

THE EFFECT OF LIGHT ON THE GROWTH  
OF SPORELINGS OF THE RED ALGAE  
*ANTITHAMNION PLUMULA* AND  
*BRONGNIARTELLA BYSSOIDES*

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(Plate I and Text-figs. 1 and 2)

It was recently suggested (Boney & Corner, 1960, 1962*a*) that, contrary to the theory of complementary chromatic adaptation, the accessory pigment phycoerythrin might not be involved in photosynthesis by sporelings of the intertidal red alga *Plumaria elegans* (Bonnem) Schm. but is used primarily as a means of protecting the plant from excess green light in the wave-band 500-540 m $\mu$ . This work, however, was done with one species only, and concerning the wider question of marine red algae in general, it seemed possible that the role of phycoerythrin might vary with ecological distribution. Thus, plants completely submerged and thereby excluded from most red light would use their accessory pigments for photosynthesis (energizing chlorophyll *a* indirectly); but plants adapted to long periods of normal daylight would be similar to *Plumaria* in relying to a much greater extent on chlorophyll *a* alone for photosynthesis, and use their accessory pigments as protection against inhibitory green light.

Evidence in support of this view has now been obtained in the present work, in which a comparative study has been made of the effect of light on the growth of sporelings of *Antithamnion plumula* (Ellis) Thur., using plant material from a persistent growth in the 'Drake's Island' Tank at the Plymouth Laboratory (in which situation it is exposed to almost normal daylight conditions during the summer months): and those of the sublittoral species *Brongniartella byssoides* (Good. et Woodw.) Schm.

METHODS

*Plant material and spore settlement.* Fruiting tufts of *Antithamnion plumula* bearing tetrasporangia were taken from the persistent summer growth at the Plymouth Laboratory: those of *Brongniartella byssoides*, also bearing tetrasporangia, were dredged from the sea-bed in the vicinity of the Duke Rock

buoy in Plymouth Sound (depth 10–12 m). In both instances settlement of spores was carried out as described earlier (Boney, Corner & Sparrow, 1959; Boney & Corner, 1959).

*Measurement of growth.* Sporelings of *Antithamnion plumula* show a pattern of early development similar to that described for sporelings of *Plumaria elegans*, and the method of measuring cell production was that described in recent work (Boney & Corner, 1962*b*). The development of sporelings of *Brongniartella byssoides* differs from the other two representative species in that the first cells of the erect filament give rise to pericentral cells with the subsequent formation of a corticated filament (Pl. 1*a*). Cell production was estimated by counting the number of 'tiers' of cells constituting the separate 'divisions' of the corticated filament, and then multiplying the number of 'tiers' by the number of cells, axial and pericentral, constituting the tier (= 5 cells/tier). In addition, the uncorticated cells immediately below the apical meristematic cell were counted and added to the cell total. Duplicate samples, each of 200–300 sporelings, were used in all experiments. The cell production per 100 sporelings per day was then calculated.

*Culture medium.* The mixture of one part Erd-Schreiber medium (Føyn, 1934) to three parts sea water was used in all experiments, as in our earlier study (Boney & Corner, 1962*a*).

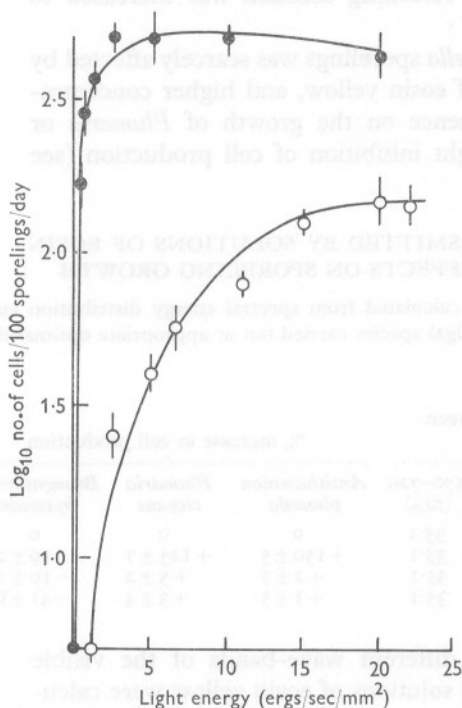
#### SCREENING EXPERIMENTS

##### *Optimum light energy*

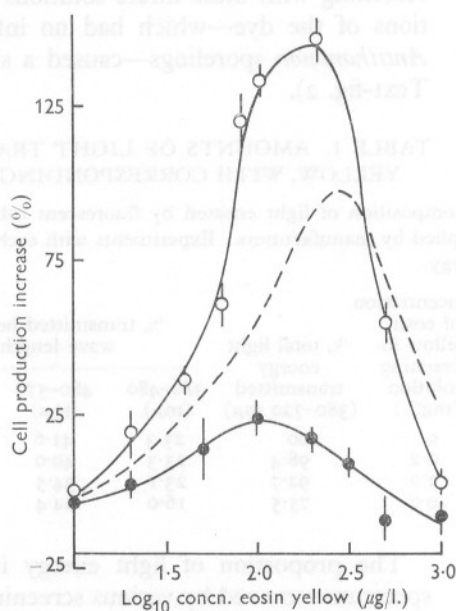
As a preliminary to the screening experiments, the optimum light energy for sporeling growth at 16° C under continuous illumination from a Mazda 'Day-light' fluorescent tube was determined for each species by means of a procedure identical with that described by Boney & Corner (1962*a*). The results are shown in Text-fig. 1. It will be seen that the maximum rate of cell production by *Antithamnion* sporelings was attained at about 17 ergs/sec/mm<sup>2</sup> in the wave-band 380–720 m $\mu$ . At light energies higher than this the sporelings began to bleach and their growth was abnormal (Pl. 1*e, f*). It will also be seen from Text-fig. 1 that the growth of *Brongniartella* sporelings reached a maximum at a lower light energy (7 ergs/sec/mm<sup>2</sup>) after which it fell off slightly as the light energy was increased. Like *Antithamnion* sporelings, those of *Brongniartella* bleached and showed abnormal growth when exposed to higher light energies (Pl. 1*b, c*).

The optimum light energies of these two species are extremely low, being only a small fraction (0.3–5.3%) of normal daylight illumination averaged over the year at Plymouth (*vide* Harvey, 1955). However, it is interesting to note that they are of the same order as the value recorded in our earlier paper for *Plumaria* (10 ergs/sec/mm<sup>2</sup> in the wave-band 380–720 m $\mu$ ). Moreover, in

the case of *Antithamnion*, the present value, which is equivalent to about 500 lux, is similar to that reported by Sundene (1959) as being most suitable for culturing this species.



Text-fig. 1



Text-fig. 2

Text-fig. 1. Cell production by sporelings of *Antithamnion* (○—○) and *Brongniartella* (●—●) grown under continuous illumination by fluorescent light of different energies. Vertical lines represent difference in duplicate values.

Text-fig. 2. Cell production by sporelings of *Antithamnion* (○—○) and *Brongniartella* (●—●) screened from a fluorescent light source by different concentrations of eosin yellow. Comparative values for *Plumaria* sporelings (---) calculated from data by Boney & Corner (1962*a*). Experiments with each species carried out at the optimal light energy for sporeling growth (*Antithamnion*, 17; *Brongniartella*, 7; *Plumaria*, 10 ergs/sec/mm<sup>2</sup> in the wave-band 380–720 m $\mu$ ). Vertical lines represent difference in duplicate values.

#### *Effect of screening with different concentrations of eosin yellow*

Experiments in which sporelings of *Antithamnion* and *Brongniartella* were screened from the light source with dilute solutions of eosin yellow were carried out exactly as described in our earlier paper on *Plumaria*. The results are shown in Text-fig. 2. It is obvious that screening *Antithamnion* sporelings caused a marked increase in cell production—even greater than that observed with *Plumaria* sporelings. Maximal stimulation of growth occurred with

concentrations of 0.1–0.2 mg dye/l., slightly lower than the corresponding values found in the *Plumaria* experiments (0.2–0.5 mg dye/l.). However, as in the studies with *Plumaria*, the effect disappeared completely when the concentration of eosin yellow in the screening solution was increased to 2.0 mg/l.

By contrast, the growth of *Brongniartella* sporelings was scarcely affected by screening with these dilute solutions of eosin yellow, and higher concentrations of the dye—which had no influence on the growth of *Plumaria* or *Antithamnion* sporelings—caused a slight inhibition of cell production (see Text-fig. 2).

TABLE 1. AMOUNTS OF LIGHT TRANSMITTED BY SOLUTIONS OF EOSIN YELLOW, WITH CORRESPONDING EFFECTS ON SPORELING GROWTH

Composition of light emitted by fluorescent tube calculated from spectral energy distribution curve supplied by manufacturers. Experiments with each algal species carried out at appropriate optimal light energy.

Concentration of eosin yellow in screening solution (mg/l.)	% total light energy transmitted (380–720 m $\mu$ )	% transmitted between wave-lengths			% increase in cell production		
		380–480 (m $\mu$ )	480–570 (m $\mu$ )	570–720 (m $\mu$ )	<i>Antithamnion plumula</i>	<i>Plumaria elegans</i>	<i>Brongniartella byssoides</i>
0	100	23.3	41.6	35.1	0	0	0
0.2	98.4	23.3	40.0	35.1	+150 $\pm$ 5	+125 $\pm$ 7	+19 $\pm$ 2
2.0	92.7	23.1	34.5	35.1	+2 $\pm$ 7	+5 $\pm$ 2	-10 $\pm$ 1
20.0	75.5	16.0	24.4	35.1	+1 $\pm$ 5	+3 $\pm$ 4	-41 $\pm$ 6

The proportion of light energy in different wave-bands of the visible spectrum removed by various screening solutions of eosin yellow were calculated from the spectral energy distribution curve of the light source and the absorption curves of the dye at different concentrations (*vide* Boney & Corner, 1962*a*). These data, together with corresponding growth measurements are shown in Table 1. It will be seen that, as in the earlier study with *Plumaria*, removal of only a very small quantity (1.6%) of the incident light caused a very large percentage increase in cell production by *Antithamnion* sporelings (150%). Moreover, this light energy was exclusively removed from the green region of the spectrum, the proportion of light energy between 480 and 570 m $\mu$  being reduced from 41.6 to 40.0 (i.e. by 3.8%). When this proportion was reduced by a greater amount (from 41.6 to 34.5, i.e. by 17%) growth of the screened material was practically normal.

The use of much heavier concentrations of eosin yellow in the screening solution (20 mg dye/l.) caused the total light energy incident on the sporelings to be reduced by 24.5%, i.e. to below the optimum value. Therefore, in order to estimate the effect of this screening solution on sporeling growth, comparison had to be made with control material illuminated by a light energy reduced by a corresponding amount. It will be seen from Table 1 that even when this

high concentration of eosin yellow was used in the screening solution—conditions under which 42% of the incident green light was removed—the growth of *Antithamnion* and *Plumaria* sporelings was unaffected.

Markedly different results were obtained with sporelings of *Brongniartella*. The exclusion of a small quantity of incident green light caused only a small increase in cell production (19%) and when a high proportion of this light energy was removed, cell production was inhibited by 41% (see Table 1).

#### DISCUSSION

The claim that the role of phycoerythrin in marine red algae normally exposed to long periods of daylight illumination is to protect them from the inhibitory effects of excess green light in the wave-band 500–540 m $\mu$  (Boney & Corner, 1962*a*) was based partly on the finding that the exclusion of very small proportions of this light (which is normally absorbed by phycoerythrin) by screening sporelings of *Plumaria* with dilute solutions of the dye eosin yellow, caused a marked stimulation of cell production. This finding has now been confirmed in identical experiments with sporelings of *Antithamnion plumula*, using material adapted to normal daylight conditions by being kept as a continuous growth just below the surface of the water in the Drake's Island Tank at the Plymouth Laboratory. By contrast, however, sporelings of the completely submerged species *Brongniartella byssoides*, which was collected from a depth of 10–12 m where it would normally be illuminated by mainly blue-green and green light, did not behave in the same way, but gave results in accordance with the theory of complementary chromatic adaptation.

These findings are of ecological interest. Thus, the weak solutions of eosin yellow used in the experiments gave a screening effect additional to that provided by the small amount of phycoerythrin in the sporelings. In nature, however, this supplementary screening could be supplied by the overlying thalli of the parent plant, and the protection so afforded could provide the growing sporeling with sufficient opportunity to become adapted to conditions of normal daylight illumination.

So far, only a small number of representative species have been studied; and attempts to follow up these growth experiments with measurements of photosynthesis by red algal sporelings have met with no success. For these reasons, therefore, the conclusions must remain tentative. However, the indications are that the ability of phycoerythrin to absorb green light is exploited by sporelings of marine red algae in two different ways. Thus, in permanently submerged species (as represented by *B. byssoides*) the phycoerythrin is used as an accessory pigment in photosynthesis: and in species which have colonized intertidal habitats (as represented by *Plumaria elegans* and our adapted species of *Antithamnion plumula*) it protects the sporelings at an early stage of growth from excess green light in the wave-band 500–540 m $\mu$ ,



these early stages being dependent on a critical balance between energy in this region of the spectrum and that present at others in the incident light.

#### SUMMARY

Sporelings of the marine red alga *Antithamnion plumula*, grown under continuous illumination from a fluorescent tube, show a maximum rate of cell production at a light energy flux of 17 ergs/sec/mm<sup>2</sup> in the range 380–720 m $\mu$ . The corresponding value for the sublittoral species *Brongniartella byssoides* is 7 ergs/sec/mm<sup>2</sup>.

Growth of *Antithamnion* sporelings is greatly increased (ca. 150%) when they are screened from the light source by dilute solutions of the dye eosin yellow (0.1–0.2 mg/l.) absorbing 2–4% of incident light in the wave-band 480–570 m $\mu$ . However, increasing the concentration of the dye in the screening solution reduces the effect. Thus, at a point where 17% of the incident green light is removed, growth returns to normal; and remains normal even when as much as 42% of the incident green light is removed.

By contrast, when sporelings of the permanently submerged species *Brongniartella byssoides* are screened with dilute solutions of eosin yellow, their growth increases only slightly (19%): and when 42% of the incident green light is removed, their growth is greatly inhibited.

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## EXPLANATION OF PLATE 1

a. *Brongniartella* sporelings; development after 4 days at 7 ergs/sec/mm<sup>2</sup> in the wave band 380-720 m $\mu$ .

b, c. *Brongniartella* sporelings after illumination for 4 days at light energies approximately ten times the value in 1 a; note the abnormal branching and bleaching of the cells in the apical region.

d. *Antithamnion* sporelings; development after 5 days at 17 ergs/sec/mm<sup>2</sup> in the wave band 380-720 m $\mu$ .

e, f. *Antithamnion* sporelings after illumination for 7 days at light energies approximately four times the value in 1 d; note the bleached condition of the cells.