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Comparative study on the extracts of *Bryum argenteum* and synthetic fungicide against some common diseases of *Zea mays*

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Manuscript details:

Received : 25.10.2017 Accepted : 21.02.2018 Published : 14.03.2018

Editor: Dr. Arvind Chavhan

Cite this article as:

Deora GS and Guhil N (2018) Comparative study on the extracts of *Bryum argenteum* and synthetic fungicide against some common diseases of maize, *Int. J. of. Life Sciences*, Volume 6(1): 217-221.

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Available online on <u>http://www.ijlsci.in</u>

ISSN: 2320-964X (Online) ISSN: 2320-7817 (Print)

ABSTRACT

The aim of the present study was to evaluate the antifungal activity of extracts of *Bryum argenteum* against the phytopathogenic fungus *Curvularia lunata*, the causual organism of maydis leaf blight disease in *Zea mays* using pour plate method. Antifungal activities of plant extract was compared with commercially available fungicides. The minimum inhibitory concentration and per cent inhibition was determined. A phytochemical screening of the extracts was also carried out to determine the antifungal substances. It was observed that the ethanolic extract of plant had a broad spectrum antifungal activity against the test fungi.

Keywords: Phytopathogenic fungi, *Bryum argenteum, Zea mays, Curvularia lunata*.

INTRODUCTION

Plant diseases create challenging problems in commercial agriculture and pose real economic threats to both conventional and organic farming systems. Synthetic fungicides bring about the inhibition of pathogens by either destroying their cell membrane either its permeability or by inhibiting metabolic processes of the pathogens and hence are extremely effective but the flip side of chemical fungicides are known to pollute the environment, soil and water besides causing deleterious effects on human health and biosphere. Hence there is a need to search for an environmentally safe and economically viable strategy for the control of diseases and to reduce the dependence on the synthetic agrochemicals.

Pates and Madsen (1995) made a study on eight bryophytes of which four possessed antimicrobial substances. Two were active against fungal species while two against bacteria.

Bodade *et al.*, (2008) evaluated the antimicrobial effect of *Plagiochasma appendiculatum*, *Thuidium cymbifolium*, *Bryum cellulare*, *Bryum argentium*

and *Racomitrium crispulum* on 12 microorganism. Solubility data & antibiotic spectra of the active plants indicated the occurrence of the variety of antibiotic substances among bryophytes.

Deora and Bhati (2007) studied on antibacterial effect of aqueous crude extract of *Plagiochasma articulatum*, *Anthoceros longi* and *Fissidens bryoides* against test organism *Xanthomonas citri in vitro*. Deora and Suhalka (2010) studied the effect of liverwort *R. gangetica* against *F. moniliforme* and found cold water extract was more effective than boiled water extract. Deora *et al.*(2010) determined the antifungal activity of a moss against certain phytopathogenic fungi.

The aim of the present study was to find out the antifungal activity of *Bryum argenteum* extracts against phytopathogenic fungi *Curvularia lunata* and comparing its per cent inhibition with that of commercially available fungicide Mancozeb. Phytochemical screening of different extract will also carried out to detect the presence of secondary metabolites.

MATERIALS AND METHODS

Plant material and extract preparation

The moss Bryum argenteum was collected in rainy season (2013) from Mt. Abu, District Sirohi (Raj.) around Nakki Lake, Guru Shikhar and Sunset point in both vegetative and sporophytic phases. Both plants were washed with distilled water to remove soil particles, attached litter and dead material. For ethanolic extract preparation, plant material weighted was grinded in mortar and pestle with equal amount of ethanol till the formation of fine paste and left for overnight then it was centrifuged and filtered. This filtrate was dried and dry residue (1gm/10ml) was used as (100%) crude extract which was serially diluted by double distilled water to prepare various concentrations from 10-100 per cent. The same method was adopted for acetonic extract preparation except grinding the plant material with acetone instead of ethanol.

Test Organism

The pure culture of test fungi *Curvularia lunata* (MTCC No.283) was obtained from the Institute of Microbial Technology (IMTECH), Chandigarh (India). This test organism was sub-cultured in laboratory at 30°C temperature to obtain its pure isolates.

Screening of Antifungal Activity

Antifungal activity of bryophyte fraction was determined by using pour plate method. The plant extracts of 3 ml was first poured into petri dishes. Then, 12 ml molten PDA was poured aseptically on the plant extract in the petri dishes and swirled round for even dispersion of the extract into the agar. The extracts were incorporated at different concentrations of 100, 80, 60, 40, 20 and control. A 5 mm mycelium agar disc of C. lunata was released into the poisoned agar/ extracts incorporated into PDA. The treatments were replicated three times, incubation period for antifungal activity was 72 hrs. The average diameter of resultant colony was measured after incubation. The growth of C. lunata mycelium on PDA without any amendment was used as control. The percent inhibition of mycelial growth by plant extract was calculated by using the formula given by Vincent (1927).

Phytochemical Analysis

Qualitative phytochemical analysis of a moss *Bryum argenteum* extract was done by the methods of Trease and Evans [2002] to detect the presence or absence of certain bioactive compounds.

RESULTS AND DISCUSSION

Antifungal activity of bryophyte extracts in acetoic and ethanolic solvent on *Curvularia lunata* are represented in Table 2&3. Although both the extracts of *Bryum agrenteum* showed varying levels of antifungal activity against the test fungi, the ethanolic extract of *Bryum argenteum* was found to be more active than other fractionated extracts.

Table1: Phytochemical screening of the Bryumargenteumextracts

Compound	Ethanolic extract	Acetonic extract
Alkaloids	-	-
Anthoquinones	-	-
Cardic Glycosides	+	+
Flavanoids	+	+
Saponins	-	-
Sterols	+	+
Terpenoids	+	+

+ = Phytoconstituents present, - = Phytoconstituents absent.

SN	Extract Concentration (%)	Colony Diameter (mm)		Per cent I	nhibition (%)
	-	Mean	SD	Mean	SD
1	Control	28.6667	0.1155	0	0
2	10	12.7333	0.1156	55.5867	0.3530
3	20	11.3667	0.0579	60.3480	0.3986
4	40	9.1333	0.0577	67.2033	0.0462
5	60	7.5667	0.0577	78.1100	0.1724
6	80	4.6000	0.1732	84.4567	0.6870
7	100	2.8333	0.2309	88.1100	0.7617
	GM	12.6571	8.2290	65.1494	18.0899
	Se	0.0756		0.2756	
	CD5%	0.2293		0.8493	
	CD1%	0.3184		1.1911	
	CV	1.03		0.73	

Fable 2 :	Effect of	ethanolic	extract	of Bryun	n argenteum	on (Curvularia	lunata
				~	0			

*,** Significant at 5% and 1% respectively

Table 3 :Effect of acetonic extract of Bryum argenteum on Curvularia lunata

SN	Extract Concentration (%)	Colony Diameter (mm)		Per cent Inhibition	n (%)
		Mean	SD	Mean	SD
1	Control	34.4333	0.0577	0	0
2	10	20.8333	0.0573	39.5033	0.2510
3	20	17.4667	0.0574	43.4600	0.1447
4	40	12.4667	0.3786	63.7900	1.1212
5	60	9.7667	0.0578	74.6333	0.1516
6	80	7.5667	0.0577	78.0200	0.2093
7	100	4.9333	0.0578	85.7667	0.1443
	GM	16.2810	9.9763	61.4989	20.3818
	Se	0.0882		0.2816	
	CD5%	0.2675		0.8678	
	CD1%	0.3715		1.2171	
	CV	0.94		0.79	

*,** Significant at 5% and 1% respectively





Deora and Guhil, 2018

SN	Extract	Per cent Inhibition (%)		Per cent Inhibition (%)		Per cent Inhibition (%)	
	Concentrati	Bryum arge	enteum	Bryum argenteum		Mancozeb	
	on (%)	Ethanolic e	xtract	Acetonic extract			
		Mean	SD	Mean	SD	Mean	SD
1	Control	0	0	0	0	0	0
2	10	55.5867	0.3530	39.5033	0.2510	89.3000	0.3615
3	20	60.3480	0.3986	43.4600	0.1447	89.3000	0.3615
4	40	67.2033	0.0462	63.7900	1.1212	89.3000	0.3615
5	60	78.1100	0.1724	74.6333	0.1516	89.3000	0.3615
6	80	84.4567	0.6870	78.0200	0.2093	89.3000	0.3615
7	100	88.1100	0.7617	85.7667	0.1443	89.3000	0.3615
	GM	65.1494	18.0899	61.4989	20.3818	89.3000	0.3029
	Se	0.2756		0.2816		0.2082	
	CD5%	0.8493		0.8678		0.6414	
	CD1%	1.1911		1.2171		0.8996	
	CV	0.73		0.79		0.40	

f able 4: Percent inhibition of mycelial growth in <i>Curvulo</i>	aria lunata against plant extract and	l synthetic fungicide.
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*,** Significant at 5% and 1% respectively



Fig.3 Percent inhibition of mycelial growth in Curvularia lunata against plant extract and synthetic fungicide

The colony diameter was decreased from lower to higher concentration, only 2.83 mm colony diameter was reported at 100 per cent concentration while it was 12.73 mm at 10 per cent. Per cent inhibition was 55.58 to 88.11 at 10 to 100 per cent concentration. While the colony diameter of acetonic extract of *Bryum argenteum* was 4.93 mm at 100 per cent concentration and 20.83 mm at 10 per cent concentration. Per cent inhibition was 39.50 to 85.76 at 10 to 100 per cent concentration. Per cent inhibition of synthetic fungicide was 89.30 in all 10 to 100 per cent concentration Both the extracts were tested for the presence of flavonoids, terpenoids, sterols, alkaloids etc, however terpenoids, sterols, flavonoids, cardiac glycosides were detected in varying degree in extracts of *Bryum argenteum* (table 1). The bryophyte extracts prepared in different solvents were found effective in reducing fungal growth as they posses various secondary metabolites which acts as antifungal agent. The present results showed similarity with the results of Deora and Bhati (2007) reported that the extract of certain bryophytes such as *Plagiochasma articulatum, Anthoceros longii, Fissidens bryoides* showed antibiotic property against *Agrobacterium tumifacians.* Deora and Guhil (2014) studied the antifungal activity of *Bryum capillare* extract against *Drechslera maydis* using hanging drop method. Deora and Guhil (2014) also evaluated the antifungal potential of moss *Bryum argenteum* and *Bryum cellulare* against the phytopathogenic fungi *Curvularia lunata* using pour plate method. Further Deora and Guhil(2016) ; Deora and Suhalka (2016,2017) made a contribution on phytochemical composition and antifungal activity of moss *Bryum cellulare* and *Philonotis revoluta* on spore germination of different test fungi and reported that moss extracts is potent to inhibit spore germination and malformation of fungal hyphae.

CONCLUSION

Results of the present study concludes that both the extract of *Bryum argenteum* exhibited varying degrees of antimicrobial activities against *C. lunata* but ethanolic extract of *Bryum argenteum* was found in this study to be most effective against the test fungi. Synthetic fungicide Mancojeb also found to be more sensitive towards *C. lunata*. Growth assessment carried out showed that growth was hindered by the extract in the test fungi. The present study suggested that bryophytes can be used as an alternative natural antifungal agent as green fungicides instead of synthetic fungicides.

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