

Management of citrus canker in Kagzi lime under nursery condition using Biorationals

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ABSTRACT :

The present study was carried out by using different bioagents and botanicals for management of citrus canker on four months old kagzi lime seedlings, Among the different botanicals treated, *Garcinia indica* fruit rind extract was found be effective in management of citrus canker under nursery condition by reducing the per cent leaves infection (81.55%), number of shoots infected (73.24%) and per cent leaf area infection (82.63%). This was followed by bioagent treatment consisting of soil and aerial application of *Lysinibacillus xylanilyticus* (VK-6B), which recorded 77.11 per cent, 71.43 per cent and 80.70 per cent, respectively.

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INTRODUCTION

Citrus is believed to have originated in the parts of Southeast Asia bordered by northeast India, Myanmar (Burma) and the Yunnan Province of China. Citrus fruits have been cultivated since ancient times, the best-known examples are the oranges, lemons, grapefruit and limes. Citrus canker is an important disease affecting citrus species caused by a bacterium *Xanthomonas citri* subsp *citri*. Infection leads to formation of lesions on the leaves and stem incase of nursery, severe infection leads to death of seedlings. It is also known that the pathogen infects the seedlings at nursery stage with an incidence of 72.22 per cent and severity of 27.55 per cent (Rashid *et al.*, 2014). The disease is extremely persistent when

it becomes established in an area. Citrus groves have been destroyed in large numbers to eradicate the disease. Hence, it is important to manage the disease at seedling stage.

MATERIAL AND METHODS

The investigation was conducted in four months old lime plants planted in green house condition at College of Horticulture Bagalkot. Totally 405 plants were used for the study. The treatments were taken up based on the results obtained under lab condition and the effective bioagents and botanicals were tested against forty five plants each and among them 15 plants were tagged for recording the disease parameters such as per cent leaves

infected, per cent leaf area infected and number of shoots infected. The plants were sprayed every fortnight interval with four spray schedule using ganesh sprayer. The procedure for mass multiplication of bioagents and preparation of botanical spray solutions are mentioned below.

Multiplication of bio agents :

Liquid mother cultures of the *Lysinibacillus xylanilyticus* (VK-6B) and KK-3 isolates were taken out in separate test tubes and by gently opening the cotton plug of the sterilized conical flask containing the prepared nutrient agar broth one loop of each liquid mother culture of two isolates were poured carefully into the liquid medium from the test tubes. Inoculated flasks were kept for multiplication in the orbital shaker at 30°C at 200 rpm for 72 hour. After 24 hours the clear medium turned turbid. Turbidity indicates the bacterial multiplication. It was used for nursery application after dilution with water (100 ml to 10 litre of water such that it reached a

population of 10⁸ cfu /ml) Vijayan *et al.* (2013).

Extraction of plant extracts :

200 g of both *Garcinia indica* and *Prosopis juliflora* were collected and washed in tap water, later rinsed in distilled water then added to the mixer and grounded at the ratio of 1:1. This mixture was kept overnight for the release of metabolites in to the solution and next day is filtered using muslin cloth. Later, this solution is diluted to 5 times the dilution *i.e.* 5 litre of water is added to 1 litre obtained solution. The solution was sprayed to the plants in nursery.

RESULTS AND DISCUSSION

The per cent leaves infected in *Garcinia indica* extract treated plants before spray was 20.05, which subsequently reduced to 18.33, 16.53, 11.63 and 3.70 per cent. This was found at par with soil and aerial application of *Lysinibacillus xylanilyticus* (VK-6B) which recorded 15.58 before spray and 14.80, 13.08,

Table 1: Effect of biorationals on development of citrus canker under nursery condition (Per cent leaves infected)										
Treatments	Treatment details	Per cent leaves infected								
		Before spray	1 st spray	% reduction	2 nd spray	% reduction	3 rd spray	% reduction	4 th spray	% reduction
T ₁	Bioagent KK-3 (soil application)	20.33 (26.79)	20.33 (26.79)	0.00	19.83 (26.44)	2.46	18.67 (25.60)	8.20	17.00 (24.35)	16.39
T ₂	<i>Lysinibacillus xylanilyticus</i> soil application	19.28 (26.05)	18.83 (25.72)	2.33	16.88 (24.27)	12.45	13.77 (21.72)	28.61	9.07 (17.52)	52.98
T ₃	Bioagent KK-3 (aerial application)	16.57 (24.01)	16.53 (24.00)	0.20	15.87 (23.48)	4.23	14.60 (22.47)	11.87	13.00 (21.13)	21.53
T ₄	<i>Lysinibacillus xylanilyticus</i> (aerial application)	19.58 (26.26)	19.20 (25.99)	1.96	17.43 (24.67)	10.98	15.27 (23.00)	22.04	11.87 (20.15)	39.40
T ₅	Bioagent KK-3 (soil + aerial application)	15.22 (22.96)	14.99 (22.77)	1.49	14.00 (21.98)	8.00	12.23 (20.46)	19.61	10.47 (18.87)	31.22
T ₆	<i>Lysinibacillus xylanilyticus</i> (soil + aerial application)	15.58 (23.26)	14.80 (22.63)	5.03	13.08 (21.21)	16.04	9.77 (18.22)	37.33	3.57 (10.88)	77.11
T ₇	<i>Garcinia indica</i> fruit rind extract @ 1:5 concentration	20.05 (26.60)	18.33 (25.35)	8.56	16.53 (24.00)	17.54	11.63 (19.94)	41.98	3.70 (11.08)	81.55
T ₈	<i>Prosopis juliflora</i> leaf extract @ 1:5 concentration	21.53 (27.64)	20.67 (27.03)	4.02	18.53 (25.49)	13.93	14.40 (22.30)	33.13	7.30 (15.68)	66.10
T ₉	Control	18.43 (25.42)	18.80 (25.70)	-1.99	19.83 (26.44)	-7.59	26.00 (30.65)	-41.05	28.77 (32.44)	-56.06
S.E. ±		0.27			0.21		0.51		0.24	
C.D. (P=0.01)		1.10			0.87		2.07		0.98	

Note: - Figure in the parentheses are arcsine transformed values

9.77 and 3.57 after 1st, 2nd, 3rd and 4th sprays, respectively. The plants sprayed with *Garcinia indica* extract recorded maximum per cent reduction in per cent leaf infection (81.55%) followed by soil and aerial application of *Lysinibacillus xylanilyticus* (VK-6B) with (77.11%). While *Prosopis juliflora* extract was the third best treatment with 66.10 per cent reduction as shown in Table 1. Similarly in case of *Prosopis juliflora* extract, the per cent leaf infection before spray was 21.53 and gradually decreased to 20.67, 18.53, 14.40 and 7.30. Whereas the untreated control recorded 56.06 per cent increase in the leaf infection from 18.43 to 18.80, 19.83, 26.00 and 28.77 after each spray.

The data on per cent leaf area infected are presented in Table 2 revealed that some of the treatments were at par with each other. The per cent leaf area infected in

Garcinia indica treatment was 13.43 before spray and later on decreased to 11.73, 10.07, 7.57 and 2.33 after 1st, 2nd, 3rd and 4th spray, respectively. Similarly in soil and aerial application of *Lysinibacillus xylanilyticus* (VK-6B), the number of leaf infected was 11.72 before spray which reduced to 10.43, 8.95, 6.97 and 2.30 after each successive spray. The highest reduction (82.63%) in per cent leaf area infected was recorded in *Garcinia indica* extract followed by soil and aerial application of *Lysinibacillus xylanilyticus* (VK-6B) @ 10⁸ cfu /ml (80.37%). The next best treatment was *Prosopis juliflora* extract with 78.13 per cent reduction. Whereas the per cent leaf infected in *Prosopis juliflora* extract treatment was 14.10 before spray which decreased to 12.85, 11.53, 8.70 and 3.08 after each successive sprays. In the untreated water control, the number of shoots

Treatments	Treatment details	Per cent leaf area infected								
		Before spray	1 st spray	% reduction	2 nd spray	% reduction	3 rd spray	% reduction	4 th spray	% reduction
T ₁	Bioagent KK-3 (soil application)	13.62 (21.65)	13.55 (21.60)	0.49	13.40 (21.47)	1.59	12.50 (20.17)	8.20	10.80 (19.18)	20.69
T ₂	<i>Lysinibacillus xylanilyticus</i> soil application	12.50 (20.70)	11.70 (20.00)	6.40	10.97 (19.33)	12.27	8.50 (16.93)	32.00	5.80 (13.93)	53.60
T ₃	Bioagent KK-3 (aerial application)	10.47 (18.87)	10.37 (18.78)	0.96	10.32 (18.73)	1.43	9.20 (17.62)	12.10	7.73 (16.06)	26.11
T ₄	<i>Lysinibacillus xylanilyticus</i> (aerial application)	12.68 (20.87)	12.28 (20.51)	3.15	11.98 (20.25)	5.52	9.13 (17.58)	27.99	6.83 (15.13)	46.12
T ₅	Bioagent KK-3 (soil + aerial application)	11.27 (19.61)	11.13 (19.49)	1.18	10.90 (19.27)	3.25	9.27 (17.72)	17.75	6.67 (14.92)	40.83
T ₆	<i>Lysinibacillus xylanilyticus</i> (soil + aerial application)	11.72 (20.01)	10.43 (18.85)	10.95	8.95 (17.40)	23.61	6.97 (15.31)	40.54	2.30 (8.72)	80.37
T ₇	<i>Garcinia indica</i> fruit rind extract @ 1:5 concentration	13.43 (21.50)	11.73 (20.03)	12.66	10.07 (18.48)	25.06	7.57 (15.96)	43.67	2.33 (8.76)	82.63
T ₈	<i>Prosopis juliflora</i> leaf extract @ 1:5 concentration	14.10 (22.06)	12.85 (21.01)	8.87	11.53 (19.85)	18.20	8.70 (17.16)	38.30	3.08 (10.11)	78.13
T ₉	Control	12.08 (20.34)	12.33 (20.56)	-2.07	12.98 (21.11)	-7.45	14.07 (22.03)	-16.41	15.67 (23.31)	-29.66
S.E.+			0.32		0.39		0.41		0.58	
C.D. (P=0.01)			1.29		1.58		1.65		2.37	

Note :- Figure in the parentheses are arcsine transformed values

infected was 12.08 before spray and increased to 12.33, 12.98, 14.07 and 15.67 at each stage.

The data on number of shoots infected is presented in Table 3 and from the results it is evident that the treatments are significantly different from each other. The number of lesions on shoot in *Garcinia indica* was 2.37 before spray which decreased to 2.27, 2.07, 1.83 and 0.63 after 1st, 2nd, 3rd and 4th spray, respectively. Similarly in soil and aerial application of *Lysinibacillus xylanilyticus* (VK-6B), the numbers of lesions were 1.40 before spray which reduced to 1.34, 1.24, 1.11 and 0.40 after each successive spray. The highest per cent reduction (73.24%) in number of lesions per shoot was found in *Garcinia indica* extract followed by soil and aerial application of *Lysinibacillus xylanilyticus* (VK-6B) @ 10⁸ cfu /ml (71.43%). The next best treatment was *Prosopis juliflora* extract which recorded 69.83 per cent reduction. Whereas in *Prosopis juliflora* extract

the number was 1.93 before spray and decreased to 1.87, 1.74, 1.56 and 0.58 after 1st, 2nd, 3rd and 4th spray, respectively. In the untreated water control, the number of lesions on shoot was 1.57 before spray and increased to 1.57, 1.60, 1.72 and 1.92 at each stage.

Garcinia indica fruit rind extract was found effective in management of citrus canker under nursery condition by recording maximum reduction in per cent leaves infected (81.55%), number of shoots infected (73.24%) and per cent leaf area infection (82.63%). The treatment consisting of soil and aerial application of *Lysinibacillus xylanilyticus* (VK-6B) recorded 76.29 per cent, 77.11 per cent, 71.43 per cent and 71.43 per cent reduction for the same parameter, respectively. *Garcinia indica* being acidic in nature has been observed as an efficient management tool in managing *Xanthomonas axanopodis* pv. *punicae* (Ramesh, 2015). As per the field observation *Garcinia indica* has

Table 3 : Effect of biorationals on development of citrus canker under nursery condition		(Number of lesions per shoot)									
Treatments	Treatment details	Number of lesions per shoot									
		Before spray	1 st spray	% reduction	2 nd spray	% reduction	3 rd spray	% reduction	4 th spray	% reduction	
T ₁	Bioagent KK-3 (soil application)	2.40	2.40	0.00	2.37	1.39	2.20	8.33	1.83	23.61	
T ₂	<i>Lysinibacillus xylanilyticus</i> soil application	2.17	2.13	1.54	2.00	7.69	1.81	16.62	1.20	44.62	
T ₃	Bioagent KK-3 (aerial application)	1.73	1.73	0.00	1.67	3.85	1.58	9.04	1.28	26.35	
T ₄	<i>Lysinibacillus xylanilyticus</i> (aerial application)	2.03	2.03	0.33	1.90	6.56	1.73	14.75	1.23	39.34	
T ₅	Bioagent KK-3 (soil + aerial application)	2.73	2.73	0.00	2.67	2.44	2.40	12.20	1.73	36.59	
T ₆	<i>Lysinibacillus xylanilyticus</i> (soil + aerial application)	1.40	1.34	4.05	1.24	11.43	1.11	20.71	0.40	71.43	
T ₇	<i>Garcinia indica</i> fruit rind extract @ 1:5 concentration	2.37	2.27	4.23	2.07	12.68	1.83	22.54	0.63	73.24	
T ₈	<i>Prosopis juliflora</i> leaf extract @ 1:5 concentration	1.93	1.87	3.10	1.74	9.83	1.56	19.14	0.58	69.83	
T ₉	Control	1.57	1.57	0.00	1.60	-1.91	1.72	-9.57	1.92	-22.34	
S.E. ±			0.09		0.06		0.06		0.05		
C.D. (P=0.01)			0.37		0.23		0.24		0.22		

prevented the formation of cankers on leaf and stem, also has shown the positive strategy in controlling the spread of spots and lesions. Similarly five plant extracts of *Hibiscus subdariffa* Linn., *Psidium guajava* Linn., *Punica granatum* Linn., *Spondias pinnata* (Linn.f.) Kurz and *Tamarindus indica* Linn. were evaluated for control of canker disease on *Citrus aurantifolia* (lime) caused by *Xanthomonas axonopodis* pv. *citri* under greenhouse condition by Chalida *et al.* (2001) and found that Tamarind extract was effective in controlling canker disease with 48 per cent over the water control with 100 per cent disease incidence. However, some bioagents *Lysinibacillus xylanilyticus* has shown a promising result along with *Garcinia indica*, According to Guang *et al.* (2008) in three field trials, *Lysobacter antibioticus* strain 13-1 reduced bacterial blight incidence of rice due to the antibiotics and density of colonization on leaves involved for biological control. Khodakaramian *et al.* (2008) reported the use of different strains of *Pseudomonas* in management of bacterial canker of citrus by reducing the number of spots on leaf under greenhouse condition.

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