## SHORT COMMUNICATION

# Distribution, pathogenicity and efficacy of some fungicides against Phytophthora parasitica

## MINA D. KOCHE\*, R. M. GADE AND Y. N. MOHOD

Department of Plant Pathology, Shri Shivaji Agriculture College, Amravati 444602, Maharashtra

Received: 22.12.2014 RMS Accepted: 17.06.2015 Published: 27.10.2015

The soil samples collected from citrus rhizosphere of different orchards and nurseries in Vidarbha region were processed in the laboratory and *Phytophthora parasitica* was isolated by leaf bait technique, soil spreading and tissue isolation method on selective medium (PARPH). All collected soil samples were found associated with *Phytophthora* in the range of 15.67 to 36.50 cfu/g of soil. The pathogenicity of *Phytophthora* was tested by inoculating mass multiplied culture of *Phytophthora* into the pots and observed characteristic symptoms of root rot viz., yellowing of leaves followed by drying of seedlings were noticed after 10 days from the inoculating mass culture of *Phytophthora* into pots. Pathogenicity was also tested on detached leaves of *Citrus jambhiri* kept in Petriplates moisture chamber. The pathogen produced water soaked chocolet/ brownish lesions on the leaf lamina within 7 days after inoculation of fungus on host leaves. Efficacy of some fungicides were tested by employing poisoned food technique. metalaxyl @(0.2%), Fosetyl-Al @(0.2%), COC @(0.3%) and B.M @(1.0%) were used and observed that complete inhibition of *Phytophthora parasitica* was recorded by metalaxyl (0.2%), Fosetyl-Al (0.2%) and COC (0.3%). Whereas, Bordeaux mixture (1.0%) was found least effective to inhibit the growth (73.22%).

Key words: Occurrence, pathogenicity, fungicides, Phytophthora parasitica

## INTRODUCTION

Citrus spp. are prone to attack by more than 150 diseases and disorders caused by fungal, viral and few bacterial pathogens right from nursery level to bearing stage resulting in incalculable losses. However a few diseases in any citrus growing area cause significant damage and require due atten-

tion for their effective management.

It would be difficult to locate any orchard in central India and other citrus cultivation belts of India, free from *Phytophthora* diseases. *Phytophthora* propagules have been recorded up to 250-350 cfu/cc soil in highly infested orchards. Every year 5-10% plants die due to severe root rot in bearing orchards (Naqvi, 2000).

Use of fungicides can mitigate the problem up to

<sup>\*</sup>Email: mdkoche@gmail.com

some level but can not eradicate it totally. Copper fungicides are used as foliar spray, drenching of basins and trunk paste to reduce *Phytophthora* diseases. Copper fungicides are effective in controlling foot rot, root rot and gummosis in citrus. Availability of systemic fungicides like metalaxyl and fosetyl-Al has given the citrus growers additional and more effective options for the management of *Phytophthora* diseases.

Soil samples were collected from different locations during June to September, 2008 from the rhizosphere of citrus orchards and nurseries in Vidàrbha region. Soil samples were taken from root zone. The samples were placed in plastic bags to maintain soil moisture, transported to laboratory, and assessed for population count of Phytophthora (Timmer et al., 1988). Soil (10g) from each sample was diluted in 90 ml water having 0.025% agar. One ml aliquot was spread on each of 10 plates of PARPH selective medium (Jefferson et al., 2000). The plates were incubated at 28°C for 2-3 days and no. of colonies of Phytophthora were counted. Soil in the second core was flooded with water, baited with pieces of citrus leaves, and placed in the incubator for 48 hr (Grimm and Alexander, 1973). The leaves were transferred to Petridishes and examined for the presence of papillate sporangia.

Purified culture of *Phytophthora parasitica* was multiplied on large scale on bajara grain. Mass multiplied culture of *Phytophthora* was inoculated at 9:1 proportion into the pots having four months old seedlings of rough lemon. The observations were recorded for mortality of seedlings due to *Phytophthora* root rot. After production of disease symptoms roots along with adhering soil were used for re-isolation to fulfill the Koch's postulates. The roots of infected seedlings along with soil were chaffed in sterilized distilled water and re-isolation was done by following serial dilution method on selective medium of *Phytophthora*.

Pathogenicity also proved by detached leaf method. For that healthy leaves of *Citrus jambhiri* were collected from Department of Plant Pathology. The leaves were washed with sterilized distilled water, then surface sterilization was done by 0.1% mercuric chloride solution and again washed with sterilized distilled water. The leaves were placed in sterilized Petriplate moisture chambers. Then leaves were inoculated with bits of 8 days old *Phytophthora* 

cultures. After inoculation Petriplates were kept at 2 °C in the BOD and the symptoms were observed after 24 hour.

Efficacy of systemic and non systemic fungicides were assessed by poisoned food technique using CMA as a basal medium against test fungus *Phytophthora parasitica*. Fungicides viz., Metalaxyl (0.2%), Fosetyl AI (0.2%), Copper oxychloride (0.3%) and B.M. (1.0%) were tested.

The average colony diameter were recorded and compared with the control treatment after 7 days of inoculation and % growth inhibition was calculated and furnished in Table 2.

A survey of citrus orchards and nurseries was done in Akola, Amravati and Nagpur districts. Almost all the samples, collected from these districts were found positive to Phytophthora. Propagule densities of Phytophthora in soil was in the range of 15.67-23.50 cfu/g soil in Akola 29.67-34.86 cfu/g soil in Amravati district and 29.11-36.50 cfu/g soil in Nagpur district. The leaf baiting technique detected more positive samples as compared to selective medium (Table 1). The isolation frequency indicated that Phytophthora was present in almost all the selected soil samples collected from rhizosphere of citrus orchards and nurseries (Table 1). Present results of association of *Phytophthora* with citrus rhizosphere confirms the findings of Naqvi, 1999; Gade, 2009 and Deokar, 2009 who reported and isolated Phytophthora palmivora, P. parasitica and P. citrophthora from the soil samples collected from citrus growing areas of Nagpur, Wardha and Amravati districts. Present results are helpful to plan management strategy district wise on the basis of inoculum level present in the soil. Characteristic symptoms of root rot viz., yellowing of leaves followed by drying of seedlings were noticed after 10 days after incorporation of mass culture of Phytophthora into pots.

Infected seedlings were collected along with root adhering soil and resorted for isolation by leaf bait method. Re-isolation yielded the same pathogen on selective medium. On the basis of morphological characters, it was confirmed as *Phytophthora parasitica* and proved that *Phytophthora parasitica* is pathogenic in nature. The present results were in agreement with the findings of Maseko and Coutinho (2002), who proved the pathogenicity of *Phytophthora* spp. on commercial citrus root stock,

Table 1: Occurrence of Phytophthora among citrus orchards in Akola, Amravati and Nagpur Districts of Vidarbha region

Loca	ations	No. of	es	Selective Med No of positive samples	ia Propagules/g soil		ole Positive in la ng technique	eaf
Dist	rict Akola							
Ako	a	06		04	15.67		05	
Ako		03		03	20.50		02	
Ako	i Jahagir	08		06	23.50		07	
	rict Amravati							
Mor	shi	03		02	29.67		03	
War	ud	03		02	34.86	Δ'	02	
Amr	avati	04		03	32.83	4	04	
Dist	rict Nagpur							
	S Katol	- 04		02	34.33		03	
Moh	pa	04		04	36.50		04	
	andri	04		02	30.33		03	
	irgaon	03		02	29.67		02	
Nag		02		02	29.11		02	

Table 2: Efficacy of different fungicides against Phytophthora parasitica. (Poisoned food technique)

Fungicides	Concentration (%)	Radial mycelial growth (mm)	% Growth inhibition
Metalaxyl	0.2	00	100
Fosetyl-Al	0.2	00	100
Copper oxychloride (COC)	0.3	00	100
Bordeaux mixture	1.0	24.10	73.22
Control		90.00	
'F' Test		Sig	
S.E. (m)±		0.55	
CD (P=0.01)		1.90	

rough lemon and troyer citrange. Gade *et al.* 2005 observed the incidence of root rot, gummosis on 35 mandarin varieties under test. Pathogenic nature of *Phytophthora* spp. in citrus was also reported by Graham and Timmer (2003).

The fungus *Phytophthora parasitica* was tested for its pathogenicity on detached leaves of *Citrus jambhiri* under moist chamber. Cent percent infection on leaf was observed within 7 days after inoculation of fungus on host leaves. The pathogen produced water soaked chocolate/ brownish lesions on the leaf lamina Kumbhare and Moghe (1976).

Table 2 indicates that, all the treatments show significant per cent growth reduction from 73.22 to 100 per cent of the test pathogen. Complete inhibition of *Phytophthora parasitica* was recorded in metalaxyl (0.2%), fosetyl-Al (0.2%) and COC (0.3%). However Bordeaux mixture (1.0%) was found least effective as compared to other fungicides to inhibit the growth of *P. parasitica* (73.22%).

Gade et al. (2008) observed complete inhibition of *P. parasitica* in metalaxyl at recommended dose.

Previous research revealed that mycelial growth of *P. nicotianae* on corn meal agar was more sensitive to fosetyl-Al and metalaxyl (Matheron and Porchas, 2000). Peshney *et al.*, (1990) observed that metalaxyl (0.1%) and copper-oxychloride (0.25%) were highly effective in controlling mycelial growth and sporangial production of *Phytophthora*.

#### REFERENCES

Deokar, Y.N., 2009. Efficacy of Pseudomonas fluorescens and Bacillus subtilis against Phytophthora spp. In citrus. M.Sc. Thesis (Unpub.) Dr. P.D.K.V. Akola (M.S.).

Gade, R.M., 2009. Biological and chemical management of Phytophthora root rot/collar rot in citrus nursery. AFPP-9<sup>th</sup> International conference on Plant Diseases Tours, France.

Gade, R.M., G.K. Giri and U.R. Sangle, 2005. Management of Phytophthora and root rot in citrus nursery. Pesticide Res. J. 17: 28-29.

Gade, R.M., V.V. Deshmukh, R. S. Ghawade and Mina Koche, 2008. Efficacy of different chemicals against Phytophthora, Pythium and Furarium solani. National Symposium on Citriculture: Emerging trends.pp.147.

Graham J.H. and A.Z. Timmer, 2003. *Phytophthora* disease of citrus. Plant disease of international importance. *Edis.ifas.ofl.edu*. **5**: 13-14.

Grimm, G.R. and Alexander, 1973. Citrus leaf pieces as traps for Phytophthora parasitica from soil slurries. Phytopathology. 63 : 540-541.

- Jefferson L. da S. Costa; John A. Menge; Willian L. Casale, 2000. Biological control of *Phytophthora* root rot of avocado with microorganisms grown in organic mulches. *Brazilian J. of Microbiol.* 31: 239-246.
- Kumbhare, G.B. and P.G. Moghe, 1976. Leaf fall disease of Nagpur oranges caused by *Phytophthora nicotianae* var. *parasitica* water house. *Curr. Sci.* **45**:561-562.
- Maseko, B. and T. Coutinho, 2002. Pathogenicity of *Phytophthora* and *Pythium* species associated with citrus root rot in South Africa. South African *J. of Bot.* **68**: 327-337.
- Matheron, M.E., and M. Porchas, 2000. Impact of azoxystrobin,
- dimethomorph, fluazinam, fosetyl-Al and metalaxyl on growth, sporulation and zoospore cyst germination of three *Phytophthora* spp. *Plant Dis.* **84**:454-458.
- Naqvi, S.A.M.H. 2000. Managing *Phytophthora* disease of citrus. *Indian Horticulture*, **44:** 5-9.
- Peshney, N.L., Moghe and P.D.Wangikar, 1990. In vitro evaluation of some fungicides against. Phytophthora nicotionae var. parasitica water house, PKV. Res. J. 14: 133-137.
- Timmer, L.W., Graham, J.H., Sandler, H.A. and S.E. Zitko, 1988. Population of *Phytophthora parasitica* in citrus orchards and tree response to fungicide applications. *Citrus Ind.* 69:40-54.