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Effect of root exudates on growth of polyphagous *Sclerotium rolfsii*

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Abstract
Twenty plants were selected and harvested their root exudates to study their antifungal potency. The results of the study showed that root exudates had marked influence on mycelial growth, colony diameter and sclerotia formation of *S. rolfsii*. Neem root exudate had maximum inhibitory effect on mycelial growth of *S. rolfsii* (58.82 %). This was followed by mango and *shatavari* root exudates which showed 54.89 per cent inhibition in each. There were no sclerotia formation on neem, mango, *shatavari*, sorghum, coconut, periwinkle and parthenium root exudates even after 15 days of inoculation. However, datura (-1.95 %), tamarind (-0.98 %) root exudates showed minimum per cent inhibition over control with profuse amount of sclerotia formation. It means datura and tamarind root exudates stimulate growth of *S. rolfsii.* Among the total samples, five promising plant root exudates *viz.* Neem, Sorghum, Mango, Coconut, *Shatavari* were selected for evaluating effect on sclerotia germination in different conc. (at 10, 20 and 50%). At 10% and 20% concentration neem root exudates whereas at 50% concentration mango root exudate found effective against *S. rolfsii*.

Keywords: *Sclerotium rolfsii*, root exudates, growth, sclerotia

Introduction
Sclerotium rolfsii is a well-known soilborne fungus that is polyphagous, ubiquitous, facultative and most destructive. It causes root rot, stem rot, collar rot, wilt and foot rot in several plants.
According to Aycock (1996), the host range of *S. rolfsii* is very broad, encompassing several horticultural and agronomical crops all over the world. It attacks over 500 species of plants from over 100 families.
Diseases caused by soilborne plant pathogens are difficult to control for several reasons. Many soilborne pathogens produce persistent resting structures that can survive in the soil for many years even in the absence of a susceptible host. Increased problem with side effects of chemical residue in soil as well as increased in resistance to fungicides has prompted the research focus on biological control in past few decades.
Root exudates mediate both positive and negative interactions in the rhizosphere. Several compounds present in root exudates play significant roles in biological processes. The plant defences induced by root exudates simply reduce susceptibility to pathogen infection, whereas in other cases these defences initiate production and release of leafy volatiles that attract predators of plant enemies. Considerable differences have been observed in the effects of root exudates on spore germination and mycelial growth; these effects vary from general stimulation, specific growth stimulation, to inhibition.
Rosmarinic acid was recently discovered in the root exudate of hairy root cultures of sweet basil provoked by *Phytophthora cinnamoni* fungal cell wall extracts (Bais *et al*., 2006) [6]. Brigham *et al*. (1999) [8] revealed cell specific production of pigmented naphthoquinones in *Lithospermum erythrorhizon*; these findings suggest the role of root exudates in defending the rhizosphere against pathogenic microorganisms.
The present investigation carried out on effect of root exudates on growth of polyphagous *Sclerotium rolfsii* with the objectives to screen for the antifungal potency of certain plants root exudates to manage *S. rolfsii.*

Materials and Method
Collection of Selected species
The study was conducted in laboratory (College of Agriculture, Pune) to know the effect of root exudates of various plants on mycelium growth and sclerotia formation which may

provide some clues for using these crops in crop rotation or intercropping. About 20 plants *viz.*, Neem, Sorghum, *Takla*, *Kaner*, Tamarind, *Ghaneri*, Periwinkle, Castor, *Landga*, Marigold, Mexican poppy, Parthenium, Datura, Cabbage, Tomato, Guava, *Gulvel*, Mango, Coconut and *Shatavari* were selected on the basis of their antimicrobial properties with the references of previous research articles.

Harvesting the root exudates from selected species of plants

The roots of selected plant species were collected and thoroughly washed with water to remove any dirt or soil particles. Then left for exudation for 24 hrs in sterilized conical flask containing sterile distilled water such that, each flask received 10 gm root with 100 ml distilled water. After 24 hrs, obtained root exudates were filtered with Whatman filter paper No.41. The exudates thus collected were assumed 100 % concentration and kept at 4 ˚C for further study.

Testing of root exudates on growth of *S. rolfsii in vitro*

The effect of root exudates on mycelial growth and sclerotia formation of *S. rolfsii* were evaluated by poison food technique along with distilled water as control. Firstly, soaked sclerotia in each root exudates having 100 % concentration and left as such overnight. After 24 hrs, transferred those sclerotia on PDA plates separately. The sclerotia soaked in sterile water served as control. The petri plates were incubated at 27±2 ˚C. The per cent inhibition of mycelial growth and sclerotia formation over control was derived.

Out of 20 samples, five plant root exudates *viz.* neem, sorghum, mango, coconut, *shatavari* were selected for evaluating effects on sclerotia germination in different concentrations. These five exudates were diluted at 10, 20 and 50% concentrations. In each concentration sclerotia were soaked for 24 hrs. Later they were transferred aseptically on sterilized petri plates containing PDA medium. The plates were incubated at 27±2 ˚C. and daily observations recorded.

Result and Discussion

The results of the study showed that exudates from the roots of plants had marked influence on mycelial growth, colony diameter and sclerotia formation of *S. rolfsii*. It is observed that root exudates of few plants had suppressed growth as well as formation of sclerotia. It is interesting that few plants' root exudates had supported the growth of *S. rolfsii.*

Root exudates of neem showed minimum mean colony diameter (28.00 mm) and statistically superior over rest of attempted treatments. After neem, colony diameter under mango and *shatavari* root exudates was observed 30.67 mm each. The colony diameter in case of sorghum root exudate was 38.00 mm.

The mycelium growth of *S. rolfsii* was high when root exudates from datura (69.33 mm), tamarind (68.67 mm) and tomato (68.00 mm) were tested as compared to the control (68.00 mm). The colony diameter of *S. rolfsii* under the treatments of remaining root exudates from other plants was lesser than control as, coconut (38.00 mm), periwinkle (43.30 mm), parthenium (44.67 mm), marigold (46.67 mm), castor (48.00 mm), mexican poppy (48.67 mm), cabbage (52.00 mm), *kaner* (53.30 mm), *gulvel* (53.33 mm), *ghaneri* (56.67 mm), guava (62.00 mm), *takla* (62.00 mm), *landga* (66.67 mm).

It is observed that, neem had maximum inhibitory effect on mycelial growth of *S. rolfsii* with 58.82 per cent inhibition over control and no sclerotia were formed. Mango and *shatavari* root exudates also suppressed the growth of *S. rolfsii* (54.89% inhibition) each. Sorghum and coconut root exudates showed 44.11 per cent inhibition over control with no sclerotia structure formation even after 15 days of inoculation.

In case of control as well as root exudates of datura (-1.95 % inhibition) and tamarind (-0.98 % inhibition) had produced profuse amount of sclerotia. It means that datura and tamarind root exudates boosted the growth and sclerotia formation of *S. rolfsii*. Tomato root exudate showed 0.00 per cent inhibition of mycelial growth of *S. rolfsii*.

In case of periwinkle and parthenium, per cent inhibitions were 36.32% and 34.30%, respectively. However, there was no sclerotia formation even after 15 days of inoculation. Root exudates of castor (29.41%), marigold (31.36%), mexican poppy (28.42%), *kaner* (21.61%) resulted mycelium inhibition with least amount of sclerotia formation.

Various studies have shown the effect of exudates from roots of plants on the germination of fungal propagules (Rovira, 1965; Eze, 1990; Odunfa, 2006) [13, 10, 11]. Rovira had noted earlier that root exudates possess commonly occurring components such as amino acids, inorganic acids and sugars that vary significantly among plant species. These compounds can have a stimulatory or inhibitory influence on fungal propagule germination and growth. According to Odunfa (2006) [11], *S. rolfsii* requires particular substances for growth and is thus host specific. Rao *et al.* (2002) had also reported that sorghum root exudates were the most effective in inhibiting mycelial growth and sclerotia germination of *S. rolfsii*.

Table 1: Effect of root exudates on growth of *S. rolfsii*

Tr. no.	Treatment	Colony diameter (5 DAI in mm)	Per cent inhibition over control	Structure formation (15 DAI)
T1	Neem	28.00	58.82	Nil
T2	*Takla*	62.00	8.82	Poor
T3	Sorghum	38.00	44.11	Nil
T4	*Ghaneri*	56.67	16.66	Fair
T5	*Kaner*	53.30	21.61	Poor
T6	Tamrind	68.67	-0.98	Profuse
T7	Periwinkle	43.30	36.32	Nil
T8	Castor	48.00	29.41	Poor
T9	Marigold	46.67	31.36	Poor
T10	*Landga*	66.67	1.95	Fair
T11	Mexican poppy	48.67	28.42	Poor
T12	Parthenium	44.67	34.30	Nil
T13	Cabbage	52.00	23.52	Fair
T14	Tomato	68.00	0.00	Profuse
T15	Datura	69.33	-1.95	Profuse
T16	Guava	62.00	8.82	Fair
T17	*Gulvel*	53.33	21.61	Poor
T18	Mango	30.67	54.89	Nil
T19	Coconut	38.00	44.11	Nil
T20	*Shatavari*	30.67	54.89	Nil
T21	Control	68.00	0.00	Profuse
SE (m)±±		0.32		
CD (0.01)		1.23		

Plate 1: Effect of root exudates in *S. rolfsii*

The data presented in Table 2 depicts, comparison among three concentrations 10, 20 and 50% of five promising plant root exudates. In each case, as concentration increased the inhibition of fungal growth also showed increasing trends. In case of neem root exudates at 10 % concentration inhibition was 21.32%, at 20% concentration inhibition raised to 33.08 %. However, at 50% concentration it was maximum 48.52% inhibition.

At 10 % concentration, treatment of neem root exudate was most effective and this was followed by mango, sorghum, *shatavari* and last coconut. At 20% concentration, neem had inhibitory action against *S. rolfsii* followed by *shatavari*, sorghum. However, at 50% conc., treatment of mango root exudate was statistically significant over other tests followed by neem, *shatavari*, sorghum.

Table 2: Effect of different concentration on growth of *S. rolfsii*.

Tr. no.	Treatment	Per cent inhibition over control		
		10%	**20%**	**50%**
T1	Neem	21.32	33.08	48.52
T2	Sorghum	9.55	29.41	44.11
T3	Mango	16.91	27.94	53.67
T4	Coconut	5.88	17.64	26.47
T5	*Shatavari*	8.08	30.88	45.58

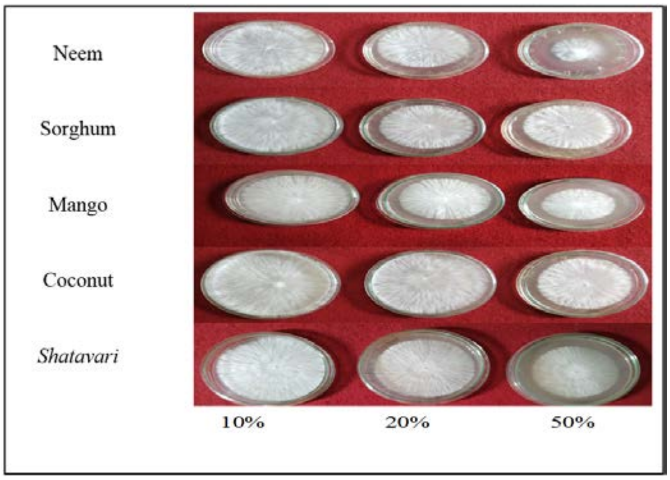

Plate 2: Effect of different conc. of promising root exudates on growth of *S. rolfsii*

Conclusion

Among twenty plant types, root exudate of neem was found effective against growth of *S. rolfsii.* This was followed by mango, *shatavari* and sorghum.

Five promising plants root exudates were tested at different concentrations, at 10 and 20% concentration neem root exudates whereas at 50% concentration mango root exudate found effective against *S. rolfsii*.

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