EXPERIMENTS USING AN ARTIFICIAL STREAM TO INVESTIGATE THE SEASONAL GROWTH OF CHALK-STREAM ALGAE

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Introduction

Seven years ago we described seasonal changes in the algae and the chemical composition of the water in chalk streams (Casey 1976, Marker 1976c). The chalk streams of southern England are hard-water and nutrient-rich, flowing over stones which range in size from large flints to small gravel. The submerged macroflora is typically dominated by *Ranunculus penicillatus* var. *calcareus* (R. W. Butcher) C. D. K. Cook, and there is a variety of emergent vegetation (Westlake et al. 1972; Ladle & Casey 1971). Beds of soft sediment collect around these plants. The gravel is disturbed and redistributed as the winter rains wash out silt and decaying macrophytes so that algal growth occurs largely on the clean gravel each year (Marker 1976a, b).

The seasonal cycle of algae shows a rapid growth of diatoms in the spring with a maximum in April or May. This is followed by a similarly rapid decline and a succession of various encrusted green and blue-green algae. Most algae grow on the bed of the stream or on macrophytes rooted in the stream-bed because the short residence time of the water prevents a sizable phytoplankton population from developing. The seasonal cycle will be influenced primarily by light, temperature, the composition of the mineral nutrients dissolved in the surrounding water and the flow of that water downstream.

During passage downstream the concentration of nutrients affects the algae both qualitatively and quantitatively and in turn the plants alter the concentration of the nutrients. In addition many different kinds of inputs from the surrounding land also change the nutrient concentrations. Sudden changes in water velocity due to rain may have dramatic effects on the amount and composition of the algae. Changes in nutrient concentration can be similarly rapid due to man's activities (Crisp 1970). Because of the erratic nature of some of these events, field studies alone are inadequate to elucidate the way in which light, temperature, and flow affect the interaction between nutrients and the seasonal cycle of the algae.

Broadly there are two ways of investigating the effects of these interacting variables (e.g. light, temperature, nutrients and flow). The first is to measure a large number of variables over many years. The main drawback of this approach lies in the essentially long-term nature of the work. It is more suited to the study of water chemistry than biology because, generally speaking, more chemical analyses can be completed in a given time, thus allowing more frequent and extensive sampling. The second method involves experiments where the investigator either isolates

part of the ecosystem, which may then be manipulated, e.g. the 'Lund tubes' (Lund 1978) or constructs self-contained artificial systems for the same purpose. However, the more they are manipulated, the more they tend to differ from the natural systems they were intended to simulate. Both of these general approaches – observational and experimental – have been applied by us, but in this article we try to give an overview of the second.

The experimental site

The experiments described here are part of a strategy of continuing research into the production ecology of chalk streams and involve the use of a large artificial recirculating stream built in a disused water-cress bed (Fig. 1; for a photograph see 43rd *Annual Report*, p. 32). To ensure that this system is not unduly artificial we make comparisons with studies on natural local streams. Essentially the artificial stream (or channel) is race-
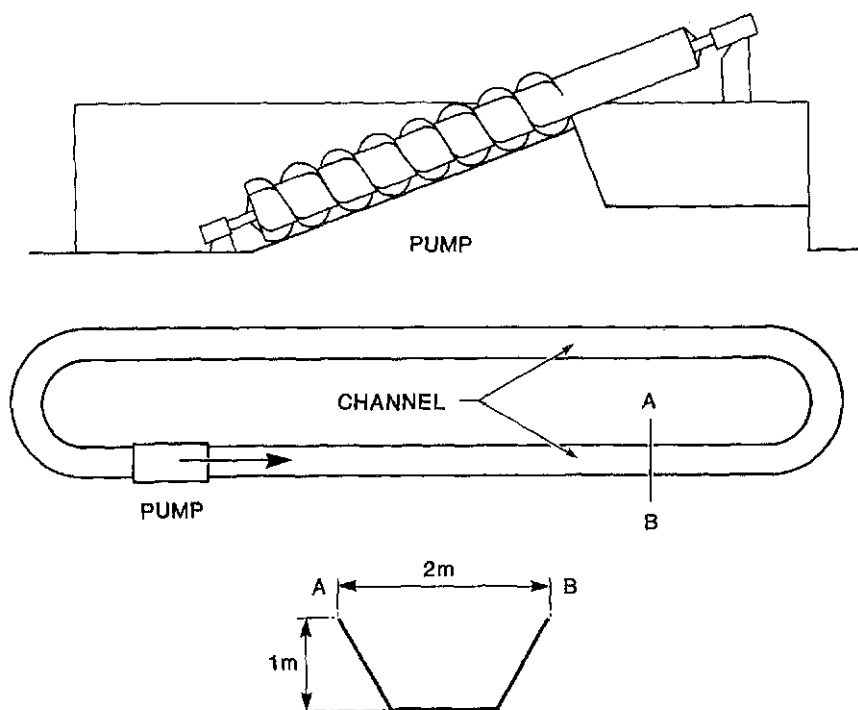

FIG. 1.   Diagram of Waterston experimental channel showing details of screw pump, plan and cross section of the channel.

track-shaped, approximately 60 m in circumference, with a trapezoidal cross-section, 1 m wide at the base and 2 m wide at the rim (Ladle et al. 1977, 1981). Water is recirculated by an Archimedian screw and in later experiments was continuously replenished by ground water from the chalk aquifer by one of two pumps. This fresh water entered just upstream of the Archimedian screw and water left the channel through an overflow slit 45 m further downstream. The smaller pump supplied water at a rate of $4 \cdot 2$ m^3 h^{-1}, the larger at $10 \cdot 2$ m^3 h^{-1}, thus allowing the turnover to be varied independently of water velocity. The rate of circulation was 1-2 orders of magnitude greater than the rate of replenishment. This rate of circulation allowed substantial velocities of water to move over large areas of substratum which could then be used for large scale destructive sampling programmes. The water that came direct from the aquifer was almost constant in chemical composition and temperature throughout the year. Variability within the experimental system was further reduced by using a constant water-velocity, and homogeneous substrata.

Experimental manipulations

The experiments described here were carried out over a period of three years between 1975 and 1978.

Initially the channel was used as a closed system with no gravel and no replenishment of water during individual experiments. Light and airborne detritus were excluded by a black polythene sheet. Temperatures ($6 \cdot 4$-$19 \cdot 0$ °C) were within the range recorded for chalk streams (Crisp et al. 1982). Oxygen concentrations were constant at 100% saturation, due to continuous reaeration by the Archimedian screw. However, large changes occurred in many of the chemical components of the water (Ladle et al. 1977). The pH increased from $7 \cdot 9$ to $8 \cdot 6$ and the calcium concentration decreased rapidly over 5 days (Fig. 2). Loss of dissolved inorganic phosphate occurred at the same time until it was undetectable after 7 days (<1 µg l^{-1} PO$_4$.P, Fig. 2). After calcium precipitation had ceased KH$_2$PO$_4$, added subsequently at concentrations equal to that in the ground-water, remained in solution. Bubbling of gaseous CO$_2$ into the water could arrest and then partially reverse the initial losses of calcium. Dr W. A. House is exploring some of the physical chemical processes involved. These include the precipitation of calcite mineral from river water, the exchange of gaseous carbon dioxide with the atmosphere, and the electrical conductance of simple aqueous systems.

In the succeeding experiments gravel (flints) was added as a natural substratum to the channel. These flints were obtained from a local quarry and were identical to those in local streams except that in the state they arrived from the quarry they were uncolonized by algae. Gravel, of nominal '40 mm diameter reject', was placed in the bottom of the channel to a depth of 500 mm at the upstream end, decreasing to 300 mm at the
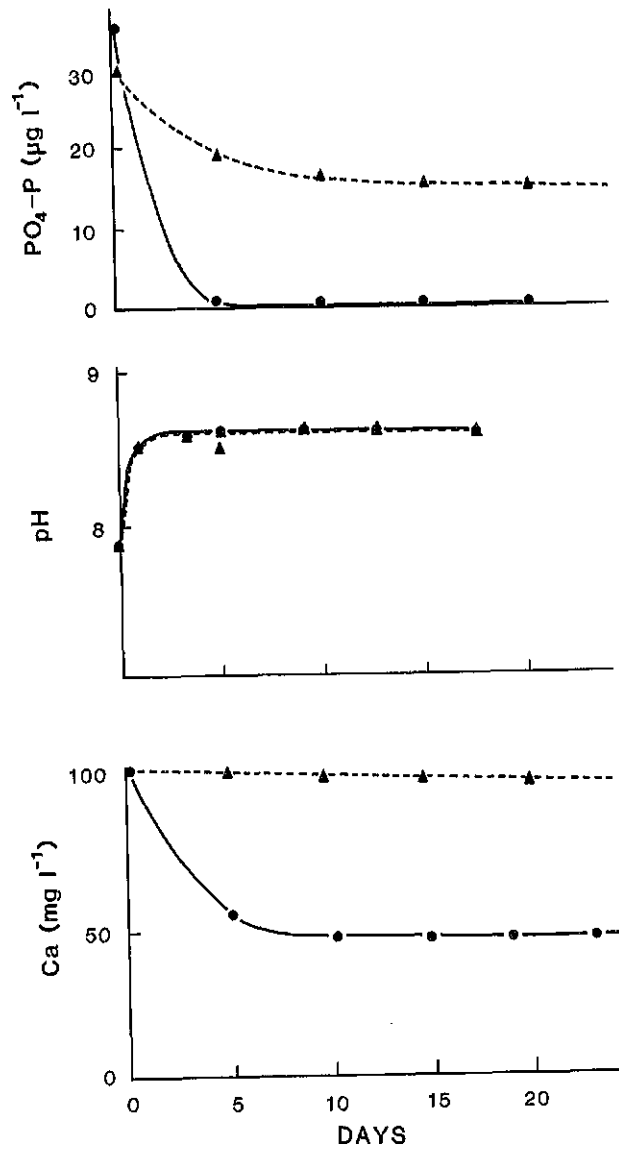
FIG 2. The effect of water recirculation on water chemistry with (-----) and without
(————————) gravel.

downstream end. The width of the bed at the surface of the gravel was c. 1·4 m, giving a total gravel area of 70 m², and the water depth above the gravel was c. 30 cm. In the absence of gravel, water velocity was 0·2 m s⁻¹ and in the presence of gravel 0·4 m s⁻¹. In the presence of gravel, the chemical changes in the water were very different and, although the pH rose again, the losses of calcium and phosphate were much less (Fig.2). Although there is a rapid loss of CO_2 in chalk streams after the water leaves the springs, it usually remains above saturation levels and there is no appreciable loss of calcium.

The next series of experiments was carried out in the dark, in the presence of gravel, but with the water being continually replenished. Interpretation of the chemical data is complicated by the fact that the water entering does not pass through the channel as a discrete 'parcel' but is continually mixed with the recirculating water. The residence time is, therefore, more accurately expressed as a 'half-life', i.e. that period of time in which 50% dilution takes place. For these experiments in the channel this period was c. 8 hours. Thus most of the differences observed will have taken place in the preceding 24 hours, provided that the rates of the contributory processes are not changing rapidly. The differences in composition observed between inflow and outflow before the dark covers were removed may be assumed to represent non-phototrophic, and probably largely non-biological, effects. Concentrations of calcium and phosphate-phosphorus, which were 100 mg l⁻¹ and 30-40 μg l⁻¹ in the inflow, in the presence of gravel, stabilized at 80 mg l⁻¹ and 10 μg l⁻¹ respectively in the outflow. There were no perceptible differences in silicate concentration between inflow and outflow.

Between 1976 and 1978 experiments were carried out in the light in two phases, with preliminary experiments between April and December 1976 and more detailed studies between April 1977 and August 1978. Here we are concerned mostly with the latter studies in which we were able to examine the seasonal changes in algae growing over gravel, subjected to constant water velocity throughout the year and with continuous replenishment of water of nearly constant composition. Gravel and flints are a particularly difficult substratum to sample, and, to ease this problem, small pots 100 mm in diameter and 100 mm deep, with circular wooden bases and sides of plastic 'garden mesh' (5 mm), were constructed. Five hundred of these were placed in the channel with their tops level with the gravel surface. These were removed at regular intervals throughout the sampling period for various chlorophyll a estimates (Marker 1972). Each time chlorophyll a was estimated, surface stones were examined for the algal flora. A combination of brushing, scraping and abrasion was required to remove the algae and even this was not totally successful. The colonization phase (April-May) was characterized by a very rapid growth of diatoms. Within five weeks chlorophyll densities had reached between
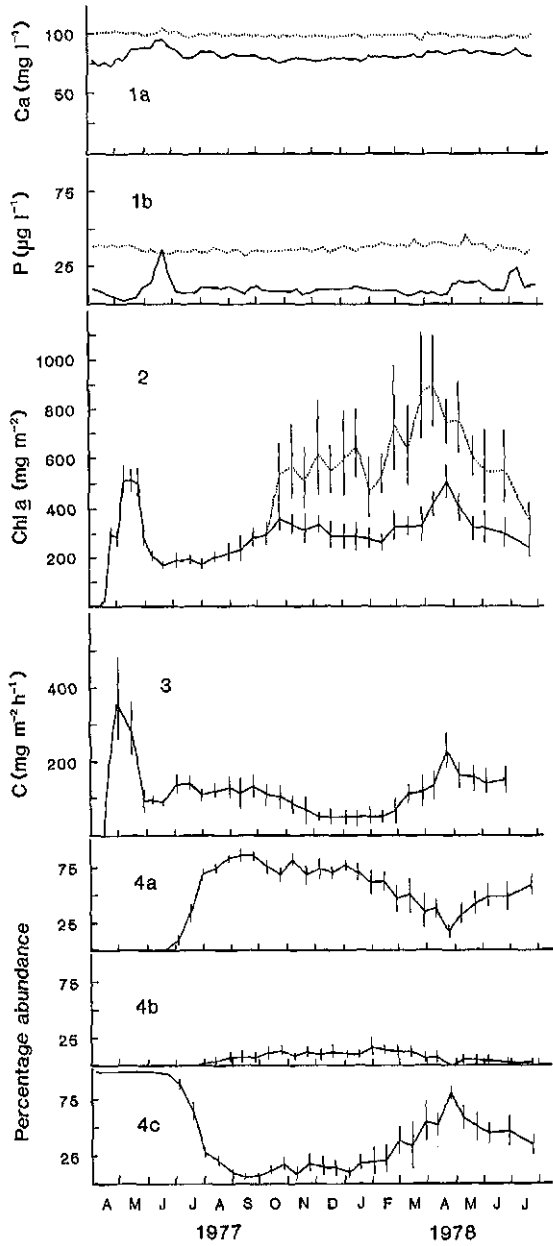
500 and 600mg m^{-2} chl. a (Fig. 3). This population was composed largely of the diatoms *Achnanthes minutissima* (Kütz), *Meridion circulare* Agardh, *Nitzschia fonticola* Grun. and *Synedra ulna* (Nitzsch) Ehr. (Fig. 4). Between 18 and 26 April chlorophyll a densities increased ten-fold and the numbers of the four dominant species appeared to have increased twenty-fold. The chlorophyll a peak in mid-May corresponded to a population density of over 10^{11} cells m^{-2}. The decline in diatoms occurred rapidly at the end of May and the chlorophyll a level was less than half its former maximum by the end of June. Concurrent observations on the dissolved chemical composition of the water leaving the channel outflow with that entering showed interesting changes over and above those which had been found earlier in the dark. Concentrations of phosphate-phosphorus fell rapidly to 1 to 2 μg l^{-1} as the diatoms grew (Figs 3 & 4). It is improbable that phosphate was ever low enough to starve the population. Phosphate was entering the channel at a rate equivalent to 35-40 mg m^{-2} day^{-1} PO$_4$.P and earlier studies (Ladle et al. 1981) with a suspended population of *Chlorella* and diatoms showed that rates of phosphate uptake by algae could be far higher than the rate of precipitation observed in the channel in the dark. However it is still possible that one of the nutrients may have been limiting the rate of diatom growth due to steep diffusion gradients through the thick algal mat. Silicate was taken up rapidly by the diatoms and the change in concentration followed the cycle of diatoms very closely. However, concentrations never fell below 0·8 mg l^{-1} Si. In local chalk streams the spring growth of diatoms follows a very similar pattern to this initial colonization phase and may bring about substantial reductions in silicate concentrations (Casey et al. 1981), but even at downstream sites concentrations below 1 mg l^{-1} Si have rarely been recorded. Other macronutrients do not appear to be totally depleted during the spring in chalk streams either.

Loss of algae through sedimentation was also estimated from material that accumulated in special sediment traps (Welton & Ladle 1979). Maximum rates of sedimentation occurred in June.

FIG. 3. Seasonal variation within the experimental channel of:
     1: (a) dissolved calcium and (b) soluble 'reactive' phosphorus;
        . . . . . inflow water, —————— outflow water.
     2: benthic chlorophyll a, ————— on the gravel alone and, ... including
        Cladophora.
     3: hourly rates of photosynthetic carbon uptake of the attached algae.
     4: percentage abundance of the major algal groups
        (a) blue-green algae
        (b) green algae
        (c) diatoms
        Vertical bars represent 95% confidence intervals.

For a period in June, outflow concentrations of phosphate and silicate exceeded inflow concentrations as the diatoms decayed and the nutrients were recycled (Figs 3, 5). For a brief period in June alkalinity (essentially bicarbonate) and concentrations of calcium in the outflow approached inflow levels. Chalk streams have a higher concentration of calcium bicarbonate than would be expected from the solubility of chalk in water
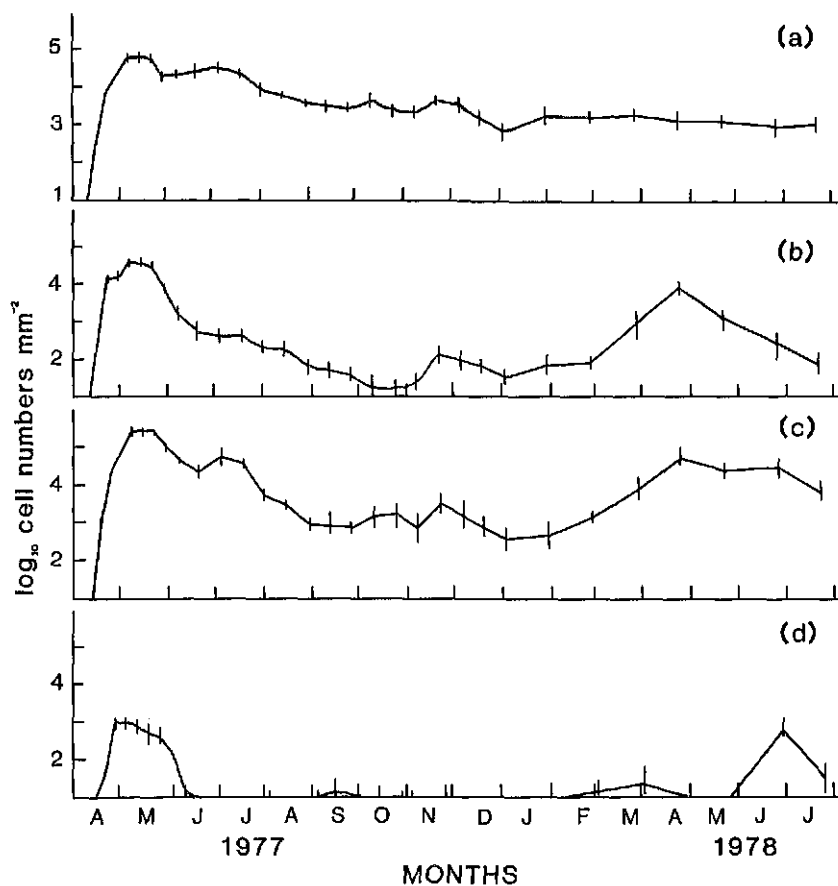

FIG. 4. Seasonal variation of four important diatoms growing on the gravel
(a) *Achnanthes minutissima*
(b) *Meridion circulare*
(c) *Nitzschia fonticola*
(d) *Synedra ulna*
Vertical bars represent 95% confidence intervals.

containing CO_2 at the levels found in chalk-stream water, and the conditions which maintain these supersaturated levels may have been temporarily present in the channel in June 1977 due to CO_2 release from the sediments, lower overall rates of photosynthesis and the presence of organic colloids.

Throughout the late summer and autumn a hard calcareous crust of green and blue-green algae developed in the channel (Fig. 3). Of these
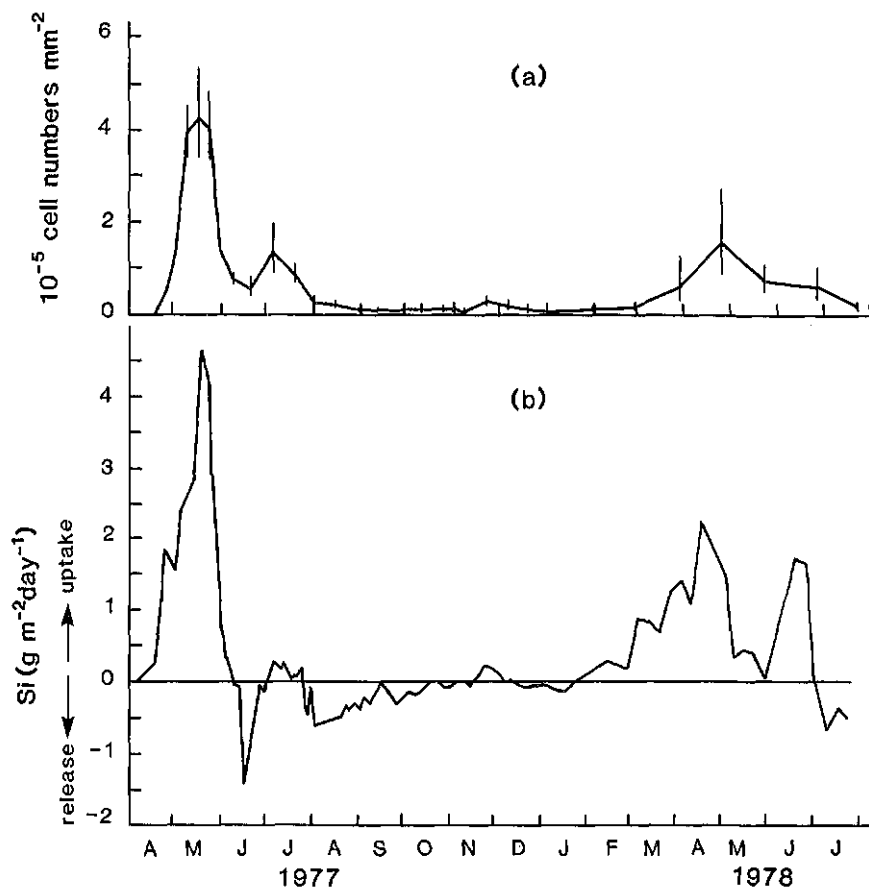

FIG. 5. (a) Seasonal variation in total diatom numbers; vertical bars represent 95% confidence intervals.

(b) Uptake and release of silicate, estimated from changes in concentration between inflow and outflow water.

Gongrosira incrustans Schmidle was the most abundant green alga and it also occurs widely in chalk streams. Current work by J. A. Rother indicates that the most important blue-green alga was *Homoeothrix varians* Geitler which is widespread in chalk streams. This was initially thought to be *Lyngbya kützingii* (Schmidle) (Marker & Casey 1982). These calcareous crusts are locally common in riffles of chalk streams. But the 'pavement' which developed in the channel was not typical and was probably a result of the undisturbed gravel bed, the absence of *Ranunculus* and the enhanced non-biologically induced carbonate precipitation.

Another unusual feature of the channel as compared with local streams was a dense growth of the filamentous alga, *Cladophora*, late in the autumn and winter of 1977/78. Some months after the main sampling programme had been completed in August 1978, densities of 285 g m^{-2} dry matter were found. This is high for waters of this type which have been drawn direct from the aquifer, with concentrations of phosphate-phosphorus between 10 and 30 μg l^{-1}.

In addition to these populations of green and blue-green algae, diatoms varied in numbers throughout the experimental period (Fig. 5). Figure 5 also illustrates the close relation between diatom cell-number and the uptake of dissolved silicate. After the initial colonization phase there were two minor periods of diatom growth – in mid-summer and in the late autumn of 1977 – followed by a major development in spring of 1978. Our earlier studies in 1976 and preliminary results of new work in 1981/82 gave broadly similar patterns but suggested that the magnitude and timing of the periods of activity may vary somewhat. The seasonal succession of algae in natural streams (Marker 1976a) is similar to the succession found in the channel, which suggests that it is unlikely to be due to seasonal variations in macronutrients or water velocity. A major difference between a natural stream, disturbed by winter spates, and the channel (undisturbed) is in the continued accumulation in the latter of biomass during the autumn and winter. However, it is clear that changes in water-velocity or new substrata are not essential for the development of diatoms in the spring, because in the channel diatoms developed on top of the calcareous crust of green and blue-green algae which had overwintered under conditions of constant water-velocity.

Interrelationships and indices of algal production

In a quite separate study, dissolved silicate has been monitored in a number of local streams over 10-15 years. A comprehensive quantitative sampling programme for benthic diatoms for a whole catchment area covering many years is totally impracticable, and measuring changes in silicate concentration may be the only possible means of monitoring the between-year and within-year variations in diatom populations. In chalk streams the marked spring growth of diatoms shows a progressive effect
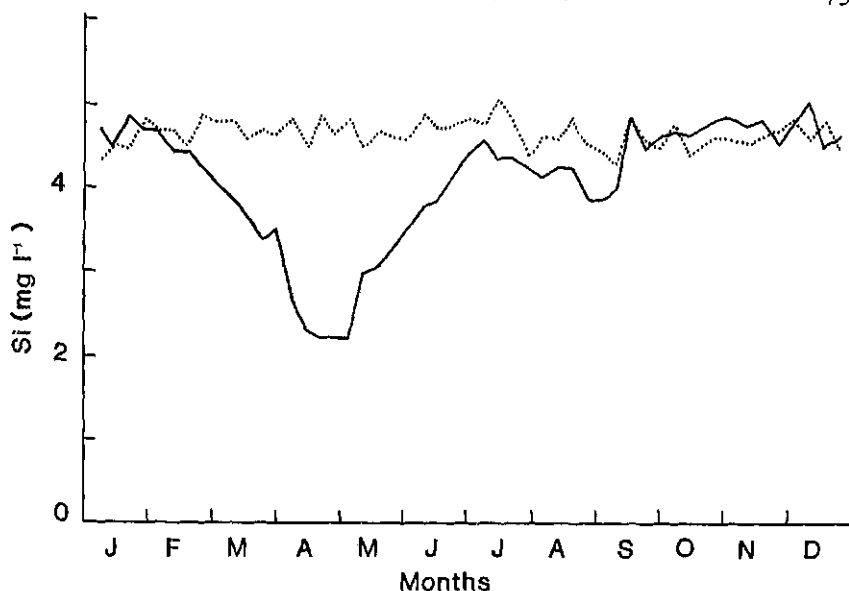
FIG. 6. Seasonal variation in silicate concentration derived from the means of weekly estimates, ... Hollybush bore-hole over 7 years (1969-75), —————— River Frome at East Stoke over 13 years (1965-77).

on the silicate concentration as the water passes downstream (Casey et al. 1981). In the downstream reaches, a late-summer or autumnal depression can also be detected (Fig. 6). Analysis showed that, in spite of considerable variation from year to year, this late-summer period of activity was statistically significant. The studies in the channel confirmed that fluctuations in diatom numbers do occur outside the spring period but that the timing and magnitude are very variable.

The measurement of production of field populations of algae is extraordinarily difficult. Numerous measurements of photosynthesis using either oxygen or ^{14}C methods have been made throughout the world, but the errors in extrapolating to algal production can be very large. Although the uptake of silicate is a measure of diatom growth, it too is subject to a wide variety of serious errors. For Bere Stream, crude extrapolation from estimates of maximum photosynthesis in April (Marker, 1976b) suggested production of c. 2 g m^{-2} day^{-1} organic matter while estimates from changes in silicate concentration yielded between 2 and 6 g m^{-2} day^{-1} organic matter (Casey et al. 1981). In the recirculating channel diatom photosynthesis was high during the initial colonization phase (Fig. 3) and uptake was calculated from ^{14}C measurements as 132 g $m^{-2}C$ for a two-month period during April and May, on a daily basis varying from zero at the

beginning of the experiment to above 4 g m^{-2} day^{-1} in mid-May. These ^{14}C uptake figures are not true estimates of production because the values represent an unknown intermediate position between net and gross photosynthesis and take no account at all of night-time respiration.

Over the same period silicate uptake was estimated as 107 g m^{-2} Si. Figure 5 shows that, even when diatom numbers were declining, uptake of silicate suggested substantial continuing production. Our figures (unpublished) suggest that actively growing populations of diatoms contain 1–2% chlorophyll a as a percentage of the organic weight. The maximum biomass in May, as measured by chl. a, would thus have corresponded to c. 30–60 g m^{-2} organic matter (600 mg m^{-2} chl. a). For this maximum biomass to match the production, inferred from silicate uptake, the silica (SiO_2) content of the diatoms would have to have averaged 85% over the two months. Anything less than this implies a gap between the maximum biomass observed and the apparent production. For example, if the silica (SiO_2) content had been only 50% of the diatom weight, production would have been over four times the maximum biomass.

Losses through sedimentation and wash-out were estimated at 40–80 g m^{-2} organic matter (c. 800 mg m^{-2} chlorophyll a and degraded derivatives). The decline in the diatom population is most unlikely to have been due either to lack of nutrients, which were continuously replenished from the bore hole, or to changes in water velocity, which remained constant. Moreover, this initial colonization stage by diatoms has been repeated on three occasions so is unlikely to be due to transitory changes in irradiance or temperature. Collaborative studies with Dr Ladle and his co-workers indicate that the decline in the diatom population owes more to the effect of grazing invertebrates (also studied by Eichenberger & Slatter 1978, Mason & Bryant 1975, Ladle et al. 1980).

Acknowledgements

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