

Original Research Article

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## Efficacy of Bio Resources on Management of Cercospora Leaf Spot in Beetroot (*Beta vulgaris* L.)

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### ABSTRACT

Beetroot (*Beta vulgaris* L.) is also known as table beet, garden beet and sugar beet. Among the diseases of beetroot Cercospora leaf spot is the most devastating foliar disease which reduces the root quality and sugar content. Management of this disease mainly depends on chemical fungicides. To reduce the use of fungicides one such attempt has been made to evaluate the effect bio resources and citral essential oil compound against cercospora leaf spot in *in-vivo* and *in-vitro* in department of Plant Pathology, SHUATS, Prayagraj, and Uttar Pradesh. Citral essential oil compound was tested *In-vitro* at different concentrations viz., 0.75%, 0.5%, 1%, 1.25% and Carbendazim 0.01% against radial growth of *Cercospora beticola*. Results revealed that the minimum radial growth was observed at a concentration of citral @ 1.25% (1.40 mm) with highest per cent inhibition (96.52%) followed by citral @ 1% (4.73 mm) with per cent inhibition (88.24%) compared to control. Application of bio resources and citral essential oil compound in *in-vivo* revealed that disease severity was recorded minimum with the treatment Microalgae + cow dung+ poultry manure + citral (13.03%) followed by fungicide Carbendazim (13.16%). The highest yield was recorded with the treatment microalgae + poultry manure + cow dung+ citral (1.47 kg/plot). The highest total soluble solid content was recorded with the treatment Microalgae + poultry manure + cow dung+ citral (13.86 °Brix) followed by poultry manure + citral (13.30 °Brix). Results revealed that bio resources and citral essential oil compound may be used as eco-friendly natural compounds to reduce the cercospora leaf spot of beetroot.

### Keywords

Cercospora, Citral,  
Bio resources, Root  
weight, Sugar  
content

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### Introduction

Beetroot (*Beta vulgaris* L.) also known as table beet, garden beet and sugar beet it is one of the major root vegetable. Belongs to the family Chenopodiaceae. It produces green

tops and swollen roots both used as a vegetable. It is a rich source of carbohydrate, calcium and vitamin C (Deuter and Grundy, 2004). Red color of roots is due to the presence of betanine pigment.

Beetroot is a versatile crop it can withstand various climatic conditions it is grown as a cool season annual crop. Russia is the top leading producer of beetroot in world. India ranks 45th in export of salad beetroot. Food and agricultural organization indicate that 30% of beets accounts for sugar production. In India beetroot is mainly cultivated in Haryana, Uttar Pradesh, Himachal Pradesh, West Bengal, Maharashtra and Tamilnadu. The total cultivable area of beetroot is 1116 hectares in Tamilnadu (Pirabu, 2014).

Beetroot crop is affected by number of fungal, bacterial, and viral diseases. The losses accounts up to 45-50% where beetroot is grown. Among the fungal diseases cercospora leaf spot caused by *Cercospora beticola* is the most destructive foliar disease. Losses due to cercospora leaf spot in root quality is reduced up to 32% and the gross sugar quality up to 42% (Shane and Teng, 1992). It mainly effects on leaf, the symptoms appeared as circular to oval shaped with ash coloured center surrounded by reddish margin. As the spots enlarge it becomes papery at the center these spots colecesce and lead to death (Sakariss *et al.*, 2010). The pathogen is able to survive for at least one year in plant debris and soil. Primarily their spores are dispersed by wind and is favored by prolong rainfall, high relative humidity and temperature up to 25°C to 35°C.

Controlling this disease mainly depends on chemical fungicides. Avoiding environmental pollution fungicide alternative applications are needed. One such attempt is the use of bio resources such as cow dung, poultry manure, microalgae and citral essential oil compounds aims in creating healthy soil, reduction in disease severity and sustaining considerable levels of yield. These organic manures are the rich source of micro and macro nutrients that regulates the availability of nutrients to plants and acts as a chelating agent (Lampkin,

1990). Poultry is the rich source of nitrogen as it increases the root yield. Microalgae act as bio fertilizer and bio stimulant that stimulate the plant growth promoting hormones (Manickavelu *et al.*, 2006). Essential oils contain anti-fungal properties that suppress the growth of the fungus. It is suggested that constituents of essential oils can be used as ecofriendly natural compounds for controlling cercospora leaf spot disease of beetroot (Abdel-kareem, 2002). Lemon grass (*Cymbopogon citratus*) essential oil contains several components including citral, geranial, neral which may have antifungal properties (Shaw, 1979). Among all the essential oil compounds citral appears to be most effective in inhibiting pathogens (Wannison *et al.*, 1996).

The present investigation was carried out to evaluate the effect of bio resources and citral essential oil compound against cercospora leaf spot and yield characters of beetroot in field conditions. Also to evaluate the effect of citral essential oil compound against radial growth of *Cercospora beticola* in *in vitro*.

## **Materials and Methods**

### **Isolation and identification of the pathogen**

The disease sample showing cercospora leaf spot symptoms in beetroot were collected from the central research field, SHUATS, Prayagraj and Uttar Pradesh. The isolation of the fungus was done using standard tissue isolation technique. Infected leaves of beetroot were cut into small bits and surface sterilized with sodium hypochlorite for 2 minutes then washed several times in distilled water and together blotted in a filter paper then transferred into a petri plate containing beetroot leaf extract agar media under aseptic condition. Beetroot leaf extract agar media was prepared by cutting fresh leaves of beetroot (200g) and boiled it in one liter of distilled water for 15 minutes. The extract

obtained was strained through a muslin cloth. Twenty grams of dextrose and 15 g of agar-agar were added to the mixture and transferred to a conical flask. The petriplates were incubated at 27± 2 °C for 3-6 days to observe the growth of the fungus. The growing fungus was examined under the microscope. Hyphal tip technique was done to purify the culture as described by (Korhonen *et al.*, 1980). Once the growth observed it is sub cultured and pure culture is maintained at 4°C for further use.

### **In-vitro evaluation of citral essential oil compound against *Cercospora beticola***

Different concentrations viz., 0.5%, 0.75%, 1%, 1.25% of citral essential oil compound and Carbendazim @0.01% were tested for their antifungal activity against mycelial growth of *Cercospora beticola* by poison food technique in *in-vitro*. The different concentrations of citral essential oil compound was added into each conical flask containing potato dextrose agar in aseptic condition and poured into a petri plate and inoculated with a 5mm disc of 7 days old culture. The petri plates was incubated at a temperature of 27±2°C and the inhibitory effect of citral essential oil against radial mycelial growth of *Cercospora beticola* was recorded at every 24hrs interval. The growth of the mycelial growth and percent inhibition was calculated using the formula (Vincent, 1947).

Where:

$$I = \frac{C - T}{C} \times 100$$

I= Inhibition percentage (%)

C= Radial growth in control plot T= Radial growth in treatment plot

### **Evaluation of bio resources and citral essential oil compound in field**

A field experiment was done at central research field, SHUATS, Prayagraj, and Uttar Pradesh during Rabi 2019-2020. The experiment was laid out in a Randomized block design with three replications and eight treatments. The bio resources such as cow dung, poultry manure, microalgae, citral essential oil compound and Carbendazim with combinations i.e., T1 Microalgae + citral, T2 Cow dung + citral, T3 Poultry manure + citral, T4 Microalgae + poultry manure + citral, T5 microalgae + cow dung + citral, T6 Microalgae + poultry manure + cow dung + citral, T7 Carbendazim @ 0.1%, T8 control were evaluated against *Cercospora* leaf spot of beetroot. The cow dung, poultry manure were applied before sowing of seeds. The application of microalgae spraying of citral essential oil compound and Carbendazim was done immediately after the appearance of symptoms i.e., 30 days after sowing (DAS) followed by two more sprays at 15 days interval. The observations were recorded before each spray and the disease severity was calculated. The disease severity or percent disease incidence was determined using disease rating scale Where, 0= No symptoms, 1= 1-10% Disease infection, 2= 11-25% Disease infection, 3= 26-50% Disease infection, 4= 51-70% Disease infection, 5= >71% Disease infection (Mayee and Datar, 1986). The percent disease control was also calculated.

$$PDI = \frac{\text{Sum of all disease ratings}}{\text{No. of observations assessed} \times \text{maximum disease rate}} \times 100$$

PDI increase or decrease over control (%) =

$$\frac{\text{Disease in control plot} - \text{Disease in treatment plot}}{\text{Disease in control plot}} \times 100$$

### Determination of root yield

Root weight was calculated after harvesting. The roots from each treatment plots were harvested separately and weighed by weighing machine and expressed as kg/plot.

### Evaluation of total soluble solids (°Brix)

Determination of total soluble solids of beetroot is done at the time of harvest. For each treatment total soluble solids was recorded using hand refractometer.

## Results and Discussion

### Effect of citral essential oil compound against radial mycelial growth of *Cercospora beticola*

The antifungal effect of citral essential oil compound at different concentrations was tested against *Cercospora beticola* using poisoned food technique. The radial growth of the fungus was recorded at every 24hrs interval. Radial growth and inhibition percentage of the pathogen varied significantly at different concentration i.e., 0.5%, 0.75%, 1%, 1.25% and Carbendazim @

0.01% as treated check compared to control. Citral @ 1.25% showed the highest inhibition in radial growth (96.52%) followed by citral at 1% (88.24%), citral at 0.5% (69.35%).

The Carbendazim at 0.01% (50.13%) and citral at 0.75% (38.70%) showed significantly less inhibition of mycelial growth compared to control (Table 1).

### Effect of bio resources and citral essential oil compound against cercospora leaf spot of beetroot

Results revealed that all the treatments were significantly reduced the disease severity of cercospora leaf spot in beetroot compared to control. The highest reduction was recorded significantly with the treatment Microalgae + poultry manure + cow dung+ citral (13.03%) followed by Carbendazim (13.16%) with per cent disease control (65.94%) and (65.60%). Least reduction was recorded in treatment microalgae + citral (17.03%) followed by microalgae + cow dung + citral (15.60) with minimum per cent disease control (55.48%) and (59.22%) compared to control respectively (Table 2).

**Table.1** Effect of citral oil on radial growth of *Cercospora beticola* at 48, 72, 96 and 120 hrs

S.No	Treatments	Radial growth (mm)				PDI (%)
		48hrs	72hrs	96hrs	120hrs	
T1	Citral @ 0.5%	10.87	16.40	20.97	24.66	<b>38.70</b>
T2	Citral @ 0.75%	6.13	9.10	10.90	12.33	<b>69.35</b>
T3	Citral @ 1%	2.47	3.13	3.77	4.73	<b>88.24</b>
T4	Citral @ 1.25%	1.17	1.27	1.30	1.40	<b>96.52</b>
T5	Carbendazim @ 0.01%	9.53	14.87	18.53	20.06	<b>50.13</b>
T6	Control	22.07	28.07	34.87	40.23	<b>0</b>
	F test	S	S	S	S	
	SEd ±	0.39	0.40	0.44	0.52	
	<b>C.D.(P=0.05)</b>	<b>1.27</b>	<b>1.30</b>	<b>1.42</b>	<b>1.63</b>	

**Table.2** Effect of bio resources and foliar spray of citral oil on disease severity of cercospora leaf spot and yield parameters in beetroot

T.No	Treatments	Before spray 30DAS	After 1 <sup>st</sup> spray 45DAS	After 2 <sup>nd</sup> spray 60DAS	After 3 <sup>rd</sup> spray 75DAS	Avg PDI	Root yield (Kg/pl ot)	TSS (°Brix)
<b>T1</b>	Microalgae + citral	6.433	10.633	15.36	17.03	55.48	1.11	<b>11.98</b>
<b>T2</b>	Cow dung + citral	6.067	9.700	13.93	15.53	59.40	1.19	<b>12.04</b>
<b>T3</b>	Poultry manure + citral	5.700	8.833	13.50	14.20	62.88	1.45	<b>13.30</b>
<b>T4</b>	Microalgae + poultry manure + citral	5.267	8.433	13.06	14.26	62.72	1.47	<b>13.21</b>
<b>T5</b>	Microalgae + cow dung + citral	6.200	8.867	14.70	15.60	59.22	1.30	<b>12.80</b>
<b>T6</b>	Microalgae + poultry manure + cow dung + citral	4.900	8.233	12.20	13.03	65.94	1.98	<b>13.86</b>
<b>T7</b>	Carbendazim	6.167	8.433	12.33	13.16	65.60	1.23	<b>12.14</b>
<b>T8</b>	Control	13.800	19.500	27.16	38.26	0	0.76	<b>11.15</b>
	F Test	S	S	S	S		S	<b>S</b>
	SE(m)±	0.36	0.52	0.58	0.68		0.10	<b>0.41</b>
	<b>CD at 5%</b>	<b>1.21</b>	<b>1.59</b>	<b>1.80</b>	<b>2.10</b>		<b>0.305</b>	<b>0.882</b>

### **Effect of bio resources and citral essential oil compound on beetroot yield**

All the treatments were significantly increased the beetroot yield when compared to control. Highest yield was recorded with the treatment Microalgae + poultry manure + cow dung+ citral (1.96 kg/plot) followed by microalgae + poultry manure + citral (1.47 kg/plot). The lowest yield was recorded with the treatment cow dung + citral (1.23 kg/plot) followed by microalgae + citral (1.11 kg/plot) compared to control (Table 2).

### **Effect of bio resources and citral essential oil compound on total soluble solids of beetroot**

Results showed that all the treatments significantly increased the total soluble solids in beetroot. The highest TSS content was recorded with the treatment Microalgae + poultry manure + cow dung + citral (13.86 °Brix) followed by poultry manure + citral (13.30 °Brix). Lowest TSS was recorded with the treatment cow dung + citral (12.04 °Brix) followed by Microalgae + citral (11.98 °Brix) compared to the control respectively (Table 2).

Beetroot is the root crop that is used as salad and vegetable. It is a versatile crop can withstand in any season. It grows well in cool season annual crop and attains best texture and quality. Beetroot roots contain 15-20% sucrose, beets accounts for 30% sugar production.

*Cercospora* leaf spot caused by *Cercospora beticola* is the major foliar disease in beetroot that decreases the root and sugar quality (Shane and Teng, 1992). The present investigation revealed that bio resources and citral essential oil compounds decreased the disease severity and increased the root and sugar quality. Results in *in-vitro* showed that

citral essential oil compound at 1.25% reduced the radial mycelial growth of *Cercospora beticola* compared to the other treatments.

Similar findings were reported by Krishna *et al.*, (2007) stated that *Cymbopogon citratus* (lemon grass oil) showed the highest inhibition compared to other essential oils. The inhibitory effect of citral oil on several fungi was also reported by Asthana *et al.*, (1988). Omer Fatouh *et al.*, (2011) reported that higher concentration of citral essential oil completely inhibited the mycelial growth of *Cercospora beticola*.

Application of bio resources in field conditions reduced the disease severity and increased the root and sugar quality of beetroot. All the treatments significantly reduced the disease severity compared to control. Highest reduction in disease severity recorded with the treatment Microalgae + poultry manure + cow dung + citral (13.03%) followed by Carbendazim (13.16%). The similar findings were reported by (French *et al.*, 1978) stated that citral essential oil compound was found more toxic against the fungi. (Krishna *et al.*, 2007) stated that citral essential oil showed the highest inhibition effect against *Cercospora arachidicola* compared to other essential oils. The increase in root weight may be due to the decrease in disease incidence. The above findings were confirmed with (Shane and Teng, 1992) he stated that highest disease incidence affects the root weight in beetroot. Insufficient nitrogen decreases the root yield higher nitrogen has a significant role to play reported by (Carter 1987, Halverson and Hartman, 1988). The reduction in disease severity of cercospora could significantly increase the sugar yield. The above recorded data was confirmed with the findings of Omer Fatouh *et al.*, (2011) in sugar beet.

In conclusion, soil application of bio resources and foliar application of citral essential oil compound and Carbendazim significantly reduced the cercospora leaf spot disease and increasing the root quality and sugar content of beetroot compared to control. This is one such attempt to reduce the environmental stress by avoiding the heavy use of chemicals to control diseases. Future line of work is needed with bio resources and plant based products for the management of cercospora leaf spot.

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