Biological Forum – An International Journal **8**(2): 410-416(2016)

ISSN No. (Print): 0975-1130
ISSN No. (Online): 2249-3239

Promoting and Inhibiting Effects of Bryophyte Extracts on the Seed Germination and Seedling Growth of *Vigna radiata and Cicer arietinum*

***Gaurav Bhadauriya*[*], *Shivom Singh*[*] *and Kajal Srivastava*[**]**

[*]*Department of Environmental Science,*
ITM University Gwalior, (Madhya Pradesh), India
[**]*Department of Biotechnology,*
Govt. Kamla Raja Girls Post Graduate College Gwalior, (Madhya Pradesh), India

(Corresponding author: Shivom Singh, shivomsingh101@gmaill.com)
(Received 05 November, 2016, Accepted 18 December, 2016)
(Published by Research Trend, Website: www.researchtrend.net)

ABSTRACT: The present study deals with the influence of bryophyte extract on seed germination and seedling growth of economically important crops *Vigna radiata* (Mung bean) and *Cicer arietinum* (Bengal gram). Seed germination tests were performed with moss (*Dicranum scoparium*) and liwerwort (*Plagiochasma appendiculatum*). Bryophytes species (moss and liverwort) were collected from the northern region of Uttarakhand (Almora and Ranikhet) of India. During study, effect of aqueous and methanolic extract of moss and liverwort on seed germination and seedling growth was recorded. The inhibition in germination rate was observed at high concentration of methanol and aqueous extract in the both seeds of Mung bean and Bengal gram, however, highly diluted aqueous extracts showed an increase in the germination and promoted the growth in both crop species. Presented data evaluate that the complete inhibition of germination with methanol extract of moss and liverwort was observed on the both crops.

Keywords: Bryophytes extract, seed germination, seedling growth, inhibition and growth promotion

INTRODUCTION

Bryophytes are usually not thought of as economically important plants although they possess many interesting properties. In ancient time, bryophytes have been used in ecology, horticulture, fuel production, household uses, medicines and even in food (Glime and Saxena, 1991). More than 70% of all medicinal compounds have derived from a small fraction of the world's biodiversity. There is great potential to exploit the bryophytes by exploring the huge chemical diversity present in these plants (Saxena *et al.,* 2006). These small plants have immense value for sustainable development and is least investigated for bioactive molecules. Yet in nature they appear to be well protected against grazing and pathogens by their chemical constitute (Glime and Saxena, 1991; Srivastava and Singh, 2007, Singh *et al.,* 2010). They have various therapeutic applications, which are currently used in the treatment (Banerjee and Sen, 1979, Saxena and Hareinder, 2004). Bryophytes are considered as a remarkable reservoir of new, natural products or secondary compounds, many of which have shown interesting biological activity. These activities of bryophytes include antimicrobial, antifungal, cytotoxic, antitumor, vasopressin antagonist, cardiotonic, allergy causing, irritancy and tumour effecting, insect anti-feedant, insecticidal, molluscicidal, pesticidal, plant growth regulatory, superoxide anion radical release inhibition and 5-lipoxygenase, calmodulin, hyaluronidase and cyclooxygenase inhibition features (Iqbal *et al.,* 2005; Sharma *et al.,* 2009). Several falvanoids such as querecetin, isoquercetin, and rutin among many other have shown effect on plant growth as reported in previous studies (Rivera-Vargas, 1993; Parvez *et al.,* 2004; Iqbal *et al.,* 2005).

Bryophyte extracts exhibit dual effects on seedling germination depending on the species. It was shown that extract of the liverwort *Porella platyphylla* inhibits the growth of radice seedlings, whereas the extract of *Brachythecium rutabulum* promotes the growth of radice seedlings (Frahm, *et al.,* 2012).

Bryophytes have unexpected high potential for applied research with implications for the improvement of crop plants (Frahm, 2004).

Keeping in view the importance of increasing use of bryophyte extracts, laboratory studies were initiated to investigate the effectiveness of bryophyte extracts for possible effects on seed germination and growth of Mung bean and Bengal gram. Thus, any possible impact on these two major crops could be further elaborated for their use in agriculture.

MATERIAL AND METHODS

A. Collection of Bryophytes

Fresh thallus of all tested bryophytes species were collected from Almora and Ranikhet, the Northern region of Kumaon, Uttarakhand, India in the month of August 2015. The Northern Himalayan region is known for a luxuriant bryophyte cover, both in frequency and diversity (Pande, 1958; Gangulee, 1969). Kumaon is situated in the state of Uttrakhand and lies between the latitude 28°44' and 30°49' N, and longitude 78°45' and 81°1' E. The topography of the area is irregular due to valleys and plateaus of various dimensions.

B. Method of Extraction

Bryophyte sample were carefully inspected to remove contaminates like soil and other plant materials. Bryophyte samples were prepared from entire green part of the thallus and washed with tap water. 5 g fresh material of bryophyte was ground in mortar and pestel with 50 ml of solvent (water and methanol) to yield a pulp and shaked in rotary shaker (200 rpm) for 12 hrs, and filtered with Whatman No. 1 filter paper. Final volume of the extract was made upto 100 cc by adding respective solvent and considered as full concentration (100%). Then these extracts were diluted to 100%, 80%, 60%, 40% and 20% concentration and were stored at 4ºC for further investigations.

C. Seed culture and Treatment

Investigated seed were collected from Rajmata Vijayaraje Scindia Krishi Vishwavidyalaya, Gwalior, MP. Seeds of Mung bean *(Vigna radiata)* and Bengal gram (*Cicer arietinum)* were surface sterilized with water: bleach (10:1 v/v) solution for 10 minutes to avoid contamination and were thoroughly rinsed several times with sterile water. For testing, Petri dishes of 9 cm were washed, dried and then sterilized in autoclave at 110 -120°C for 1 hour. Whatman No.1 filter papers were kept in each Petri dish and eight seeds of Mung bean and Bengal gram were placed in separate Petri dishes at equal distances. All the experimental Petri dishes were kept at room temperature of 27°C for 288 h. Supplementary light was provided by cool white lamps, 400 µE m^{-2} s^{-1}, 400-700 nm, with a 16/8 h day/night cycle.

10 ml of each extract was added to the Petri dishes and each treatment was repeated three times. Seeds soaked in distilled water were used as control. 10 ml distilled water was applied to each Petri dish during the experiment to keep the Watman paper moist for seedling development for a period of 288 hrs.

Germination, root length and shoot length of seedling were recorded after every 24 hrs intervals upto 288 hrs. Emergence of radical was considered as criterion for seed germination. Average root and shoot lengths for each treatment were calculated.

RESULTS AND DISCUSSION

Effect of the aqueous and methanol extracts of moss (*D. scoparium*) and liwerwort (*P. appendiculatum*) on seed germination was analyzed in Mung bean and Bengal gram and was presented in Table (1&2). The germination was completed within 24 hrs in Mung bean and 48 hrs in Bengal gram under controlled conditions. The low concentration (20% and 40%) of aqueous extracts of both bryophytes (Table 1&2) resulted in promoting effect on the germination of Mung bean and Bengal gram seeds. In contrast, there was 100% inhibition of germination of both crop seeds treated with methanol extracts of bryophytes (Table 1 & 2). Furthermore, at higher concentration (80-100%) of aqueous extract of undertaken bryophytes, the results show decrease in germination of experimental crops during 24 hrs. However, on increasing time duration (after 48 hrs), 100% germination was observed in seeds of Mung bean and Bengal gram treated with aqueous extract (Table 1 & 2).

The aqueous extracts of moss and liverwort at low concentration (20-40%) promote the length of the roots and shoots of Mung bean and Bengal gram seedlings in comparison to control; whilst at higher concentration (60, 80 and 100%) of the bryophytes extract (aqueous) had inhibition of growth of seedlings compared with the control (Fig. 1-8).

In present study the following results were obtained undiluted aqueous moss and liverwort extract showed a distinct inhibition of the growth of the both undertaken crops i.e. Mung bean and Bengal gram (Fig. 1-8). Diluted aqueous extracts (20-40%) showed prominent promoting effect over to control. Moreover, methanol extracts revealed a complete inhibition at the concentration of 100, 80 and 60%, but, in *D. scoparium* a slightly germination at a dilution of 20 and 40% in both the experimental crops. The effect of bryophytes to promote germination of seeds and seedlings of undertaken crops is, somehow, complicated to interpret.

Table 1: Effect of bryophytes extract on germination percentage of Mung bean.

Bryophytes	Treatment (Hrs)	Aqueous extract						Methanolic extract					
		100%	80%	60%	40%	20%	Control (Water)	100%	80%	60%	40%	20%	Control (Methanol)
P. appendiculatum	24	50	75	100	100	100	100	-	-	-	-	-	-
	48	75	75	100	100	100	100	-	-	-	-	-	-
	96	100	100	100	100	100	100	-	-	-		-	-
	120	100	100	100	100	100	100	-	-	-		-	-
	144	100	100	100	100	100	100	-	-	-	-	-	-
D. scoparium	24	50	75	75	75	100	100	-	-	-	-	-	-
	48	75	75	100	100	100	100	-	-	-	-	-	
	96	100	100	100	100	100	100	-	-	-	25	50	-
	120	100	100	100	100	100	100	-	-	-	25	50	-
	144	100	100	100	100	100	100	-	-	-	25	50	-

Table 2: Effect of bryophytes extract on germination percentage of Bengal gram.

Bryophytes	Treatment (Hrs)	Aqueous extract						Methanolic extract					
		100%	80%	60%	40%	20%	Control (Water)	100%	80%	60%	40%	20%	Control (Methanol)
P. appendiculatum	24	-	-	-	-	-	-	-	-	-	-	-	-
	48	50	75	100	100	100	100	-	-	-	-	-	-
	96	75	100	100	100	100	100	-	-	-	-	-	-
	120	100	100	100	100	100	100	-	-	-	-	-	-
	144	100	100	100	100	100	100	-	-	-	-	-	-
D. scoparium	24	-	-	-	-	-	-	-	-	-	-	-	-
	48	75	75	100	100	100	100	-	-	-	-	-	-
	96	100	100	100	100	100	100	-	-	-	25	50	-
	120	100	100	100	100	100	100	-	-	-	50	50	-
	144	100	100	100	100	100	100	-	-	-	50	50	-

Present study validate the results of Sharma *et al.*, (2009), who reported the complete inhibition of germination in 100% acetone and methanol extract of most of the bryophyte species, whereas, with the decrease in concentration of acetone extracts, increase in germination was observed. They also depicted that; on doing dilution of aqueous extract, increase in germination was observed. However, study presented by Farm *et al.* in 2012 shows results both in agreement and in contrast.

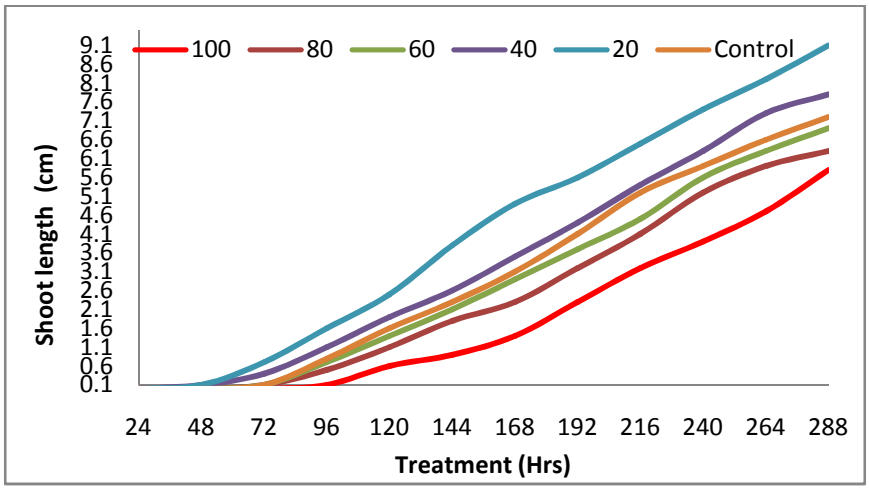

Fig. 1. Shoot length of Mung bean treated with *P. appendiculatum* aqueous extract.

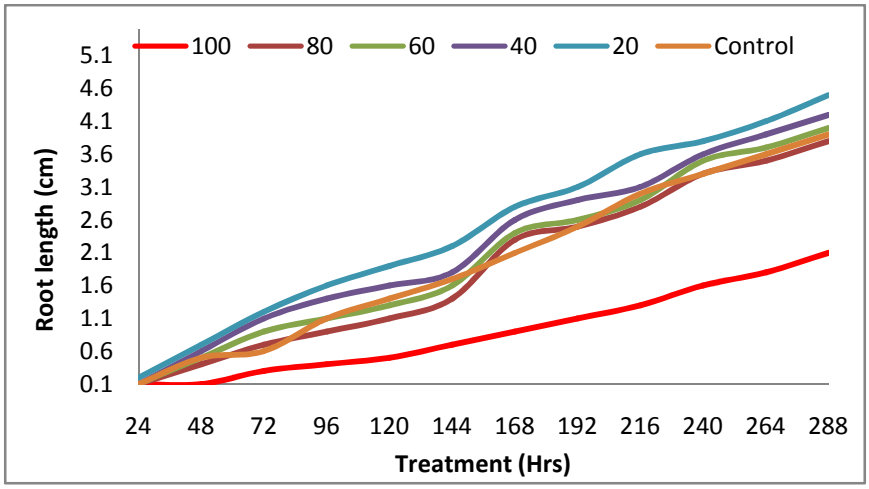

Fig. 2. Root length of Mung bean treated with *P. appendiculatum* aqueous extract.

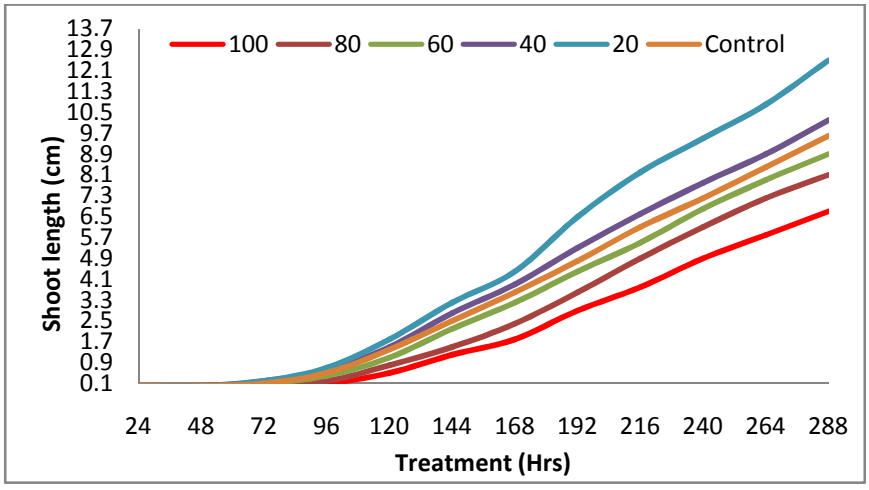

Fig. 3. Shoot length of Bengal gram treated with *P. appendiculatum* aqueous extract.

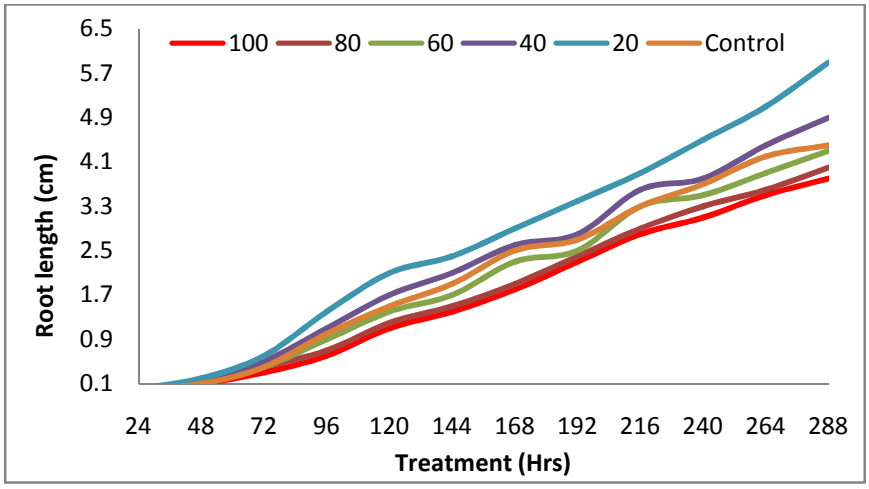

Fig. 4. Root length of Bengal gram treated with *P. appendiculatum* aqueous extract.

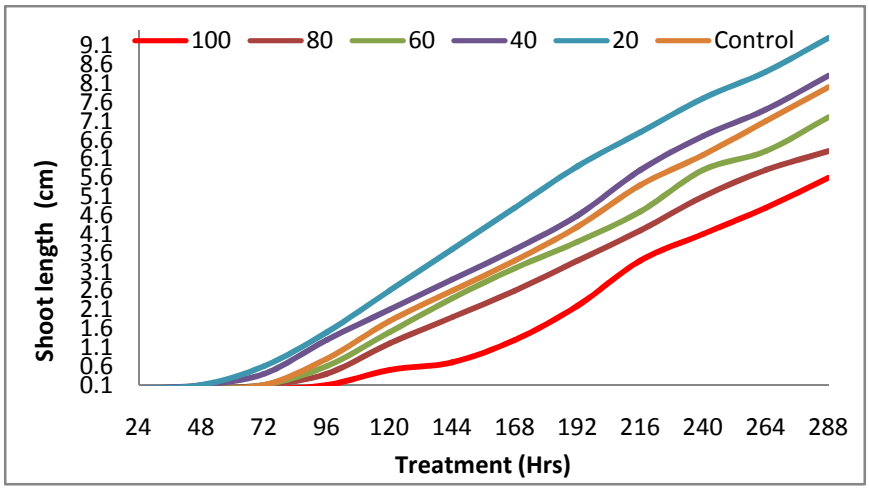

Fig. 5. Shoot length of Mung bean treated with *D. scoparium* aqueous extract.

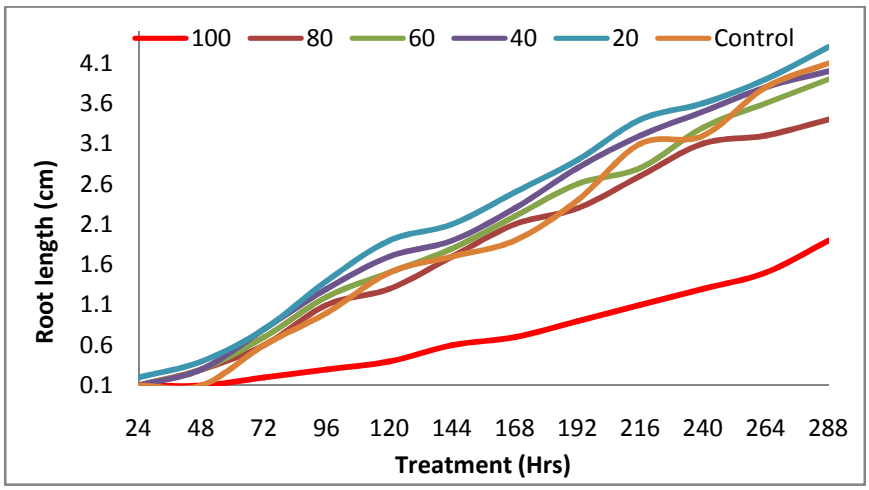

Fig. 6. Root length of Mung bean treated with *D. scoparium* aqueous extract.

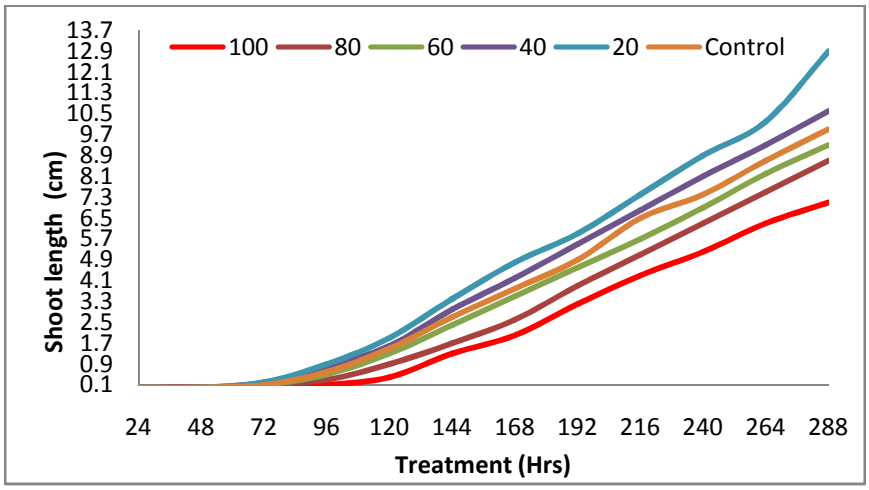

Fig. 7. Shoot length of Bengal gram treated with *D. scoparium* aqueous extract.

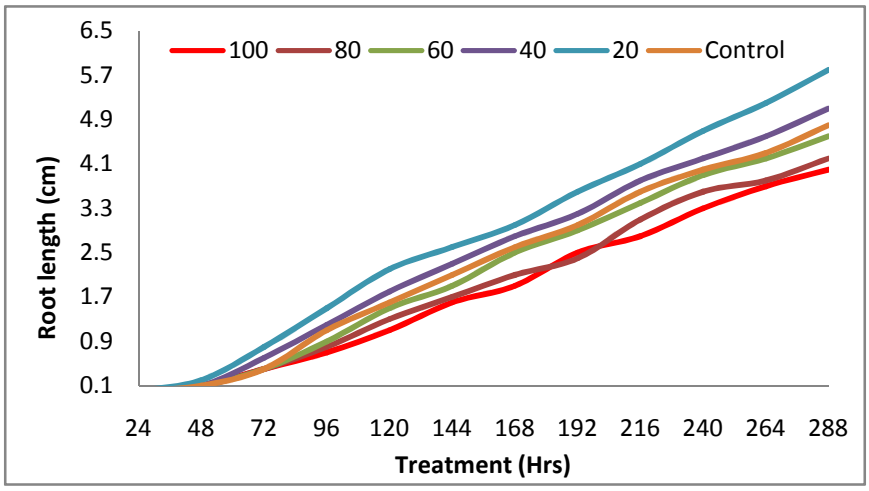

Fig. 8. Root length of Bengal gram treated with *D. scoparium* aqueous extract.

According to them in wheat crops aqueous extract of bryophytes showed promoting effect, whereas, in radish, alcoholic extract showed promoting effect over aqueous extract of bryophytes. Huneck and Meinunger (1990) tested 52 species of mosses and 29 species of liverworts on growth regulation activity. Study done by them illustrated that, different concentrations of the bryophyte extracts vary the amount of promotion viz. inhibition and hence, it is difficult to explain. It was concluded from the study that, the growth regulation activity depends on the concentrations, i.e. Inhibiting growth at higher and promoting growth at lower concentrations of bryophyte extract done in aqueous and alcoholic solvent. Same results were also suggested by Matsuo *et al.* (1981) and Asakawa (1982). They have long evolutionary history and acquired a collection of biochemicals that may be a substantial source of antimicrobial activity and may offer a gene bank for production of proteins, enzymes, fatty acids and sugars allowing crops to survive under drought and cold conditions. Bryophytes have a germination promoting effect along with potential commercial aspect. The prospective of bryophytes for applied research with implication for agriculture and mankind still is not fully valued and explored; the research in this field provides clear indications of exciting new uses for bryophytes in the near future.

ACKNOWLEDGEMENTS

The authors wish to express their gratitude to Dr. Khursheed Ahmad Wani, Department of Environmental Science, ITM University, Gwalior for providing needful help and support.

REFERENCES

Asakawa, Y. (1982). Chemical constituents of Hepaticae. In progress in the chemistry of organic natural products Vienna: Springer **42**: 1-285.

Banerjee, R.D & Sen, S.P. (1979). Antibiotic activity of bryophytes. *The Bryologis*. **82**: 141-153.

Frahm, J.P. (2004). Recent developments of commercial products from bryophytes. *The Bryologist.* **107**: 277-283.

Frahm, J.P., Risse, S. & van Saan-Klein, B. (2012). Are bryophyte extracts inhibiting or promoting seed growth?. *Archive For Bryology*. **127**: 1-10.

Gangulee, H.C. (1969). Mosses of eastern India and adjacent regions. III *Bull. Botanical Society of Bengal*. **23**: 131-134.

Glime, J.M. & Saxena, D.K. (1991). Uses of Bryophytes. Today & Tomorrow's Printers & Publishers, New Delhi. 100 pp.

Huneck, S. &Meinunger, L. (1990). Plant growth regulatory activities of bryophytes: a contribution to the chemical ecology of mosses and liverworts. Pp. 289-298.

Iqbal, Z., Golisz, A., Furubayashi, A., Nasir, H. & Fujii, Y. (2005). Allelopathic potential of buckwheat. *Fourth World Congress of Allelopathy*. Wagga Wagga, Australia.

Matsuo, A., Nadaya, K., Nalayama, M. & Hayashi, S. (1981). Plant growth in hibitors isolated from the Liverwort *Plagiochila ovalifolia*. *Nippon Kagaku Karshi*. 665-670.

Pande, SK. (1958). Some aspects of Indian Hepaticology. *Journal of Hattori Botanical Laboratory*. **24**: 115-202.

Parvez, MM., Tomita-Yokotani, K., Fujii, Y., Konishi, T., & Iwashina, T. (2004). Effects of quercetin and its seven derivatives on the growth of *Arabidopsis thaliana* and *Neurospora crassa*. *Biochemical Systematics and Ecology*. **32**: 631-635.

Rivera-Vargas, L.I., Schmitthenner, A.F. & Graham, T.L. (1993). Soybean flavonoid effects on and metabolism by *Phytophthora sojae. Phytochemistry*. **32**: 851-857.

Saxena, DK. & Kaur, H. (2004). Uses of Bryophytes, *Resonance.* **9**: 56-65.

Saxena, DK., Singh, S., & Srivastava, K.(2006). Distribution of some mosses in Nainital, Almora and Pithoragarh district of Kumaon Region, India. *Environment Conservation Journal*. **7**(1-2): 83-87.

Sharma, A., Bargali, K. & Pande, N. (2009). The allelopathic potential of bryophyte extract on seed germination and seedling growth of *Bidens biternata*. *Nature and Science*, **7**(6): 30-38.

Singh, S., Srivastava, K. & Khanna, DR. (2010). Potential of *Plagiochasma appendiculatum* on inhibition of certain economically important plant pathogens. *Biological Forum – An International Journal.* **2**(2): 122-129.

Srivastava, K. & Singh, S. (2007). Pharmaceutical Products and anti-microbial activity of Bryophytes: Uses of Green Brain. *In: Medicinal Plants: Conservation, Cultivation and Utilization*, Editors: A. K. Chopra, D. R. Khanna, G. Prasad, D. S. Malik and R. Bhutani., Daya Publication House, New Delhi. pp-103-111.