THE USE OF SCALED CHRYSOPHYTES IN LONG TERM MONITORING PROGRAMS FOR THE DETECTION OF CHANGES IN LAKEWATER ACIDITY

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(Received May 8, 1991; accepted January 20, 1993)

Abstract. The addition of a biomonitoring component to a routine chemical monitoring program would enhance the potential of the monitoring effort to measure shifts in lakewater acidity. We provide evidence that changes in the species composition of scaled chrysophytes can provide an effective means for identifying and quantifying changes in lakewater acidity. As a result, it is consluded that a scaled chrysophyte component would provide an excellent addition to a long term monitoring effort. Specific and subspecific taxa of scaled chrysophytes are differentially distributed along a pH gradient. The distributions along a pH gradient of many of the well defined and cosmopolitan taxa are summarized and the taxa arranged into different pH groups. The importance of pH, relative to other variables, in controlling the distribution of scaled chrysophytes is discussed utilizing evidence from both neolimnological and paleolimnological studies. A plan for the incorporation of a biomonitoring component utilizing scaled chrysophytes into a long term monitoring program for acidity is outlined. The plan includes analyses using discrete water samples, sediment traps and sediment cores. Several programs are outlined and discussed based on the frequancy of sampling events.

1. Introduction

It is well know that shifts in lakewater acidity can result in measurable changes in the resident flora and fauna of a given waterbody (Almer *et al.*, 1974; Beamish, 1976; Hendrey *et al.*, 1976; Hendrey and Wright, 1976; Schofield, 1976; Yan and Stokes, 1978; Conway and Hendrey, 1982; Havens *et al.*, 1986). Routine chemical monitoring programs do not necessarily provide the best means of estimating the effects of changes in acidity on aquatic ecosystems. This is especially true of monitoring programs that consist of a small number of samples per year; in this case valuable information on lakewater acidity may be missed. For example, episodic acidification events that occur between sampling visits may go undetected in a chemical monitoring program. Likewise, although the mean pH of a waterbody may vary significantly from one year to the next, the extent of the variation may not be detected with a few measurements of pH. Daily or weekly pH fluctuations, caused by storm events, spring snow melt or a change in primary productivity, may also result in erroneous conclusions.

Aquatic organisms experience daily and long term fluctuations in acitity. As a

Water, Air, and Soil Pollution **71**: 357–376, 1993.

result, species composition is constantly molded by lakewater pH. Thus, if the alterations in the species composition caused by shifts in pH could be identified and quantified, they would add significantly to our understanding of the effects on the ecosystem. The question becomes, 'which group(s) of organisms best reflect both short term oscillations and long term pH shifts?'.

The predominance of the Chrysophyceae in dilute, poorly buffered, oligotrophic lakes is a characteristic common to many lake regions (Willen, 1969; Hutchinson, 1967; Schindler and Holmgren, 1971; Schindler and Nighswander, 1970; Kling and Holmgren, 1972; DeNoyelles and O'Brien, 1978; Findlay, 1978; Conway and Hendrey, 1982; Siver and Chock, 1986; Sandgren, 1988). Not only do the Chrysophyceae often dominate the number of species in oligotrophic lakes, but they usually comprise a maximum percantage of the biomass as well (Schindler and Holmgren, 1971; Kling and Holmgren, 1972; DeNoyelles and O'Brien, 1978; Conway and Hendrey, 1982; Wetzel, 1983; Sandgren, 1988). Species of Chrysophyceae with siliceous scales are common and often abundant in oligotrophic lakes (Takahashi, 1978; Kristiansen, 1986; Roijackers and Kessels, 1986).

Shifts in the relative importance of each algal class represented in the phytoplankton are well documented with increasing lakewater acidity. The greatest changes in the composition of phytoplankton communities are typically found between a pH of 5 and 6 (Almer *et al.*, 1974; Almer *et al.*, 1978; Yan and Stokes, 1978; Hörnström *et al.*, 1973; Smol, 1986). As a general rule, the Chrysophyceae, Dinophyceae and sometimes the Cryptophyceae increase in importance as the lakewater pH drops below 6.0 (Almer *et al.*, 1978; Yan and Stokes, 1978; Hendrey *et al.*, 1981; Conway and Hendrey, 1982). Planktonic diatoms and blue-green algae often becomes less important, especially below pH 5 (Almer *et al.*, 1978; Battarbee, 1984; Smol, 1986; Charles, 1985; Davis and Smol, 1986). Since the Chrysophyceae are an integral component of the phytoplankton floras of poorly buffered oligotrophic lakes, and can remain important below pH 5 to 5.5, they should be considered in a pH monitoring program.

In this paper we review the evidence that scaled chrysophytes are linked directly to lakewater pH, that many well defined species have definitive distributions along a pH gradient, and that species composition can be used to signal both short and long term shifts in lakewater acidity. In effect, we present evidence that changes in the composition of scaled chrysophytes are excellent biological indicators of pH.

We will restrict our discussion to species with siliceous scales classified in the Paraphysomonadaceae (Preisig and Hibberd, 1983) and Synurophyceae (Andersen, 1987), and refer to them as scaled chrysophytes. We will also rely on studies where electron microscopy was used to identify species. Because scaled chrysophytes are euplanktonic organisms, all of our comments relate to lakes.

2. The Importance of pH in Controlling the Distribution of Scaled Chrysophytes

Kristiansen (1986) listed three prerequisites an organism must fulfill to be a useful biological indicator. First, it must be taxonomically well defined. Second, the organism must be easily identified and not be confused with other taxa. Third, it must be distributed within a 'narrow ecological spectrum'. In this context, the occurrence of the organism must be controlled by the environmental factor in question, in this case, pH. Thus, the relative importance of lakewater acidity in governing the distribution of a given species must be known prior to its use as an indicator of lakewater pH.

There are many qualitative remarks in the literature relating the distribution of species of scaled chrysophytes to lakewater pH. Many of the statements are observations such as 'the organism was found in an acidic lake' or 'or was collected in a bog', and are not accompanied by actual pH readings or any additional environmental data (e.g. temperature). Although such remarks will be valuable in supporting hypotheses characterizing the distributions of individual taxa along a pH gradient, in themselves they are of little importance in defining criteria for establishing a long term program to monitor shifts in pH.

An important milestone in the study of scaled chrysophytes was the use of electron microscopy as a tool for identifying organisms by virtue of their siliceous scales and bristles (Asmund and Kristiansen, 1986). With electron microscopy a precise and unequivocal means for identifying specific and subspecific taxa of scaled chrypsophytes has evolved (e.g Takahashi, 1978; Wee, 1982; Nicholls, 1982; Asmund and Kristiansen, 1986; Siver, 1991a). As a result, most species fulfil the first two prerequisites listed above.

Many studies utilizing electron microscopy have provided records that can be used directly or indirectly to describe the distributions of taxa along environmental gradients (see Kristiansen, 1986; Roijackers and Kessels, 1986; Smol *et al.*, 1984; Siver and Hamer, 1989; Siver, 1989a. 1991a for a review). A few such studies are of particular relevance because they provided valuable insights into the importance of lakewater pH in controlling the distribution of scaled chrysophytes.

In a detailed seasonal study of scaled chrysophytes in three ponds, Takahashi (1967) concluded that pH was the most important factor controlling the distribution of individual species. Kristiansen (1975) provided pH and temperature frequency distribution records for species of *Synura* and concluded that some taxa were more commonly found in acidic (e.g. *S. sphagnicola*) or alkaline (e.g. *S. uvella*) waters, whereas others (e.g. *S. petersenii*) were indifferent to pH. In a similar fashion Takahashi (1978) arranged 33 widely distributed species, into the pH categories of Hustedt (1939) and concluded that the distribution of many species were restricted along a pH gradient. Ito and Takahashi (1982) reported that the maximum development of *Spiniferomonas* species occurred between pH 6 to 7. Kristiansen (1985, 1986, 1988) reviewed the occurrence of species in alkaline habitats with a pH above 8.

The above cited works provided evidence for the role of pH in controlling the distributions of scaled chrysophyte species. However, more recently, a number of studies have utilized multivariate statistical procedures to document the relative importance of pH as compared to other parameters such as temperature, nutrient levels, dissolved organic carbon and heavy metals. The studies can be divided into two categories; those relating extant populations to lakewater conditions measured at the time of collection (Roijackers and Kessels, 1986; Siver and Hamer, 1989) and those where surface sediment remains were correlated with present day lakewater characteristics (Smol *et al.*, 1984; Dixit *et al.*, 1988, 1989, 1990; Cumming *et al.*, 1991). In all of the investigations, lakewater pH was shown to be a primary factor governing the occurrence of scaled chrysophytes.

In a study of 50 waterbodies from the Netherlands relating the distributions of scaled chrysophytes to lakewater pH, alkalinity and temperature, Roijackers and Kessels (1986) found that the first principal component was dominated by pH and accounted for 57% of the total variance in the dataset. The second principal component accounted for an additional 35% of the variance and was correlated most closely with water temperature. Roijackers and Kessels (1986) concluded that the lakewater pH controlled the presence of scale-bearing taxa in a given waterbody, whereas the water temperature controlled the resultant biomass. In a study of 62 lakes Siver and Hamer (1989) used principal component analysis to address the relative importance of pH and other limnological parameters in controlling the occurrence of scaled chrysophytes as a group, as well as for individual genera and species. The lakewater pH or pH in combination with specific conductance, was found to strongly dominate the first principal component for each taxonomic unit, and was instrumental in controlling the occurrence of 23 of the 25 taxa tested. Water temperature and total phosphorus levels were believed to be more important in determining when, and to what degree, a species would develop in a given waterbody.

Smol *et al.* (1984) examined the distribution of scaled chrysophytes from the surface sediments of 38 lakes in relation to 17 limnological characteristics and found that the first reciprocal averaging axis was highly correlated with pH ($r = 0.79$) and pH-related parameters (e.g. log alkalinity, pCa and pMg). Correlations of the first reciprocol averaging axis with the average lake depth, summer epilimnion temperature, elevation, nitrate level and aluminum concentration were statistically significant; however, these factors explained lesser percentages of the total variance. The conductance, water color, Secchi disc depth, total phosphorus, chlorophyll *a*, sulfate and silica levels were not significantly correlated with the first reciprocal averaging axis. In similar studies from Ontario (Dixit *et al.*, 1988, 1989), northern New England (Dixit *et al.*, 1990) and Norway (Cumming *et al.*, 1991), lakewater pH was also documented as an important variable controlling the distribution of scaled chrysophytes.

The effect(s) that factors that covary with pH may play in the distribution of scaled chrysophyte taxa are unknown. It is possible that one or more factors covarying

with pH may exert equal or greater influence on the distribution of a given species than pH. If this is true, the pH inference would be correct only if the correlation between the covarying factor(s) and the pH remained constant (Davis and Smol, 1986; Siver and Hamer, 1989).

The above cited works, representing critical quantitative studies from different geographic regions clearly support the idea that scaled chrysphytes will be valuable candidates for use in pH biomonitoring programs.

3. What Are the Most Valuable Scaled Chrysophyte Taxa for Use in Monitoring the Effects of Acid Deposition?

Many species of scaled chrysophytes have definitive and well documented distributions along a continuum of pH (Siver, 1989a). As a result, assemblages of taxa can be identified and subsequently used to monitor pH. For discussion purposes we divide taxa into four groups. Group 1, the low pH group, consists of *Synura sphagnicola, S. echinulata, Mallomonas canina, M. hindonii, M. pugio, M. paludosa, M. acaroides* v. *muskokana* and *M. hamata*. Taxa in this group are found predominantly in acidic waters (Smol *et al.*, 1984; Roijackers and Kessels, 1986; Siver, 1989a,b; Siver and Hamer, 1989; Charles and Smol, 1988; Dixit, 1986; Dixit *et al.*, 1988). The maximum frequency of occurrence and weighted mean pH values for species within this group are consistently reported to be below pH 6.0. *Mallomonas canina, M. hindonii* and *M. paludosa*, each with a weighted mean pH below 5.1 (Siver, 1989a), are probably the best representatives of a category termed 'strict acidobionts'. *Mallomonas acaroides* v. *muskokana* and *S. sphagnicola* are more commonly encountered and are distributed over a wider range of pH, however, their frequency of occurrence also drops significantly above a pH of 5.5 to 6.0 (e.g. Roijackers and Kessels, 1986; Siver, 1988a, 1989a,b; Charles and Smol, 1988; Dixit *et al.*, 1988). Although Dixit *et al.* (1988) found *M. acaroides* v. *muskokana* primarily in waterbodies from the Sudbury, Ontario region between a pH of 5 and 6, this organism has often been reported in floras of Adirondack lakes below pH 5.0 (Siver, 1988a, 1989b; Charles and Smol, 1988).

Synura sphagnicola has been observed in both clearwater and humic stained (bog) acid lakes (Dürrschmidt, 1980; Kristiansen, 1975; Smol *et al.*, 1984; Roijackers and Kessels, 1986; Siver, 1987, 1988a). Because it tends to be more abundant in acid bog habitats, Dixit *et al.* (1988) suggested that *S. sphagnicola* may become a useful indicator of organic vs. mineral acid inputs. In a similar but opposite manner a change in the abundance of *M. hamata*, a species reported to be lacking from humic habitats (Smol *et al.*, 1984; Siver, 1988a; Dixit, 1986), also may indicate the origin of acidification in a lake. In addition, *M. hamata* appears to be sensitive to elevated metal concentrations (Dixit *et al.*, 1988).

Of the taxa within the low pH group, *Synura echinulata* has been reported over the largest pH range; it has been observed, although rarely, above pH 8 (Kristiansen, 1975, 1988). Nonetheless, *S. echinulata* is most often found in oligotrophic waters,

acidic in nature and low in alkalinity (Kristiansen, 1975; Smol *et al.*, 1984; Siver, 1988a; Roijackers and Kessels, 1986).

With further study, other rarer taxa, such as *Spiniferomonas crucigera* Takahashi and *Chrysodidymus synuroideus* Prouse, may be found to belong in the low pH group. Based on available records *Spiniferomonas crucigera* is restricted to acidic localities with a pH range of 4.8 to 6.7 (Kristiansen, 1978; Ito and Takahashi, 1982; Eloranta, 1985; Siver, 1988a) and has a weighted mean pH of 5.3. *Spiniferomonas abei* has often been reported from acidic habitats below a pH of 7 (Kristiansen, 1978; Nicholls, 1981; Skogstad, 1982; Ito and Takahashi, 1982). Charles and Smol (1988) reported *C. synuroideus* in surface sediments from 51% of the lakes they surveyed in the Adirondacks and found it to be an acidibiontic taxon with a weighted mean pH of 5.49. Dixit (1986) and Dixit *et al.* (1988) also reported *C. synuroideus* in surface sediments from Sudbury lakes as an acidobiont. However, Siver (1988a) did not record this species in live samples from the Adirondacks and records from the literature suggest a higher mean pH (Dürrschmidt, 1982). Thus, although *C. synuroideus* is a probable acidobiont, further work is needed to verify the extent of its distribution along a pH gradient. *Synura mammillosa* (Takahashi, 1978; Roijackers and Kessels, 1986) and *Synura macracantha* (Charles and Smol, 1988) are two additional taxa with affinities for low acidic waters; however, they are rarely reported and may be confused with other species (Siver, 1989a).

Group 2 consists of taxa primarily reported from acidic softwater localities above a pH of 5 to 5.5; the frequency of occurrence of these taxa drops significantly as the pH shifts below the 5 to 5.5 interval or rises above 7.0 (Siver, 1989a). Species in this group, herein referred to as the 'mid pH group', include *Mallomonas punctifera, M. galeiformis, M. heterospina, Spiniferomonas bourrellyi, S. serrata, Synura spinosa* and *Chrysosphaerella longispina*. Except for *M. punctifera*, the weighted mean pH values for these taxa are most often reported between a pH of 5.9 and 6.5 (Siver, 1989a). A change in the abundance of organisms within the mid pH group would most likely signal a shift in lakewater acidity within the critical range of pH 5 to 6.5.

In Connecticut (Siver, 1989a) and Adirondack (Smol *et al.*, 1984; Charles and Smol, 1988; Siver, 1988a) waterbodies, *M. punctifera* was restricted to a narrow pH range of 5 to 7. However, based on literature records its range extends above pH 8.0 (Kristiansen, 1986) and it has been reported to have a weighted mean pH above 7.0 (Dixit *et al.*, 1988). Charles and Smol (1988) and Siver (1991a) suggested the possibility that different ecotypes or underscribed varieties may exist. Such an idea is supported by the fact that *M. punctifera* has been reported as both a warm (Siver, 1989a) and cold (Roijackers and Kessels, 1986) water species.

Initial work suggests that a host of rarer taxa may be correctly placed in the mid pH group. *Mallomonas transsylvanica* Péterfi and Momeu and *M. calceolus* Bradley are primarily restricted to a pH range of 5 to 7 (Bradley, 1964; Kristiansen, 1978; Cronberg and Kristiansen, 1980; Dürrschmidt, 1982; Kling and Kristiansen, 1983) and would disappear below pH 5. *Mallomonas allantoides, M. flora, M. cor-*

contica, M. papillosa, M. doignonii, M. allorgei and *M. lychenensis* also appear to have affinities for acidic localities (see Siver, 1989a and references therein).

Group 3, the pH indifferent group, consists of *Mallomonas akrokomos, M. crassisquama, M. caudata, Synura petersenii, Synura uvella, Synura curtispina, Synura mollispina, Spiniferomonas trioralis, Spiniferomonas bilacunosa, Spiniferomonas coronacircumspina, Spiniferomonas cornuta* and *Chrysosphaerelle brevispina*. Taxa in this group have their center of distribution and weighted mean pH around pH 7 (Siver. 1989a; Wee and Gabel, 1989). Some of the most commonly observed and widespread species of scaled chrysophytes, such as *S. petersenii, M. caudata, M. crassisquama* and *S. trioralis*, are included within this group; these species are found over a wide range of pH. A few of the species, such as *S. coronacircumspina, S. cornuta* and *S. petersenii* have been recorded from acid environments (pH < 5.0), however, the remaining taxa in this group disappear below the 5 to 5.5 pH interval (Siver, 1989a); this is a characteristic Group 3 shares with Group 2.

It is well documented that the occurrence of *Mallomonas crassisquama* drops significantly as the pH lowers below 5 to 5.5 (Siver and Skogstad, 1988; Smol *et al.*, 1984; Roijackers and Kessels, 1986; Hartmann and Steinberg, 1986). Dixit *et al.* (1988), however, reported populations of *M. crassisquama* from surface sediments of lakes with a pH below 5. Several studies have made reference to a form of *M. crassisquama* with large, heavily silicified scales that had a striking resemblance to wingless scales of *Mallomonas pseudocoronata* (Siver and Skogstad, 1988; Jacobsen, 1985; Kling and Kristiansen, 1983). It is now known that this latter form is a separate species, *Mallomonas duerrschmidtiae* Siver, Hamer and Kling, that has a weighted mean pH of 5.76, and is primarily distributed below pH 7 (Siver *et al.*, 1990). The presence of *Mallomonas duerrschmidtiae* may account for the reports of *M. crassisquama* in waters below pH 5.

Group 4, the high pH group, consists of taxa with their maximum distributions and weighted mean pH above 7.0, and that are rarely reported below pH 6.5 (Siver, 1989a; Wee and Gabel, 1989). This group consists of *Mallomonas corymbosa, M. pseudocoronata, M. alpina, M. tonsurata, M. acaroides* v. *acaroides* and *M. portaeferreae. Mallomonas elongata* is an additional species that is often reported from alkaline waters (Asmund, 1959; Asmund and Hilliard, 1961; Wujek *et al.*, 1977; Roijackers and Kessels, 1986; Smol *et al.*, 1984), and has a weighted mean pH close to or above 7 (Siver, 1989a).

It is clear that since many species of scaled chrysophytes are distributed over a narrow pH range, they would be excellent bioindicators. There are groups of taxa found along the entire natural pH spectrum, thus offering continuous 'tools' for monitoring and identifying shifts in lakewater pH. Even though some lake regions in North America have not been thoroughly surveyed for chrysophytes, based on available data, it is believed that each potentially valuable indicator species discussed in this paper is distributed throughout North America; most have world-wide disributions. A good example is the small State of Connecticut, where all 15 worldwide species of the genus *Spiniferomonas* have been found (Siver, 1988b,c). The frequencies

of occurrence, as represented from literature records, for taxa referred to in this paper have been presented elsewhere (Siver, 1989a).

4. Can Scaled Chrysophytes Signal Shifts in Lakewater pH?

In this section we provide evidence from studies of extant and fossil populations of scaled chrysophytes documenting their value as both qualitative and quantitative indicators of changes in lakewater pH over short (months) and long (years) time frames.

In recent years there has been a great emphasis placed on analyzing stratigraphic profiles of siliceous microalgal populations from lake sediments in order to reconstruct historical lakewater conditions (e.g. Bradbury, 1975; Battarbee, 1979; Munch, 1980; Davis *et al.*, 1983; Davis and Anderson, 1985; Charles and Norton, 1986; Smol *et al.*, 1986; Charles *et al.*, 1989). Much of this research has focused on the reconstruction of past pH conditions in order to establish trends in changes in lakewater acidification caused by naturally occurring events, disturbances to the watershed or acid deposition (Davis and Smol, 1986). Although most studies have utilized diatoms, scaled chrysophytes have become an integral part of such paleolimnological efforts (Smol, 1986; Davis and Smol, 1986; Charles and Smol, 1988; Dixit *et al.*, 1989, 1990; Cumming *et al.*, 1991). As a consequence of the paleolimnological research, a rich volume of ecological data for many scaled chrysophyte taxa, especially species dominating poorly buffered lakes, has been assembled (see Smol, 1986, 1990 for a review).

The results of downcore stratigraphic analysis utilizing chrysophyte remains for seven small acidic lakes, believed to have been adversely affected by acid deposition, are summarized in Table I. Curiously enough, in all but one of the lakes (Little Echo), the lower strata were always overwhelmingly dominated (over 90% of the total scale counts) by the common species, *Mallomonas crassisquama*. Theoretically, such a finding implies that the pH levels during most of the pre-industrial revolution period were greater than 5 and in all probability above 6.0. Sharp shifts in species dominance from *M. crassisquama* to acidophilous and acidobiontic taxa were observed in each of the lakes (Table I). In most cases, some combination of *Mallomonas acaroides* v. *muskokana, Mallomonas hamata, Mallomonas hindonii, Synura echinulata* and *Synura sphagnicola* accounted for the majority of scales in recent sediments. Smaller pulses of other slightly acidic to circumneutral species, such as *Mallomonas galeiformis* (reported as *M. trummensis*), *Mallomonas punctifera* and *Mallomonas calceolus* were often found prior to the sharp rise in the more acid-tolerant organisms. In each lake, the changes in the abundances of the taxa between respective strata reflected shifts in lakewater pH.

There is now equally strong evidence to indicate that scaled chrysophytes can also signal upward shifts in lakewater pH, i.e. lake recovery. Little Echo Pond is a highly colored, acidic bog lake with a pH between 4.2 and 4.5. The lower strata, upper strata (Smol, 1986) and present day flora (Siver, 1988a) are overwhelmingly

TABLE I

Summaries of paleolimnological studies using scaled chrysophytes to infer historical lakewater pH values. Only the dominant species for both the lowermost strata (oldest sediments) and the uppermost strata (youngest sediments) are given. The pH catagories are as suggested by the author(s) and follow the system of Hustedt (1939): ACB = acidobiontic; ACF = acidophilic; IND = indifferent (circumneutral)

Reference	Lake/Location	Current pH	Dominant taxa			
			Lower Strata	pH Category	Upper Strata	pH Category
(1) Smol *et al.*, 1984b	Deep Lake, Adirondacks	4.7	*M. crassisquama*	IND	*S. echinulata*	ACF
					M. hamata	ACB
					M. hindonii	ACB
(2) Christie and Smol, 1986	Upper Wallface, Adirondacks	4.7–5.0	*M. crassisquama*	IND	*M. echinulata*	ACF
					M. hamata	ACB
					M. hindonii	ACB
(3) Charles *et al.*, 1987	Big Moose, Adirondacks	4.6–5.0	*M. crassisquama*	IND	*M., hindonii*	ACB
					M. hamata	ACB
(4) Smol, 1986	Little Echo, Adirondacks	4.2–4.5	*M. acaroides*	ACF	*M. acaroides*	ACF
			S. sphagnicola	ACF	*S. sphagnicola*	ACF
(5) Hartmann and Steinberg, 1986	Kleiner Arbersee, ALPS	5.0	*M. crassisquama*	IND	*S. echinulata*	ACF
					S. sphagnicola	ACF
(6) Dixit *et al.* unpub. data	Lake Bonneville, Quebec	5.1	*M. crassisquama*	IND	*S. echinulata*	ACF
	Lake Truite Rouge, Quebec	5.9	*M. crassisquama*	IND	*M. acaroides*	ACF
			M. allorgei	IND	*M. galeiformis* [a]	ACF
					S. echinulata	ACF
					S. petersenii	IND
					M. hamata	ACB

[a] Reported by Dixit *et al.* (unpub. data) as *M. trummensis*

dominated by *M. acaroides* v. *muskokana* and *S. sphagnicola*, taxa belonging to the low pH group. However, a pulse of scales from the circumneutral species, *M. caudata* was observed in a stratum that corresponded to a series of liming events in the 1960's (Smol, 1986).

The pH of Swan Lake, located in the Sudbury (Ontario) region, has increased from a low of 3.9 (1977) to a high of 5.7 (1986), following a known decrease in inputs from acid deposition (Dixit *et al.*, 1989). In sediments from Swan Lake dated pre-1940, the relative amounts of species of scaled chrysophytes was similar. A major shift towards a predominance of species in the low pH group commenced circa 1940, corresponding to an increase in mining and smelting operations (Dixit *et al.*, 1989). A second shift, back to more circumneutral taxa, correlated with known reductions in SO_2 emissions. Based on a pH inference model developed from lakes in the Sudbury region (Dixit *et al.*, 1988), changes in calculated values of chrysophyte-inferred pH were highly correlated with measured values (Dixit *et al.*, 1989).

Multiple regression models for pH inference based on the remains of scaled chrysophytes from surface sediments have yielded r^2 values of 0.70 and 0.74 for lakes in the Adirondacks (Charles and Smol, 1988) and Sudbury, Ontario region (Dixit *et al.*, 1988), respectively. Highly significant models with r^2 values ranging from 0.88 to 0.94 have been developed using both chrysophyte and diatom remains (Charles and Smol, 1988).

Davis and Smol (1986) identified several potential problems involved with using siliceous algae (chrysophytes and diatoms) for inferring historical lakewater conditions. One shortcoming, the lack of ecological data to adequately assess the value of individual species of scaled chrysophytes as indicators of pH, has been largely overcome by the recent surge of work on scaled chrysophytes. Davis and Smol (1986) also pointed out that further work was needed to better understand the preservation of scales under various environmental regimes. The dissolution of scales may be dependent on the species, scale thickness, the conditions of preservation and/or the age of the sediments. Haworth (1983, 1984) reported high concentrations of chrysophyte scales in the top 30 cm of sediments from Blelham Tarn; however, no scales were found below the 30 cm stratum. Kristiansen (1986) suggested that the disappearance of scales reported by Haworth may be the results of dissolution of scales in the older sediments. In sediments from several Swedish lakes, Cronberg (1982a,b) reported a rich concentration of cysts of *Mallomonas eoa*, but no scales were found. The overwhelming dominance of *M. crassisquama* in older sediments could be related, in part, to a differential dissolution and/or breaking of scales.

The siliceous resting stage common to all chrysophytes, known as the stomatocyst, is species-specific and known to preserve well in lake sediments (Nygaard, 1956; Kristiansen, 1986). Stomatocyst microfossils are becoming increasingly important in paleolimnology (Smol, 1990) for inferring historical changes in lakewater (e.g. Adam and Mahood, 1981; Battarbee *et al.*, 1980; Carney and Sandgren, 1983; Smol, 1985; Duff and Smol, 1988; Rybak,, 1986; Rybak *et al.*, 1991), and will no doubt

add to our knowledge of lake ontogeny, especially in systems where scale preservation is poor. The use of stomatocysts is somewhat limited because only 30 of the 430 morphotypes can be identified to the species level (Kristiansen, 1986; Cronberg, 1986).

Siver and Hamer (1990) recently demonstrated that highly significant pH inference models, with r^2 values above 0.8, can also be developed using data based on discrete samples of live populations. In a 5 yr study of a small lake in Connecticut, Siver and Hamer (1992) found that the most significant changes in the scaled chrysophyte flora correlated with spring declines in pH. During the 5 yr period the acidobionts *Mallomonas pugio* and *M. canina* were observed only when the pH dipped below 5.5.

In conclusion, the available evidence from both paleo and neolimnological studies clearly indicate that since scaled chrysophytes would be a powerful tool for measuring shifts in lakewater pH, they would be a valuable group for use in the long term monitoring of lakes. Future areas of research, such as understanding the effects of covariables on species distributions, will only further enhance our abilities to interpret pH change in lake systems using scaled chrysophytes.

5. Proposed Sampling Methodologies

5.1. Introduction

As discussed above, there are several reasons why the focus of a long-term monitoring program (LTMP) incorporating scaled chrysophytes should be placed on analysis at the specific and subspecific levels. First, the distributions of many taxa along a pH gradient are well described from different lake regions. Second, many species are found over a rather narrow pH interval and, as such, they are very sensitive to shifts in pH. Third, the reported sensitivities of most species to changes in pH are similar in many lake regions. Fourth, the taxonomy is very sophisticated; even though electron microscopy is necessary for the proper identification of many species, the identifications are usually accurate and unambiguous. Fifth, because the siliceous remains persist as microfossils, they can also be used to infer paleolimnological conditions.

We propose that live populations of scaled chrysophytes serve as the primary basis of the long-term monitoring program (LTMP). We further propose that the program be supplemented with a one-time analysis of scaled chrysophytes from a short sediment core representing approximately 100 to 200 yr. Supplementing a LTMP with a one-time analysis of a sediment core would provide an historical framework. This is especially attractive since the preparation of samples for extant and microfossil populations is similar (see below). The incorporation of both microfossils and living populations would result in: (a) the immediate preparation of a 'present day' data base from which future shifts in lakewater acidity could be monitored; (b) an estimation of any long-term historical changes and; (c) the

collection of additional data (e.g. seasonality data) that could be used to fine-tune statistical models.

5.2. Response variable

A suitable response variable must be applicable to both living and microfossil assemblages, and one that can result in correct enumeration of species composition. We propose that the abundance of scales be used as the counting parameter. Scales are relatively easy to indentify and they can be estimated from both living and microfossil assemblages using similar preparation methods. Scale counts are the response variable used in paleolimnological investigations (e.g. Smol, 1986; Charles and Smol, 1988; Dixit *et al.*, 1988; Cumming *et al.*, 1991). Scale counts for many of the important taxa now can be transformed into cell concentrations (Siver, 1991b). Thus, either scale or cell counts could be expressed on a per volume of water (living) or per unit of sediment (microfossil) basis.

5.3. Sampling program

Programs consisting of one or four sampling events per year are considered. We propose that sediment traps and discrete water samples be used to sample the extant flora. A short sediment core would be taken once on the initial visit to the lake when the sediment trap(s) are deployed. On subsequent sampling events the contents of the trap(s) are harvested and the trap reset. On each visit an integrated tube sample of the euphotic zone is taken along with a net tow (6 or 10 μm mesh). The water samples will be subsequently used for quantitative analysis; the net samples are solely used for aiding in identification purposes.

Sediment cores and trap samples are advantageous because they integrate the remains of populations that grew and sedimented throughout a defined period of time, and over the entire water column. Trap collections could be further divided into 'seasonal' samples in a 4× per yr program. In shallow lakes, as well as in some deep lakes, the resuspension of sediment into the traps may limit their use, especially during periods of mixing. Discrete water samples from the euphotic zone require minimal collecting time and would provide an additional evaluation tool, especially if the use of the traps is not suitable or they are lost. It is inevitable that some taxa will be missed. Even though the missing taxa may differ between the trap, core and discrete water samples, data collected from each will provide equally sophisticated models (Siver and Hamer, 1990). Each approach will serve to complement the others and provide a robust means for measuring shifts in lakewater pH.

Sampling protocols would be identified for all geographic regions. A spring overturn date is proposed if a sampling regime of 1× per yr is chosen. Proposed sampling periods for dimictic lakes where a 4× per yr regime is selected are: minimum temperature (under the ice); spring overturn maximum temperature (late summer); and fall overturn. Modifications for monimictic lakes or where under-ice sampling is not possible, are summarized in Marmorek *et al.* (1988).

5.4. Field and laboratory procedures

Field and sample preparation techniques are similar to those used for the diatoms and are only briefly summarized.

It is proposed that a sediment core be taken from the deepest part of the basin with a modified K-B gravity corer (Brinkhurst *et al.*, 1969; Glew, 1988, 1989) equipped with a 6.5 cm ID plexiglass tube (Blomquist, 1985) and extruded on shore using a vertical extrusion system. The core can be sectioned in 0.5 or 1 cm intervals, each placed in a Whirl-pac bag (Smol *et al.*, 1984), and maintained according to protocols outlined by Charles and Whitehead (1987). The number of sediment sections analyzed would be determined by available funding and the need for historical information. Sections not analyzed can easily be archived and also used for dating purposes

We propose that a single sediment trap assembly, equipped with three cylindrical tube traps, each with an aspect ratio (height:diameter) of between 5 and 10, be positioned 1 to 2 m from the bottom using an anchor-buoy system (see Bloesch and Burns, 1980; Blomqvist and Hakanson, 1981; Flower, 1990). The tops of two of the cylindrical tube traps can be capped while the third is emptied. A subsurface buoy is attached to each trap assembly. Additional means for facilitating the location of a trap may include the use of an independent surface buoy, a submerged line leading from the trap to a known point on shore, radio control devices or standard triangulation procedures. The method used to locate a trap will be dependent, in part, on the size and accessibility of the lake.

Water samples are to be taken with an integrated tube sampler. A depth of twice the Secchi disc depth is proposed for simplicity. Water and net samples are fixed immediately with acid Lugol's (Wee, 1983); formalin or another preservative can be added later.

A total of 100–200 mL of each water sample is concentrated with centrifugation. Known volumes of each concentrated water, sediment trap and core sample should be digested using the strong acid method to remove organic matter, rinsed, reconcentrated, and used to prepare slides for quantitative analyses (Battarbee, 1986). Subsamples of each digested sample are air dried onto a number of cover slips, preferable using Battarbee trays (Battarbee, 1986), mounted on glass slides using Hyrax and labelled. It is suggested that 500 scales from a known area be enumerated under oil imeersion using phase-optics. Using appropriate conversion factors, the absolute concentration of scales and cells can be calculated.

There are no up-to-data comprehensive taxonomic guides for scaled chrysophytes. However, there are many recent taxonomic papers that collectively provide EM micrographs of all accepted taxa. Two general guides that include numerous taxa are ones by Wee (1982) and Takahashi (1978). Recent books by Asmund and Kristiansen (1986) and Siver (1991a) for the genus *Mallomonas* are available. Variability in the structure of scales and bristles is discussed in many of the taxonomic papers.

Species identifications are to be verified with EM prior to their enumeration. Either SEM or TEM can be used. For SEM, aliquots for each type of sample can be dried onto aluminum foil, coted with gold and/or palladium (Siver, 1987), and observed directly with SEM. For TEM, aliquots are dried on formvar/carbon coated copper grids. For the live samples the appropriate net samples can be analyzed to aid in identifications.

Photographic records of both light and electron micrographs, cross referenced to the type of sample, date, and lake will be a critical aid in species identification and quality assurance / quality control (QA/QC) procedures. Slide preparations, aluminum foil samples, the stubs used for SEM, and the TEM grids can be easily archived.

5.5. Additional considerations

QA/QC protocols similar to those outlined by Charles and Whitehead (1987) can be directly applied to procedures used in sample collection, preparation and taxonomy. Inter and intra-core, trap and water sample variability can also be determined.

Many of the ordination and regression techniques are referred to above. The multivariate technique known as CANOCO, recently developed by ter Braak (1986), is a theoretically superior method of ordination that has also been modified to provide a more appropriate means of using data sets in multiple regression analysis (Stevenson *et al.*, 1989). Weighted averaging techniques outlined by Birks *et al.* (1990) can be used to prepare appropriate inferential models.

As is true with diatoms, the use of microfossil remains could provide an immediate means of estimating natural variability within a given region (Smol, 1990). Patchiness is not considered a problem in the core or trap samples since they integrate over the entire water column and throughout the year. The use of a tube sample would also effectively integrate populations from all depths throughout the euphotic zone.

6. Recommendations

The collection, preparation and analysis of both live and sediment samples for scaled chrysophytes could easily be incorporated, at a minimal expense, into an existing long-term monitoring program. Field collections of live samples can be made quickly and would require little training. Even through the collection of sediment samples would require additional time, it is a straight-forward procedure that would be done only once.

Few equipment items are needed for the proper analysis of chrysophyte populations. Water sampling devices, gravity corers and sediment traps are relatively inexpensive. The preparation of samples for microscopical analyses would require the use of a centrifuge, fumehood, warming tray and a research grade microscope. Electron microscope facilities are normally rented.

The estimated cost, in terms of person-hours per sample, for three combinations

TABLE II

Proposed cost estimate analysis for the inclusion of scaled chrysophytes into a long term monitoring program for the detection of chronic or episodic acidification or recovery events. Three combinations are presented that differ in their method of collection (i.e. discrete samples or sediment traps) and inclusion of microfossil populations. All combinations assume a scale abundance measurement as the response variable and identification of taxa with scanning electron microscopy. Travel to/from sites is not included. Estimates are given in person-hours per sample and are separated on the basis of field *vs* laboratory and technician (T) *vs* skilled expert (SK) requirements. Microfossil analyses and the setting of traps is done only on the initial visit

Combination	Scaled Chrysophyte Group	Method of Collection
A	Live	Discrete samples
B	Live	Sediment traps
C	Live and microfossil	Discrete, Trap, Core

Combination	Field or Laboratory	Method of Collection	Live or Microfossil	Process	Person-hours per Sample	Personnel Requirements	Total Time Allotment per Sample
A	Field	Discrete	Live	Collection	0.25	T	
	Laboratory			Sample preparation	0.5	T	
				Sample analysis	3–4	SK	
				Data entry	0.5	T	
							4.25–5.25
B	Field	Trap	Live	Set trap	2	T	
				Collection	2	T	
	Laboratory			Sample preparation	0.5	T	
				Sample analysis	3–4	SK	
				Data entry	0.5	T	
							8–9
C	Field	Discrete	Live	Collection	0.25	T	
		Trap	Live	Collection	2	T	
		Core	Microfossil	Collection	3–4	T	
	Laboratory	Discrete	Live	Total from A	4–5	T/SK	
		Trap	Live	Total from B	4–5	T/SK	
		Core	Microfossil	Sample preparation	0.5	T	
				Sample analysis	3–4	SK	
				Data entry	0.5	T	
							17.25–21.25

of methods (i.e. integrated tube, traps or cores) and organismal assemblages (live or microfossil) are suggested in Table II. All combinations assume scale abundance as the response variable and vertification of taxa using electron microscopy. Ideally, the sampling program outlined above (combination C, Table II) would yield the maximum degree of information and allow for present day and historical inferences. Removing the microfossil component would not alter the inference capabilities of methods based on living populations; it would simply remove the historical aspects.

The use of either integrated tube samples or sediment traps would yield equally significant and complementary results. Tube samples should be taken, regardless of the program, since only a minimal (0.25 hr) allotment of time is needed to make the collection. Samples should always be archived. Even through the time allotment per sample is approximately doubled if microfossil populations are incorporated (Table II), the analyses are necessary on only the initial visit; on subsequent visits the person hours per sample figure is reduced to 5 to 9 hr.

The person-hours per sample estimates calculated for scaled chrysophytes are overestimated in the sense that no additional field work would be necessary to include all other groups of phytoplankton. Also, preparation of samples for counting chrysophytes and diatoms could be done simultaneously. We feel that the amount of information obtainable is directly proportional to the true expenditure. Thus, the feasibility of implementing combination C as opposed to A or B is essentially a function of available funding.

7. Conclusions

Biomonitoring provides the best means of measuring the response of an aquatic ecosystem to shifts in lakewater acidity. Changes in lakewater pH beyond natural variability, whether unidirectional or oscillating, whether subtle or episodic and whether long or short lived, could easily go undetected in a routine chemical monitoring program, but would inevitably leave their mark on the organisms in the ecosystem. The question becomes, 'Of the myriad of organisms in a given system which ones could best reflect both unidirectional long term trends of 'small' magnitude and short term oscillations of 'larger' magnitude?'.

Phytoplanktic organisms are usually autotrophic, possess simple morphologies and have relatively high rates of reproduction. These three features set them apart from most other groups of organisms and make them especially attractive as candidates for biological monitoring. First, their autotrophic nature implies that a response of the organism to changing lakewater acidity was a direct effect on the cell and not one related to a change in food supply (i.e. an indirect effect). Second, because they are short lived organisms with relatively high turnover rates, each individual cell is exposed to the range in lakewater pH expected to occur over a short (e.g. a week) time period, as opposed to a much greater range in the case of longer lived organisms (e.g. fish). As such, by virtue of their inherent life cycles, phytoplankton theoretically can offer a much finer scale by which to

monitor lakewater chemistry. The time frame of the lifespan of an algal cell is similar in magnitude to the time scale in which measurable changes in pH are detected. An additional advantage of using an organism with a short lifespan is that collections made between years most likely represent independent populations. This means that the idea of independence, often assumed in most statistical procedures, is valid. Such may not be true for longer lived organisms (i.e. fish). Lastly, because phytoplankton are structurally simple, mostly single celled in nature, a change in pH would trigger an immediate effect on the cell. By virtue of their simple morphologies, the cell lacks a complex physiological system for buffering change.

As outlined in this paper, there are many advantages for using scaled chrysophytes in a LTMP. The major points can be summarized as follows. First, there are a relatively large number of well defined taxa, that can be identified accurately and are cosmopolitan in their distributions. Second, the distributions of many taxa along a pH gradient are well described. Many are found over a rather narrow pH range marked at their extremes by abrupt decreases in occurrence. Third, the same organisms are applicable in all geographical areas. Fourth, pH has been repeatedly shown to be an important environmental factor controlling the distributions of scaled chrysophytes. Fifth, because their siliceous cell walls persist in sediments they can also be used to infer historical shifts in pH and provide a means by which to estimate natural variability. Sixth, there is a wealth of studies that clearly demonstrate that scaled chrysophytes can identify shifts in lakewater acidity. Seventh, scaled chrysophytes are truly planktonic and therefore reflect their ambient environment. Eighth, QA/QC procedures can readily be applied to all aspects of the proposed sampling program. Lastly, field and laboratory procedures necessary for implementing a LTMP are simple and inexpensive.

The levels of sophistication of the potential inferences that would be made from data collected by each combination outlined above (Table II) would be equal. The differences are simply in the types of inferences that could be made. The inclusion of historical data (i.e. the microfossil analyses) is not needed in a LTMP, but highly recommended. A sampling program utilizing either integrated tube samples (combination A) or sediment traps (combination B) would each provide a mechanism for detecting acidification or recovery at an early stage, assessing regional trends, and monitoring episodic events.

References

Adam, D. P. and Mahood, A. D.: 1981, *Geological Society of America Bulletin* **92**, 839.

Almer, B., Dickson, W., Ekstrom, C., Hornstrom, E., and Miller, U.: 1974, *Ambio* **3**, 30.

Almer, B., Dickson, W., Ekstrom, C., and Hornstrom, E.: 1978, 'Sulfur pollution and the Aquatic Ecosystem', in Nriagu, J. O. (ed.), *Sulfur in the Environment. Part II, Ecological Impacts.* John Wiley & Sons, New York, pp. 271–311.

Anderson, R. A.: 1987, *Amer. J. Bot* **74**, 337.

Asmund, B.: 1959, *Dansk Botanisk Arkiv* **18**, 1.

Asmund, B. and Hilliard, D. K.: 1961, *Hydrobiologia* **17**, 237.
Asmund, B. and Kristiansen, J.: 1986, *Op. Bot.* **85**, 1–128 pp.
Battarbee, R. W.: 1979, 'Diatoms in Lake Sediments', in Berglund, B.E. (ed.), *Paleohydrological Changes in the Temperate zone in the Last 15000 Years* Sub project B, Vol. II, pp. 177–225.
Battarbee, R. W.: 1984, *Philosophical Transactions of the Royal Society of Londen, Series B***305**, 451.
Battarbee, R. W.: 1986, 'Diatom Analysis', in Berglund, B. E. (ed,), *Handbook of Holocene Palaeoecology and Palaeohydrology* John Wiley & Sons, New York, pp. 527–570.
Battarbee, R. W., Cronberg, G., and Lowry, S.: 1980, *Hydrobiologia* **71**, 225.
Beamish, R. J.: 1976, *Water, Air, and Soil Pollution* **6**, 501.
Birks, H. J. B., Line, J. M., Juggins, S., Stevenson, A. C., and ter Braak, C. J. F.: 1990, *Phil. Trans. R. Soc. London. B.* **327**, 263.
Bloesch, J. and Burns, N. M.: 1980, *Schweiz. Z. Hydrol.* **42**, 15.
Blomquist, S.: 1985, *Sedimentology* **32**, 605.
Blomquist, S. and Håkanson, L.: 1981, *Arch. Hydrobiol.* **91**, 101.
Bradbury, J. P.: 1975, 'Diatom Statigraphy and Human Settlement in Minnesota', Special Paper 171, The Geological Society of America, Boulder, Colorado.
Bradley, D. E.: 1964, *J. Gen. Microbiol.* **37**, 321.
Brinkhurst, R. O., Chua, K. E., and Batoosingh, E.: 1969, *J. Fish. Res. Bd. Can.* **26**, 2581.
Carney, H. J. and Sandgren, C. D.: 1983, *Hydrobiologia* **101**, 195.
Charles, D. F.: 1985, *Ecology* **66**, 994.
Charles, D. F. and Norton, S. A.: 1986, 'Paleolimnological Evidence for trends in atmospheric deposition of acids and metals', in 'Acid Deposition: Long-Term Trends', National Academy Press, Washington, D. C., pp. 335–431.
Charles, D. F. and Whitehead, D. R.: 1987, 'Paleoecological Investigation of Recent Lake Acidification (PIRLA): Interim Report', Electric Power Research Institute, Palo Alto, pp. 1–414.
Charles, D. F. and Smol, J. P.: 1988, *Limnology and Oceanography* **33**, 1451.
Charles, D. F., Battarbee, R. W., Renberg, I., van Dam, H., and Smol, J. P.: 1989, 'Paleoecological Analysis of Lake Acidification Trends in North America and Europe Using Diatoms and Chrysophytes', in Adriano, D. (ed.), *Acid Precipitation, v.2.* Springer-Verlag, New York.
Conway, H. L. and Hendrey, G. R.: 1982, 'Ecological Effects of Acid Precipitation on Primary Producers', in D'Itri, F.M. (ed.), *Acid Precipitation, Effects on Ecological Systems* Ann Arbor, Michigan.
Christie, C. E. and Smol, J. P.: 1986, *Hydrobiologia* **143**, 355.
Cronberg, G.: 1982a, *Hydrobiologia* **86**, 185.
Cronberg, G.: 1982b, *Folia Limnologica Scandanavica* **18**, 1.
Cronberg, G.: 1986, 'Chrysophycean Cysts and Scales in Lake Sediments: A Review', in: Kristiansen, J. and Andersen, R. A. (eds.), *Chrysophytes: Aspects and Problems* Cambridge University Press, Cambridge, pp. 281–315.
Cronberg, G. and Kristiansen, J.: 1980, *Bot. Notiser* **133**, 595.
Cumming, B. F., Smol, J. P., and Birks, H. J. B.: 1991, *Nord, J. Bot.* **11**, 231.
Davis, R. B., Norton, S. A., Hess, C. T., and Brakke, D. F.: 1983, *Hydrobiologia* **103**, 113.
Davis, R. B. and Anderson, D. S.: 1985, *Hydrobiologia* **120**, 69.
Davis, R. B. and Smol, J. P.: 1986, 'The Use of Sedimentary Remains of Siliceous Algae for Inferring Past Chemistry of Lake Water - Problems, Potential and Research Needs', in Smol, J. P., Battarbee, R. W., Davis, R. B., and Merilainen, J. (eds.), *Diatoms and Lake Acidity* Dr. W. Junk, Dordrecht, The Netherlands, pp. 291–300.
DeNoyelles, F. and O'Brien, W. J.: 1978, *Archiv für Hydrobiologie* **81**, 137.
Dixit, S. S.: 1986, 'Algal Microfossils and Geochemical Reconstructions of Sudbury Lakes: A Test of the Paleo-Indicator Potential of Diatoms and Chrysophytes', PhD Thesis, Queen's University, Kingston, Ontario, pp. 1–190.
Dixit, S. S., Dixit, A. S., and Evans, R. D.: 1988, *Canadian Journal of Fisheries and Aquatic Sciences* **45**, 1411.
Dixit, S. S., Dixit, A. S., and Smol, J. P.: 1989, *Can. J. Fish. Aquat. Sci.* **46**, 1667.
Dixit, S. S., Smol, J. P., Anderson, D. S., and Davis, R. B.: 1990, *J. Paleolimnol.* **3**, 269.
Duff, K. E. and Smol, J. P.: 1988, *Canadian Journal of Botany* **66**, 1117.
Durrschmidt, M.: 1990, *Nova Hedwigia* **33**, 353.

Durrschmidt, M.: 1982, *Archiv für Hydrobiologie Supplement* **63**, 121.
Eloranta, P.: 1985, *Memoranda Societas pro Fauna et Flora Fennica* **62**, 41.
Findlay, D. L.: 1978, *Can. Fish, Mar. Serv. M. S. Report* **466**, I-IV + 1.
Flower, R. J.: 1990, *Hydrobiologia* **214**, 311.
Glew, J. R.: 1988, *J. Paleolimn.* **1**, 235.
Glew, J. R.: 1989, *J. Paleolimn.* **2**, 241.
Hartman, H. and Steinberg, C.: 1986, *Hydrobiologia* **143**, 87.
Havens, K. and Decosta, J.: 1986, *Hydrobiologia* **137**, 211.
Haworth, E. Y.: 1983, *Hydrobiologia* **103**, 131.
Haworth, E. Y.: 1984, 'Stratigraphic Changes in algal Remains (Diatoms and Chrysophytes) in the Recent Sediments of Blelham Tarn, English Lake District', in Haworth, E. Y., and Lund, J. W. G. (eds.), *Lake Sediments and Environmental History*, University of Minnesota Press, Minneapolis, pp. 165–190.
Hendrey, G. R. and Wright, F. R.: 1976, *J. Great Lakes Res.* **2 (Suppl. 1)**, 192.
Hendrey, G. R., Baalsrud, K., Traaen, T. S., Laake, M., and Raddum, G.: 1976, *Ambio* **5**, 224.
Hendrey, G. R., Yan, N. D., and Baumgartner, K. J.: 1981, *Responses of Freshwater Plants and Invertebrates to Acidification*, Internat. Sym. for Inland Waters and Lake Restoration, Portland, Maine.
Hornstrom, E., Ekstom, C., Miller, U., and Dickson, W.: 1973, *SNV 1973* **4**.
Hustedt, F.: 1939, *Arch. Hydrobiol. Suppl.* **16**, 1–394.
Hutchinson, G. E.: 1967, *A. Treatise on Limnology, 2. Introduction to Lake Biology and the Limnoplankton*, John Wiley & Sons, New York.
Ito, H. and Takahashi, E.: 1982, *Japanese Journal of Phycology* **30**, 272.
Jacobsen, B. A.: 1985, *Nord. J. Bot.* **5**, 381.
Kling, H. J. and Holmgren, S. K.: 1972, *Can. Fish. Mar. Serv. Tech. Report* **337**, 1.
Kling, H. J. and Kristiansen, J.: 1983, *Nord. J. Bot.* **3**, 269.
Kristiansen, J.: 1975, *Verh. Internat. Verein. Limnol.* **19**, 2709.
Kristiansen, J.: 1978, *Bot. Tidsskr.* **73**, 71.
Kristiansen, J.: 1985, *Verh. int. Verein. rheor. angew. Limnol.* **22**, 2826.
Kristiansen, J.: 1986, *British Phycological Journal* **21**, 425.
Kristiansen, J.: 1988, *Hydrobiologia* **161**, 171.
Munch, C. S.: 1990, *Freshwater Biology* **10**, 61.
Nicholls, K. H.: 1981, *Canadian Jounnal of Botany* **59**, 107.
Nicholls, K. H.: 1982, *Nova Hedwigia* **34**, 80.
Nygaard, G.: 1956, *Folia Limnol. Scand.* **8**, 32.
Preisig, H. R. and Hibberd, D. J.: 1983, *Nord. J. Bot.* **3**, 695.
Roijackers, R. M. and Kessels, H.: 1986, *Nord. J. Bot.* **6**, 373.
Rybak, M.: 1986, *Hydrobiologia* **140**, 67.
Rybak, M., Rybak, I., and Nicholls, K.: 1991, *J. Paleolomnol.* **5**, 19.
Sandgren, C. D.: 1988, 'The Ecology of Chrysophyte Flagellates: Their Growth and Perennation Strategies as Freshwater Phytoplankton', in Sandgren, C. D. (ed.), *Growth and Reproductive Strategies of Freshwater Phytoplankton*, Cambridge Univ. Press, Cambridge, pp. 9–104.
Schindler, D. W. and Nighswander, J. E.: 1970, *J. Fish. Res. Bd. Can.* **27**, 2009.
Schindler, D. W. and Holmgren, S. K.: 1971, *J. Fish. Res. Bd. Can.* **28**, 189.
Schofield, C. L.: 1976, *Ambio* **5** 228.
Siver, P. A.: 1987, *Nord. J. Bot.* **7**, 107.
Sivel, P. A.: 1988a. *Canadian Journal of Botany* **66**, 1391.
Siver, P. A.: 1988b, *Br. phycol. J.* **23**, 379.
Siver, P. A.: 1988c, *Nord, J. Bot.* **8**, 205.
Siver, P. A.: 1989a, *Can. J. Bot.* **67**, 2120.
Siver, P. A.: 1989b, *Beiheft zur Nova Hedwigia* **95**, 111.
Siver, P. A.: 1991a, *The Biology of Mallomonas: Morphology, Taxonomy and Ecology*, Kluwer Academic Publishers, Dordrecht, pp. 1–230.
Siver, P. A.: 1991b, *J. Paleolimnology* **5**, 219.
Siver, P. A. and Chock, J. S.: 1986, 'Phytoplankton Dynamics in a Chrysophycean Lake', in Kristiansen, J. and Andersen, R. A. (eds.), *Chrysophytes: Aspects and Problems*, Cambridge University Press, Cambridge, pp. 165–183.

Siver, P. A. and Skogstad, A.: 1988, *Nord, J. Bot.* **8**, 99.
Siver, P. A. and Hamer, J. S.: 1989, *Limnol. Oceanogr.* **34**, 368.
Siver, P. A. and Hamer, J. S.: 1990, *Can. J. Fish. Aquat. Sci.* **47**, 1339.
Siver, P. A., Hamer, J. S. and Kling, H.: 1990, *J. Phycol.* **26**, 728.
Siver, P. A. and Hamer, J. S.: 1992, *J. Phycol.* **28**, 186.
Skogstad, A.: 1982, 'Synuraceae-floraen i 27 lokaliteter i Oslo-området. Hovedfagsoppgave', Ph.D. Thesis, University of Oslo, Norway.
Smol, J. P.: 1985, *Hydrobiologia* **123**, 199.
Smol, J. P.: 1986, 'Chrysophycean Microfossils as Indicators of Lakewater pH', in Smol, J. P., Battarbee, R. W., Davis, R. B. and Meriläinen, J. (eds.), *Diatoms and Lake Acidity*, Dr. W. Junk, Dordrecht, The Netherlands, pp. 275–287.
Smol, J. P.: 1990, 'Paleolimnology: Recent Advances and Future Challenges', in De Bernardi, R. (ed.), *Scientific Perspectives in Theoretical and Applied Limnology*, Mem. Ist. Ital. Idrobiol. **47**, 253.
Smol, J. P., Charles, D. F., and Whitehead, D. R.: 1984, *Canadian Journal of Botany* **62**, 911.
Smol, J. P., Battarbee, R. W., Davis, R. B., and Meriläinen, J.: 1986, *Diatoms and Lake Acidity*, Dr. W. Junk Publishers, Dordrecht, pp. 1–307.
Stevenson, A. C., Birks, H. J. B., Flower, R., and Battarbee, R. W.: 1989, *Ambio* **18**, 228.
Takahashi, E.: 1967, *Bulletin of the Yamagata University (Agricultural Science)* **5**, 99.
Takahashi, E.: 1978, *Electron Microscopical Studies of the Synuraceae (Chrysophyceae) in Japan, Taxonomy and Ecology,* Takai University Press, Tokyo.
ter Braak, C. J. F.: 1986, *Ecology* **67**, 1167.
Wee, J. L.: 1982, *Bibliotheca Phycologia* **62**, 1.
Wee, J. L.: 1983, *Transactions of the American Microscopical Society* **102**, 68.
Wee, J. L. and Gabel, M.: 1989, *Am. Midl. Nat.* **121**, 32.
Wetzel. R. G.: 1983, *Limnology*, 2nd (ed.) Saunders College Publishing, New York, pp. 1–767.
Willen, T.: 1969, *Oikos* **20**, 67.
Wujek, D. E., Gretz, M., and Wujek, M. G.: 1977, *The Michigan Botanist* **16**, 191.
Yan, N. D. and Stokes, P.: 1978, *Environmental Conservation* **5**, 93.