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J. Nat. Prod. Plant Resour., 2014, 4 (2):56-61
(http://scholarsresearchlibrary.com/archive.html)

ISSN : 2231 – 3184
CODEN (USA): JNPPB7

Studies on pigment chlorophyll isolation and estimation of different bryophytes for their biochemical properties

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ABSTRACT

Amount of chlorophyll concentration content studied in the six species of the classes anthocerotae, hepaticae and musci from the different localities of Purander fort, Maharashtra. It has been found that the total chlorophyll content is more in Pogonatum aloides Hedw. of musci compared to Anthoceros erectus Kash., Folioceros khandalensis Kash. of anthocerotae and Plagiochasma articulatum Kash. P. simulensis Kash. of hepaticae. The most maximum amount of chlorophyll-a is in P. simulnensis and minimum amount of chlorophyll-a is in A.erectus. Similarly, chlorophyll-b is higher in F. khandalensis and lower in P. articulatum. Chlorophyll activity is higher in hepaticae class. The light plays an important key role overall in this present chlorophyll estimation. Chlorophyll a:b ratio in P. articulatum shows maximum amount and less in A. erectus, whereas Funaria hygrometrica Hedw. has medium ratio.

Keywords: Chlorophyll, Biochemical properties, Isolation and estimation, Bryophytes.

INTRODUCTION

Chlorophyll is a green compound found in leaves and green stems of plant and it is also very important green pigment found in higher plants and bryophytes also. They found in damp shaded and humid localities with aquatic environment at plane and altitude region. It gives green colour to leaves and absorbs light that is used in photosynthesis. Chlorophyll is found in high concentrations in chloroplasts of plant cells. Chlorophyll molecules are specifically arranged in and around photosystem that are embedded the thalyakoid membrane of chloroplast. The chloroplast of bryophytes carries a mixture of chlorophyll and carotenoid pigments closely (Strokes, 1864). Breaking of chloroplast envelopes the formation of vesicles in thylakiods & quick degradation of linonenic acid (Adamoon and Coweorkers, 1988). This may be response in breaking of chloroplast envelopes of the formation of vesicles in thalyakoids and quick degradation linonenic acid. The light parameter changes the chlorophyll concentration in bryophytes. According to Kllio and Valane (1975) continuous light over a long period of time can looses their chlorophyll.

Kuiper (1978) shown the role of tocopherol an antioxidant helps to detect photoxidation process and protect chlorophyll amount and it play significant role to protect chlorophyll amount in bryophytes. When such a less temperature & high light conditions prevails,then the chlorophyll content protected. Photosynthesis observed in bryophytes an optimum amount and decreases the rate rapidly.Hence, high CO_2 and O_2 ratio declines with ethylene

production and stimulates chlorophyll synthesis with high photosynthesis ratio. Photorespiration would serve as energy source to protect the chlorophyll form over excitation by the intensity of light (Bidwell,1979). Zeher (1979) recorded the high rate of photosynthesis & respiration in winter season.

Total chlorophyll increases in summer season (Kreshaw and Webber, 1986). The stroma of thalyakoids destroyed much like destruction in dark shady places. Therefore, photoperiod can play a significant an important role in development, productivity, and acclimation and other aspects of bryophytes in classes like Anthocerotae, Hepaticae and Musci respectively (Scarnio,1986). Bryophytes cannot changes their leaf thickness but possible for the changes in chlorophyll concentration. In the blue green light environment can causes major increases of the content of chlorophyll (Czeczuga,1987).

As light intensity changes with climate & seasons per year. Some bryophytes changes their chlorophyll concentration based on the amount of leaf. The amount of green tissue of concentrated chlorophyll per dry weight is higher (Mishler and Oliver, 1991). The bryophytes grows in low light which will enhances the chlorophyll-b concentration and thus chlorophyll a:b ratio reduced and those lacks the lights it tuned in to red colors and photosynthetic apparatus permanently damaged (Tuba et al.,1996). Totally, High intensities damages chlorophyll and DNA. Chlorophyll degradation is routinely observed response to stress chiefly in elevated concentrations of various heavy metals. Thus changes of chlorophyll content and pigment ratio are important indicators.

MATERIALS AND METHODS

Fresh plant collected forms the different sites of Purandar at different altitude. Well grown fresh bryophytes were collected with their fresh thallus in plastic polythene bags. The material of each collected plant cleaned by removing soil and allowed to air dried and allowed to store for preservation in petri plates for specific room temperature. The estimation was done as per Arnon (1949) method. 0.5gm fresh plant material were homogenized in 20 ml of 80% chilled acetone with the help of mortar and pestle in dark. A pinch of MgCO3 powder added. The extract filtered through Whatman no.1 filter paper. Final volume of the filtrate was made to 100 ml with 80% acetone in conical flask wrapped with black carbon paper. Absorbance readings done at the 645 nm and 663 nm wavelengths respectively with 80% acid used as a blank.

In the Spectrophotometric analysis shows that Funaria hygrometrica shows that the maximum variation at 645 nm and invertly, Plagiochasma areticulatum shows minimum amount chlorophyll and similarly at 663nm, Plagiochasma simulnensis shows maximum amount of chlorophyll ans minimum in Pogonatum aloides due to as per light effect.

RESULTS AND DISCUSSION

The result of present investigations of chlorophyll-a, chlorophyll-b and total chlorophyll and its ratio shown in the following table No.1.

Tables No.1 : Chlorophyll-a, chlorophyll-b and total chlorophyll and its ratio

Sr. No.	Name of species	Chlorophyll-a	Chlorophyll -b	Total Chloro-phyll	Chlorophyll a:b ratio
1)	*Anthoceros erectus* Kash.	0.128	0.084	0.146	1.52
2)	*Folioceros khandalensis* Kashs	0.165	0.113	0.193	1.46
3)	*Plagiochasma simulnensis Kash.*	0.229	0.052	0.169	4.40
4)	*Plagiochasma articulatum* Kash	0.359	0.043	0.228	8.34
5)	*Pogonatum aloides* Hedw.	0.349	0.058	0.237	6.01
6)	*Funaria hygrometrica* Hedw.	0.212	0.051	0.159	4.15

Concentrations of chl-a is more than chl-b in all the given species in table no.1. But in the class of Hepatacea, Plagiochasma articulatum shows the maximum amount of chl-a while in the class Anthocerotae Anthoceros erectus shows less amount of chlorophyll as compared to amount of chl-b out of six species. Maximum amount of chl-b were occurred in the Folioceros khandalensis and minimum in Plagiochasma simulnensis. Maximum total chlorophyll obsereved in the Pogonatum aloides and less in Anthoceros erectus**.** Chlorophyll a:b ratio recorded maximum in Plagiochasma articulatum. Less in Anthoceros erectus whereas Funaria hygrometrica has medium ratio. All the graphs A, B and C shows various chlorophyll concentrations as per their values as per result table with

different species. The total chlorophyll amount is higher in Pogonatum aloides and lower in Anthoceros erectus. The chlorophyll amount calculated respectively by following formula with chl-a,chl-b and total chlorophyll as per Arnon (1949) method.

Photo plates of collected bryophytes from Purandhar fort, Maharashtra, India

Anthoceros erectus Kash

Folioceros khandalensis Kash.

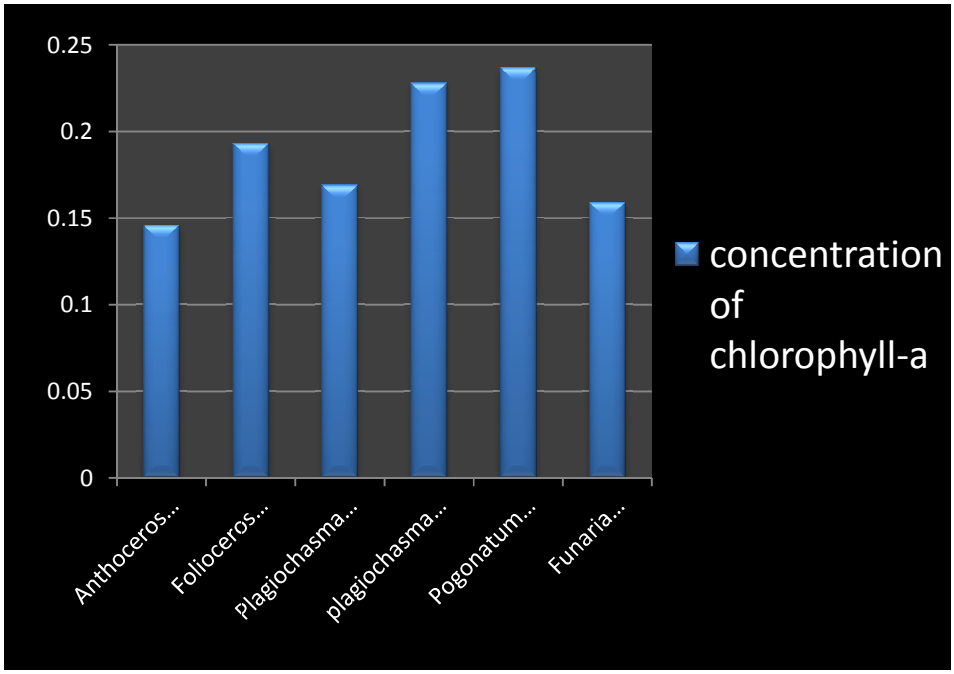
Plagiochasma Kash.

Plagiochasma simulnensis Kash.

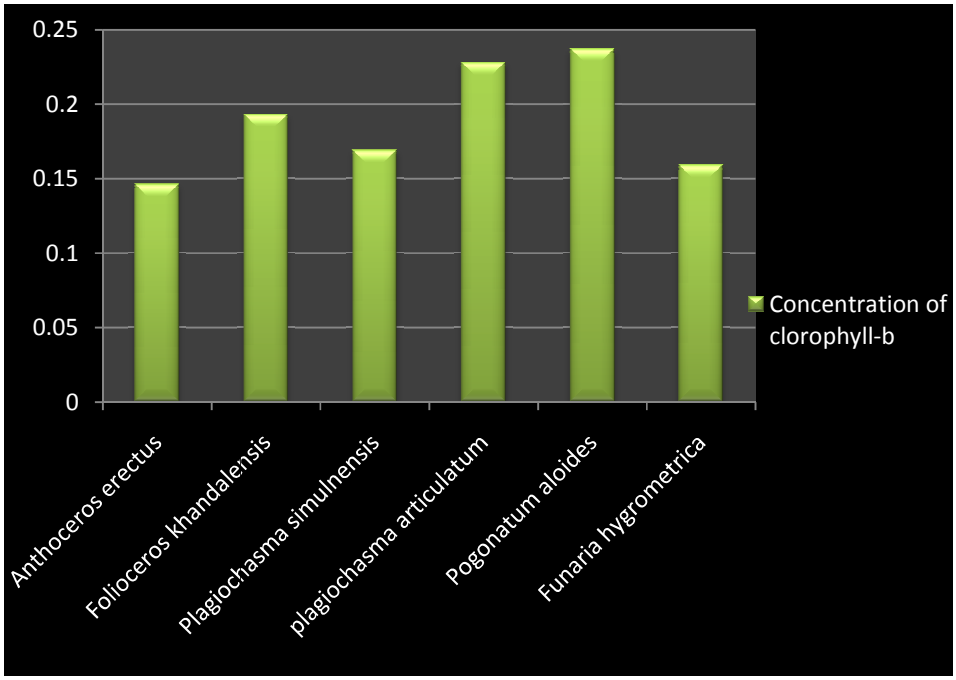
Funaria hygrometrica Hedw.

Pogonatum aloides Hedw

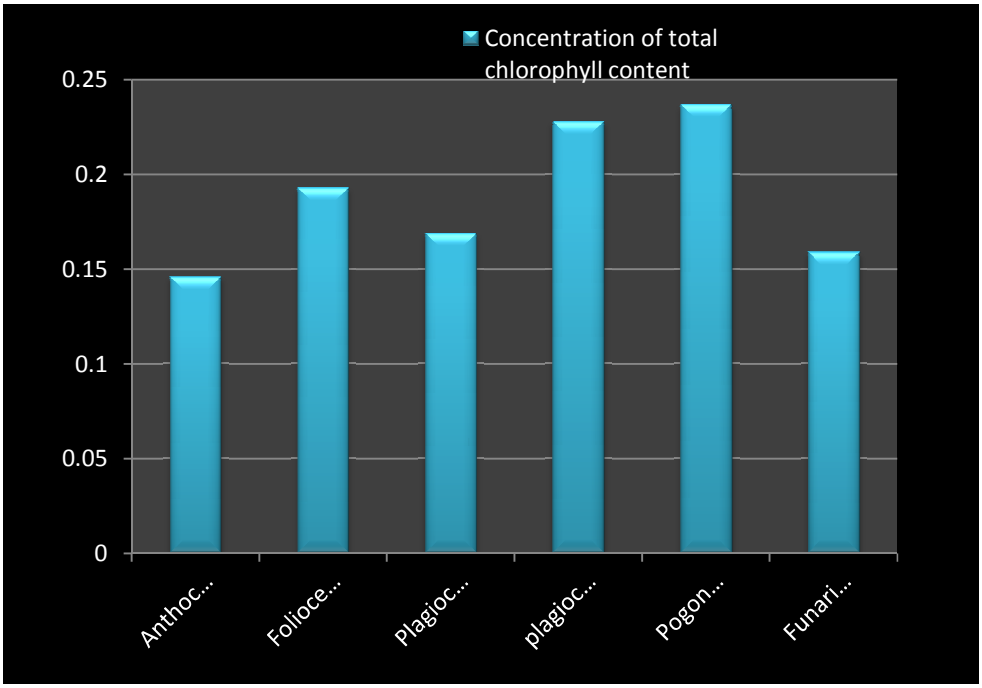
Graph-A showing concentration of chlorophyll-a



Graph-B showing concentration of chlorophyll-b



GRAPH C: Showing concentration of 'Total chlorophyll content' in collected plants



CONCLUSION

The plant material collected from different places at Purander region during period of shiny morning, the plant specimen rinsed, cleaned, dried and store paper bags at room temperature. Before the chlorophyll estimation soil is totally separated from the associated thallus form each plant.

In this Chlorophyll Estimation Arnon (1949) method is used for the chlorophyll estimation. The result shows a significant variation and the chlorophyll amount in each collected species of bryophytes. Chl-a, Chl-b and total chlorophyll amount shows different values with maximum and minimum amount proportion.

The light (Blue, Green, White & Red) plays a significant role in this chlorophyll estimation. The amount of chlorophyll is changes in different classes of bryophytes due to light effect. The light parameter changes the chlorophyll concentration in selected plants due to different climatic conditions. Photoperiod changes due to low light as per seasonal changes. Low light increases chl-b concentration. Low and high light effects on chlorophyll molecules and affects on the total chlorophyll concentration amount. Leaves content changes, their morphology changes due to high light and low light. The increase and reduction of chlorophyll amount in every plant. During blue green light chlorophyll amount enhances. Continue supply of light affects on growth of plants, hence chlorophyll amount decreases immediately. High light also make molecular and genetic changes and this affects on DNA structure and plenty of loss of chlorophyll.

Acknowledgements
Authors are sincerely grateful to UGC, University Grants Commission, Western Circle, Pune for providing financial support. We are indebted to Hon. Dr. C.V. Murumkar, The Head and Principal, Tuljaram Chaturchand College, Baramati for providing necessary research facilities.

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