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SEED PRIMING WITH EXTRACTS OF SOME PLANT SPICES IN THE MANAGEMENT OF ROOT PATHOGENS OF MUNG BEAN

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ABSTRACT

Natural products are considered as possible alternative for managing plant diseases. Fungicidal activity of four spices namely of *Trachymyspermum ammi* (L.), *Nigella sativa* (L.), *Foeniculum vulgare* (Mill.) and *Cinamonum verum* (J. Persl.) were tested against root rot fungi viz., *Fusarium oxysporum*, *Rhizoctonia solani* and *Macrophomina phaseolina* using paper disc method. The results of *in vitro* assay exhibited that *M. phaseolina* and *F. oxysporum* were inhibited by 100 % w/v extract of *F. vulgare* while *T. ammi* was more effective in inhibition of *Rhizoctonia solani* at 100 % concentration. The results of in *vivo* experiments revealed that root, shoot length and weight was maximum when *N. sativa* was primed at 100 % w/v concentration while all spices except *N. sativa* showed complete reduction of the colonization of *R. solani* in mung bean roots. However, *C. verum* and *F. vulgare* reduced the colonization of *M. phaseolina* and *F. oxysporum* in mung bean roots.

Key words: aqueous extract, paper disc method, Root infecting fungi, seed priming,

INTRODUCTION

Seed priming is one of the most effective technology to enhance rapid development of the seed and better yield (Harris *et al*., 2007). Seed priming technique has been practiced in many countries including Pakistan, China and Australia and proved to be cheaper cost effective strategy to increase growth of allocated crops (Farooq *et al*., 2009). It is a pre-sowing seed treatment, performed in controlled conditions while emergence of radical is avoided by drying before sowing (Basra *et al*., 2006).

Although many strategies have been suggested as well as developed for the control of pathogenic diseases. Plants parts in the form of powder were used for controlling plant diseases as well as preservative in food and other stuff. They are also used as a fungicide which has great importance and needs more attention (Bodde, 1982). Spices are recognized for the prevention of a microbial deterioration of food etc. *Trachyspermum ammi* L. (Ajwian) belonging to family Apiaceae is a highly valued spice which contains 2–4.4% brown colored oil known as ajwain oil. The main component of this oil is thymol, which is used in the treatment of gastro-intestinal ailments, and also exhibits fungicidal, antimicrobial and anti-aggregatory effects on human (Singh and Singh, 2000; Sivropoulou, *et al.*, 1996). *Foeniculum vulgare*, (Fennel), is a well-known medicinal plant containing trans-anethole, estragole, fenchone, and α-phellandrene with the variable concentration depending on the phenological state and origin of the plant (Senatore *et al.*, 2013). *Cinnamomum zeylanicum* (L.), commonly known as cinnamon, is endemic to Sri Lanka. Apart from these, β-caryophyllene, linalool, cinnamaldehyde and other terpenes are present (Paranagama, 1991). *Nigella sativa* is a herbaceous plant which is also known as black seed, contains up to 36–38% fixed oil, with saponins, alkaloids, proteins and essential oils making up the rest of the composition (Burtis and Bucar, 2000). Black seed extract or oil has been reported to possess antimicrobial activity including antioxidant as well as antitumor activity (Morsi, 2000).

The main purpose of controlling plant root rot diseases and to improve the health and growth of the plants as well as improving the production and quality of plant by reducing the parasitic pathogen. The objective of this research was to perform antifungal of some botanical plant parts (spices) used against root rot diseases of mung bean.

MATERIALS AND METHODS

Seeds of *Trachymyspermum ammi* L., *Nigella sativa* L., *Foeniculum vulgare* (Mill.) and *Cinamonum verum* (J. Persl.) were purchased from the local market of Karachi. These spices were than grind to form fine powder and

prepare aqueous extract with different concentrations. Pure cultures of *Fusarium oxysporum*, *Rhizoctonia solani* (Kuhn), *Macrophomina phaseolina* (Tassi) Goid were obtained from KUCC (Karachi University Culture Collection) and maintained on Potato Dextrose Agar (PDA) using antibiotics, penicillin and streptomycin, to avoid bacterial growth.

In vitro experiment of aqueous extract of spices was performed by paper disc method to observe its antifungal property. In paper disc method, 6 mm disc of root rot fungi were placed in the centre of Potato Dextrose Agar (PDA) filled petri plate and at the other three corners, disc of different concentration of aqueous extract of spices were placed. The fourth corner was received disc soaked in sterilized distilled water for control. There was three replicates of each treatment and were incubated at 30°C for 5-6 days.

A pot experiment was performed in the screen house of Department of Botany, University of Karachi, where pots were filled with 300 g sandy loam soil (having sand 72%, silt 16% and clay 12%). Seeds of mungbean were surface sterilized with 1% of calcium hypocloride for 2-3 minutes, rinsed it under running tap water and then dried. Seeds were primed with 50, 75 and 100% of each spice extract for 5-7 minutes and after drying, five treated seeds were sown into each pot. Seeds primed with sterilized distilled water were served as control. Each treatment was replicated three times and was kept for 30 days. After thirty days of germination, plants uprooted for observation of growth parameters and colonization of root infecting fungi.

RESULTS AND DISCUSSION

In Vitro Studies

All spices gave proper zone of inhibition against *R. solani* at all concentrations. However, maximum zone of inhibition was recorded by *T. ammi* at 100% w/v against *R. solani* followed by *F. vulgare* extract at 100 % w/v. In case of *F. oxysporum* and *M. phaseolina*, maximum zone of inhibition was recorded by 100% *F. vulgare* extract. However, *T. ammi* extract showed no effect on *F. oxysporum* and *M. phaseolina* growth (Table 1). Similar report was given by Kalleli *et al.* (2020) where antifungal activity from oil of fennel seeds was investigated against *F. oxysporum* f. sp. *lycopersici* where oil inhibited 83% mycelium growth and sporulation up to 97%. This may be due to chemical components like fenchone, estragole, limonene and trans-anethole present in essential oil of fennel (Kalleli *et al.*, 2020).

Table 1. Inhibition of root rot fungi by aqueous extracts of spices.

Treatments		*Macrophomina phaseolina*	*Fusarium oxysporum*	*Rhizoctonia solani*
Spices extract	Concentration (%)	Zone of inhibition (mm)		
Control	0	0	0	0
Nigella sativa	100	0	1.2	1.5
N. sativa	75	0	1.1	1.1
N. sativa	50	0	0.7	0
Foeniculum vulgare	100	1.2	1.5	1.9
F. vulgare	75	1.1	1.4	0.9
F. vulgare	50	0.9	1.3	0
Cinamonum verum	100	1.9	1.3	1.8
C. verum	75	1.4	1.3	1.7
C. verum	50	1.0	0.8	0.8
Trachymyspermum ammi	100	0	0	2.0
T. ammi	75	0	0	1.8
T. ammi	50	0	0	1.4

Pot experiment

Seed priming of mung bean seeds revealed that improved growth was achieved when all spices with different concentrations was used in contrast to control. Maximum shoot and root length was attained when seeds primed with *N. sativa* at 100 % w/v followed by 75 % w/v ($P<0.05$). However, *N. sativa*, *F. vulgare* and *C. verum* at 100 % w/v gave improved shoot and root weight compared to control ($P < 0.05$). Result showed that as the concentration increased, growth parameter of mung bean was also increased (Table 2). *N. sativa* extracts produce active role in improving growth and productivity of maize genotypes (Neelum and P1543) at different concentrations (Allah Ditta *et al.*, 2021). Colonization of roots by *M. phaseolina* was reduced when seed primed with *C. verum* at 100 % w/v while *F. vulgare* at 100 % w/v showed reduced *F. oxysporum* colonization on roots. However, all spices except *N. sativa* showed completely controlled the colonization of *R. solani* (Table 3). According to Khan *et al.* (2003), *N. sativa* produced inhibitory effect against *C. albicans.* It may be due to presence of active ingredients like oleic acid and β–sitosterol leading to an excellent antifungal activity (Asdadi *et al.*, 2014).

Table 2. Seed priming with aqueous extracts of spices on the growth of mung bean.

Spices extract	Concentration (%)	Shoot Length (cm) ± SD	Shoot Weight (g) ± SD	Root Length(cm) ± SD	Root Weight(g) ± SD
Control	0	11 ± 1.68	0.12 ± 0.028	3.6 ± 0.7	0.02 ± 0.005
N. sativa	100	16.6 ± 0.47	0.16 ± 0.017	5.8 ± 0.36	0.04 ± 0.005
N. sativa	75	15.6 ± 0.60	0.15 ± 0.03	5.1 ± 0.75	0,04 ± 0.01
N. sativa	50	14.3 ± 1.77	0.13 ± 0.005	4.6 ± 0.32	0.03 ± 0.02
F. vulgare	100	16.4 ± 0.50	0.16 ± 0.02	4.9 ± 1.19	0.04 ± 0.01
F. vulgare	75	14.7 ±1.20	0.15 ± 0.01	4.7 ± 0.23	0.03 ± 0.02
F. vulgare	50	13.4 ± 0.52	0.14 ± 0.01	4.4 ± 0.47	0.02 ± 0.01
C. verum	100	12.6 ± 1.55	0.16 ± 0.2	5.7 ± 0.52	0.05 ± 0.01
C. verum	75	14.6 ± 1.65	0.13 ± 0.1	4.6 ± 0.7	0.03 ± 0.005
C. verum	50	12.6 ± 1.55	0.12 ± 0.028	3.8 ± 0.83	0.03 ± 0.01
T. ammi	100	14.2 ±1.38	0.14 ± 0.02	4.0 ± 0.26	0.04 ± 0.01
T. ammi	75	12.9 ±1.35	0.13 ± 0.005	3.9 ± 0.7	0.04 ± 0.01
T. ammi	50	12.1 ± 1.46	0.13 ± 0.02	3.6 ± 0.58	0.02 ± 0.005
$LSD_{0.05}$ Spices		1.05	0.01	0.50	0.009
Concentration		1.05	0.01	0.50	0.009

Table 3. Seed priming with aqueous extract of spices in the control of root rot fungi of mung bean.

Spices extract	Concentration (%)	*Macrophomina phaseolina* Mean ± SD	*Fusarium* spp. Mean ± SD	*Rhizoctonia solani* Mean ± SD
Control	0	33 ± 8.0	35.6 ± 9.23	55.3 ± 19.23
N. sativa	100	16.4 ± 8.35	19.1±11.9	2.7 ± 4.79
N. sativa	75	19 ± 5.19	21.7 ± 17.0	5.3 ± 9.23
N. sativa	50	30.3 ± 9.23	24.7 ± 14.2	5.3 ± 9.23
F. vulgare	100	22.1 ± 12.60	5.3 ± 8.50	0 ± 0
F .vulgare	75	24.3 ± 14.43	8.1 ± 8.00	0 ± 0
F. vulgare	50	27.4 ± 17.04	24.6 ± 8.50	2.7 ± 4.79
C. verum	100	10.8 ± 4.44	8.1 ± 8.00	0 ± 0
C. verum	75	22.1 ± 1.26	22 ± 5.19	0 ± 0
C. verum	50	24.6 ± 8.50	22 ± 19.05	8.3 ±14.4
T. ammi	100	16.4 ± 8.35	11.1 ± 12.7	0 ± 0
T. ammi	75	19.1 ± 12.6	24.6 ±8 .50	0 ± 0
T. ammi	50	22.0 ± 5.1	30.3 ± 9.23	0 ± 0
$LSD_{0.05}$Spices		8.30	65.82	5.75
Concentration		8.30	65.82	5.75

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