Bryophytes, the oldest lineages among extant land plants, are the second largest phylum of land plants after angiosperms among, and inhabit every continent. These plants invade variety of habitats and are exposed to different environmental and biotic factors with the advantage of miniature nature. They possess dominant leafy or thalloid plant body representing a phase in evolution of plant migration from water to land. But we still do not know much about the role of bryophytes and that at the species level. *Plagiochasma, Asterella, Cyathodium, Targionia* and *Anthoceros* are dominant bryophytes in Western Ghat region of Maharashtra and grow luxuriantly in rainy season.

In order to defend adverse environmental factors many of the secondary metabolites especially terpenoids and phenolic compounds of quite numerous chemical structures are synthesized (Herout, 1990). These secondary metabolites made them unpalatable to herbivores and secondly as defence agents in the plant fight for survival (from pathogens). It is very common observation that generally they are free from attack of any pathogen or insect pest, i.e. they are immune to any pathogenic attack. A general lack of commercial value, small size, and inconspicuous place in the ecosystem have made the bryophytes appear to be of no use to most people. But literature survey suggests a variety of uses by various cultures around the world (Glime and Saxena, 1990). Contemporary plant scientists are considering bryophytes as sources of genes for modifying crop plants to withstand the physiological stresses of the modern world. These simple but unique plants are used in pharmaceutical products, in horticulture, for household purposes, and are also ecologically important.

Taking into account all the above considerations, in the present work some physiological and biochemical work was undertaken which includes chlorophyll, carotenoids, soluble sugars, starch, proline and nitrogen contents. Since the plants are accumulators of heavy metals, mineral status along with heavy metal concentration has also been studied. In order to know the status of secondary metabolites polyphenols, lipids, terpenoids and flavonoids have been investigated qualitatively and quantitatively. The antioxidant properties in the species have been examined which include the enzymes like catalse, peroxidase, superoxide dismutase, polyphenol oxidase and phenylalanine ammonia lyase. Study of
pesticide potential includes in vitro antibacterial and antifungal effects with different extracts (in different solvents viz. ethanol, methanol and dichloromethane) of bryophytes against two routine laboratory used fungi, two phytopathogenic fungi and two plant pathogenic bacteria. Allelopathic studies of these plants have also been carried out. Under this, effect of aqueous extracts from these plants on germination and seedling growth of jowar and wheat has been studied upto 120 h in different modes of application (germination and seedling growth in aqueous extracts, pre-sowing soaking in distilled water (6h) and further germination in bryophyte extracts and pre-sowing soaking in bryophyte extracts (6h) and further germination and seedling growth in distilled water.

The significant findings of the present work are summarized as follows

1. In the bryophytes studied the values of chlorophyll a:b ratio were less than 1, suggesting their adaptation to shaded habitat. The reduced chlorophyll a:b ratio may be due to increased levels of chlorophyll b, a typical shade adaptation. It also suggests that liverworts are more shade-adapted than hornwort A. subtilis.

2. The carotenoid content of these bryophytes was in between 7.79-10.16 mg / 100g fresh wt.

3. The levels of reducing and non-reducing sugars and starch were lower as compared to those of higher plants. The levels of starch were much higher than those of total (soluble) sugars in all the bryophytes.

4. The concentrations of free amino acids and amides were higher in T. hypophylla, followed by A. subtilis and rather at the similar levels in A. wallichiana. In C. cavernarum amino acids and amides content was 1.28 and 1.51 mg g⁻¹ dry tissue respectively and the lowest concentration of these constituents was observed in P. intermedium (1.31 and 1.12 mg g⁻¹ dry tissue respectively).

5. Qualitative analysis of amino acids revealed that A. wallichian, C. cavernarum and A. subtilis contained 7 types of different amino acids, P. intermedium and T. hypophylla contained 6 and 5 types of amino acids respectively. Valine was observed to be common in all the five bryophytes. Other amino acids present were amino-n-butyric
acid, threonine, cysteine, tryptophane, leucine, tyrosine, aspartate, phenylalanine and methionine. In *P. intermedium* valine, glutamic acid and 4 unidentified amino acids were found. Besides in *C. cavernarum* out of 7 amino acids, 2 were unidentified and one cysteine which was only found in this bryophyte. Maximum number of amino acids identified in *A. subtilis* was 6 out of 7 in which leucine and tyrosine were uncommon to all bryophytes analysed. Total 5 amino acids were identified out of 7 with alanine as uncommon to all bryophytes. *T. hypophylla* showed only 5 types of amino acids out of which only two were identified as valine and phenylalanine.

6. *T. hypophylla* recorded the highest amount of proline (11.29 mg g\(^{-1}\) dry tissue) while about half and fairly similar amount was present *A. subtilis* and *A. wallichiana*. This was followed by *C. cavernarum* with 4.33 mg g\(^{-1}\) dry tissue and the lowest being in *P. intermedium* (3.93 mg g\(^{-1}\) dry tissue). Highest level of proline in *T. hypophylla* suggests that it is more drought tolerant than the other bryophytes studied.

7. The studies on antioxidant enzymes showed that activity of catalase was the highest as compared to rest of the other antioxidative enzymes. The highest activity was observed in *P. intermedium* and *A. subtilis* nearly at the same level, however, the lowest but almost similar in *T. hypophylla* and *C. cavernarum*. Activity of catalase in *A. wallichiana* was in between but closer to the lowest one.

8. The activity of peroxidase was the lowest in all bryophytes as compared to that of catalase and SOD. The maximum activity was observed in *A. wallichiana* followed by *P. intermedium* and *A. subtilis*. The activity of this enzyme in *A. subtilis* was followed by *C. cavernarum* and finally the lowest activity was recorded in *T. hypophylla*.

9. The present results show that PPO activity in *C. cavernarum* was the highest among the bryophytes studied while its activity was the lowest in *A. wallichiana*.

10. Results of activity of SOD clearly indicated that *T. hypophylla* has shown the highest SOD activity than any other bryophyte. While *A. subtilis*, *A. wallichiana* and *C. cavernarum* have shown slight lower values for SOD activity. *P. intermedium* has the lowest SOD activity.
11. Activity of PAL was the highest in the hornwort, *A. subtilis* while it was more or less at similar levels in case of *P. intermedium*, *A. wallichiana* and *C. cavernarum* with the lowest being in *T. hypophylla*. It is one of the possibilities that since *A. subtilis*, a hornwort which grows very close to the moist soil surface of the ground (its natural habitat) which is a wide source of bacterial and fungal pathogens, the activity of PAL was the highest in it. Among the free radical scavenging enzymes (catalase, peroxidase and SOD) catalase seems to be the major enzyme exhibiting the highest specific activity. SOD (superoxide dismutase) also has recorded quite considerable activity in all bryophytes and a hornwort. Peroxidase showed the lowest activity in all these plants. It can be concluded from these observations that the bryophytes have well developed free radical scavenging mechanism.

12. The highest concentration of polyphenols was 1g 100g\(^{-1}\) fresh tissue in *P. intermedium* and that the lowest observed in *C. cavernarum* was 0.17 g 100g\(^{-1}\) fresh tissue. *A. wallichiana* and *T. hypophylla* have moderately similar concentration while hornwort, *A. subtilis* contains 0.39 g of polyphenols 100g\(^{-1}\) fresh tissue.

13. Qualitative results of polyphenols in bryophytes revealed that maximum number of phenolic compounds were found in *P. intermdium* i.e. 9 out of which 2 compounds were epicatechin+gallic acid and p-hydroxiquinolene and remaining 7 were unidentified. Thereafter, *T. hypophylla* contained 7 phenolic compounds which were unidentified. *A. subtilis* showed the presence of Epicatechin+gallic acid and gallic acid with four unidentified phenolics. In *A. wallichiana* 4 phenolic compounds and in *C. cavernarum* 3 bands were detected but could not be identified.

14. The results on quantitative estimation of flavonoids showed that the level of total flavonoids in *P. intermedium* and *T. hypophylla* was almost similar (respectively 2.96 and 3.05 mg of rutine equivalents g\(^{-1}\) fresh wt). The highest amount of total flavonoids was observed in *A. wallichiana* (3.17 mg of rutine equivalents g\(^{-1}\) fresh wt) while it was the lowest in case of *C. cavernarum* (1.95 mg of rutine
equivalents g\textsuperscript{-1} fresh wt). \textit{A. subtilis} contains 2.70 mg of rutin equivalents g\textsuperscript{-1} fresh wt of total flavonoids.

15. Qualitative estimation of flavonoids showed that \textit{P. intermedium} has maximum number (seven) of flavonoid components followed by \textit{A. wallichiana} with six flavonoids. \textit{A. subtilis} and \textit{T. hypophylla} contain four different flavonoids. The lowest variety of flavonids is found in \textit{C. cavernarum}. Compounds (iii), (i) and (ii) respectively in \textit{P. intermedium}, \textit{C. cavernarum} and \textit{T. hypophylla} are with approximate similar Rf values (0.63) and hence common to those bryophytes. While compounds (ii) and (i) are common with almost similar Rf values (0.71 and 0.72) in \textit{P. intermedium} and \textit{A. subtilis} respectively. All other flavonoid compounds are different and these all are not found in one or the other bryophytes studied.

16. Quantitative estimation of lipids revealed that \textit{P. intermedium} contains the highest amount of total lipids than rest of the bryophytes and \textit{T. hypophylla} is following with the lipid concentration of 2.41% dry wt. Other two liverworts i.e. \textit{C. cavernarum} and \textit{A. wallichiana} contain rather low levels of lipids. As \textit{A. subtilis} belongs to anthocerotae it lacks any oil bodies (Dembitsky and Rezanka, 1995) and hence may be the reason for its low lipid content.

17. Separation of phospholipids in solvent system II [Chloroform:Methanol : Distilled water (65:25:04 v/v)] was much better with more number than in solvent system I [Chloroform : Methanol : Acetic acid : Distilled water (85:15:10:04 v/v)]. All the five bryophytes contain 6-8 number of phospholipids except \textit{A. subtilis} which has only 3 (in II solvent system) and 5 (in solvent system I) compounds.

18. Qualitative analysis of neutral lipids showed that only few compounds appear to be common to all bryophytes. \textit{T. hypophylla} has the maximum 8 neutral lipids and \textit{A. wallichiana} has 5 lipids it while remaining three bryophytes have 3 neutral lipids. The neutral lipids identified are phosphotidyl glycerol in \textit{P. intermedium} and \textit{C. cavernarum} (Rf value 0.28). In \textit{T. hypophylla} lipid component with Rf value 0.44 (v) appear to be phosphotidyl ethanolamine.
19. The level of polar terpenoids was found higher than that of non-polar ones. *T. hypophylla* and *C. cavernarum* have similar type and higher level of polar terpenoids while that of *P. intermedium* is slightly lower in that case. *A. wallichiana* and *A. subtilis* have lower but rather similar concentration of polar terpenoids. *P. intermedium* contained higher concentration of non-polar terpenoids while next comes *A. subtilis*, *A. wallichiana* and *T. hypophylla* in that order. The lowest level of non-polar terpenoids is found in *C. cavernarum*. It is also found that *A. wallichiana* and then *A. subtilis* contain lower levels of polar + non-polar terpenoids and their highest concentration is observed in *T. hypophylla* (about 10 % dry wt).

20. Each of the bryophytes, except *C. cavernarum* (2 numbers), contains 8-10 number of terpenoid compounds.

21. Effect of aqueous extracts of bryophytes on germination and seedling growth in wheat and jowar without pre-sowing soaking of seeds reflected that in wheat usually inhibition of germination was observed at the initial stages (24h) by all the bryophytes. Similarly, during initial stages (24h) inhibition of germination of jowar seeds was observed in aqueous extracts of *P. intermedium*, *T. hypophylla*, *C. cavernarum* however, it was stimulated by *A. wallichiana* and *A. subtilis*. During the later stages (48-120h) no significant effect of all bryophytes is observed on germination of jowar seeds.

During initial stages seedling growth of wheat was markedly affected by aqueous extracts of bryophytes, *P. intermedium*, *A. wallichiana*, *C. cavernarum* and *A. subtilis* at all concentrations. However, during latter stages (48 to 120 h) the growth was remarkably stimulated (both shoot as well as root growth). *T. hypophylla* extract at all concentrations, except direct extract, however, exhibited inhibitory effects.

The results of aqueous extracts of different bryophytes on jowar seedlings without pre-sowing soaking treatment showed that during initial stages of seedlings' growth (0-24 h) there was inhibition by all bryophytes while, during the later stages (48-120 h) it was stimulated at all concentrations of *P. intermedium*, *A. wallichiana* and *T. hypophylla*. Higher concentrations of *C. cavernarum* and *A.
*subtilis* caused to decrease the rate of increase in size but still it was above the control.

22. Results of pre-sowing soaking of wheat seeds in distilled water for 6h and then treatment with aqueous extracts of bryophytes on germination showed that at all stages the lower concentration (3:1 and 1:1) of *P. intermedium*, *T. hypophylla*, *C. cavernarum* and *A. subtilis* stimulated germination but there was no significant effect by *A. wallichiana*. Higher concentration of *A. subtilis* extract caused to stimulate it in jowar at all stages of germination while inhibitory effect was observed by aqueous extracts of *T. hypophylla* and *C. cavernarum*. There was no significant effect of observed *P. intermedium* and *A. wallichiana* extracts on germination of this plant.

It was observed that all extracts of bryophytes at higher concentrations had inhibitory effect on wheat seedling growth during initial stages. However, during the later stages (48-120h), all concentrations of all bryophytes were either stimulatory or without any effect.

Extracts of *C. cavernarum* and *A. subtilis* appeared to be inhibitory for jowar seedlings at all stages (24-120h) and that of *A. wallichiana* was inhibitory particularly after 48 h of growth. Aqueous extract of *T. hypophylla* had stimulatory effects during 48 h and there was no consistency of results observed in case of aqueous extract of *P. intermedium*.

23. Pre-sowing soaking of wheat seeds in aqueous extracts of bryophytes demonstrated that there was stimulation in germination at the lower concentrations (3:1 and 1:1) of *P. intermedium*, *A. wallichiana* and *A. subtilis* extracts during the initial (24 h) stage. However there was significant effect on the germination of wheat seeds in *T. hypophylla* and *C. cavernarum* extracts. There was inhibition of germination of jowar seeds at all concentrations of *P. intermedium*, *A. wallichiana*, *T. hypophylla* and *C. cavernarum*. However, no significant effect of *A. subtilis* was observed. On the contrary aqueous extract of this bryophyte was stimulatory at higher concentrations.
Pre-sowing soaking of wheat seeds in aqueous extracts of *P. intermedium*, *A. wallichiana*, *T. hypophylla* and *C. cavernarum* at all concentrations resulted into inhibition of seedling growth during initial stages (24 h) whereas, there was stimulation at all concentrations by *A. subtilis*. During later stages of growth (48-120 h) all concentrations of *A. wallichiana*, *T. hypophylla* and *A. subtilis* were stimulatory while the lower concentrations of *P. intermedium* and *C. cavernarum* were slightly stimulatory.

During the early stages of growth there was significant inhibition of jowar seedlings by all concentrations of *P. intermedium*, *A. wallichiana*, *T. hypophylla* and *C. cavernarum*. Even the aqueous extract of *A. subtilis* has slight effect. Same was true during the later stages of growth (48-120 h) of jowar seedlings at all concentrations of *P. intermedium* and *A. wallichiana*. However, the higher concentrations of *T. hypophylla*, *C. cavernarum* and *A. subtilis* had slightly stimulatory effects.

It can be concluded that early stages of germination followed by initial phase of seedlings development appear to be sensitive to the bryophyte extracts during any mode of application (germination and seedling growth directly in aqueous extracts, pre-sowing soaking in distilled water and that in aqueous extracts of bryophytes). During later stages of seedling growth (48-120 h) most of the bryophytes at moderate to higher concentrations appear to be stimulatory.

24. *In vitro* antimicrobial activity of extracts of different bryophytes in different solvents revealed that the methanol extract of all the bryophytes possesses very good antifungal activity against almost all the fungal pathogens tested. Extracts in dichloromethane were next in that order. Ethanol extracts produced the lowest antifungal activity against three fungi (*A. flavus*, *F. oxysporium* and *M. phaseolina*). However, these extracts were found stimulatory for *F. oxysporium* and *M. phaseolina* at lower concentration (200 µl) and that inhibitory at higher concentration (400 µl).

25. All types of extracts of all bryophytes produced remarkable antifungal activity against *Aspergillus niger* and methanol extracts were the most effective than those of ethanol and dichloromethane.
Ethanol extracts of *P. intermedium* and *T. hypophylla* (at lower concentration) and *A. subtilits* and *A. wallichiana* (at higher concentration) were most effective while the methanol extracts of *A. subtilis* and dichloromethane extracts of *A. wallichiana* and *P. intermedium* demonstrated good antifungal activity.

26. *Aspergillus flavus* appeared quite resistant microbe to all extracts of bryophytes. Methanol extracts of *P. intermedium* and *A. wallichiana* exhibited a very good antifungal activity. Ethanol extracts of *A. subtilis*, *P. intermedium* and *A. wallichiana* produced rather good antifungal activity.

27. Dichloromethane extracts of almost all bryophytes were the most effective against *Fusarium oxysporium*. Ethanol extract of *T. hypophylla* was quite effective against *F. oxysporium*, while, methanol extract of *P. intermedium* was the most effective against this fungus.

28. The extracts of bryophytes did not produce any significant effect on *Microfomina phaseolina* except, for all extracts of *P. intermedium*, which showed remarkable effect on this fungus. Similarly, ethanol and methanol extracts of *A. subtilis* showed good *in vitro* antifungal activity.

29. *Erwinia carotovora* was more susceptible to ethanol extracts of all the bryophytes studied and *P. intermedium* extract was more effective. Dichloromethane extracts were much effective over methanol extracts of bryophytes *A. wallichiana* had greater activity. Methanol extracts *P. intermedium* and *T. hypophylla* were quite remarkable.

30. *Xanthomonas citri* did not respond so significantly to the extracts of bryophytes studied. Only higher concentration of ethanol extracts of *T. hypophylla* and *C. cavernarum* showed some antibacterial activity. Methanol and dichloromethane extracts, however, did not produce any inhibition of growth of these bacteria. Very poor antibacterial activity was recorded by *C. cavernarum* extracts in methanol and dichloromethane.