves of this plant are reported containing anti-pyretic, anti-inflammatory, anti-ulcer (Pal *et al.*, 1995), anti-tumour (Makonnen *et al.*, 1997), anti-spasmodic (Caceres *et al.*, 1992), diuretic (Morton, 1991), cholesterol lowering (Mehta *et al.*, 2003), hepatoprotective, anti-oxidant, anti-diabetic (Ruckmani *et al.*, 1998), anti-hypertensive (Dahot, 1988), anti-bacterial and anti-fungal activities (Nickon *et al.*, 2003) due to the presence of 4-(α -L-rhamnopyranosyloxy) benzyl isothiocyanate, 4-(4'-O-acetyl- α -L-rhamnopyranosyloxy) benzyl isothiocyanate, 4-(α -L-rhamnopyranosyloxy) benzyl glucosinolate, niazimicin, pterygospersin and benzyl isothiocyanate (Nepolean *et al.*, 2009). *M. oleifera* is being employed for the treatment of various ailments in the indigenous system of medicine, generally in South Asia (Farooq *et al.*, 2007).

Present study was carried out to explore the fungicidal effectiveness of *M. oleifera* on growth and inhibition of root infecting fungi on crop plants.

Materials and Methods

Collection of material: Unblemished leaves, stem, fruit and seeds of *Moringa oleifera* Lam. were collected from the campus of University of Karachi. All plant parts were dried and powdered by using an electric grinder and stored each plant powder in glass jar respectively.

Extract preparation: 10g of *M. oleifera* plant parts (stem, leaves, fruit and seeds) were soaked in 90 mL distilled water, separately and left it for overnight. The concentration of the extract thus prepared was used as stock solution (100% v/w). The suspension was filtered through Whatman's filter paper No.1 into 250 mL Pyrex

flask. The filtrate was further diluted with distilled water to prepare 75, 50 and 25% v/w concentrations.

In vitro: Paper disc and agar well diffusion methods were used to study the inhibition of *M. phaseolina*, *R. solani* and *F. oxysporum* with aqueous extracts of different concentrations of plant parts. In the paper disc method, sterilized filter paper disc (6mm in diameter) were soaked in 25, 50 and 75% v/w concentrations of stem, leaves, fruits and seed extracts respectively. Three different concentrations of discs were placed on three sides of Petri plate. The fourth disc containing distilled water served as control (Nair *et al.*, 2005). The disc of each root rot fungus (5mm in diameter) was placed in the center, respectively. Similarly, in agar well diffusion method, four wells (5mm in diameter and 2.5mm deep) were made on the surface of the agar medium using a sterilize borer. Three wells were filled with 50 μ L of different concentrations (25, 50, 75 % v/w) of aqueous extracts of *M. oleifera* stem, leaves, fruits and seeds respectively, whereas forth one well with distilled water serve as control. A disc of each test fungus was placed in center respectively. Each root infecting fungus replicated thrice and plates were incubated for 5-6 days at room temperature (27-33°C) and then measured the zone of inhibition in millimeters (Ghazala *et al.*, 2003). The percent growth inhibition over control was determined (Lokeshia & Benagi, 2007).

Soil properties: Soil was obtained from an experimental plot of Department of Botany (University of Karachi), sieved to remove pebbles and was filled in the pots (8 cm diameter) containing 300g. Soil was sandy loam containing (sand 72%, silt 16% and clay 12%), total nitrogen 0.083% (Mackenzie & Wallace, 1954), pH 7.2, moisture holding capacity 47% (Keen & Rakzowski, 1922), 4-8 sclerotia/g of *M. phaseolina* (Sheikh & Ghaffar, 1975), 12-15% of *R. solani* (Wilhelm, 1955) and *Fusarium* spp., 2700 cfu/ g (Nash & Synder, 1962).

In vivo: Cowpea (*Vigna unguiculata* (L.) Walp cv. Lobia-2000) and mash bean (*Vigna mungo* (L.) Hepper cv. NM-97) seeds were sterilized in 1% Ca(OCl)₂ for 2-3 minutes, washed twice in sterilized distilled water to remove entire dust particles. Consequently washed seeds were treated with 25, 50 and 75% v/w extracts of plant parts (stem, leaves, fruits and seeds), respectively, for 5-10 minutes and then air dried for \geq 3 hours in the laminar flow chamber, whereas untreated seeds served as control. In soil drenching, plant extracts of stem, leaves, fruits and seeds with the concentrations of 25, 50 and 75 % v/w drenched (20 mL) separately in each pot, soil without extracts served as control. Similarly, plant parts (stem, leaves, fruit and seeds) powdered at 0.1, 1.0 and 2.5% w/w were amended in the soil and watered 2-3 days for the decomposition of organic substrate. Soil without amendment regarded as control. Each treatment was replicated thrice and four seeds were sown in each pot. The pots were randomly placed in the screen house of the Department of Botany (KU) and data was recorded after one month.

Estimation of growth parameters and incidence of root rot fungi: After 30 days of germination, plants were carefully uprooted and growth parameters recorded in terms of shoot length and weight, root length and weight, number of nodules. Roots were washed thrice in sterilized water and surface sterilized by 1% Ca(OCl)₂ for 2-3 minutes and each root was cut into five pieces. Transferred each root pieces on poured potato dextrose agar (PDA) supplemented with antibiotics, penicillin @ 100,000/L and streptomycin @ 200 mg/L to inhibit bacterial growth. Incubate the Petri plate for one week at room temperature (32-36°C). Colonization percentage of root rot fungi was recorded (Short *et al.*, 1980).

Statistical analysis: The data were analyzed for three way analysis (ANOVA) as per experimental design. Means were separated using Duncan's multiple range test (DMRT) at $p < 0.05$ (Sokal & Rohlf, 1995) using "Statistica" software. Significant differences within the means of treatments and controls were calculated using the LSD statistical test.

Result

In vitro, the experiment was performed to observe antifungal activity of *M. oleifera* parts by using the paper disc and well methods. Most effective result was achieved by leaves extract using all concentrations showed excellent inhibition of root rot fungi by both methods. Maximum inhibition in the control of *M. phaseolina*, *R. solani* and *F. oxysporum* was recorded by stem extract when used at 75% v/w followed by 50 and 25% v/w concentrations. However, seed extract showed least inhibition of *F. oxysporum* at 75 and 50% v/w concentrations, whereas it was failed to inhibit *M. phaseolina* and *R. solani* mycelium as compared to control. No zone of inhibition of root rot fungi was observed by fruit extract used in all concentrations. Both paper disc and well methods showed effective fungicidal results in the screening of *M. oleifera* parts using different concentrations (Table 1 and Fig. 1).

In vivo complete germination of cowpea and mash bean plants showed by *M. oleifera* leaves when used in seed treatment and soil drenching methods in 25 and 50% w/w and amended in soil at 0.1 and 1.0% v/w. Seeds of cowpea and mash bean when treated with 75% v/w leaves extract of *M. oleifera* showed better growth parameters followed by stem extract at 25% v/w, whereas the highest number of nodules was recorded by both stem and leaves extracts used at 25% v/w concentrations. Complete suppression of root rot fungi was shown by leaves extract used in 25 and 50% v/w concentrations. Maximum inhibition of *R. solani*, *M. phaseolina* and *Fusarium* spp. colonization was noticed by the stem extract in all concentrations on both crop plants. Plant growth of cow pea and mash bean were enhanced when leaves powder amended in soil at 0.1% w/w but stem powder showed better plant growth when amended in soil at 1.0% w/w. Colonization of *M. phaseolina*, *Fusarium* spp. and *R. solani* was completely suppressed by *M. oleifera* leaves powder amended in soil at 1.0 and 2.5% w/w on mash bean plant, but in

cowpea it showed at 2.5% w/w. Stem powder amended in soil at 0.1, 1.0 and 2.5% w/w showed significant inhibition of root rot fungi. In case of soil drenching method, shoot length and weight, root length and weight, number of nodules of cowpea and mash bean plants increases due to the *M. oleifera* leaves and stem extracts drenched at 25% v/w in the soil followed by 50 and 75% v/w concentrations recorded in both crop plants. Excellent control of root rot fungi on both crop plants were observed by all concentrations of leaves extract drenched in soil followed by stem extract (Fig.

2). It was noticed that when the seed and fruit extracts and powders of *M. oleifera* used as soil amendment, seed treatment and soil drenching methods showed poor plant growth and unable to inhibit the colonization of *Fusarium* spp., *R. solani* and *M. phaseolina* on cowpea and mung bean plants (Tables 2 and 3).

Among all *M. oleifera* plant parts, leaves powder and extract had shown fungicidal effects in controlling remarkably root rot fungi, and progressively improved plant growth when used in seed treatment, soil drenching and soil amendment methods.

Table 1. *In vitro*, inhibition of root rot fungi by aqueous extracts *Moringa oleifera* parts of with different concentrations by using paper disc and well diffusion methods.

Treatments	Concentrations/ Growth inhibition (MIC)												
	<i>Macrophomina phaseolina</i> (%) (mm)				<i>Fusarium oxysporum</i> (%) (mm)				<i>Rhizoctonia solani</i> (%) (mm)				
	Control	A	B	C	Control	A	B	C	Control	A	B	C	
Paper disc method													
Seed extract	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	7.30 ± 1.53	2.22 ± 1.73	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	
Fruit extract	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	
Leaves extract	0.0 ± 0.0	80.00 ± 2.65	68.90 ± 1.00	62.22 ± 1.00	0.0 ± 0.0	93.33 ± 1.00	84.44 ± 1.00	79.30 ± 1.53	0.0 ± 0.0	97.80 ± 1.00	97.11 ± 0.58	95.60 ± 1.00	
Stem extract	0.0 ± 0.0	53.30 ± 4.58	40.00 ± 1.00	32.70 ± 2.08	0.0 ± 0.0	26.67 ± 1.00	22.22 ± 1.00	17.04 ± 2.08	0.0 ± 0.0	42.90 ± 2.09	23.71 ± 1.53	18.44 ± 1.53	
Well method													
Seed extract	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	11.80 ± 1.53	8.90 ± 1.00	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	
Fruit extract	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	0.0 ± 0.0	
Leaves extract	0.0 ± 0.0	72.60 ± 0.58	62.90 ± 1.53	54.00 ± 1.53	0.0 ± 0.0	97.11 ± 0.58	94.00 ± 1.53	92.70 ± 2.31	0.0 ± 0.0	95.6 ± 0.57	93.30 ± 1.00	86.70 ± 2.65	
Stem extract	0.0 ± 0.0	63.70 ± 4.93	57.04 ± 1.53	44.44 ± 1.00	0.0 ± 0.0	32.70 ± 1.53	24.44 ± 1.00	13.33 ± 1.00	0.0 ± 0.0	51.80 ± 2.52	38.44 ± 3.79	31.80 ± 2.08	
LSD_{0.05}:	Concentration =	0.911				0.823				0.715			
	Plant parts =	0.911				0.823				0.715			
	Methods =	0.842				0.596				0.523			

Where; MIC=Minimum inhibitory concentration, ± Standard deviation and Concentrations of plant extracts: A=75% v/v, B= 50% v/v, C= 25% v/v

Discussion

M. oleifera parts, especially leaves powder and extract revealed pronounced effect and possess antifungal activity against *M. phaseolina*, *R. solani* and *F. oxysporum* both *in vitro* and *in vivo* experiments and were found to be best in all concentrations. *M. oleifera* leaves contain a number of phytochemicals such as; saponins, flavonoids, phenolic and tannins compounds exhibit antimicrobial activities (Sato *et al.*, 2004; Cushine & Lamb, 2005; Mboti *et al.*, 2009). Due to this, it would suggest that it exert anti-fungal activities observed in this research could be attributed to such compounds. Leaves extract of *M. oleifera* was found to be effective in controlling growth of fungi such as *Basidiobolus haptosporus* and *Basidiobolus ranarums* (Nwosu & Okafor, 1995). Leaves extract in chloroform showed positive suppression against *Escherichia coli* (MTCC 443), *Pseudomonas aeruginosa*, *Staphylococcus aureus* (MTCC 3160), *Streptococcus pyogenes* (MTCC 442), *Aspergillus niger* (MTCC 1781) and *Candida albicans* (MTCC 181) as compared to petroleum ether which was failed to inhibit bacterial and fungal strains (Devendra *et al.*, 2011). *M. oleifera* leaves in aqueous and ethanol extracts possess antibacterial activity by producing zone of inhibition using

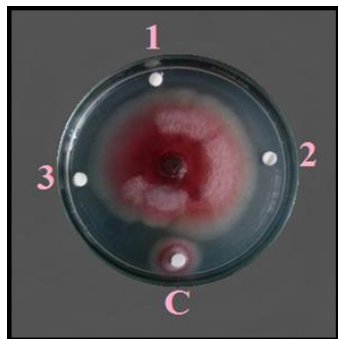
disc diffusion method against *Escherichia coli*, *Proteus vulgaris* and *Salmonella typhi* respectively, whereas acetone and chloroform extracts did not show any antibacterial activity (Gomashe *et al.*, 2014). Leaves of *M. oleifera* showed excellent fungicidal activity against root rot fungi when used at 0.1, 1.0, 2.5% w/w amended in soil and treated with seeds or drenched in soil at 75, 50 and 25% v/w concentrations but also improve the growth parameters of cow pea and mash bean plants. Growth of crop plants increased when *M. oleifera* leaves were applied because it exhibit plant growth regulators (Foidle *et al.*, 2001) containing rich source of ascorbate, calcium, potassium, zeatin and phenolic compounds (Basra *et al.*, 2011) considered as natural crop growth enhancer. Ikram & Dawar (2013) reported that using *Prosopis juliflora* leaves and stem powder @ 1.0% w/w were effective in the inhibition of root rot fungi and in the improvement of all growth parameters of cowpea and mung bean plants. Organic amendments are usually used for the improvement of crop plants and increasing agricultural productivity by suppressing plant parasitic nematode and fungi (Alam, 1990; Stone *et al.*, 2003). Application of organic amendments to soil is considered as a key point to improve the quality and health of soil by supplying important micronutrients (Timsina &

Conner, 2001). Physically organic amendments addition brought changes in water retention, permeability, drainage, water infiltration, structure and aeration of soil (Davis & Wilson, 2005). Seed treatment is an effective method as it protects seed from seed-borne and soil-borne pathogens and enables the seed to germinate and become established as a healthy seedling (Chang & Kommedahl, 1968). Using seed treatment and soil drenching methods, all parts of *M. oleifera* extracts have positive potential in improving plant growth and germination as well as reduced root infecting fungi, but seed extract resulted in minimum suppression of root infecting fungi at 75% v/w concentration when drenched in soil. Seed extracts of *M. oleifera* showed antimicrobial activity against *Aspergillus niger* and *Candida albicans* (Abdulmoneim & Abu-Zaid, 2011) but in our research it failed to suppress root rot fungi particularly *R. solani*, *Fusarium* spp. and *M. phaseolina*. Soil drenching with garlic extract increases summer squash flower number (Helmy, 1992). Similarly, Mohanta *et al.* (2007) reported that using aqueous and organic solvent extracts of the *Semecarpus anacardium* Linn. possess inhibitory effect against *Staphylococcus aureus*. Mangang & Chhetry (2012) investigated that cold water extracts of *Artemisia vulgaris*, *coix lacryma jobi*, *Lantana camera*, *Michelia champaka*, *Passiflora foetida*, *punica granatum* and *Strobilanthes flaccidifolius* showed 50% or more mycelial inhibition of *R. solani*. Amendment of *Avicennia marina* stem and leaves powder pellets extensively controlled root rot diseases caused by pathogenic fungi in cowpea and brinjal plants (Tariq & Dawar, 2011). Similarly, Akhter *et al.* (2006) observed that the extracts of *Adhatoda vasica* and *Zingiber*

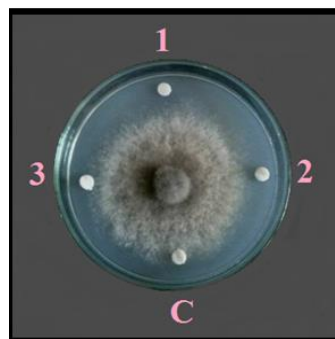
officinale, *Piper betle*, *Azadirachta indica* and *Vinca rosea* in combination with cow dung and *Calotropis procera* (leaf) extract in combination with cow urine possess high ability to inhibit conidial germination of *Bipolaris sorokiniana*. Suleiman & Emua (2009) reported that among *Zingiber officinale*, *Aloe vera*, *Garcinia kola* and *Azadirachta indica* extracts, *Aloe vera* showed 60% inhibition of mycelial growth of *Pythium aphanidermatum*. Extracts of *Polyalthia longifolia* showed antifungal activity against *Macrophomina phaseolina* (Datar, 1999). Therefore, different methods of formulation using plant extract possess antifungal activity to control soil borne pathogens (Hadar *et al.*, 1992; Chung & Miller, 1995; Muchovej & Pa-covsky, 1997; Ahmed & Nimer, 2002).

Chemical control of plant pathogens has proved effective, but the majority of these agrochemicals is extremely costly and exhibit lethal effects. Present research showed antifungal effect of *M. oleifera* leaves remarkably control root rot fungi caused by *Fusarium* spp., *R. solani* and *M. phaseolina* but also enhanced plant growth. Hence, by using plant resources for its antifungal activity is good for the development of viable mode of agriculture in an organic farming system and more improvement in crop yield on cheaper bases can easily be done. For that reason, it is suggested that *M. oleifera* leaves needs to be introduced into fields on a large scale which represent an environment friendly strategy for controlling root rot fungi as a substitute for chemical fungicides. One more advantage of using the aqueous leaves extract that, it can easily use and is affordable to resource-limited farmers.

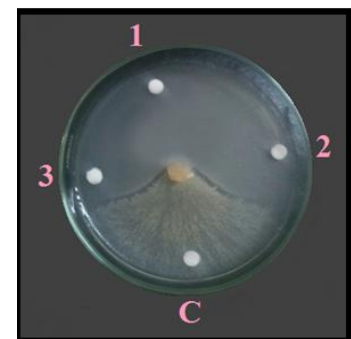
(Agar paper disc diffusion method)



F. oxysporum

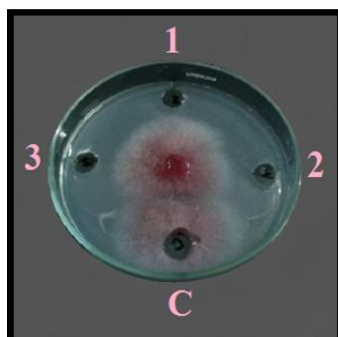


M. phaseolina

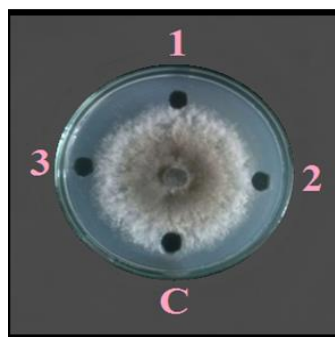


R. solani

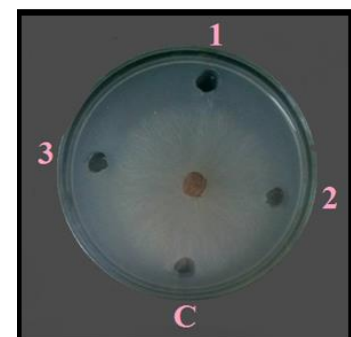
(Agar well diffusion method)



F. oxysporum



M. phaseolina



R. solani

Fig. 1. *In vitro*, inhibition of root rot fungi by aqueous extracts of *Moringa oleifera* leaves using paper disc and well diffusion methods. where, 1= 75% v/w concentration; 2=50% v/w concentration; 3= 25% v/w concentration; C= Control

Table 2. Methods of application by using *Moringa oleifera* parts on growth parameters of cowpea plant.

Methods	Treatments	Conc. (%)	Germination (%) ± SD	Shoot length (cm) ± SD	Shoot weight (g) ± SD	Root length (cm) ± SD	Root weight (g) ± SD	Number of nodules ± SD
Soil amendment	Control	0.0	53.00±11.53	14.46±1.84	0.57±0.34	10.09±5.03	0.48±0.22	11±4.58
		0.1	73.00±11.54	20.06±1.38	1.44±0.31	16.23±1.56	0.66±0.19	18±2.51
	Seed powder	1.0	40.00±20.00	15.90±2.59	1.22±0.71	10.29±1.96	0.41±0.11	03±2.51
		2.5	26.67±11.54	5.16±3.91	0.49±0.39	4.70±1.73	0.17±0.07	04±2.00
	Fruit powder	0.1	60.00±20.00	20.20±1.60	1.46±0.52	15.81±1.59	0.73±0.05	15±1.52
		1.0	40.00±20.00	16.69±4.88	1.01±0.23	12.87±0.32	0.68±0.03	13±2.64
		2.5	40.00±20.00	14.17±4.67	0.71±0.17	8.33±0.85	0.27±0.04	05±3.05
		0.1	100±0.0	27.65±4.83	2.01±0.07	18.39±0.83	0.97±0.05	25±2.08
	Leaves powder	1.0	100±0.0	21.12±1.29	2.68±0.26	15.58±2.58	0.57±0.05	24±4.00
		2.5	86.70±11.54	20.13±5.50	1.08±0.04	9.04±1.90	0.34±0.09	15±2.00
	Stem powder	0.1	100±0.0	25.40±1.27	2.09±0.13	13.50±2.71	0.57±0.09	18±1.52
		1.0	86.70±11.54	17.58±5.42	1.66±0.56	16.04±2.02	0.54±0.03	22±5.56
		2.5	40.00±20	15.09±1.14	0.54±0.12	9.91±2.48	0.35±0.18	13±4.72
Seed treatment	Control	0.0	40.00±20	17.10±2.95	0.51±0.06	11.22±1.22	0.29±0.06	14±4.93
		75	73.33±30.55	16.46±3.01	0.51±0.18	12.28±1.50	0.20±0.07	12±2.52
	Seed extract	50	86.67±23.09	15.12±1.23	0.62±0.30	13.29±0.87	0.21±0.03	14±1.53
		25	100±0.0	21.81±0.93	1.93±0.77	15.25±0.43	0.42±0.04	26±2.52
	Fruit extract	75	33.33±11.54	14.35±2.88	0.43±0.19	12.50±1.34	0.11±0.03	13±1.53
		50	73.33±11.54	15.80±2.02	0.66±0.16	14.29±1.13	0.18±0.01	14±1.53
		25	93.33±11.54	20.63±3.31	0.95±0.03	17.53±0.62	0.24±0.04	17±1.53
		75	93.33±11.54	29.67±2.00	3.24±0.62	15.53±0.70	0.62±0.02	28±6.00
	Leaves extract	50	100±0.0	30.68±0.86	3.99±0.65	20.10±0.62	0.77±0.06	33±5.00
		25	100±0.0	34.56±4.62	5.10±0.69	19.57±1.24	0.90±0.15	48±8.72
	Stem extract	75	46.70±11.55	16.20±2.17	0.75±0.29	11.50±2.59	0.32±0.04	14±2.00
		50	73.33±30.75	21.13±5.47	2.05±0.55	18.32±2.32	0.42±0.04	28±5.51
		25	100±0.0	25.57±3.86	2.89±0.45	20.12±4.43	0.56±0.04	56±5.86
Soil drenching	Control	0.0	66.67±11.55	15.86±0.4	1.03±0.34	9.15±1.19	0.22±0.08	12±4.93
		75	13.33±11.54	6.20±5.60	0.41±0.36	6.10±5.37	0.05±0.04	02±1.53
	Seed extract	50	20.00±20.00	9.70±8.40	0.45±0.39	8.11±7.39	0.07±0.07	03±3.00
		25	80.00±20.00	16.20±3.54	0.61±0.083	9.70±1.21	0.15±0.03	07±1.53
	Fruit extract	75	73.33±11.55	12.77±0.96	0.41±0.07	9.22±0.68	0.097±0.03	09±0.58
		50	66.67±11.54	14.07±0.47	0.48±0.06	10.14±0.77	0.16±0.02	10±2.08
		25	73.33±11.54	18.53±1.72	0.56±0.02	12.36±2.03	0.17±0.02	12±1.53
		75	86.67±11.55	22.44±1.26	1.08±0.04	13.83±0.93	0.35±0.04	43±1.00
	Leaves extract	50	100±0.0	25.21±1.39	1.63±0.13	14.93±0.56	0.36±0.02	48±4.00
		25	100±0.0	28.34±1.78	2.83±0.30	17.61±0.65	0.44±0.04	54±2.00
	Stem extract	75	66.67±23.09	18.44±1.49	0.92±0.06	14.76±2.92	0.29±0.03	25±3.00
		50	80±0.0	20.79±1.35	1.07±0.05	12.15±1.60	0.37±0.05	35±3.00
		25	100±0.0	26.36±2.33	1.99±0.24	16.04±1.54	0.62±0.03	44±2.08
LSD_{0.05}:								
Concentrations=			6.842	1.438	0.160	1.169	0.046	1.770
Plant parts=			6.842	1.438	0.160	1.169	0.046	1.770
Methods=			5.926	1.246	0.138	1.013	0.039	1.533

Where; Conc. = Concentrations, SD = ± Standard deviation

Table 3. Methods of application by using *Moringa oleifera* parts on growth parameters of mash bean plant.

Methods	Treatments	Conc. (%)	Germination (%) \pm SD	Shoot length (cm) \pm SD	Shoot weight (g) \pm SD	Root length (cm) \pm SD	Root weight (g) \pm SD	Number of nodules \pm SD
Soil amendment	Control	0.0	73.33 \pm 11.55	13.57 \pm 1.46	0.26 \pm 0.03	12.12 \pm 2.34	0.16 \pm 0.03	06 \pm 3.10
		0.1	93.33 \pm 11.55	20.81 \pm 1.63	0.30 \pm 0.12	14.09 \pm 3.73	0.24 \pm 0.075	10 \pm 5.51
	Seed powder	1.0	86.67 \pm 11.54	19.52 \pm 1.38	0.39 \pm 0.03	8.44 \pm 0.72	0.24 \pm 0.055	03 \pm 1.0
		2.5	66.67 \pm 30.55	9.57 \pm 2.62	0.04 \pm 0.01	3.48 \pm 1.77	0.06 \pm 0.015	00 \pm 0.0
		0.1	86.67 \pm 23.09	19.78 \pm 1.45	0.43 \pm 0.056	12.55 \pm 4.18	0.20 \pm 0.04	13 \pm 20.0
	Fruit powder	1.0	93.33 \pm 11.55	18.56 \pm 3.15	0.39 \pm 0.067	14.93 \pm 0.29	0.23 \pm 0.071	17 \pm 6.43
		2.5	73.33 \pm 11.54	14.87 \pm 0.98	0.28 \pm 0.04	9.21 \pm 0.62	0.15 \pm 0.026	10 \pm 2.08
		0.1	100 \pm 0.0	20.91 \pm 3.92	0.47 \pm 0.056	16.85 \pm 1.43	0.35 \pm 0.03	18 \pm 1.0
	Leaves powder	1.0	100 \pm 0.0	28.13 \pm 0.78	1.04 \pm 0.057	21.14 \pm 1.35	0.59 \pm 0.076	34 \pm 2.08
		2.5	100 \pm 0.0	21.14 \pm 0.55	0.45 \pm 0.09	15.17 \pm 4.33	0.28 \pm 0.04	26 \pm 2.08
		0.1	100 \pm 0.0	18.24 \pm 0.61	0.52 \pm 0.035	14.29 \pm 1.18	0.26 \pm 0.015	23 \pm 10.07
	Stem powder	1.0	100 \pm 0.0	20.39 \pm 1.74	0.65 \pm 0.031	16.25 \pm 2.54	0.33 \pm 0.042	24 \pm 2.0
2.5		93.33 \pm 11.54	16.65 \pm 1.41	0.33 \pm 0.05	9.64 \pm 5.76	0.24 \pm 0.053	15 \pm 2.52	
Seed treatment	Control	0.0	80.00 \pm 20.0	15.73 \pm 0.69	0.51 \pm 0.04	9.30 \pm 2.62	0.14 \pm 0.02	11 \pm 3.2
		75	93.33 \pm 11.55	17.23 \pm 0.39	0.72 \pm 0.21	12.18 \pm 1.94	0.11 \pm 0.026	14 \pm 2.65
	Seed extract	50	100 \pm 0.0	18.31 \pm 0.45	0.60 \pm 0.072	8.69 \pm 1.77	0.15 \pm 0.015	15 \pm 1.0
		25	100 \pm 0.0	17.93 \pm 0.17	0.52 \pm 0.032	10.05 \pm 2.56	0.16 \pm 0.038	16 \pm 2.08
		75	73.33 \pm 11.55	14.65 \pm 0.54	0.54 \pm 0.015	8.45 \pm 0.46	0.13 \pm 0.042	09 \pm 2.0
	Fruit extract	50	80 \pm 20.00	15.84 \pm 0.89	0.51 \pm 0.026	7.49 \pm 0.15	0.07 \pm 0.021	05 \pm 1.53
		25	93.33 \pm 11.55	20.33 \pm 0.98	0.46 \pm 0.021	7.81 \pm 0.52	0.10 \pm 0.02	10 \pm 1.0
		75	100 \pm 0.0	22.93 \pm 1.42	1.88 \pm 0.038	19.94 \pm 1.26	0.76 \pm 0.021	41 \pm 2.05
	Leaves extract	50	100 \pm 0.0	27.07 \pm 0.61	1.85 \pm 0.03	20.54 \pm 2.71	0.87 \pm 0.05	48 \pm 3.51
		25	100 \pm 0.0	28.17 \pm 0.72	1.78 \pm 0.06	25.78 \pm 1.47	0.86 \pm 0.02	46 \pm 1.0
		75	100 \pm 0.0	20.02 \pm 0.35	1.44 \pm 0.11	19.81 \pm 0.43	0.41 \pm 0.03	32 \pm 3.52
	Stem extract	50	100 \pm 0.0	21.61 \pm 0.78	0.96 \pm 0.015	18.41 \pm 1.11	0.35 \pm 0.021	25 \pm 1.53
25		100 \pm 0.0	17.15 \pm 1.64	0.91 \pm 0.031	17.58 \pm 1.94	0.31 \pm 0.076	20 \pm 2.08	
Soil drenching	Control	0.0	80.00 \pm 0.0	16.07 \pm 1.81	0.28 \pm 0.04	10.67 \pm 0.89	0.13 \pm 0.032	07 \pm 3.21
		75	40.00 \pm 20.00	13.81 \pm 1.67	0.25 \pm 0.015	9.93 \pm 1.35	0.07 \pm 0.026	04 \pm 1.53
	Seed extract	50	26.67 \pm 11.54	14.93 \pm 2.27	0.21 \pm 0.03	6.49 \pm 2.63	0.09 \pm 0.035	08 \pm 2.08
		25	73.33 \pm 11.54	17.12 \pm 0.52	0.31 \pm 0.026	11.39 \pm 0.85	0.15 \pm 0.025	14 \pm 1.53
		75	33.33 \pm 11.54	12.33 \pm 0.89	0.29 \pm 0.03	6.29 \pm 0.54	0.07 \pm 0.01	03 \pm 0.58
	Fruit extract	50	53.33 \pm 11.54	13.64 \pm 0.54	0.29 \pm 0.038	9.01 \pm 1.16	0.09 \pm 0.036	06 \pm 1.0
		25	66.67 \pm 11.54	16.33 \pm 0.66	0.26 \pm 0.01	8.89 \pm 0.42	0.12 \pm 0.026	12 \pm 1.53
		75	86.67 \pm 11.55	23.93 \pm 1.68	1.03 \pm 0.02	23.46 \pm 2.18	0.68 \pm 0.038	25 \pm 2.65
	Leaves extract	50	100 \pm 0.0	26.77 \pm 2.00	1.45 \pm 0.32	20.53 \pm 0.79	0.73 \pm 0.04	44 \pm 7.21
		25	100 \pm 0.0	33.60 \pm 1.63	2.02 \pm 0.04	24.66 \pm 1.13	0.83 \pm 0.04	52 \pm 3.51
		75	66.67 \pm 11.55	19.05 \pm 1.85	0.71 \pm 0.03	12.46 \pm 0.91	0.36 \pm 0.015	18 \pm 3.0
	Stem extract	50	80 \pm 20.00	20.86 \pm 1.82	0.89 \pm 0.03	13.42 \pm 0.84	0.42 \pm 0.035	22 \pm 2.65
25		100 \pm 0.0	26.54 \pm 1.09	1.09 \pm 0.06	15.94 \pm 0.83	0.47 \pm 0.02	28 \pm 3.51	
LSD_{0.05}: Concentrations=			5.569	0.723	0.033	0.960	0.017	1.504
Plant parts=			5.569	0.723	0.033	0.960	0.017	1.504
Methods=			4.823	0.626	0.029	0.832	0.014	1.302

Where; Conc. = Concentrations, SD = \pm Standard deviation

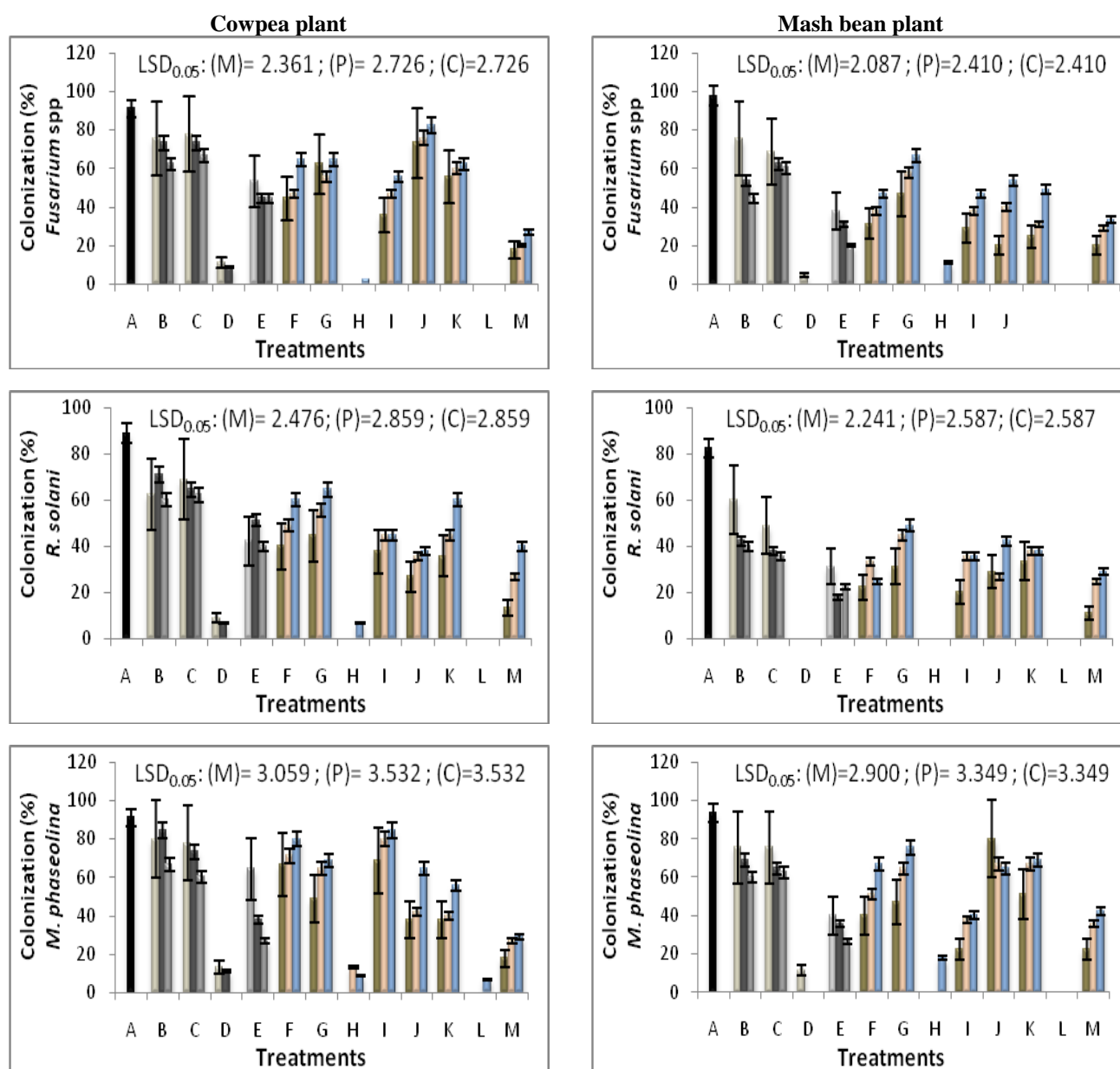


Fig. 2. Methods of application by using *Moringa oleifera* parts in the control of root rot fungi on crop plants.

Control ■ Soil amendment: 0.1% □ 1.0% ■ 2.5% w/w concentrations;
 Seed treatment and soil drenching: 75% ■ 50% ■ 25% v/w concentrations.
 (M)= Methods, (P) = Plant parts, (C) = Concentrations

where, A= Control, B= Soil amended with 0.1%; 1.0 and 2.5% w/w *M. oleifera* seed powder, C = Soil amended with 0.1%; 1.0 and 2.5% w/w *M. oleifera* fruit powder, D= Soil amended with 0.1%; 1.0 and 2.5% w/w *M. oleifera* leaves powder, E= Soil amended with 0.1%; 1.0 and 2.5% w/w *M. oleifera* stem powder, F= Seed treated with 75%; 50% and 25% v/w *M. oleifera* seed extract, G= Seed treated with 75%; 50% and 25% v/w *M. oleifera* fruit extract, H = Seed treated with 75%; 50% and 25% v/w *M. oleifera* leaves extract, I = Seed treated with 75%; 50% and 25% v/w *M. oleifera* stem extract, J=Soil drenched with 75%; 50% and 25% v/w *M. oleifera* seed extract, K= Soil drenched with 75%; 50% and 25% v/w *M. oleifera* fruit extract, L=Soil drenched with 75%; 50% and 25% v/w *M. oleifera* leaves extract, M= Soil drenched with 75%; 50% and 25% v/w *M. oleifera* stem extract.

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