

THE PRODUCTION OF ALGAE GROWING ON GRAVEL IN A CHALK-STREAM

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Little is known of the algae growing in chalk-streams. They are not as readily seen as the dense growths of water crowfoot (*Ranunculus penicillatus* var. *calcareus*) which are a major feature of these streams. Moreover the ease with which algae may be swept away by the current or eaten by animals may mask a fast growth rate. This study was concerned with the algae growing over the surface of stones — the epilithic algae — and represents part of the work of the River Laboratory on the energy flow of the chalk-stream ecosystem (Westlake et al. 1972).

The main study site (Site A) was on Bere Stream at Bere Heath (see p. 29, Fig. 1) where the macrophytes are not normally cut. Instead, wild watercress, *Rorippa nasturtium-aquaticum* var. *siifolium*, grows over the surface of the *Ranunculus* until 50–70% of the stream surface is covered (Ladle & Casey 1971; Ladle, Bass & Jenkins 1972). The *Ranunculus* plants die back under this dense cover and during the winter weed and sediment beds are washed away. The large areas of submerged gravel left behind occupy about 70% of the river bed in the late winter but contract to only 20% by late summer as the *Ranunculus* plants grow again. A second reach (Site B) 1 km upstream is similar except that the *Ranunculus* is cut in May and watercress does not develop to the same extent. Comparative studies were carried out on Dockens Water (Site C) and on Ober Water (Site D), two soft-water streams, about 30–40 km to the east in the New Forest. The bed of these streams is covered by gravel and macrophytes occupy about 5% of the stream bed.

Biomass

Owing to the accumulation of organic and inorganic detritus, the biomass of benthic algae cannot be measured directly by weighing, and the estimation of cell numbers was considered too time-consuming. Estimates of algal biomass were therefore based on analyses of chlorophyll *a*, in spite of the difficulties of relating chlorophyll concentrations to algal weight. Pigments were extracted in methanol and then transferred to 90% acetone for measurement by absorption spectrophotometry (Marker 1972). Corrections were made for the presence of chlorophyll degradation products.

At both Sites A and B, on Bere Stream, the seasonal changes were characterized by a maximum cover of chlorophyll *a* in April (Fig. 1). Its size (150–300 mg chlorophyll *a* m⁻²) varied from site to site and from year to year because of the variable climatic conditions and different flow patterns caused by changing growth patterns of the *Ranunculus* (Marker 1976a). Many pennate diatoms contributed to the rapid growth during March and April, but of the larger species *Diatoma vulgare*, *Navicula*

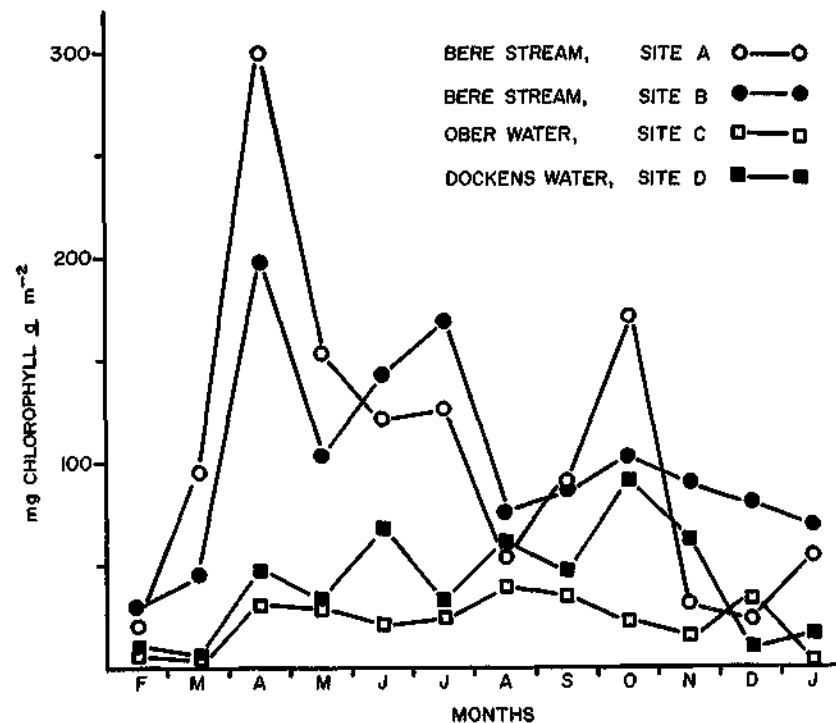


FIG. 1. Seasonal variation in the chlorophyll *a* cover, corrected for phaeopigments, of the epilithic flora during 1972, in Bere Stream, Dockens Water and Ober Water. (Reprinted with permission from *J. Ecol.*, 64.)

avenacea, *N. gracilis* and *Synedra ulna* were significant. The decline of the diatom population at the end of April was not coupled with the depletion of any of the major nutrients (K, N, P or Si) and dissolved silicate concentrations rarely dropped below 2 mg Si l⁻¹ during April (see Casey pp. 28–35). Smaller and unpredictable growths of diatoms occasionally occurred outside April and *Melosira varians* could frequently be found in the side channels at any time during the summer. The summer biomass rarely rose above 75 mg chl. *a* m⁻² and could be as low as 30 mg m⁻². Lime-encrusted algae, similar to those which have been recorded in hard-water English streams, were found during the summer (Fritsch 1949). Although *Vaucheria* was also common, *Cladophora* was never quantitatively important in Bere Stream.

Two neighbouring soft-water streams (Sites C and D), which have considerably lower nutrient levels than the chalk-streams, provided an interesting comparison (Marker 1976a). The summer chlorophyll *a*

cover was significantly lower than in Bere Stream and there appeared to be no pronounced increase in the spring. In winter biomass was low because winter spates disturbed the stream bed and exposed uncolonized gravel (Fig. 1).

If it is assumed that diatoms contain approximately 2% of their organic dry weight as chlorophyll *a*, then even during mid-April the biomass of epilithic algae amounted to no more than 15 g dry weight m⁻² of exposed gravel. The maximum biomass of *Ranunculus* is much larger and has been estimated at 100-500 g m⁻² in a number of streams (Dawson 1976).

Suspended algae

Small chalk-streams do not carry a natural phytoplankton and centric diatoms do not occur in the large numbers so characteristic of the larger English rivers. Site A on Bere Stream is little more than 3 km from the main source and the retention time up to this point is only two hours. Even the River Frome has a retention time of probably little more than three days. However, maximum concentrations of chlorophyll *a* still occur in the spring and are due to pennate diatoms which have become detached from the river bed. Maximum concentrations of chlorophyll *a*, varying between 6 and 50 mg chl. *a* m⁻³, were found in April each year observations were carried out (in Bere Stream in 1968, 1969 and 1970, in the River Piddle in 1968 and 1969 and in the River Frome in 1969, 1970, 1971 and 1972). It appears that populations of benthic diatoms occur regularly in the spring in these hard-water streams. The chlorophyll concentration declined rapidly after April and could be as low as 1 mg chl. *a* m⁻³ in the late summer months. The lower densities of benthic algae in the summer, and their more secure attachment to the substratum, will partly explain these low concentrations. *Simulium* larvae filter large quantities of suspended solids from the water (Ladle, Bass & Jenkins 1972). This effect may mask a much larger movement downstream of suspended algae, although inverse correlations between insect densities and chlorophyll concentrations have not been demonstrated.

Photosynthesis and production

Production and growth cannot be calculated from estimates of biomass because of the effects of respiration, grazing, drift, etc. Estimates of production were, therefore, calculated from direct measurements of *in situ* rates of photosynthesis. Flints from the bed of the stream were enclosed in small perspex chambers approximately 15 cm in diameter, and water was circulated in the chamber by a small propeller driven by a 6V motor (Plate 2). The intensity of light reaching the bed of the stream was measured with a small selenium photocell. The rate of photosynthesis was estimated from the change in oxygen concentration in the chambers and respiration was estimated using identical but opaque chambers.

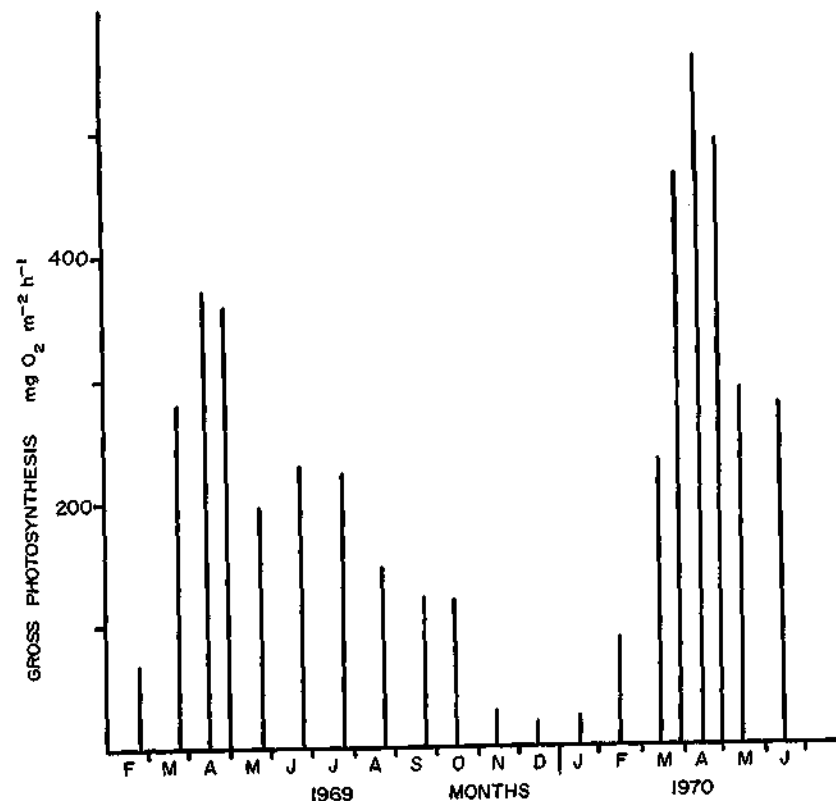


Fig. 2. Seasonal variation in gross photosynthesis of the epilithic algae in Bere Stream. (Reprinted with permission from *J. Ecol.* 64.)

Maximum rates of photosynthesis in Bere Stream (Site A) were observed in April and coincided with the period of maximum biomass (Fig. 2). The rate of photosynthesis per unit quantity of chlorophyll, which is the specific rate, decreased rapidly as the biomass increased. Specific rates were highest in midsummer and lowest in the autumn and winter (Fig. 3). These changes are important because they directly affect the calculation of production from photosynthetic rates and illustrate the difficulties of predicting changes in biomass (Marker 1976b).

From repeated measurements over a day, conversion factors were obtained for calculating 24-hour rates from short term studies. If it is assumed that the photosynthetic quotient is unity (i.e. volume of O₂ liberated is equal to the volume of CO₂ absorbed) and that the carbon content of algae is approximately 50% of the dry organic matter, figures

for daily gross production may be calculated. Gross photosynthesis over 24 hours less community respiration over 24 hours will give a minimum figure for net production (Table 1). The difference in net production between 1969 and 1970 is due to the large differences in biomass.

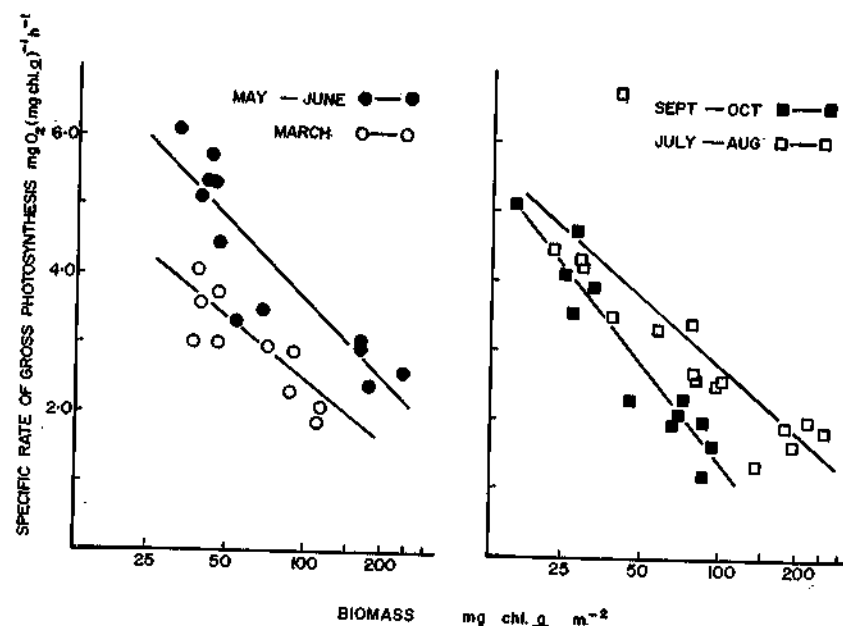


FIG. 3. Seasonal variation in the relationship between the specific rate of gross photosynthesis and biomass.

TABLE I

Annual production of epilithic algae in Bere Stream. Values are expressed as g dry organic matter and calculated from gross photosynthesis.

	1969		1970	
	gross production	net production	gross production	net production
g m ⁻² exposed gravel	384	180	519	250
g m ⁻² river bed	156	76	246	124

Light

Some of the studies suggested that light directly affected the rates of photosynthesis of algae growing on the gravel. On one occasion during March, the rate of photosynthesis of diatoms, containing 80-90 mg

chl. *a* m⁻², was reduced by 30% when the light intensity was artificially lowered from 100 to 30 W m⁻² (i.e. 30 000 to 10 000 lux). In a more general approach, the analysis of data from a series of studies during winter and spring showed that the effect of light intensity on photosynthesis was highly significant (Marker 1976b).

Both the effect of light intensity on the rates of photosynthesis and the size of the algal populations themselves suggest that there must be a considerable attenuation of light by the mats of benthic algae. Direct measurements of light transmission are not possible because the substratum is opaque. Moreover it is not possible to remove the algae quantitatively as an undisturbed layer, because many species are found growing in the pits and crevices of the weathered flint surfaces. In a different approach, algae were grown on transparent glass surfaces submerged in Bere Stream and the transmission of light measured using a selenium photocell. During April, light transmission through algal layers with 50 mg chl. *a* m⁻² was between 55 and 60%, and with 100 mg chl. *a* m⁻² between 30 and 40%. During May and June transmission varied from 35% in 1973 to 65% in 1974 with populations containing 50 mg chl. *a* m⁻². This variation may have been due to the irregular growth of algae in the summer as discrete patches of lime-encrusted algae appeared on the glass surfaces.

A direct comparison of the growth of algae on glass slides with the growth of algae on gravel showed a broadly similar pattern with diatoms growing rapidly in March and April. However, there were distinct quantitative differences. The maximum biomass found on glass slides was frequently two or three times that found on gravel during April. In contrast, densities of algae on slides during May and June were frequently less than on gravel. Consequently the interpretation of the transmission data is not entirely straightforward.

Flow

Flow may also affect the rate of photosynthesis by directly controlling the diffusion of gases and nutrients. In the river, velocities over the gravel can vary widely, from near zero to over 1 m s⁻¹, owing to the influence of the weed beds. The circulation of water in the photosynthesis chamber was adjusted so that loosely attached diatoms were not removed from the gravel. Presumably, therefore, the diffusion of gases and nutrients to the algae was not slower than in the river. No attempt was made in these studies to vary the circulation of water in the photosynthesis chambers and as a result the more subtle effects of flow may have been missed. A much more elaborate sampling programme would have been needed to take this into account.

Conclusion

The epilithic algae reach their maximum biomass in April when chlorophyll estimates suggested that biomass was 10-15 g m⁻² of dry

organic matter. Annual production was calculated to be over fifteen times greater than biomass. The estimation of net primary production is always difficult for benthic floras and comparisons are especially difficult when different methods are used. But these figures contrast sharply with those for *Ranunculus* which has a ratio of annual production to maximal seasonal biomass of 1:16 (Dawson 1976). The accumulation of algal biomass is apparently being prevented. Some organic matter may be excreted; some algae will be washed off the bed of the stream by the current and grazing by herbivorous invertebrates will also tend to prevent the accumulation of algae. These factors may also affect the accumulation of *Ranunculus* but grazing may be less important and the effect of scour more seasonal in its impact.

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PLATE 2. Photosynthesis chambers and light meter in position on the bed of Bere Stream.

ACTINOMYCETES AND ACTINOPHAGE IN FRESH WATER

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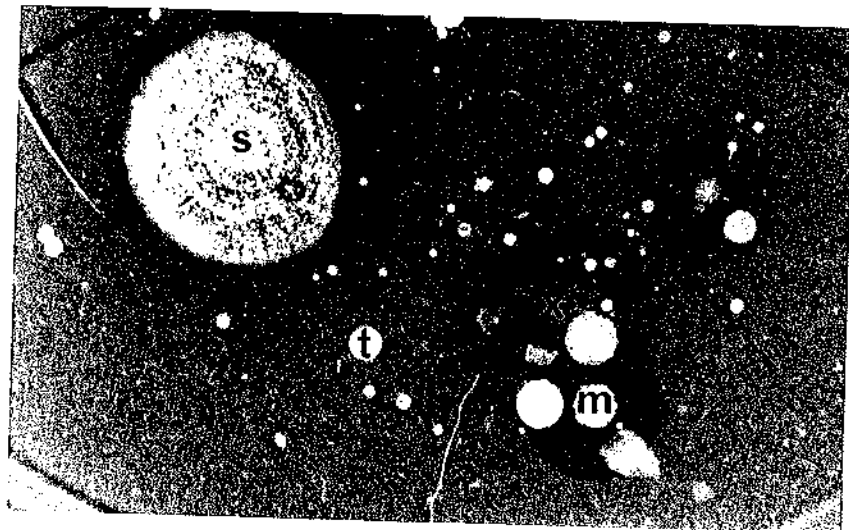


PLATE 3. A river water sample plated on to chitin-actidione agar. Growing colonies of *Streptomyces* (s) and *Micromonospora* (m) show hydrolysis zones, as also do myxobacteria (x). Other bacteria (t) are growing but not producing hydrolysis zones. ($\times 3$).

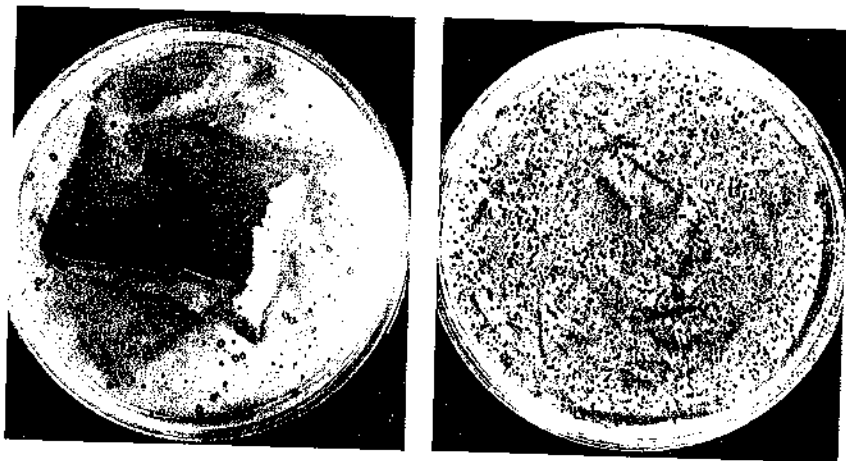


PLATE 4. Plaque formation in *Actinoplanes missouriense* (left) and in *Streptomyces griseus* (right) following phage recovery from local oak leaf litter and stream water respectively. ($\times 0.6$).

The Actinomycetales, commonly referred to as actinomycetes, are a group of micro-organisms which have a true branching mycelium and so resemble the fungi, but are small in size and have other characters more like those typical of bacteria. They thus form a group lying in classification half-way between the fungi and the bacteria. If the mycelium is short lived in sub-culture, or even becomes absent altogether, it is difficult to decide whether the strain should be called an actinomycete or a bacterium. Strains now included in the genera *Nocardia* and *Mycobacterium*, or more loosely placed within this taxonomic area (e.g. Nocardioform-Lspi, discussed below) frequently give rise to such a dilemma. Again, actinomycetes resemble fungi because they may release motile spores from sporangia, but these spores are as small (about $1 \mu\text{m}$ diameter) as motile bacteria.

Several other rather fundamental characters of actinomycetes are those of procaryotic cells (such as those of bacteria and blue-green algae) rather than those of eucaryotic cells (as found in fungi, green algae and higher plants); procaryotic characters are their lack of membrane-bounded organelles such as nuclei and mitochondria, their frequent susceptibility to phage-virus and their type of antibiotic sensitivity. They are killed by many of the antibiotics which also kill bacteria.

Records of direct observations made on aquatic actinomycetes growing in their natural milieu are virtually non-existent so far, although I did find a few sporangia of *Actinoplanes missouriensis* Couch on twigs left exposed in a stream for several weeks, after I had been encouraged to search by a high level of recovery of this species in agar-plated washings from parallel exposures. Considering more indirect procedures, the technique of damp incubation has potential, and by using this on damp allochthonous leaf material collected on the shore of Windermere I was able to observe the development of a variety of sporangial forms of the family Actinoplanaceae. Although *Actinoplanes* was the dominant form recovered, strains of *Spirillospora* and of other unidentified genera were also present (Willoughby, 1969).

In isolating actinomycetes from nature on to nutrient growth media, addition of actidione (cycloheximide) eliminates fungi but unfortunately there are no antibiotics which will satisfactorily segregate growing actinomycetes and bacteria as two distinct groups. However, in making general surveys of actinomycetes in fresh water, it is probably better to allow the full spectrum of procaryotic cells (both bacterial and actinomycete) to develop on the initial isolation medium rather than to attempt such a segregation. Where a single actinomycete only is searched for, the use of a more selective method becomes a possibility. Thus Cross & Attwell (1974) have employed the antibiotic novobiocin (in conjunction