Research Paper :

In vitro sensitvity of plant protection chemicals and fertilizers to *Ralstonia solanacearum*, the causal agent of bacterial wilt in ginger

RESHMY VIJAYARAGHAVAN AND KOSHY ABRAHAM

International Journal of Plant Protection, Vol. 4 No. 1 (April, 2011) : 99-102

See end of the article for authors' affiliations

Correspondence to :
RESHMY VIJAYARAGHAVAN
Krishi Vigyan Kendra, Ambalavayal, WAYANAD, (KERALA) INDIA
Email:drreshmydhanesh@gmail.com

SUMMARY

***In vitro* inhibitory effect of antibiotics, fungicides, insecticides and fertilizers on growth of *R. solanacearum* was tested. All the antibiotics except Cephalaxin were inhibitory to the pathogen. Copper fungicides tested inhibited the pathogen. The higher two concentrations of Fytolan and 0.2 per cent of Kocide showed more inhibition of the pathogen. All the insecticides tested were not inhibitory to the pathogen. Among fertilizers, Factomphos exhibited maximum inhibition of the pathogen followed by Rajphos, urea and MoP.**

Vijayaraghavan, Reshmy and Abraham, Koshy (2011). *In vitro* sensitvity of plant protection chemicals and fertilizers to *Ralstonia solanacearum*, the causal agent of bacterial wilt in ginger. *Internat. J. Pl. Protec.*, **4**(1): 99-102.

Key words : *Ralstonia solanacearam*, Fungicides, Insecticides, Antibiotics, *In vitro* sensitivity

Received : ***October, 2010***
Accepted : ***December, 2010***

India, the world's largest producer, consumer and exporter of ginger, has a predominant position in the global market and accounts for 50 per cent of world's total production. One of the important factors which limit the production of ginger is the occurrence of diseases. Bacterial wilt, incited by *Ralstonia solanacearum* Yabuuchi (Smith), is at present considered as a very serious threat in most of the ginger growing areas. The pathogen infects over 450 plant species including many economically important crops other than ginger. Because of its destructive potential to various crops, the control of *R. solanacearum* has attracted much research attention (Hayward, 1994). As ginger is a remunerative crop, farmers are adopting all available methods to increase the productivity of the crop where plant protection measures play an important role. Even after adapting to various cultural and chemical means, bacterial wilt of ginger continues to be a complex disease to manage. However, every effort has to be made to reduce the losses due to the disease by adopting suitable management strategy. Hence, a study was conducted to find out the *in vitro* inhibitory effect of antibiotics, fungicides, insecticides and fertilizers on the growth of *R. solanacearum*.

MATERIALS AND METHODS

The *in vitro* compatibility of *R. solanacearum* to antibiotics, fungicides, insecticides and fertilizers commonly used in ginger plots were studied by Filter paper disc method in the year 2007 at College of Horticulture, Vellanikkara, KAU, Thrissur.

Antibiotic sensitivity with bacterial isolates was assessed with Hi Media antibiotic discs. Three replications were maintained and observations on the inhibition zone around the discs were taken after 48h. In the case of fungicides, insecticides and fertilizers, in order to get a desired concentration, the required quantity was added to 100 ml sterile water and autoclaved filter paper discs of 4mm diameter were soaked in these various concentrations of plant protection chemicals / fertilizers for a period of 30 min. Fertilizers were exposed to UV light for a period of 45 min. to lessen the

contamination. Three discs of antibiotic, fungicide, insecticide and fertilizer were placed on sterile King's B (KB) medium seeded with the pathogen, *R. solanacearum*. Control consisted of KB medium alone inoculated with the antagonist. Three replications were maintained for each isolate. The inoculated Petri dishes were incubated at room temperature and the observations on inhibition zone around the discs were taken after 48h.

RESULTS AND DISCUSSION

Data on the *in vitro* sensitivity of *R. solanacearum* to the effect of other antibiotics, fungicides, insecticides and fertilizers to the pathogen are presented in Table 1, 2 and 3.

Antibiotics :

Observations (Table 1) on the *in vitro* sensitivity of *R. solanacearum* to 11 different antibiotics showed that except for Cephalaxin, all the antibiotics at different concentrations inhibited the pathogen. It was noticed that the higher concentration of the antibiotic exerted maximum inhibition. Highest concentration of Streptocycline, Gentamicin, Kanamycin and Tetracycline exhibited maximum inhibition. The inhibitory effect of various antibiotics was studied by various workers. Rani (1994) observed the inhibitory effect of Ambistryn-S and Chloromycetin against *R.solanacearum* of ginger under *in vitro* conditions. She also observed that plants treated with Ambistryn-S, Terramycin and Chloromycetin showed minimum wilt incidence. Similarly, pre-planting and pre-storage treatment in Streptocycline (200 ppm) along with Dithane M-45 (0.25%) and Bavistin (0.1%) delayed the disease development. The effectiveness of Streptomycin and Streptopenicillin over other antibiotics against wilt pathogen of ginger both under *in vitro* and *in vivo* conditions has been well elucidated by Singh *et al*. (2000) whereas Penicillin G, tetracycline and plantomycin did not inhibit the pathogen at any of the concentrations tested. According to Sambasivam (2003), various isolates of *R.solanacearum* infecting ginger were resistant to Ampicillin and Rifampicin but sensitive to Chloramphenicol and Kanamycin. However, Carbenicillin, Nalidixic acid, streptomycin sulphate and Tetracycline showed a varied response with the isolates. Similarly, several antibiotics like Oxytertacycline, Tetracycline, Penicillin G and Streptomycin inhibited *R.solanacearum* (Goorani *et al*., 1978). He *et al.* (1983) reported that all the strains of *P. solanacearum* were resistant to Penicillin, Viomycin and Chloramphenicol. Gunawan (1989) observed the suppression of *R. solanacearum* with Streptomycin sulphate. Likewise, Akbar (2002) reported that Ampicillin, Streptomycin sulphate and Kanamycin inhibited the pathogen whereas Rifampicin, Chloramphenicol and Oxytertacycline failed to restrict the growth of the pathogen. Kumar and Sarma (2004) noticed that all the ginger isolates tested were resistant to Tetracycline, Polymixin B sulphate and Chloramphenicol.

Table 1 : *In vitro* sensitivity of *R. solanacearum* to antibioties

Sr. No.	Antibiotics	Concentration (ppm)	*Inhibition zone (mm)
1.	Chloramphenicol	0.1	17.67^{h} (4.26)
		0.25	20.33^{f} (4.57)
		0.50	23.0^{e} (4.85)
2.	Gentamicin	0.1	24.0^{de} (4.95)
		0.3	27.67^{c} (5.31)
		0.5	30.33^{b} (5.55)
3.	Rifampicin	0.05	23.33^{e} (4.88)
		0.15	24.67^{d} (5.02)
		0.3	27.67^{c} (5.31)
4.	Kanamycin	0.01	12.33^{i} (3.58)
		0.05	24.67^{d} (5.02)
		0.3	30.33^{b} (5.55)
5.	Ampicillin	0.1	18.67^{g} (4.38)
		0.25	23.67^{de} (4.92)
6.	Streptomycin	0.1	23.67^{de} (4.92)
	sulphate	0.25	26.67^{c} (5.21)
7.	Tetracycline	0.05	30.33^{d} (5.55)
		0.3	31.67^{b} (5.67)
8.	Penicillin G	0.1	24.67^{d} (5.02)
9.	Nalidixic acid	0.3	20.33^{f} (4.57)
10.	Oxytetracycline	0.3	12.33^{i} (3.58)
11.	Cephalaxin	0.3	0^{j} (0.71)

* Mean of three replications. In each column figures followed by same letters donot differ significantly according to DMRT. Figures in paranthesis are √x+0.5 transformed values

The different concentrations of Streptocycline restricted the growth of the pathogen, exhibiting an inhibition zone (IZ) of above 25 mm with the maximum at 400 ppm concentration. The inhibitory effects of Streptomycin and streptocycline on *Pseudomonas solanacearum* has been observed by Shivappashetty and Rangaswami (1971). Paul (1998) observed that 250 and 500ppm concentration of Ambistryn, Oxytetracycline and Streptocycline inhibited *R.solanacearum* under *in vitro* conditions. The crop sprayed with Streptocycline solution (100 ppm) at regular intervals of 15 days also reduced the incidence of bacterial wilt in ginger (Dohroo, 2001). Likewise, seed rhizomes treated with Streptocycline 200 ppm for 30 min. and shade dried before planting was

found effective in reducing the wilt incidence (Anandaraj *et al*., 2005).

Fungicides :

Amongst the eight fungicides tested, only the copper fungicides *viz.,* Fytolan, Kocide and Shield in that order showed inhibitory property towards *R. solanacearum* (Table 2). Higher two concentrations of Fytolan and 0.2 per cent concentration of Kocide exhibited more inhibition of the pathogen. The lowest concentration of Shield did not show any inhibitory effect. The inhibitory effect of Bordeaux mixture towards the pathogen has already been reported (Rani, 1994). Similarly, the effectiveness of Bordeaux mixture, Copper oxychloride, Thiride, Blue copper, Kocide, Nabem, Maneb, Dithane M-45, Captan, and Thiram against *R. solanacearum* has also been noticed by various workers (Jyothi, 1992; Akbar, 2002; Anandaraj *et al*., 2005).

Table 2 : *In vitro* sensitivity of *R.solanacearum* to fungicides

Sr. No.	Fungicide	Concentration (%)	*Inhibition zone (mm)
1.	Fytolan	0.2	15.5^{c}
		0.3	18.5^{b}
		0.4	21.0^{a}
2.	Kocide	0.1	8.5^{f}
		0.15	10.0^{e}
		0.2	18.5^{b}
3.	Shield	0.5	0.0^{g}
		1.0	10.0^{e}
		1.5	13.5^{d}

* Mean of three replications. In each column figures followed by same letters do not differ significantly according to DMRT

Insecticides :

A total of eight insecticides were evaluated against the pathogen as well and it was observed that all the insecticides tested were not inhibitory to the growth of *Ralstonia solanacearum.* Not much literature is available in this line.

Fertilizers :

Contradictory to the result of effect of insecticides towards the pathogen, it was noticed that in general, all the fertilizers tested inhibited the pathogen (Table 3). The highest concentration of Factomphos exhibited maximum inhibition followed by that of Rajphos, Urea and MoP. Thus, results of the study clearly indicated that chemical fertilizer application has an indirect effect in checking the growth and multiplication of the pathogen.

Table 3 : *In vitro* sensitivity of *R.solanacearum* to fertilizers

Sr. No.	Fertilizer	Concentration (%)	*Inhibition zone (mm)
1.	Urea	1.0	12.00^{ef}
		1.5	14.67^{d}
		2.0	20.33^{bc}
2.	Factomphos	2.0	14.67^{d}
		2.5	19.00^{c}
		3.0	23.67^{a}
3.	MoP	2.0	11.33^{f}
		2.5	13.67^{de}
		3.0	18.67^{c}
4.	Rajphos	2.0	11.33^{f}
		2.5	14.83^{d}
		3.0	21.00^{b}
5.	DAP	2.0	5.67^{g}
		2.5	11.33^{f}
		3.0	13.67^{de}

*Mean of three replications.
In each column figures followed by same letters do not differ significantly according to DMRT.
MoP : Muriate of potash, DAP: diammonium phosphate

The usefulness of application of chemical fertilizers and soil ameliorants in checking the activity of the pathogen in ginger and other crops has also been documented. Vudhivanich (2002) noticed a decrease in the population of bacterial wilt pathogen of ginger in the soil amended with urea and calcium oxide and the decrease was attributed to toxicity of ammonium, ammonia and nitrate degraded from urea. Calcium oxide is added to convert the ammoniacal form into nitrite (NO_2) which is highly toxic to the growth of the pathogen. According to Hepperly *et al.* (2004), addition of gypsum along with triple super phosphate at the time of planting ginger rhizomes helped in production of bacterial wilt free ginger rhizomes. Devi (1978) observed that sawdust along with urea combined with Agrimycin (200 ppm) showed reduction in wilt incidence of tomato. Moreover, application of urea and lime minimized the wilt incidence in solanaceous and cucurbitaceous vegetables. Combination of Actigard (acibenzola-S-methyl) with the S-H mixture (bagasse, rice husk, oyster shell powder, urea, KNO_3, calcium super phosphate and mineral ash) significantly reduced bacterial wilt of tomato caused by *R. solanacearum* (Anith *et al*., 2004).

Authors' affiliations:
KOSHY ABRAHAM, Department of Plant Pathology, College of Horticulture, Kerala Agricultural University, Vellanikkara, THRISSUR (KERALA) INDIA

REFERENCES

Akbar, K.I. (2002). Integrated management of bacterial wilt of tomato caused by *Ralstonia solanacearum* (Smith) Yabuuchi *et al.,* M.Sc (Ag.) Thesis, Kerala Agricultural University, Thrissur, Kerala (India).

Anandaraj, M., Devasahayam, S., Zacharaiah, J.T., Eapen, J.S., Sasikumar, B., Sreenivasan, V. and Thankamani, C.K. (2005). *Ginger*. (eds. Rajeev, P. and Kandiannan, K.)., Kochi, Extension pamphlet, 8p.

Anith, K.N., Momol, M.T., Kloepper, J.W., Marois, J.J., Olson, S. M. and Jones, J.B. (2004). Efficacy of plant growth-promoting rhizobacteria, acibenzolar-S-methyl, and soil amendment for integrated management of bacterial wilt on tomato. *Pl. Dis.,* **88**: 669-673.

Devi, L.R. (1978). Bacterial wilt of tomato in Kerala - Host range and survival of the pathogen and control. Ph.D. Thesis, Kerala Agricultural University, Thrissur, Kerala (India), 163p.

Dohroo, N.P. (2001). Fast spreading disease of ginger, *Agriculture Tribune*, Monday, January 1, 2001, Chandigarh, India, 4pp.

Goorani, M.A., Abo-El-Dahab, M.K. and Wagin, E.E. (1978). Test *in vitro* and in pots with certain chemicals for inhibition of *Pseudomonas solanacearum*. *Zentralbl. Bacteriol. Perasitenkd. Infektions kr. Hyg.,* **133** : 235-239.

Gunawan, O.S. (1989). The effect of concentration of Streptomycin sulphate antibiotic on multiplication of *Pseudomonas solanacearum* E.F. Smith. *Bull. Penilitian Hortikultura,* **17**: 110-111.

Hayward, A.C. (1994). The host of *Pseudomonas solanacearum*. In : *Bacterial wilt : The disease and its causative agent, Pseudomonas solanacearum* (ed. By Hayward, A.C. and Hartman, G.L.), CAB International, Wallingford, UK, pp.9-24.

He, L.Y., Sequeira, L. and Kelman, A. (1983). Characteristics of strains of *Pseudomonas solanacearum* from China. *Pl. Dis*., **67**: 1357-1361

Hepperly, P., Zee, F., Kai, R, Arakawa, C., Meisner, M., Kratky, B., Hamamoto, K. and Sato, D. (2004). Producing bacterial wilt free ginger in green house culture, Soil and Crop Management, Cooperative Extension Service, College of Tropical Agriculture and Human Resources, University of Hawaii at Manoa, p.6.

Jyothi, A.R. (1992). Characterization and management of bacterial wilt of chillies caused by *Pseudomonas solanacearum* E.F. Smith, M.Sc. (Ag.) Thesis, Kerala Agricultural University, Thrissur, Kerala (India).

Kumar, A. and Sarma, Y.R. (2004). Characterization of *Ralstonia solanacearum* causing bacterial wilt in ginger. *Indian Phytopath.,* **57**: 12-17

Paul, S.T. (1998). Biochemical and biological basis of resistance in solanaceous vegetables against bacterial wilt of *Ralstonia solanacearum* (Smith) Yabuuchi *et al.,* Ph.D. Thesis, Kerala Agricultural University, Thrissur, Kerala (India).

Rani, G.L. (1994). Management of bacterial wilt of ginger (*Zingiber officinale* Rosc.) incited by *Pseudomonas solanacearum* (Smith) Smith, M.Sc. (Ag.) Thesis, Kerala Agricultural University, Vellanikkara, Thrissur, Kerala (India).

Sambasivam, P.K. (2003). Characterization of *Ralstonia solanacearum* (Smith) Yabuuchi *et al.*, causing bacterial wilt in ginger using molecular markers. M.Sc. (Ag.) Thesis, Kerala Agricultural University, Thrissur, Kerala (India).

Shivappashetty, K.S. and Rangaswami, G. (1971). *In vitro* and *in vivo* activities of streptocycline on bacterial blight of rice caused by *Xanthomonas oryzae*. *Indian Phytopath.,* **24**: 145-152.

Singh, P.P., Singh, V.B., Singh, H.P. and Rajan, S. (2000). Genetic diversity in ginger (*Zingiber officinale* Rosc.) with reference to essential oil content. *J. Spices & Aromatic Crops,* **9**: 161-164

Vudhivanich, S. (2002). Effect of soil amendment with urea and calcium oxide on survival of *Ralstonia solanacearum* , the causal agent of bacterial wilt or rhizome rot of ginger, *Kasestart J. Natural Sci.*, **36** : 242-247.

****