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STUDIES ON GUAVA (*PSIDIUM GUAJAVA* L.) DRYING/WILT DISEASE IN ORCHARDS OF PUSHKAR VALLEY

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A serious decline in number of guava trees was noted due to drying/wilt disease, which significantly reduced the guava orchard area in Pushkar valley, Isolations were made on 2% PDA from different parts of plant namely root, root bark, stem bark, stele (><2cm), twigs, leaf and apex portion of branch, Fungal cultures showing maximum similarity were purified and were sent to Indian Type Culture Collection, I.A.R.I. and identified as *Fusarium oxysporum* Schl. f. sp *psidii, Fusarium solani* Mart. Sacc., *Rhyzoctonia solani* Kuhn. and *Pestalotiopsis disseminata. F. oxysporum* and *F. solani* were proved to be pathogenic while *R. solani* and *P. disseminata* were associated fungi for drying/wilt disease of guava. Mother extract of Bilva (5%) and Sadabahar (2%), Metal salts of magnesium sulfate and Borax each 10^{-4} M were proved effective in inhibiting the mycelial growth of pathogens and associated fungi *in vitro* using poisoned food technique. These were at par with effective fungicides bavistin and topsin-M.

Keywords : Control; Drying/wilt disease; Fungal pathogen; *Psidium guajava* L.

Introduction

Guava (*Psidium guajava* L.) is an important fruit crop of subtropical regions and cultivated in India as a cash crop and known for its rich vitamins and nutritive values[1]. In Rajasthan guava is cultivated in 1593 hectares with 22255 tones production[2].

The incidence of wilt disease in guava plantation of Varanasi and adjacent districts of U.P. ranged from 3.9 to 30 per cent[3]. It suffers badly from the dreaded wilt disease. The exact cause of the disease is not fully understood, but the pathogens like *Fusarium oxysporum* f. sp. *psidiii, F. solani, Macrophomina phaseoli, Rhizoctonia bataticola, Cephalosporium* sp. and *Gliocladium roseum* are reported to be the cause of the disease[4]. Since 1991 a serious decline of guava has been noted due to drying/wilt disease in Pushkar valley. Due to this malady several orchards have been cut down in the area. Therefore, a systemic study on isolation, pathogenicity and evaluation of non-conventional chemicals along with effective fungicides were undertaken *in vitro* and their results are reported here in.

Materials and Methods

Isolation and Pathogenicity : Isolations were made from different parts of the infected plants viz., root, root bark, stele bark, steal (>< 2 cm), twigs, leaf and apex portion of branch on 2 per cent potato dextrose agar (PDA) medium. Fungal culture showing maximum similarity were purified and identified at IARI as *Fusarium oxysporum* Schl. f. sp *psidii, F. solani* Mart. Sacc., *Rhizoctonia solani* Kuhn. and *Pestalotiopsis disseminata.* Pathogenicity experiment was conducted in pots of cage house, Air-dried grounded soil sterilized with 2 per cent formaldehyde solution[5] was used for filling the pots (12" dia.), these pots were sterilized with 0.5% copper sulphate solution. *F. oxysporum* was multiplied on maize meal medium[6] while *R. solani and F. solani* on oat meal sand medium[7] incubated at 25 °C for a fortnight to ensure good fungal growth. The prepared inoculum was mixed with sterilized soil @ 10 per cent to which guava 3-7 months old plants were transplanted. The inoculated plants were kept under regular watch and data of infection/replication were recorded.

Growth inhibition test : Leaf extract of asoka (*Polyalthia longifolia* L.), bilva (*Aegel marmelos* correa. ex Roxb) and sadabahar (*Vinca rosea* L.); were prepared and used as mother extracts (ME) at concentration of 5, 5 and 2%, respectively[8]. Metal salts of magnesium sulphate ($MgSO_4 7 H_2O$),

manganese sulphate ($MnSO_4.H_2O$) and borax as sodium tetraborate ($Na_2B_4O_7.10H_2O$) each used in 10^{-4} M concentration. Cultural filtrate of *Trichoderma harzianum* (100%), neem oil (800 ppm), along with fungicides carbendazim (bavistin 50 WP, 0.1%), mancozeb (indofil-M-45, 0.2%) and thiophanate methyl (topsin - M 70 WP, 0.1%) were used for *invitro* toxicity studies by using poisoned food technique[9] in three replication against *F. solani* and *F. oxysporum* (pathogenic), *R. solani* and *P. disseminata,* the associated fungi causing drying/wilt of guava and compared with control. the observation on mycelial growth was recorded by measuring fungal growth diagonally and per cent inhibition of growth was calculated.

Results and Discussion

Data (Table 1) showed that fungi isolated from different parts of the infected plants showing drying/wilt symptoms, *F. oxysporum* f.sp. *psidii* and *F. solani* were highly pathogenic to guava as the Koch's postulates proved for these fungi and they caused disease (100% infection) in all inoculated plants while *R. solani* showed infection up to 33.33 per cent, it is associated fungus and could not be re-isolated during pathogenicity test.

It is evident from the data (Table 2) that all the given treatments were effective in inhibition of mycelial growth of all fungi studied *in vitro.* Among these treatments topsin-M was found highly effective against all test fungi in inhibiting average mycelial growth (85.67%) followed by carbendazim (84.46%), *V. rosea* (75.30%), *A. marmelos* (67.21%), magnesium sulphate (64.81%) and manganese sulphate (62.02%) while borax (33.20%) found less effective as compared with check (4.04%). Maximum mycelial growth inhibition of *F. oxysporum* was recorded with carbendazim (85.94%), topsin-M (85.94%) and ME of *V. rosea* (81.15%) and were at par with each other. In *F. solani,* carbendazim (80.02%) and topsin-M (84.86%) were found effective in inhibiting the mycelial growth and were at par to each other. Magnesium sulphate (metal salts) and ME of *V. rosea* and *A. marmelos* were found effective in inhibiting the mycelial growth of *R. solani* (85.94%), and were at par with carbendazim and topsin-M. Neem oil was also found effective in reducing average (50.38%) growth of all test fungi. *T. harzianum* inhibited the growth of all test fungi but it was highly antagonistic against *R. solani* (71.80%) than *F. oxysporum* f.sp. *psidii* (67.05%), *P. disseminata* (27.24%) and less against *F. solani* (19.18%).

Maximum inhibition of mycelial growth of *P. disseminata* causing leaf and twig blight of guava was recorded with ME of *V. rosea* (85.94%), metal salts of $MnSO_4$ (82.33%) and both the systemic fungicides were at par in their efficacy.

Fungal cultures showing maximum similarity were purified and sent to ITCC, IARI, New Delhi and were identified as *F. oxysporum* f. sp. *psidii, F. solani, R. solani* and *P. disseminata F. oxysporum* f. sp. *psidii* and *F. solani* were proved to be pathogenic while *R. solani* and *P. disseminata* were associated fungi for drying/wilt disease of guava as they could not re-isolated during pathogenicity. This may be due to invasion of roots, plant became weak, followed the defoliation of leaves and death of twigs, possibly involvement of toxin like substance present in root[7]. The exact cause of disease is not fully understood earlier, but the pathogens like. *F. oxysporum* f. sp. *psidii,*

Table 1. Pathogenicity of guava seedlings against *F. oxysporum, F. solani.* and *R. solani.*

Fungi inoculated	No. of Plant inoculated	No. of plant infected	Infection (%)
F. oxysporum f. sp. *psidii*	6	6	100
F. solani	6	6	100
R. solani	6	2	33.33

Table 2. Effect of non-conventional chemicals and fungicides on mycelial growth inhibition causing drying/wilt disease of guava *in vitro*.

Non-conventional chemicals/fungicides	Conc.	Percent Mycelial growth inhibtion				
		F. oxysporum	*F. solani.*	*R. solani*	*P. disseminata*	Mean
Carbendazim	0.1%	85.94	80.02	85.94	85.94	84.46
Topsin-M	0.1%	85.94	84.86	85.94	85.94	85.67
Mancozeb	0.2%	9.09	57.77	65.34	38.93	42.78
Neem oil	800 ppm	49.08	53.56	59.96	38.93	50.38
$MgSO_4 7H_2O$	10^{-4}M	38.93	70.76	85.94	63.60	64.81
$MnSO_4 H_2O$	10^{-4}M	56.25	42.15	67.35	82.33	62.02
$Na_2 B_4 O_7 10H_2O$ (Borax)	10^{-4}M	19.20	27.24	47.45	38.93	33.20
P. longifolia (M.E.)	5%	38.25	38.93	49.03	44.71	42.73
A. marmelos	5%	67.10	67.63	85.94	48.18	67.21
V. rosea	2%	81.15	48.16	85.94	85.94	75.30
T. harzianum (CF)	100%	67.05	19.18	71.80	27.24	46.32
Control	-----	4.05	4.05	4.05	4.05	4.05
Mean	-----	50.17	49.53	66.22	53.73	

	CD 5%
Fungi (F)	3.11
Treatments (T)	5.40
Interaction (F x T)	10.80

F. solani, R. bataticola, Macrophomina phaseoli, Cephalosporium sp. and *Gliocladium roseum and reported to be the cause of the disease* [1, 10, 11].

The present study indicated that all fungicides tested for the inhibition of mycelial growth of fungi under study found highly effective. Joubert and Frean[12] have conducted similar studies using 14 fungicides including carbendazim. They found that all test fungicides controlled the fungus, *F. oxysporum* f. sp. *psidii* causing wilt of guava. Later on, the effectiveness of 10 pesticides including carbendazim, mancozeb and topsin-M in reducing population of *F. oxysporum* f. sp. *psidii, F. solani* and *R. solani* has been observed to be highly effective in sterilized and in un-sterilized soil[13]. These findings corroborate the results of present investigation. Some workers have also tried heavy metals against *F. oxysporum* f. sp. *psidii in vitro*. Dwivedi[14] has reported that mercury, cadmium, copper, cobalt, zinc, iron, calcium and manganese effectively inhibit the mycelial growth of *F. oxysporum* f. sp. *psidii in vitro*. In present investigation, metal salts of $MgSO_4$, $MnSO_4$, and borax were found effective in reducing the mycelial growth of test fungi.

In recent years, many phyto-extracts are being used as fungicide for the control of various plant pathogens *in vitro* and *in vivo*. In the present study, *V. rosea* and *A. marmelos* ME were most effective in inhibiting mycelial growth of guava drying/ wilt pathogens while *P. longifolia* ME was found least effective. It has been reported earlier that phyto-extracts of garlic, soapnut (*Sapindus trifoliata*) and *P. longifolia* were highly fungitoxic *in vitro* against *F. moniliforme* and other fungi [9, 15].

In the modern era, where hazards of pollution are increasing day by day, the biological control will help to reduce it. In present ir vestigation, *T. harzianum* was found antagonistic with all test fungi in inhibition of mycelial growth. Inhibition of fungal growth may be either due to the

production of toxin by the *T. harzianum* or coiling of hyphae against the hypae of *Fusarium spp.* and other fungi. *In vitro* antagonism between guava wilt pathogens and *Trichoderma* spp. have been established earlier and found that *T. lingorum* and *T. viride* inhibited the growth of *F. oxysporum* f. sp. *psidii* and *F. solani* by 70 and 60 per cent, respectively[16] which supported the findings of present study.

In the light of above results, use of ME of *V. rosea, A. marmelos, T. harzianum,* $MgSO_4$ and $MnSO_4$ would be part of our future strategy to combat against pathogenic and associated fungi causing drying/wilt disease of guava, along with both the systemic fungicides (bavistin & topsin-M) and mancozeb recommended and cited in the literature already. However, it is unresolved problem of guava[17].

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