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In vitro efficacy of different botanicals, bioagents, chemicals against *Xanthomonas axonopodis* pv.*citri* by turbidiometrical method

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ABSTRACT

Citrus canker disease of acid lime caused by *Xanthomonas axonopodis* pv. *citri* is an important disease in many parts of MH region .The bacterium infects the twigs, petioles, fruit stalks and fruits. Action of botanicals,bioagents,chemicals against *Xac* was studied by turbidiometrically at 24hrs.intervals upto 96hrs by spectrophotometer at 620nm.At 96 hours of incubation least bacterial growth(0.232OD) was exhibited in copper-oxychloride + streptomycin sulphate (0.2 % + 200ppm) followed with copper-oxychloride + streptomycin sulphate (0.2% + 100ppm) 0.266 OD statistically superior over all treatments. Similar findings are observed after 24h growth in copper oxychloride + streptomycin sulphate (0.2 % + 200ppm) followed with copper-oxychloride +streptomycin sulphate (0.2 % + 100ppm) (0.303, 0.306, respectively. In botanicals and bioagents neem seed kernel extract (5%) was effective in reducing the growth of bacteria with 0.446 OD at 96 h followed by *Pseudomonas flourescence* $1x10^8$ cell and *Bacillus subtilis* $1x10^8$ cell with 0.506 and 0.486 OD, respectively.

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INTRODUCTION

Citrus is an important value as fruit crop. Present day citrus is delectable, juicy, and seedless is of great nutritional significance as well (Khan *et al.*, 1992). Additionally, it possesses enormous therapeutic qualities (Chaudhry *et al.*, 1992). Citrus is a member of Rutaceae family and grown in varying densities in countries with tropical or subtropical climates. Citrus plant is attacked by a number of diseases like citrus canker, gummosis, citrus decline, citrus tristeza virus, greening, etc. Citrus canker is caused by "*Xanthomonas campestris* pv. *citri*" that is probably the worst enemy to citrus plants (Sahi *et al*., 2007). Citrus canker disease is of regular occurrence on several citrus cultivars in varying degrees of incidence depending on the climatic conditions. The bacterium, *Xanthomonas* causes different symptoms ranging from pustules to necrotic lesions consisting of erumpent corky tissue surrounded by water soaked tissues and yellow

halo on leaves, stems and fruits (Zekri *et al.*, 2005; Graham *et al.*, 2004; Das, 2003). *Xanthomonas axonopodis* pv. *citri* is a rod shaped, gram negative bacterium with single polar flagellum.

MATERIAL AND METHODS

The citrus canker diseased sample were collected from Akola district of Maharashtra state. The isolate of pathogen were obtained from infected leaves,twigs of acid lime showing typical symptoms of citrus canker. These isolate were isolated by tissue isolation method. A bacterial suspension of each specimen was then cultured on NA medium. Following incubation, colonies similar to *Xanthomonas* were maintained on NA medium at room temperature by adopting subsequent sub culturing at periodical, regular intervals. Three days old cultures were used for further studies.

Identification of the pathogen :

The identification of the pathogen involved in causing of citrus canker in acid lime was determined by conducting studies on its morphological, biochemical, cultural and physiological features of the pathogen as per standard microbiological procedures.

Efficacy of botanicals,bioagents,chemicals against *Xac* was studied by turbidiometrically at 24hrs. intervals upto 96hrs by spectrophotometer at 620nm.

RESULTS AND DISCUSSION

Efficacy of chemicals, botanicals and bioagents was studied turbidiometrically. The results are given in Table 1 and Fig 1. At 96 hours of incubation least bacterial growth (0.232OD) was exhibited in copper oxychloride + streptomycin sulphate (0.2 % + 200ppm) followed with copper oxychloride +streptomycin sulphate (0.2 % + 100ppm) 0.266 OD statistically superior over all treatments. Similar findings are observed after 24h growth in T_6 and T_5 (0.303, 0.306OD), respectively. In botanicals and bioagent neem seed kernel extract (5%) was effective in reducing the growth of bacteria with

Table 1 : Efficacy of chemicals, botanicals and bioagents against *Xanthomonas axonopodis* pv. *citri* by Turbidometric method at 620nm

Sr. No.	Treatments	Conc.	24 hrs	96Hrs
T_1	Streptomycin sulphate	100 ppm	0.383	0.293
T_2	Streptomycin sulphate	200 ppm	0.373	0.286
T_3	Copper oxychloride	0.2%	0.364	0.273
T_4	Copper oxychloride	0.3%	0.353	0.270
T_5	COC+Streptomycin sulphate	0.2%+100 ppm	0.306	0.266
T_6	COC+Streptomycin sulphate	0.2%+200 ppm	0.303	0.232
T_7	Kasugamycin	100 ppm	0.381	0.286
T_8	Kasugamycin	200 ppm	0.366	0.274
T_9	Agrimycin	100 ppm	0.373	0.293
T_{10}	Agrimycin	200 ppm	0.376	0.283
T_{11}	COC + Agrimycin	0.2%+100 ppm	0.351	0.292
T_{12}	COC + Agrimycin	0.2%+200 ppm	0.343	0.283
T_{13}	COC+ Kasugamycin	0.2%+100 ppm	0.323	0.276
T_{14}	COC+Kasugamycin	0.2%+200 ppm	0.344	0.276
T_{15}	Neem extract	1%	0.561	0.393
T_{16}	Neem extract	2.5%	0.576	0.403
T_{17}	Neem extract	5%	0.596	0.446
T_{18}	*Pseudomonas fluorescens*	$1x10^8$ cfu /ml	0.615	0.506
T_{19}	*Bacillus subtilis*	$1x10^8$ cfu /ml	0.636	0.486
T_{20}	Control		0.756	0.951
	Blank		0.260	0.260
	'F' test		Sig.	Sig.
	S.E. (m) $\pm$		0.002	0.001
	C.D. (P = 0.01)		0.007	0.003

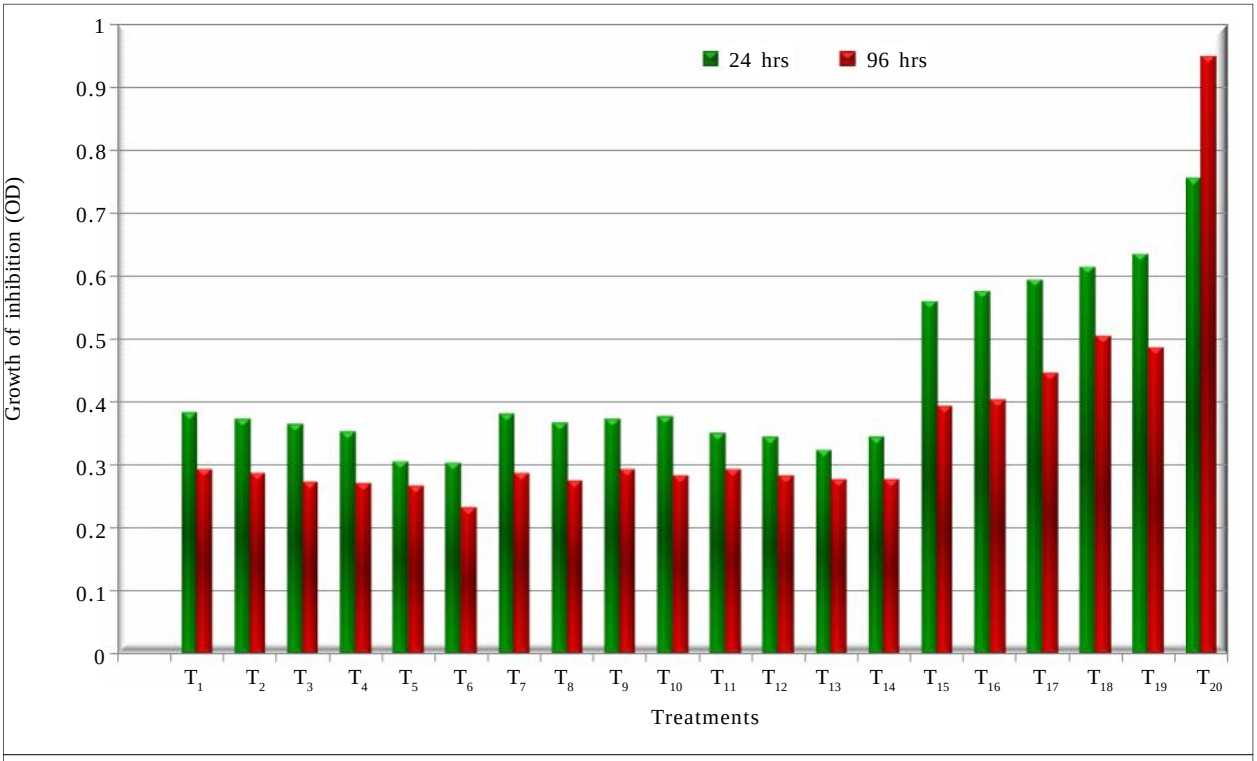

Fig. 1 : Efficacy of chemicals, botanicals and bioagents against *Xanthomonas axonopodis* pv. *citri* by turbidometric method at 620nm

0.446 OD at 96 h followed by *Pseudomonas flourescence* 1x10^8 cell and *Bacillus subtilis* 1x10^{8}cell with 0.506 and 0.486 OD, respectively. The present findings are in agreement with Sharma *et al*. (1980). *In vitro* evaluation of eight chemicals against *Xanthomonas vesicatoria* by paper disc and turbidometric method and described that, combination of streptocycline and copper sulphate was most effective in inhibiting the growth of pathogenic organism in turbidometric method. Das (2005) who assessed different of chemicals, antagonist and botanicals *in vitro* against *Xanthomonas axonopodis* pv. *citri* by measuring the growth turbidometrically at 620nm and reported maximum growth of inhibition after 96 hrs of incubation in COC (0.3%) + streptomycin sulphate (100ppm) *i.e*. 0.230 OD.

Conclusion :

Lower OD were found in chemical treatment *i.e.* copper oxychloride 0.2% + streptomycin sulphate 200ppm followed by concentration COC (0.2%)+Streptomycin sulphate (100ppm) by turbidometrical method *i.e.* 0.232 OD and 0.266 OD, respectively.

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