

INHIBITORY EFFECT OF PLANT EXTRACTS AND PLANT OILS ON *XANTHOMONAS ORYZAE* PV *ORYZAE*, THE BACTERIAL BLIGHT PATHOGEN OF RICE

RAJI P.¹, SUMIYA K. V.², DHANYA S.³, REMYA K.⁴ & NARAYANANKUTTY M. C.⁵

¹Associate Professor (Plant Pathology), Regional Agricultural Research Station, Pattambi, Kerala, India
²Assistant Professor (Plant Pathology), Krishi Vigyan Kendra, Pattambi, Kerala, India
^{3,4}Project Fellow, Regional Agricultural Research Station, Pattambi, Kerala, India

⁵Professor (Horticulture), Associate Director of Research, Regional Agricultural Research Station, Pattambi, Kerala, India

ABSTRACT

Bacterial blight caused by *Xanthomanas oryzae* PV *oryzae* is a major disease of rice causing yield losses in all the major rice growing countries. The disease remains as one of the major production constraints in India also. The present study was conducted to evaluate plant oils and plant extracts against bacterial blight pathogen *Xanthomonas oryzae* PV *oryzae*. Five plant oils and twenty five plant extracts were tested *in vitro*. Extract of garlic bulb (*Allium sativum*) recorded highest zone of inhibition, followed by tamarind fruit (*Tamarindus indica*), gooseberry fruit (*Phyllanthus emblica*), green mango (*Mangifera indica*) and lemon juice (*Citrus aurantifolia*). Among the plant oils tested, five per cent and one percent concentrations of palmarosa oil (*Cymbopogon martinii*) exhibited highest inhibition of the pathogen followed by lemongrass oil (*Cymbopogon flexuous*), cinnamon oil (*Cinnamomum zeylanicum*) and vetiver oil (*Chrysopogon zizanioides*). These inhibitory plant oils and plant extracts can be tested in the field and can be utilized for developing botanical formulations for the management of bacterial blight of rice.

KEYWORDS: Bacterial Blight, Plant Extracts, Plant Oils, Rice, Xanthomonas oryzae PV oryzae

INTRODUCTION

Bacterial blight of rice caused by *Xanthomonas oryzae* PV *oryzae* is a serious disease of rice in majority of the rice growing countries of the world. The disease is causing yield loss of 20 - 30 per cent annually in Asia and Africa (Jena, 2013). In India, bacterial blight is a major production constraint in irrigated and low land ecosystems. The disease is causing yield losses in major rice growing states of the country. Generally the stage between maximum tillering to booting is highly susceptible to the disease. Kresek and leaf blight are the two phases of infection. The leaf blight symptom is commonly seen in Kerala, however in recent years there is an increase in appearance of kresek phase also. Almost all the rice varieties cultivated in Kerala are susceptible to the disease. The chemical control using antibiotics are not giving satisfactory result. High rainfall and humid conditions prevailing in the state and the susceptibility of cultivated varieties has resulted in epidemics of BLB in major rice growing tracts of Palakkad and Aleppey districts of the state. Considering the environmental and health hazards of the chemicals particularly the antibiotics and the problem of development of resistance in the pathogen, the plant derived products are viable alternatives for disease management. The antimicrobial properties of plant oils have been reported by several researchers (Pattnaik *et al.*, 1996; Bansod & Rai, 2008; El- Barotty *et al.*, 2010; Amini *et al.*, 2012). The efficacy of plant extract for the management of plant diseases were reported

(Nguefack *et al.*, 2013; Sehajpal *et al.*, 2009; Jabeen *et al.*, 2009). The present study was conducted to evaluate the antimicrobial activity of plant extracts and plant oils against *Xanthomonas oryzae* PV *oryzae*.

MATERIALS AND METHODS

In Vitro Evaluation of Plant Extracts against Xanthomonas Oryzae PV Oryzae

Twenty five plant extracts were tested *in vitro* against *Xanthomonas oryzae* PV *oryzae*. 10 per cent water extracts of the plant materials were prepared and filter sterilized. PSA medium mixed with *Xanthomonas oryzae* PV *oryzae* culture suspension was plated in petriplates. 10 mm sized sterilized filter paper discs were dipped in filter sterilized plant extracts and placed at the centre of the bacteria inoculated medium plated. Plates were incubated at room temperature. The plant extracts tested were rhizome extract of ginger (*Zingiber officinale*), and turmeric (*Curcuma longa*), bulb extract of garlic (*Allium sativum*) and onion (*Allium cepa*), leaf extracts of *Aloe vera*, Lantana (*Lantana camara*), tamarind (*Tamarindus indica*), neem (*Azadiracta indica*), thulasi (*Ocimum sanctum*), curry leaf (*Murraya koenigii*), papaya (*Carica papayas*), eupatorium (*Chromolaena odorata*), murikoodi (*Hemigraphis colorata*), panikoorka (*Plectranthus amboinicus*), cherula (*Aerva lanata*) and thippali (*Piper longum*) fruit extracts of green fruit of tamarind (*Tamarindus indica*), gooseberry (*Phyllanthus emblica*), lemon (*Citrus aurantifolia*), tomato (*Solanum lycopersicum*) and dry powder extract of pepper (*Piper nigrum*) and asafoetida (*Ferula assafoetida*). Sterilized PSA medium was melted and bacterial culture suspension was mixed with it and plated. The filter paper discs dipped in the plant extracts were placed at the centre of the medium and plates were incubated at room temperature. Three replications of each treatment were maintained. Observation on zone of inhibition was measured on 3rd day. Measurements were square root transformed. The analysis of variance was performed and means were separated by Fischer's LSD test.

In Vitro Evaluation of Plant Oils against Xanthomonas Oryzae PV Oryzae

An *in vitro* study was carried out to test the efficacy of five plant oils *viz.*, Lemongrass oil (*Cymbopogon flexuous*), Palmarosa oil (*Cymbopogon martinii*), Vetiver oil (*Chrysopogon zizanioides*), Cinnamon oil (*Cinnamomum zeylanicum*), and Maroti oil (*Hydnocarpus pentadra*) against *Xanthomonas oryzae* PV *oryzae* by filter paper disc method. Oils were emulsified with 0.05 per cent tween 80 in sterilized distilled water to get 1% and 5% concentrations. Sterilized filter paper discs of 10 mm size were dipped in this. The control sets were prepared by sterilized distilled water instead oils. 48 hours old culture of the pathogen was suspended in sterile water and mixed with melted PSA medium and plated. The filter paper discs dipped in oils were placed at the centre of the petriplate and incubated at room temperature. Three replications were maintained for each treatment. Observations on diameter of the inhibition zone were measured on 3rd day. Measurements were square root transformed. The analysis of variance was performed and means were separated by Fischer's LSD test.

RESULTS AND DISCUSSIONS

The effect of plant extracts on *Xanthomonas oryzae* PV *oryzae is* given in table 1. Among the 25 plant extracts tested against *Xanthomonas oryzae* PV *oryzae*, extract of garlic bulb (*Allium sativum*) recorded highest zone of inhibition (3.1 cm), followed by tamarind fruit (*Tamarindus indica*) (1.96 cm), gooseberry fruit (*Phyllanthus emblica*) (1.62 cm), green mango (*Mangifera indica*) (1.55 cm) and lemon juice (*Citrus aurantifolia*), (1.57 cm). The efficacy of plant extracts against rice diseases have been reported by various researchers (Kamalakannan *et al.*, 2001 and Biswas, 2007). The

Inhibitory Effect of Plant Extracts and Plant Oils on *Xanthomonas Oryzae* PV *Oryzae*, The Blight Pathogen of Rice

inhibitory effect of *Allium sativum* against *Rhizoctonia solani*, the sheath blight pathogen of rice was reported by Sehajpal *et al.* (2009).

Among the plant oils tested, palmarosa oil recorded highest zone of inhibition of 2.45cm and 2.02 cm in 5 per cent and 1 per cent concentrations respectively (Table 2). The palmarosa oil (5%) was equally effective as streptocycline against *X. oryzae* PV *oryzae* (2.40 cm). Palmarosa oil one per cent exhibited an inhibition of 2.02 cm. This was followed by one and per cent of lemon grass oil (1.15 and 1.18 cm), cinnamon oil (1.10 and 1.80 %), and vetiver oil (1.13 and 1.17%). The inhibitory effect of plant oils against plant pathogen has been reported by several workers. (Nguefack *et al.*, 2005; Soylu *et al.*, 2006). The Bengyella *et al.* (2011) reported the antifungal activity of essential oil of *Ocimum gratissimum* against Alternaria *padwickii* and *Bipolaris oryzae*. The inhibitory effect of essential oil of *Cymbopogon citratus* against rice pathogen *Alternaria padwickii*, *Bipolaris oryzae* and *Fusarium moniliforme* was reported by Nguefack *et al.* (2008). Tripathi *et al.* (2008) reported the inhibitory effect of essential oils of *Ocimum sanctum*, *Prunus persica* and *Zingiber officinale* against *Botrytis cinera*. The essential oils of *Ocimum sanctum*, *Cymbopogon citrates*, *Cymbopogon martini* and *Pelargonium graveolens* were reported to be inhibitory to *Colletotrichum musae* and *Botryodiplodia theobromae*, causal agents of anthracnose of banana (Muthukumar and Renganathan, 2012). Sharma *et al.* (2013) reported the inhibition of mycelia growth of *Sarocladium oryzae in vitro* and reduction in sheath rot severity in field by citronella oil.

Plant oils and extracts with antimicrobial activity offer eco friendly option for disease management. The plant oils and plant extracts showing significant inhibition of bacterial blight pathogens has to be tested for its efficacy in the field. Further the active principles responsible for the inhibitory effect can also be studied for developing formulations.

CONCLUSIONS

Bacterial blight caused by *Xanthomonas oryzae* PV *oryzae* is a major production constraint in all the major rice growing countries. Management of the disease is difficult as there are no resistant varieties suitable to the state of Kerala. Recommended antibiotics are not giving satisfactory results in the situations of sudden outbreaks. In this study, Palma Rosa oil (*Cymbopogon martinii*) and extract of garlic bulb (*Allium sativum*) exhibited highest inhibition of *Xanthomonas oryzae* PV *oryzae*. Plant oils and plant extracts identified inhibitory to *Xanthomonas oryzae* PV *oryzae* from this study can be tested in the field and can be utilized for developing botanical formulations for the management of bacterial blight of rice.

ACKNOWLEDGEMENTS

The financial support provided by Govt. of India under DARE/ICAR 100 crore institutional grants is thankfully acknowledged.

REFERENCES

- 1. Amini, M., Safaie, N., Salmani, M. J., and Shams Bakhsh, M. (2012). Antifungal activity of three medicinal plant essential oils against some phytopathogenic fungi. *Trakia Journal of Sciences*. 10 (1), 1-8.s
- 2. Bansod, S., & Rai, M. (2008). Antifungal activity of essential oils from Indian medicinal plants against human pathogenic *Aspergillus fumigatus* and *A. niger. World Journal of Medical Sciences*, 3 (2): 81-88, 2008

- Bengyella, L., Nguefack, J. and Pranab, R. (2011). Evaluation of antifungal potential of Ocimum gratissimum extracts on two seed borne fungi of rice (Oryza sativa L.) in Cameroon. *Asian Journal of Biological Sciences*. 4: 306-311
- Biswas, A. (2007). Evaluation of neem formulations against sheath blight disease of rice. Indian Journal of Plant Protection, 35(2), 296-298.
- El Baroty, G. S., El Baky, H.H.A., Farag, R.S. and Salch, M. A. 2010. Characterisation of antioxident and antimicrobial compounds of cinnamon and ginger essential oils. *African Journal of Biochemistry Research*. 4(6), 167-174
- Jabeen, R., Ashraf, M., & Ahmad, I. (2009). Evaluating the effects of cold water diffusates against *Xanthomonas* oryzae pv.oryzae causing bacterial leaf blight in rice. *Archives of Phytopathology and Plant Protection*, 42 (2), 137 187.
- Jena, K. K. (2013). Stacking and molecular characterization of major genes towards broad spectrum resistance to virulent bacterial blight pathogen in rice. Proceedings of fourth international conference on bacterial blight on rice. December 2- 4th, 2013 held at CSIR Centre for Cellular and Molecular Biology, Hyderabad, India.
- 8. Kamalakannan, A., Shanmugam, V., Surendran, N., & Srinivasan, R. (2001). Antifungal properties of Plant extracts against *Pyricularia grisea*, the rice blast pathogen. Indian Phytopathology, 54(4), 490-492
- 9. Muthukumar, A. and Renganathan, R.2012. In vitro and in vivo evaluation of plant oils against anthracnose pathogen Colletotrichum musae and Botryodiplodia theobromae. Indian *Journal of Plant protection*. 2.91-94
- Nguefack, J. Somda,I. Mortensen,C.N. and Zollo,P.H.A. 2005. Evaluation of five essential oils from aromatic plants of Cameroon for controlling seed borne bacteria of rice (*Oryza sativa* L.). Seed Science technology. 33: 397-407
- Nguefack, J., Leth, V., Dongmo J. B. L., Torp, J., Zollo, P. H. A., and Nyasse, S. (2008). Use of three essential oil seed treatment against seed born fungi of rice. (*Oryza sativa* L). *American-Eurasian Journal of Agriculture and Environmental Science* 4 (5), 554-560.
- Nguefack, J., Wulff. G. E., Dongmo, J. B. L., Fouelefack, F. R., Fotio, D., Mbo, J.and Torp, J. 2013. Effect of plant extracts and an essential oil on the control of brown spot disease, tillering, number of panicles and yield increase in rice. *European Journal of Plant Pathology*. 137: 871-882.
- Pattnaik, S., Subramaniyam, V. R., & Kole, C. (1996). Antibacterial and antifungal activity of ten essential oils *in vitro*. *Micribios*, 86, 237 46.
- Sharma, L., Sharma, K. K & Sinha, A. P. (2013). Potential application of botanicals, essential oils and natural products against *Sarocladium oryzae*. Annals of Plant Protection Science, 21 (1), 109 – 113
- Sehajpal. A., Arora, S., & Knur, P. (2009). Evaluation of plant extracts against *Rhizoctonia solani* causing sheath blight of rice. The Journal of Plant Protection Science, 1, 25-30.
- Soyulu, E.M., Soylu, S. and Kurt, S. 2006. Antimicrobial activities of the essential oils of various plants against tomato late blight disease agent *Phytophthora infestans*. *Mycopathologia*. 161, 119-128

Inhibitory Effect of Plant Extracts and Plant Oils on *Xanthomonas Oryzae* PV *Oryzae*, The Blight Pathogen of Rice

 Tripathi, M., Dubey, N. K. and Shukla, A.K. (2008). Use of some essential oils as post harvest botanical fungicides in the management of grey mould of grapes caused by *Botrytis cineria*. World Journal of Microbial *Biotechnology* 24, 39-46.

APPENDICES

Sl. No	Treatments (Plant Extracts)	Diameter of Inhibition Zone (Cm)
1	Ginger (Zingiber officinale)	0.0 (1.00)
2	Garlic (Allium sativum)	3.1 (2.02)
3	Aloe vera	0.0 (1.00)
4	Asafoetida (Ferula assafoetida)	0.0 (1.00)
5	Lantana (Lantana camara)	0.0 (1.00)
6	Tamarind leaf (Tamarindus indica)	0.0 (1.00)
7	Turmeric (Curcuma longa)	0.0 (1.00)
8	Pepper (Piper nigrum)	0.0 (1.00)
9	Gooseberry (<i>Phyllanthus emblica</i>)	0.0 (1.00)
10	Thippali (Piper longum)	0.0 (1.00)
11	Tamarind fruit (Tamarindus indica)	1.96 (1.71)
12	Neem (Azadiracta indica)	0.0 (1.00)
13	Tulsi (Ocimum sanctum)	0.0 (1.00)
14	Gooseberry (Phyllanthus emblica)	1.62 (1.61)
15	Lemon Juice (Citrus aurantifolia)	1.57 (1.60)
16	Tomato (Solanum lycopersicum)	0.0 (1.00)
17	Curry leaf extract (Murraya koenigii)	0.0 (1.00)
18	Papaya Leaf (Carica papaya)	0.0 (1.00)
19	Eupatorium (Chromolaena odorata)	0.0 (1.00)
20	Murikoodi (Hemigraphis colorata)	0.0 (1.00)
21	Green mango extract (Mangifera indica)	1.55 (1.59)
22	Panikoorka (Plectranthus amboinicus)	0.0 (1.00)
23	Cheroola (Aerva lanata)	0.0 (1.00)
24	Irumbanpuli (Averrohoa bilimbi)	0.0 (1.00)
25	Onion (<i>Allium cepa</i>)	0.0 (1.00)
26	Control	0.0 (1.00)
	CD (0.05 %)	0.018

Table 1: Effect of Plant Extracts on Xanthomonas Oryzae PV Oryzae

Values in parenthesis are $\sqrt{x+1}$ transformed. Each value is the mean of three replications

 Table 2: Effect of Essential Oils on X Oryzae PV Oryzae

Treatments (Plant Oils)	Dose	Zone of Inhibition (Cm)
T ₁ - Lemon grass (<i>Cymbopogon flexuous</i>)	1.0 (%)	1.15 (1.46)
T ₂ - Lemon grass (<i>Cymbopogon flexuous</i>)	5.0 (%)	1.18 (1.47)
T ₃ - Palmarosa (<i>Cymbopogon martinii</i>)	1.0 (%)	2.02 (1.73)
T ₄ - Palmarosa (<i>Cymbopogon martinii</i>)	5.0 (%)	2.45 (1.85)
T ₅ Vetiver (Chrysopogon zizanioides)	1.0 (%)	1.13 (1.45)
T ₆ . Vetiver (<i>Chrysopogon zizanioides</i>)	5.0 (%)	1.17 (1.46)
T ₇ - Cinnamon (<i>Cinnamomum zeylanicum</i>)	1.0 (%)	1.10 (1.44)
T ₈ - Cinnamon (<i>Cinnamomum zeylanicum</i>)	5.0 (%)	1.80 (1.67)
T ₉ - Maroti (<i>Hydnocarpus pentadra</i>)	1.0 (%)	0.00 (1.00)
T ₁₀ . Maroti (Hydnocarpus pentadra)	5.0 (%)	0.00 (1.11)
T ₁₁ - Streptocycline	100 ppm	2.40 (1.84)
T ₁₂ . Control	-	0.00 (1.00)
CD (0.05 %)		0.075

Values in parenthesis are $\sqrt{x+1}$ transformed. Each value is the mean of three replications