Int.J.Curr.Microbiol.App.Sci (2018) Special Issue-6: 1905-1909

International Journal of Current Microbiology and Applied Sciences
ISSN: 2319-7706 **Special Issue-6 pp. 1905-1909**
Journal homepage: http://www.ijcmas.com

Original Research Article

Efficacy of Bioagents and Botanicals against *Xanthomonas axonopodis* pv. *punicae* causing Bacterial Blight of Pomegranate

K.T. Apet*, R.C. Agale, O.S. Thakur and M.R. Tambe

Department of Plant Pathology, College of Agriculture, VNMKV,
Parbhani - 431402 (MS), India
**Corresponding author*

Keywords

Xanthomonas axonopodis pv. *punicae*, Bacterial blight, Pomegranate, Bioagents, Botanicals and inhibition zone technique

A B S T R A C T

Bioagents *viz.*, *T. hamatum*, *B. subtilis*, *P. fluorescence*, *T. virens*, *T. harzianum* and *T. viride* and botanicals *viz.*, Shatavari leaf extract, Sadaphuli leaf extract, Adulasa leaf extract, Karanj leaf extract, Ashwagandha leaf extract, Behada leaf extract, Ritha leaf extract were tested against *Xanthomonas axonopodis* pv. *punicae* a pathogen of oily spot of pomegranate by inhibition zone technique. Maximum inhibition was observed due to the *P. fluorescence* (12.82 mm), while minimum with *T. virens* (2.38 mm), similarly maximum inhibition zone was observed due to leaf extract of botanical Sadaphuli (8.22 %) followed by Ritha (6.50%) inhibition at 10 per cent concentration.

Introduction

In recent years the bacterial blight of pomegranate caused by *Xanthomonas axonopodis* pv. *punice* is of the important disease of pomegranate causing heavy losses in yield and quality of fruits in pomegranate. The survey conducted in Pune and Sangli district of Maharashtra has reported that the farmers are using bleaching powder, Bordeaux mixture, di ammonium phosphate, urea, farmyard manure to manage the disease without any success (Pawar and Kadam, 2012).

Plant extract and bioagents are widely screened and used for its antogonostci activity against pathogenic microorganism (Britto *et al.*, 2011; Azzebegi, *et al.*, 2010; Bonyadi *et al.*, 2009; Raju *et al.*, 2013). Therefore, the present studies were under taken and this will help to formulate the disease management strategies.

Materials and Methods

Before preparation of leaf extract leaves each plant species were dipped in the 0.1 % mercuric chloride ($HgCl_2$) for one minute. Leaf extracts were prepared by grinding 100 gm washed leaves of each plant species in 100ml distilled water with mixture-cum grinder. These were then filtered through Whatman No.1 filter paper using funnel and volumetric flask (100 ml capacity). The final clear filtrate obtained was treated as 100 per

cent concentration of standard leaf extract. The desired quantity required for preparation of 10 and 20% concentration was taken from this 100 per cent standard leaf extract. These leaf extracts were then evaluated *in vitro* against *X. axonopodis* pv. *punicae* by applying inhibition zone technique (Giri *et al.*, 2008).

Three replication for each concentration of leaf extract were maintained. Plates containing YGCA medium with bacterial suspension without any leaf extract were maintained as control. All these petriplates were incubated at 28 ± 2 °C for 48 hours. Observation on growth of test pathogen and per cent inhibition over control was calculated by the formula of Vincent, 1947.

The antagonistic potential of bioagents *viz*., *T. hamatum*, *B. subtilis*, *P. fluorescence*, *T. virens*, *T. harzianum* and *T. viride* were assessed *in vitro* against *X. axonopodis* pv. *punicae* by Inhibition zone method. Biocontrol agents were collected from department Plant of Pathology, VNMKV, Parbhani. For this 30 ml of YGCA medium was poured in sterilized petriplates and allowed to solidify. Seeded broth with *X. axonopodis* pv. *punicae* was spread over the petriplates with the help of sterilized spreader.

A 5 mm diameter well was prepared in the centre of the petriplate with the help of sterilized cork borer and 2 ml of broth containing bio-agents were poured in that well. Sterilized distilled water was used as a control. Observations regarding inhibition zone formed by biocontrol agents against *X. axonopodis* pv. *punicae* were recorded 5-7 days after inoculation by incubating at 28±2^{o}C (Raju, 2010). Observation on growth of test pathogen and per cent inhibition over control was calculated by the formula of Vincent (1947).

Results and Discussion

The result presented in Table 1 revealed that Sadaphuli leaf extract at 10 per cent was found most effective against *X. axonopodis* pv. *punicae* by forming 8.22 per cent inhibition. Ritha leaf extract was found second best effective plant extract which showed 6.50 per cent inhibition followed by Adulasa leaf extract (5.35%), Shatavari leaf extract (4.52%), Karanj leaf extract (4.11%), Ashwagandha leaf extract (4.11%) and Behada leaf extract 3.28 per cent inhibition.

Similarly, the result presented in Table 1 and Figure 1 revealed that Sadaphuli leaf extract at 20 per cent was found most effective for controlling *X. axonopodis* pv. *punicae* by forming 9.06 per cent inhibition. Ritha leaf extract was found second best effective plant extract which showed 8.22 per cent inhibition followed by Adulasa leaf extract (6.99%), Shatavari leaf extract (6.58%), Karanj leaf extract (5.75 %), Ashwagandha leaf extract (5.36%) and Behada leaf extract 5.36 per cent inhibition.

The data presented in Table 2 and Figure 2 revealed that *P. fluorescence* produced maximum metabolites in the culture medium resulting in the production of inhibition zone of 12.82 mm followed by *B. subtilis* 10.24.

Amongst fungal antagonists *T. viride* though inhibited the bacterium, but the inhibition zone produced half to that of bacterial antagonists i.e. 6.63 mm followed by *T. hamatum* 3.29mm, *T. harzianum*2.4mm. *T. virens* was the least effective producing only 2.38 mm inhibition zone.

In present investigations the bioagents and botanicals observed effective in inhibition of growth of bacterium *X. axonopodis* pv. *punicae.*

Table.1 *In vitro* efficacy of botanicals against *X. axonopodis* pv. *punicae*

Tr. No.	Treatments	Mean bacterial growth* (mm) at conc.		% inhibition of bacterial growth	
		10 %	20%	10 %	20%
T_1	Shatavari leaf extract (*Asparagus racemosa*)	85.93	84.08	4.52 **(12.27)**	6.58 **(14.86)**
T_2	Sadaphuli leaf extract (*Catharanthus roseus*)	82.60	81.85	8.22 **(16.66)**	9.06 **(17.51)**
T_3	Adulasaleaf extract (*Adhatoda vasica*)	85.18	83.71	5.35 **(13.37)**	6.99 **(15.33)**
T_4	Karanj leaf extract (*Pongamia pinnata*)	86.30	84.82	4.11 **(11.69)**	5.75 **(15.33)**
T_5	Ashwagandha leaf extract (*Withania somnifera*)	86.30	85.18	4.11 **(11.69)**	5.36 **(13.38)**
T_6	Behada leaf extract (*Terminalia belerica*)	87.04	85.18	3.28 **(10.43)**	5.36 **(13.38)**
T_7	Ritha leaf extract (*Sapindus mukorossi*)	84.07	82.60	6.50 **(14.77)**	8.22 **(16.66)**
T_8	Control	90	90	0.00	0.00
	S.E. +	**0.89**	**0.51**	**0.51**	**0.29**
	C.D.(P=0.05)	**2.68**	**1.53**	**1.55**	**0.89**

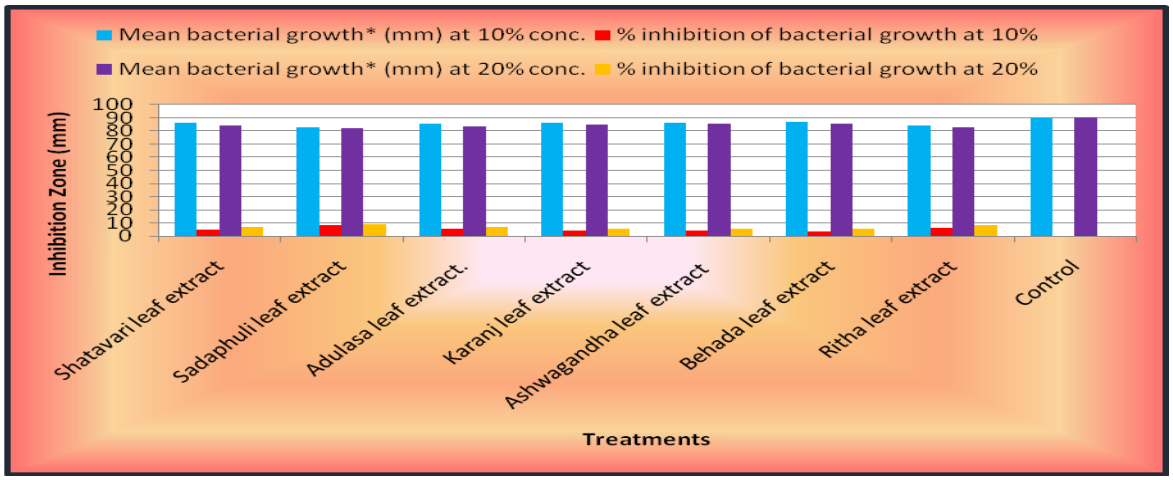
* Mean of three replications Figures in parenthesis are arcsin values.

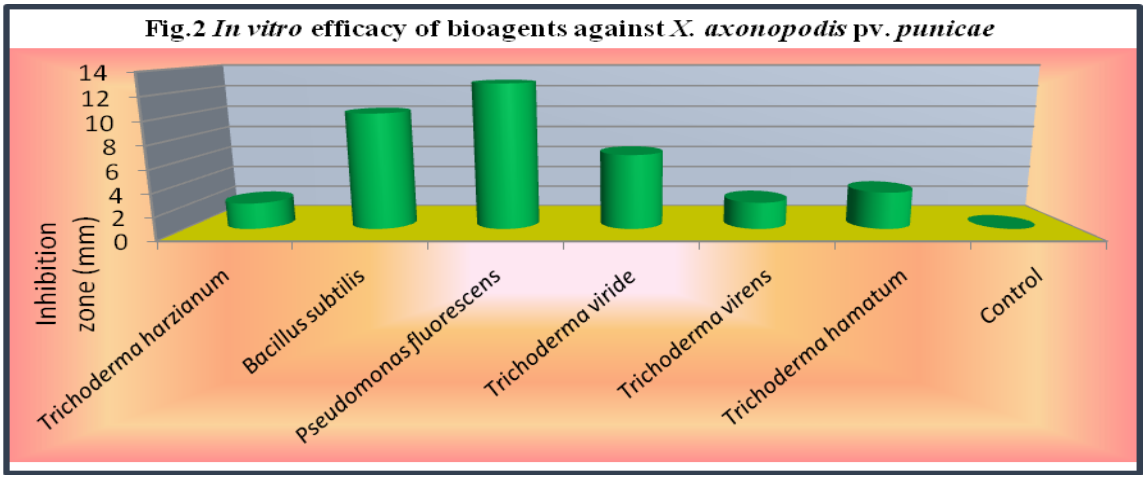
Table.2 *In vitro* efficacy of bioagents against *X. axonopodis* pv. *punicae*

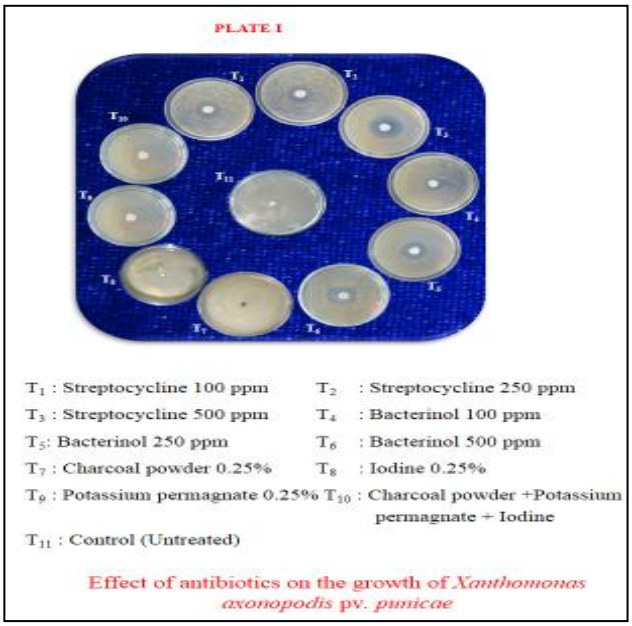
Tr. No.	Bioagents	Mean inhibition zone *(mm)
T_1	*Trichoderma harzianum*	2.4 **(8.91)**
T_2	*Bacillus subtilis*	10.22 **(18.66)**
T_3	*Pseudomonas fluorescens*	12.82 **(20.98)**
T_4	*Trichoderma viride*	6.63 **(14.92)**
T_5	*Trichoderma virens*	2.38 **(8.87)**
T_6	*Trichoderma hamatum*	3.29 **(10.45)**
T_7	Control	0.00 **(00)**
	SE±	**0.05**
	CD 0.05	**0.17**

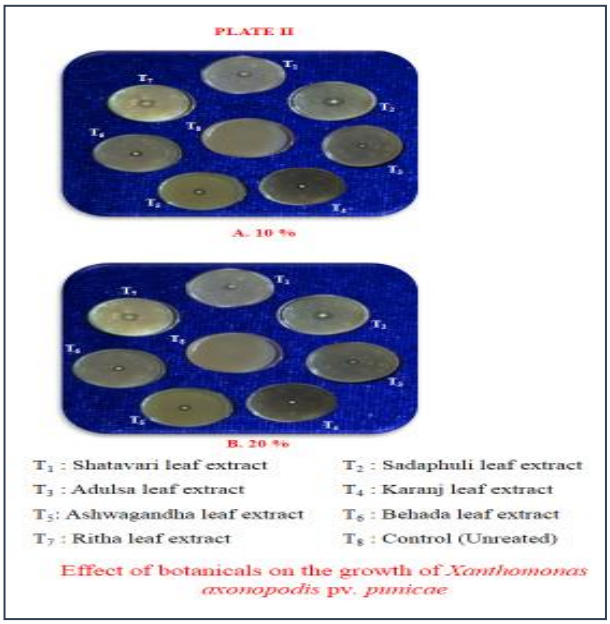
* Figures in parenthesis are arcsin values

Fig.1 *In vitro* efficacy of botanicals against *X. axonopodis* pv. *punicae*









The results obtained on control strategies correlates with the results of earlier worker (Granage, 1985; Laha *et al.,* 1992; Hulloli *et al.,* 1998; Manjula *et al.,* 2002; Apet *et al.,* 2013; Sajid, 2013).

References

Anonymous, 2015.Vision 2050, by NRC pomegranate, Solapur, pp.29

Apet, K.T., Suryawanshi, A.P., Gholve, V.M., More A.S. and Kuldhar D.P. 2013. Efficacy of bioagents and antibiotics against *X. axonopodis* pv. *punicae* causing oily spot of pomegranate, *J. Pl. Dis. Sci.* 8 (2): 209-211

Giri, G. K., Gade, R. N., Gulhane, A. R. and Das, S. 2008. Efficacy of bioagents and botanicals and chemicals against citrus canker *(X. axonopodis* pv. *citri) J. Pl. Dis. Sci.* 3(2): 249-250.

Grainge, M., Berger, L. and Ahmed, S. 1985. Effect of extracts of *Artabotrys uncinatus* and *Allium sativum* on *Xanthomonas campestris* pv. *oryzae*. *Current Sci.,* 54(2): 90p.

Hingorani, M. K. and Mehta, P. P. 1952. Bacterial leaf spot of pomegranate. *Ind. Phytopath*. 5:55-56

Hingorani, M. K. and Singh, N. J. 1959. *Xanthomonas Punicae* sp. *Nov. on punica granatum* L. *Ind. J. Agril. Sci.* 29: 45-48.

Hulloli, S. S., Singh, R. P., and Verma J. P. 1998. Management of bacterial blight of cotton induced by *Xanthomonas axonopodis* pv *malvacearum* with use of neem based formulations. *Ind. Phytopath.* 51(1):21-25.

Laha, G. S., Singh, R. P. and Verma, J. P. 1992. Biocontrol of *Rhizoctonia solani* in cotton by fluorescent *Pseudomonas*. *Ind. Phytopath*, 45: 412-415.

Manjula, C. P. and Khan, A. N. A. 2003. Incidence of bacterial blight of Pomegranate (*Punica granatum* L.) in Karnatka. *Indian Phytopath*, 56 (3): 341.

Raju, J., Benagi, V. I., Jayalakshmi, K., Angadi, S.G., Basha, H. and Giri M. S. 2013. Evaluation of chemicals, bioagents and botanicals for management of bacterial blight caused by *Xanthomonas axonopodis* pv. *punicae* under field conditions, *Bioinfolet.* 10 (2B): 714 -717.

Sajid, M., Rashid, A., Ehtishan-ul-haq, M., Talha Javed, M., Jamil, H., Mudassir M., Farrq, M., Ahamad, F., Latif, M.C., Ahamad M., Ahamad M. and Kamaran, A. 2013. *In vitro* evaluation of chemicals and plant extracts against colony growth of *Xanthomonas axonopodis* pv. *malvacearum* causing bacterial blight of cotton. *Euro. J. Exp. Bio*., 3(1): 617-621.

Vincent, J. M. 1927. Distortion of fungal hyphae in the presence of certain inhibitors. Nature. 159: 180.