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**INFLUENCE OF STREAM FLOW ON EPILITHIC DIATOM
COMMUNITIES AND ITS CONSEQUENCE FOR WATER QUALITY
MONITORING**

by

Christopher J. Penny

(B.Sc. Hons)

**A Dissertation submitted in partial fulfilment of the requirements for the degree
of Master of Science in Advanced Ecology**

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Department of Biological Sciences

University of Durham

1993



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ABSTRACT

Epilithic diatom communities and relevant physico-chemical variables were studied between 3 May and 16 July 1993 in Harwood Beck, an oligotrophic stream in Upper Teesdale, and the River Browney, a lowland eutrophic river. The effect of stormflow on diatom-based water quality indices was assessed.

In both rivers epilithic chlorophyll *a* ($\mu\text{g cm}^{-2}$) was highly variable and very low after mid-June. In Harwood Beck there was a significant difference in epilithic chlorophyll *a* between cobbles and boulders, with the greatest difference after stormflow. Epilithic chlorophyll *a* in the River Browney was initially high, but declined markedly after stormflow. River Browney chlorophyll *a* was inversely correlated with current speed on cobbles and total inorganic nitrogen on cobbles and boulders. Uptake of inorganic nitrogen may be directly related to epilithic diatom biomass. A sharp rise in the percentage of *Cocconeis placentula* between mid-June and mid-July suggests that grazing may be an important factor limiting algal biomass. The possible roles of algal grazers, *Cladophora glomerata* and silicate concentration, in progressively limiting diatom biomass during the summer are discussed.

Percentage composition changes of taxa after stormflow are generally related to taxa morphology and mode of adherence to the substrate. There were no significant differences in the percentages of the five most common taxa between cobbles and boulders.

Percentage changes in taxa were not significantly correlated with nutrients, with the exception of *Navicula gregaria* and *N. lanceolata* (in the River Browney) which were inversely correlated with the concentration of total inorganic nitrogen. There appears to be a succession of dominant species, possibly related to the influence of flow and nutrient concentrations, which tended to increase during the study period.

Diatom water quality indices remained relatively stable over the 10 week period. Effectively equal water quality classifications were derived from the Specific Pollution Index (SPI) and Generic Diatom Index (GDI). The zoning system proposed by Round (1993) may be broadly useful, but requires careful interpretation.

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CHAPTER 1

INTRODUCTION

1.1 Biological Monitoring

There is a need for compatible, reliable and practical methods to assess biological water quality (Descy, 1979). Biological and chemical monitoring of pollution can and should supplement each other, although, in theory, either could provide reasonable indications of the effects of pollutants. Chemical monitoring has limitations associated with temporal and spatial sampling and its inability to account for the potential synergistic effects of pollutants and their bioaccumulation in ecosystems (Spellerberg, 1993). Organisms react to fluctuations in water quality which may be missed by intermittent chemical analysis.

An organism can act as a biological indicator because its distribution and abundance is related to particular environmental variables and/or because it has the capacity for accumulating pollutants in its tissues. Good indicator species have the following qualities:

- a. They are easily sampled and identified.
- b. They are cosmopolitan in distribution.
- c. There must be abundant autoecological data.
- d. They often have economic importance as a resource, nuisance or pest.
- e. They should be easily cultured in the laboratory and have low genetic and niche variability.

(Hellawell, 1986).

Bioaccumulative indicators have additionally desirable characteristics not found with invertebrates or diatoms; i.e. long life cycles, sufficient tissue mass for analysis, and concentration of the pollutant(s) above the environmental level(s) (Hellawell, 1986).

Sampling of macro-invertebrates has been routinely used for many years by U.K. water regulatory bodies to supplement chemical analysis. Invertebrate populations exhibit a varied response to both toxic and organic pollutants and are relatively sedentary. Heavy pollution affects whole taxonomic groups of invertebrates; specific differences are only important in cases of mild organic pollution (Mason, 1991). The current method for nationally reporting biological water quality using macro-invertebrates is the Biological Monitoring Working Party (BMWP) score. The BMWP score is the sum of scores (1 to 10) assigned to individual families according to their sensitivity to pollution; the least tolerant families score 10. Average Score Per Taxon (ASPT) values are calculated by dividing the BMWP by the number of taxon. Larger samples are likely to include more families and species (Mason, 1991). BMWP performs optimally with faster flowing waters over a fairly varied stony / gravelly bed (typically riffles). However, when applied to deep slow flowing sites or to restricted habitats, the BMWP scores may underestimate water quality (Extence & Ferguson, 1989). Standardisation of kick sample time and mesh net gauge is necessary, as increasing sampling effort leads to substantial increases in the BMWP score, due to a fuller representation of the macro-invertebrate community. Quantitative sampling of macro-invertebrates is difficult due to their patchy distribution; large numbers of samples are needed to make reasonable estimates of population densities (Hellawell, 1986). The summer hatch of many taxa, especially sensitive groups such as Plecoptera (stoneflies) means that species absence is not always associated with water quality (Jeffries & Mills, 1990). However, in a performance evaluation of the BMWP score system at 268 unpolluted running water sites on 41 rivers, ASPT values were less influenced by season and sample size than BMWP scores (Mason, 1991). The ASPT values indicated that seasonal variations were relatively slight compared to the variations due to environmental parameters (Armitage *et al.*, 1983).

Multivariate analysis has been used to classify clean U.K. running water sites on the basis of the macro-invertebrate fauna. Subsequently, RIVPACS (River Invertebrate Prediction and Classification System) allowed prediction of the probability of a given species or family occurring from environmental data. ASPT values can be predicted using 5 environmental variables in a multiple regression equation which explains 67.7% of the total variation. Therefore, observed ASPT values can be compared with predicted values in the assessment of water quality (Mason, 1991). If

comparable systems of data interpretation were developed for other organisms then biological monitoring of pollution could become more representative of the whole river ecosystem. Algal monitoring is more appropriate in some circumstances, such as the assessment of trophic status. Implementation of the EC Municipal Wastewater Treatment Directive will legally require algal monitoring (Harding & Hawley, 1991).

Monitoring with macrophytes has been used successfully, especially for the analysis of heavy metal concentrations, but for the general surveillance of organic pollution they are less sensitive than invertebrates or diatoms. The only macrophyte species appearing to greatly increase in range with organic pollution is the pondweed *Potamogeton pectinatus*, which is very tolerant (Mason, 1991). Macrophytes are stationary and visible to the naked eye, but seasonal variations in biomass and diversity are high. Macrophyte communities are largely governed by climate, geology and soil type and many freshwater systems such as high altitude streams have sparse floras (Hellawell, 1986).

Fish, being mobile organisms, tend to avoid pollution; species tend to be either present or absent with little gradation in between (Jeffries & Mills, 1990). A major disadvantage of using fish for routine environmental surveillance is the intensive and time consuming sampling effort required. The principal use of fish in biological monitoring work is tissue analysis to assess the health risks associated with the bioaccumulation of heavy metals and pesticides.

1.2 Diatoms in water quality monitoring

Diatoms have figured in river water quality studies since 1902. Contrasting ecological tolerances and distributions of many diatom taxa have long been recognised (Cox, 1991). Both short and long term (i.e paleolimnological) environmental changes can be inferred from the flora (Steinberg & Schiefele, 1988). Pollution often leads to great variations in the communities: it is difficult to obtain reliable data to establish tolerance limits of the species. Diatom species distribution is such that no two sites are alike, but because presence/absence can be added to abundance data, it is easier to define communities by the indicator species (Round, 1993). For example, "clean" waters

draining acid/rock soils support a flora dominated by small *Eunotia* spp., especially *E. exigua* (e.g. Llyn Brienne, Central Wales). Clean calcareous sites tend to be dominated by *Amphora pediculus* or *Achnanthes* (e.g. R. Tees, R. Avon), whereas "dirty" sites may be dominated by *Nitzschia palea* and *Gomphonema parvulum* (e.g. R. Mersey, R. Don). Many "dirty" sites support numerous small *Nitzschia* and *Navicula* spp. but these are difficult to identify accurately (Round, 1993).

Explanations of species distribution are usually based on correlation^s of relative abundance with water quality variables. Recent approaches have investigated diatom distributions in relation to a wider range of factors with defined ranges and optima and have used statistical tests to detect significant correlation^s (Cox, 1991). In general, indicator species are present throughout the year, but biomass and relative abundance^s change seasonally or continually (Round, 1993).

Biomonitoring systems based on correlation^s between measured environmental variables and species abundance often lack laboratory experiments to demonstrate causal relationships. Diatoms are easily cultured and are therefore amenable to experimental verification of their reaction (tolerance) to nutrients, pollutants etc. (Round, 1988). Freshwater diatom communities are highly diverse in relation to other communities and therefore the sensitivity of data interpretation is potentially greater (Coste *et al.*, 1991).

Round (1993) concludes that diatoms are very sensitive to water chemistry, eutrophication and pollution but relatively insensitive to physical features of the environment. However, there is much evidence in the literature to show that current velocity has a marked affect upon community composition (see 1.3.1). Diatoms are cosmopolitan and ubiquitous organisms; they are usually the most abundant autotrophic organisms in rivers and present along the whole length of a river/stream throughout the year. The same diatom species are found in many parts of the world and a massive literature exists (Round, 1991).

Studies on diatom assemblages associated with a variety of substrata indicate that contrasting microhabitats have distinctive floras (Round, 1992). Leclercq and Maquet (1987) have argued that indices based on diatom composition give more precise and valid predictions than benthic macro-invertebrates because they react directly to organic pollutants, whereas macro-invertebrates are more

influenced by substratum or current conditions. In eutrophic waters the influence of the substrata on the diatom community may be lower due to high nutrient availability (Cox, 1991). Unlike invertebrates, diatoms have no specific food requirements or specialist habitat niches (Round, 1992). However, algae in general show less sensitivity to pesticides and heavy metals than invertebrates (Hellowell, 1978).

The epilithic diatom community is sometimes visible as a brown mucilaginous coating, particularly during the spring bloom period. However, the epilithic diatom biomass can be very low or practically absent on some substrates and samples may need to be pooled to obtain a sufficient quantity for analysis. Usually a relatively small sample from the upper surface of a rock in the stream bed offers a good representation of the whole epilithic community, but replication is advisable (Round, 1993).

Diatom taxonomy is better studied than that of most algal groups (Whitton, 1991). Cell counting by microscope is rapid and accurate; random counts give an excellent assessment of the flora as cell densities are very high and permanent slides enable long term storage for future analyses. The advantage of examining live versus dead cells is that live cells can be associated with prevailing conditions whereas accumulated dead or inwashed specimens may have grown under different conditions. However, the specific identification of live cells is often more difficult as valve features may be obscured by organic matter. The estimation of the live/dead cell ratio remains problematical (Coste *et al.*, 1991).

The significance of slight morphological differences are largely unknown and the assessment of specific status can be highly subjective; life cycle morphological change may be confused with specific differences (Cox, 1991). Identifying species by girdle rather than valve views of the frustules or basing the identification on only one valve often requires considerable experience. Assemblage composition data are often quantified as relative abundances of taxa. However, this is related to the number of taxa present and does not take account of cell size and density. Cell size affects the species contribution to biomass and its division rate; smaller cells often have faster turnover rates; replication times are typically between 1 and 5 days (Cox, 1991).

1.3 Factors affecting diatom communities

If diatoms are to be used for routine water quality monitoring, all the environmental factors affecting them need to be clearly understood. The factors are often highly inter-related with synergistic or antagonistic interactions. For example, an increase in flow rate or current velocity will be associated with greater stream depth and therefore increased light attenuation. This may lead to lower temperatures and reduced photosynthesis, thus affecting the dissolved oxygen concentration. However, higher flow and current also create more turbulence which allows the water to become reoxygenated from the atmosphere.

Patrick (1977) carried out fundamental research and has published a comprehensive review of the environmental factors affecting stream communities. Discussion of diatom community response to flow rate is virtually absent from the literature, but there are many references to the effect of current velocity.

1.3.1 Flow rate and current speed

In fast flowing streams *Achnanthes minutissima*, *Achnanthes microcephala* and *Diatoma hiemale* are most often quoted as the dominants. Enhanced flow may also remove any contaminant silt / mucilage flora. Researchers have shown that colonisation (adhesion) is slower in fast flow than slow flow and it may take a few weeks for full floral development (Round, 1993).

The nutrient renewal rate and removal rate of potentially auto-toxic excretory products both increase with current (Patrick, 1977). Very low currents allow detritus accumulation (Wendker, 1992). Biomass and immigration rates of diatoms show an inverse relationship with current velocity (Antoine & Benson - Evans, 1982).

Schumacher & Whitford (1965) in Patrick (1977) found that the phosphorus uptake of *Eunotia pectinalis* at 0.18 m s^{-1} was about 7 times greater than in still water. Hustedt (1939) and others in

Patrick (1977) have noted that the life form morphology of some diatom taxa is affected by current speed.

In fast flowing waters, strongly attached forms of diatoms such as *Achnanthes* and *Cocconeis* are more common than in slower flowing waters where *Melosira varians* and many species of *Synedra*, *Gomphonema* and *Cymbella* are more typical (Patrick, 1977). Anyam (1990) found *Cocconeis* and *Diatoma* to be more abundant in stronger currents and *Navicula* and *Nitzschia* to be more abundant in weaker currents.

Antoine and Benson-Evans (1982) studied the effect of current velocity on the growth of benthic algal communities under controlled laboratory conditions using artificial channels. Filamentous and coccoid green algae grew better at lower current velocities; chlorophyll *a*, a correlate of algal biomass was inversely related to current. The Bacillariophyta species all reacted differently to the current velocities in the channels (i.e. 0.76 m s⁻¹, 0.88 m s⁻¹ and 1.48 m s⁻¹) and their distribution was related to their size and powers of adhesion. Species such as *Cocconeis placentula* and *Nitzschia palea* showed a marked inverse relationship to current velocity. *Synedra pulchella* was found in significant numbers at 0.76 m s⁻¹ but was not detected at 0.88 m s⁻¹ or 1.48 m s⁻¹. *Diatoma vulgare*, *Cocconeis placentula* var. *euglypta* and *Cymbella* (= *Reimeria*) *sinuata* showed optimum biomass at 0.76 m s⁻¹. *Melosira varians*, *Achnanthes lanceolata* and *Gomphonema parvulum* flourished best at 0.88 m s⁻¹, whereas *Fragilaria vaucheriae* (= *F. capucina* var. *vaucheriae*) grew better at 1.48 m s⁻¹ (Antoine & Benson-Evans, 1982).

There is a lack of information on the reaction of diatom communities to current velocity in natural streams (i.e. with factors such as detritus accumulation, nutrient availability and current dependent grazing of the herbivorous insects). Understanding the reaction of diatom taxa to variations in current velocity is important as it may affect water quality assessment. Zimmermann (1961) found that saprobian values (an organic pollution index) increased with current velocity where the chemical quality of the water was identical. Wendker (1992) found that diversity and evenness increased between current velocities of 0.005 m s⁻¹ and 0.5 m s⁻¹; much lower current velocities than Antoine & Benson - Evans were experimenting with.

1.3.2 Light

The intensity and duration of light necessary for optimum growth varies according to the diatom species under consideration. Therefore, the seasonal variation in diatom community structure is partly related to sunshine hours and other climate variables (i.e. temperature and precipitation). Sunshine seems to favour the development of *Cyclotella meneghiniana*, *Fragilaria capucina* and *Navicula cryptocephala*. Thus, bankside shading, the depth of the epilithon and the quality and quantity of suspended solids may significantly affect the diatom flora. Stream turbidity disturbs the ^{turbulence} sediments (increasing light attenuation due to suspended solids), but may also affect the micro-current patterns (Patrick, 1977). From a study on the River Ithon in Wales, Anyam (1990) showed that at stream depths below 1 m light intensity was not an important factor in controlling epilithic development; depth and biomass were not significantly correlated.

1.3.3 Temperature

Diatoms seem to have varying ranges of temperature tolerance. At optimum temperatures species diversity and biomass may increase. Temperature increases affect chemical diffusion rates and lower the water's dissolved oxygen capacity; these environmental changes may affect reproductive rates and metabolism (Patrick, 1977).

1.3.4 Oxygen

Temperature, light, nutrient levels and other environmental conditions seem to be important in determining the rates of photosynthesis and therefore oxygen production. Chohnoky (1968) has emphasised that *Achnanthes minutissima* requires a high oxygen concentration and correlated its⁹ increasing abundance with the re-oxygenation of water downstream of an organic pollution input (Chohnoky, 1968; in Patrick, 1977).

More study is needed to determine the extent to which oxygen generation by photosynthesis mitigates the effects of low dissolved oxygen. However, in severely eutrophic situations, with excessive algal growth, stream flow is impeded and available light is minimal. Therefore, photosynthesis cannot compensate for the deoxygenated conditions (Patrick, 1977).

1.3.5 pH

pH affects the carbonate-bicarbonate buffering system: at low pH carbon available for diatom growth is in the form of CO_2 or HCO_3^- , whereas at high pHs it is in the form of bicarbonate and carbonate. Hustedt (1956) has classified diatoms according to pH preference: alkalibionte (pH > 7), alkaliphile forms (pH +/- = 7), acidophile (pH < 7), acidobionte (pH < 5) and indifferent (wide tolerance) (Patrick, 1977). There has been extensive work using pH tolerance of lake diatoms to reconstruct pH change from fossil assemblages (Batterbee, 1986).

1.3.6 Nutrients

Ammonia, nitrate, and phosphate are utilized by diatoms in varying amounts. Many species of algae can accumulate large amounts of various nutrients under favourable conditions and therefore may not be dependent upon the external medium for some time after a nutrient has been reduced to a sub-optimal level. Species such as *Melosira varians*, *Synedra ulna* and *Cocconeis placentula* become very common in the presence of high nitrate concentrations (2 - 3 mg l⁻¹) in stream waters in eastern parts of the U.S.A (Patrick, 1977). Either phosphate or nitrate may be a limiting nutrient, although little is known about how the N:P ratio affects different species.

Silicate is required for the formation of diatom frustules. Different species require varying amounts of silicate; silicate is much more available in alkaline than in acid waters. The silicate (Si O₂) content of diatoms can vary between 30% and 70% of the organic weight (Patrick, 1977).

1.3.7 Substrata

There may be a slight effect of stone type (geology) on the colonising diatom flora; sandstone is thought to be most conducive to colonisation, especially in the early stages, due to surface pitting (Round, 1993). There is slight evidence that stone size affects the biomass of diatoms: lower on small stones (Duffer and Dorris, 1966, in Round 1993); the floristic composition relative to stone size needs checking.

1.3.8 Density-dependent factors

Abiotic factors regulate potential biomass levels, whereas biotic factors directly regulate realised biomass levels (Steinman, 1992).

Katoh (1992) studied the correlation between cell density and the dominant growth form of epilithic diatom assemblages. At an intermediately polluted site the relative abundance of adnate diatoms (*Cocconeis* and *Achnanthes* spp.) decreased, and rosette forming diatoms (*Achnanthes minutissima*, *Fragilaria capucina* var. *vaucheriae*) increased, with algal cell density. The relative abundance of stalked diatoms (e.g. *Gomphonema*) increased with algal cell density up to 10^5 cells.mm⁻². Benthic invertebrates which selectively graze the epilithic algae (favouring stalked taxa) modify the community composition such that adnate species tend to become dominant (Katoh, 1992).

1.3.8.1 Competition

The development of diatom communities on substrates is relatively well studied. Diatoms are usually the most important component of the epilithic algal community. Katoh (1992) recognises four general stages in the development of epilithic algal communities:

- a. Slime development of organic matter and bacteria.

b. Development of adnate algal layer.

Initially the community is a two dimensional one, with adnate species such as *Achnanthes lanceolata* and *Cocconeis placentula* forming a flat 'pavement like' growth over the substrate surface. Several researchers agree that *Achnanthes lanceolata* and *Cocconeis placentula* never co-dominate a substrate: the first coloniser out-competes the other. At this stage the number of microhabitats (as defined by the variations in current structure, nutrients and light effects, etc.) are relatively few (Patrick, 1977).

c. Development of vertically positioned algal layer (stalked or rosette forming taxa).

As the adnate layer develops, an irregular micro-current pattern develops and species begin to stand upright by producing jelly pads or jelly stalks. Thus, a three dimensional community with several microhabitats, maintaining a higher species diversity is created (Patrick, 1977). Lamb & Lowe (1981, 1987) and Poff *et al.* (1990) all found that 3-dimensional growth was greater at lower current velocities.

d. Development of filamentous algal layer.

Diatoms excrete organic compounds which are either autotoxic, heterotoxic or stimulatory. Autoantibiosis has been observed by von Denffer (1948) in *Nitzschia palea*. Jorgensen (1956) found that *Asterionella formosa* and *Nitzschia palea* produced antibiotics against each other when grown together (Patrick, 1977).

Interactions between algal populations influence community composition: e.g. by competition for nutrients and light, area and space, growth inhibition or stimulation by extracellular products and growth promotion by the creation of three-dimensional structures. Algae other than diatoms (e.g. *Cladophora glomerata*) may have a greater influence on the diatom communities than factors such as organic pollution or pH (Elber & Schanz, 1990).

1.3.8.2 Parasitism and predation

Observations by Patrick *et al.* (1977) have shown that certain insect larvae will select certain species of diatoms such as *Rhoicosphenia curvata*. Patrick (1977) also notes that protozoans, particularly ciliates, select diatoms of a given shape and size as their food.

Winterbourn *et al.* (1992) studied the relationship between algal biomass accumulation, invertebrate colonization and stream-water pH in three regions of England and Wales. The abundance of Chironomidae larvae (the main epilithic invertebrates) was positively related to algal pigment concentration (chlorophyll *a* and phaeopigments) over a wide pH range (Winterbourne *et al.*, 1992).

Contrary to the findings of Marker and co-workers who propose silicate as the controlling factor, Steinman (1992) asserts that biomass level appears to be ultimately constrained by light level and herbivory. In a woodland stream irradiance level and grazer density (mainly the snail *Elimia clavaeformis*) were manipulated in a factorial design to examine the relative effects of biotic and abiotic factors on periphyton biomass and taxonomic structure. Large or upright diatoms became more abundant when grazer density was reduced and light intensity increased.

1.4 Water Quality Indices

Indices are an important management tool for interpreting water quality in terms of community composition. Indices reduce floristic data to a numerical form by classifying species/genera according to pollution sensitivity, water quality indicating value and relative abundance. Various indices have been devised for diatoms: (Kolkwitz & Marsson, 1908, Patrick *et al.*, 1954, Patrick & Hohn, 1956, Fjordingstad, 1964, Descy, 1979, Schwertfeger, 1980, Leclercq & Maquet, 1987, Watanabe, 1988, etc.). Most tend to be based on a limited number of water quality variables associated with organic pollution (Cox, 1991). Different experts apply different weightings to the commonly used variables (i.e. pH, biochemical oxygen demand, phosphate, nitrate, temperature,

conductivity and dissolved oxygen). The saprobic system is based on the nutrient chemicals ammonia, nitrate and phosphate. Diatom indices have been widely criticised, mainly due to the mathematical expressions used. Ratings give incomplete information, but for water management purposes are useful; all indices and chemical data are to some extent reductionist. A major problem with most indices is that they are poor at separating the effects of eutrophication, heavy metal contamination and organic pollution (Round, 1992).

Diversity has been shown by many workers (e.g. Lange-Bertalot, 1979) to be unsuitable for water quality monitoring and also requires time consuming large counts in order to include all the rare taxa (Round, 1993). Washington (1984), in Round (1991a), found that only 3 out of 18 diversity indices were valuable. Archibald (1972), in Round (1991a) found diversity was not closely correlated with water quality.

Watanabe (1981) developed the diatom community index (DCI). This index is based on his idea that the degree of water pollution in rivers actually changes gradually and continuously and therefore can not be classified into definite classes (Sumita & Watanabe, 1983).

Many researchers have compared indices applied to invertebrates and diatoms. A good correspondence was found between assessments obtained from diatoms and oligochaetes with physico-chemical parameters (orthophosphate) despite these two groups of organisms having different habitats. In comparisons between chemical and diatom indices on the Rhone basin, Descy and Coste (1988) demonstrated biologically accurate estimates of water quality using diatoms (Coste *et al.*, 1991).

This study uses 3 indices: SPI (Species Pollution Index, Coste in CEMAGREF, 1982), GDI (Generic Diatom Index, Rumeau and Coste, 1988) and the zoning system proposed by Round (1993).

1.5 Aims

The effect of flow on the epilithic diatom community may be related to the size of the substrate as well as the hydrological regime of the stream. Therefore community changes were studied over

time on cobbles and boulders at two sites (upland and lowland). It was thought that diatom biomass may be higher, and less variable, on boulders which remain stable at higher flow rates than cobbles. Therefore, variations in diatom community structure and diatom-based water quality indices may decrease with substrate stability.

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The aim of this project is to investigate how the epilithic diatom community responds to flow events and consequently how these events may affect the classification of water quality by commonly used diatom water quality indices.

CHAPTER 2

MATERIALS AND METHODS

2.1 Physico-chemical measurements

Stream width was measured to the nearest 0.1 m at the same position on each field visit. Stream depth was measured at 3 positions mid-stream and maximum depth was estimated to +/- 5 cm. Temperature ($^{\circ}\text{C}$) and conductivity ($\mu\text{S cm}^{-1}$) were measured using a WTW (Wissenschaftlich-Technische Werkstätten) meter (model FC910).

A calibrated Ott current meter was positioned at the point of fastest current with the propeller facing the flow direction at about a third of the depth below the surface. Current was measured over a 60 second period. Flow data ^{were} obtained from National River Authority automatic recording stations (25 m downstream of the Harwood Beck sample site and 4 km downstream of the River Browney sample site).

pH was determined using a WTW meter (model pH91). Total alkalinity was determined by titrating 50 ml or 100 ml of stream water with 0.02 M HCl to an end point of pH 4.2 and calculated using the following equation:

$$\text{Alkalinity (mg l}^{-1}\text{ CaCO}_3) = \frac{\text{volume of HCl added to end point (ml)} \times \text{normality of acid} \times 50\,000}{\text{volume of sample (ml)}}$$

$$\text{Total alk. (meq l}^{-1}\text{)} = \text{alk. (mg l}^{-1}\text{ CaCO}_3) \times 0.0499 \quad (\text{Golterman } et\ al., 1978)$$

2.2 Collection of water samples

Separate water samples for phosphorus and inorganic nitrogen analysis were collected in 250 ml, polypropylene bottles, which had been washed in 10% H_2SO_4 for at least an hour, rinsed several times in distilled water and rinsed twice with stream water before filling. Samples were returned to the laboratory in an ice box.

2.3 Collection of diatom samples

Cobbles were defined as being between 64 and 150 mm in diameter and boulders over 256 mm in diameter according to the Wentworth scale (Gordon *et al.*, 1992). Diatoms were sampled

quantitatively from the upper surface of cobbles and boulders collected from a defined 10 m reach. Most of the cobbles and boulders sampled were sandstones and tended to have smooth, flat surfaces. Stones coated with silt or with macroscopic algal growths were avoided in order to select the epilithic flora and so that between rock diatom biomass variability measurements were more reliable. The sampling method was devised after careful consideration of the methods from Douglas (1958) and Thorpe & Williams (1980). The sampling area was defined using the base of a cleanly cut 125 ml polythene bottle (area $\approx 20 \text{ cm}^2$). The diatoms were transferred to sample bottles using a stiff nylon brush and distilled water. Replication of sampling from 4 cobbles and 4 boulders was used to assess biomass and chlorophyll variation. The samples were transported to the laboratory in an ice box.

2.4 Laboratory procedures

Water for phosphorus analysis was filtered immediately on arrival through Whatman GF/F filters washed with Milli-Q (MQ) water. Filtrable Reactive Phosphorus (FRP or orthophosphate) and Filterable Total Phosphorus (TFP) were determined on the same day using the molybdenum-blue method of Murphy and Riley as modified by Eisenreich *et al.* (1975) with a detection limit in the order of $1 \mu\text{g l}^{-1} \text{ P}$.

Water for Nitrite, Nitrate and Ammonia analyses was deep frozen so that several weeks samples could be analysed together. Nitrite was determined using a method based on N-1-naphthylethylenediamine dihydrochloride (Stainton *et al.*, 1977). Nitrate was reduced to nitrite by a cadmium-copper couple and analysed as for nitrite. Ammonia was determined by the indophenol blue method (Stainton *et al.*, 1977). The detection limit of these methods is in the order of $5 \mu\text{g l}^{-1} \text{ N}$.

2.5 Biological analysis

The diatom samples were each made up to 100 ml with distilled water and from the homogenous suspension 10 ml was stored for diatom analysis, 30 ml was frozen for chlorophyll extraction and 60 ml (for biomass) was filtered through 4.7 cm Whatman GF/C, glass microfibre filters of known dry weight. The filters were then put back in the oven overnight at $105 \text{ }^\circ\text{C}$ and allowed to cool for a few hours in a desiccator before being re-weighed to estimate biomass.

30 % of the quantitative sample for chlorophyll determination was filtered through 2.5 cm GF/C (glass microfibre) filters. Chlorophyll a content was estimated by extraction in 5 ml of 90 % methanol at 70 °C. The extraction was carried out in 30 ml snap cap bottles covered with foil to exclude light, and samples were stored in a cool dark place to prevent chlorophyll breakdown. This was repeated to obtain 10 ml samples for which absorbance was measured at 665 nm and 750 nm using a Shimadzu dual-beam spectrophotometer. 0.1 ml of 0.1 M HCl was then added, and the samples were left for one hour in the refrigerator, before absorbance at 665 and 750 nm was re-measured in order to estimate the phaeophytin content. The amount of chlorophyll a in µg (C) was calculated as follows:

$$C = (A_b - A_a) \cdot [R/R-1 \cdot K \cdot V/L]$$

Where:

A_b = absorbance of extract at 665 nm before acidification less the absorbance at 750 nm

A_a = absorbance of extract at 665 nm after acidification less the absorbance at 750 nm.

R = maximum acid ratio (i.e. A_b / A_a) for extracts containing no phaeopigments.
= 1.59 for 90 % methanol in 1×10^{-3} M HCl

K = 1000x the reciprocal of the specific absorption coefficient (SAC) of chlorophyll a at 665nm
= 12.99

V = volume of solvent used to extract the sample = 10 ml.

L = path length of the cuvette = 4 cm.

Diatoms contain chlorophyll a and chlorophyll c. The chlorophyll a content of diatoms lies in the range of 0.3-2 % of the dry weight of algae including diatoms (Werner, 1977).

From 3 June, biomass measurements were discontinued as chlorophyll a was thought to give a better estimate of the diatom biomass; biomass and chlorophyll a were poorly correlated. As the 10 % stored for slides proved to be an inadequate quantity to obtain slides of suitable density for counting, 50% of the quantitative samples were subsequently frozen for chlorophyll analysis and 50% stored for diatom slide preparation. However, even the 50% or more stored for diatom slides was rarely enough, due to the decline in biomass from late May, and slides were produced from pooled cobble and boulder samples.

2.6 Diatom slide preparation

The method is based on that of J.R. Carter (pers comm.). Batterbee (1986) explains the general principles. Glassware was scrupulously cleaned and separate stirring rods were used for each sample in order to prevent cross-contamination. The samples were allowed to settle in the refrigerator, the supernatant decanted and the sediment transferred to a boiling tube and allowed to settle. Dilute HCl was used to dissolve calcareous material and the samples were centrifuged in distilled water. Organic matter was digested by the addition of 5 ml concentrated H₂SO₄, two crystals of potassium permanganate and 10 ml of saturated oxalic acid. The solution was allowed to sediment overnight and was then centrifuged in distilled water several times to remove all traces of acidity. A few drops of this solution were heated on a cover slip until dry and mounted on a slide using naphrax, a high resolution mountant (refractive index = 1.74). Live slides did not generally allow the precision of identification to species level due to the valves being obscured by organic matter. Sometimes sonication of the sample was required to break up the mucilage and disperse the diatom valves and frustules.

2.7 Diatom counts

Cobble and boulder samples were pooled in order to obtain slides of sufficient density for counting. Slides of counting density could only be produced for 10 replicates. Approximately 200 valves per slide were counted. This allowed representation of over 90 % of the taxa present to be recorded by percentage. Counting errors were minimised by only identifying the diatom by valve view and only if over 50 % of the valve was visible (valves sometimes fragment). Any remaining taxa were identified by scanning the slide for an additional 10 minutes but were recorded as being present only and not assigned a percentage value.

Taxonomic identification to species level was achieved using Barber and Haworth (1978) and Krammer and Lange-Bertalot (1986, 1988 and 1991).

2.8 Diatom guild classification

Molloy (1992) classified diatom taxa into guilds, distinguished on the basis of morphological features associated with their mode of attachment. When the classification system is applied to the diatom flora of the River Browney and Harwood Beck the following 6 guilds account for most diatom taxa:

1. *Achnanthes*: small size, monoraphid, generally prostrate orientation to the substrate.
2. *Cocconeis*: concave, monoraphid; prostrate orientation to the substrate and large amounts of mucilage (strong adherence).
3. Adnate: adjacent to substrate surface without being prostrate or erect (including *Rhoicosphenia*, *Amphora* and *Surirella*).
4. *Navicula* spp.: biraphid, generally prostrate, frequently motile.
5. Stalked: stalk forming genera including *Cymbella* and *Gomphonema*.
6. Erect: perpendicular to substrate without stalks; often forming rosettes. Generally araphid or pseudoraphid: *Fragilaria*, *Diatoma*, *Synedra*, *Asterionella*, *Meridion*.

2.9 Pollution Indices

2.9.1 The Coste SPI and GDI were calculated according to the following formula:

$$\text{(Diatom Index) ID} = \frac{\sum a_j \cdot v_j \cdot i_j}{\sum a_j \cdot v_j}$$

where a = relative abundance (\approx %), v = indicator value (1 = poor indicator, 3 = good indicator) and i = sensitivity to pollution (5 = very sensitive, 1 = tolerant) (Coste *et al.*, 1991).

ID > 4.5 : 'best biological quality, no pollution'.

ID = 4-4.5 : 'almost normal quality (slight changes in the community, slight pollution)'.

ID = 3-4 : 'more important changes in the community, decrease of the sensitive species, moderate pollution or significant eutrophication'.

ID = 2-3 : 'resistant species dominant, decrease or disappearance of the sensitive species (reduced

diversity), heavy pollution'.

ID = 1-2 : 'marked dominance of a few resistant species (many species disappear), very heavy pollution'. (Descy, 1979).

2.9.2 Round (1993)'s Zone System

Table 2.1. Round (1993)'s system of zonation for running water sites based on epilithic diatom community composition.

Zone	Definition	Dominant diatoms
1	Clean water in uppermost reaches	<i>Eunotia exigua</i> <i>Achnanthes microcephala</i>
2	Nutrient richer and somewhat higher pH pH 5.6-7.1, alkalinity (mg l ⁻¹ CaCO ₃) 2.8 - 5.7 Leclercq (1977)	<i>Ceratoneis arcus</i> <i>Fragilaria capucina</i> <i>Achnanthes minutissima</i>
3	Nutrient rich pH 6.5-7.3, alkalinity 5.0-23.3 Leclercq (1977, 1988)	<u>downstream zonation</u> : <i>Achnanthes minutissima</i> (upper region) <i>Cymbella minuta</i> (middle region) <i>Cocconeis placentula</i> (lower-region) <i>Reimeria sinuata</i> (lower-region) <i>Amphora pediculus</i> (lower-region)
4	Eutrophic with restricted flora due to the detrimental influx of materials	<i>Gomphonema parvulum</i> distinguished by the relative absence of: <i>Amphora</i> / <i>Cocconeis</i> & <i>Reimeria</i>
5	Flora grossly restricted by detrimental influx of materials	Small <i>Navicula</i> and <i>Nitzschia</i> species <i>Gomphonema parvulum</i> <i>Gomphonema auger</i> <i>Navicula accomoda</i> <i>Navicula goeppertiana</i> <i>Amphora veneta</i>

CHAPTER 3

FIELD SITES

3.1 Introduction

Sampling was undertaken between 3 May and 17 July 1993. The river Browney (NZ 222 455), a lowland eutrophic stream, was sampled weekly and Harwood Beck (NY 849 309), an oligotrophic stream in Upper Teesdale (altitude: 380 m), was sampled fortnightly until 3 June when both sites were sampled approximately every 10 days.

3.3 River Browney

The River Browney drains a millstone grit and coal measure catchment of approximately 178.5 km². The landscape is undulating agricultural land used for both crops and pasture and the nutrient inputs are from agricultural runoff and a small sewage treatment works about 5 km upstream of the sampling site.

Fig. 3.1. River Browney study site (3 October 1993)



Table 3.1 River Browney chemical data (pers.comm., M.G. Kelly).

Date	Concentrations mg l ⁻¹ :								
	Na	K	Mg	Ca	Fe	Zn	Si	O2	BOD 5 day
24 April 1992	25	4.5	19.5	44	0.17	0.014	-	13.5	1.4
28 July 1992	43.5	6.95	24.5	67.5	0.12	0.006	1.75	13.2	-
22 October 1992	39	7.15	19	51.5	0.21	0.01	6.38	-	1.9
22 January 1993	25.5	4	12.5	37	0.32	0.016	2.6	-	3
27 April 1993	21	3.9	12.5	34.5	0.33	0.011	6.8	13.4	1.9
Mean	30.8	5.3	17.6	46.9	0.23	0.011			
Standard deviation	9.8	1.6	5.1	13.3	0.09	0.004			

(Note: Metal concentrations were determined using atomic absorption spectrophotometry; silicate (= molybdate reactive silicon) was determined using the Ascorbic Acid Reduction Method; Dissolved oxygen was measured with a WTW meter and BOD was determined by incubation in the dark for 5 days at 20°C in the presence of allylthiourea).

Table 3.2. Some River Browney physico-chemical parameters over study period.

Statistic	Temp. (°C)	Cond. (µS cm ⁻¹)	pH	Alk. (meq l ⁻¹)
Mean	11.3	544	?	5.7
Standard Deviation	3.2	89	0.4	1.8
Range	8.6	272	1.1	5.4
Minimum	6.6	394	7.1	2.7
Maximum	15.2	666	8.2	8.1

3.2 Harwood Beck

Harwood Beck drains a catchment area of 25.1 km² where the geology is of the carboniferous limestone series. The catchment area consists of moorland and rough grazing land and the topography is hilly.

The two streams are of similar dimensions but the difference in altitude and the much greater degree of bankside shading at the River Browney site are key physical factors. The altitude extremes and marked contrast in land use between sites can be considered as fundamental factors affecting nutrient status.

Table 3.3 Harwood Beck chemical data (pers.comm., M.G. Kelly).

Date	Concentrations mg l ⁻¹								O2	5-day BOD
	Na	K	Mg	Ca	Fe	Zn	Si			
24 April 1992	5.6	0.96	3.4	38	0.07	0.026			12.6	< 1.0
28 June 1992	5.2	1.25	4.45	47.5	0.03	0.019	1.05		11.6	1.0
22 October 1992	4.8	0.9	3.5	38	0.15	0.001	1.55		12.9	1.1
22 January 1993	5.9	0.65	1.75	15.5	0.26	0.043	0.48			< 1.0
27 April 1993	5.1	0.75	2.45	31.5	0.2	0.028	1.55		13.8	< 1.0
Mean	5.32	0.9	3.11	34.1	0.14	0.023				
Standard deviation	0.43	0.23	1.04	11.9	0.09	0.015				

(methods as for Table 3.1)

Table 3.4 Some physico-chemical parameters for Harwood Beck over study period.

Statistics	Temp. (°C)	Cond. (µS cm ⁻¹)	pH	Alk. (meq l ⁻¹)
Mean	11.5	244		6.2
Standard Deviation	3.6	92		3.6
Range	8.8	234	1.5	10.1
Minimum	5.7	112	6.9	1.9
Maximum	14.5	346	8.4	12.0

Fig. 3.2. Harwood Beck study site (14 August 1993)



CHAPTER 4

RESULTS

4.1 Flow

The maximum flow at both sites was recorded on 14 May (6 days before next visit). There were two smaller stormflow events in Harwood Beck at the end of May and mid-July. A comparison of Figures 4.1 and 4.2 shows the maximum flow in the River Browney to be about four times that in Harwood Beck.

Figure 4.1. Flow rate of River Browney at Burn Hall over the study period.

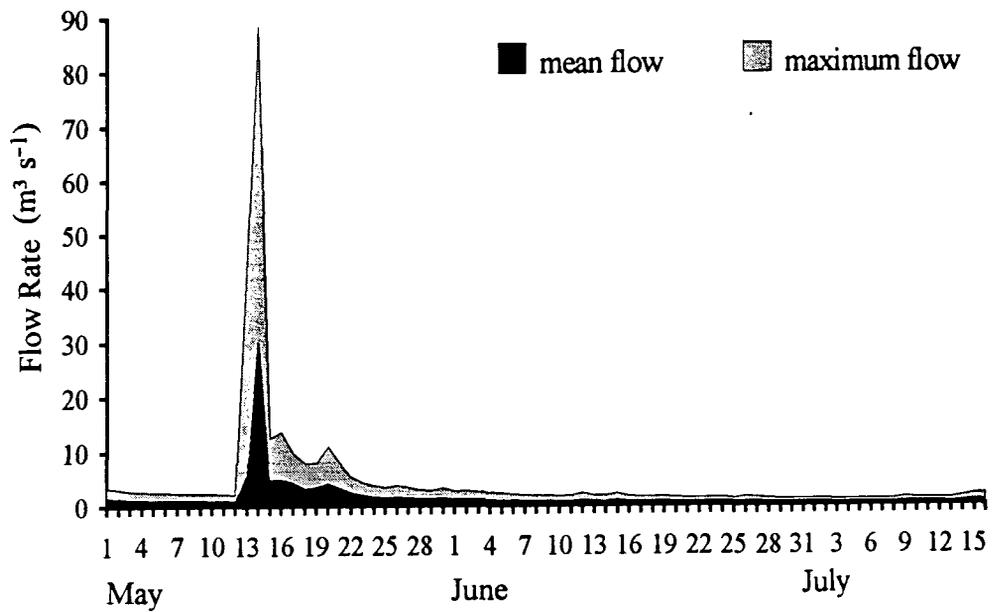
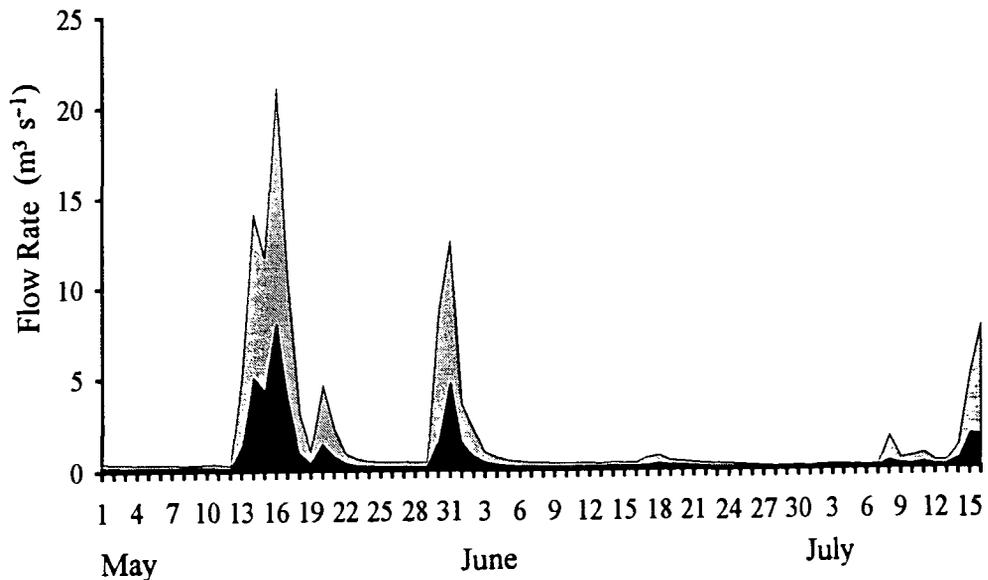


Figure 4.2. Flow rate of Harwood Beck over study period.



4.2 Other environmental conditions over study period

The River Browney and Harwood Beck had similar widths, depths and temperatures over the study period (Table 4.1). The range in conductivity was approximately $250 \mu\text{S cm}^{-1}$ at both sites, but the minimum recorded in the River Browney ($394 \mu\text{S cm}^{-1}$) was higher than the maximum in Harwood Beck ($346 \mu\text{S cm}^{-1}$). The mean conductivity in the River Browney is over twice that of Harwood Beck which probably reflects the higher calcium and magnesium levels of the River Browney (i.e. 'harder' water). This is supported by the data in Tables 3.1 and 3.3. Current speed varied by a factor of two at the River Browney and a factor of four at Harwood Beck.

Table 4.1. Physical measurements of the River Browney (B) and Harwood Beck (H).

Date	Site	Stream depth (m)	Stream width (m)	Temperature ($^{\circ}\text{C}$)	Conductivity ($\mu\text{S cm}^{-1}$)	Current (m s^{-1})
03 May	B	0.32	10.3	8.4	465	0.47
12 May	B	0.38	11.0	10.9	559	0.45
20 May	B	0.45	9.0	6.6	394	0.90
26 May	B	0.35	8.4	7.3	494	0.76
03 June	B	0.26	8.2	12.4	523	0.80
17 June	B	0.25	7.3	13.2	624	0.55
23 June	B	0.29	7.4	12.5	646	0.59
06 July	B	0.15	7.2	14.9	666	0.55
16 July	B	0.35	7.4	15.2	524	0.89
Mean		0.31	8.5	11.3	544	0.66
Standard deviation		0.03	0.5	1.1	30	0.06
Minimum		0.15	7.2	6.6	394	0.45
Maximum		0.45	11.0	15.2	666	0.90
Date	Site	Stream depth (m)	Stream width (m)	Temperature ($^{\circ}\text{C}$)	Conductivity ($\mu\text{S cm}^{-1}$)	Current (m s^{-1})
03 May	H	0.26	8.5	5.7	278	0.59
20 May	H	0.28	8.2	7.0	209	0.91
03 June	H	0.43	9.6	12.7	137	0.38
17 June	H	0.14	7.4	13.3	314	0.34
23 June	H	0.22	6.6	14.5	310	0.33
06 July	H	0.12	6.3	13.3	346	0.27
16 July	H	0.50	10.2	13.9	112	1.13
Mean		0.28	8.1	11.5	244	0.56
Standard dev.		0.05	0.55	1.4	35	0.13
Minimum		0.12	6.3	5.7	112	0.27
Maximum		0.50	10.2	14.5	346	1.13

Mean values of pH and Total Alkalinity are approximately equal for the River Browney and Harwood Beck (Table 4.2). The major difference in the chemistry of the two sites is nutrient status. Harwood Beck is generally oligotrophic whilst the River Browney is eutrophic. Preliminary data analysis did not reveal any significant correlations of pH, alkalinity, conductivity and temperature with other variables.

Table 4.2. Chemical measurements of the River Browney (B) and Harwood Beck (H).

NH₄ - N, ammonium-nitrogen; NO₂ - N, Nitrite; NO₃ - N, Nitrate; TIN, Total Inorganic Nitrogen (= nitrite, nitrate and ammonium); FTP, Filterable Total Phosphorus; FRP, Filterable Reactive Phosphorus; -, Unknown

DATE	SITE	pH	Total Alk. meq l ⁻¹	NH ₄ - N µg l ⁻¹	NO ₂ -N µg l ⁻¹	NO ₃ N µg l ⁻¹	TIN µg l ⁻¹	FTP µg l ⁻¹	FRP µg l ⁻¹	N: TFP	TIN: FRP
03- May	B	8.2	7.8	40	12	503	555	325	170	1.5	3.3
12- May	B	7.5	5.7	155	36	514	705	488	339	1.1	2.1
20- May	B	7.6	2.7	108	17	1008	1133	188	159	5.4	7.1
26- May	B	7.9	5.2	81	20	836	937	226	192	3.7	4.9
02-June	B	7.8	4.5	70	45	1311	1426	375	310	3.5	4.6
17-June	B	7.3	6.8	183	239	1191	1613	472	445	2.5	3.6
23-June	B	7.8	6.5	317	100	1248	1665	535	508	2.3	3.3
06-July	B	8.1	8.1	159	258	1098	1515	393	365	2.8	4.2
16-July	B	7.1	4.3	372	108	927	1407	778	647	1.2	2.2
Mean		7.7	5.7	165	93	960	1217	420	348	2.7	3.9
Standard		0.1	0.6	38	32	99	135	59	55	0.5	0.5
Minimum		7.1	2.7	40	12	503	555	188	159	1.1	2.1
Maximum		8.2	8.1	372	258	1311	1665	778	647	5.4	7.1
DATE	SITE	pH	Total Alk. meq l ⁻¹	NH ₄ - N µg l ⁻¹	NO ₂ -N µg l ⁻¹	NO ₃ N µg l ⁻¹	TIN µg l ⁻¹	FTP µg l ⁻¹	FRP µg l ⁻¹	N: TFP	TIN: FRP
03- May	H	8.4	12.0	<5	< 1.0	54	55	16.5	9.9	3.3	5.5
20- May	H	8.3	4.0	11	< 1.0	91	103	22.2	8.7	4.1	11.8
02-June	H	7.8	2.7	10	1.7	41	53	11.2	< 5.0	3.7	-
17-June	H	8.0	7.6	17	< 1.0	25	42	< 5.0	< 5.0	-	-
23-June	H	8.2	7.9	17	< 1.0	11	28	< 5.0	< 5.0	-	-
06-July	H	7.8	7.5	11	< 1.0	83	94	24.0	20.0	3.5	4.7
16-July	H	6.9	1.9	20	< 1.0	17	37	14.8	13.1	1.1	2.8
Mean		7.9	6.2			46	59	< 14	< 9.5		
Standard dev.		0.2	1.3								
Minimum		6.9	1.9			11	28	< 5.0	< 5.0		
Maximum		8.4	12.0			91	103	24	20.0		

4.3 Flow and nutrients

Concentrations of FTP and FRP tend to increase in the water during low flow periods

(Fig. 4.3). No clear relationship is apparent between mean flow rate and the concentration of Total Inorganic Nitrogen ($\mu\text{g l}^{-1}$), but in the River Browney TIN gradually increases over the spring and summer, reaching a peak level of over 1.6 mg l^{-1} by the end of June (Fig. 4.4)

Figure 4.3. Relationship between mean flow rate in River Browney and Filtrable Total Phosphorus.

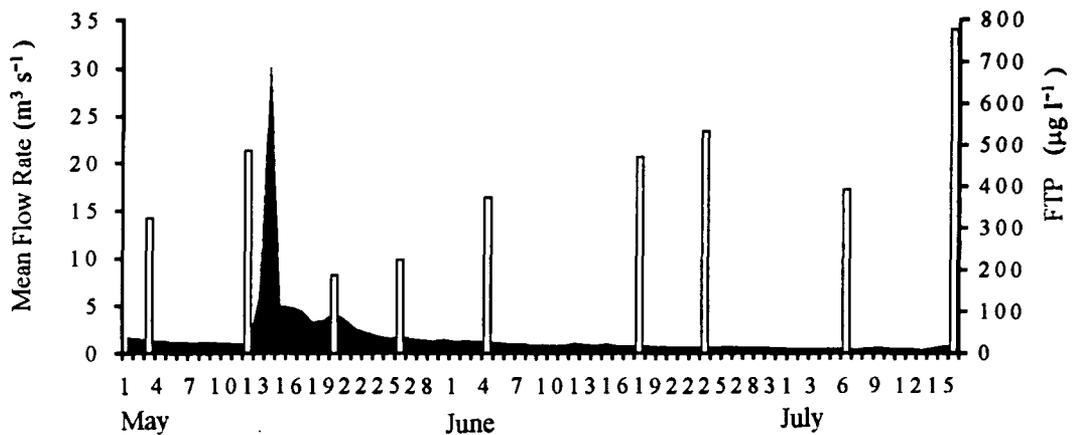


Figure 4.4. Relationship between mean flow rate in River Browney and Total Inorganic Nitrogen.

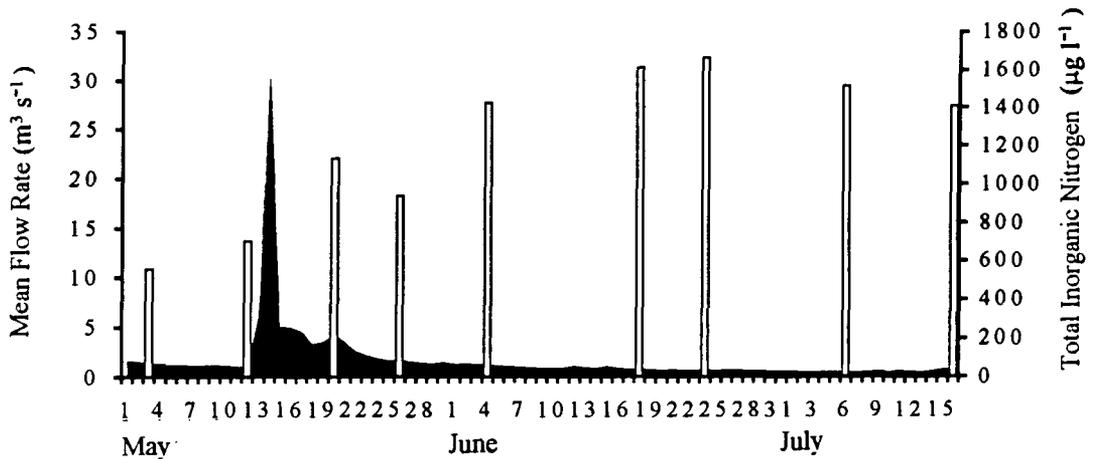
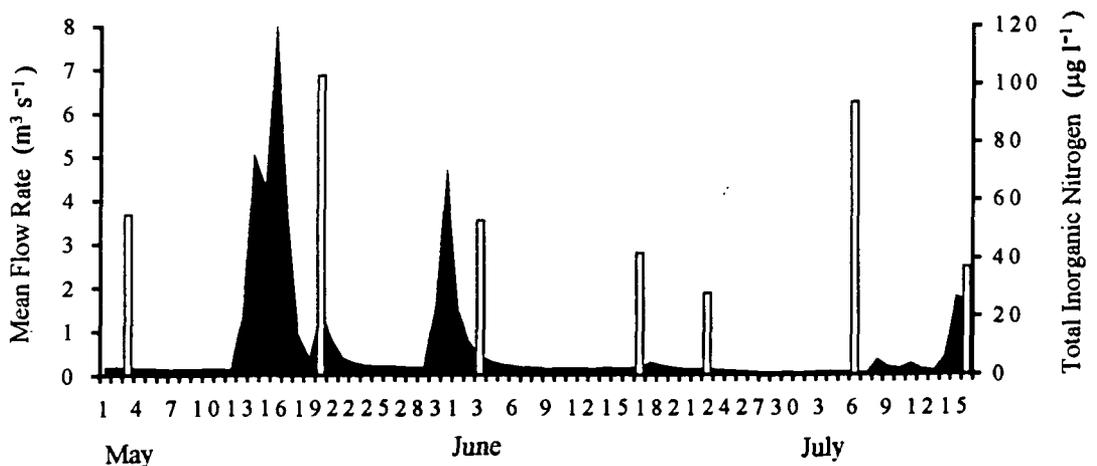


Figure 4.5. Relationship between mean flow rate in Harwood Beck and Total Inorganic Nitrogen.



4.4 Chlorophyll *a* during study

4.4.1 River Browney

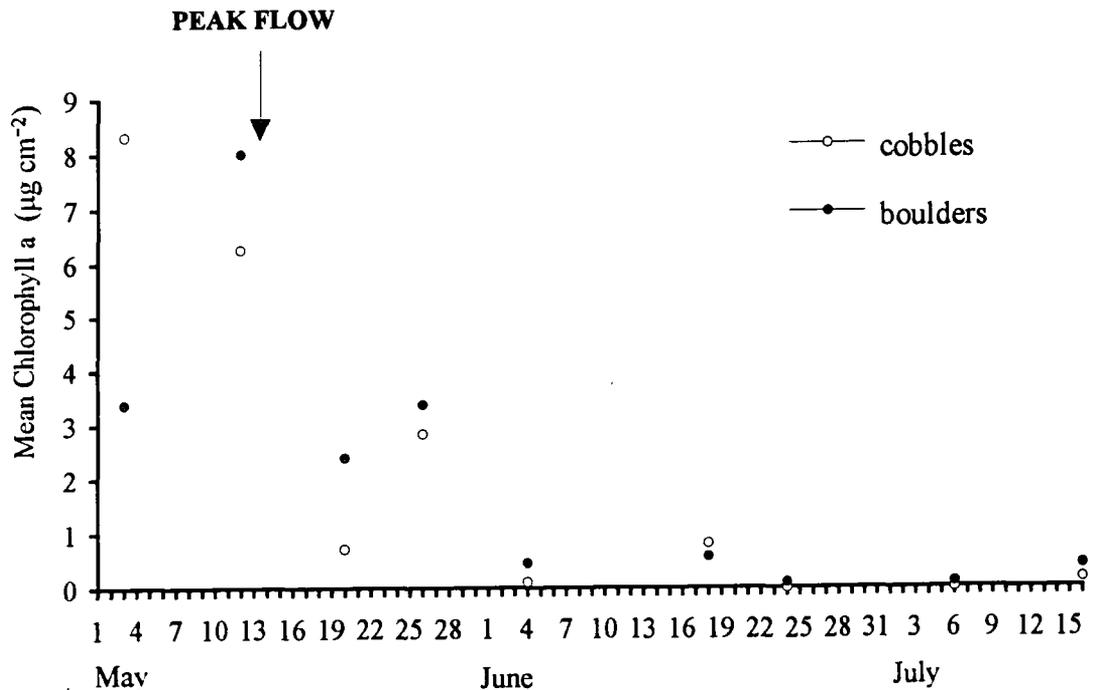
Chlorophyll *a* ($\mu\text{g cm}^{-2}$) is highly variable on both cobbles and boulders. However a steep drop in chlorophyll *a* occurred after the major storm on 20 May (Fig. 4.6). An analysis of variance on \log_{10} transformed values shows significant differences in chlorophyll *a* between dates ($p < 0.01$), but not between cobbles and boulders (Table 4.3).

Table 4.3. Analysis of variance of \log_{10} transformed chlorophyll *a* on cobbles and boulders over time.

Source of Variation	Sum of Squares	degrees of freedom	Mean Square	F statistic	F critical
Date	11.265	5	2.253	3.355	2.477
Substrate	0.012	1	0.012	0.018	4.113
Interaction	2.748	5	0.550	0.818	2.477
Error	24.176	36	0.672		
Total	38.201	47			

Two sampling dates (23 June and 6 July) are omitted from the analysis as there was insufficient material for chlorophyll extraction. There is a close relationship between chlorophyll *a* on boulders and cobbles ($r = 0.73$, $p < 0.05$).

Figure 4.6. Mean chlorophyll *a* ($\mu\text{g cm}^{-2}$) on cobbles and boulders in the River Browney.



There is a slight recovery in chlorophyll *a* (26 May) during a period of low flow following the storm (Fig. 4.6). Chlorophyll *a* is not significantly higher on cobbles or boulders during low flow periods, but appears to be higher on boulders during high flow periods. On 3 May, during a low flow spring bloom period, mean chlorophyll *a* on cobbles was over twice as high than on boulders.

4.4.2 Harwood Beck

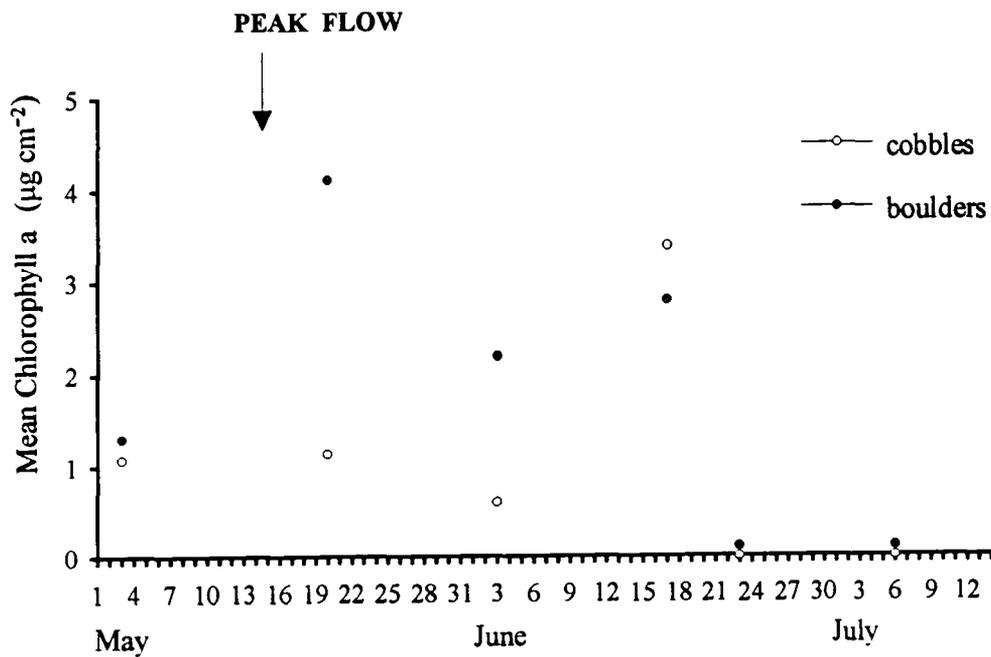
The chlorophyll *a* is highly variable on both cobbles and boulders. An analysis of variance on \log_{10} transformed values shows significant differences in the chlorophyll *a* between dates and between cobbles and boulders ($p < 0.01$ and $p < 0.05$, respectively). The less intense stormflow at Harwood Beck may have been between cobble and boulder stability thresholds and therefore caused a greater difference in chlorophyll *a* on cobbles and boulders.

Table 4.4. Analysis of variance of \log_{10} transformed chlorophyll *a* on cobbles and boulders over time.

Source of Variation	Sum of squares	degrees of freedom	Mean square	F statistic	F critical
Date	6.910	3	2.303	15.237	3.009
Substrate	0.682	1	0.682	4.512	4.260
Interaction	0.557	3	0.186	1.227	3.009
Error	3.628	24	0.151		
Total	11.777	31			

Maximum chlorophyll *a* was recorded on boulders after the storm on 20 May (Fig. 4.7). This co-occurred with the maximum recorded nutrient level. However, chlorophyll *a* on cobbles remained low. By 17 June, after 2 weeks of low flow, mean chlorophyll *a* on cobbles reached a maximum (in very low nutrient conditions). From 23 June chlorophyll *a* remained very low ($< 0.5 \mu\text{g cm}^{-2}$) in Harwood Beck (Figs. 4.6 and 4.7).

Figure 4.7. Mean chlorophyll *a* ($\mu\text{g cm}^{-2}$) on cobbles and boulders in Harwood Beck.



4.4.3 Relationship with other environmental variables.

Chlorophyll *a* was generally poorly correlated with physical variables. The only statistically significant correlation occurred between chlorophyll *a* on cobbles and current velocity in the River Browney.

However, in the River Browney, mean chlorophyll *a* on cobbles and boulders were both inversely correlated with Total Inorganic Nitrogen ($r = -0.9$, $p < 0.01$; $r = -0.8$, $p < 0.05$ respectively), but not with TFP ($r = -0.2$). At Harwood Beck, Total Inorganic Nitrogen was inversely correlated with chlorophyll *a* on boulders, but not on cobbles (Table 4.5).

Table 4.5. Chlorophyll *a* correlation with current speed, \log_{10} mean flow rate, TIN and TFP.

		Chlorophyll <i>a</i> ($\mu\text{g cm}^{-2}$)	
		Cobbles	Boulders
River Browney	Current (ms^{-1})	-0.79	-0.59
	Log 10 mean flow	-0.23	-0.07
Harwood Beck	Current (ms^{-1})	-0.61	-0.27
	Log 10 mean flow	-0.58	-0.26
River Browney	Total Inorganic N ($\mu\text{g l}^{-1}$)	-0.90	-0.80
	TFP ($\mu\text{g l}^{-1}$)	-0.20	-0.20
Harwood Beck	Total Inorganic N ($\mu\text{g l}^{-1}$)	0.08	0.78
	TFP ($\mu\text{g l}^{-1}$)	-	-

4.5 Species composition

Tables 4.6 and 4.7 present the results of diatom counts from cobbles and boulders at the two sites.

A total of 37 taxa were identified from the River Browney including 2 varieties of *Fragilaria capucina*. The maximum number of taxa found in a single count was 27 from the pooled boulder sample of 3 May. *Achnanthes minutissima* (mean percentage = 23%), *Amphora pediculus* (17%), *Navicula gregaria* (13%) and *Navicula lanceolata* (9.6%) all occur with over twice the mean percentage of other taxa. 10 species (29% of the taxa) occurred on 3 or less occasions with an abundance of 1% or less. *Gyrosigma acuminatum*, *Nitzschia linearis* and *Nitzschia liebetruithii* were only found by scanning the slide after the count.

The taxa exclusively present in the River Browney (i.e. not Harwood Beck) were:

Gomphonema parvulum, *Gyrosigma acuminatum*, *Melosira varians*, *Navicula capitata*, *Navicula tripunctata*, *Nitzschia dissipata*, *Nitzschia inconspicua*, *Nitzschia liebetruithii*, *Nitzschia linearis* and *Surirella brebissonii*.

Table 4.6. Percentage of epilithic diatom taxa in the River Browney.

C = Cobbles; B = Boulders; + = recorded when scanning the slide after counting; - = absent
 F = Frequency of occurrence; AV = Mean Percentage; SD = Standard Deviation

Diatom taxa	Date:	03-May		12-May		20-May		26-May		03-June	
	Percentage (%)	C	B	C	B	C	B	C	B	C	B
<i>Achnanthes lanceolata</i>		3.1	2.3	0.9	3.6	5	1.9	5.6	6	5.2	14.2
<i>Achnanthes minutissima</i>		15.4	11.2	14.3	16.8	28.1	17.1	11.1	35	43.4	40.9
<i>Amphora pediculus</i>		28.7	13.8	25.9	17.2	10	18.8	15.1	29	13.4	8.6
<i>Ceratoneis arcus</i>		-	-	-	-	0.6	1.3	-	-	0.4	-
<i>Cyclotella meneghiniana</i>		-	+	-	-	-	-	-	-	0.4	-
<i>Cocconeis pediculus</i>		-	-	-	0.4	-	-	-	-	-	-
<i>Cocconeis placentula</i>		0.5	3.2	1.8	0.8	15	2.6	7.1	3	2.8	2.5
<i>Cymbella affinis</i>		-	0.5	-	-	0.6	0.7	0.8	-	2.8	-
<i>Cymbella delicatula</i>		1	2.8	-	-	0.6	0.7	0.4	-	2.8	-
<i>Cymbella microcephala</i>		0.5	2.8	0.4	-	8.7	8.4	0.4	+	7.6	5.6
<i>Cymbella minuta</i>		1	0.5	0.9	+	1.8	0.7	1.2	0.5	+	0.5
<i>Reimeria sinuata</i>		-	-	2.1	-	-	1.3	3.2	-	4	2.5
<i>Diatoma moniliformis</i>		+	+	-	+	-	-	-	0.5	+	+
<i>Diatoma tenuis</i>		-	+	-	-	-	-	-	0.5	+	1
<i>Diatoma vulgare</i>		-	0.5	0.4	0.4	+	-	-	-	-	-
<i>Fragilaria capucina</i> var. <i>cap.</i>		1	0.9	+	+	-	-	0.4	+	-	2
<i>Fragilaria cap.</i> var. <i>vaucheriae</i>		-	1	-	-	0.6	-	-	-	1.2	0.5
<i>Gomphonema olivaceoides</i>		0.5	+	1.8	0.8	1.2	0.7	-	-	-	0.5
<i>Gomphonema olivaceum</i>		0.5	0.9	0.4	2.4	-	-	1.2	+	1.2	1
<i>Gomphonema parvulum</i>		+	+	+	+	1.2	-	0.8	-	+	-
<i>Gomphonema</i> spp.		-	2.3	0.4	-	1.2	-	-	-	0.4	-
<i>Gyrosigma acuminatum</i>		-	+	-	-	-	-	-	-	-	-
<i>Meridion circulare</i>		-	-	-	-	-	-	-	0.5	-	+
<i>Melosira varians</i>		-	0.5	-	-	-	-	-	-	-	-
<i>Navicula capitata</i>		-	-	-	-	-	-	-	-	-	-
<i>Navicula exigua</i>		-	-	-	-	-	-	-	-	-	-
<i>Navicula gregaria</i>		40.5	17	27.2	36	11.8	12.2	24.8	1.5	9.6	9.6
<i>Navicula lanceolata</i>		2.1	26.4	16.5	14.8	7.5	22.5	19.2	8	2.8	5.1
<i>Navicula tripunctata</i>		-	+	-	-	+	-	-	-	-	-
<i>Nitzschia dissipata</i>		0.5	0.9	+	0.8	1.2	0.7	-	-	-	0.5
<i>Nitzschia inconspicua</i>		-	-	3.1	0.4	-	1.3	0.8	2.5	0.4	-
<i>Nitzschia liebetruthii</i>		-	-	-	-	-	-	-	-	-	-
<i>Nitzschia linearis</i> var. <i>linearis</i>		-	-	+	+	-	-	-	-	-	-
<i>Nitzschia palea</i>		2.6	1.9	0.9	2	-	-	0.8	3.5	+	3.5
<i>Rhoicosphenia abbreviata</i>		+	10.1	1.7	2	4.3	9.1	6.7	9	1.6	1
<i>Surirella brebissonii</i>		2.1	0.5	1.3	1.6	0.6	-	0.4	0.5	-	0.5
<i>Synedra ulna</i>		-	-	-	-	-	-	-	-	-	-
NUMBER OF TAXA:		18	27	21	20	20	16	18	17	22	20

Table 4.6 continued.

	Date:		17-June		23-June		06-July		16-July		F	AV	SD
	Percentage (%)		C	B	C	B	C	B	C	B			
Diatom taxa													
<i>Achnanthes lanceolata</i>	1.6	1.5	9.3	0.5	5.8	5.6	6.1	4.1	18	4.6	3.3		
<i>Achnanthes minutissima</i>	33.6	27.7	11.5	41	8.4	8.7	24	21.2	18	23	12		
<i>Amphora pediculus</i>	21.2	32.6	6.2	34	9.1	11.6	3.9	8.9	18	17	9.4		
<i>Ceratoneis arcus</i>	-	0.5	-	-	-	-	0.6	-	5	0.7	0.4		
<i>Cyclotella meneghiniana</i>	0.8	+	8.7	2.5	0.5	0.5	-	1.2	9	1.6	2.8		
<i>Cocconeis pediculus</i>	1.2	0.5	3.1	2	2.1	+	-	1.2	8	1.3	1		
<i>Cocconeis placentula</i>	4.1	3.1	12.4	3.5	35.6	38.5	30.6	26	18	11	13		
<i>Cymbella affinis</i>	-	-	-	0.5	1.6	1	0.6	-	9	1	0.8		
<i>Cymbella delicatula</i>	-	0.5	-	1	-	-	8.3	1.2	10	1.9	2.4		
<i>Cymbella microcephala</i>	2.5	-	-	3.5	2.1	2	6.1	3.6	15	3.6	3		
<i>Cymbella minuta</i>	0.8	-	0.6	-	-	+	1.7	1.8	15	0.8	0.6		
<i>Reimeria sinuata</i>	8.3	4.6	13.7	-	1.5	3.4	0.6	-	11	4.1	3.8		
<i>Diatoma moniliformis</i>	+	-	-	0.5	1	-	0.6	-	10	0.3	0.4		
<i>Diatoma tenuis</i>	-	-	-	-	-	-	-	-	4	0.4	0.5		
<i>Diatoma vulgare</i>	-	-	1.8	-	-	0.5	-	0.6	7	0.6	0.6		
<i>Fragilaria capucina</i> var. <i>cap.</i>	-	1	-	-	+	-	0.6	-	10	0.6	0.6		
<i>Fragilaria cap.</i> var. <i>vaucheriae</i>	1.2	-	-	-	+	-	0.6	-	7	0.7	0.4		
<i>Gomphonema olivaceoides</i>	-	1.5	-	1	-	-	0.6	-	10	0.9	0.5		
<i>Gomphonema olivaceum</i>	0.8	8.7	1.2	2	2	1	2.8	1.2	16	1.7	2		
<i>Gomphonema parvulum</i>	-	-	0.6	1	-	-	-	0.6	10	0.4	0.5		
<i>Gomphonema</i> spp.	2.1	0.5	9.3	-	-	-	-	-	7	2.3	3.2		
<i>Gyrosigma acuminatum</i>	-	-	-	-	-	+	-	-	2				
<i>Meridion circulare</i>	-	-	-	-	-	-	-	-	2	0.3	0.3		
<i>Melosira varians</i>	-	-	-	-	-	-	-	0.6	2	0.9	0.5		
<i>Navicula capitata</i>	-	-	-	0.5	-	-	-	-	1				
<i>Navicula exigua</i>	0.4	-	-	-	-	-	-	-	1				
<i>Navicula gregaria</i>	3.8	6.6	1.2	1	7.9	8.7	3.9	8.9	18	13	12		
<i>Navicula lanceolata</i>	3.8	5.6	9.9	0.5	12.5	6.2	2.8	6.5	18	9.6	7.5		
<i>Navicula tripunctata</i>	-	-	0.6	-	-	+	-	-	4	0.2	0.3		
<i>Nitzschia dissipata</i>	-	+	0.6	-	0.5	-	-	-	10	0.6	0.4		
<i>Nitzschia inconspicua</i>	-	-	-	-	-	-	-	-	6	1.4	1.1		
<i>Nitzschia liebethuthii</i>	-	-	-	-	0.5	-	-	-	1				
<i>Nitzschia linearis</i> var. <i>linearis</i>	-	-	-	-	-	-	-	-	2				
<i>Nitzschia palea</i>	2.5	2.5	0.6	3	1.6	1.5	1.7	4.1	16	2	1.1		
<i>Rhoicosphenia abbreviata</i>	11.3	2.6	7.5	2	6.3	10.3	3.9	7.1	18	5.4	3.7		
<i>Surirella brebissonii</i>	+	-	1.2	-	1	0.5	+	0.6	14	0.8	0.6		
<i>Synedra ulna</i>	+	-	-	-	-	-	-	-	1				
NUMBER OF TAXA:	20	18	19	18	20	19	20	18					

A total of 36 taxa was identified from Harwood Beck over 3 months including 3 varieties of *Fragilaria capucina* along with several small *Navicula* spp. (one taxon) which could not all be identified to species with confidence. The maximum number of taxa identified from any one slide at Harwood Beck was 21 (17 June, boulders). The flora is characterised by the dominance of *Achnanthes minutissima* with *Navicula* spp. being sub-dominant. *Ceratoneis arcus*, *Cymbella delicatula*, *Cymbella microcephala* and *Gomphonema olivaceum* were also present on all sampling occasions. 16 species (47% of the taxa) were found on 3 or less occasions (Table 4.7).

Taxa exclusive to Harwood Beck (i.e not found in the River Browney) were: *Fragilaria crotonensis*, *Fragilaria capucina* var. *gracilis*, *Fragilaria famelica*, *Gomphonema acuminatum*, *Navicula exilis* and *Navicula submolesta*.

Table 4.7. Percentage of epilithic diatom taxa in Harwood Beck.

C = Cobbles; B = Boulders; + = recorded when scanning the slide after counting; - = absent
F = Frequency of occurrence; AV = Mean Percentage; SD = Standard Deviation

Diatom taxa	Date:		03-May		20-May		03-June		17-June	
	Percentage (%)		C	B	C	B	C	B	C	B
<i>Achnanthes lanceolata</i>	-	-	1	0.4	0.6	1.9	+	2.3		
<i>Achnanthes minutissima</i>	57.8	47.7	63.7	78.1	81.5	77.1	62.8	60.2		
<i>Amphora pediculus</i>	-	-	1.5	0.4	-	-	-	-		
<i>Ceratoneis arcus</i>	1.2	0.6	0.5	2.7	0.6	0.4	0.4	0.5		
<i>Cyclotella meneghiniana</i>	-	-	0.5	-	-	-	-	-		
<i>Cocconeis pediculus</i>	-	-	-	-	-	-	-	-		
<i>Cocconeis placentula</i>	-	-	0.5	0.4	+	-	-	0.5		
<i>Cymbella affinis</i>	0.3	1.2	-	2.7	0.3	1.5	2.1	3.7		
<i>Cymbella delicatula</i>	1.2	9.3	6.9	3.2	3.2	3.8	7.1	18.7		
<i>Cymbella microcephala</i>	4.8	1.8	2	2.3	1.8	8.8	2.3	1.4		
<i>Cymbella minuta</i>	0.3	0.6	-	-	0.3	0.4	+	0.9		
<i>Cymbella silesiaca</i>	-	-	1	-	-	+	-	0.5		
<i>Reimeria sinuata</i>	-	0.6	-	-	0.6	-	-	0.9		
<i>Diatoma moniliformis</i>	0.3	0.3	-	-	0.3	0.4	-	+		
<i>Diatoma tenuis</i>	0.3	-	-	-	0.3	-	0.4	-		
<i>Diatoma vulgare</i>	-	-	+	-	-	-	-	+		
<i>Fragilaria crotonensis</i>	+	-	-	+	-	-	-	-		
<i>Fragilaria capucina</i> var. <i>cap.</i>	-	0.6	-	-	0.6	0.8	0.4	0.5		
<i>Fragilaria cap.</i> var. <i>gracilis</i>	0.3	-	-	-	-	-	-	-		
<i>Fragilaria cap.</i> var. <i>vaucheriae</i>	0.6	-	-	-	-	-	-	-		
<i>Fragilaria famelica</i>	-	-	-	-	-	-	-	-		
<i>Gomphonema acuminatum</i>	-	-	-	-	-	-	-	-		
<i>Gomphonema olivaceoides</i>	1.2	0.9	-	-	0.3	0.8	1.1	1.7		
<i>Gomphonema olivaceum</i>	0.3	3.2	1	1.3	1.6	2.3	0.4	1.7		
<i>Gomphonema</i> spp.	-	-	5	-	0.3	+	-	0.9		
<i>Meridion circulare</i>	-	-	-	-	+	-	-	-		
<i>Navicula exilis</i>	0.6	-	-	-	-	-	-	-		
<i>Navicula gregaria</i>	-	-	1.5	0.4	-	0.4	-	+		
<i>Navicula accomoda</i>	-	-	-	0.4	-	-	6	2.3		
<i>Navicula lanceolata</i>	-	-	1.5	-	-	-	-	-		
<i>Navicula submolesta</i>	18	17.4	-	-	-	-	13.1	4.6		
<i>Navicula</i> spp.	12.8	15.8	13.9	8.1	8.3	3.3	3.5	0.5		
<i>Nitzschia palea</i>	-	-	-	-	+	-	-	-		
<i>Nitzschia</i> spp.	-	-	0.5	-	-	-	-	-		
<i>Rhoicosphenia abbreviata</i>	-	-	-	-	-	-	-	-		
<i>Synedra ulna</i>	-	-	-	-	+	-	0.4	0.5		

NUMBER OF TAXA: 16 13 16 13 19 15 15 21

Table 4.7 continued.

Date:	23-June		06-July		16-July		F	AV	SD
	Percentage (%)		C	B	C	B			
Diatom taxa									
	-	-	-	-	5.6	0.4	8	1.5	1.8
<i>Achnanthes lanceolata</i>	-	-	-	-	-	-	2	0.9	0.8
<i>Achnanthes minutissima</i>	84	82.7	58.5	60.7	59.7	61.7	14	67	11.4
<i>Amphora pediculus</i>	-	-	-	-	-	-	1	0.5	
<i>Ceratoneis arcus</i>	1	0.4	0.8	0.6	2.6	1.8	14	1.0	0.8
<i>Cyclotella meneghiniana</i>	-	-	-	-	-	-	1	0.0	
<i>Cocconeis pediculus</i>	-	-	-	+	-	-	1	0.0	
<i>Cocconeis placentula</i>	0.5	-	0.8	-	2	0.9	8	0.7	0.6
<i>Cymbella affinis</i>	1.9	1.1	4.5	2.8	5.6	5.3	13	2.5	1.8
<i>Cymbella delicatula</i>	2.9	1.9	6.2	5	6.6	3.1	14	5.7	4.4
<i>Cymbella microcephala</i>	1.9	1.9	9	10	7.2	5.9	14	4.4	3.2
<i>Cymbella minuta</i>	0.5	-	0.8	-	-	0.4	9	0.5	0.3
<i>Cymbella silesiaca</i>	-	-	-	-	-	-	3	0.5	0.5
<i>Reimeria sinuata</i>	+	-	-	-	1	0.4	6	0.6	0.4
<i>Diatoma moniliformis</i>	1	-	0.4	0.6	-	-	8	0.4	0.3
<i>Diatoma tenuis</i>	-	-	-	-	1.5	0.4	5	0.6	0.5
<i>Diatoma vulgare</i>	-	-	-	-	-	-	2		
<i>Fragilaria crotonensis</i>	+	+	0.4	0.6	-	-	6	0.2	0.2
<i>Fragilaria capucina</i> var. <i>cap.</i>	-	-	2.1	2.8	2.6	5.3	9	1.7	1.6
<i>Fragilaria cap.</i> var. <i>gracilis</i>	-	-	-	-	-	-	1	0.3	
<i>Fragilaria cap.</i> var. <i>vaucheriae</i>	0.5	0.4	-	0.6	-	+	5	0.4	0.2
<i>Fragilaria famelica</i>	-	0.8	-	-	-	-	1	0.8	
<i>Gomphonema acuminatum</i>	-	-	-	0.6	-	-	1	0.6	
<i>Gomphonema olivaceoides</i>	-	0.4	-	-	0.5	1.8	9	1.0	0.5
<i>Gomphonema olivaceum</i>	1.4	0.4	4.1	2.8	3.6	1.8	14	1.9	1.2
<i>Gomphonema</i> spp.	-	-	-	-	-	-	4	1.6	2.3
<i>Meridion circulare</i>	-	-	-	-	-	0.4	2	0.2	
<i>Navicula exilis</i>	-	-	-	-	-	-	1	0.6	
<i>Navicula gregaria</i>	-	-	-	-	-	-	4	0.6	0.6
<i>Navicula accomoda</i>	-	-	-	-	-	-			
<i>Navicula lanceolata</i>	-	-	+	-	+	0.4	4	0.5	0.7
<i>Navicula submolesta</i>	-	-	-	-	-	-			
<i>Navicula</i> spp.	4.4	10	12	12.9	6.1	10.4			
<i>Nitzschia palea</i>	-	-	-	-	-	-	1	0.0	
<i>Nitzschia</i> spp.	-	-	-	-	-	-	1	0.5	
<i>Rhoicosphenia abbreviata</i>	-	-	-	-	1	-	1	1	
<i>Synedra ulna</i>	-	-	0.4	0.6	1	+	7	0.4	0.3
NUMBER OF TAXA:	13	11	14	14	16	18			

Paired t - tests were applied to taxa percentage data of the River Browney and Harwood Beck for taxa present on all sampling occasions. There were no statistically significant differences between the observed percentages of these taxa on cobbles or boulders (Tables 4.8 and 4.9).

Table 4.8. Paired t-Tests between cobble and boulder percentages

Taxon	t value (calculated)	t Critical two-tail
<i>Achnanthes minutissima</i>	- 0.72	2.31
<i>Amphora pediculus</i>	- 1.06	2.31
<i>Cocconeis placentula</i>	1.74	2.31
<i>Navicula gregaria</i>	0.83	2.31
<i>Navicula lanceolata</i>	- 0.54	2.31

4.5.1 Species changes in the River Browney

For the 5 most common taxa, mean cobble and boulder percentages were plotted against the mean daily flow rate over the sampling period (Figs. 4.8 - 4.12).

The percentage of *Achnanthes minutissima* increased after the storm (Fig. 4.8) suggesting that it is more firmly attached to stones than other taxa and /or it is a colonising species. A slight decline in the percentage of *Amphora pediculus* (Fig. 4.9) was observed following the storm, but no overall relationship is evident. The percentage of *Cocconeis placentula* also increases after the storm with a massive increase in percentage during July (Fig. 4.10). By contrast, the percentage of *Navicula gregaria* and *Navicula lanceolata* decline following the storm (Figs 4.11 and 4.12), although the percentage of *N. lanceolata* remains stable before and after stormflow.

Figure 4.8. Percentage change of *Achnanthes minutissima* in the River Browney (vertical bars) plotted against mean flow rate (shaded area).

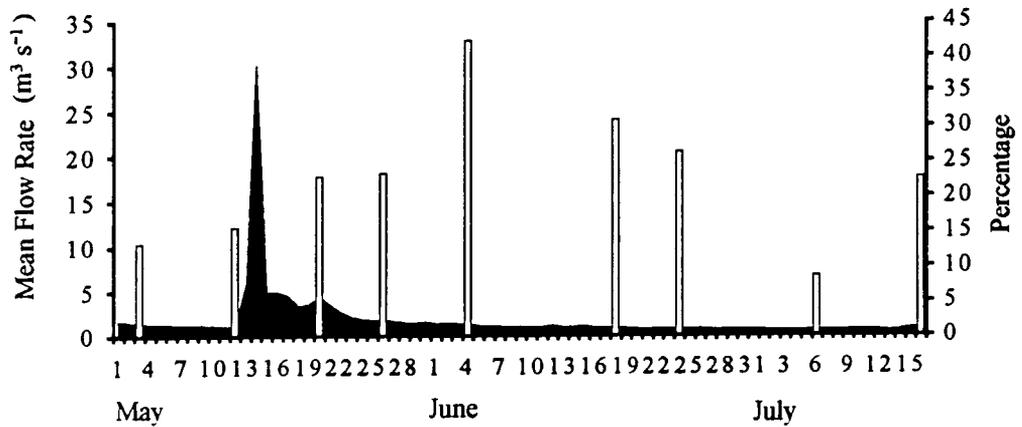


Figure 4.9. Percentage change of *Amphora pediculus* against River Browney mean flow rate.

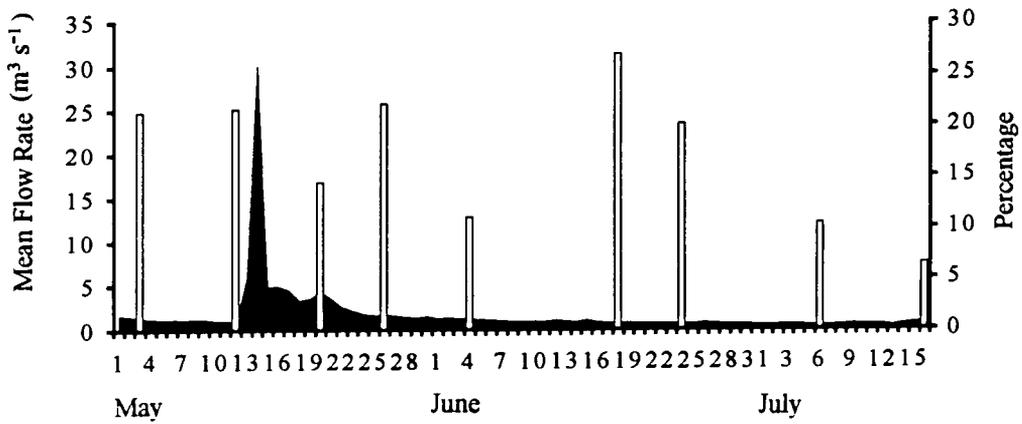


Figure 4.10. Percentage change of *Cocconeis placentula* against River Browney mean flow rate.

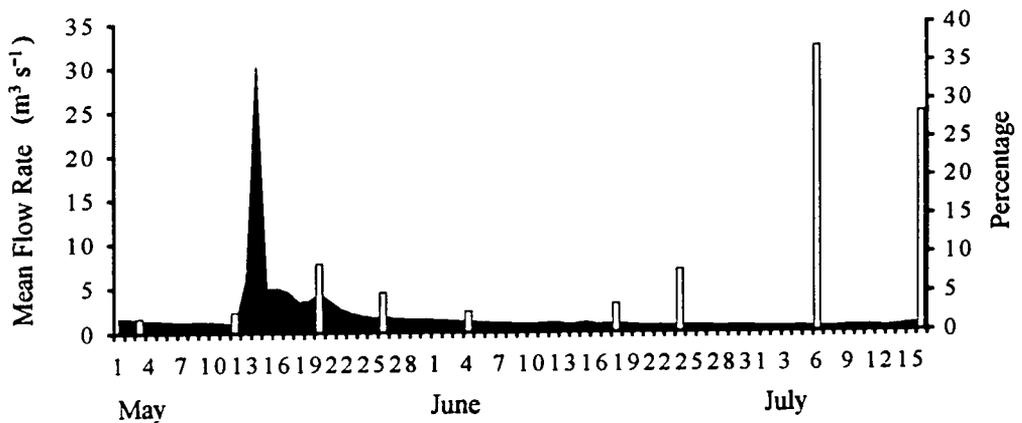


Figure 4.11. Percentage change of *Navicula gregaria* against River Browney mean flow.

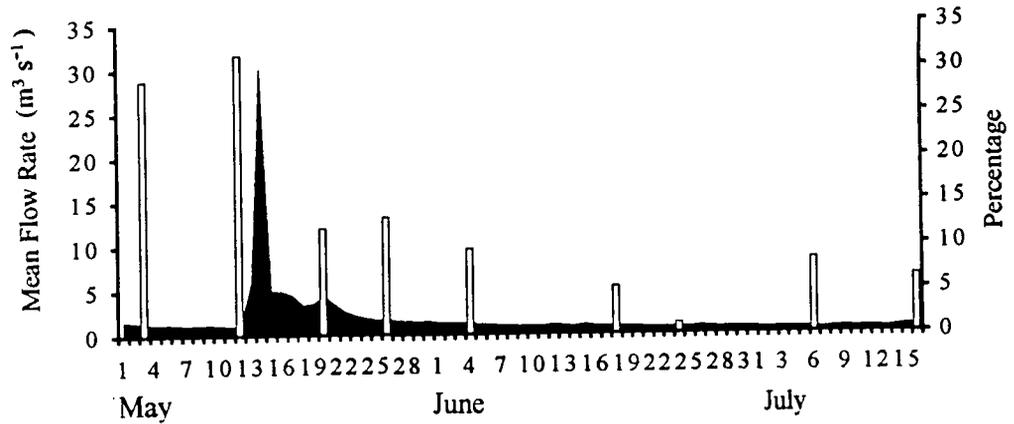
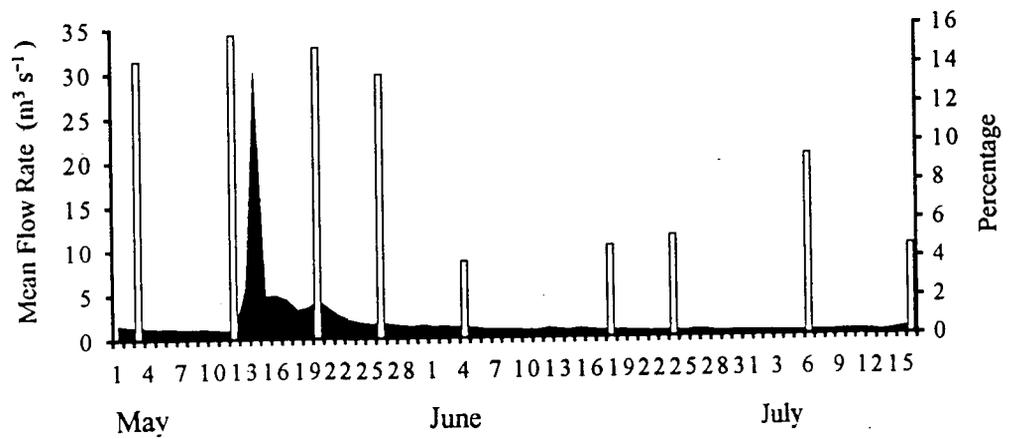


Figure 4.12. Percentage change of *Navicula lanceolata* against River Browney mean flow rate.



The large decline in the percentage of *Navicula gregaria* and *N. lanceolata* during June and July appears to be related to an increase in Total Inorganic Nitrogen. Figures 4.13 & 4.14 illustrate the close relationship between Total Inorganic Nitrogen and the percentage of *N. gregaria* ($r = 0.937$, $p < 0.001$) and *N. lanceolata* ($r = 0.9$, $p < 0.001$).

Figure 4.13. Correlation between the percentage of *Navicula gregaria* in the River Browney and Total Inorganic Nitrogen.

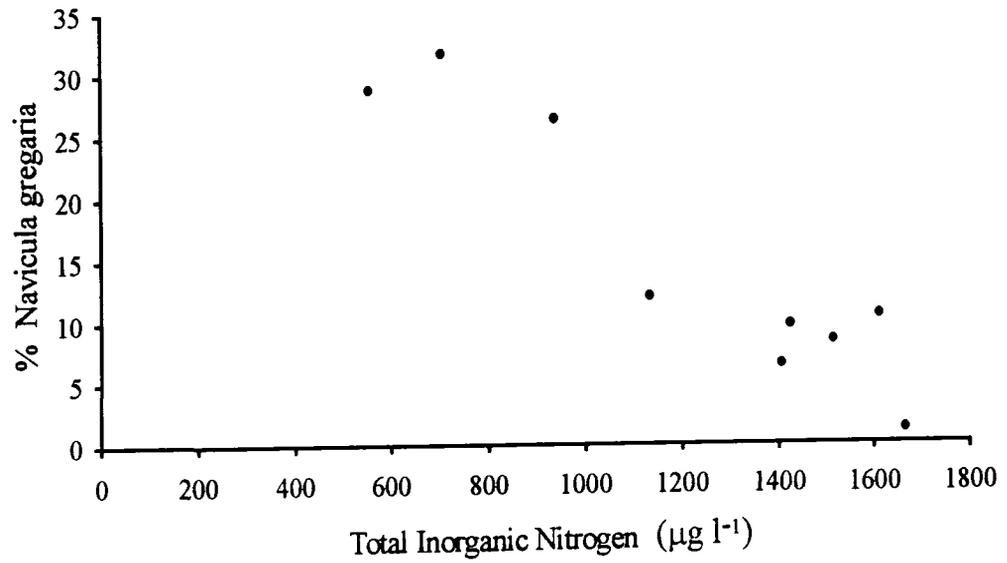
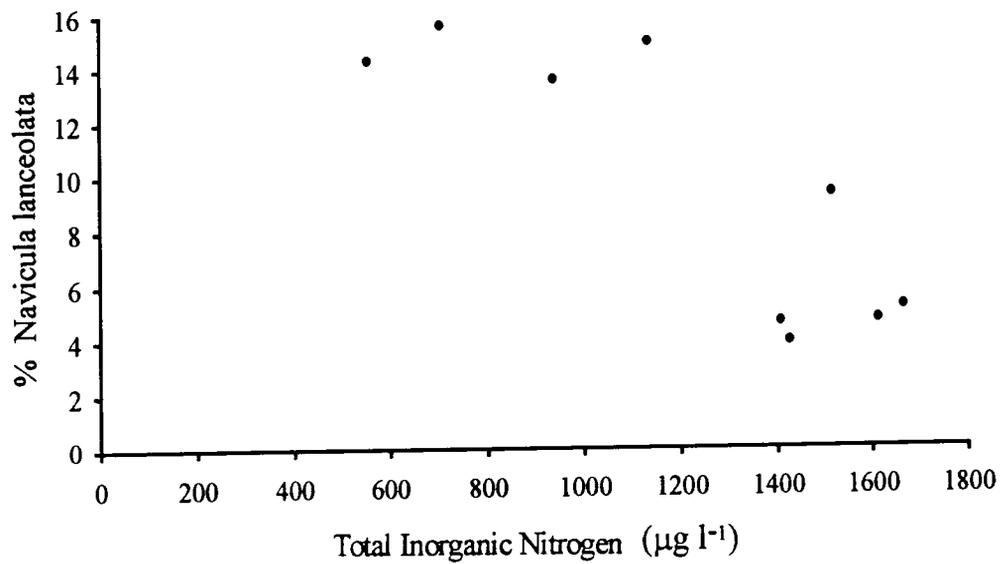


Figure 4.14. Correlation between the percentage of *Navicula lanceolata* in the River Browney and Total Inorganic Nitrogen.

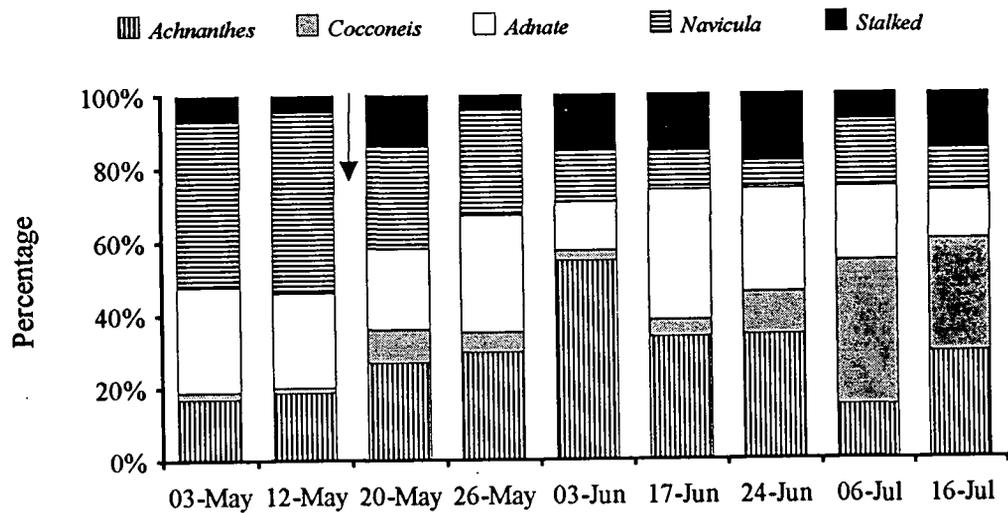


For other taxa Total Inorganic Nitrogen and percentage were poorly correlated.

4.5.2 Change in Diatom Guilds of the River Browney

Analysis of the diatom community guild structure indicated that the strongly adhered *Cocconeis* and *Achnanthes* taxa and stalked taxa increased in percentage after the storm, whereas the motile *Navicula* species decreased in percentage. A slight decrease in the percentage of adnate taxa (i.e. *Rhoicosphenia*, *Amphora* and *Surirella*) was observed.

Figure 4.15. Mean percentage of River Browney diatoms classified by guild. ↓ = storm.



4.5.3 Species changes in Harwood Beck

For the five most common taxa mean percentage on each sample date was plotted against mean daily flow rate data over the sampling period (Figs. 4.16 to 4.20). No significant differences were observed between percentages of these taxa on boulders or cobbles.

Table 4.9. Paired t-Tests between cobble and boulder diatom percentages

Taxon	t value (calc.)	t Critical two-tail
<i>Achnanthes minutissima</i>	-0.01	2.45
<i>Cymbella delicatula</i>	-0.69	2.45
<i>Cymbella microcephala</i>	-0.37	2.45
<i>Cymbella</i> spp.	0.36	2.45
<i>Navicula</i> spp	0.74	2.45

The percentage of *Achnanthes minutissima* shows a tendency to increase after stormflow events (Fig. 4.16), whereas there was a marked decrease in percentage of *Navicula* spp. after storm events (Fig. 4.20). No effect of flow rate on the percentage of *Cymbella microcephala* (Fig. 4.18) or *Cymbella delicatula* (Fig. 4.17) was evident, but the percentage of *C. microcephala* increases substantially in July. The overall percentage of *Cymbella* spp. remained stable during stormflow periods (Fig. 4.19).

Figure 4.16. Percentage change of *Achnanthes minutissima* with mean flow rate in Harwood Beck.

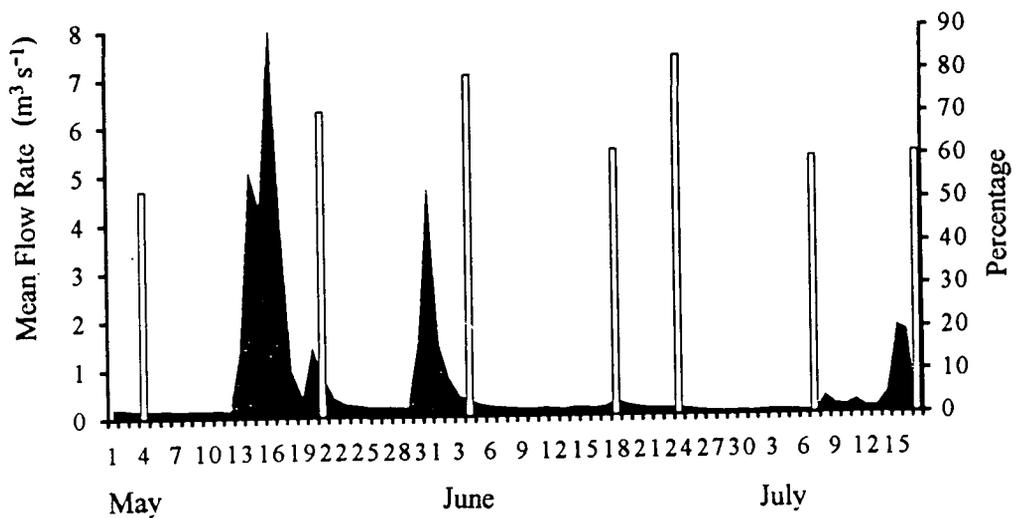


Figure 4.17. Percentage change of *Cymbella delicatula* with mean flow rate in Harwood Beck.

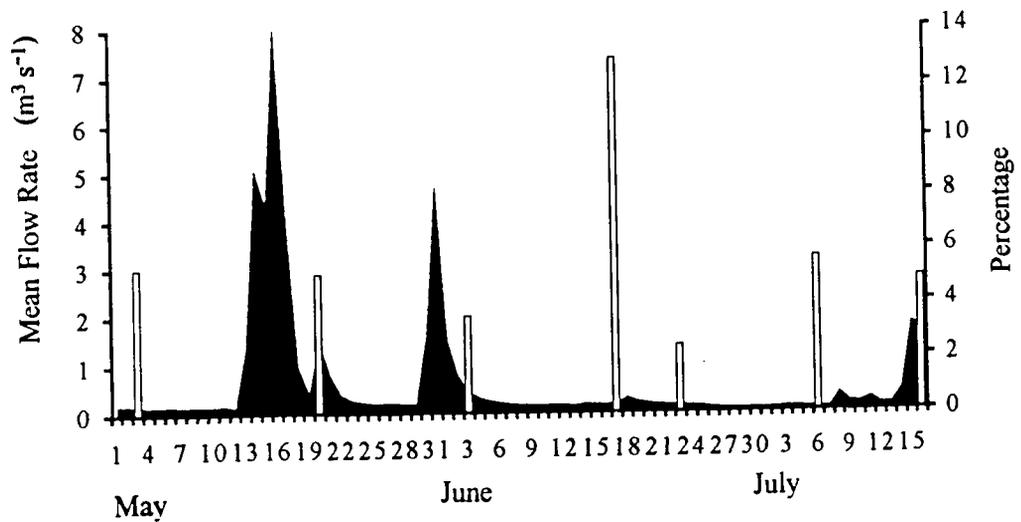


Figure 4.18. Percentage change of *Cymbella microcephala* with mean flow rate in Harwood Beck.

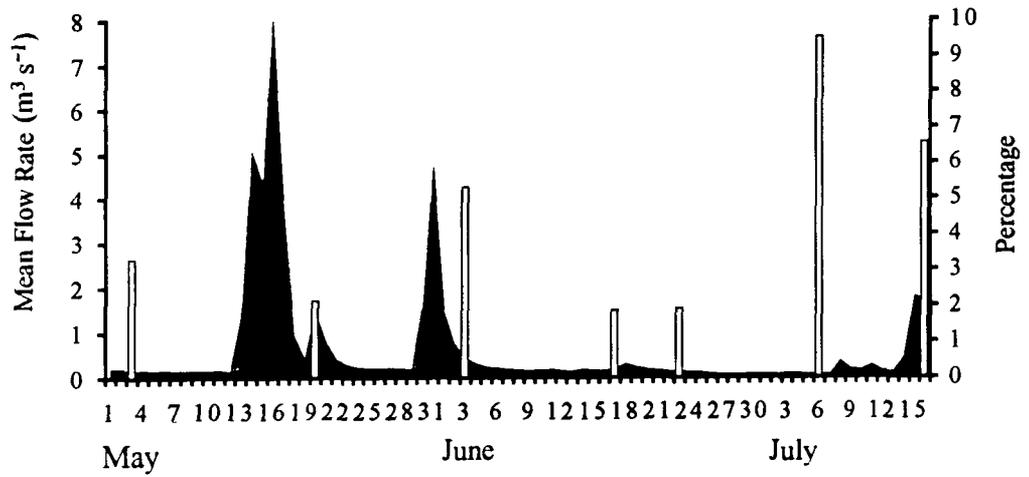


Figure 4.19. Percentage change of *Cymbella* spp. with mean flow rate in Harwood Beck.

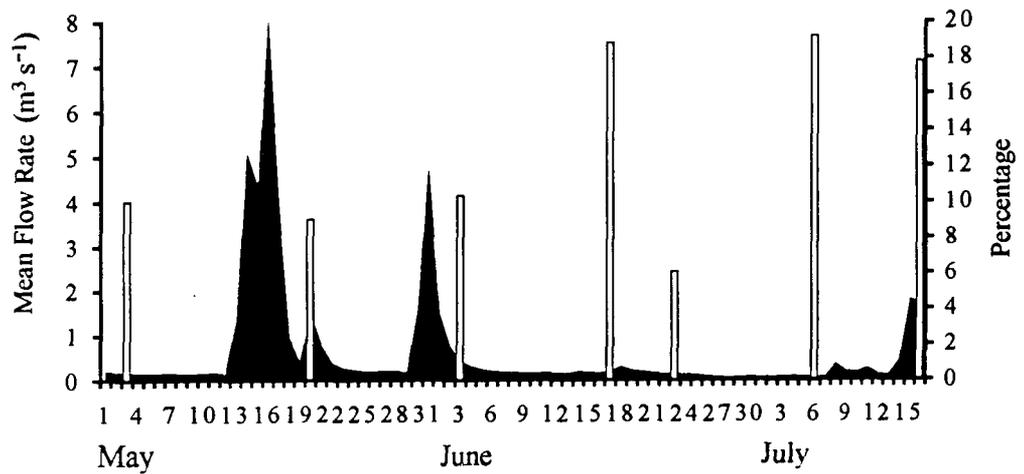
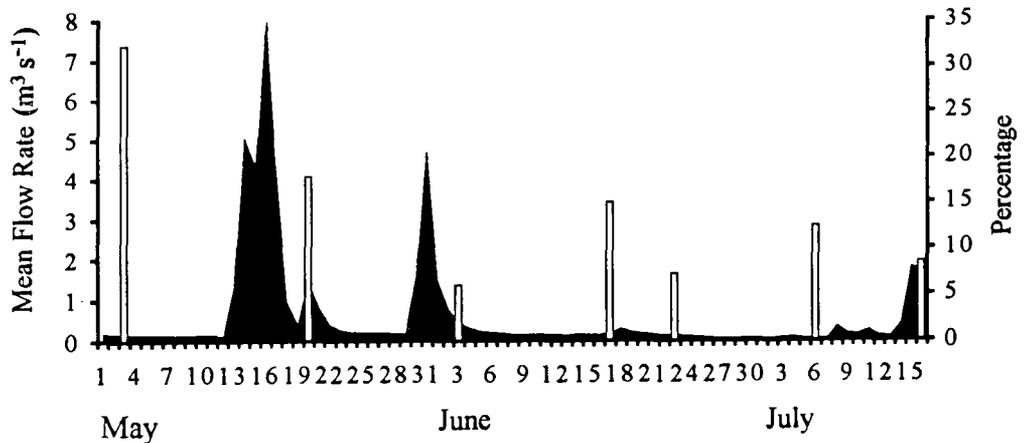


Figure 4.20. Percentage change of *Navicula* spp. with mean flow rate in Harwood Beck.



4.5.4 Changes in Diatom Guilds of Harwood Beck

There were differences in the communities on cobbles and boulders in Harwood Beck, which may be related to flow (Figs. 4.21 and 4.22). After the major storm event of mid-May, the percentage of stalked taxa (i.e. *Gomphonema* and *Cymbella*) increased on cobbles and decreased on boulders. On 20 May erect taxa (without stalks) such as *Fragilaria*, *Diatoma*, *Synedra* and *Meridion* were virtually eliminated on both cobbles and boulders. However, on all other sampling occasions some of these taxa were present. The percentage of *Navicula* spp. is also reduced following high flow events. Although the percentage of *Achnanthes* is seen to increase following the storm (particularly on boulders) it also shows a substantial increase on 23 June following a sustained low flow period. This may be related to the very low nutrient conditions at this time (Table 4.2). Total Inorganic Nitrogen is minimum and P is below detection limit ($< 5 \mu\text{g l}^{-1}$).

Figure 4.21. Percentage of Harwood Beck diatoms on cobbles classified by guild.

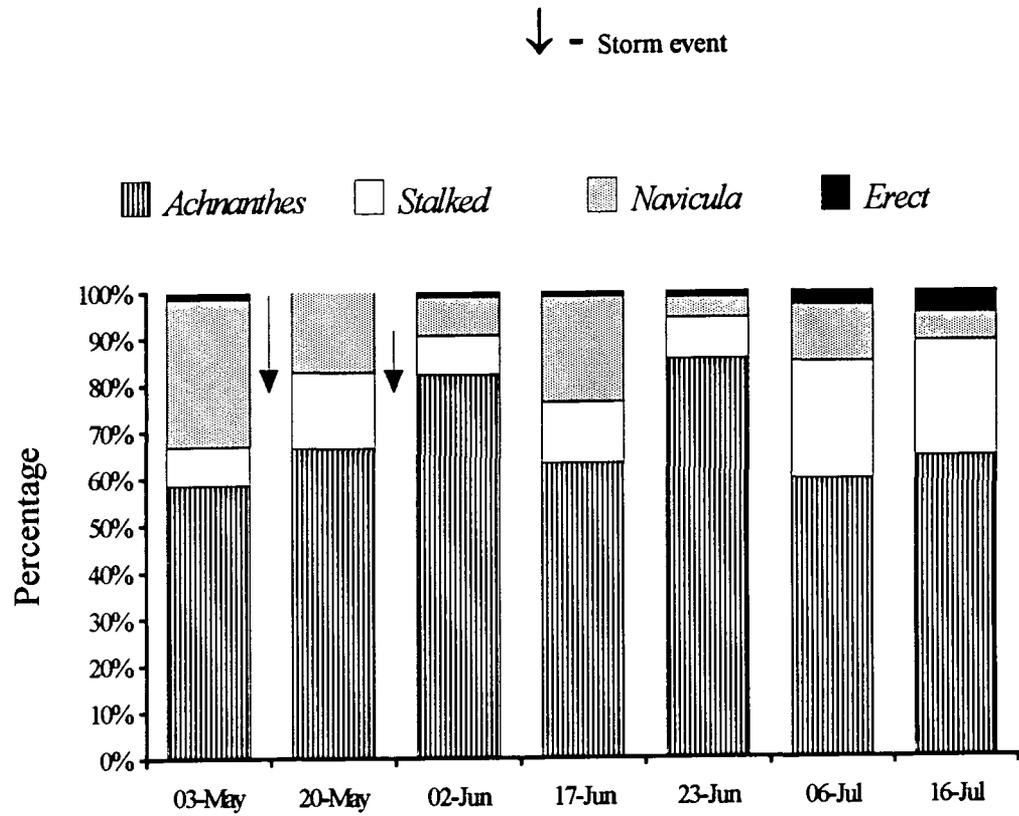
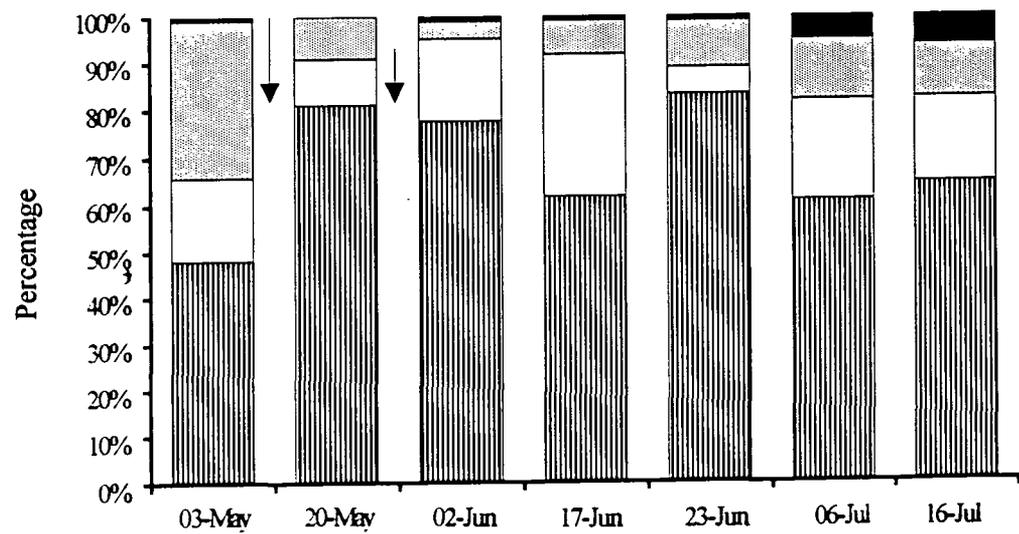


Figure 4.22. Percentage of Harwood Beck diatoms on boulders classified by guild.



4.6 Diatom Pollution Indices

4.6.1 Coste SPI and GDI for the River Browney

SPI remains relatively stable over time (compared with GDI) with a mean water quality of 3.9 (GDI = 3.7), as shown by the arrows (Figs. 4.23 and 4.24). There is close agreement between SPI and GDI on cobbles ($r = 0.93$), but not on boulders ($r = 0.44$). The variation in SPI was significantly correlated with the percentage of *Achnanthes minutissima* (cobbles, $r = 0.87$; boulders, $r = 0.71$) which is highly sensitive to pollution ($S = 5$) but of poor indicator value ($V = 1$). *A. minutissima* was not correlated with the GDI. Both SPI and GDI remain more stable on boulders and GDI is more variable on both cobbles and boulders (Table 4.10).

Table 4.10. Standard deviations of SPI and GDI on cobbles and boulders in the River Browney.

	SPI Cobbles	SPI Boulders	GDI Cobbles	GDI Boulders
Standard deviation	0.25	0.19	0.35	0.24

Figure 4.23. Species Pollution Index (SPI) in the River Browney over study period.

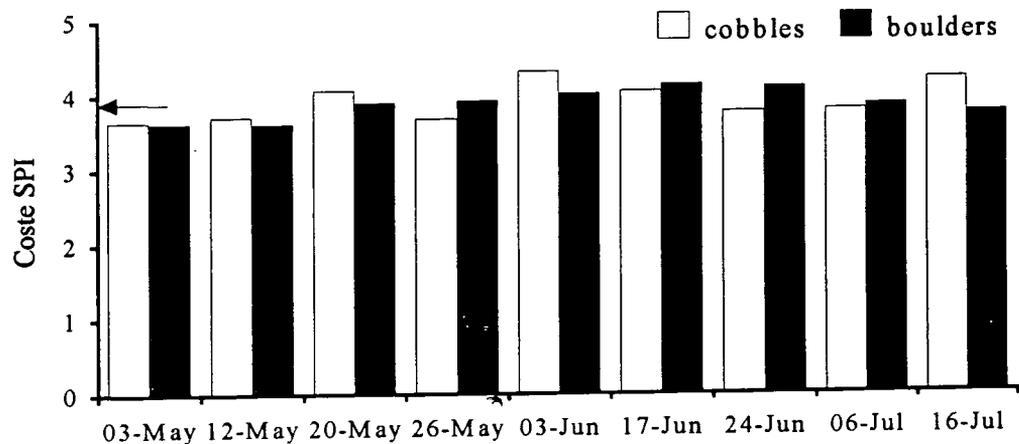
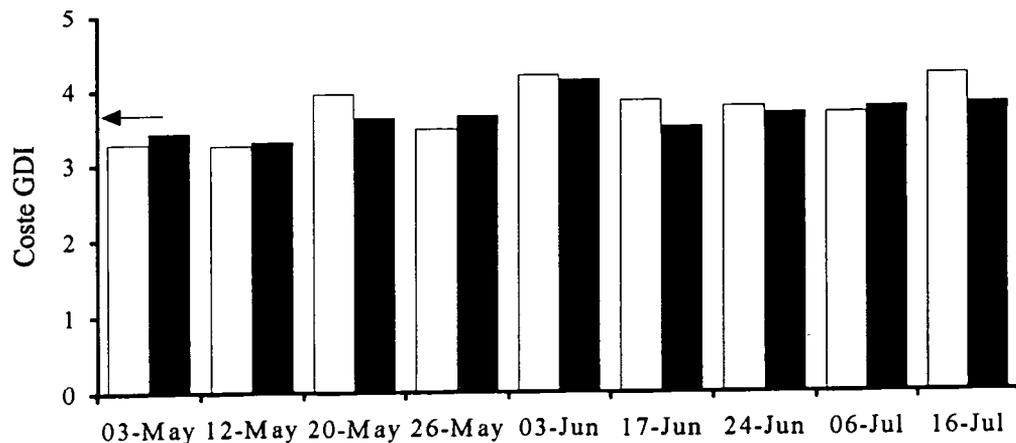


Figure 4.24. Generic Diatom Index (GDI) in the River Browney over study period.



4.6.2 Coste SPI and GDI for Harwood Beck

SPI values are more variable at Harwood Beck than in River Browney, but are still more stable on boulders (Table 4.11). The GDI values for Harwood Beck are less variable than SPI values (Table 4.11). As for the River Browney, SPI values of Harwood Beck are correlated with the percentage of *Achnanthes minutissima* ($r = 0.62$, $p > 0.1$ and $r = 0.89$, $p < 0.01$ for cobbles and boulders, respectively). GDI values are also well correlated with *A. minutissima* ($r = 0.76$, $p < 0.05$, $r = 0.88$, $p < 0.01$ for cobbles and boulders respectively). SPI and GDI values are significantly correlated ($r = 0.8$, $p < 0.05$ for cobbles; $r = 0.96$, $p < 0.001$ for boulders).

Table 4.11. Standard deviations of SPI and GDI on cobbles and boulders in Harwood Beck.

	SPI cobbles	SPI boulders	GDI cobbles	GDI boulders
Standard deviation	0.42	0.28	0.19	0.21

Fig. 4.25. Species Pollution Index (SPI) in Harwood Beck over study period.

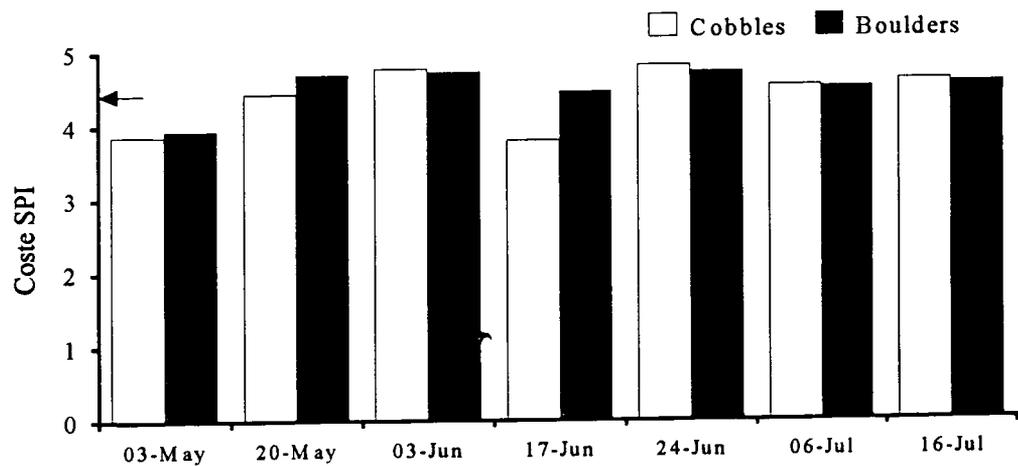
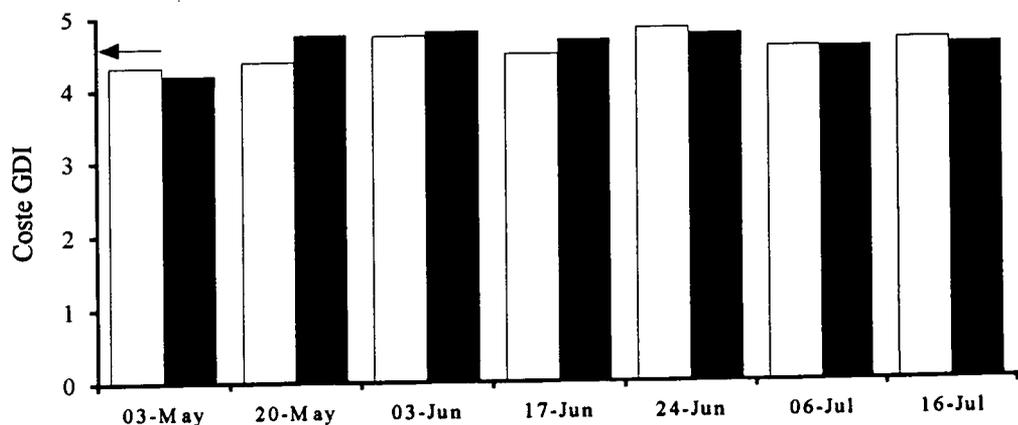


Figure 4.26. Generic Diatom Index (GDI) in Harwood Beck over study period.



4.7 Round (1993)'s zonation system for running water sites

4.7.1 Classification of River Browney

The River Browney diatom community is characteristic of Rounds' zone 3 ('nutrient rich') classification. All of the sub-zonal dominant diatom taxa occurred as dominant in the River Browney on one or more occasions, with the exception of *Reimeria sinuata* which occurred at a maximum of 13.7 % (cobbles) on 23 June. There appears to be a succession of dominant species, possibly related to the influence of flow on nutrient concentrations which tended to increase during the summer. *Amphora pediculus* was dominant before the mid-May storm event and *Achnanthes minutissima* was dominant later in May and on all 3 sampling dates in June. *Cocconeis placentula* was dominant in July.

4.7.2 Classification of Harwood Beck

On the basis of the dominant diatom taxa used in Rounds' zone classification system Harwood Beck can be considered as a zone 2 site (i.e. 'nutrient richer and somewhat higher pH'), because *Achnanthes minutissima* was clearly dominant on all sampling occasions.

CHAPTER 5

DISCUSSION

5.1 Introduction

It is clear that stormflow affects epilithic biomass and diatom community composition. A sharp decline in chlorophyll *a* followed heavy stormflow in the River Browney, but in Harwood Beck lighter stormflow and consequent higher nutrient availability may have encouraged biomass accrual on boulders. At both sites chlorophyll *a* was over twice as high on boulders as on cobbles, following stormflow. However, the difference between cobble and boulder chlorophyll *a* over the entire study period was only significant in Harwood Beck. Despite high temporal variability in chlorophyll *a* and taxon percentages, diatom-based water quality indices remained stable.

5.2 Variation in chlorophyll *a* / biomass

Fluctuations in algal populations may be caused by various factors and simple correlations are difficult to interpret causally (Marker and Willoughby, 1988). The decline in chlorophyll *a* observed in June (Figs 4.6 and 4.7) may be due to interactions between a number of factors including: current speed, flow rate, light intensity, herbivory, available nutrients and the growth of algae other than diatoms.

Chlorophyll *a* on River Browney cobbles showed a significant inverse correlation to current velocity (Table 4.5). This is in agreement with other studies based on both field measurements (Anyam, 1990) and laboratory experiments (Antoine and Benson-Evans, 1982). However, in the natural stream situation this relationship is complicated by the interactions of detritous accumulation, nutrient availability, and current-dependant grazing of herbivorous insects. For example, Horner and Welch (1981) showed a relationship between current speed, orthophosphate concentration and biomass measurement, as chlorophyll *a*, in a stream. In the River Browney FTP and FRP concentrations were inversely correlated with flow rate (Fig. 4.3), but the correlation with chlorophyll *a* was negative and insignificant. The correlation may be due to dilution of nutrient inputs from agricultural runoff and the upstream sewage output at higher flows.

Marker (1976) found that hard-water, nutrient rich streams ($\text{pH} \cong 8$), supported a higher biomass than softwater, near neutral streams. Although the River Browney has harder water and higher nutrient concentrations than Harwood Beck this relationship is complicated by the differences in hydrological regime. Chlorophyll a was substantially higher in the River Browney than in Harwood Beck, before stormflow in mid-May (Figs 4.6 and 4.7). However, mean peak stormflow was approximately four times higher in the River Browney than in Harwood Beck (Figs 4.1 and 4.2). Subsequent chlorophyll a levels in Harwood Beck were higher than or comparable to chlorophyll a levels in the River Browney. From 23 June, in a period of sustained low flow, the very low chlorophyll a (below $0.5 \mu\text{g cm}^{-2}$) in both streams (Figs 4.6 and 4.7) suggests that other factors were preventing biomass recovery.

Although there was no significant difference in chlorophyll a between cobbles and boulders over the entire study period, Figs 4.6 and 4.7 show chlorophyll a to be substantially higher on boulders shortly after stormflow, suggesting that boulders provided a more stable substrate. The stormflow in Harwood Beck between 13 and 19 May was probably only heavy enough to cause disturbance of cobbles, because boulder chlorophyll a was at a maximum on 20 May. If cobbles remained stationary a similar increase in chlorophyll a on cobbles might have been expected. The maximum difference between cobble and boulder chlorophyll a also occurred on 20 May, and the second largest difference occurred on 3 June, immediately after another slightly smaller flow event.

Maximum boulder chlorophyll a on 20 May could be partly attributable to the higher nutrient conditions on this date. However, Horner *et al.* (1990) found that constant current velocities up to 0.6 m s^{-1} enhanced biomass accrual on stationary substrata, with further increases resulting in substantial biomass reduction. Harwood Beck current speed on the 20 May was measured as 0.91 m s^{-1} at a third of stream depth at the point of apparently fastest current. However, the epilithon was deeper and sampled from various positions in the central stream area where current speed may have been as low as 0.6 m s^{-1} . High variability in chlorophyll a may be partially associated with spatial replication of cobble and boulder samples. Horner *et al.* (1990) also found that sudden increases in velocity increased instantaneous loss rates by an order of magnitude or more, although marked biomass reductions were not apparent a day after velocity change. Therefore, recording current velocity gradient prior to sample collection could provide useful data. River Browney current velocity

on 20 May was the same (0.90 m s^{-1}), but a greater preceding current velocity gradient was probably responsible for the marked chlorophyll *a* loss.

The River Browney was subject to considerable bankside shading, whereas Harwood Beck was highly exposed. The collapse in epilithic diatom biomass occurred two to three weeks earlier in the River Browney (Figs 4.6 and 4.7), so it is possible light intensity may have been a significant factor limiting the recovery of River Browney chlorophyll *a* in early June. However, in a study chlorophyll *a* was influenced to a much greater degree by grazing (of the snail *Elimia clavaeformis*) rather than irradiance. Highest levels of areal specific primary productivity occurred in 'high light / low grazing' treatments. Biomass levels were similar between 'high light / high grazing' and 'low light / high grazing' treatments. Once grazing pressure exceeded a certain threshold, biomass levels remained low irrespective of plentiful abiotic resources (Steinman, 1992). Marker and Casey (1982) showed that maximum chironomid larvae population density coincided with the collapse of the diatom population during the first two weeks in June.

The slightly alkaline and relatively clean waters of Harwood Beck and the River Browney may support rich invertebrate populations. Mulholland *et al.* (1986) showed marked increases in algal biomass as stream pH fell from 7 to 4 (particularly at lower pH values). This may be due to the lower numbers of grazing invertebrates in acid waters. Winterbourne *et al.* (1992) found that invertebrates thought to feed mainly by grazing and scraping algae from stones, including many mayflies and snails, are notably absent from acid streams.

Steinman *et al.* (1991) performed laboratory experiments to examine the interactive effects of nutrients (N & P) and herbivory (by the snail *Elimia clavaeformis*). The experiments indicated that nutrient uptake by algae was reduced when grazing intensity increased, due to the lower biomass levels. In the River Browney, chlorophyll *a* on cobbles and boulders shows a significant inverse correlation with TIN (Table 4.5).

Nutrient limitations are often critical in the control of community structure and processes in stream ecosystems (Chessman *et al.*, 1992). In a study using nutrient diffusing artificial substrata Chessman *et al.* (1992) showed that N was the primary nutrient limiting algal biomass and in many cases P was the secondary limiting nutrient. Ratios of total inorganic nitrogen to reactive phosphorus in stream waters showed a consistent relationship to limiting nutrients. In some cases,

exact ratios could not be calculated because nutrient concentrations were below detection limits. At ratios above 17 by mass, phosphorus was always limiting, or results were inconclusive. At ratios below 7 by mass nitrogen was limiting. Measurement of stream water nutrient status could be used to predict the limiting nutrient in most cases, unless factors other than nutrients (e.g. light and grazing) were limiting (Chessman *et al.*, 1992). In the River Browney TIN : FRP ratios, were below 7.1 (Table 4.2).

Diatom populations in many temperate streams and rivers show a marked bloom in spring and autumn, with much lower populations in summer, when green algae are often more prolific (Marker and Willoughby, 1988). Physiological tests indicated that *Cladophora* growth was likely to be limited by N during late summer and early autumn (Lohman and Priscu, 1992). In the River Browney *Cladophora* may have been favoured by low TIN : FRP in late June and July. However, TIN : FRP was similarly low in early May (Table 4.2).

Seasonal variations in silicate concentrations in streams are well documented (Marker, 1976; Casey *et al.*, 1981; Marker and Casey, 1982, M.G. Kelly, pers. comm.). There is a general trend of highest silicate levels in the spring and autumn, at times of diatom 'blooms', and lowest levels in mid-summer, when there is a widespread collapse in epilithic diatom biomass. The minimum silicate requirements of diatoms for growth and reproduction need further investigation in order to further assess the importance of silicate availability as a limiting factor.

5.3 Variations in community structure

In Harwood Beck, N and P values were very low and frequently below the detection limit of determination techniques. N and P could not be correlated to percentage changes in individual taxa, although species such as *Navicula gregaria* and *N. lanceolata* were found only when N and P were relatively high. Pringle (1990) observed little change in periphyton taxonomic structure when inorganic nitrate and phosphate were added in relatively small amounts (i.e from 40 to 80 $\mu\text{g l}^{-1}$ NO_3^- - N and from 2.5 to 12.5 $\mu\text{g l}^{-1}$ PO_4^- - P). The apparent resistance of algal taxonomic structure may arise from metabolic flexibility in nutrient uptake rates (Steinman *et al.*, 1991).

Higher available nitrogen concentration when biomass is low may explain the inverse correlation of *N. gregaria* and *N. lanceolata* with total inorganic nitrogen in the River Browney (Figs

4.13 and 4.14), because *Navicula* spp. appear to decrease in percentage composition when epilithic diatom biomass declines as they are loosely attached motile species of the upper epilithon. Figures 4.11 and 4.20 show the loosely attached nature of *Navicula* spp. Fig. 4.12 shows the percentage of *N. lanceolata* falling from a stable $\approx 30\%$ to $\approx 10\%$ at the time of the collapse in River Browney chlorophyll *a* (Fig. 4.6).

Stalked (*Gomphonema* and *Cymbella*) taxa and smaller taxa growing directly on the substratum (*Achnanthes*) tended to be most adapted to resisting the mechanical force of current velocity (Wendker, 1992). This is supported by Figures 4.15, 4.21 and 4.22 which show substantial changes in morphological guild structure following stormflow (i.e. relative increases in stalked taxa and *Achnanthes* accompanied by the decrease in *Navicula* spp.).

Steinman *et al.* (1991) found the influence of grazing on diatom community structure to be a direct function of growth form. Prostrate growth forms (e.g. *Cocconeis*) are well adapted to high grazing pressure (Steinman, 1992). Stalked taxa may be more susceptible to grazing than closely adhered forms such as *Cocconeis placentula*. When biomass is low due to high herbivore activity prostrate growth forms become dominant (Steinman *et al.*, 1991). The massive percentage increase of *C. placentula* in the River Browney during July suggests that grazing may be an important process (Fig. 4.1). Table 5.1 indicates that *Achnanthes minutissima* may be more resistant to flow while *Cocconeis placentula* is more resistant to grazing. On 3 June and 6 July, pH, nutrients and chlorophyll *a* were similar (Table 5.1). On 3 June *A. minutissima* was dominant at a maximum of 42% (in high current), while *C. placentula* was minimal (2.6%). On 6 July *A. minutissima* was at its lowest (8.5%) while *C. placentula* was dominant (37%).

Table 5.1. Cross-tabulated River Browney data

Date	pH	TIN ($\mu\text{g l}^{-1}$)	FRP ($\mu\text{g l}^{-1}$)	Current (m s^{-1})	Chl. <i>a</i> ($\mu\text{g cm}^{-2}$)		% <i>A.min.</i>	% <i>C. plac.</i>
					cobbles	boulders		
3 June	7.8	1426	310	0.80	0.11	0.45	42	2.6
6 July	8.1	1515	365	0.55	< 0.10	< 0.10	8.5	37

5.4 Variation in diatom indices

5.4.1 River Browney

Figures 4.23 and 4.24 show that the SPI and GDI values for the River Browney become slightly higher following stormflow, due to the percentage increase in *Achnanthes minutissima*. The SPI and GDI are in close agreement, particularly on cobbles, but both remained more stable on boulders.

The Round zone 3 ('nutrient rich') classification is in general agreement with Costes' diatom indice classification (SPI and GDI = 3-4 = 'more important changes in the community; decrease of the sensitive species; moderate pollution or significant eutrophication'). Round (1993) considers his zone 3 classification to correspond with the pH and alkalinity classification by Leclercq and Manquet (1987) (i.e. pH 6.5 - 7.3; alk. 5.0 - 23.3); however, the data collected from the River Browney during this study do not support this (pH 7.1 - 8.2; alk. 2.7 - 8.1).

5.4.2 Harwood Beck

Figures 4.25 and 4.26 show that Harwood Beck SPI and GDI values become slightly higher following stormflow, due to the percentage increase in *Achnanthes minutissima*. The cobble SPI values were lowest (i.e. below 4) on 3 May before stormflow (minimum % *A. min.*, maximum % *Navicula* spp.) and 17 June (relatively high percentage of *Cymbella delicatula*); the GDI remains more stable. With these exceptions the SPI and GDI are in close agreement with diatom indice values between 4 and 5.

As for the River Browney, pH (6.9 - 8.4) and alkalinity (1.9 - 12) data from this study do not fit the ranges adopted by Round for this classification (pH 5.6 - 7.1; alk 2.8 - 5.7). In addition, with Round (1993)'s system, Harwood Beck is classified as a zone 2 site (defined as being 'nutrient richer'); this is misleading as Harwood Beck was oligotrophic when sampled. Harwood Beck fits the zone 1 description ('Clean water of low pH in uppermost reaches') which corresponds to the classification by Coste's SPI and GDI (= 4.0 - 4.5 = 'almost normal quality, slight changes in the community, slight pollution or I.D. => 4.5 = Best biological quality, no pollution'). *Achnanthes microcephala* is present in low abundance, but *Eunotia exigua* is absent. Round describes these species as small celled and firmly attached and this description is suitable for *Achnanthes minutissima*

which occurs widely in clean waters. Round (1993) does acknowledge that all workers do not distinguish between *A. microcephala* and *A. minutissima*. Harwood Beck also has a moderate abundance of small *Navicula* spp. which can be misleading when using Round's system as they can be the dominant taxa in grossly polluted water.

5.5 Concluding comments

Temporal variation in epilithic diatom communities seems to be predominantly associated with flow induced changes in diatom biomass and dissolved nutrient concentrations, during spring and early summer. The prevention from recovery and summer collapse in diatom biomass may be related to increased grazing pressure, low silicon concentration and perhaps increased competition with other algae such as *Cladophora*. However, this has little effect on the interpretation of water quality. The maximum TIN : FRP was recorded following stormflow at both sites (Table 4.2). Dilution of FRP due to high flow volume and lower uptake of TIN may have been related to the flow-induced decline in algal biomass.

The diatom-based SPI and GDI water quality indices generally remained relatively stable (i.e. within one class of water quality), despite high variability in flow regime and chlorophyll *a*. GDI was more stable than SPI at Harwood Beck and produced well defined water quality classifications for both sites. Changes in the percentage of *Achnanthes minutissima* (of poor indicating value) accounted for much of the variation. The results indicate greater stability of SPI and GDI on boulders, which is highly likely to be due to the greater stability of boulders during flow events. It is not possible to comment on the stability of indices within stormflow periods and sampling at these times would be dangerous and unnecessary.

Effectively equivalent water quality classifications are derived from the SPI and GDI. Although this conclusion is limited to 2 sites it supports observations made by Coste *et al.* (1991) in France, and Kelly (pers. comm.) in other U.K. waters. Round (1993)'s zoning system is qualitative and requires careful interpretation, due to differences in taxonomy and the pH and alkalinity ranges.

CHAPTER 6

SUMMARY

- (1) Epilithic diatom communities and relevant physico-chemical variables were studied over spring and summer in Harwood Beck and the River Browney. The effect of stormflow on diatom-based water quality indices was assessed.
- (2) The River Browney and Harwood Beck sites were very similar in channel dimensions, temperatures, pH and Total Alkalinity. The main physical differences between sites were altitude, magnitude of stormflow events and the degree of bankside shading.
- (3) The most important difference in chemistry of the two sites is in nutrient status:
River Browney mean TIN \cong 1200 $\mu\text{g l}^{-1}$, mean FRP \cong 350 $\mu\text{g l}^{-1}$; Harwood Beck mean TIN \cong 60 $\mu\text{g l}^{-1}$; mean FRP \cong 10 $\mu\text{g l}^{-1}$
- (4) The difference in water hardness is of secondary chemical importance:
River Browney mean conductivity = 544 $\mu\text{S cm}^{-1}$; standard deviation = 30 $\mu\text{S cm}^{-1}$; Harwood Beck mean conductivity = 244 $\mu\text{S cm}^{-1}$; standard deviation = 35 $\mu\text{S cm}^{-1}$
- (5) The River Browney may be N limited (TIN : FRP is usually less than 7)
- (6) Chlorophyll *a* ($\mu\text{g cm}^{-2}$) is highly variable on cobbles and boulders. Analysis of variance of River Browney \log_{10} chlorophyll *a* data shows a significant difference in chlorophyll *a* ($\mu\text{g cm}^{-2}$) between dates, but not between cobbles and boulders, over the entire study period. Chlorophyll *a* ($\mu\text{g cm}^{-2}$) of the River Browney epilithon was inversely correlated with current speed ($r = 0.79$, $p < 0.05$ for cobbles) and total inorganic nitrogen ($r = -0.9$, $p < 0.01$ for cobbles; $r = -0.8$, $p < 0.05$ for boulders). Uptake of inorganic nitrogen may be directly related to epilithic diatom biomass.

At Harwood Beck chlorophyll *a* ($\mu\text{g cm}^{-2}$) is significantly different between dates, and between cobbles and boulders, over the entire study period. Differences in chlorophyll *a* ($\mu\text{g cm}^{-2}$) between cobbles and boulders of Harwood Beck may be largely attributable to the relative stability of boulders and instability of cobbles during stormflow. Following stormflow, chlorophyll *a* is over twice as high on boulders at both sites.
- (7) *Cladophora glomerata* may be competing with epilithic diatoms for light and space and may be favoured by higher N levels in June and July.

- (8) *Cocconeis placentula* and *Achnanthes minutissima* are strongly adhered species which increase in percentage following stormflow. By contrast, motile *Navicula* spp. generally show a substantial decrease in percentage following stormflow.
- (9) Stalked *Cymbella* and *Gomphonema* spp. appear to be intermediate in strength of adherence / resistance to stormflow and maintain a relatively constant percentage representation in the flora over stormflow periods.
- (10) A sharp increase in the percentage of *Cocconeis placentula* between mid-June and mid-July is related to the collapse in biomass level as it is a firmly attached prostrate species which colonises stones with a pavement like growth. It is thought to be less vulnerable to algal grazers than other species of vertical growth form and therefore likely to sharply increase in percentage if herbivory is the principle cause of biomass decline.
- (11) Previous data from the River Browney, Harwood Beck and many other running waters indicate that silicate levels are much lower in summer than in spring and autumn and this may be an important factor limiting diatom growth.
- (12) Despite the effects of flow rate upon species percentage and the community change associated with highly variable biomass levels, the Specific Pollution Index (SPI) and Generic Diatom Index (GDI) remain relatively stable over the study period (i.e. seasonally stable).
- (13) The SPI and GDI classify the River Browney as 'slightly to moderately polluted or significantly eutrophic' and Harwood Beck as either 'slightly polluted' or of the 'best biological quality'. This is in general agreement with the Round zoning system which classifies the River Browney as zone 3, ('nutrient rich') and Harwood Beck as zone 2.
- (14) The SPI is significantly correlated with the percentage of *Achnanthes minutissima* at both sites. Differences in the SPI and GDI before and after stormflow and between cobbles and boulders are within ≈ 0.5 classification unit. The SPI and GDI tend to remain more stable on boulders.

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APPENDIX 1

Nomenclature follows Krammer and Lange - Bertalot (1986 - 91) with a few exceptions (Round *et al.*, 1990), mainly associated with their controversial treatment of the Fragilariaceae.

Genera	Species	Authorities	
<i>Achnanthes</i>	<i>lanceolata</i>	(Brébisson) Grunow in Cleve and Grunow 1880	
	<i>minutissima</i>	Kützing 1833	
	<i>oblongella</i>	Öestrup 1902	
<i>Amphora</i>	<i>pediculus</i>	(Kützing) Grunow 1880	
<i>Cyclotella</i>	<i>meneghiniana</i>	Kützing 1844	
<i>Cocconeis</i>	<i>pediculus</i>	Ehrenberg 1838	
	<i>placentula</i>	Ehrenberg 1838	
<i>Cymbella</i>	<i>affinis</i>	Kützing 1844	
	<i>delicatula</i>	Kützing 1849	
	<i>microcephala</i>	Grunow in Van Heurck 1880	
	<i>minuta</i>	Hilse ex Rabenhorst 1862	
	<i>silesiaca</i>	Bleisch in Rabenhorst 1864	
<i>Diatoma</i>	<i>moniliformis</i>	Kützing 1833	
	<i>tenuis</i>	Agardh 1812	
	<i>vulgaris</i>	Bory 1824	
<i>Fragilaria</i>	<i>crotonensis</i>	Kitton 1869	
	<i>capucina</i> var. <i>capucina</i>	Desmazières 1825	
	<i>capucina</i> var. <i>gracilis</i>	(Öestrup) Hustedt 1950	
	<i>capucina</i> var. <i>vaucheriae</i>	(Kützing) Lange-Bertalot 1980	
	<i>famelica</i>	(Kützing) Lange-Bertalot 1980	
<i>Gomphonema</i>	<i>acuminatum</i>	Ehrenberg 1832	
	<i>angustatum</i>	(Kützing) Rabenhorst 1864	
	<i>angustum</i>	Agardh 1831 <i>non</i> Kützing 1844	
	<i>olivaceoides</i>	Hustedt 1950	
	<i>olivaceum</i>	(Hornemann) Brébisson 1838	
	<i>parvulum</i>	(Kützing) Kützing 1849	
<i>Gyrosigma</i>	<i>acuminatum</i>	(Kützing) Rabenhorst 1853	
<i>Hannaea</i> *	<i>arcus</i>	(Ehrenberg) Patrick 1966	
<i>Meridion</i>	<i>circulare</i>	(Greville) C.A. Agardh 1831	
<i>Melosira</i>	<i>varians</i>	Agardh 1827	
<i>Navicula</i>	<i>capitata</i>	Ehrenberg 1838	
	<i>exigua</i>	(Gregory) Grunow in Van Heurck 1880	
	<i>exilis</i>	Kützing 1844	
	<i>gregaria</i>	Donkin 1861	
	<i>lanceolata</i>	(Agardh) Ehrenberg 1838 <i>non sensu</i> Kützing <i>nec sensu</i> Hustedt	
	<i>meniscus</i>	Schumann 1867	
	<i>submolesta</i>	Hustedt 1949	
	<i>tripunctata</i>	(O.F. Müller) Bory 1822	
	<i>Nitzschia</i>	<i>dissipata</i>	(Kützing) Grunow 1862
		<i>inconspicua</i>	Grunow 1862 <i>pro parte</i>
		<i>liebetruthii</i>	Rabenhorst 1864
<i>linearis</i> var. <i>linearis</i>		(Agardh) W. Smith 1853	
<i>Reimeria</i>	<i>palea</i>	(Kützing) W. Smith 1856	
	<i>sinuata</i>	(Gregory) Kociolek and Stoermer 1987	
	<i>Rhoicosphenia</i>	(C. Agardh) Lange-Bertalot 1980b	
<i>Surirella</i>	<i>brebissonii</i>	Krammer and Lange-Bertalot 1987	
<i>Synedra</i>	<i>pulchella</i>	Kützing 1844	
	<i>ulna</i>	Ehrenberg 1830	

* *Hannaea* = *Ceratoneis* = *Fragilaria* (in Lange-Bertalot)

APPENDIX 2

Chlorophyll and biomass data

Chlorophyll *a* ($\mu\text{g cm}^{-2}$)

Date	River Browney Cobbles		Boulders		Harwood Beck Cobbles		Boulders	
	Mean	Standard error	Mean	Standard error	Mean	Standard error	Mean	Standard error
03 May	8.32	2.55	3.36	1.35	1.07	0.42	1.3	0.475
12 May	6.25	2.48	8.01	3.85				
20 May	0.695	0.30	2.385	1.08	1.14	0.41	4.11	1.65
26 May	2.815	1.35	3.365	1.87				
03 June	0.105	0.05	0.445	0.19	0.6	0.185	2.185	0.585
17 June	0.81	0.13	0.57	0.185	3.365	0.72	2.785	0.775
23 June	< 0.1		< 0.1		< 0.1		< 0.1	
06 July	< 0.1		< 0.1		< 0.1		< 0.1	
16 July	0.15	0.025	0.405	0.06	0.10	0.025	0.29	0.08

Biomass (mg cm^{-2})

	River Browney Cobbles		Boulders		Harwood Beck Cobbles		Boulders	
	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation	Mean	Std. deviation
03 May	2.05	1.45	0.85	0.45	0.75	0.45	1.45	1.15
12 May	2.5	1.8	4.5	3.9				
20 May	0.42	0.2	0.52	0.475	0.44	0.34	1.71	0.595
26 May	0.51	0.345	0.31	0.36				

APPENDIX 3

Diatom counts from replicate samples

River Browney = B; Harwood Beck = H; Cobble = c; Boulder = b; + = present; - = absent

Diatoms	Date: 20 May				3 June		17 June			16 July		
	Replicates: Bc2	Bc3	Bb1	Bb4	Bb2	Hc1	Hb2	Hc2	Hc3	Hb3	Hc3	Hb1
	Percentage: %	%	%	%	%	%	%	%	%	%	%	%
<i>Achnanthes lanceolata</i>	2.2	1.8	0.8	5.1	6.5	+	-	-	-	-	3.9	0.6
<i>Achnanthes minutissima</i>	46.9	9.6	17.8	48	8.8	51.4	71.6	55.7	67	84	61.7	66.3
<i>Achnanthes oblongella</i>	-	-	-	-	-	-	0.4	-	-	-	-	-
<i>Amphora pediculus</i>	1.1	2.6	31.2	7.2	22.9	-	-	-	-	-	-	-
<i>Ceratoneis arcus</i>	1.7	-	-	0.4	-	2	2.3	2.5	1.8	0.4	3.3	0.6
<i>Cyclotella meneghiniana</i>	1.7	-	-	-	3.2	-	-	-	-	-	-	-
<i>Cocconeis pediculus</i>	-	-	-	0.4	1.8	-	-	-	-	-	-	-
<i>Cocconeis placentula</i>	5	7	2.5	3.8	13.8	-	-	-	-	-	-	0.3
<i>Cymbella affinis</i>	3.3	1.8	-	-	0.9	1.8	-	1	-	1.9	1.1	4.1
<i>Cymbella delicatula</i>	5	2.6	-	0.4	1.4	11.3	2.3	18.1	8.3	0.8	7.8	3.8
<i>Cymbella microcephala</i>	10.6	2.6	1.7	-	2.3	3.5	6	6.4	3.6	0.8	3.9	4.5
<i>Cymbella minuta</i>	1.1	-	-	-	2.3	8.3	+	-	1.8	0.8	-	1
<i>Cymbella silesiaca</i>	-	-	-	-	-	-	1.9	0.5	-	0.8	0.6	-
<i>Diatoma moniliformis</i>	-	-	-	-	-	1	1.2	2	2.2	1.5	0.6	0.6
<i>Diatoma tenuis</i>	-	-	0.4	0.4	0.5	0.5	0.8	-	0.9	-	-	-
<i>Diatoma vulgare</i>	-	-	-	-	0.5	0.5	-	-	-	-	-	-
<i>Fragilaria crotonensis</i>	-	-	-	-	-	-	1.2	+	+	-	-	-
<i>Fragilaria capucina</i> var. <i>cap.</i>	-	-	-	-	0.5	-	-	1	1.3	-	2.8	2.5
<i>Fragilaria cap.</i> var. <i>vaucheriae</i>	2.8	0.9	0.4	0.4	-	2	0.4	1	0.9	0.4	-	-
<i>Fragilaria famelica</i>	-	-	-	-	-	0.5	-	0.5	-	-	-	-
<i>Gomphonema angustatum</i>	-	-	-	-	-	-	0.4	1	0.9	-	2.2	-
<i>Gomphonema angustum</i>	-	-	-	-	-	2.5	-	-	-	0.8	-	-
<i>Gomphonema olivaceoides</i>	0.6	0.9	-	-	-	2	3.5	-	-	0.4	1.1	-
<i>Gomphonema olivaceum</i>	1.7	-	2.1	5.1	0.9	8.8	-	4.4	0.9	0.4	-	6.3
<i>Gomphonema parvulum</i>	-	-	-	0.8	-	-	-	-	-	-	-	-
<i>Gomphonema</i> spp.	-	2.6	0.8	0.4	-	-	-	-	-	-	1.2	1.3
<i>Meridion circulare</i>	-	-	-	-	0.5	-	0.4	-	+	-	0.6	-
<i>Melosira varians</i>	-	-	-	-	0.9	-	-	-	-	-	-	-
<i>Navicula capitata</i>	-	-	-	-	1.4	-	-	-	-	-	-	-
<i>Navicula exilis</i>	-	-	-	-	-	-	-	-	0.4	-	-	-
<i>Navicula gregaria</i>	5.6	18.5	6.4	6.8	12.4	-	-	-	-	-	-	-
<i>Navicula lanceolata</i>	5.6	41.2	11.9	8.9	6	-	-	-	-	-	-	-
<i>Navicula meniscus</i>	-	-	-	3.4	-	-	-	-	-	-	-	-
<i>Navicula</i> spp.	-	-	-	-	-	2.9	6	5.4	9.6	7.4	8.9	8.1
<i>Nitzschia dissipata</i>	-	0.9	0.4	-	-	-	-	-	-	-	-	-
<i>Nitzschia inconspicua</i>	-	-	0.8	-	-	-	-	-	-	-	-	-
<i>Nitzschia linearis</i> var. <i>linearis</i>	-	-	-	-	-	0.5	-	-	-	-	-	-
<i>Nitzschia palea</i>	1.7	0.9	2.5	0.8	2.8	-	-	-	-	-	-	-
<i>Reimeria sinuata</i>	2.8	0.9	1.7	5.9	0.9	-	-	-	-	-	0.6	-
<i>Rhoicosphenia abbreviata</i>	0.6	5.2	18.2	1.4	6	-	-	-	-	-	-	-
<i>Surirella brebissonii</i>	-	-	0.4	0.4	2.8	-	-	-	-	-	-	-
<i>Synedra pulchella</i>	-	-	-	-	-	-	1.2	-	-	-	-	-
<i>Synedra ulna</i>	-	-	-	-	-	0.5	0.4	0.5	0.4	-	-	-
NUMBER OF TAXA:	18	16	17	19	23	18	17	15	16	13	15	13

APPENDIX 4

Spreadsheet for calculating diatom water quality indices (SPI and GDI)
(Worked example from 16 July: Harwood Beck, boulders)

B = Boulders; SPP = Species; GEN = Genera; S = Sensitivity; V = Indicator value; A = Abundance

Diatom	Harwood Beck:					B				
	SPP.		B %			GEN.		B %		
	S	V	A	SVA	AV	S	V	A	SVA	AV
<i>Achnanthes lanceolata</i>	4	1	0.4	1.6	0.4	5	1	0.4	2	0.4
<i>Achnanthes minutissima</i>	5	1	61.7	308.5	61.7	5	1	61.7	308.5	61.7
<i>Amphora pediculus</i>	4	2		0	0	3	2		0	0
<i>Ceratoneis arcus</i>	5	2	1.8	18	3.6	5	2	1.8	18	3.6
<i>Cyclotella meneghiniana</i>	2	1		0	0	3	1		0	0
<i>Cocconeis pediculus</i>	4	2		0	0	4	1		0	0
<i>Cocconeis placentula</i>	4	1	0.9	3.6	0.9	4	1	0.9	3.6	0.9
<i>Cymbella affinis</i>	4	2	5.3	42.4	10.6	5	1	5.3	26.5	5.3
<i>Cymbella delicatula</i>	5	2	3.1	31	6.2	5	1	3.1	15.5	3.1
<i>Cymbella microcephala</i>	4	2	5.9	47.2	11.8	5	1	5.9	29.5	5.9
<i>Cymbella minuta</i>	4	2	0.4	3.2	0.8	5	1	0.4	2	0.4
<i>Cymbella silesiaca</i>	5	2		0	0	5	1		0	0
<i>Cymbella sinuata</i>	5	1	0.4	2	0.4	5	1	0.4	2	0.4
<i>Diatoma moniliformis</i>	0	0		0	0	4	1		0	0
<i>Diatoma tenuis</i>	5	3	0.4	6	1.2	4	1	0.4	1.6	0.4
<i>Diatoma vulgare</i>	4	1		0	0	4	1		0	0
<i>Fragilaria crotonensis</i>	4	1		0	0	4	1		0	0
<i>Fragilaria capucina</i> var. <i>cap.</i>	4	1	5.3	21.2	5.3	4	1	5.3	21.2	5.3
<i>Fragilaria cap.</i> var. <i>gracilis</i>	4	1		0	0	4	1		0	0
<i>Fragilaria cap.</i> var. <i>vaucheriae</i>	3	1		0	0	4	1		0	0
<i>Fragilaria famelica</i>	4	1		0	0	4	1		0	0
<i>Gomphonema angustatum</i>	4	1		0	0	3	2		0	0
<i>Gomphonema angustum</i>	5	1		0	0	3	2		0	0
<i>Gomphonema olivaceoides</i>	5	3	1.8	27	5.4	3	2	1.8	10.8	3.6
<i>Gomphonema olivaceum</i>	5	2	1.8	18	3.6	3	2	1.8	10.8	3.6
<i>Gomphonema</i> spp.	3	2		0	0	3	2		0	0
<i>Meridion circulare</i>	5	2	0.4	4	0.8	5	2	0.4	4	0.8
<i>Navicula exilis</i>	3	1		0	0	3	1		0	0
<i>Navicula gregaria</i>	3	1		0	0	3	1		0	0
<i>Navicula accomoda</i>	1	3		0	0	3	1		0	0
<i>Navicula lanceolata</i>	3	1	0.4	1.2	0.4	3	1	0.4	1.2	0.4
<i>Navicula submolesta</i>	2	2		0	0	3	1		0	0
<i>Navicula</i> spp.	3	1	10.4	31.2	10.4	3	1	10.4	31.2	10.4
<i>Nitzschia palea</i>	1	3		0	0	1	1		0	0
<i>Nitzschia</i> spp.	1	1		0	0	1	1		0	0
<i>Rhoicosphenia abbreviata</i>	4	1		0	0	4	1		0	0
<i>Synedra pulchella</i>	3	3		0	0	3	1		0	0
<i>Synedra ulna</i>	3	1		0	0	3	1		0	0

Total : 566.1 124

488.4 106.2

$$\text{SPI} = 566.1 / 124 = 4.584$$

$$\text{GDI} = 488.4 / 106.2 = 4.599$$

