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A diatom species index for bioassessment of Australian rivers

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Abstract. The Diatom Index for Australian Rivers (DIAR), originally developed at the genus level, was reformulated at the species level with data from diatom sampling of rivers in the Australian Capital Territory, New South Wales, Queensland, South Australia and Victoria. The resulting Diatom Species Index for Australian Rivers (DSIAR) was significantly correlated with the ARC_E (Assessment of River Condition, Environment) index developed in the Australian National Land and Water Resources Audit (NLWRA), and with nine of the ARC_E's constituent indices and sub-indices, across 395 river reaches in south-eastern Australia. These correlations were generally stronger than those shown by the biological index that was used to assess river condition in the NLWRA, the ARC_B (Assessment of River Condition, Biota) index based on macroinvertebrates and the Australian River Assessment System (AUSRIVAS). At a finer spatial scale, DSIAR was strongly and significantly correlated with measures of catchment urbanisation for streams in the eastern suburbs of Melbourne, Victoria. DSIAR scores across south-eastern Australia bore little relationship to the latitude, longitude or altitude of sampling sites, suggesting that DSIAR is not greatly affected by macro-geographical position. In addition, DSIAR scores did not vary greatly among small-scale hydraulic environments within a site. DSIAR appears to have potential as a broad-scale indicator of human influences on Australian rivers, especially the effects of agricultural and urban land use, and also for impact studies at a local scale. Further evaluation is warranted to test the sensitivity of the index to natural variables such as catchment geology, and to assess its performance in northern, western and inland Australia.

Additional keywords: biological monitoring, biotic index, water quality.

Introduction

Diatoms are used widely to monitor fresh waters, particularly in Europe and North America (e.g. Potapova and Charles 2002; Prygiel 2002). In Australia, considerable use has been made of freshwater diatoms in palaeolimnological studies aimed at reconstruction of past climates and historical or pre-historical changes in water quality (Gell *et al*. 2005), and various case studies of responses of diatoms to particular anthropogenic stressors have been reported (see references below). However, diatoms are seldom included in routine, large-scale biological assessment of Australian fresh waters, a field that is heavily focussed on macroinvertebrates (e.g. Davies 2000). For example, diatoms do not form part of the 'Index of Stream Condition' used in the State of Victoria (DSE 2005), the 'Sustainable Rivers Audit' of streams in the Murray–Darling Drainage Division (MDBC 2004) or the 'Ecosystem Health Monitoring Program' in south-eastern Queensland (EHMP 2005). This is curious, since diatoms have several attributes that should render them useful in bioassessment of Australian fresh waters. They are easily and quickly collected, can be stored as permanent mounts on microscope slides that require little storage space or maintenance, and appear to respond to a wide range of anthropogenic stressors such as thermal pollution (Chessman 1985), sewage disposal (Chapman and Simmons 1990; Dela-Cruz *et al*. 2006), upstream impoundment (Growns and Growns 2001), secondary salinisation (Blinn and Bailey 2001; Blinn *et al*. 2004), and urban stormwater (Sonneman *et al*. 2001; Newall and Walsh 2005).

The limited adoption of diatoms for bioassessment in Australia is probably related to a scarcity of methods with demonstrated capability for effective, routine application in this country. Although diatom indices developed in other continents have sometimes been applied in Australia (e.g. Newall and Walsh 2005), their use has not been tested widely and may be problematic. For example, Newall *et al*. (2006) found that the European Indice Biologique Diatomées (Lenoir and Coste 1996) showed no apparent relationship to catchment disturbance in the Kiewa River, Victoria. Chessman *et al*. (1999*b*) developed preliminary bioassessment methods for diatoms in the eastern parts of New South Wales (NSW) and Victoria, which included a Diatom Index for Australian Rivers (DIAR). For this index, 55 genera, defined according to Round *et al*. (1990), were assigned numbers ranging from 1 to 10 to reflect their inferred sensitivity to common anthropogenic stressors. DIAR was intended as a generalised indicator of human influence, rather than an indicator of specific stressors such as salinity (cf. Philibert *et al*. 2006). DIAR scores, calculated as the average of the sensitivity values of the genera present in a standard sample, and expressed relative to predicted scores in the absence of anthropogenic stress,

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differed significantly between near-pristine reference sites and sites exposed to human influence. The quotient of observed and predicted values of DIAR also correlated significantly with alkalinity, electrical conductivity, hardness and pH (Chessman *et al*. 1999*b*).

In the present paper, we extend DIAR to a species-level version (Diatom Species Index for Australian Rivers, or DSIAR) with data from four Australian states and the Australian Capital Territory (ACT). A species-level version offers the potential for greater responsiveness to anthropogenic stress by incorporating information on variation in sensitivity among species within a genus. It also circumvents problems caused by continuing frequent changes in taxonomic definitions of diatom genera. We test the new index by examining its association with environmental variables, including independent data on anthropogenic alteration of Australian rivers derived from a recent nation-wide assessment (the National Land and Water Resources Audit).

Materials and methods

Datasets used for index derivation

DSIAR was developed with diatom data from several recent surveys of streams in the ACT, NSW, Queensland, South Australia (SA) and Victoria undertaken by the authors and others (see Acknowledgements). These included sampling in SA and Victoria during 1994–1999 (Philibert *et al*. 2006), studies of the condition of rivers in several regions of NSW during 1999–2003 (Chessman 2002 and unpublished; Philibert *et al*. 2006) and sampling in the ACT, NSW, Queensland and Victoria in 2001–2003 for a comparative evaluation of bioassessment methods undertaken by the former Co-operative Research Centre for Freshwater Ecology (Marchant *et al*. 2006). Collectively these studies generated over 1100 diatom samples from more than 600 sites, covering a wide range of environments from pristine protected areas to agricultural and urban surroundings and spread over about a quarter of the Australian continent (Fig. 1). The dataset of Chessman *et al*. (1999*b*) was not used because diatom identification for that study was to genus level only.

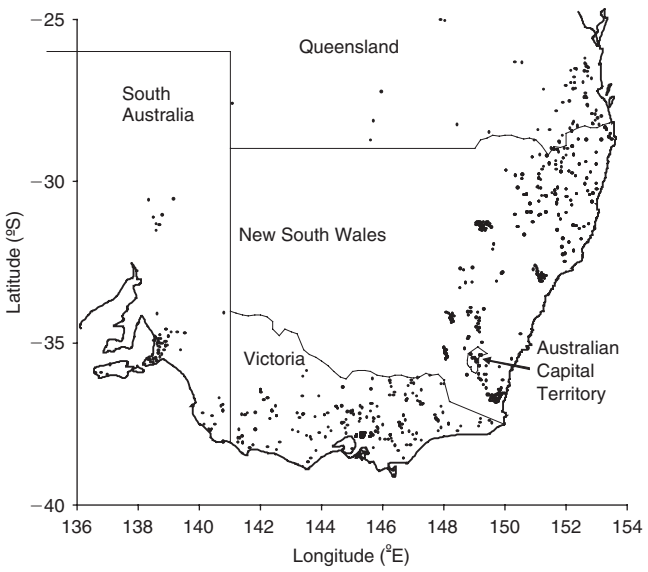

Fig. 1. Location of diatom sampling sites in south-eastern Australia.

Diatoms were sampled from both flowing water and pools, mostly from hard substrata with sharpened wooden spatulas but sometimes from mud surfaces with pipettes (Chessman *et al*. 1999*a*). Often two or three samples were taken at a site at the same time, but from different hydraulic environments – 'riffles' (defined by the presence of unbroken standing waves), 'runs' (unbroken waves moving downstream), 'glides' (moving water with a flat surface) and 'pools' (still water) – and sometimes from different substrata (rocks, submerged wood, aquatic macrophytes and sediments). Samples were preserved in the field in ethanol or Lugol's iodine.

In the laboratory, diatom frustules were cleaned and mounted in Naphrax on microscope slides. Usually ∼300 valves per sample (mean = 314; s.d. = 109) were identified to species or sub-species level by transect scanning under a microscope at a magnification of 1000×. In some samples where diatoms were sparse, the number counted was less (minimum = 3; 5th percentile = 63). Identification followed standard international keys (Krammer and Lange-Bertalot 1986, 1988, 1991*a*, 1991*b*; Lange-Bertalot and Metzeltin 1996; Reichardt 1999; Krammer 2000; Witkowski *et al*. 2000; Lange-Bertalot 2001) and Australian keys (Gell *et al*. 1999; Sonneman *et al*. 2000). All samples were analysed by a single laboratory at the University of Adelaide.

Derivation of sensitivity values

Sensitivity values (SVs) intended to reflect sensitivity or tolerance to anthropogenic stress were derived objectively for all identified species in the manner described by Chessman (2003) for macroinvertebrates. This approach is an iterative gradient analysis, similar to reciprocal averaging, in which preliminary SVs are used to assign scores to a set of samples collected across a gradient of anthropogenic stress, and the SVs and sample scores are alternately and repeatedly revised until both stabilise. It requires datasets in which the dominant variation is associated with human disturbance rather than natural spatial and temporal patterns. In order to reduce the influence of natural gradients, analyses were done separately for specific combinations of geographic region and season. If an original study covered a broad geographic area and more than one season, data were subdivided. For example, 1994–1999 data from Victoria were divided into the north-east, north-west, south-east and south-west parts of the state, and autumn and spring data were analysed separately. This subdivision process resulted in 37 datasets for analysis: 18 from NSW, three from Queensland, three from South Australia, 12 from Victoria, and one from the Border Rivers and surrounding areas spanning the NSW–Queensland border. The number of sites per dataset ranged from 5 to 44 (mean of 21).

Each dataset for a particular combination of geographic region and season was treated as follows. First, abundances of diatom species in each sample were expressed as proportions of the total number of valves counted, with any varieties or formae of the same species amalgamated. If more than one sample had been taken from a site at the same time, proportional abundance data were averaged across all simultaneous samples. The starting SV for each species was set as the DIAR SV for the corresponding genus (Chessman *et al*. 1999*b*), and preliminary

site scores were calculated as abundance-weighted averages of the SVs of the species recorded from each site. Rank correlation coefficients were then calculated between these initial scores and the relative abundances of each species. Since it is mathematically impossible for a species with few occurrences in a dataset to achieve a very large positive or negative correlation, each correlation coefficient was divided by the maximum positive coefficient that is theoretically possible for a species recorded with the same frequency. The resulting quotients therefore had a possible range from −1 (for a species with a negative correlation coefficient equivalent to the theoretical maximum for its occurrence frequency) to +1 (for a species with a positive correlation equivalent to the possible maximum for its frequency). These quotients were then used to assign revised SVs to the species. The species with the highest positive quotient, suggestive of the greatest sensitivity to anthropogenic disturbance, was assigned an initial DSIAR SV of 100. The species with the lowest negative quotient, suggestive of the greatest tolerance, was assigned an initial SV of 1. The other species were scaled between these extremes in proportion to their quotients. The use of quotients rather than raw correlation coefficients avoided the risk of rare species being assigned mid-range SVs simply because of their rarity.

The revised SVs were used to calculate revised site scores and the process of recalculating SVs and scores was repeated several times until the SVs stabilised. Final sets of SVs were derived by averaging SVs obtained from each of the 37 individual datasets. Standard deviations of final SVs were calculated for those species represented in more than one dataset.

Calculation of index scores

Final SVs were used to calculate DSIAR scores for each sample in the datasets used for index derivation, and for other diatom data, in two forms. Scores weighted by proportional abundance – hereafter DSIAR-w scores – were calculated by the multiplication of the average proportional abundance of each species (on a scale of 0 to 1) by its DSIAR SV and the summing of the resulting products. These scores therefore estimate the sensitivity of the average individual in a sample. Unweighted scores – hereafter DSIAR-uw scores – were calculated by the simple averaging the SVs of all the species recorded in a sample, and therefore estimate the sensitivity of the average species. Both types of DSIAR scores have a possible range of 1–100. High DSIAR scores signify a flora considered to be sensitive to common anthropogenic stressors, implying that the level of these stressors is likely to be low (i.e. that river condition is comparatively natural). Conversely, low scores are interpreted as indicating a flora that tolerates anthropogenic stress, or even responds positively to it, and hence the likely presence of such stress.

Relationships of DSIAR scores to geographic location and habitat

In order to assess broad-scale geographic variation, DSIAR scores for individual samples were plotted against the latitude, longitude, and altitude of the sampling sites. To assess within-site variation, scores for samples collected from rocks at the same site and time, but from different hydraulic environments, were compared by Pearson correlation and paired-sample *t*-tests. Hydraulic environments were compared pair-wise rather than collectively (e.g. by analysis of variance) because the mix of environments sampled varied among sites, and hence the data were unbalanced. Samples for which fewer than 200 valves were counted were excluded from the latter analyses because of the possibility that DSIAR scores would be unstable at low counts. There were insufficient co-incident samples for meaningful comparisons of rocks with other substrata.

Relationships of DSIAR scores to anthropogenic stressors

As a test of the expected relationship between DSIAR scores and anthropogenic stressors, scores calculated for samples in the datasets used for index derivation were related to independent measures of human influence obtained from the Assessment of River Condition (ARC), a recent, continental-scale evaluation of fluvial environments and catchments for Australia's National Land and Water Resources Audit (Norris *et al*. 2001). The ARC includes an environment index (ARC_E) amalgamated from four constituent indices (a catchment disturbance index, a hydrological disturbance index, a habitat index and a nutrient and suspended sediment load index). These four indices were in turn calculated from a series of sub-indices (Table 1). The source data for the sub-indices were primarily cartographic data, satellite imagery, stream-flow monitoring data and numerical modelling. The ARC_E indices and sub-indices were generated as means for river reaches, which averaged 14 km in length but ranged up to 180 km. The ARC also includes a biological index, ARC_B (Assessment of River Condition, Biota), based on aquatic macroinvertebrates. All ARC indices and subindices are scaled from 0 to 1, where 1 represents an estimated natural (or at least pre-European) state and 0 represents an estimated state under a high degree of human influence (see Norris *et al*. (2001) for further details).

DSIAR scores for 395 ARC reaches containing diatom sampling sites were associated with ARC_E index and sub-index scores by the calculation of Pearson correlation coefficients. For this analysis DSIAR scores, in both weighted and unweighted forms, were averaged for all diatom samples from each reach. The hydrological disturbance index and its sub-indices were excluded from analysis because they were available for fewer than 20% of the reaches with diatom data. Correlations were also calculated for ARC_B, for comparison with DSIAR.

As a further test of the relationship between DSIAR and human influence, index scores were calculated for diatom samples collected in a previous study of the impact of urbanisation on streams in the eastern suburbs of Melbourne, Victoria (Newall and Walsh 2005). In that study, four diatom samples were taken in each of two sampling periods (February–March 2002 and October–November 2002) from submerged rocks (and sometimes other hard substrata) at 16 independent sites with various levels of urban development in their catchments. DSIAR scores were averaged for the four replicates at each site in each sampling period and the averages were regressed against two measures of likely anthropogenic stress: drainage connection and effective imperviousness. These variables reflect the extent of artificial hard surfaces in a catchment (roofs, roads, car parks, etc.) and their connection to streams via stormwater pipes (see Newall and Walsh (2005) for details). Values of drainage connection and effective imperviousness (measured on a scale of 0–1) were

Table 1. Subindices and indices constituting the Assessment of River Condition, Environment (ARC_E) index (Norris *et al.* 2001)

Index	Subindex	Description
Catchment disturbance	Infrastructure	Weighted average of areal extent of infrastructure such as roads, railroads, and utilities within reach watershed
	Land use	Weighted average of areal extent of land uses such as intensive agriculture, urbanisation, dryland cropping, forestry, and grazing within reach watershed
	Land cover change	Loss of forest cover during the period 1990–1995 within reach watershed
Hydrological disturbance	Change in mean annual flow	Deviation of total flow volume from modelled natural volume
	Change in flow duration curve	Deviation of monthly flow duration curves from modelled natural curves
	Change in seasonal amplitude	Deviation of seasonal flow range from modelled natural range
	Change in seasonal periodicity	Deviation of seasonal timing of high and low flows from modelled natural timing
Habitat	Bedload condition	Deviation of modelled current bedload from modelled natural load
	Riparian	Tree cover within 100 m of stream
	Connectivity	Calculation based on the occurrence of artificial barrier structures (e.g. dams and weirs) and levees
Nutrient and suspended sediment load	Suspended sediment	Deviation of modelled current suspended sediment load from modelled natural load
	Total P	Deviation of modelled current phosphorus load from modelled natural load
	Total N	Deviation of modelled current nitrogen load from modelled natural load

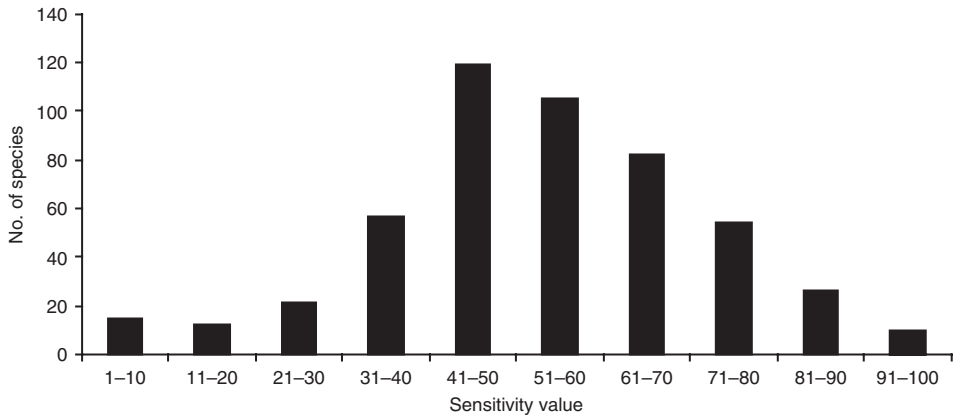

Fig. 2. Frequency distribution of derived sensitivity values for 501 diatom species.

transformed to fourth roots before analysis because the raw values were highly skewed.

Results

Sensitivity values

SVs were derived for 501 species (Appendix 1). The final (mean) SVs of these species had an approximately normal distribution (Fig. 2), and only species that occurred in 10 or fewer datasets had final SVs below 20 or above 80 (Fig. 3). The standard deviation (s.d.) of the SVs derived for individual species from different datasets also stabilised with increasing prevalence (Fig. 3), and 73% of the s.d.s were below the s.d. of SVs generated randomly between 1 and 100 (s.d. = 29). Intra-generic variation in mean SVs was sometimes low; for example, SVs were generally high in *Brachysira*, *Eunotia*, *Fragilaria* and *Frustulia*. However, a wide range of SVs occurred in most genera comprising many species (Appendix 1).

Relationships of DSIAR scores to geographic location and habitat

DSIAR scores for individual samples had very weak relationships to the latitude, longitude and altitude of the sampling sites (Fig. 4). A few sites at high altitudes had particularly high scores (>70) for both the weighted and the unweighted form of the index. These sites were all in eastern Victoria.

Four hydraulic environments had sufficient qualifying samples for meaningful comparisons of DSIAR scores among samples taken from different hydraulic environments but at the same site and time, and from the same substratum (rocks). These were pools ($n = 193$), riffles ($n = 138$), runs ($n = 92$) and glides ($n = 48$). DSIAR-uw scores for coincident samples were highly correlated for all possible pairs of these environments, and did not show any consistent bias (Fig. 5). For DSIAR-w the consistency between hydraulic environments was generally lower (Fig. 6). Paired *t*-tests found no significant differences between environments for DSIAR-uw ($P > 0.05$ in all cases), but for DSIAR-w, differences were significant between pools and runs,

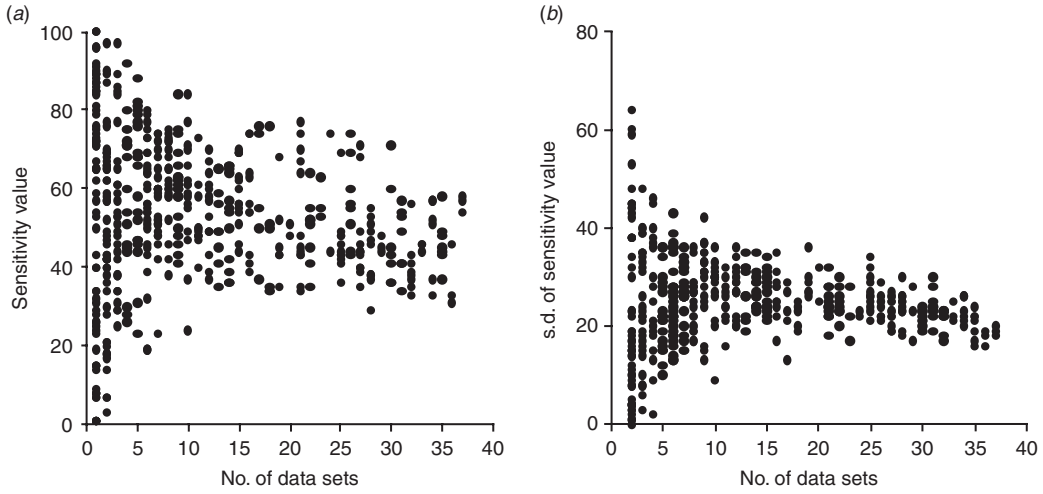

Fig. 3. Relationships between the number of datasets in which a diatom species occurred and (*a*) its derived sensitivity value and (*b*) the standard deviation (s.d.) of the sensitivity value.

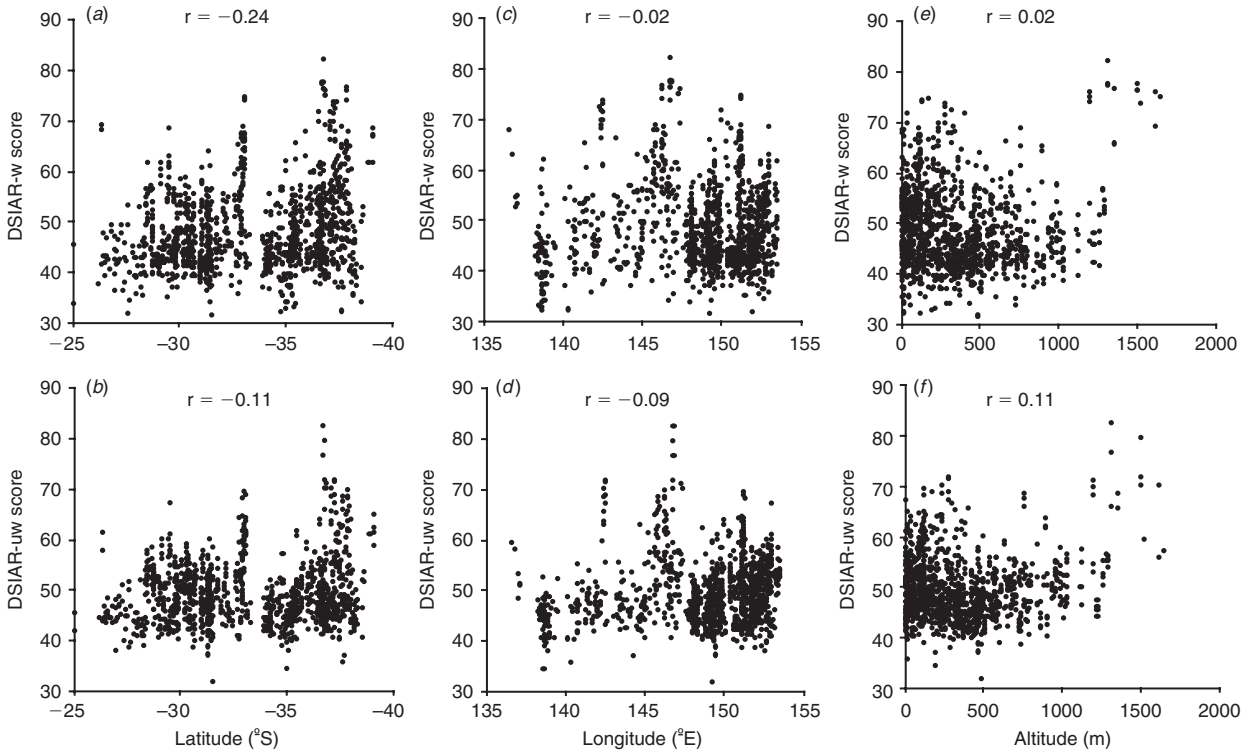

Fig. 4. Relationships of Diatom Species Index for Australian Rivers (DSIAR) scores for individual samples (weighted – w, and unweighted – uw) to the (*a*, *b*) latitude, (*c*, *d*) longitude and (*e*, *f*) altitude of sampling sites, with associated Pearson correlation coefficients.

and between riffles and glides ($P = 0.001$ in both cases). On average, scores were 1.5 units higher for runs than for pools ($n = 62$), and 2.7 units higher for riffles than for glides ($n = 30$).

Relationships of DSIAR scores to anthropogenic stressors

The number of diatom samples per ARC reach ranged from 1 to 54 and averaged 2.9; more than a third of the reaches (151) had only one sample. Average DSIAR scores (Fig. 7) were significantly correlated with the ARC_E index and with nine of its constituent indices and sub-indices (Table 2). These correlations were generally similar for the weighted and unweighted forms of DSIAR, but only the unweighted form was significantly correlated with the connectivity sub-index. Where both DSIAR and the ARC_B index based on macroinvertebrates were significantly correlated with an ARC_E index or sub-index, the Pearson correlation of DSIAR was usually about twice as great as that of

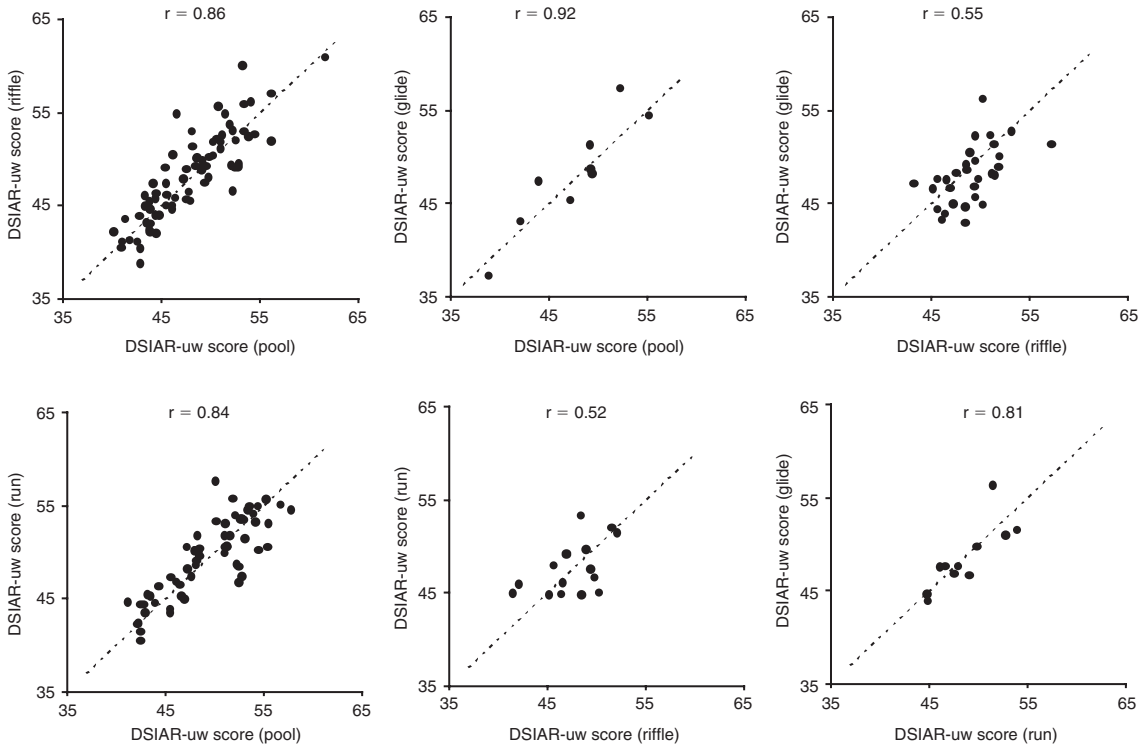

Fig. 5. Relationships between unweighted Diatom Species Index for Australian Rivers (DSIAR) scores for samples collected from rocks in different hydraulic environments at the same site and time, with associated Pearson correlation coefficients. Dotted lines indicate equality of scores from the habitats being compared.

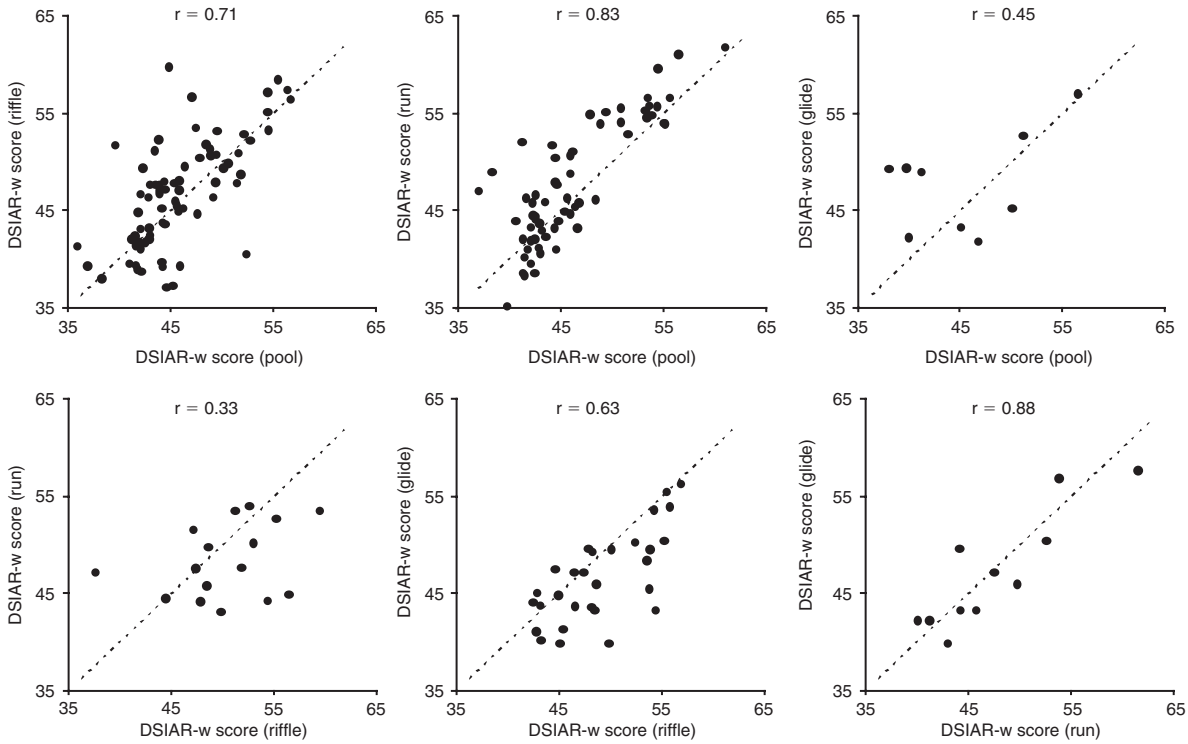

Fig. 6. Relationships between weighted Diatom Species Index for Australian Rivers (DSIAR) scores for samples collected from rocks in different hydraulic environments at the same site and time, with associated Pearson correlation coefficients. Dotted lines indicate equality of scores from the habitats being compared.

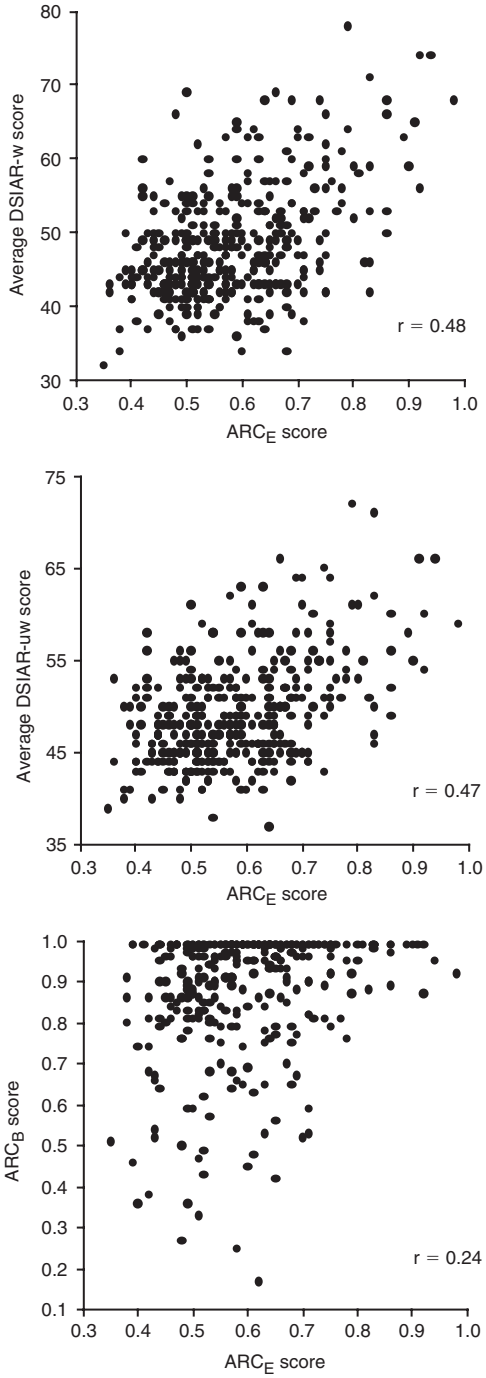

Fig. 7. Relationships of average Diatom Species Index for Australian Rivers (DSIAR) scores (weighted – w, and unweighted – uw) to ARC_E (Assessment of River Condition, Environment) index scores for ARC reaches, with associated Pearson correlation coefficients. For comparison, the relationship is shown between ARC_E scores and ARC_B (Assessment of River Condition, Biota) index scores based on macroinvertebrates and the Australian River Assessment System (AUSRIVAS).

ARC_B (Table 2). DSIAR was significantly correlated with three metrics with which ARC_B was not (the catchment disturbance index and the connectivity and riparian subindices) and conversely, the ARC_B was significantly correlated with three with which DSIAR was not (the infrastructure, land cover change and bedload condition sub-indices) (Table 2).

For the data from Newall and Walsh (2005) for streams in the eastern suburbs of Melbourne, DSIAR-uw was highly correlated with both drainage connection and effective imperviousness (Fig. 8; $P < 0.01$ in all cases). Patterns were similar for DSIAR-w, but weaker (Fig. 9). Thirty-six of 288 species and morphospecies in the Melbourne dataset had no assigned DSIAR grade, and so these had to be omitted from the calculations. However, these represented fewer than 4% of the total number of individuals.

Discussion

The strength of association between DSIAR and the ARC_E index and its constituent indices and sub-indices suggests that DSIAR can serve as a broad-scale indicator of anthropogenic stress related to catchment land use and associated nutrient enrichment of streams in south-eastern Australia. The maximum Pearson correlation coefficient of 0.50 between DSIAR and ARC_E components is within the range of significant correlation coefficients (0.29–0.75) reported for associations between diatom indices and the percentages of catchments covered by agricultural or urban land within individual states or ecoregions of the USA (Fitzpatrick *et al*. 2001; Fore and Grafe 2002; Wang *et al*. 2005). Aside from the sampling variability inherent in biological data, five main factors probably limit the broad-scale strength of association between a bioassessment metric like DSIAR and physically or chemically based indices of human disturbance. First, at large spatial scales, many anthropogenic stressors are likely to impinge on biological communities, and if bioassessment metrics are responding to a variety of stressors, which differ from place to place, a tight relationship between a biological metric and any particular physical or chemical index would not be expected. Second, the physical and chemical indices may not fully encapsulate the actual stressors that most influence biological communities. For example, the ARC_E nutrient sub-indices express modelled differences in long-term nitrogen and phosphorus loads between current and natural conditions. However, riverine diatom assemblages are likely to associate more strongly with baseflow nutrient concentrations than with long-term loads. Third, physical and chemical indices such as those in the ARC_E are limited by the data and modelling methods on which they are based. Fourth, if the physical and chemical indices express large-scale temporal and spatial averages (e.g. for ARC reaches up to 180 km long), they may not be a good reflection of small-scale and short-term environmental conditions at the places and times when biological samples are taken. Finally, the bioassessment metric may be affected by natural environmental gradients as well as by anthropogenic factors.

Our results also need to be seen in the context of broad-scale relationships between other bioassessment metrics and physical and chemical indices of human influence on Australian rivers. For example, a multimetric index based on fish had an R^2 value of only 0.10 for its linear relationship with an index of anthropogenic catchment disturbance at sites across NSW (Harris and Silviera 1999). The macroinvertebrate-based Australian River Assessment System (AUSRIVAS) has been widely used for broad-scale bioassessment of Australian rivers (e.g. Smith *et al*.

Table 2. Pearson coefficients of correlation between average DSIAR scores (weighted – w, and unweighted – uw) and ARC_E (Assessment of River Condition, Environment) index and sub-index scores for ARC reaches

For comparison, correlations are shown with ARC_B (Assessment of River Condition, Biota) index scores based on macroinvertebrates and the Australian River Assessment System (AUSRIVAS). The range of values is given for each index across all reaches with diatom data, together with the number of reaches with data available for each index (*n*). Only statistically significant correlation coefficients are listed: $*P < 0.05$; $**P < 0.01$, $***P < 0.001$

Index	Range	*n*	Correlation with:		
			DSIAR-w	DSIAR-uw	ARC_B
ARC_E	0.35–0.98	374	0.48***	0.47***	0.24***
Catchment disturbance	0.04–0.96	374	0.30***	0.31***	–
Infrastructure	0.46–0.98	374	–	–	−0.12*
Land use	0.58–1.00	374	0.50***	0.46***	0.18**
Land cover change	0.98–1.00	372	–	–	0.17**
Habitat	0.19–1.00	374	0.29***	0.33***	0.17**
Bedload condition	0.00–1.00	395	–	–	0.20***
Riparian	0.00–1.00	327	0.33***	0.34***	–
Connectivity	0.00–1.00	374	–	0.11*	–
Nutrient and suspended sediment load	0.10–1.00	374	0.43***	0.40***	0.25***
Suspended sediment	0.10–1.00	370	0.21***	0.19***	0.20***
Total P	0.19–1.00	374	0.44***	0.41***	0.23***
Total N	0.16–1.00	374	0.46***	0.42***	0.25***

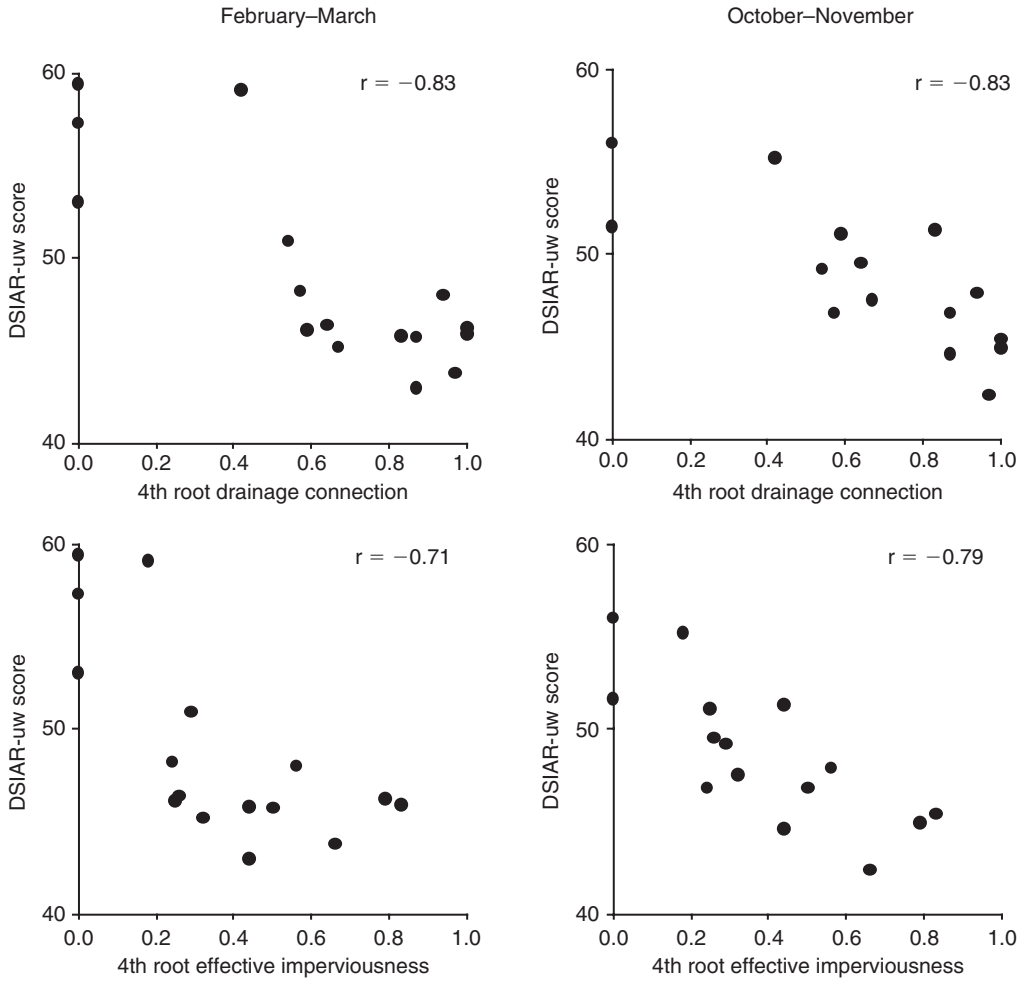

Fig. 8. Relationships of average Diatom Species Index for Australian Rivers (DSIAR) unweighted scores to drainage connection and effective imperviousness at 16 sites on streams in the eastern suburbs of Melbourne, with associated Pearson correlation coefficients (raw data from Newall and Walsh 2005).

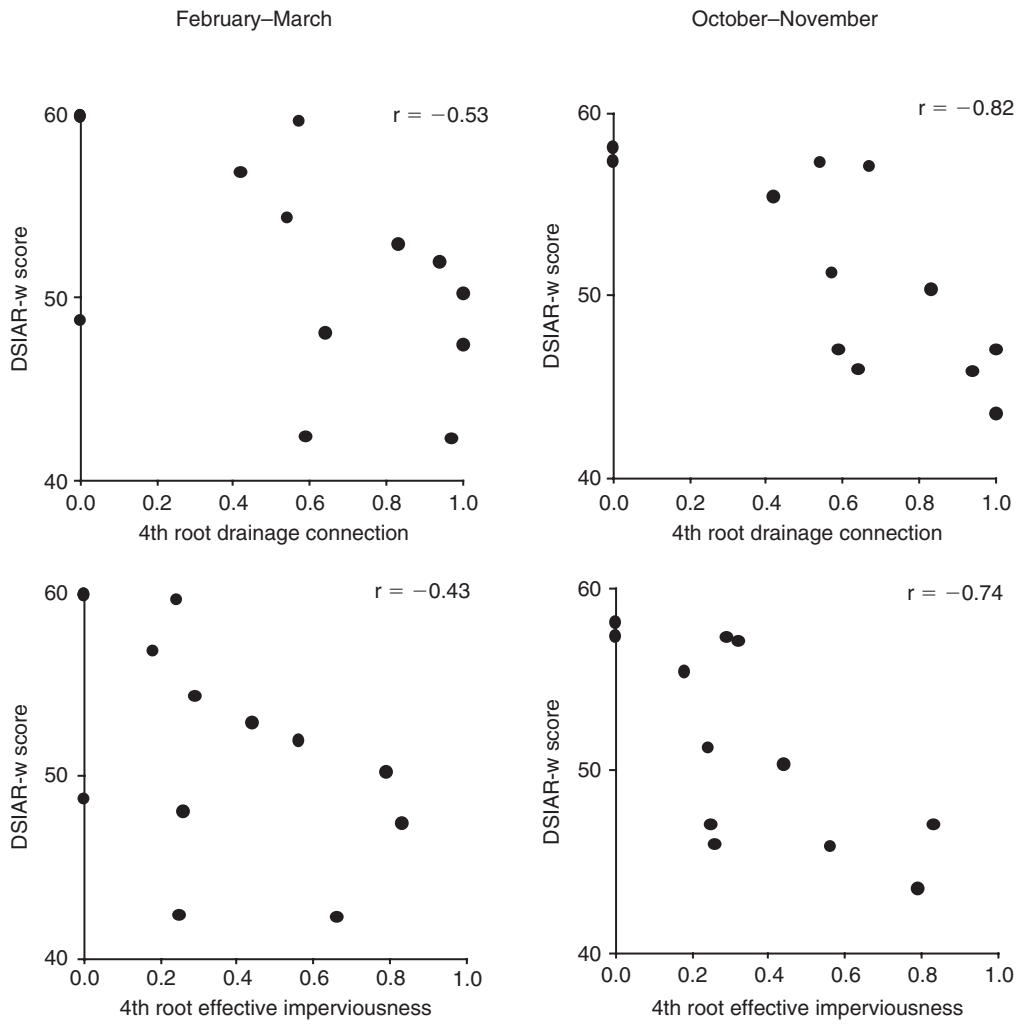

Fig. 9. Relationships of average Diatom Species Index for Australian Rivers (DSIAR) weighted scores to drainage connection and effective imperviousness at 16 sites on streams in the eastern suburbs of Melbourne, with associated Pearson correlation coefficients (raw data from Newall and Walsh 2005).

1999; Turak *et al*. 1999). Yet the ARC_B index, which is derived largely from AUSRIVAS assessments (Norris *et al*. 2001), had much weaker associations than DSIAR with ARC_E and most of its constituents, across the same set of ARC reaches. ARC_B was substantially less strongly correlated than DSIAR for eight of the ARC_E indices and sub-indices, and significantly more strongly correlated for only three (Table 2). Moreover, in one of these three instances, for the infrastructure sub-index, the correlation with ARC_B was negative, implying counter-intuitively that catchments with more human infrastructure such as roads have rivers in more natural biological condition. A notable feature of ARC_B was that it often attained the maximum possible value of 1 (implying no human impact) for reaches where ARC_E was below 0.5, implying substantial human impact. By contrast, DSIAR had very few high values for reaches with ARC_E <0.5 (Fig. 7).

The results for the streams in the eastern suburbs of Melbourne affected by urban development imply that DSIAR also has potential merit for more localised studies. Walsh (2006) noted that the effective imperviousness of catchments in this region is strongly associated with a wide range of physical, chemical, biochemical and biological impacts on stream ecosystems. Since the calculation of effective imperviousness is not a quick or simple task (see Walsh *et al*. 2004), a bioassessment metric such as DSIAR that is highly correlated with effective imperviousness and drainage connection has the potential to serve as a useful surrogate for prediction of ecological impact in streams affected by urban development. The unweighted version of DSIAR seems preferable to the version that was weighted according to the proportional abundances of the diatom species, because the unweighted version tended to be more strongly correlated with physical and chemical variables and less prone to differences between hydraulic habitats.

An advantage of diatoms for bioassessment of streams is that even with species-level identification, costs are low (Descy and Coste 1991; Stevenson and Pan 1999). The routine sampling methods for stream diatoms that are currently used by the senior author, which involve the collection of one composite

sample from flowing water and one from still water, require less than 15 min per site (B. Chessman, unpubl. data). By contrast, the AUSRIVAS protocols for sampling of macroinvertebrates require up to a hour of field or laboratory sub-sampling per sample, in addition to the time required to collect the bulk sample. However, species-level identification of diatoms does require considerable training and experience. Since genus-level assessments can provide adequate sensitivity in some circumstances (Kelly *et al*. 1995; Growns 1999; Hill *et al*. 2001; Wunsam *et al*. 2002), further development of genus-level diatom indices for Australian conditions could be beneficial, especially when the genus-level taxonomy eventually stabilises.

The development of methods to estimate natural, location-specific values of DSIAR might improve capacity to interpret the index via allowance for natural spatial and temporal variation in attainable scores. Although DSIAR scores did not seem to be greatly affected by macro-geographical position or within-site variation in hydraulic conditions, they might well respond more strongly to other natural environmental gradients. For example, the particularly high scores recorded for some high-altitude sites in eastern Victoria suggest influence by a regional factor such as catchment geology. Natural variation could be assessed by extensive sampling of reference sites with low levels of human disturbance, in regions where such sites still exist (cf. Chessman *et al*. 1999*b*). In regions where human disturbance is ubiquitous, other approaches may be needed (cf. Chessman and Royal 2004). It would also be useful to obtain further datasets for the derivation of sensitivity values, especially for the rarer species. The fact that the standard deviations of SVs were highest for species that occurred infrequently suggests that the mean SVs calculated for these species may be less reliable. However, because these species are rare, they have little influence on DSIAR scores in most cases.

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Appendix 1. Average sensitivity values (SVs) for diatom species

Standard deviations (s.d.) are given for all species present in more than one data set (Frequency > 1)

Species	Frequency	SV	s.d. of SV	Species	Frequency	SV	s.d. of SV
Achnanthes brevipes	7	51	32	*Caloneis schumanniana*	3	31	35
Achnanthes coarctata	3	57	35	*Caloneis silicula*	10	56	31
Achnanthes conspicua	9	69	33	*Caloneis undulata*	3	29	26
Achnanthes cotteriensis	2	47	15	*Cavinula pseudoscutiformis*	1	76	
Achnanthes exigua	31	41	28	*Chamaepinnularia bremensis*	13	45	32
Achnanthes grischuna	1	19		*Chamaepinnularia evanida*	1	96	
Achnanthes holsatica	1	1		*Chamaepinnularia soehrensis*	8	68	22
Achnanthes imperfecta	5	68	21	*Cocconeis pediculus*	9	46	33
Achnanthes impexa	5	88	10	*Cocconeis placentula*	36	33	16
Achnanthes inflata	2	43	44	*Cocconeis pseudothumensis*	7	48	27
Achnanthes kryophila	3	61	34	*Craticula accomoda*	12	57	36
Achnanthes lemmermannii	2	21	4	*Craticula buderi*	1	67	
Achnanthes nitidiformis	1	9		*Craticula cuspidata*	11	58	22
Achnanthes nodosa	4	80	20	*Craticula halophila*	25	48	23
Achnanthes oblongella	26	69	21	*Craticula halophilioides*	4	92	2
Achnanthes pericava	1	31		*Craticula molestiformis*	17	55	21
Achnanthes scotica	4	54	37	*Craticula riparia*	6	64	24
Achnanthes subexigua	10	70	21	*Ctenophora pulchella*	18	50	25
Achnanthidium biasolettianum	6	43	36	*Cyclostephanos dubius*	5	69	22
Achnanthidium minutissimum	37	57	19	*Cyclostephanos invisitatus*	1	8	
Actinella eunotioides	3	97	6	*Cyclostephanos tholiformis*	16	43	35
Actinocyclus normanii	9	63	42	*Cyclotella atomus*	7	52	32
Adlafia bryophila	14	66	24	*Cyclotella meneghiniana*	32	41	22
Adlafia minuscula	13	59	35	*Cyclotella pseudostelligera*	21	34	22
Amphipleura pellucida	2	18	23	*Cyclotella stelligera*	15	51	32
Amphora coffeaeformis	25	41	30	*Cymatopleura solea*	6	55	27
Amphora commutata	1	92		*Cymbella affinis*	12	58	27
Amphora delicatissima	7	60	36	*Cymbella aspera*	10	84	9
Amphora exigua	1	43		*Cymbella caespitosa*	1	26	
Amphora holsatica	6	32	13	*Cymbella cistula*	26	55	22
Amphora libyca	26	43	25	*Cymbella cymbiformis*	1	15	
Amphora montana	15	54	34	*Cymbella delicatula*	8	74	27
Amphora ovalis	4	46	45	*Cymbella ehrenbergii*	2	66	3
Amphora pediculus	36	31	19	*Cymbella falaisensis*	2	3	3
Amphora sabiniana	2	42	12	*Cymbella hantzschiana*	2	58	60
Amphora thumensis	3	84	8	*Cymbella helvetica*	9	63	16
Amphora veneta	29	44	23	*Cymbella laevis*	5	54	34
Aneumastus tusculus	6	52	35	*Cymbella naviculiformis*	2	42	38
Anomoeoneis sphaerophora	7	42	23	*Cymbella perpusilla*	2	38	32
Anorthoneis excentrica	3	34	32	*Cymbella subhelvetica*	1	32	
Asterionella formosa	7	59	29	*Cymbella tumida*	31	49	25
Asterionella ralfsii	2	40	22	*Denticula elegans*	1	50	
Aulacoseira alpigena	2	48	53	*Denticula subtilis*	3	38	40
Aulacoseira ambigua	4	75	21	*Denticula tenuis*	9	50	35
Aulacoseira crenulata	6	50	39	*Diadesmis confervacea*	19	45	27
Aulacoseira granulata	25	46	32	*Diadesmis contenta*	14	36	24
Aulacoseira italica	10	58	36	*Diadesmis gallica*	1	53	
Aulacoseira subarctica	4	28	15	*Diatoma moniliformis*	2	69	42
Bacillaria paxillifer	32	39	23	*Diatoma tenue*	22	55	30
Brachysira brebissonii	5	77	15	*Diatomella balfouriana*	12	49	29
Brachysira irawanoides	1	25		*Diploneis elliptica*	12	43	30
Brachysira neoexilis	6	77	28	*Diploneis ovalis*	9	48	31
Brachysira procera	2	65	38	*Diploneis parma*	22	52	28
Brachysira styriaca	17	76	23	*Diploneis pseudovalis*	2	89	5
Brachysira vitrea	21	70	25	*Diploneis smithii*	15	44	30
Caloneis aerophila	4	26	19	*Diploneis subovalis*	7	72	28
Caloneis bacillum	25	42	34	*Encyonema brehmii*	1	85	
Caloneis lauta	4	71	46	*Encyonema gracile*	30	71	24
Caloneis macedonica	1	96		*Encyonema mesiana*	8	65	28
Caloneis molaris	2	30	4	*Encyonema minuta*	34	52	26

(*Continued*)

Appendix 1. (Continued)

Species	Frequency	SV	s.d. of SV	Species	Frequency	SV	s.d. of SV
Encyonema muelleri	1	72		*Gomphonema acutiusculum*	6	60	23
Encyonema obtusum	1	34		*Gomphonema affine*	23	63	17
Encyonema silesiacum	34	57	22	*Gomphonema amoenum*	2	62	19
Encyonema tenuissimum	1	34		*Gomphonema angustum*	35	37	17
Encyonopsis amphicephala	11	73	24	*Gomphonema augur*	8	38	25
Encyonopsis cesatii	5	44	22	*Gomphonema bohemicum*	8	70	24
Encyonopsis microcephala	26	56	26	*Gomphonema clavatum*	32	56	23
Encyonopsis perborealis	12	49	29	*Gomphonema clevei*	14	66	26
Encyonopsis tynnii	1	71		*Gomphonema exilissimum*	6	61	18
Entomoneis alata	10	61	32	*Gomphonema gracile*	30	60	22
Entomoneis costata	13	49	31	*Gomphonema helveticum*	3	89	3
Eolimna minima	28	29	18	*Gomphonema insigne*	3	59	24
Eolimna subminuscula	25	36	25	*Gomphonema lagenula*	28	51	23
Epithemia adnata	30	43	23	*Gomphonema micropus*	19	68	27
Epithemia cistula	2	34	43	*Gomphonema minutum*	22	51	24
Epithemia sorex	31	41	19	*Gomphonema olivaceum*	7	49	26
Epithemia turgida	1	29		*Gomphonema parvulum*	37	54	18
Eunotia arcus	6	54	36	*Gomphonema productum*	5	80	15
Eunotia bilunaris	26	74	24	*Gomphonema pseudoaugur*	10	62	30
Eunotia camelus	1	57		*Gomphonema pseudotenellum*	8	58	29
Eunotia circumborealis	1	79		*Gomphonema subtile*	2	69	10
Eunotia exigua	16	74	25	*Gomphonema truncatum*	34	51	26
Eunotia faba	9	74	15	*Gomphonema utae*	1	90	
Eunotia fallax	5	68	23	*Gyrosigma acuminatum*	25	44	21
Eunotia formica	6	75	18	*Gyrosigma attenuatum*	16	54	31
Eunotia implicata	15	69	21	*Gyrosigma nodiferum*	5	47	28
Eunotia incisa	15	70	22	*Gyrosigma obscurum*	5	52	36
Eunotia intermedia	3	86	15	*Gyrosigma parkeri*	5	66	17
Eunotia minor	24	74	23	*Gyrosigma scalproides*	1	84	
Eunotia muscicola	9	84	13	*Gyrosigma spenceri*	12	53	31
Eunotia naeglii	7	63	33	*Gyrosigma strigilis*	2	36	44
Eunotia paludosa	10	66	30	*Hantzschia amphioxys*	23	55	22
Eunotia pectinalis	15	69	30	*Hantzschia distinctepunctata*	11	47	26
Eunotia praerupta	9	66	31	*Hantzschia virgata*	1	85	
Eunotia pseudoserra	1	100		*Haslea spicula*	15	60	31
Eunotia serpentina	21	74	24	*Hippodonta capitata*	27	46	25
Eunotia silvae	1	91		*Karayevia clevei*	27	35	25
Eunotia tecta	2	69	18	*Karayevia laterostrata*	6	80	28
Eunotia tenella	8	61	27	*Kobayasiella jaagii*	1	72	
Fallacia indifferens	6	19	21	*Kobayasiella subtilissima*	1	80	
Fallacia pygmaea	21	43	24	*Kolbesia kolbei*	5	81	19
Fallacia tenera	28	38	19	*Kolbesia ploenensis*	10	59	30
Fistulifera saprophila	2	14	10	*Lemnicola hungarica*	10	61	30
Fragilaria biceps	6	56	43	*Luticola cohnii*	1	7	
Fragilaria bidens	5	47	27	*Luticola goeppertiana*	22	35	23
Fragilaria capucina	37	58	20	*Luticola mutica*	26	60	29
Fragilaria crotonensis	8	75	17	*Luticola pseudokotschyi*	9	53	25
Fragilaria nanana	6	79	22	*Mastogloia elliptica*	16	56	28
Fragilaria tenera	17	74	13	*Mastogloia smithii*	17	50	29
Fragilariforma virescens	18	76	19	*Mayamaea agrestis*	12	39	32
Frustulia creuzbergensis	3	77	17	*Mayamaea atomus*	30	36	21
Frustulia disjuncta	1	100		*Mayamaea excelsa*	1	65	
Frustulia gaertnerae	1	72		*Mayamaea muraliformis*	6	64	19
Frustulia longinqua	1	100		*Melosira nummuloides*	2	28	38
Frustulia rhomboides	27	71	27	*Melosira varians*	35	48	24
Frustulia vulgaris	31	57	30	*Meridion circulare*	16	74	24
Frustulia weinholdii	1	87		*Microstatus maceria*	1	88	
Geissleria decussis	21	43	24	*Navicella pusilla*	16	64	28
Geissleria schoenfeldii	4	72	36	*Navicula absoluta*	3	48	8
Gomphoneis herculeana	3	25	18	*Navicula alineae*	1	8	
Gomphonema acuminatum	27	49	19	*Navicula angusta*	8	60	28

(*Continued*)

Appendix 1. (Continued)

Species	Frequency	SV	s.d. of SV	Species	Frequency	SV	s.d. of SV
Navicula bjoernoeyaensis	1	74		*Navicula trivialis*	12	67	22
Navicula breitenbuchii	1	75		*Navicula vandamii*	11	48	27
Navicula capitatoradiata	26	44	25	*Navicula veneta*	33	44	22
Navicula cari	12	60	20	*Navicula ventralis*	7	23	27
Navicula cincta	19	52	36	*Navicula viridula*	35	44	22
Navicula complanata	1	89		*Navicula wildii*		1	87
Navicula crucicula	7	53	27	*Naviculadicta difficillima*	14	65	29
Navicula cryptocephala	36	46	18	*Neidium affine*	10	49	33
Navicula cryptotenella	33	46	25	*Neidium ampliatum*	4	75	18
Navicula digitoradiata	2	40	34	*Neidium dubium*	1	62	
Navicula digitulus	1	65		*Nitzschia acicularioides*	2	57	45
Navicula duerrenbergiana	10	37	30	*Nitzschia acicularis*	29	45	28
Navicula eidrigiana	2	46	45	*Nitzschia acidoclinata*	20	51	32
Navicula erifuga	18	34	20	*Nitzschia acula*	7	65	26
Navicula expecta	5	79	16	*Nitzschia aequorea*	2	59	17
Navicula germanii	7	52	34	*Nitzschia agnita*	25	43	24
Navicula gottlandica	7	60	15	*Nitzschia alpina*	1	23	
Navicula gregaria	27	43	23	*Nitzschia amphibia*	27	44	19
Navicula heimansioides	25	69	26	*Nitzschia angustata*	2	65	16
Navicula hintzii	1	26		*Nitzschia angustatula*	7	60	31
Navicula incertata	11	40	16	*Nitzschia angustiforaminata*	8	65	35
Navicula kotschyi	9	58	28	*Nitzschia archibaldii*	27	40	26
Navicula lacustris	1	73		*Nitzschia aurariae*	3	42	33
Navicula lanceolata	25	48	29	*Nitzschia austriaca*	1	87	
Navicula laterostrata	3	65	19	*Nitzschia bacillum*	3	61	39
Navicula leptostriata	15	56	29	*Nitzschia capitellata*	28	55	24
Navicula libonensis	11	50	24	*Nitzschia clausii*	26	51	22
Navicula longicephala	2	70	30	*Nitzschia closterium*	1	40	
Navicula medioconvexa	1	8		*Nitzschia communis*	2	42	48
Navicula menisculoides	6	65	26	*Nitzschia commutata*	2	66	15
Navicula menisculus	28	37	25	*Nitzschia desertorum*	16	41	22
Navicula notha	6	66	20	*Nitzschia dissipata*	31	41	23
Navicula oligotraphenta	2	76	1	*Nitzschia diversa*	6	56	22
Navicula peregrina	8	59	27	*Nitzschia draveillensis*	2	77	2
Navicula perminuta	9	58	37	*Nitzschia dubia*	4	68	9
Navicula phyllepta	14	57	23	*Nitzschia elegantula*	15	46	24
Navicula porifera	1	7		*Nitzschia filiformis*	32	43	22
Navicula praeterita	6	69	16	*Nitzschia flexa*	5	31	29
Navicula pseudoventralis	3	49	21	*Nitzschia flexoides*	1	1	
Navicula radiosa	21	67	18	*Nitzschia fonticola*	29	48	17
Navicula radiosafallax	12	70	21	*Nitzschia fossilis*	9	41	30
Navicula recens	21	64	32	*Nitzschia frustulum*	32	37	17
Navicula reichardtiana	3	51	31	*Nitzschia fruticosa*	2	62	38
Navicula rhynchocephala	27	68	23	*Nitzschia gessneri*	7	70	15
Navicula rostellata	9	43	26	*Nitzschia graciliformis*	16	54	25
Navicula salinarum	8	46	28	*Nitzschia gracilis*	34	57	21
Navicula salinicola	2	42	45	*Nitzschia hantzschiana*	9	62	31
Navicula schmassmannii	6	54	31	*Nitzschia homburgensis*	2	42	17
Navicula schroeteri	32	35	23	*Nitzschia hybrida*	1	1	
Navicula silicula	3	53	39	*Nitzschia incognita*	8	42	20
Navicula slesvicensis	1	89		*Nitzschia inconspicua*	34	37	23
Navicula splendicula	7	60	17	*Nitzschia intermedia*	20	48	25
Navicula striolata	1	1		*Nitzschia lacuum*	24	44	22
Navicula stroemii	2	57	9	*Nitzschia liebetruthii*	30	36	19
Navicula submuralis	12	49	34	*Nitzschia linearis*	35	45	21
Navicula subrhynchocephala	7	58	20	*Nitzschia lorenziana*	11	48	28
Navicula subrotundata	2	67	0	*Nitzschia microcephala*	30	45	20
Navicula symmetrica	1	40		*Nitzschia modesta*	1	15	
Navicula tenelloides	25	43	26	*Nitzschia nana*	14	54	30
Navicula tridentula	2	20	14	*Nitzschia obtusa*	9	75	24
Navicula tripunctata	6	39	27	*Nitzschia ovalis*	1	14	

(*Continued*)

Appendix 1. (Continued)

Species	Frequency	SV	s.d. of SV	Species	Frequency	SV	s.d. of SV
Nitzschia palea	35	49	22	*Planothidium engelbrechtii*	3	34	23
Nitzschia paleacea	31	54	22	*Planothidium frequentissimum*	31	34	24
Nitzschia paleaeformis	18	51	23	*Planothidium granum*	22	45	23
Nitzschia perminuta	27	44	22	*Planothidium lanceolatum*	28	49	26
Nitzschia perspicua	1	28		*Pleurosigma angulatum*	2	7	8
Nitzschia prolongata	3	54	35	*Pleurosigma elongatum*	12	37	20
Nitzschia pseudofonticola	6	45	37	*Pleurosigma nodiferum*	5	76	21
Nitzschia pumila	15	62	25	*Pleurosigma salinarum*	6	19	14
Nitzschia pura	8	62	36	*Psammothidium abundans*	10	77	20
Nitzschia pusilla	19	35	26	*Psammothidium bioretii*	3	46	40
Nitzschia radicula	13	41	26	*Psammothidium helveticum*	11	50	32
Nitzschia recta	28	38	23	*Psammothidium marginulatum*	2	97	4
Nitzschia reversa	15	56	24	*Psammothidium sacculum*	14	64	27
Nitzschia rosenstockii	4	51	30	*Psammothidium subatomoides*	13	65	22
Nitzschia sigma	21	48	28	*Pseudostaurosira brevistriata*	28	46	26
Nitzschia sigmoidea	9	60	37	*Pseudostaurosira zeilleri*	14	42	24
Nitzschia sinuata	14	55	35	*Reimeria sinuata*	21	44	28
Nitzschia sociabilis	13	35	21	*Rhoicosphenia abbreviata*	34	33	20
Nitzschia solita	14	50	26	*Rhopalodia acuminata*	2	21	15
Nitzschia subacicularis	15	63	27	*Rhopalodia brebissonii*	25	49	27
Nitzschia subcapitellata	6	46	36	*Rhopalodia constricta*	7	51	32
Nitzschia sublinearis	1	71		*Rhopalodia gibba*	33	46	21
Nitzschia suchlandtii	8	60	19	*Rhopalodia musculus*	23	53	28
Nitzschia supralitorea	19	43	30	*Rossithidium linearis*	8	72	20
Nitzschia tropica	6	51	28	*Rossithidium petersenii*	4	60	25
Nitzschia tubicola	18	35	24	*Rossithidium pusillum*	22	64	20
Nitzschia umbonata	4	45	38	*Sellaphora bacillum*	3	38	14
Nitzschia valdecostata	13	56	32	*Sellaphora laevissima*	2	49	59
Nitzschia valdestriata	13	49	19	*Sellaphora pupula*	31	53	24
Nitzschia vermicularis	11	49	22	*Sellaphora seminulum*	32	33	18
Nitzschia wuellerstorffii	1	65		*Sellaphora vitabunda*	7	48	28
Nupela lapidosa	3	29	22	*Simonsenia delognei*	2	87	4
Nupela rumrichorum	1	59		*Stauroneis anceps*	16	39	17
Nupela tristis	1	89		*Stauroneis kriegeri*	7	60	26
Opephora olsenii	8	58	22	*Stauroneis obtusa*	3	44	10
Pinnularia appendiculata	12	64	27	*Stauroneis pachycephala*	6	73	17
Pinnularia borealis	13	65	26	*Stauroneis phoenicenteron*	3	53	48
Pinnularia braunii	7	74	26	*Stauroneis producta*	1	24	
Pinnularia decrescens	1	81		*Stauroneis smithii*	2	17	19
Pinnularia divergens	2	52	64	*Staurophora salina*	2	18	23
Pinnularia divergentissima	4	71	40	*Staurophora wislouchii*	2	68	11
Pinnularia gibba	16	63	29	*Staurosira construens*	30	41	23
Pinnularia intermedia	3	85	16	*Staurosira elliptica*	27	44	22
Pinnularia interrupta	9	60	31	*Staurosirella leptostauron*	10	55	29
Pinnularia legumen	7	46	33	*Staurosirella pinnata*	35	43	19
Pinnularia mesolepta	1	86		*Stenopterobia curvula*	11	61	30
Pinnularia microstauron	6	73	13	*Stenopterobia densestriata*	2	90	14
Pinnularia obtusa	8	72	19	*Stephanodiscus hantzschii*	17	37	27
Pinnularia similis	5	58	30	*Stephanodiscus niagarae*	2	7	8
Pinnularia subcapitata	22	52	25	*Stephanodiscus parvus*	10	24	30
Pinnularia viridiformis	7	68	26	*Surirella amphioxys*	1	85	
Pinnularia viridula	6	69	25	*Surirella angusta*	28	53	25
Placoneis clementis	15	55	28	*Surirella biseriata*	5	82	12
Placoneis constans	11	48	21	*Surirella bohemica*	1	15	
Placoneis elginensis	27	39	26	*Surirella brebissonii*	21	45	22
Placoneis exigua	5	23	22	*Surirella elegans*	9	61	35
Placoneis hambergii	2	43	1	*Surirella linearis*	8	73	17
Placoneis placentula	4	44	28	*Surirella minuta*	10	71	26
Placoneis pseudanglica	3	67	22	*Surirella ovalis*	12	61	27
Planothidium daui	4	30	13	*Surirella patella*	7	74	18
Planothidium delicatulum	32	38	22	*Surirella robusta*	10	47	27

(*Continued*)

Appendix 1. (Continued)

Species	Frequency	SV	s.d. of SV	Species	Frequency	SV	s.d. of SV
Synedra acus	28	47	30	*Tryblionella calida*	21	35	26
Synedra parasitica	15	69	23	*Tryblionella debilis*	14	36	31
Synedra ulna	35	58	16	*Tryblionella gracilis*	1	54	
Tabellaria flocculosa	21	77	23	*Tryblionella hungarica*	19	43	29
Tabularia fasciculata	27	40	20	*Tryblionella levidensis*	5	46	36
Thalassiosira weissflogii	14	39	29	*Tryblionella littoralis*	8	52	29
Tryblionella aerophila	1	1		*Tryblionella punctata*	1	1	
Tryblionella apiculata	22	41	26				