Indian Journal of Plant Sciences ISSN: 2319–3824
An Open Access, Online International Journal Available at http://www.cibtech.org/jps.htm
2018 Vol. 7(4) October-December, pp.42-48/Deora and Deora
Research Article

STUDIES ON PHYTOCHEMICAL SCREENING OF CERTAIN BRYOPHYTES OF RAJASTHAN

G.S. Deora and *Sandhya Deora
Department of Botany, University College of Science
Mohanlal Sukhadia University Udaipur 313001 (Rajasthan)
**Author for Correspondence*

ABSTRACT

Bryophytes are lower group of plants which generally grow in moist and shady places during rainy season. The present study was carried out to find out the presence of various phytochemicals which plays a decisive role in the protection of plants against the growth of different types of pathogens. The presence of secondary metabolites such as terpenoids, flavonoids, saponins, cardiac glycosides and sterols, anthoquinones and alkaloids were determined by using different types of tests in the aqueous, ethanolic and methanolic extracts of *Asterella angusta* and *Hyophila involuta.* Both *A. angusta* and *H. involuta* showed high intensity of colors in presence of terpenoids, flavonoids, saponins, cardiac glycosides and in methanolic extract followed by ethanolic and aqueous crude extracts.

INTRODUCTION

Plants are the store house of many types of phytochemicals as a means of protection against insects, fungi, bacteria and viruses. When we ingest these plant chemicals or phytochemical extracts they offer us some of that same strong protection (Vandell, 1999).

Plants and plant parts provide a good source of therapeutically active compounds which are rich in valuable bioactivities of antioxidants, anti-inhibitory, antimicrobial, and antitumor activities (Maridass *et al*., 2008).

The word flavonoid is derived from Latin word *Flavus* means yellow. Flavonoid is recognized as having properties which are beneficial to human health (Middleton, 1998). According to De Groot and Rauten (1998) more than 4000 varieties of flavonoids have been identified. Terpenoids play an important role in traditional herbal remedies and are under investigation for antibacterial, antineoplastic and other pharmaceutical functions. Terpenoids appear to be ubiquitous in bryophytes (Asakawa *et. al*., 1981).

The steroids and sterols are produced from terpenoids precursors. Phytosterols are special forms of steroids with a hydroxyl group at position-3 and a skeleton derived from cholestane. So far 15 sterols have been isolated from bryophytes. Sitosterol and stigmasterol occur in all investigate taxa. Campesterol is more common in liverworts and rest of the sterols occur in mosses (Asakawa, 1980).

Cardiac glycosides are drugs used in the treatment of congestive heart failure and cardiac arrhythmia. These glycosides are found as secondary metabolites in several bryophytes.

The aim of this study was to find out the presence of phytochemicals which are responsible for the antibiotic activity of these moisture loving plants.

Keywords: *Phytochemical Screening, Bryophytes, Extracts, Phytochemicals, Secondary Metabolites*

MATERIALS AND METHODS

Preparation of extracts:

The washed and cleaned plant materials were dried on blotting paper in laboratory at room temperature to remove extra moisture. After drying the plant materials were grinded using pestle and mortar. Sunlight

exposure was avoided to prevent the loss of volatile compounds. For aqueous extract preparation, weighted plant material was grinded in pestle and mortar with equal amount of double distilled water (DDW) till the formation of a fine paste. The same method was adopted for ethanol and methanol extract preparation except the grinding of plant material with ethanol and methanol respectively instead of double distilled water.

Centrifugation:

The smooth pastes of plants were filtered through Whatman's filter paper no. 1 and centrifugation was done at 6000 rpm for 20 minutes.

Filtration:

After centrifugation the supernatant was filtered again using Whatman's filter paper no.1 and stored in conical flasks for further use.

Phytochemical screening:

The aqueous ethanolic and methanolic extracts of *Hyophila involuta* (a moss) and *Asterella angusta* (a liverwort) were subjected to various phytochemical tests. The methods of Evans (1997) and Raaman (2006) were used to detect the presence of a flavonoids, terpenoids, alkaloids, anthraquinones, cardiac glycosides, saponins, sterols.

RESULTS AND DISCUSSION

The phytochemical screening of present investigation revealed the presence of cardiac glycosides, flavonoids, saponins, sterols and terpenoids in both *Asterella angusta* and *Hyophila involuta.* The intensity of color was more in methanolic followed by ethanolic and aqueous extracts suggested that more phytochemicals were dissolved in methanolic extract followed by ethanolic and aqueous extracts (Table 1-2; photo plate 1-4).

The systematic screening of plant species with the purpose of discovering new bioactive compounds is a routine activity in several laboratories. Plants and plant parts have been provided a good source of phenolic compounds, nitrogen compounds, vitamins, terpenoids, saponins and some other secondary metabolites which are rich in valuable bioactivities of antioxidant, anti-inflammatory, antitumor, antimutagenic, anticarcinogenic and antimicrobial activities (Maridass *et.al.* , 2008). Bryophytes contain a large number of terpenoids and aromatic compounds. Many researchers have established the presence of these active chemicals in bryophyte extracts.

Seven pure flavonoids from five moss species were isolated and identified by Basil *et al*., (2003). Some of these shown pronounced antibacterial activity.

Asakawa *et al.*, (2013) reported the volatile compounds of 25 taxa of the liverwort family Frullaniaceae from New Zealand, Australia and South America by GC-MS analysis.

The presence of flavonoids, terpenoids, cardiac glycosides and saponins have been reported in various liverworts and mosses (Deora and Guhil, 2016; Deora and Vishwakarma, 2016; Deora and Suhalka, 2017).

Thus the results of the present study as well as the survey of literature and the comparison of this work with many other research work have made it easy to conclude that most of the bryophytes contain terpenoids, flavonoids and many other aromatic compounds. So far only five percent of the total bryophytes have been studied chemically. Although liverworts are small plant groups, therefore a number of terpenoids and phenolic compounds, several of which show interesting biological activity.

Table 1: Showing results of phytochemical tests applied on various extracts of *Asterella angusta* (A liverwort)

Class of Phytochemicals	Phytochemical Test Applied	Observations	Results	Intensity of colour* in *Asterella angusta* extracts		
				Aqueous	Ethanol	Methanol
Alkaloids	Mayer's and Hager's Test	No precipitate	Phytochemical absent	–	–	–
Anthoquinones	Borntrager's Test	No layer formation	Phytochemical absent	–	–	–
Cardiac glycoside	Keller killeni Test	Brown ring	Phytochemical present	+	+	++
Flavonoids	Ferric Chloride Test	Green colour	Phytochemical present	+	++	+++
	Lead Acetate and Gelatin Test	White precipitate	Phytochemical present	+	++	+++
	Alkaline Reagent and Sodium Hydroxide Test	Yellow fluorescent colour	Phytochemical present	+	++	+++
Saponins	Froth Test	No froth	Phytochemical absent	–	–	–
Sterols	Salkowaski Test	Reddish brown colour	Phytochemical present	+	++	+++
	Liebermann-Burchardt Test	Brown ring	Phytochemical present	+	+	++
Terpenoids	Salkowaski Test	Lower layer turned yellow	Phytochemical present	+	+	++
	Liebermann-Burchardt Test	Deep red colour	Phytochemical present	+	+	++
	Phenyl hydrazine test	Yellow-orange colour	Phytochemical present	+	+	++

**Intensity of colour: (-): No colour; (+): Light colour; (++): More intense colour; (+++): Most intense colour among the three types of extracts tested.*

Table 2: Showing results of phytochemical tests applied on various extracts of *Hyophila involuta* (A moss)

Class of Phytochemicals	Phytochemical Test Applied	Observations	Results	Intensity of colour* in *Hyophila involuta* extracts		
				Aqueous	Ethanol	Methanol
Alkaloids	Mayer's and Hager's Test	No precipitate	Phytochemical absent	–	–	–
Anthoquinones	Borntrager's Test	No layer formation	Phytochemical absent	–	–	–
Cardiac Glycoside	Keller killeni Test	Brown ring	Phytochemical present	+	++	+++
Flavonoids	Ferric Chloride Test	Green colour	Phytochemical present	+	++	+++
	Lead Acetate and Gelatin Test	White precipitate	Phytochemical present	+	++	+++
	Alkaline Reagent and Sodium Hydroxide Test	Yellow fluorescent colour	Phytochemical present	+	++	+++
Saponins	Froth Test	No froth	Phytochemical absent	–	–	–
Sterols	Salkowaski Test	Reddish brown colour	Phytochemical present	+	++	+++
	Liebermann-Burchardt Test	Brown ring	Phytochemical present	+	++	+++
Terpenoids	Salkowaski Test	Lower layer turned yellow	Phytochemical present	+	++	+++
	Liebermann-Burchardt Test	Deep red colour	Phytochemical present	+	++	+++
	Phenyl hydrazine Test	Yellow-orange colour	Phytochemical present	+	++	+++

**Intensity of colour: (-): No colour; (+): Light colour; (++): More intense colour; (+++): Most intense colour among the three types of extracts tested.*

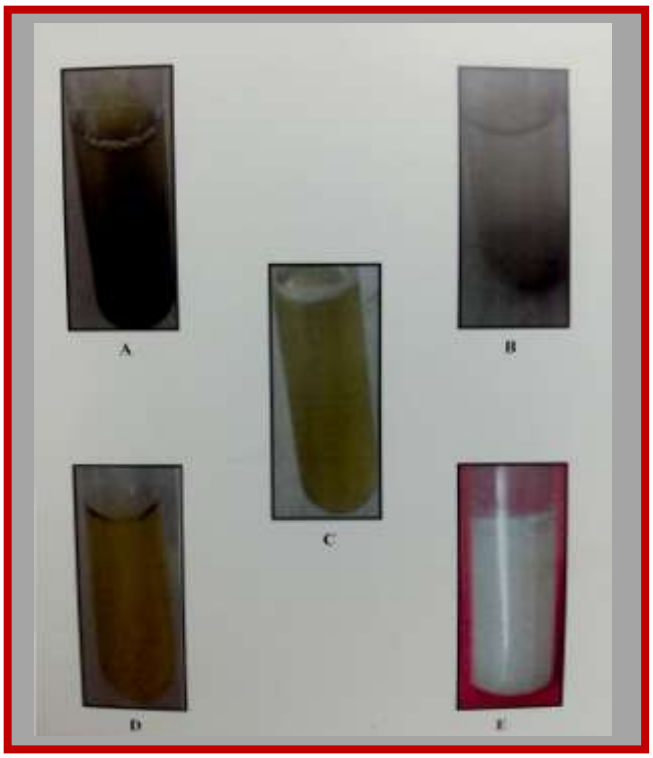

Photo plate 1

Tests for flavonoids:
(A) Ferric chloride test (B) Lead acetate test (C) Alkaline reagent test (D) Sodium hydroxide test (E) Gelatin test

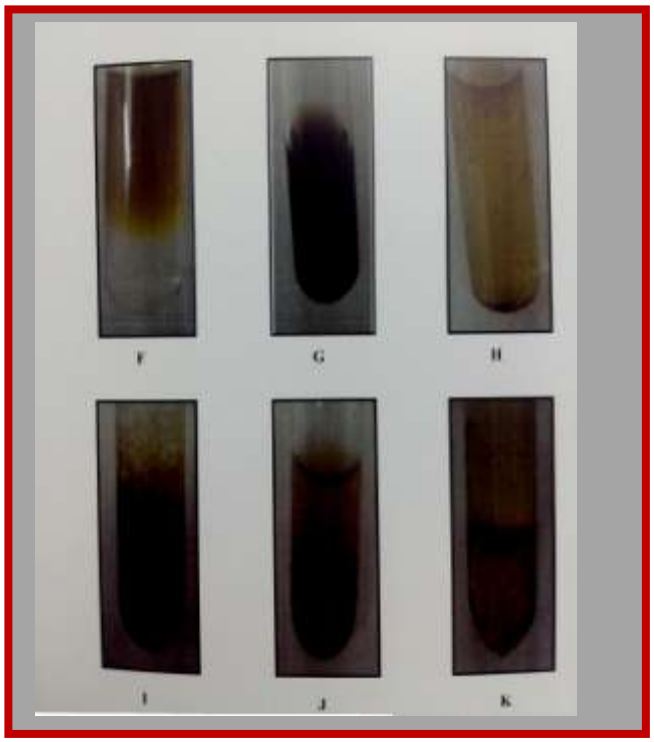

Photo plate 2

Tests for terpenoids: (F) Salkowaski test (G) Liebermann-Burchardt test (H) Phenylhydrazine test.

Test for sterols:
(I) Salkowaski test (J) Liebermann-Burchardt test.
Test for Cardiac Glycoside:
(K) Keller Killeni test

Photo plate 1- 2 Phytochemical screening of *Asterella angusta*

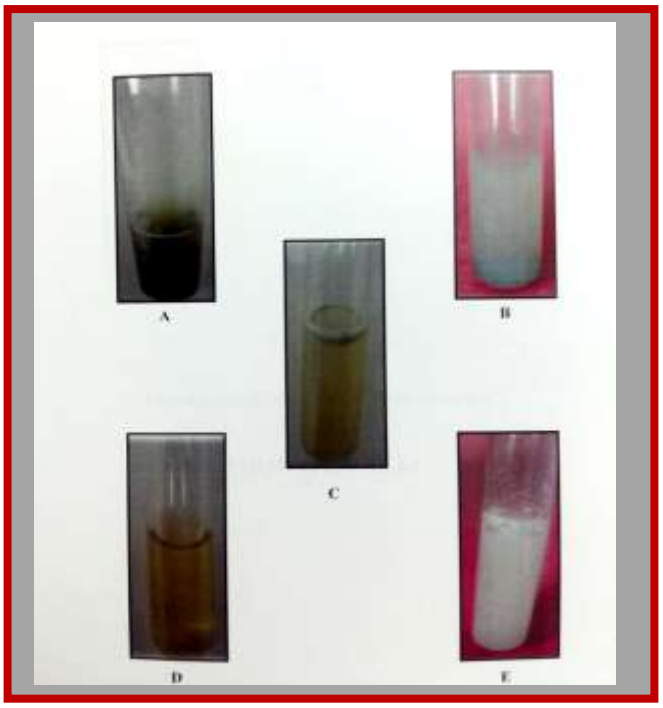

Photo plate 3

Tests for flavonoids:
(A) Ferric chloride test (B) Lead acetate test (C) Alkaline reagent test (D) Sodium hydroxide test (E) Gelatin test

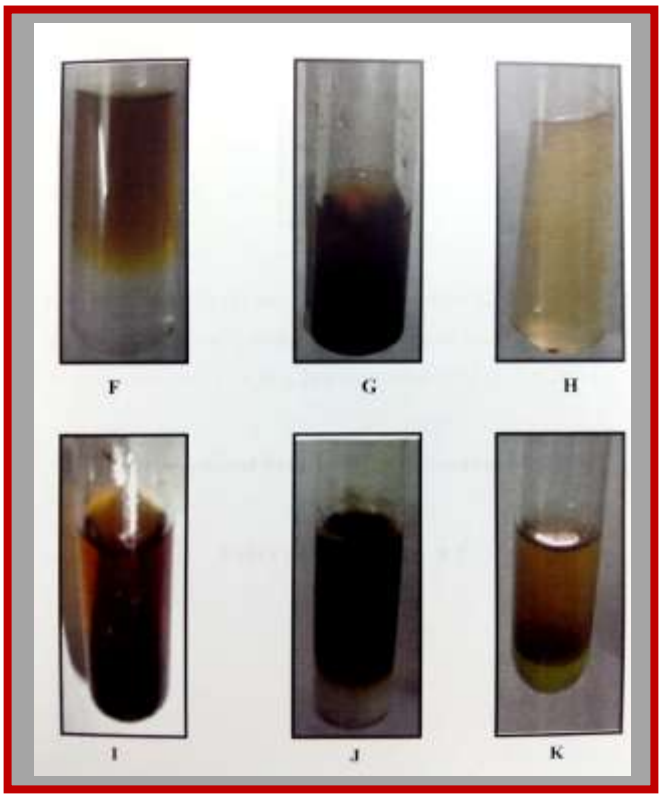

Photo plate 4

Tests for terpenoids:
(F) Salkowaski test (G) Liebermann-Burchardt test (H) Phenylhydrazine test.
Test for sterols:
(I) Salkowaski test (J) Liebermann-Burchardt test.
Test for Cardiac Glycoside:
(K) Keller Killeni test

Photo plates 3-4: Phytochemical screening of *Hyophila involuta*

REFERENCES

Asakawa Y, **Ludwiezuk A** and **Nagashima F (2003)**. Phytochemical and biological studies of bryophytes. *Phytochemistry* **91** 52-80.

Asakawa Y, **Toyota M** and **Takemoto T (1980)**. Four new sacculatane type diterpenoids from *Trichocoleopsis sacculata* and *Pellia endiviaefolia. Phytochemistry* **19**(8) 1799-1803.

Asakawa Y, **Toyota M** and **Takemoto T** and **Mues R (1981)**. Aromatic esters and terpenoids of the liverworts in the genera *Trichocolea*, *Neotrichocolea*, and *trichocoleopsis*. *Phytochemistry* **20**(12) 2695-2699.

Basile A, **Sorbo S**, **Lopez-Saez JA** and **Cobianchi R (2003)**. Effects of seven pure flavonoids from mosses on germination and growth of *Tortula muralis* Hedw. (Bryophyte) and *Raphanus sativus* (Magnoliophyta). *Phytochemistry* **62**(7) 1145- 1151.

De Groot H and **Rauten UC (1998)**. Tissue injury by reactive oxygen species and the protective effects of flavonoids. *Fundam clin pharmacol* **12** 249-255.

Deora GS and **Guhil N (2016).** Studies on antifungal potential of *Bryum cellulare* (A moss) crude extracts against spore germination of fungus *Curvularia lunata*. *International Journal of Pharmaceutical Sciences and Research* **7**(1) 353-357

Deora GS and **Suhalka D (2017)**. Estimation of quercetin by High Performance chromatography and antifungal activity of moss *Philonotis revoluta*. *International Journal of Pharmaceutical Sciences and Research* **8**(1) 294-300.

Deora GS and **Vishwakarma G (2016**). Antimicrobial efficacy of moss *Bryum argenteum* (Hedw.) (Bryale: Bryaceae) against plant pathogen *Pseudomonas syringae* (PV.) (Pseudomonadales: Pseudomonadaceae). *Journal of Applied Life Sciences International* **5**(1) 1-8.

Evans WC (1991). Trease and Evans Pharmacognosy. Harcourt Brace and company. Asia Pvt. Ltd**.**

Maridass M, Ghandhikumar S and Raju G (2008). Preliminary phytochemical analysis of *Diospyros species*. *Ethnobotanical leaflets* **12**: 868-872.

Raaman N (2006). *Phytochemical techniques*. New India publishing 318pp.

Vandell CJ (1999). *Phytochemical testing*. *Food product design* (February). 77-84.