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Antifungal and antibacterial activity of some mosses from Maharashtra, India.

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ABSTRACT

In this study, the antifungal and antibacterial effect of 5 different mosses were tested in vitro against 4 bacterial and 4 fungal strains. For the extraction, methanol, ethanol, and water were used as solvents. The highest antimicrobial effect was seen in methanol extracts, and lowest level of antimicrobial effect was seen in aqueous extracts. *Bryum coronatum Schwaegr and Steeriophyllum anceps* (Bosch .et Lac.) mosses showed the highest activity. The results showed that *Trichoderma viride* and *Penicillium notatum* were found to be more sensitive than the other test strains like *Aspergillus niger* Tiegh and *Fusarium oxysporum* Schlecht. *Staphylococcus aureus strain* was sensitive against *Steeriophyllum anceps* (Bosch .Et Lac.), extracts, and resistant against *Bryum coronatum* Schwaegr and *Hypnum reflexum F.E. Tripp* extracts. *Escherichia coli* was resistant against *Hyophila involuta* (Hook.) Jaeg .Methanolic extract of *Macromitrium sulcatum* (Hook.) Brid and *Bryum coronatum* Schwaegr shows maximum zone of inhibition against all tested bacteria. All the results were compared with standard antibiotic discs viz. Nastatin (10 µg) and ampicillin (10 µg).

Key words: Moss, antimicrobial effect, disc diffusion assay.

INTRODUCTION

Bryophytes are the oldest known land plants in the world. They are also called as plant amphibians of the plant kingdom. They are placed taxonomically between algae and pteridophytes. They are distributed further into three classes, Hepaticae (liverworts, 6000 species), Anthocerotae (hornworts, 300 species) and Musci (mosses, 14 000 species). Based on the morphological characters (branching patterns and location of sexual organs), the mosses has been divided into two major groups as acrocarpous mosses and pleurocarpous mosses. Many bryophytes exhibit antimicrobial effects against fungi and bacteria (Basile *et al*., 1999; Frahm and Kirchhoff, 2002; Ilhan *et al*., 2006; Sabovljevic *et al*., 2006; Subhisha and Subramoniam, 2005; Bodade *et al*., 2008; Dulger *et al.,* 2009).

Most of the species of mosses are not damaged by bacteria, fungi, snails insect larvae and mammals because, biological compounds like aliphatic compounds, oligosaccharides, polysaccharides, acids, phenylquinone and aromatic and phenolic substances in bryophytes are protected against these organisms (Asakawa, 1981, 1984, 2001). Therefore, mosses have the potential for medical use, *Fissidens* species were used as diuretic and hair growth stimulating drugs in China (Asakawa, 1981). In India mosses like *Bryum sp, Philonotis sp* and *Polytrichum juniperinum* which are used to heal burns, bruises and wounds (Ilhan *et al*., 2006). The aim of this study was to determine the antifungal and antibacterial effects of some mosses.

MATERIALS AND METHODS

Plant material:

The present study is based on the mosses like *Bryum coronatum* Schwaegr, *Steeriophyllum anceps* (Bosch .Et Lac.), *Hypnum reflexum* F.E. Tripp, *Hyophila involuta* (Hook.) Jaeg, and *Macromitrium sulcatum* (Hook.) Brid collected from various localities such as Bhimashankar, Kaas-Satara, Khandala, Lawasa, Lonawala, Mahabaleshwar, Pachgani, Purandar, Aundh and Sinhagad regions of Western Ghats of Maharashtra. The material was collected during July, 2017 to September, 2018 from shady places along the sides of foot paths, wet walls, wet soils and tree trunks.

Preparation of the extracts

Samples of mosses were treated with 0.8% Tween 80 aqueous solution to remove epiphytic hosts found on the plant surface. Then, the samples were washed in tap and distilled water, and dried on filter paper. Extracts of the identified moss species like *Hypnum reflexum* F. E. Tripp, *Steeriophyllum ancens* (Bosch et Lac.) Broth, *Macromitrium sulcatum* Brid, *Bryum coronatum* Schwaegr, and *Hyophila involuta* (Hook.) Jaeg was prepared. The samples were extracted with different solvents like ethanol, methanol and distilled water. Extraction made within four days. Plant material (100 g) was dried in open air at room temperature and extract grinded in mortar and pestle with methanol, ethanol and distilled water separately. Finally, filtrates were used to screen antibacterial and antifungal activities by applying extracts saturated discs.

Test microorganisms and their maintenance

All eight microbial strains are tested for their response to mosses extracts. They are obtained from National Fungal Culture Collection of India (NFCCI), Agharkar Research Institute Pune, India. These included four bacterial strains viz. *Bacillus substilis* (NFCCI NCIM 2697), *Escherichia coli* (NFCCI 2067), *Pseudomonas aeruginosa* (NFCCI 2200), *Staphylococcus aureus* (NFCCI 2492) and four fungal strains viz. *Aspergillus niger* (NFCCI 3114), *Fusarium oxysporum* (NFCCI 1276), *Penicillium notatum* (NFCCI 1072), and *Trichoderma viride* (NFCCI 1139). Prior to use tested organisms, they are sub cultured on nutrient agar (NA), and Saboraud's dextrose agar (SDA), Hi-media to ensure their viability and adequate density

Determination of antimicrobial activity

The plant extracts were tested for antibacterial and antifungal activities through the disc diffusion method, according to the National Committee for Clinical Laboratory Standards (NCCLS, 1997). Mueller-Hinton Agar (MHA) and Sabouraud Dextrose Agar (SDA), sterilized and cooled to 4 to 50°C, were distributed in sterilized Petri dishes. The filter paper discs (6 mm in diameter, Whatman No: 1) were individually impregnated with the extract solutions and then placed onto the agar plates, which had previously been inoculated with tested microorganisms (100 µl). Plates were inoculated with bacteria, incubated at 37°C for 24 h and 30°C for 48 h for the fungi. The diameter of inhibition zones were measured in mm. All the tests were performed in duplicate and the inhibition zones were compared with those of reference discs. Reference discs used for control are Nastatin (10µg) for fungi and ampicillin (10 µg) for bacteria.

RESULTS AND DISCUSSION

In this study, the antimicrobial effects of five mosses like *Hypnum reflexum* F. E. Tripp, *Steeriophyllum ancens* (Bosch et Lac.) Broth, *Macromitrium sulcatum* Brid, *Bryum coronatum* Schwaegr, and *Hyophila involuta* (Hook.) having three different solvent extract (methanol, ethanol and distilled water), were compared with standard antibiotics used as positive controls. Antimicrobial activity of plant extracts in different solvents on test microorganisms are given in Table 1 and Table 2

Table-1 Antifungal screening of mosses against test fungi using disc diffusion method.

Sr No	Name of moss species	Moss extract in the following solvent (MIC zone in mm)											
		Distilled water				Ethanol				Methanol			
		F1	F2	F3	F4	F1	F2	F3	F4	F1	F2	F3	F4
1	*Hypnum reflexum* F. E. Tripp	8	7	**11**	9	9	9	9	**10**	9	8	9	8
2	*Bryum coronatum* Schwaegr	2	3	5	9	6	8	7	7	6	7	7	7
3	*Steeriophyllum anceps* (Bosch et Lac.) Broth	7	2	6	6	6	6	5	4	6	7	6	**13**
4	*Hyophila involuta* Hook.) Jaeg	4	5	6	6	9	3	6	6	7	6	5	4
5	*Macromitrium sulcatum* Brid	6	7	4	6	7	8	6	7	6	7	8	7
6	Antibiotic-Nastatin(10µg)	5	6	4	5	5	6	4	5	5	6	4	5

F-1 *Fusarium oxysporum,* F2- *Aspergillus niger*, F3-*Trichoderma viride,* F4-*Penicillium notatum.*
mm- Millimeter, NI- No inhibition,

Table-2 Antibacterial screening of mosses against test bacteria using disc diffusion method.

Sr No	Name of moss species	Moss extract in the following solvent (MIC zone in mm)											
		Distilled water				Ethanol				Methanol			
		B1	B2	B3	B4	B1	B2	B3	B4	B1	B2	B3	B4
1	*Hypnum reflexum* F. E. Tripp	1	3	3	4	6	6	3	6	3	6	7	4
2	*Bryum coronatum* Schwaegr	**1**	3	3	4	9	8	9	11	5	9	**13**	9
3	*Steeriophyllum anceps* (Bosch et Lac.) Broth	9	4	3	5	7	9	5	8	6	7	6	4
4	*Hyophila involuta* Hook.) Jaeg	6	3	**NI**	2	6	6	9	11	9	3	6	6
5	*Macromitrium sulcatum* Brid	3	5	4	3	5	5	6	11	11	9	11	7
6	Antibiotic-ampicillin(10µg)	6	8	8	6	6	8	8	6	6	8	8	6

B1- *Staphylococcus aureus*, B2- *Pseudomonas aeruginosa*, B3- *Escherichia coli, B4- Bacillus subtilis.* mm- Millimeter, NI- No inhibition.

The results indicated that methanolic and ethanolic extracts of all mosses had inhibition effect against all test fungi. Methanolic extract of *Steeriophyllum ancens* shows maximum zone of inhibition (13mm) against *Penicillium notatum* and minimum zone of inhibition (2mm) in aqueous extracts against *Aspergillus niger*. Aqueous extract of *Hypnum reflexum* shows maximum zone of inhibition (11mm) against *Trichoderma viride* and minimum zone of inhibition (7mm) against *Aspergillus niger.*

The antimicrobial test results revealed that *Bryum coronatum* and *Steeriophyllum ancens* extracts had a potential activity against all microorganisms, except *Fusarium oxysporum* and *Aspergillus niger.* The highest antifungal effect was shown in methanol extracts, aqueous showed the lowest level of antifungal effect.

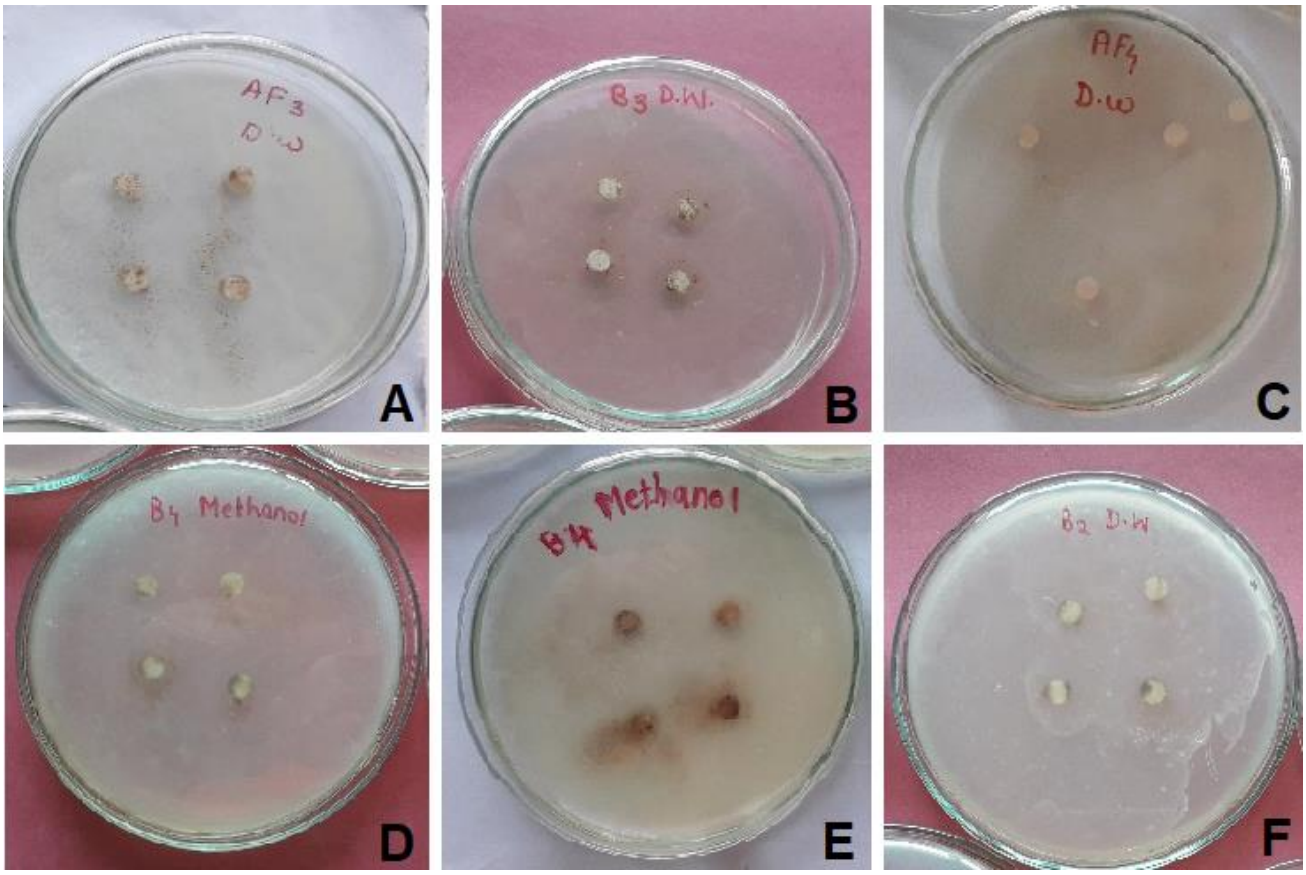

Figure 1: Antifungal and antibacterial activities of various mosses extracts.
A: *Hypnum reflexum* **B:** *Hyophila involuta* **C:** *Bryum coronatum* **D:** *Bryum coronatum* **E:** *Bryum coronatum*n
F: *Hyophila involuta*

The results showed that *Trichoderma viride* and *Penicillium notatum* were found to be more sensitive than the studied test strains (*Aspergillus niger* and *Fusarium oxysporum*). *Staphylococcus aureus* was sensitive against *Steeriophyllum anceps* extracts, this strain was resistant against *Bryum coronatum* and *Hypnum reflexum* extracts. *Escherichia coli* was resistant against Hyophila involuta .Methanolic extract of *Macromitrium sulcatum* and *Bryum coronatum* shows maximum zone of inhibition against all tested bacteria.

These mosses samples showed inhibition effect against both the gram positive and negative bacteria. Our results revealed that the selected mosses might possess a novel antimicrobial agents. The results obtained are similar to some researcher's report that extracts from mosses displayed antifungal activities (Castaldo-Cobianchi *et al*., 1988; Bodade *et al.,* 2008). This study help in the discovery of new antibiotics that could serve as selective agents against infectious diseases.

REFERENCES:

Asakawa Y (1981) Biologically active substances obtained from bryophytes. J.*Hattori Bot. Lab*. 50: 123-142.

Asakawa Y (1984) Some biologically active substances isolated from Hepaticae: terpenoids and lipophilic aromatic compounds. *J. Hattori. Bot. Lab*. 56: 215-219.

Asakawa Y (2001) Recent advances in phytochemistry of bryophytes acetogenins, terpenoids and bis(bibenzyl)s from selected Japanese, Taiwanese, New Zealand, Argentinean and European liverworts. *Phytochemistry*, 56: 297-312.

Basile A, Giordano S, Lopez-Saez, JA, Castaldo Cobianchi R (1999) Antibacterial Activity of pure flavonoids isolated from mosses. Phytochemistry, 52: 1479-1482.

Bodade RG, Borkar PS, Saiful Arfeen MD, Khobragade CN (2008) In vitro Screening of bryophytes for antimicrobial activity. *J. Med. Plants*, 7(4): 23-28.

Castaldo CR, Giordano S, Basile A, Violante U (1988) Occurrence of antibiotic activity in *Conocephalum conicum*, *Mnium umdulatum* and *Leptodictyum riparium* (Bryophyta). *Giorn. Bot. Ital*. 122: 303-311.

Dulger B, Hacıoglu N, Uyar G (2009) Evaluation of Antimicrobial Activity of Some Mosses from Turkey. *Asian J. Chem*. 21(5): 40934096.

Frahm JP, Kirchhoff K (2002) Antifeeding effects of bryophyte extracts from Neckera crispa and *Porella obtusata* against *Arion lusitanicus*. Cryptogam. Bryol. 23: 271-275.

Glime J (2007) *Bryophyte Ecology*, Vol. 1, Michigan Technological University (MTU), Botanical Society of America (BSA), International Association of Bryologists (IAB), published online at http://www.bryoecol.mtu.edu.

Goffinet B, Shaw AJ (eds.) (2009) Bryophyte Biology. Cambridge University Press, Cambridge, UK. p. 565.

Dlhan S, Savaroglu F, Colak F, Filik Dscen C, Erdemgil FZ (2006) Antimicrobial Activity of Palustriella commutata (Hedw.) Ochyra Extracts (Bryophyta) *Turk. J. Biol*. 30: 149-152.

NCCLS (National Committee for Clinical Laboratory Standards) (1997) Performance Standards for antimicrobial disk susc0eptibility test. (VIth ed.). Wayne PA. Approved Standard. M2-A6.

Sabovljevic A, Sokovic M, Sabovljevic M, Grubisic D (2006) Antimicrobial activity of *Bryum argenteum*. Fitoterapia, 77: 144-145.

Subhisha S, Subramoniam A (2005) Antifungal activities of a steroid from *Pallavicinia lyellii,* a liverwort. *Indian J. Pharmacol.* 37(5): 304308.