

In-vitro Screening of Botanicals against *Bipolaris sorokiniana* (Sacc.) Shoemaker of Wheat

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Abstract: *Bipolaris sorokiniana* (Sacc.) Shoemaker is an important fungal pathogen affecting the production of wheat (*Triticum aestivum* L.). Conventional management strategies using chemicals are expensive as well as hazardous. This has necessitated the search for alternatives in botanicals. In-vitro studies were conducted to evaluate the efficacy of aqueous extracts of different plants against foliar blight of wheat. Nine different plant materials viz, garlic (*Allium sativum* L.), periwinkle (*Vinca rosea* L.), ginger (*Zingiber officinale* L.), neem (*Azadirachta indica* L.), milkweed (*Calotropis gigantea* L.), datura (*Datura metel* L.), onion (*Allium cepa* L.), marigold (*Tagetes erecta* L.) and tulsi (*Ocimum sanctum* L.) were tested at five aqueous concentrations viz., 0.5, 1, 2, 2.5 and 5%. Garlic bulb extract was found superior among all the test botanicals followed by periwinkle and onion.

Keywords: Wheat, foliar blight, *Bipolaris sorokiniana* and botanicals

INTRODUCTION

Wheat (*Triticum aestivum* L.) is counted among the 'big three' cereal crops. A number of abiotic as well as biotic stresses pose serious threat to successful cultivation of this crop. Among the biotic stresses, insect pests and diseases are the major limiting factors. Diseases can be due to bacteria, fungi, viruses, nematodes or a complex of them. The fungal pathogen *Bipolaris sorokiniana* (Sacc.) Shoemaker is the causal agent of various diseases like head blight, seedling blight, foliar blight / spot blotch, common root rot and black point of wheat, barley, other small cereal grains and grasses (Zillinsky, 1983; Wiese, 1998). Among all the diseases caused by this pathogen, foliar blight of wheat is considered as one of the most important diseases. Substantial economic loss in wheat production has occurred due to the severity of foliar blight, affecting the livelihood of millions of small farmers in warm and humid regions of South Asia. Indian subcontinent has 10 million ha of affected land, out of which India alone has 9 million ha, most of which is in the rice - wheat cropping system (Nagarajan and Kumar, 1998). The injudicious and indiscriminate use of chemical fungicides in the crop

causes phytotoxicity, health hazards and increase in the selection pressure of the pathogen. Hence the study was undertaken to screen the efficacy of different plant products *in vitro* keeping in view their utility in formulating an eco-friendly management strategy in field.

MATERIAL AND METHODS

The experiment was conducted at the Department of Mycology and Plant Pathology, Banaras Hindu University, Varanasi in a completely randomized design. Diseased wheat leaf samples showing the typical blight symptoms were collected from the Agriculture Research Farm, and brought to the laboratory. The symptomatic leaf samples were surface sterilized with 1% sodium hypochlorite. These leaves were cut into small pieces (1-2cm) having lesions with the help of sterilized scissors. The leaf bits were transferred into a sterilized moist chamber and incubated at 25±2°C. The moist chamber was prepared by placing blotting paper in a 9 cm diameter sterilized Petri-plate, moistening it with sterile distilled water and autoclaving it at 121°C. The spore formation took place on the third day of incubation

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(De Wolf *et al.*, 1998). After observing conidial crops on the transferred leaf bits under a stereoscopic binocular microscope, a single conidium from each leaf bit was transferred separately by a fine needle into Petri-dishes poured with Potato Dextrose Agar medium (Peeled potato: 200g, Dextrose: 20g, Agar-Agar: 20g, Distilled water: 1000ml) acidified with 4-5 drops of 25% lactic acid. Nine different plant materials were collected and their stock solutions were prepared in sterile distilled water (1:1) by crushing them using mortar and pestle and filtering the extract through muslin cloth. Five different aqueous concentrations i.e., 0.5, 1.0, 2.0, 2.5 and 5.0% of each botanical were prepared from the stock solution by subsequent dilutions. In order to prepare spore suspension, 200 micro litres (μ l) of sterile distilled

water was taken in a clean sterilized cavity block and some spores of *B. sorokiniana* were harvested with the help of a fine acrylic brush from 7 day old culture. And the volume of the water was adjusted in such a fashion to have 200-300 spores in 5 μ l. The spore suspension was homogenised by using a micro pipette. Five micro litre of the spore suspension was poured into the cavity slides already containing 65 μ l of different concentrations (0.5, 1.0, 2.0, 2.5 and 5.0%) of each of the test botanicals. The sterile distilled water without botanicals served as control. The cavity slides were placed in moist chambers and incubated at $25\pm 1^\circ\text{C}$. After 24 hours, the germination of spores was observed under a microscope and the per cent inhibition of spore germination was computed using the following formula:

$$\text{Inhibition \% of conidial germination} = \frac{\text{Total no. of conidia} - \text{No. of germinated conidia}}{\text{Total no. of conidia}}$$

Sl.No	Common Name	Botanical Name	Family	Parts used
1	Garlic	(<i>Allium sativa</i> L.)	Amaryllidaceae	Clove
2	Periwinkle	(<i>Vinca rosea</i> L.)	Apocynaceae	Leaves
3	Ginger	(<i>Zingiber officinale</i> L.)	Zingiberaceae	Rhizome
4	Neem	(<i>Azadirachta indica</i> L.)	Meliaceae	Leaves
5	Milkweed	(<i>Calotropis gigantea</i> L.)	Apocynaceae	Leaves
6	Datura	(<i>Datura metel</i> L.)	Solanaceae	Leaves
7	Onion	(<i>Allium cepa</i> L.)	Amaryllidaceae	Bulb
8	Marigold	(<i>Tagetes erecta</i> L.)	Asteraceae	Leaves
9	Tulsi	(<i>Ocimum sanctum</i> L.)	Lamiaceae	Leaves

RESULTS AND DISCUSSION

A perusal of data on the effect of the test botanicals at different concentrations i.e., 0.5, 1.0, 2.0, 2.5 and 5.0 per cent showed varying effect on the spore

germination of *B. sorokiniana* (Table 1). The spore germination was noticed to be reduced with increase in the concentration irrespective of the botanicals over control. Out of the nine test botanicals, garlic clove

Table 1
Effect of botanicals on spore germination of *Bipolaris sorokiniana* in vitro

Sl.No.	Botanicals	% spore germination after 24 hours				
		Concentrations				
		0.5	1.0	2.0	2.5	5.0
1	Garlic	51.2(45.7)	11.0(19.4)	4.4(12.1)	3.2(10.3)	0.0(0.0)
2	Periwinkle	73.7(59.1)	65.4(54.0)	57.8(49.5)	51.0(45.6)	36.9(37.4)
3	Ginger	75.1(60.1)	67.6(55.3)	61.3(51.6)	55.6(48.2)	50.1(45.1)
4	Neem	81.0(64.2)	75.2(60.1)	65.0(53.7)	62.4(52.2)	55.1(47.9)
5	Milkweed	81.1(64.2)	80.4(63.7)	75.0(60.0)	70.3(57.0)	69.3(56.4)
6	Datura	86.8(68.7)	85.9(67.9)	77.7(61.8)	77.2(61.5)	76.6(61.0)
7	Onion	90.0(71.6)	80.0(63.4)	60.0(50.8)	55.0(47.9)	50.0(45.0)
8	Marigold	90.0(71.6)	85.0(67.2)	85.0(67.2)	85.0(67.2)	85.0(67.2)
9	Tulsi	90.0(71.6)	90.0(71.6)	90.0(71.6)	90.0(71.6)	90.0(71.6)
10	Control*	100.0(90.0)	100.0(90.0)	100.0(90.0)	100.0(90.0)	100.0(90.0)

*Sterile distilled water used as control

CD(p=0.05)

Botanicals: 0.578 Concentrations: 0.431 Botanicals \times Concentrations: 0.292

Figures in parentheses represent arcsine transformed values.

extract showed complete inhibition of spore germination followed by periwinkle, onion and ginger at 5.0 per cent concentration. It was also noticed that the test botanicals showed significant increase in spore germination up to 2.0 per cent concentration. Among crude extracts of onion, neem, milkweed, datura, marigold and tulsi, onion extract was found to be superior to the others in its effect on the spore germination of *B. sorokiniana*. The efficacy of the garlic may be attributed to thiosulfinates, particularly allicin. Slusarenko *et al.* (2008) reported the effectiveness of garlic juice against a range of plant pathogenic bacteria, fungi and oomycetes *in-vitro* and reported that allicin has effectively controlled seed borne *Alternaria* spp. in carrot. Bach *et al.* (2014) reported that use of garlic extract induces systemic resistance in barley plants against *B. sorokiniana*. The exploitation of plant products for the management of plant diseases have achieved greater significance in recent times due to its readily available nature, easy biodegradability, non-phytotoxicity, besides inducing resistance in host.

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