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Research Article

Influence of Environmental Factors on Tetraspores Shedding and Diurnal Periodicity in *Padina tetrastromatica Hauck*. (Phaeophyceae) along the Visakhapatnam Coast, East Coast of India

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Abstract: Studies on the tetraspores shedding were made for a period of one and half year from August 1993 to January 1995 in *Padina tetrastromatica* growing along the Visakhapatnam coast. Experiments were conducted on the effect of environmental factors such as exposure to air (desiccation), Salinity, light intensity, photoperiod and wavelength on tetraspores shedding and diurnal periodicity was studied under laboratory conditions. The spore shedding in this brown alga with peak shedding of tetraspores in submerged conditions of fronds, 35‰ salinity, 1000 lux light intensity, white light and 24h light (at low light intensity of 1000 Lux) were found to be optimum conditions for peak shedding of tetraspores in *Padina tetrastromatica* growing along the intertidal region of Visakhapatnam Coast. Environmental parameters such as exposure to air, salinity and different wavelengths did not affect the diurnal periodicity in *Padina tetrastromatica*. But periodicity in tetraspores shedding was changed in different light intensities and different light and dark cycles.
Keywords: *Padina tetrastromatica* Hauck, Tetraspores shedding, Diurnal periodicity, Environmental factors, Jodugullapalem, Visakhapatnam coast..

INTRODUCTION

Padina tetrastromatica is one of the most important members of the algae in Visakhapatnam Coast with respect to their relative size, abundance and its distribution on the rocky shores. The importance of Indian marine algae as food lies in their essential amino acids, vitamins and minerals [1]. This alga is considered to be a potentially useful seaweed having food (salad vegetable), Pharmaceutical and fertilizer value. Despite the recognised importance of this alga, information on the major environmental factor, which effects its growth, tetraspores shedding is fragmentary. In order to gain an insight of tetraspores shedding capacities, the effect of desiccation, salinity, light intensity, photoperiod and wavelength, have been followed under controlled conditions in the laboratory. Although at *Padina tetrastromatica* is common on the Visakhapatnam Cost, the quantity is inadequate for this commercial use. The above studies are very essential before undertaking cultivation of any alga. The purpose of the present study is to ascertain optimal level for those environmental parameters which are most important in influencing liberation of tetraspores in *Padina tetrastromatica*. The results obtained on these aspects are presented and discussed in this paper. Several authors studied the spore shedding and other aspects on brown algal members in different geographical regions of the world [2-8]. Studies on sporulation play a vital role in the field of mariculture to generate the algal populations in the natural habitats. In the present investigation studies were made on the tetraspores shedding from *Padina tetrastromatica* in different environmental parameters at in Jodugullapalem along the Visakhapatnam coast was made for a period of one and half years from August 1993 to January 1995, is presented in this paper.

MATERIAL AND METHODS

Visakhapatnam is situated on the east coast of India between the latitude 17˚ 40’ 30’’ and 17˚ 45’ N longitudes 83˚ 16’ 25” and 83˚ 21’ 30”E. The coastline is sandy with outcrops of rocky boulders in different regions. Depending upon the physical nature of the substratum four stations were selected in an earlier investigation [9]. Materials for this study were collected during the spring tide periods from Jodugullapalem region where large accessible boulders occur with dense growth of algae. *Padina tetrastromatica* was collected for carrying out the laboratory experiments during the years August 1993 to January 1995.Tetraspores liberation experiments were carried out with the fertile material, washed with sterilized sea water and placed in

the petriplates filled with sea water. The tetraspores liberation experiments were conducted for 24 hours at room temperature (30 ± 2^0C) and petri-dishes were illuminated by fluorescent lamps at 1000 lux for 8 hours during the day time from 10 to 18 hours. The tetraspores liberated daily counted and after 24 hours were transferred to a measuring cylinder with a pipette. The tetraspores suspension was then diluted to a known volume depending upon the Quantity of tetraspores liberated in the Petri-dishes. Taking a subsample of 1c.c tetraspores suspension into a plankton counting chamber, the tetraspores were counted. Average values of three counts were used for computing the tetraspores output in each experiment. Fresh weight of the thalli was taken and the tetraspores output was expressed as tetrospores / gr. Fr. Wt. /day. Data on the diurnal changes in tetraspores shedding were collected by changing the material kept in the Petri-dishes at four hours intervals. The tetraspores liberated at 4 hours intervals i.e. at 22h, 02h, and 06h. 10h, 14h and 18h, were counted as described above. Ten experiments (replicates) were conducted with *Padina tetrastrornatica* to study the diurnal changes in tetraspores shedding.Experiments were conducted on the effect of environmental factors on tetraspores shedding and diurnal periodicity of tetraspores shedding from this marine alga. In the experiments conducted to study the exposure to air, the fronds were blotted to remove the water on the surface of the fronds and exposed to air in the laboratory and also in the open air during the day time. At the time of conducting these experiments the temperature in the laboratory was 30± 2°C and the relative humidity varied from 60 to 65%. At 15 minute intervals (0, 15, 30, 45, 60, 75 minutes) the materials thus exposed to air were transferred to Petri-dishes filled with seawater and the tetraspores output was estimated after 24 hours as mentioned in the earlier works (Subba Rangaiah, 1983). Seawater collected from the inshore area was adjusted to 100‰ salinity by exposing to sun light to make up the stock solution. Lower grades were prepared from this stock solution by the addition of requisite quantity of distilled water. Tetraspores output was estimated at 0‰, 05‰,15‰ 25 ‰ 35 ‰ 45 ‰ 55‰, 65‰ and 75 ‰ salinities, maintaining the Petri-dishes at room temperature 30 ± 2^0C under 8 hours day length with 1000 lux light intensity. Effect of light intensity on tetraspores output were investigated at room temperature using light intensities of 0 (dark), 1000 lux, 2000 lux, 3000 lux and 4000 lux light. To study the effect of photoperiod on the tetraspores shedding, experimental sets were subjected to 0:24, 4:20, 8:16, 12:12, 16:8, 20:4, 24:0(L: D cycles) in separate light and dark chambers. Based on the changes observed in the tetraspores output per day, experiments on diurnal periodicity were conducted selecting certain periods of exposure to air (0,15,30,45 and 60 minutes), salinities (25 ‰ 35 ‰ 45 ‰ 55‰ and 65‰), light intensities (0 (dark), 1000 lux, 2000 lux and 3000 lux light intensity), photoperiod (0:24, 4:20, 8:16, 12:12, 16:8, 20:4, 24:0) (L: D cycles) in separate light and dark chambers. In all above experiments, the data collected were expressed as tetraspores per gr.fr.wt/day, to observe the quantity of tetraspores liberation under diverse environmental conditions.

RESULTS

Data collected on the influence of environmental factors such as exposure to air (desiccation), salinity, light intensity, photoperiod and wavelength on tetraspores shedding and diurnal periodicity were presented in the Fig. 1,2, 3,4,5,6,7,8, 9 and 10 respectively.

Factors influencing on tetraspores shedding

Data collected on the effects of desiccation, salinity, light intensity, Photoperiod and wavelength observed on the liberation of tetraspores are shown in figs. 1, 2, 3, 4 and 5 respectively.

Exposure to air (Desiccation)

Changes observed in the tetraspores output of *Padina tetrastromatica* in control (O minute exposure) and at different periods of exposure to air at room temperature (30 ± 2^0C) in the laboratory is shown in Fig 1. In experiments conducted in shade i.e. in the laboratory, tetraspores shedding was seen up to75 minutes exposure (Fig. 1). Maximum spore output was observed in control where fronds were submerged for 24 h duration and the number of tetraspores liberated decreased with increase in the duration of exposure of fronds to air at laboratory temperature. The output of tetraspores was very low from the fronds exposed to 15, 30, 45 and 60 minutes respectively and no tetraspores liberation observed at 75 minutes of exposure.

Salinity

Effects of salinity on tetraspores shedding of Padina tetrastromatica are presented in Fig. 2. Tetraspores output varied markedly in different salinities of seawater tested and there was no liberation at 0 ‰, 15‰ and 75 ‰ salinities. The tetraspores liberation was observed from 25 to 60 ‰ with minimum number of tetraspores at25 and 60 ‰ salinities. Peak output of tetraspores was found at 35 ‰. But considerable number of tetraspores was also seen liberating from the fronds at 25‰ and 45 ‰ salinities (Fig. 2).

Light intensity

The quantity of tetraspores liberated from the fronds of *Padina tetrastromatica* exposed to dark to three different light intensities ranging from 0 lux to 4000 lux are presented in Fig. 3. Tetraspores output was varied in different light intensity raging from 0 Lux to 4000 lux. Peak shedding of tetraspores was found at 1000 lux and considerable number at 2000 lux light intensity and from there onwards the quantity of tetraspores liberated decreased gradually. Very low output of tetraspores was observed at 0 Lux and 3000

Lux and tetraspores output was totally inhibited at 4000 Lux light intensity.

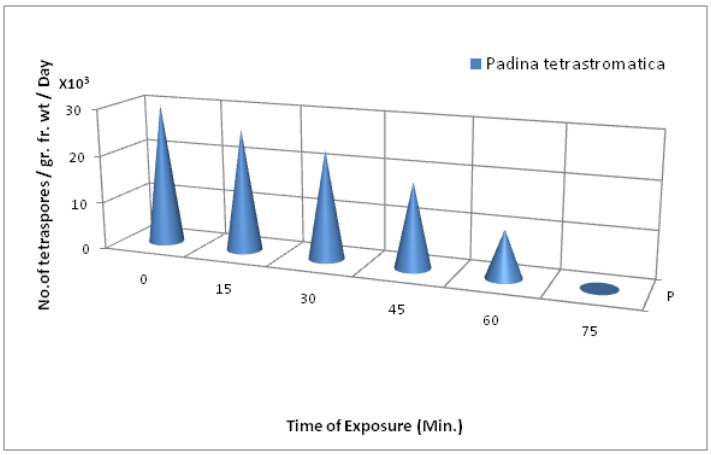

Fig. 1: Effect of desiccation on the tetraspores shedding of *Padina tetrastromatica*

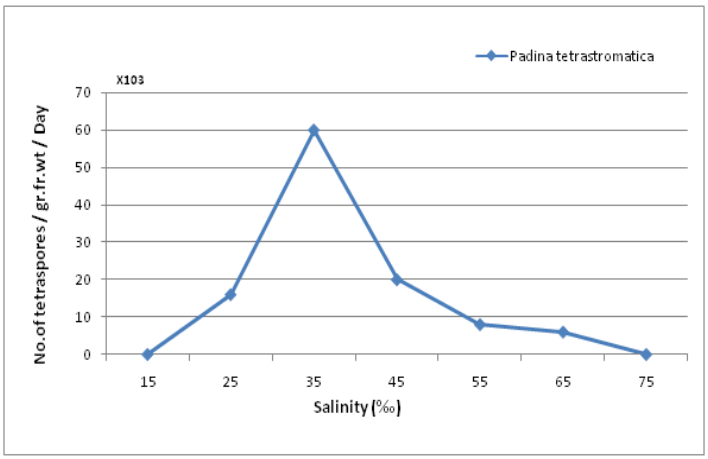

Fig.2.Effect of Salinity on the tetraspores shedding of *Padina tetrastromatica*

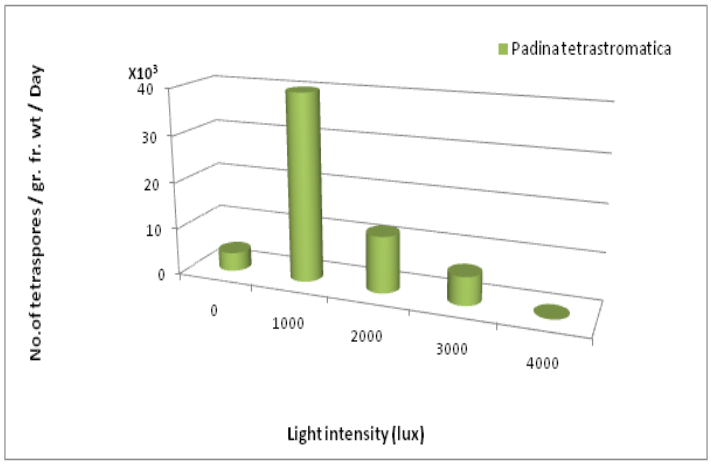

Fig. 3: Effect of light intensity on the tetraspores shedding of *Padina tetrastromatica*

Photoperiod

The effects of light and dark regimes on tetraspores shedding are presented in Fig. 4. These photoperiod experiments on *Padina tetrastromatica* was conducted at light intensity 1000 lux. Peak output of tetraspores varied with the duration of light intensity received by the plants subjected to different L: D regimes (Light: Dark). In *Padina tetrastromatica* at light intensity 1000 lux ,tetraspores output increased from 0 : 24 (L:D cycle) with maximum output at 24 : 00 (L:D cycle). Tetraspores output decreased with further increase in the photoperiod at this light intensity.

Wavelength

Effects of different wavelength of light on tetraspores production in *Padina tetrastromatica* are presented in Fig. 5. Maximum shedding of tetraspores was noticed in control (White light) and tetraspores output was decreased gradually from yellow to red.

Factors influencing diurnal periodicity

Data collected on the effects of desiccation, salinity, photon flux density, Photoperiod and Wavelength observed on the diurnal periodicity in the liberation of tetraspores are shown in figs. 6, 7, 8, 9 and 10 respectively.

Exposure to air (Desiccation)

Data obtained on the diurnal periodicity by exposing the fronds of *Padina tetrastromatica* from control (submerged condition) to 60 minutes. Peak shedding of tetraspores was observed in the fronds of 0 minute exposure between 06:00 h and 10:00 h without any change in the normal shedding period (Fig.6). The quantity of tetraspores liberated decreased in the alga with increased in the duration of exposure. Considerable tetraspores liberation was observed in between 10:00 h and 14:00 h.

Salinity

Influence of five different salinities on the diurnal periodicity of tetraspores release is depicted in Fig.7. The peak output of tetraspores was observed in *Padina tetrastromatica* between 0600 and 1000 h in salinities ranging from 35- 45‰ salinity without any shift in the time of peak shedding of tetraspores in a day. But at 25 and 65‰, the diurnal variations are not prominent since very less number of tetraspores was liberated from the fronds.

Light Intensity

Diurnal periodicity varied in the different light intensities. Liberation of tetraspores from the fronds kept in dark and in four different light intensities viz. 0 lux, 1000 lux, 2000 lux and 3000 lux are presented in the Fig. 8. The peak shedding of tetraspores was observed between 06:00h to 10:00h in 1000 Lux and 3000 Lux light intensities. Prominent peak with more number of tetraspores was obtained between 10:00h to 14:00h in 2000 Lux light intensity.

Photoperiod

Diurnal periodicity varied in the different photoperiods. Liberation of tetraspores from the fronds kept in seven different photoperiod is presented in the Fig. 9. The peak shedding of tetraspores was observed between 06:00h to 10:00h in 24L: 0D. This normal pattern of tetraspores shedding was changed in photoperiod 0L: 24D and 04L: 20D, maximum tetraspores shedding between 10:00h to 14:00h.

Wavelength

No change in the diurnal periodicity was observed at different wavelengths are presented in fig.no.10. Peak number of tetraspores was liberated in white light wavelength between 06:00h and 10:00hs.

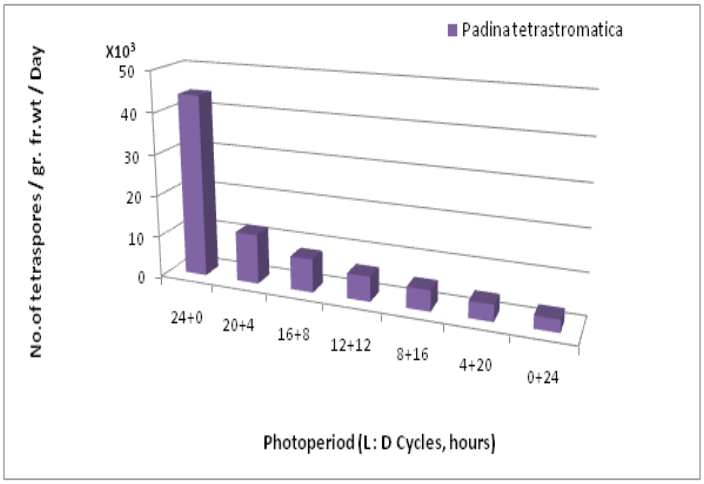

Fig. 4: Effect of Photoperiod on the tetraspores shedding of *Padina tetrastromatica*

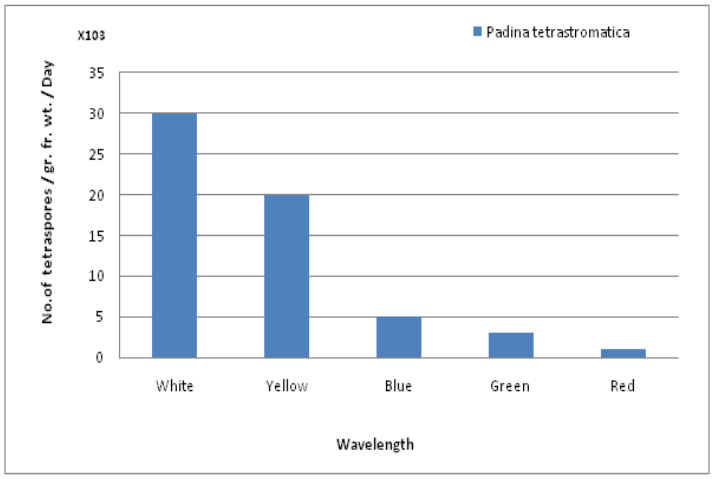

Fig. 5: Effect of wavelength on the tetraspores shedding of *Padina tetrastromatica*

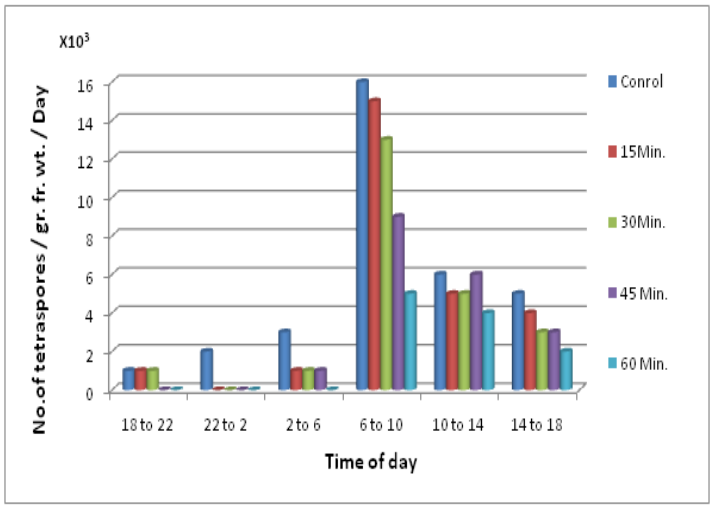

Fig. 6: Effect of different desiccations on diurnal periodicity in the liberation of tetraspores of *Padina tetrastromatica*

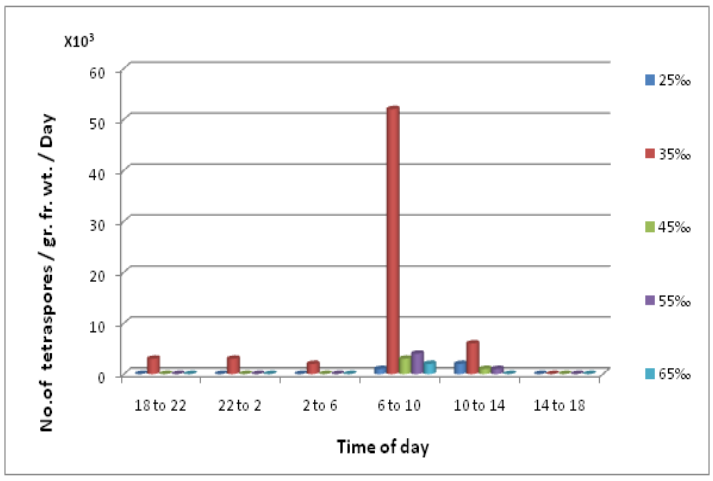

Fig. 7: Effect of different salinities on diurnal periodicity in the liberation of tetraspores of *Padina tetrastromatica*

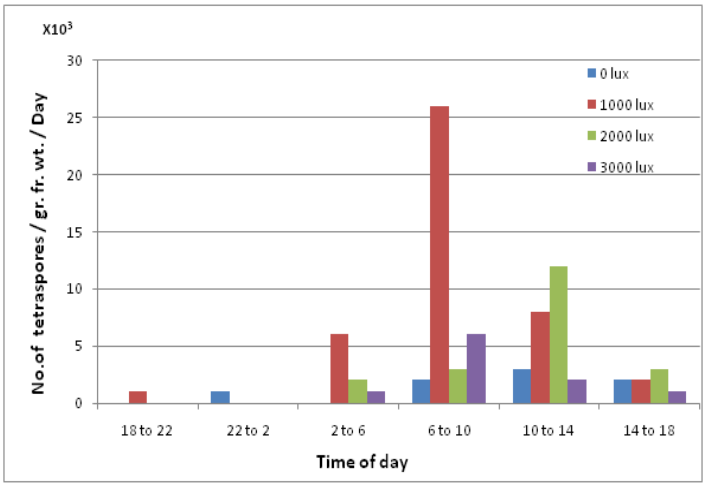

Fig. 8: Effect of different light intensities on diurnal periodicity in the liberation of tetraspores of *Padina tetrastromatica*

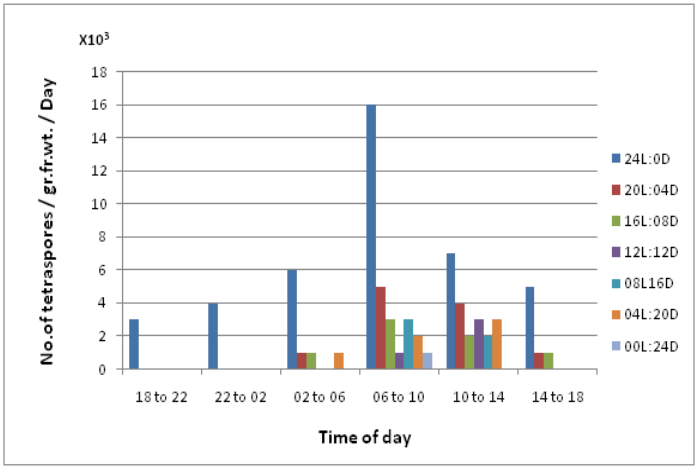

Fig. 9: Effect of different photoperiods on diurnal periodicity in the liberation of tetraspores of *Padina tetrastromatica*

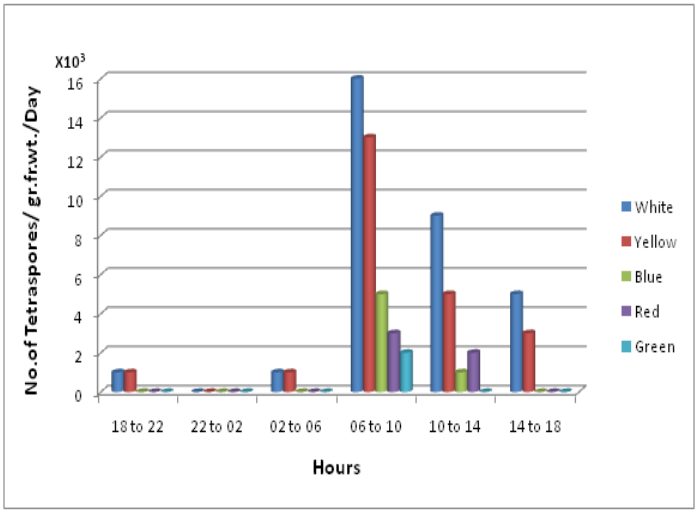

Fig. 10: Effect of different wavelengths on diurnal periodicity in the liberation of tetraspores of *Padina tetrastromatica*

DISCUSSION

In the present study, information was gathered on tetraspores shedding of *Padina tetrastromatica* growing in the infra littoral fringe zone of the intertidal region of Visakhapatnam coast. The variations of spore production in the influence of environmental factors on tetraspores shedding and diurnal pattern of tetraspores liberation were studied by conducting the experiments under laboratory conditions. In the present study tetraspores shedding abilities of *Padina tetrastromatica was* influenced by some environmental factors such as exposure to air (desiccation), salinity, light intensity, photoperiod and wavelength. Withstanding ability of different marine algae to these environmental parameters depend on the vertical distribution of algae on rocky surfaces. The eco-physiological investigation of spore shedding on Indian marine algae was studied by several authors [10-15] and a study on spore shedding of brown algae was fragmentary [8, 16-18]. In the present study tetraspores shedding in *Padina tetrastromatica* was observed only for 60 min inside the laboratory with peak tetraspores shedding in 0 Lux light intensity (dark). Spore output gradually decreased in the fronds of *Padina tetrastromatica* (Fig.1), indicating that emergence of fronds during low tides adversely affects the spore liberation and that the submerged condition is favourable in this brown alga. These observations on *Padina tetrastromatica* agree with the findings on *S. ilicifolium* [8] and also depend on the distribution of this alga in the intertidal habitat. Salinity of the seawater influences tetraspores shedding in *Padina tetrastromatica* the optimum salinity range observed for the maximum shedding in *Padina tetrastromatica* was 35 ‰. Several studies reveals the effect of salinity on tetraspores shedding and observed different optimum ranges [8, 10, 14-17, 19]. Tetraspores liberation in *Padina tetrastromatica* occurred in the light intensities ranging from 0 to 4000 Lux with peak shedding at 1000 Lux. Similar trend was reported [8, 18, 20], observed that the time of peak liberation of spores in the fronds of *Gloiopeltis* species exposed to air for 2 to 6 h was accelerated by 10 h. In the present study in *Padina tetrastromatica* showed delay in the peak shedding of tetraspores for about 4h in the fronds exposed for 60 minutes. In the previous studies made by [21] on *Gelidium pusillum*, where 4h delay in spore shedding was observed in fronds exposed for 45 minutes. Variations in the salinity did not affect diurnal periodicity pattern in the members of Dictyotales and species of *Sargasssum* [8, 16]. In the present study also maximum numbers of tetraspores were liberated at 1000 lux light intensity. The combined effects of light intensity and day length or photoperiod may also play an important role in spore liberation. In the present study, long day conditions are favourable for tetraspores shedding in *Padina tetrastromatica*. From the above observation, it is clear that these interacting effects show that the amount of energy received by plants influences spore release and this periodicity in the discharge of spores is due to photosynthetic effect as described by [22]. Peak output of tetraspores was observed in white light in *Padina tetrastromatica.* As similar result reported by [23] in *Wrangelia argus*. Minimum numbers of spores were released in yellow light, in contrary to the observation made by [24] in Gigartinales and [23] in *Centroceros clavulatum* and *Polysiphonia platycarpa*.

The results obtained on factors influencing diurnal periodicity are very interesting. The effect of light intensity and quality of light controlled the peak liberation of tetraspores in a day are changed to 10:00 to 14:00 in the experiments conducted with *Padina tetrastromatica* (Fig. 8 and Fig. 9). But exposure to air (Desiccation), salinity and wavelength have no influence on the normal daily periodicity in *Padina tetrastromatica* (Fig. 6, 7 and 10). Changes observed in the time of peak output of spores are tabulated below to show the relationship between the factors like desiccation, salinity, light intensity, photoperiod and wavelength on diurnal periodicity of tetraspores liberation.

Table 1:Time of maximum liberation of tetraspores in a day of *Padina tetrastromatica*

1. Desiccation	
Control (Zero)	6 AM – 10 AM
15 Minutes	6 AM – 10 AM
30 Minutes	6 AM – 10 AM
45 Minutes	6 AM – 10 AM
60 Minutes	6 AM – 10 AM
2. Salinity	
15‰	6 AM – 10 AM
25‰	6 AM – 10 AM
35‰	6 AM – 10 AM
45‰	6 AM – 10 AM
55‰	6 AM – 10 AM
65‰	6 AM – 10 AM
3. Light intensity	* 10 AM – 2 PM
0 Lux (Dark)	6 AM – 10 AM
1000 Lux	* 10 AM – 2 PM
2000 Lux	6 AM – 10 AM
3000 Lux	
4. Photoperiods	
24 D: 00 L	* 10 AM – 2 PM
20 D: 04 L	* 10 AM – 2 PM
16 D: 08 L	6 AM – 10 AM
12 D: 12 L	6 AM – 10 AM
08 D: 16 L	6 AM – 10 AM
04 D: 20 L	6 AM – 10 AM
00 D: 24 l	6 AM – 10 AM
5. Wavelength	
White	6 AM – 10 AM
Yellow	6 AM – 10 AM
Blue	6 AM – 10 AM
Green	6 AM – 10 AM
Red	6 AM – 10 AM

* Indicates, change over the normal periodicity.

The present study agrees with the observations of [25] on Gigartinales and [21] on some Gelidiales. But, however, 4h delay was observed in the fronds exposed for 45 minutes in *Gelidium pusillum* [21]. Different salinities did not affect diurnal periodicity pattern in the members of Gigartinales and Gelidiales studied respectively by [21] and [25]. Similar trend was observed in the present study also where there is no effect of salinity on diurnal variation in the tetraspores shedding in *Padina tetrastromatica*. Data collected on tetraspores shedding under different light and dark conditions indicate that the spore shedding exists continuously for 2 to 3 days in *Padina tetrastromatica.* In the present study, it is clearly seen that the rhythm of spore shedding is persistent in continuous light, continuous dark and also in different light and dark conditions. The above study clearly indicates that submerged conditions, long days, low light intensities and normal seawater are favourable for the maximum liberation of tetraspores in *Padina tetrastromatica*. The observations of the present investigation agree with the above findings. When the fronds of *Padina tetrstromatica* exposed up to 1000 lux, 2000 lux and 3000 lux, there was no change in the peak period of shedding of tetraspores. In this respect the present study agrees with the results of [8, 16, 23, 26]. It seems that light intensity did not have any effect on the diurnal periodicity of tetraspores shedding in Padina tetrastromatica.

CONCLUSION

The Present study on different factors, it can be concluded that the submerged condition of fronds, light intensity of 1000 lux, salinities around normal sea water (35 ‰), 24h light photoperiod and white light wavelength are favourable for maximum tetraspores shedding of *Padina tetrastromatica* growing along the intertidal region of Visakhapatnam coast. These experimental findings closely agree with the environmental conditions existing in the intertidal habitat at Jodugullapalem of the Visakhapatnam coast. It is interesting to note that the quantity of tetraspores liberated in *Padina tetrastromatica* of the present study is almost less than half when compared to the studies made by [10]. This change may be due to increase in the temperature (2-3^{o}C) in the nature, and indiscriminate discharge of industrial effluents in to the sea. If this process continues, we do hope that in future there will be a drastic change in the seaweeds of Visakhapatnam coast towards decrease in the vegetation as well as in tetraspores shedding.

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