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Eco-friendly management of cabbage black rot disease by botanicals

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Abstract
Cabbage is the most abundantly consumed vegetable crop among the crucifers globally. Black rot disease (*Xanthomonas campestris* pv. *campestris*) is the principal yield-limiting and destructive pathogen affecting worldwide cabbage production. The use of eco-friendly management practice is of paramount importance to residue toxicity on the host, environmental pollution and harmful effect on consumer's health. The present study consists of eighteen different botanicals for black rot disease management under *in-vitro* conditions. Highest per cent inhibition was showed by tulsi leaf extract (31.58 mm) followed by mint (26.35 mm) and garlic (18.71 mm) and the lowest per cent inhibition was observed with leaf extract of galanga (9.56 mm). In vivo evaluation of Tulsi leaf extract (at 10%) known to reduce 84.77 per cent disease incidence as compared to control and recorded maximum plant growth and yield parameters.

Keywords: Eco-friendly management, cabbage black rot disease, botanicals

Introduction
Cabbage (*Brassica oleracea* var. *capitata* L.) is a leafy winter vegetable grown for its edible enlarged terminal bud. In addition to several minerals such as calcium, phosphorus, potassium, sulphur and iron present in cabbage, it also contains high percentage of protein and vitamins A, B and C. The crop suffers severely (>60% loss) from black rot, one of the most serious bacterial diseases with its worldwide occurrence (Williams, 1998) [3]. Hence, there is a need to develop suitable eco-friendly management strategies. Though, the control of the disease using chemicals has been found to give satisfactory results, however, the application of the chemicals for the control of the disease is not encouraged due to environmental hazards and development of acquired resistance by the pathogen. Another non-chemical method is the botanicals, which are of natural origin and bio-degradable in nature and do not leave any toxic residues or byproducts to accumulate in the environment. Also, the cost involved in botanical pesticides production and application is much less compared to chemical pesticides.

Material and Methods
Bio-assay with botanicals
Eighteen different botanical extracts *ie*. neem, tulsi, garlic, vasaka, ashwagandha, babchi, galanga, munguruvalli, long pepper, senna, sarpagandha, shatavari, mint, kalmegh, brahmi, coleus, turmeric and lemon grass were tested for their efficacy against black rot disease management caused by *X. c.* pv. *campestris* on cabbage both *in vitro* and *in vivo* conditions.

In vitro assay of botanicals
The extracts of eighteen different plant leaves and bulbs were used to examine the inhibitory effect on bacterial growth. The fresh plant material were collected and washed first in tap water followed by distilled water; 100 grams of fresh sample was chopped and macerated in a surface sterilized pestle and mortar by adding 100 ml of sterile water (1:1 w/v). The extract was filtered through two layers of muslin cloth, filtrate thus obtained was used as a stock solution. 5, 7.5 and 10 per cent each of plant extract was prepared by mixing 5, 7.5 and 10 ml of stock solution with 95, 92.50 and 90 ml of sterilized distilled water, respectively.
The bacterial suspension (48 hr old) of *X. c.* pv. *campestris*, multiplied in nutrient broth (20 ml) was mixed with molten nutrient agar medium (1000 ml) contained in an Erleyenmayer's flask, so as to get the thick growth of bacteria on the medium, 15 to 20 ml of seeded medium was poured onto the sterilized petriplates and allowed to solidify. 6 to 8 mm diameter well was made in each agar plate using sterilized cork borer.

50 µl of plant extracts with different concentrations were loaded into wells in petriplates separately and sterile distilled water was used as control check. Then inoculated plates were incubated at 32 °C for 48 hours. The Observations were recorded periodically on inhibitory zones (diameter in mm) produced by plant extracts against *X. c.* pv. *campestris* around the wells and then analyzed statistically.

In vivo assay of botanicals

A pot experiment was carried out for the management of black rot of cabbage. There were 12 treatments replicated 3 times with complete Randomized design. 30 days old cabbage (variety saint) seedlings was planted in pot containing sterilized soil. Cabbage seedlings were artificially pre inoculated with black rot causing bacterial suspension of *X. c.* pv. *campestris* (5×10^6 cfu/ml).

Based on the *in vitro* condition, effective treatment tulsi leaf extract botanical was selected for management. Each treatment was sprayed 5 times at an interval of 15 days initiating the first spray at the onset of disease on leaves. Observations on the incidence of disease on leaves were recorded before and after spray of botanical using 0–5 scale. And growth parameters *viz*., head diameter (cm), head weight (g), leaf area index (cm^2) and root length (cm) were recorded and analyzed statistically.

Result and Discussion

In vitro efficacy of botanical

The results showed significant difference among the treatments for black rot disease management in cabbage. Among the tested botanicals, tulsi leaf extract (@ 10% Conc.) was found to be most effective and recorded maximum inhibitory effect (31.58 mm) (Table 1) followed by mint (26.35 mm). It is mainly due to presence of basic active ingredients (methyl cinnamate, citral, eugenol, linalool, camphor and methyl chavicol) and presence of secondary metabolites (alkaloids, tannins, steroids, saponins and terpenoids). Similar results were reported by reported by Sharma and Patel (2017) [2].

In vivo efficacy of botanical

Tulsi leaf extract (at 10%) were tested under *in vivo* condition to known the efficacy of botanical against black rot disease. The results showed 84.77 per cent (Table 2) disease reduction as compared to control and recorded maximum plant growth and yield parameters like leaf area (0.71 cm2), root length (17.54 cm), head diameter (24.70 cm) and head weight (1840 g) (Table 3). This might be due to the presence of secondary metabolites (alkaloids, volatiles oils, tannins, saponin, flavonoids and sterols) Deepa and Kanika, 2014 [1].

Table 1: *In vitro* efficacy of botanicals against *Xanthomonas campestris* pv. *campestris*

Treatments	Common name	Botanical name	Inhibition zone (mm)			Mean
			5 (%)	7.5 (%)	10 (%)	
T_1	Neem	*Azadirachta indica*	10.48(18.89)	11.00(19.37)	11.33(19.67)	10.94
T_2	Tulsi	*Ocimum sanctum*	26.90(31.24)	29.60(32.96)	38.23(38.19)	31.58
T_3	Garlic	*Allium sativum*	14.49(22.37)	19.50(26.21)	22.15(28.08)	18.71
T_4	Vasaka	*Adhathoda vasica*	11.27(19.62)	14.08(22.04)	15.42(23.12)	13.59
T_5	Ashwagandha	*Withania somnifera*	10.50(18.91)	11.15(19.51)	11.42(19.75)	11.02
T_6	Babchi	*Psoralea corylifolia*	09.25(17.71)	12.23(20.47)	16.25(23.77)	12.58
T_7	Galanga	*Alpinia galangal*	07.18(15.54)	10.50(18.91)	11.00(19.37)	9.56
T_8	Munguruvalli	*Cissus quandragularies*	0.00(0.07)	0.00(0.07)	0.00(0.07)	0.00
T_9	Long pepper	*Piper longum*	0.00(0.07)	00.00(0.07)	0.00(0.07)	0.00
T_{10}	Senna	*Cassia angustifolia*	10.17(18.60)	16.10(23.66)	16.75(24.16)	14.34
T_{11}	Sarpagandha	*Rauvolfia serpentine*	10.33(18.75)	13.45(21.51)	15.75(23.38)	13.18
T_{12}	Shatavari	*Asparagus racemosus*	0.00(0.07)	00.00(00.07)	00.00(00.07)	0.00
T_{13}	Mint	*Mentha arvensis*	24.15(29.43)	26.00(30.66)	28.90(32.52)	26.35
T_{14}	Kalmegh	*Andrographis paniculata*	0.00(0.07)	0.00(0.07)	0.00(0.07)	0.00
T_{15}	Brahmi	*Centella asiatica*	10.57(18.97)	15.35(23.05)	16.35(23.85)	14.09
T_{16}	Coleus	*Coleus forskohlii*	10.10(18.53)	11.50(19.82)	15.15(22.91)	12.25
T_{17}	Turmeric	*Curcuma longa*	16.37(23.43)	17.18(24.49)	19.27(26.04)	17.61
T_{18}	Lemon grass	*Cymbopogon flexuosus*	13.27(21.36)	17.25(24.54)	20.08(26.62)	16.87
T_{19}	Streptocyclin (0.05%) + Copper oxychloride (0.3%)		40.00(39.23)	40.23(39.37)	39.92(39.18)	40.05
T_{20}	Control		0.00(0.07)	0.00(0.07)	0.00(0.07)	0.00
	S. Em±		0.11	0.15	0.20	
	CD at 1%		0.43	0.52	0.76	

Values in parentheses indicate Square root transformed values

Table 2: *In-vivo* efficacy of tulsi leaf extract against of black rot disease of cabbage

Treatments	Treatment details	Before spray	Per cent Disease Incidence (Spray)					
			1st	2nd	3rd	4th	5th	% Disease reduction
T_1	Tulsi leaf extract	1.74	3.23	4.26	4.94	5.57	6.43	84.77
T_2	Standard check (Streptocycline + Copperoxychloride)	1.70	2.23	2.87	3.25	3.98	4.20	90.05
T_3	Control (Distilled water spray)	1.82	18.25	22.47	29.26	35.22	42.23	
	S.Em.±	0.03	0.04	0.04	0.05	0.04	0.07	
	C.D. @ 1%	0.10	0.14	0.14	0.18	0.17	0.24	

Table 3: Effect of tulsi leaf extract on growth and yield parameters of cabbage

Treatments	Treatments details	Growth parameter		Yield parameter	
		Leaf area index (cm^2)	Root length (cm)	Head diameter (cm)	Head weight (Kg)
T_1	Tulsi leaf extract (at 10%)	0.71	17.54	24.70	1.84
T_2	Standard check (Streptocycline 0.05% + Copper Oxychloride 0.3%)	0.61	15.32	22.78	1.65
T_3	Control (Distilled water spray)	0.37	8.16	18.55	0.65
	S.Em.±	0.02	0.07	0.17	0.06
	C.D. @ 1%	0.06	0.27	0.65	0.22

Conclusion

Botanicals are safe, eco friendly and cost effective means of managing the crop diseases effectively. Among the botanicals Tulsi leaf extract (at 10%) was found to be best treatment to reduce cabbage black rot disease incidence there by enhance the plant growth and yield, which is mainly due to presence of secondary metabolites. Therefore, certainly plant extracts represent a potentially valuable and eco-friendly crop protection tool in high-value cropping systems in horticulture crop production.

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