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Effect of extracts of various plant parts on seed mycoflora and seed germination of tomato

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ABSTRACT

Total sevetneen fungi were found to be associated with the seeds of tomato. The seeds of tomato var. local showed maximum seed mycoflora with maximum per cent incidence. The common and dominant fungi recorded were *Aspergillus niger, Aspergillus flavus, Fusarium moniliforme, Rhizopus nigricans, Curvularia lunata* and *Alternaria alternata.* The common and dominant seed borne fungi were found to be inhibitory for seed germination and caused great loss in seedling vigor, seed and seedling rots of tomato var. local. The root, stems, leaf and bark extracts of some common and easily available plants were screened for the bio-control seed mycoflora associated with tomato. The extracts of all the test plants were found to be inhibitory in more or less degree for the incidence of seed mycoflora while with a few exceptions, they were found to be stimulatory for seed germination.

Key words : Tomato, seed mycoflora, Seed germination, Seedling vigor, Seedling emergence

INTRODUCTION

Solanaceae family includes a large number of annual or perennial herbs, shrubs, small trees and climbers. More than seventy species belonging to twenty-one genera are found in India. Economically the family is fairly important, as it comprises of several crops of food value, medicinal value, vegetables and ornamentals. Several plants of this family are cultivated all over the world for their economic importance.

Tomato (*Lycoperiscon esculentum*) is the most widely grown vegetable crop. It is grown through out the year in Marathwada region in fields, gardens, small home gardens and by market gardeners for fresh consumption of fruits (berries) as well as for processing purposes. Tomato has an outstanding vitamin contents like ascorbic acid (vitamin C), vitamin A, Thiamine (vitamin B) and riboflavin (vitamin B_2). Tomato fruits are used in many ways.

In the present study, ten local and easily available plants in the near by area were selected for their root, stem, leaf and bark extracts and the effects of these extracts on seed mycoflora and seed germination were studied.

MATERIALS AND METHODS

Collection of seed samples:

The methods described by Neergaard (1973) has been adopted for the collection of seed samples. Accordingly, seed samples of different var. of tomato (50 g each) were collected from ripe dried fruits from field, storehouses, market places and research centers. A composite seed sample for each of the var. was prepared by mixing the individual seed samples together and preserved in gunny bags at room temperature during the studies.

Detection of seed mycoflora:

The seed-borne fungi of different varieties of seeds of tomato were detected by moist blotter (B) and agar (A) plate methods as recommended by ISTA (1966), De Tempe (1970), Neergaard (1973) and Agarwal *et al.* (1976). The procedure of moist blotter (B) and agar (A) plate methods are described as below.

Identification of seed-borne fungi:

The seed-borne fungi were preliminary identified on the basis of sporulation characters like asexual or sexual spores or fruiting structures. Detailed examination of fungal characters was done under compound microscope and their identification was confirmed with the help of related manuals (Subramanian, 1971, Neergaard and Mathur, 1980, Jha, 1993 and Mukadam *et al.,* 2006). Pure cultures of the identified fungi were prepared and maintained on PDA (Potato dextrose agar) slants for further experiments.

Effect of culture filtrates on per cent seed germination, root length, shoot length and seedling emergence:

Production of toxin was studied by growing some common and dominant seed-borne fungi of plants like *Alternaria alternata, Aspergillus flavus, Curvularia lunata* and *Fusarium moniliforme* on liquid GN medium of pH 5.6 for ten days.

Twenty five ml of the medium was poured in 100 ml

Borosil glass conical flasks, autoclaved and inoculated separately with 2 ml spore suspension of the test seed-borne fungus that was maintained on PDA slants for seven days. The flasks were incubated at room temperature (27±1^{0}C) for ten days. After incubation, the culture filtrates were collected in pre-sterilized culture bottles from the flasks by filtering the contents through Whatman filter paper No.1 and treated it as crude toxin preparation.

Collection of plant material for extracts:

During the present studies, ten common and easily available plants in the vicinity like *Acacia nilotica, Adhatoda zeylanica, Annona squamosa, Azadirachta indica, Curcuma longa, Lawsonia inermis, Murraya koenigii, Ocimum sanctum, Terminalia bellerica* and *Terminalia chebula* were selected. Their identification was confirmed using the 'Flora of Marathwada' (Naik, 1998). The roots, stems, leaves and barks of the selected plants were collected separately, surface sterilized with 0.1 % $HgCl_2$ and washed repeatedly with sterile distilled water for several times and kept for drying in hot air oven (Metalab) at 60^{0}C temperature for 48 hours. After drying, the roots, stems, leaves and barks were preserved separately in polythene bags at room temperature (27± 1^{0}C) during the studies.

The dried roots, stems leaves and bark of selected plants were crushed separately into fine powder with the help of blender (Remi). 5 g powder each of the plant parts was dissolved separately in 100 ml sterilized hot distilled water in 250 ml Borosil glass conical flasks. The flasks were kept in oven (Metalab) for 24 hours at 60^{0}C and the content was filtered through Whatman filter paper No.1. The filtrates were used as 5% aqueous plant extracts.

Effect of extracts of various plant parts on seed mycoflora and seed germination:

During the present studies, the seeds of different varieties of tomato were placed on blotters in Petri plates as described earlier and with the root, stem and leaf extracts (5%) of the selected plants. Per cent seed germination and associated seed mycoflora were recorded on seventh day. Seed plates moistened with sterile distilled water served as control.

RESULTS AND DISCUSSION

Seeds of four different varieties of tomato were screened for incidence of fungi for seven days by Agar plate and Blotter test methods.

Results presented in Table 1 show that in all seventeen fungi were recorded from seeds of different verities of tomato.

Table 1 : Incidence of mycoflora on seeds of different varieties of tomato by agar plate (A) and blotter test (B) methods (after 7 days of incubation)

Sr. No.	Seed mycoflora	% Incidence of seed mycoflora							
		Tomato local		Pusa Ruby		PKM-1		Lakshmi	
		A	B	A	B	A	B	A	B
1.	*Alternaria alternata*	37	27	33	23	27	19	21	17
2.	*Alternaria solani*	30	25	24	21	19	18	17	14
3.	*Aspergillus niger*	72	48	67	43	61	37	51	31
4.	*Aspergillus flavus*	62	52	57	46	53	42	47	37
5.	*Aspergillus fumigatus*	-	10	-	07	-	04	-	03
6.	*Rhizopus stolonifer*	28	26	23	21	19	18	11	13
7.	*Rhizopus nigricans*	48	33	41	27	33	23	27	19
8.	*Fusarium solani*	15	12	11	08	09	05	06	03
9.	*Fusarium moniliforme*	50	30	47	26	41	22	33	19
10.	*Rhizoctonia solani*	10	05	09	03	07	02	04	02
11.	*Penicillium digitatum*	10	05	08	04	07	03	04	02
12.	*Chaetomium* species	-	-	-	-	-	-	-	-
13.	*Curvularia lunata*	45	30	39	23	31	19	27	13
14.	*Pythium debaryanum*	05	-	04	-	03	-	03	-
15.	*Phytophthora capsici*	-	10	-	09	-	07	-	05
16.	*Cladosporium herbarum*	-	-	-	-	-	-	-	-
17.	*Helminthosporium speciferum*	-	-	-	-	-	-	-	-

Table 2 : Effect of culture filtrates (CF) on seed germination, root length, root rot, shoot length, shoot rot (on blotter) and seedling emergence of tomato variety-local (after ten days of incubation)

Sr. No.	CF of common and dominant seed borne fungi	Tomato variety-Local					
		Seed germination (%)	Root length (mm)	Root rot	Shoot length (mm)	Shoot rot	Seedling emergence (%)
1.	*Alternaria alternata*	27	6.2	+++	20.8	+++	18
2.	*Aspergillus flavus*	17	4.5	+++	18.8	+++	13
3.	*Curvularia lunata*	61	48.9	+	47.2	+	32
4.	*Fusarium moniliforme*	39	32.9	++	44.6	+	18
5.	Control (Sterile GN-medium)	100	112	-	100	-	56

+++ = Severe rot ++ = Moderate rot + = Low rot
\- = No rot

Of the seeds of tomato varieties screened for incidence of fungi tomato var. local showed maximum incidence of fungi followed by varieties Pusa Ruby, PKM-1 and Lakshmi.

Seeds on agar plates shown more fungi than the on the blotters.

The common and dominant fungi recorded were *Aspergillus niger, Aspergillus flavus, Fusarium moniliforme, Rhizopus nigricans, Curvularia lunata* and *Alternaria alternata.*

Culture filtrates of some common and dominant seed borne fungi were studied on seed germination, seedling emergence and seedling health of Solanaceous plants. Ten day old culture filtrates obtained from seed borne fungi grown on GN medium were used in experiments to see their effects on seed germination, seedling emergence and seedling health after ten days.

Results presented in Table 2 show that there was maximum inhibition of seed germination in culture filtrate of *Aspergillus flavus* (seed germination 17%) followed by *Alternaria alternata* (27%), *Fusarium moniliforme* (39%) and *Curvularia lunata* (61%) over the control (100%).

From the results it is also clear that, the seeds treated with culture filtrate of *Aspergillus flavus* showed maximum reduction in root length (4.5 mm, control 112.0 mm), shoot length (18.8 mm, control 100 mm) and maximum root rot and shoot rot. Where as the seeds treated with culture filtrate of *Curvularia lunata* showed minimum reduction in root length (48.9 mm, control 112 mm), shoot length (47.2 mm, control 100 mm) and minimum root rot and shoot rot.

It is also clear from the results that there was maximum inhibition of seedling emergence in culture filtrate of *Aspergillus flavus* (13%) followed by *Alternaria alternata* (18%), *Fusarium moniliforme* (18%) and *Curvularia lunata* (32%) over the control (56%).

Table 3 : Effect of extracts of various plant parts on per cent seed mycoflora and per cent seed germination of Tomato var. local on blotter paper (after seven days)

Sr. No.	Source plant	Part used for extracts	% Seed mycoflora	% Seed germination
1.	*Acacia nilotica*	Root	75	41
		Stem	79	39
		Leaf	66	47
		Bark	68	57
2.	*Adhatoda*	Root	60	39
	zeylanica	Stem	65	51
		Leaf	43	67
3.	*Annona squamosa*	Root	51	55
		Stem	43	62
		Leaf	27	73
4.	*Azadirachta indica*	Bark	16	92
		Leaf	14	89
		Kernel	18	95
5.	*Curcuma longa*	Dried rhizome	46	71
		Leaf	61	35
6.	*Lawsonia inermis*	Root	57	49
		Stem	80	28
		Leaf	36	64
7.	*Murraya koenigii*	Root	16	89
		Stem	25	79
		Leaf	06	91
8.	*Ocimum sanctum*	Root	07	94
		Stem	10	92
		Leaf	04	95
9.	*Terminalia*	Root	50	38
	bellerica	Bark	54	50
		Leaf	59	27
10.	*Terminalia*	Root	47	43
	chebula	Bark	44	55
		Leaf	43	31
	Control (sterile distilled water)	--	100	33

During the present studies, the seeds of tomato var. local were placed on blotters in Petri plates and moistened with root, stem and leaf extracts of different plants (total ten plants). The plates were incubated for seven days at room temperature and the incidence of seed mycoflora and seed germination was studied. The plates moistened with sterile distilled water served as control. The results are presented in Table 3.

From the results (Table 3) it is evident that, the root, stem and leaf extracts of all the test plants were found to be inhibitory in more or less in the same degree for the incidence of seed mycoflora while with a few exceptions, they were found to be stimulatory for seed germination.

The seeds treated with leaf extracts of *Azadirachta indica,* leaf and root extracts of *Ocimum sanctum* and leaf extracts of *Murraya koenigii* showed very reduced incidence of seed mycoflora and maximum seed germination while, the seeds treated with the stem and root extracts of *Lawsonia inermis* and *Acacia nilotica*, leaf extract of *Curcuma longa* and *Terminalia bellerica* showed maximum incidence of seed mycoflora and reduced seed germination.

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