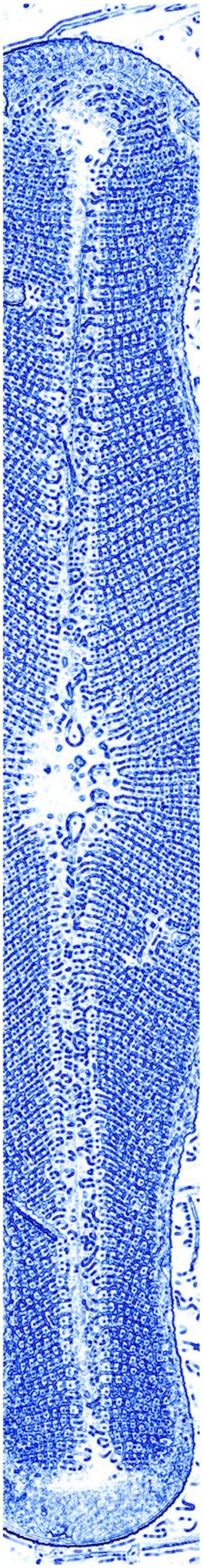

Increase in nuisance blooms and geographic expansion of the freshwater diatom *Didymosphenia geminata:*

Recommendations for response

White Paper, January 2007

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FOREWORD

In May 2006, EPA Region 8 and the Federation of FlyFishers co-sponsored an international symposium on an emerging issue, the phenomenon of *Didymosphenia geminata*. A concerned group of international scientists, resource managers, aquatic professionals, conservation groups, consulting firms, and state, federal and tribal agencies gathered to bring together the current knowledge of this microscopic diatom. The meeting was exceptional for the diversity of interests of participants, all joined by a common concern about a rather small organism, and its behavior and potential impacts. This white paper is an outcome of that meeting and the expressed need by participants to document the issues and make recommendations in responding to the change in behavior of *D. geminata*. We hope this document provides a basis to address research and management needs and to stimulate understanding of an amazing biological phenomenon.

ENDORSEMENT

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DISCLAIMER

EXECUTIVE SUMMARY

The diatom *Didymosphenia geminata* (Lyngbye) Schmidt is emerging as an organism with an extraordinary capacity to impact stream ecosystems on a global scale. In recent years, streams in New Zealand, North America, Europe, and Asia have been colonized by unprecedented masses of "didymo" and its extracellular stalks. This diatom is able to dominate stream surfaces by covering up to 100% of substrate with thicknesses of greater than 20 cm, greatly altering physical and biological conditions within streams. This species is expanding its geographic range in North America and the rate that nuisance blooms are reported by the public and local media are increasing, yet little scientific investigation of the phenomenon in North America has been initiated.

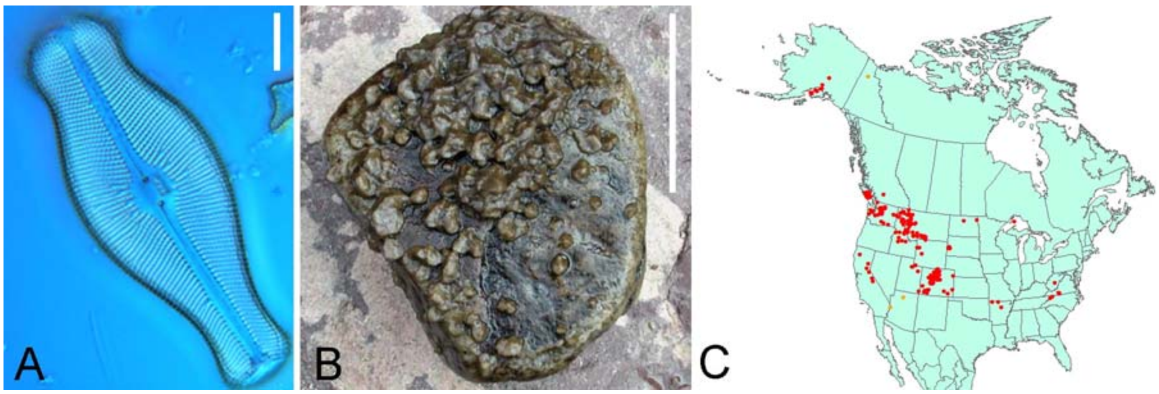

FIGURE 1. A. Image of *D. geminata* cell under the light microscope. Scale bar is equal to 10 microns. **B.** Cobble from stream showing typical growth habit. Scale bar is approximately 10 cm. **C.** Map showing the confirmed distribution records of *D. geminata* in North America.

PROBLEM

A global community of scientists, land managers, and anglers have reached consensus views on realized and potential threats of Didymosphenia geminata. *We recognize a growing body of evidence that* D. geminata *is:*

- *the only freshwater diatom to exhibit large scale invasive behavior, and a persistent phenomenon on a global scale*
- *a species with the biological capacity to produce inordinate amounts of stalk material (extracellular mucopolysaccarides) with unique properties*
- *a significant biological impact to stream ecosystem function, with the ability to alter foodweb structure and hydraulics of streams and rivers*
- *an organism that has expanded its ecological range and tolerance*
- *exhibiting a pattern of growth with potential impact to fisheries*
- *a significant strain on regional and national economies through impacts to tourism, fisheries, and hydropower*
- *an organism for which we lack basic biological and ecological knowledge*

SUMMARY OF RECOMMENDATIONS

- *Develop an aggressive education and outreach program to change user behavior in order to minimize spread of* D. geminata *on a global scale.*
- *Determine if there has been a genetically based physiological change in* D. geminata *that is linked to a nuisance strain.*
- *Trace the relationships of nuisance outbreaks and those records can be compared with models of predicted global distribution using molecular markers.*
- *Determine the degree to which the spread of* D. geminata *is aided by specific human vectors on waders or other gear.*
- *Track the geographic distribution of* D. geminata *on a global scale using effective and proper documentation of sites and voucher material.*
- *Determine the ecological conditions under which excessive biomass is produced in low nutrient streams and rivers, over short periods of time.*
- *Develop strategies to mitigate existing blooms.*
- *Determine the unique composition, structure, and cellular processes that produce the* D. geminata *stalk, which is responsible for its negative ecosystem impacts.*
- *Determine it perturbations in signal, regulatory, and/or synthetic pathways of stalk production by cymbelloid diatoms has resulted in increased production in* D. geminata.
- *Evaluate the apparent resistance of the stalk to degradation by bacteria and fungi, and determine ecosystem effects of stalk material.*
- *Verify the direct and indirect impacts of* D. geminata *and its stalks to aquatic macroinvertebrates and fish.*
- *Resolve the impacts of* D. geminata *at both high and low densities and whether there are threshold levels of nuisance growths.*

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INTRODUCTION

Didymosphenia geminata (Lyngbye) Schmidt was originally described from the Faroe Islands, north of Scotland. This diatom was very common in Scotland, Sweden and Finland (Cleve 1894-1896) and in the Kanchou region of China, *D. geminata* formed massive accumulations (Skvortzow 1935). While historic growth patterns include episodic formation of large masses, growth patterns now differ by having greater spatial coverage and temporal persistence. Until recently, this diatom was restricted to low nutrient waters, but now it occurs in more nutrient-rich streams and rivers. In many regions of North America, *D. geminata* now forms nuisance benthic growths that extend for greater than 1 km and persist for several months of the year. Furthermore, *D. geminata* has appeared to expand its geographic range within North America and Europe and recently invaded New Zealand. Under nuisance bloom conditions, *D. geminata* cells produce copious amounts of extracellular stalk material that form thick benthic mats. To the observer, these mats appear as fiberglass insulation, tissue paper, “rock snot”, brown shag carpet, or sheep skins covering the streambed (Fig.2).

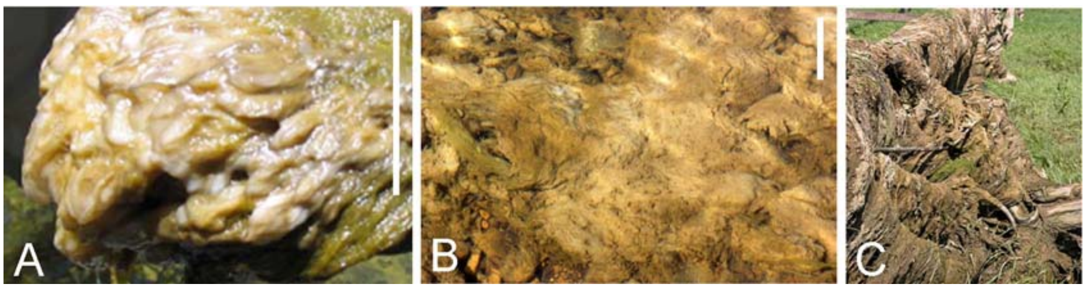

FIGURE 2. A. Stream cobble covered with *D. geminata* and stalks 5 cm thick. Scale bar equal to approximately 10 cm. **B.** Streambed covered with *D. geminata*. Note that rocks and cobbles are hardly visible. Scale bar equal to approx. 10 cm. **C.** Dried stalks on docks. (Images by Erica Shelby, Arkansas Department of Environmental Quality).

BIOLOGY

Didymosphenia geminata is a diatom, which is a type of single-celled algae. Diatoms are remarkable organisms, unique for their silica (SiO_2) cell walls, which are often well-preserved in sediments making diatoms useful as environmental indicators (Smol & Stoermer 1998). Diatoms are found in nearly every freshwater and marine aquatic habitat and contribute a large percentage of the global carbon budget through photosynthesis. In both oceans and freshwaters, diatoms are one of the major groups of organisms within the plankton (including other algae, bacteria, and protozoa) and also grow attached to surfaces. Diatoms store chrysolaminarin (ß1,3 linked glucan) as well as accumulate lipid within the cell. Lipids are an oil-rich source of energy, which make diatoms a valuable food for other organisms. The life history of diatoms includes both vegetative and sexual reproduction (reviewed in Edlund & Stoermer 1997), although the sexual stage has not been documented in *D. geminata* (but see Skabichevsky 1983).

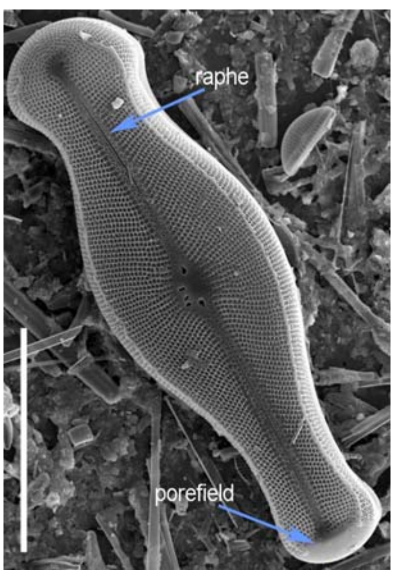

FIGURE 3. Scanning electron micrograph of the silica cell wall of *D. geminata*. The raphe is composed of the two slits that run along the apical axis of the cell. The cell secretes mucopolysaccarides through the raphe in order to move on surfaces. At the base of the cell is the porefield, through which the stalk is secreted. Scale bar equal to 50 µm (Image by Sarah Spaulding, US Geological Survey).

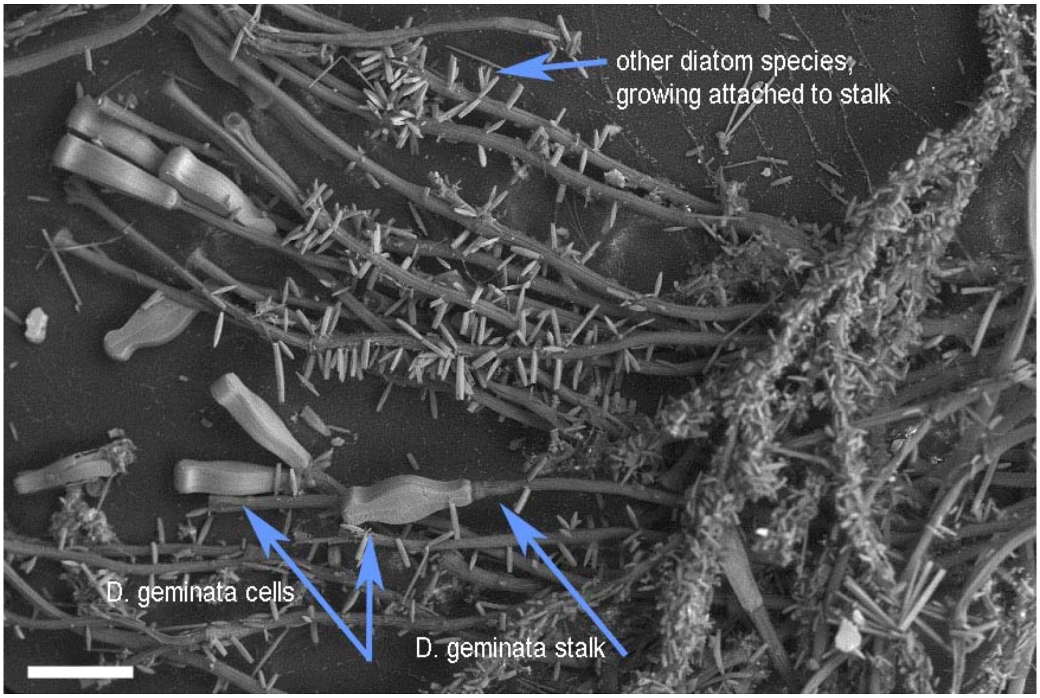

FIGURE 4. Scanning electron micrograph of *D. geminata* cells and their mucopolysaccaride stalks. The stalks produced within the cell are many times the length of the cell itself. Note the smaller diatoms growing attached onto the stalks. Scale bar equal to 100 µm (Image by Sarah Kiemle, Michigan Technological University).

Valve morphology of the genus *Didymosphenia* has been well documented (Dawson 1973a, 1973b, Antoine & Benson-Evans 1983, Stoermer et al. 1986, Metzeltin & Lange-Bertalot 1995). *Didymosphenia* is considered within the cymbelloid, rather than gomphonemoid, lineage of diatoms (Kociolek & Stoermer 1993). Cells possess a raphe, a structure that allows the cells to move on surfaces. The cells also possess an apical porefield, through which a mucopolysaccaride stalk is secreted (Fig. 3). The stalk may attach to rocks, plants, or any other submerged substrate. When the diatom cell divides (i.e. vegetative reproduction), the stalk also divides, eventually forming a dense mass of branching stalks. It is not the diatom cell itself that is responsible for the negative impacts of *D. geminata*, but the massive production of extracellular stalk (Fig. 4).

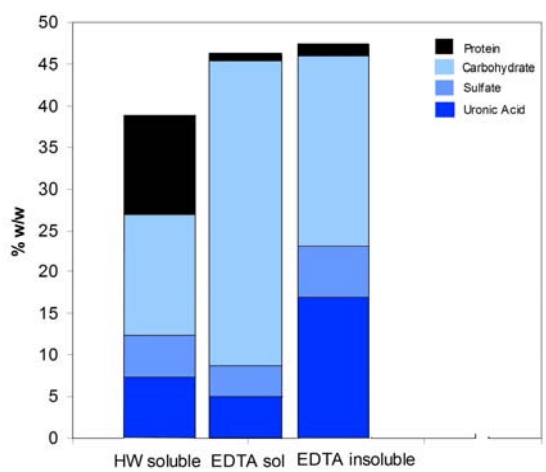

FIGURE 5. Biochemical composition of three fractions of the *D. geminata* stalk (hot water soluble, EDTA soluble, EDTA insoluble). The three fractions differ in their percentage composition of urionic acid, sulfate compounds, carbohydrate, and protein (Data from Michael Gretz, Michigan Technological University).

Extracellular polymeric substances (EPS) that comprise the stalk are predominantly composed of polysaccarides and protein (Fig. 5). They are complex, multi-layered structures that are resistant to degradation. The degree to which internal (genetic) and external (environmental) change initiates the high level of stalk production is unknown, yet resolving the mechanisms of stalk production is crucial for determining ecological impacts, physiological regulation, and control of *D. geminata*. We have little understanding of the biology and ecological roles of *D. geminata*, and we need basic information to determine the causes and conditions that lead to nuisance blooms and the geographic expansion of this diatom. The first step is to determine the signal and regulatory genes in *D. geminata* that may be activated in response to environmental cues to result in excessive stalk production. Other cymbelloid diatoms produce stalks that are close in chemical composition to *D. geminata*. These species (notably varieties of *Cymbella mexicana*) may also produce excessive amounts of stalks, leading to nuisance growths.

GEOGRAPHIC DISTRIBUTION

North America

In North America, historical reports of *D. geminata* are sparse and voucher specimens are uncommon. Although it is not possible to state the historical range of this diatom with confidence, historical distributions were considered to be northern circumboreal in cold, oligotrophic waters. The earliest published records of *D. geminata* from North America on Vancouver Island, British Columbia (Cleve 1894-1896); however there are no notes on its abundance. Nearly one hundred years later, *D. geminata* formed nuisance blooms in the Heber River and over a period of five years had spread to twelve other watersheds on Vancouver Island (Sherbot & Bothwell 1993, Bothwell et al. 2005).

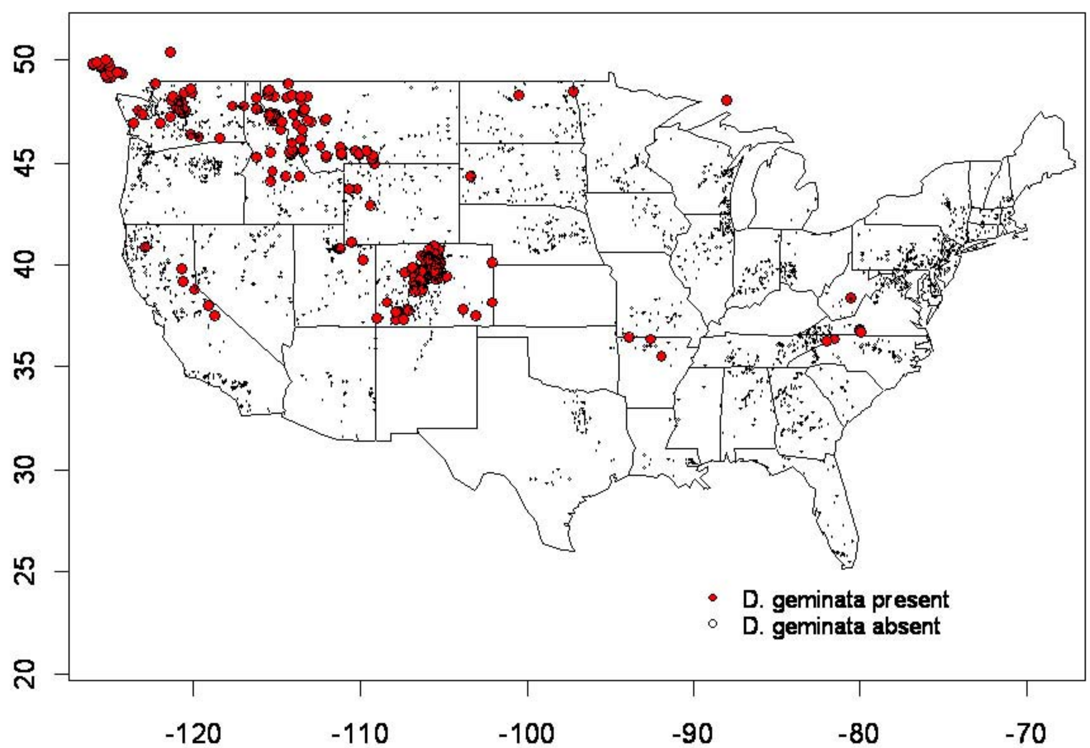

FIGURE 6. Confirmed presence and absence records of *D. geminata* in the United States. A total of 4569 samples were included and *D. geminata* is present in 283 sites. Records are based on data from USGS National Water Quality Assessment (NAWQA), EPA Environmental Monitoring and Assessment (EMAP), and samples from other studies. (Map by Sarah Spaulding, US Geological Survey).

Since that time, nuisance blooms have waned, but the diatom is present in more streams. In *The Diatoms of the United States*, Patrick & Reimer (1975) reported only one state, Virginia, as the distribution for *D. geminata* in the United States. More recent works consider *D. geminata* as present in rivers in the western United States (Bahls 2004). A pattern of expanding range and nuisance populations has developed in North America over the past several years (Fig. 6) (Pryfogle et al. 1997, Holderman & Hardy 2004, Shelby 2006), as well as in Europe and New Zealand (Fig. 7).

Europe

In European countries, reports are variable concerning the extent of *D. geminata* in streams and rivers. Northern and western rivers of the United Kingdom are subject to large masses of *D. geminata*, but the growths are considered to be a natural phenomenon and have been recorded for over 150 years. There are no reports of geographic expansion or increase in biomass of *D. geminata* (Whitton & Crisp 1984). Likewise, although masses of *D. geminata* increased with regulation of streamflow (Skulberg 1982), the formation of blooms is considered a normal event.

In Icelandic rivers, *D. geminata* formed large blooms beginning in the early 1990's (Jónsson et al. 2000). Blooms had no relation to bedrock geology or specific conductance, that is, the distribution and biomass of extensive mats appeared to be unrelated to water chemistry. Since the 1990's, populations of *D. geminata* have decreased, or remained stable. Icelandic rivers are vital to the salmon fishery and there was concern that the masses of *D. geminata* would negatively impact spawning, yet there is no clear evidence of a negative influence of *D. geminata* on fish stock.

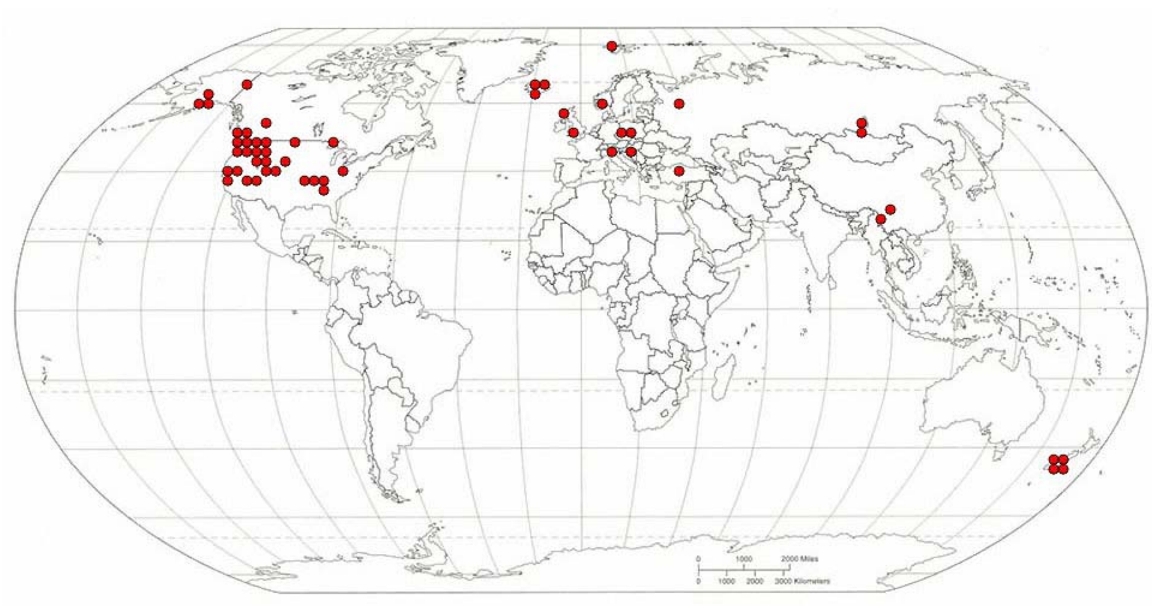

FIGURE 7. Confirmed presence and published records of *D. geminata* from around the world. Dots do not represent number of reports, but show rough geographic area of populations. (Map by Sarah Spaulding, US Geological Survey).

High abundances of *D. geminata* were documented in several rivers of the Carpathian, Gorce, and Tatra mountains of Poland (Kawecka & Sanecki 2003, Noga 2003). Observations of extensive growths and their expansion to new watersheds was contrasted to observations from the 1960's when *D. geminata* was present, but occurred in low abundance. The rivers where *D. geminata* formed large masses in recent years are impacted by anthropogenic nutrient input, with river concentrations of nitrate (NO_3) ranging from 1.7 to 3.8 mg/l and phosphate (PO_4) ranging from 13 to 100 µg/l (Kawecka & Sanecki 2003). The discovery of nuisance *D. geminata* populations in high nutrient waters was the first recognition that the species was appearing outside its recognized ecological range.

Similar to rivers in Poland, the eutrophic Tisa River in Serbia was reported to contain *D. geminata* throughout most of the year (March through November) (Subakov-Simić & Cvijan 2004). The Tisa River had temperatures above 20 °C for three months of the year, as well as high concentrations of ammonia (NH_3) (0.67 mg/l) and metals. This report also presents evidence that *D. geminata* is able to grow well at high temperatures and in polluted sites. Such a finding is repeated in the Değirmendere River in Turkey, where irrigation return flows, municipal wastes, and other inputs heavily influence water chemistry (Kara & Şahin 2001). At this site, *D. geminata* was found in high abundance for several months of the year.

New Zealand

The first confirmed record of *D. geminata* in the southern hemisphere was in October of 2004, in the lower Waiau River of the South Island of New Zealand (Kilroy 2004). Despite a proactive response of containment by the New Zealand government, within 18 months *D. geminata* spread to 12 rivers on the South Island (Fig. 8) and formed excessive blooms in several sites. The blooms in New Zealand demonstrate that *D. geminata* is an aggressive invasive species with dramatic ecological, economic, social, hydropower, recreational, and aesthetic impacts (Kilroy et al. 2005a, 2005b, 2005c, 2006, Campbell 2005, Branson 2006).

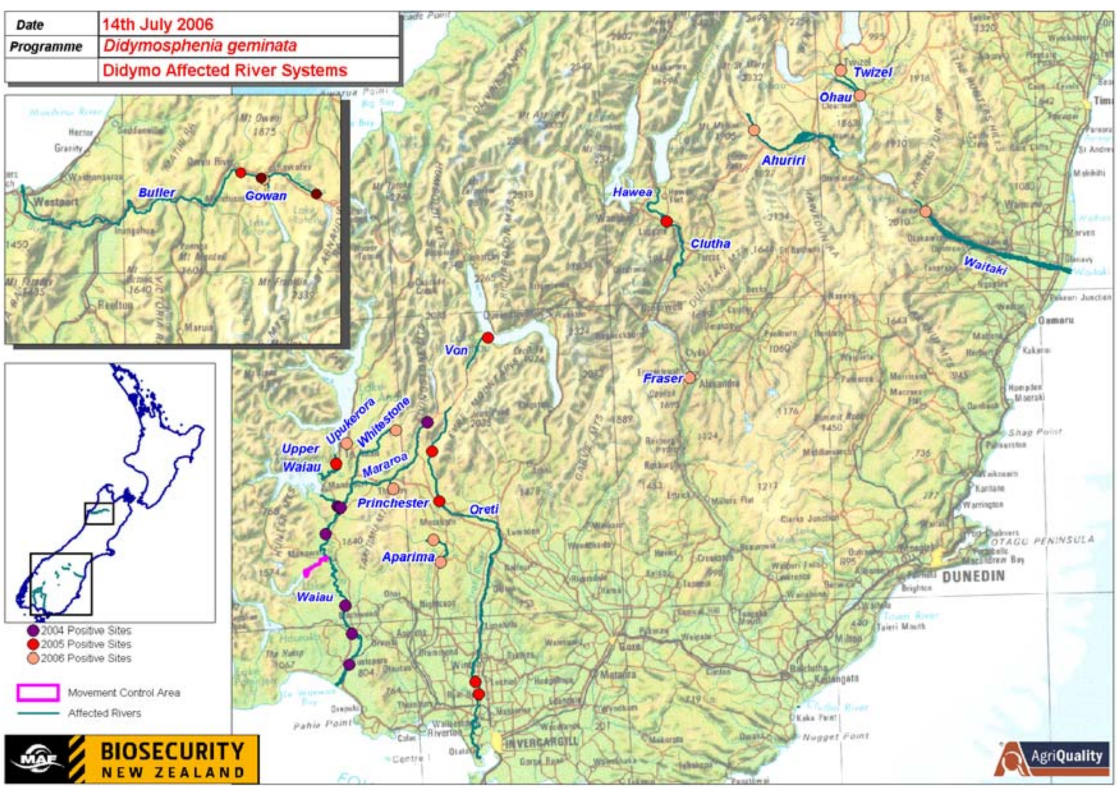

FIGURE 8. Confirmed presence of *D. geminata* in New Zealand as of July 2006.

Biosecurity New Zealand, the branch of government responsible for invasive species, identified *D. geminata* as harmful and of great concern. There is widespread agreement that *D. geminata* was introduced through human activity, in fact, penalties of up to five years imprisonment and a fine of $100,000 NZ are in place for intentionally spreading *D. geminata*. As of November 2006, *D. geminata* has not been confirmed in any locations on the North Island of New Zealand.

Prior to the incursion in New Zealand, knowledge of ecosystem roles and impacts of *D. geminata* was primarily anecdotal (Kilroy 2004). At the present time, several scientific and technical studies have been completed, or are in progress to address identification, detection, distribution, containment, impact, and control or eradication of *D. geminata* in New Zealand (see Appendix A for Biosecurity NZ website). As a result of work in New Zealand, the ability of *D. geminata* to survive outside water and requirements to decontaminate aquatic gear from live cells has been experimentally established. The range of *D. geminata* in terms of hydraulic habitat, temporal changes in biomass, and relation to density of benthic invertebrates has been investigated (Kilroy et al. 2005d). The interaction of flows and the likelihood of *D. geminata* transport to vital hydropower sites in Lake Manapouri were established (Biggs et al. 2005, Sutherland et al. 2005). Studies to determine the effects of *D. geminata* on native fish and invertebrates (benthic and drift) and water quality (dissolved oxygen and pH) are in progress. Other studies will address impacts of *D. geminata* on productivity of trout, develop molecular detection methods, and establish efficient monitoring efforts.

Global view of suitable stream habitats
A global distribution map based on ecological niche models shows suitable stream habitats for *D. geminata* on every continent except Antarctica (Fig. 9) (McNyset & Julius, 2006). While historical records in North America are poor, this map presents very different picture from the distribution of *D. geminata* (given in the United States as Virginia) (Patrick & Reimer, 1975). We now know that *D. geminata* can thrive in a wide range of physical and chemical conditions within rivers and spread by humans is of concern. Rivers in the southern hemisphere are particularly at risk to new introduction and invasion. Appropriate agency personnel in Australia, Argentina, Chile and Peru should be notified and made aware of the potential ecological damage and urgency of implementing decontamination procedures.

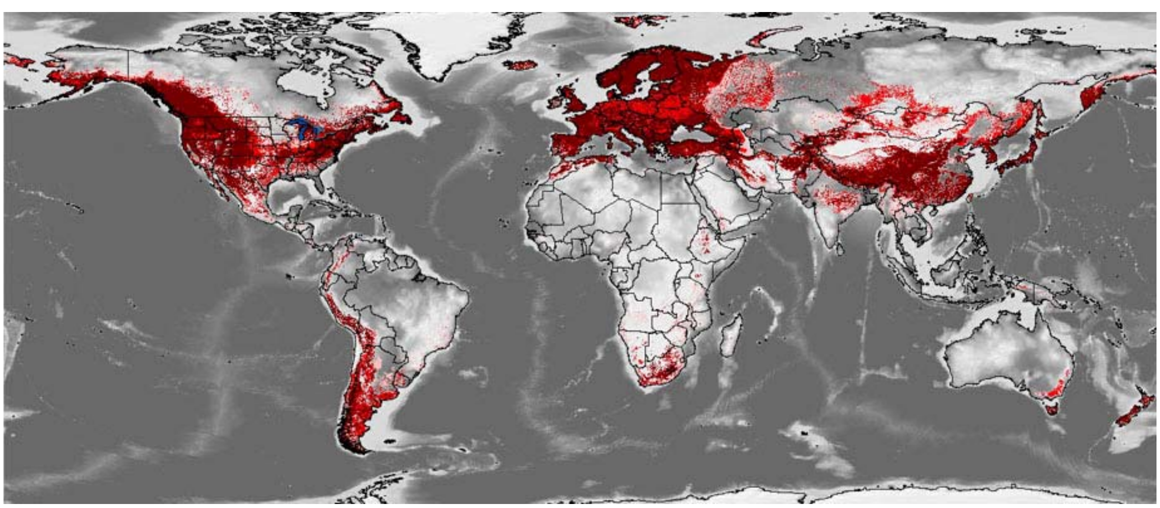

FIGURE 9. Map of the world showing regions where suitable stream habitats for *D. geminata* are located. Results for Australia are preliminary (Map by Kris McNyset, US Environmental Protection Agency).

ECOLOGICAL RELATIONSHIPS

The physical, chemical, and biological properties of streams and their organisms are intimately tied (Hynes 1975). *Didymosphenia geminata* both influences the stream environment and is controlled by environmental features. This diatom is capable of producing such great amounts of stalk that the mats covering the stream bed result in changes in ecological properties of the stream (e.g., species diversity, population sizes, nutrient pools) (Larned et al. 2006). Algal, invertebrate, and fish species diversity and population sizes may be altered. In addition, high growth rates and extensive mats of *D. geminata* may impact ecological processes such as ecosystem metabolism and nutrient cycling. Stalk and algal biomass, formation of nuisance blooms, legacy of stalks, interactions with invertebrates, interactions with fish, control by water chemistry and hydrology, impact on dissolved oxygen, and seasonal cycles are all part of how this organism exerts its influence on its stream and how it is also controlled by environmental features.

Stalks responsible for high biomass

A comparison of *D. geminata* biomass as ash free dry mass (AFDM) and chlorophyll *a* confirms that the mats are accumulations of stalks with a thin surface layer of cells (Larned et al. 2006). The AFDM biomass of *D. geminata* was measured to be 250 times greater than the chlorophyll *a* biomass. The comparison also indicates that the ecological interactions related to *D. geminata* are primarily due to the impact of the extracellular stalks, not the cells themselves. Blooms of *D. geminata* generate biomass and chlorophyll values many times those found in non-bloom conditions. Furthermore, AFDM biomass is produced at a level considered indicative of a biologically impaired river. In New Zealand rivers, an analysis of AFDM and chlorophyll *a* exceeded national guidelines for periphyton biomass (Table 1) (Kilroy et al. 2005a). The guidelines are intended to maintain high quality angling and fish habitat, and values are much higher than in non-*D. geminata* streams in New Zealand and elsewhere.

TABLE 1. Minimum and maximum ash free dry mass (AFDM) and chlorophyll *a* (Chl. *a*) for periphyton cover from studies in New Zealand rivers. Sites with large masses of *D. geminata* are shown in bold text. (Data from Kilroy et al. 2005a).

River	AFDM(g/m^2) min.	AFDM(g/m^2) max.	Chl. *a* (mg/m^2) min.	Chl. *a* (mg/m^2) max.	Reference
Mararoa	**18**	**1171**	**145**	**1029**	Kilroy et al. 2005a
Lower Waiau	**34**	**210**	**157**	**1155**	Kilroy et al. 2005a
Ohau	10	63	~2	~55	Biggs & Hickey 1994
Quebec streams	2.4	22.6	5.1	54.6	Bourassa & Cattaneo 1998
Mataura	~2.5	45	-	-	Biggs et al. 1998
Waiau	-	-	~0.3	~200	Biggs et al. 1998

Nuisance blooms

Although *D. geminata* occurs in both lakes and flowing waters, nuisance blooms are only known in streams and rivers. In contrast to historical, episodic growths of *D. geminata*, nuisance blooms are masses of cells and stalks that extend for greater than 1 km and persist for several months of the year. During a nuisance bloom, *D. geminata* cells produce copious amounts of extracellular stalk material.

For the purposes of this document, the phrase “nuisance bloom” refers to growths in sites where *D. geminata* was considered within its native range (northern boreal and high elevation sites), but where benthic mats are extensive spatially and temporally (Table 2). For example, nuisance blooms in Rapid Creek, South Dakota are present over a 5 to 10 km reach, at 30 to 100% coverage, for over 4 months of the year and are recurring. “Invasive blooms” refer to appearance of *D. geminata* in locations with no previous record (e.g., New Zealand) and denotes the behavior of an introduced non-indigenous species.

TABLE 2. Some of the rivers in North America with documented nuisance blooms. The list is likely to be much more extensive. Documenting the occurrence and extent of blooms is problematic based on existing monitoring efforts because measurements may not be made at the appropriate spatial scale.

PROVINCE/STATE	RIVER	YEARS	REFERENCE
Alberta	Red Deer Bow	Late 1990's	A. Kirkwood, U. Calgary
Arkansas	Red White	2005	Shelby 2006
British Columbia	Heber Englishman Nahmint Stamp/Somass Puntledge	1985 1993 1995-1997 1993,95,96,01,02 1997,02,03	British Columbia, Ministry of Environment
California	American Fork	Mid 1990's	S. Spaulding, US Geological Survey S. Lehr, California Dept. Fish & Game
Montana	Kootenai	2001-?	Holderman & Hardy 2004
Tennessee	Clinch South Holston	2005	T. Baker, Tennessee Valley Authority
South Dakota	Rapid Creek	2002	J. Shearer, South Dakota Dept of Game Fish & Parks
Virginia	Jackson Smith	2006 2006	S. Smith, Virginia Dept. of Game and Inland Fisheries

In a broad sense, nuisance algal blooms are typically directly related to anthropogenic increases in nutrient input to surface waters (Schindler 1977, Anderson et al. 2002). Increased concentrations of nitrogen and phosphorus result in adverse effects due to excessive primary production of algae. Cyanobacterial (blue-green algae) blooms are a well-known phenomenon in freshwater with high quantities of phosphorus (Jacoby et al. 2000, Bowling 1994, Hecky & Kilham 1988). In contrast, blooms of *D. geminata* are unlike other algal blooms, because they are associated with nutrient-poor waters. Notably, many *D. geminata* blooms have occurred in stream habitats generally considered pristine or with limited ecological disturbance (Jónsson et al. 2000, Sherbot and Bothwell 1993).

In North America, documenting the occurrence and extent of *D. geminata* is problematic. Standard counting techniques for diatom analysis underestimate the presence of *D. geminata* in the western United States by at least 50% (S. Spaulding, U.S. Geological Survey unpublished data). Compared to other diatom species, *D. geminata* has much larger cells (80-150 μm in length); yet smaller cells dominate the diatom community (Fig. 10). Counting procedures intended to evaluate diatom species in the periphyton are often based on a fixed count (e.g., 300 cells counted), which favor small, numerous species. An alternative technique is to note the presence of *D. geminata* cells, even if they do not appear in standard analysis. Interestingly, *D. geminata* never comprises greater than 3% of the diatom community in western streams (EPA Environmental Monitoring and Assessment Program data), even within samples collected from nuisance blooms. Reports of data using biovolume avoid part of the problem (Jónsson et al. 2000), by reporting abundance in terms of biomass, rather than number of cells.

A measure such as the visual biovolume index (Kilroy et al. 2005d) is a preferred method to estimate the abundance and impact of *D. geminata*. The visual biovolume index is a measure of the percent coverage of *D. geminata* on a cross section of stream channel, multiplied by the thickness of the mat. The index takes the amount of extracellular stalk into account and is more appropriate for documenting the extent of nuisance blooms. In order to track the geographic distribution of *D. geminata* on a global scale, it is important to use effective and proper documentation of sites and archive voucher samples.

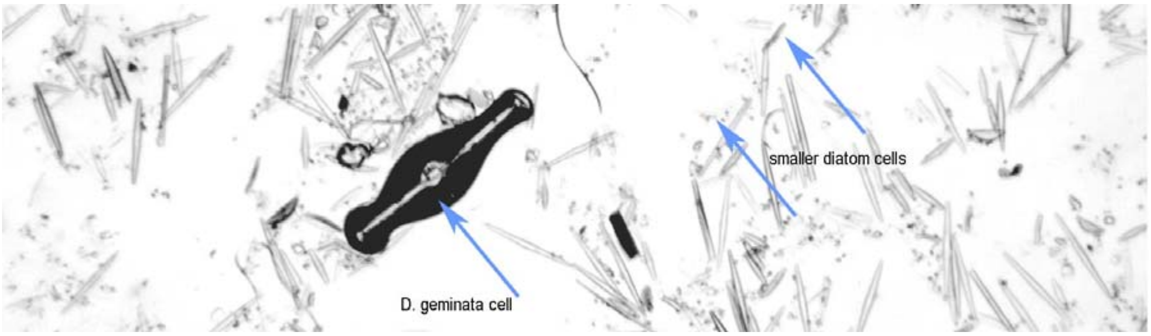

FIGURE 10. Microscope image of diatoms showing relative size of cells. *Didymosphenia geminata* is underestimated in terms of presence because standard counting techniques are directed at small species. (Image by S. Spaulding, U.S. Geological Survey).

Legacy of stalks

The extracellular stalk of *D. geminata* is a complex, multi-layered structure, resistant to degradation in streams. Observations in Colorado streams show that stalks persist up to 2 months following a peak in growth of *D. geminata* (S. Spaulding, U.S. Geological Survey unpublished data). In effect, the stalks persist in the stream longer than the cells that produced them (Fig. 11). Furthermore, the stalks trap fine sediment within their dense matrix and change the nature of the stream substrate. This coating of the stream benthos may then act to control the algae and invertebrate species able to feed and move on those surfaces. The legacy of the *D. geminata* stalks is a potentially strong influence on stream community composition. It is important to evaluate the apparent resistance of the stalk to degradation by bacteria and fungi, and determine ecosystem effects of stalk material.

FIGURE 11. Image of a rock coated with *D. geminata* stalks and fine sediment. The cells of the diatom are no longer present, but the stalks continue to determine the nature of the stream substrate. (Image by S. Spaulding, U.S. Geological Survey).

Interactions with invertebrates

Abundance and diversity of benthic macroinvertebrates are likely to be affected by *D. geminata* in two ways: direct trophic interactions and habitat interactions (Larned et al. 2006). Direct trophic interaction refers to utilization of *D. geminata* as a food source. Macroinvertebrate species that consume *D. geminata* are expected to be favored over those species that don't eat *D. geminata*. Habitat interaction refers to utilization of stream surfaces by macroinvertebrates.

Species that require exposed sediment are expected to be negatively impacted by extensive coverage of *D. geminata.* Recent work in New Zealand, Colorado, and Montana indicates that there are complex interactions between *D. geminata* and benthic macroinvertebrates.

Results from New Zealand rivers indicate that both number of species and density of invertebrates was greater with higher *D. geminata* coverage (Larned et al 2006). However, few of the species present were characteristic of high river health. With lower amounts of *D. geminata* coverage, invertebrate abundance and diversity increased, forming a more even distribution of species within the community. In contrast, results from Colorado rivers indicate that high densities of *D. geminata* were related to a decline in total macroinvertebrate richness (T. Schmidt, personal communication). In these rivers, the macroinvertebrate community was dominated by chironomids (midge fly larvae). Analysis of macroinvertebrate gut contents showed that mayfly, stonefly, caddisfly, and chironomid larvae consumed *D. geminata*, but that the presence of *D. geminata* in guts was related to body size. That is, the results suggest that small macroinvertebrates were not able to consume *D. geminata*. Results from surveys in Montana (D. Beeson, personal communication) showed dramatic increases in *D. geminata* cover (up to 100% over monitoring period of 1998-2003). This study also indicated an increase in diptera taxa (including midge fly larvae) and loss of mayflies, stoneflies, and caddisflies.

These initial results suggest that the impact of *D. geminata* on aquatic macroinvertebrates is directly related to temporal and spatial extent of nuisance blooms. If *D. geminata* masses are capable of altering the taxonomic composition and size of benthic macroinvertebrates present in the drift, that relationship represents a trophic level impact. Further work should resolve the differences in impacts of *D. geminata* at both high and low densities and whether there are threshold levels of nuisance growths. In addition, it would be beneficial to determine the extent to which macroinvertebrate grazing reduces *D. geminata* abundance. An open question is the degree to which macroinvertebrates are physically able to move through the masses of stalks to gain access to the nutritious cells.

Interactions with fish

Studies on the effects of *D. geminata* on native New Zealand fish are in progress (Larned et al. 2006). Given large amounts of non-nutritious stalk material present on stream substrates in affected areas, *D. geminata* is predicted to have deleterious effects on native fish. Fish that inhabit benthic habitats, consume benthic prey, and nest beneath or between cobbles are expected to be the most impacted because they utilize the same habitat as *D. geminata* (Larned et al. 2006). Nuisance growths of *D. geminata* have the potential to impact fisheries through food web interactions with aquatic macroinvertebrates. That is, if the favored food sources for fish are impacted in a negative way, fish will also be impacted negatively.

Water chemistry

Water chemistry is typically considered a controlling factor for diatom distribution and abundance, particularly nutrient concentrations and pH. Historically, *D. geminata* was considered to be restricted to oligotrophic (low nutrient) and low temperature waters, and a broad range of conductance in the European Alps (Krammer & Lange-Bertalot 1986). Although historical values of chemical and physical parameters in relation to *D. geminata* biomass were not recorded, there is a widespread understanding among diatomists and aquatic ecologists that *D. geminata* had narrow environmental tolerances. Therefore, one of the commonly noted observations about this diatom is the expansion of its ecological tolerance to a broader physical and chemical range (Kawecka & Sanecki 2003, Kilroy 2004).

Preliminary data from a random survey of streams in the western United States (Stoddard et al. 2005) show that *D. geminata* is present in a wide range of freshwater conditions (Fig. 12). These data are presented based on presence/absence of *D. geminata* in the western Environmental Monitoring and Assessment Program (EMAP) pilot. Rather than being restricted to cold temperatures, *D. geminata* is present in waters from 4 to 27 °C, and shows a temperature range greater than what was previously observed. The relation of *D. geminata* presence to pH is narrow, with *D. geminata* found in waters at, or above, a pH of 7.

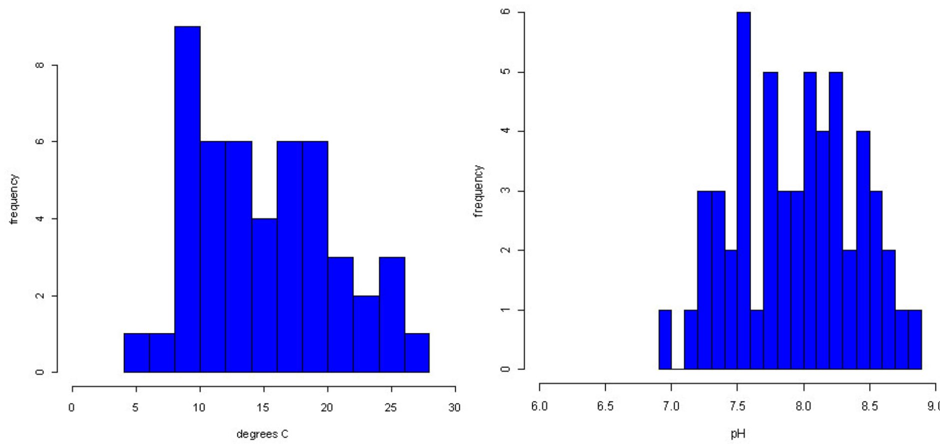

FIGURE 12. A) Water temperature versus frequency of sites with *D. geminata* present in western streams of the United States. B) pH versus frequency of sites with *D. geminata* present. (Data from EPA EMAP Western Pilot for 2000-2003.)

The range of specific conductance and acid neutralizing capacity (ANC) at sites with *D. geminata* present are both broad (Fig. 13). These data demonstrate a wide range of tolerance from electrolyte poor to concentrated waters, although *D. geminata* occurs more often at lower values of conductance and ANC.

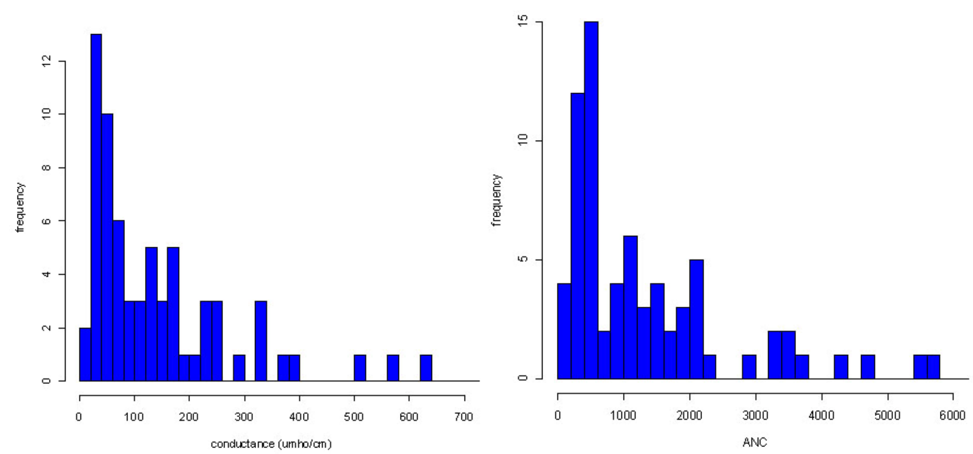

FIGURE 13. A) Conductance (µmho/cm) versus frequency of sites with *D. geminata* present in western streams of the United States. B) Acid neutralizing capacity (ANC) versus frequency of sites with *D. geminata* present. (Data from EPA EMAP Western Pilot for 2000-2003.)

Although *D. geminata* occurs most frequently in waters with low total phosphorus (< 2 µg/l) and low nitrate (< 1 mg/l) (Fig. 14), it can also be found where both of these nutrients are present at very high concentrations. These values show where *D. geminata* is present, but give no indication of the biomass or growth rate in association with nutrient concentration. Furthermore, it is unknown if *D. geminata* is limited by either of these important nutrients in any streams in North America. In New Zealand, nutrient enrichment experiments indicate that growth of *D. geminata* is limited by nitrogen, phosphorus, or both nutrients within most of its current range (Larned et al. 2006). In other words, with greater concentrations of either nutrient growth would be stimulated. Increased loading of nutrients to affected rivers by watershed sources is expected to result in increased growth of *D. geminata*.

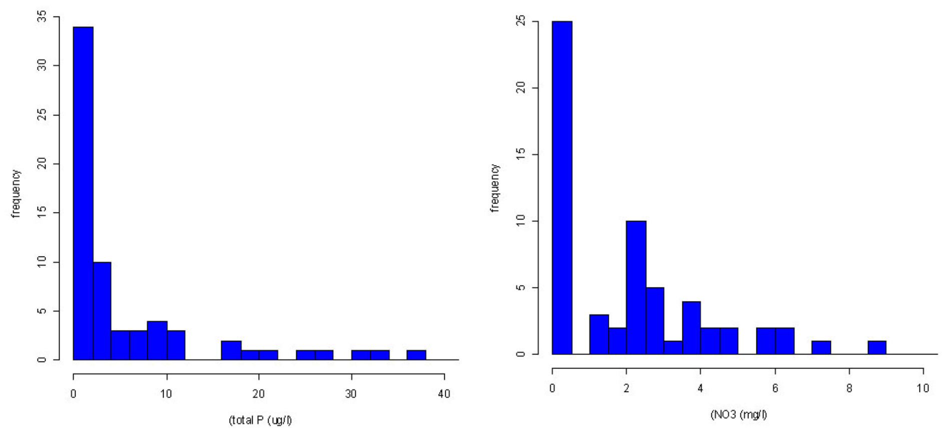

FIGURE 14. A) Total phosphorus (µg/l) versus frequency of sites with *D. geminata* present in western streams of the United States. B) Nitrate (mg/l) versus frequency of sites with *D. geminata* present. (Data from EPA EMAP Western Pilot for 2000-2003.)

Hydraulic range

Didymosphenia geminata thrives in a wide range of hydraulic conditions (Fig. 15) (Kilroy et al. 2005c). The hydraulic range is striking, because dense mats of the alga are able to grow in slow moving, shallow waters as well as waters with greater depth and velocity than could be safely measured by technicians. In the Mararoa and Waiau rivers, masses of *D. geminata* were greatest at water velocities of approximately 0.5 m/s. With stable flow, biomass of *D. geminata* tends to

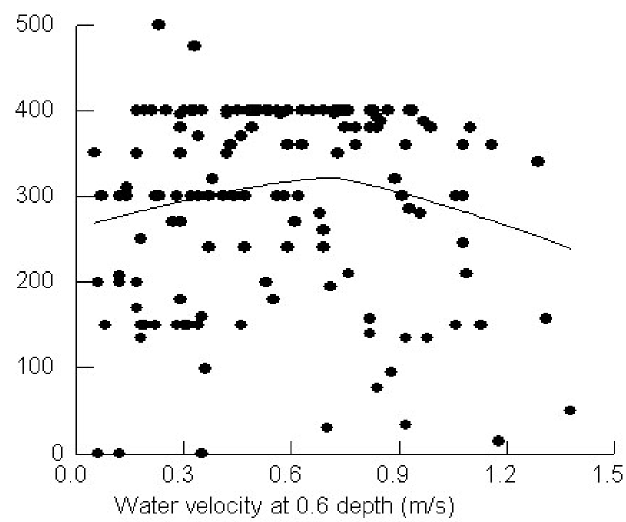

FIGURE 15. Water velocity versus visual biovolume index in the Mararoa River, New Zealand. The pattern indicates there is no relation between water velocity and visual biovolume index. *Didymosphenia geminata* forms dense mats (high visual biovolume index) from low to high water velocities. (Data from Kilroy et al. 2005c).

increase. In fact, the best hydrological predictor of *D. geminata* biomass is number of days since a flood greater than 75 to 100 m^3/s. In other words, large floods scour the river bed and return biomass to a low level. However, in order to reduce cell biomass, floods must be high enough to cause the rocks on the streambed to mobilize (Larned et al. 2006), scouring the cells from rock surfaces.

In North America and Europe, high density blooms are frequent in rivers directly below impoundments (Skulberg 1982, Dufford et al. 1987, Kawecka & Sanecki 2003). A monthly survey of rivers in Alberta, Canada suggests that *D. geminata* occurs with higher frequency in locations where flow and temperature is regulated by dams compared to non-regulated rivers (A. Kirkwood, University of Calgary, personal communication). In these river reaches, stable flows and fairly constant temperatures favor development of large masses of *D. geminata*. Restoration of historic, or pre-impoundment, natural flows in rivers may mitigate nuisance blooms, as well as restore river condition.

A biological paradox
Recent work on *D. geminata* blooms has resulted in a remarkable observation. Within the masses of extracellular stalks and cells, concentrations of dissolved oxygen are supersaturated with respect to the atmosphere (Larned et al. 2006). Determining the source of nutrients and flux of oxygen within the algal mats is likely to reveal how *D. geminata* attains its remarkable biomass. Typically, the concentration of dissolved oxygen within algal mats formed by other species is not supersaturated, but oxygen concentrations may be quite low as cells respire and decompose. In contrast, peak values of dissolved oxygen are present well below the surface of the *D. geminata* mats. Larned et al. propose that these algal mats contain other photosynthetic organisms that are actively producing oxygen. They suggest that a unique assemblage of organisms is able to utilize high concentrations of dissolved nutrients produced in organic matter at the bases of mats, and then transfers these nutrients to *D. geminata* cells. An investigation of the processes within the mat matrix will lead to addressing the biological paradox of how *D. geminata* produces excessive biomass in low nutrient streams and rivers, over short periods of time.

Molecular markers
Craig Cary, University of Waikato, New Zealand is leading an effort to elucidate a genetic marker that allows a quick, inexpensive, and reliable method for determining the presence of *D. geminata* within a watershed. The Cary laboratory has already been successful in determining DNA sequences unique to *D. geminata*, and the method is promising for monitoring efforts (Cary et al. 2006). Following this work, the expanding distribution of *D. geminata* has prompted a genetic survey to determine: 1) How genetically related are populations of *D. geminata* around the world? 2) Are there one, or more, “source” populations that are able to spread to new sites? 3) Has there been a genetic change in one or more populations that have led to invasive behavior? In spring of 2006, Cary initiated a broad request to the scientific and management community to contribute samples of *D. geminata* for a global population study. Samples have been contributed from the broadest distribution possible, including but not limited to, representative samples from Asia, Europe, and North America.

In determining if there has been a genetically based physiological change linked to a nuisance strain, it is important to consider different types of change. Physiological changes may include general physiological changes, including such processes as photosynthetic capacity and ecological tolerances. Such changes might allow expansion into new habitats. In addition, there may be specific changes in mode of production of stalk, that is, alterations in the synthesis of extra polymeric substances (EPS). Have the signal and regulatory genes in *D. geminata* become activated in response to environmental cues to result in excessive stalk production? It is critical to determine if enzymes (such as glycan synthase and glycosyl transferase) are responsible for the excessive stalk production. Other diatom species produce stalks and may also have the potential to become nuisance species if EPS production becomes "turned on".

RANGE EXPANSION

The mechanisms for *D. geminata* to expand its range to new watersheds are not well understood. Early suggestions that increases in UV-B radiation was tied to the expansion were not supported (Sherbot & Bothwell 1993, Wellnitz et al. 1996, Rader & Belish 1997). Recent work illustrates the capacity of *D. geminata* to survive outside of the stream environment as well as potential vectors in its spread. Cells are able to survive and remain viable in cool, damp, dark conditions for at least 40 days (Kilroy 2005). Fishing equipment, boot tops, neoprene waders, and felt-soles in particular, all provide a site where cells remain viable, at least during short term studies (Kilroy et al. 2006). At the same time, prime destinations for fishing are becoming more popular with anglers. Rather than frequent a favorite local fishing site, it is now common that anglers travel to multiple, or distant destinations for fishing vacations. Moreover, they may be fishing in a river less than twenty four hours after leaving their local rivers in North America, and unknowingly spreading *D. geminata*.

FIGURE 16. *D. geminata* is able to survive on boot tops, neoprene waders, and felt-soles and may be spread to distant sites (Image by Sarah Spaulding, US Geological Survey).

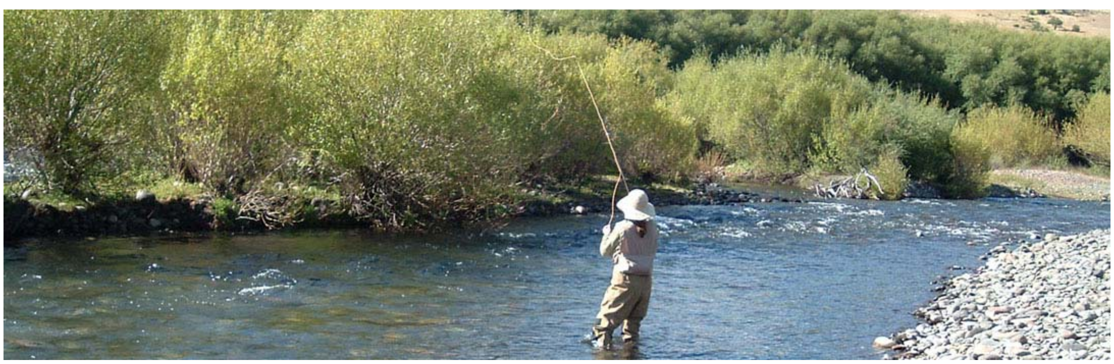

FIGURE 17. An increasing number of anglers from North America are visiting other continents to fish, as illustrated by this tourist in Rio Malleo, Argentina. Ecological models predict that rivers in South America and Australia contain suitable habitat for *D. geminata*. (Image by Matt Wilhelm, Federation of Fly Fishers).

For aquatic organisms, the relationship between the spread of invasive species to recreation is well established (e.g., Eurasian water milfoil (*Myriophyllum spicatum* L.) and zebra mussels (*Dreissena polymorpha*)) (e.g., Madsen et al. 1988, Strayer et al. 1996, Vitousek et al. 1997, Schneider et al. 1998, Johnson et al. 2001). Gear and equipment used in aquatic recreation is being tested for its role in spreading *D. geminata*, but it is possible that humans transport *D.*

geminata in other ways (e.g., boats including jet skis, water transport for rural fire fighting, irrigation, water diversions, waterfowl hunting, and float airplanes). Determining the likely risk of such vectors may be valuable for targeting control programs and public messages about decontamination.

ECONOMIC IMPACT

While *D. geminat*a is not considered invasive in the United States, the diatom's nuisance blooms has economic impacts. The human population of western United States is closely dependent on a system of canals to transport water for hydropower generation, agriculture, and human consumption. Nuisance algae, including *D. geminata*, regularly thrive on the stable substrate and flow regime of canal systems (Pryfogle et al. 1997). In some canal systems, managers implement regular removals by scraping *D. geminata* growths from the concrete surfaces of canals (Fig. 18).

FIGURE 18. Stalks of *D. geminata* clog a grate in a water supply canal in California (Image by Peter Pryfogle, Idaho National Laboratory).

Didymosphenia geminata is often reported by recreationalists to land managers as being unsightly (see Appendix D). The stalks are frequently mistaken for raw sewage, leading homeowners and recreationalists to complain to local water treatment plants. Many communities rely on tourism dollars that are generated by outdoor recreation. Natural resource opportunities represent important economic value, yet they may be vulnerable to damage by the spread of nuisance species. In the United States, the cost to control and eradicate nuisance and invasive species is estimated at $120 billion annually, with $1 billion from the impacts of invasive zebra mussels alone (Pimentel et al. 2005).

Upon the appearance of *D. geminata* in New Zealand in October 2004, Biosecurity New Zealand initiated a national incursion response based on the potential losses to the national economy. The presence of *D. geminata* threatens the opportunities for tourists to experience clear, unimpacted rivers. Commercial eel fisheries, water supplies, tourism, and biodiversity values are projected to be impacted and economic losses are estimated at between NZ $57 and 285 million over a period of eight years (Branson 2006).

CONTROL TECHNIQUES

Biosecurity New Zealand is currently pursuing a series of experimental trials to test biocides for potential control of *D. geminata* within streams and rivers in New Zealand (Jellyman et al. 2006). In order to test the efficacy of various biocides, *D. geminata* was grown on artificial substrates and placed in experimental stream channels. Several biocides were tested on *D. geminata*. The mats were exposed to each biocide for a period of one hour and the viability of algal cells determined at various time periods, up to 28 days after treatment. Mortality of fish in the experimental stream channels was also assessed. Of the five biocides tested, chelated copper had the greatest negative effect on *D. geminata* for all contact times. In the next stages, the tolerance limits of fish to chelated copper will be established. Although copper compounds have a long history of use as algaecides the United States, in lakes, reservoirs, and to a lesser extent, flowing waters, they have not been evaluated for control of *D. geminata* outside of New Zealand.

REDUCE THE SPREAD

Plants, animals and microscopic organisms, including *D. geminata*, may adhere to waders, boots, boats, float tubes, and angling gear. Cleaning gear before traveling between bodies of water, whether between nearby streams or for international destinations, is crucial. Decontamination of gear is the only way to prevent the spread and subsequent introduction of *D. geminata* into new watersheds. While decontamination will not destroy all invasive species, cleaning procedures minimize the possibility of spread. These simple treatments effectively destroy *D. geminata* algal cells (Kilroy 2005):

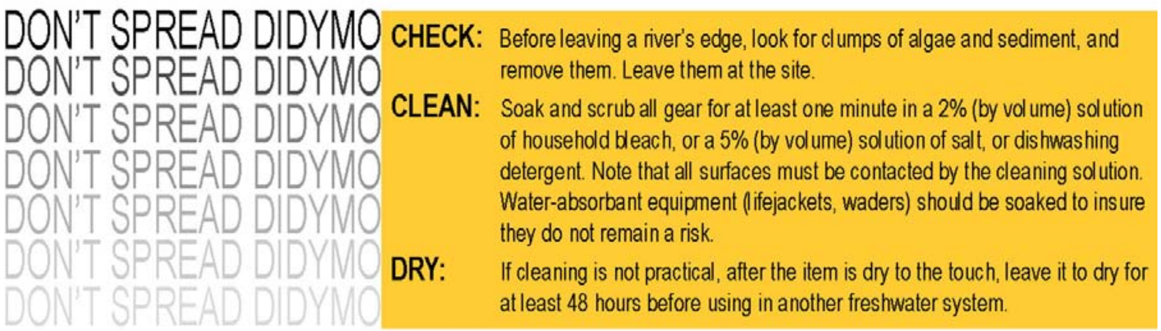

FIGURE 19. Recommended methods to prevent spread of *D. geminata* (Graphic from EPA information sheet).

An aggressive education and outreach program is required to change water resource user behavior in order to minimize spread of *D. geminata* on a global scale. A public awareness campaign, directed at freshwater anglers, boaters, professional guides, and other recreationalists must be integrated with existing invasive species programs. Freshwater resource users, including ecologists, water managers, fisheries biologists, and other scientists, need to be aware of the threat and should practice decontamination procedures to prevent the spread. Furthermore, members of the United States Aquatic Nuisance Species (ANS) Task Force must be informed of the distribution and impact of *D. geminata*, and include this organism within the scope of nuisance and invasive species within the United States.

SUMMARY

Didymosphenia geminata now has a broad distribution in North America, a condition that appears to have developed in the past ten to twenty years. Although the diatom is more common in the western United States, it is also forming large growths in rivers in the eastern United States and Canada. This diatom was known to produce large masses since the earliest historical records, but now the blooms are over a greater area in the Northern Hemisphere and spreading across rivers in the Southern Hemisphere While *D. geminata* was formerly considered to have narrow ecological tolerances, it is now present in streams exhibiting a wide range of chemical characteristics. It is capable of growing throughout most of the year in streams with low to high NO_3 concentrations (< 1 mg/l - > 8 mg/l), low to high temperatures (4 - 27 °C), and within a broad range of light exposure. The diatom forms an unknown number of nuisance blooms in North America, covering benthic surfaces for greater stream reaches than 1 to 2 km. The diatom is invasive in New Zealand, and is rapidly expanding to new watersheds, despite aggressive control measures.

Didymosphenia geminata causes us to question our fundamental understanding of streams and rivers. First, *D. geminata* presents a biological paradox; how is excessive biomass produced in low nutrient streams and rivers, over short periods of time? Second, *D. geminata* produces a mucopolysaccaride stalk that appears to be resistant to biodegradation by bacteria and fungi. What is the unique composition and structure of the stalk, and how does the stalk itself play a role in the success of this organism? Third, *D. geminata* has direct and indirect impacts across aquatic trophic levels. What is the long-term significance of stalks that are resistant to decomposition and trap fine sediment past the life span of the organism? Fourth, there are suggestions that macroinvertebrates and fish respond to nuisance levels of *D. geminata* with community and population level shifts in composition, abundance, and size class. What are the trophic impacts of *D. geminata*? Finally, is there a genetically based physiological change in this organism that is linked to a nuisance strain? Molecular markers present the opportunity to trace the genetic relationships of nuisance outbreaks and those records can be compared with models of predicted global distribution.

Scientists, conservationists and natural resources managers are concerned about nuisance blooms of *D. geminata* and change in behavior of this organism or appearance of a nuisance strain. As an outcome of the International *Didymosphenia* Symposium in Bozeman, Montana, two goals are clear:

1) Develop an outreach effort to inform and involve the public and government agencies
2) Develop an approach to research that will allow us to address the behavior and impacts of this organism.

This document and the following recommendations are intended to accomplish those goals.

RECOMMENDATIONS

- An aggressive education and outreach program is required to change water resource user behavior in order to minimize spread of *D. geminata* on a global scale.
    - A public awareness campaign, directed at freshwater anglers, boaters, professional guides, and other recreationalists must be integrated with existing invasive species programs.
    - Freshwater resource users, including ecologists, water managers, fisheries biologists, and other scientists, need to be aware of the threat and should practice decontamination procedures to prevent the spread.
    - Members of the United States Aquatic Nuisance Species (ANS) Task Force must be informed of the distribution and impact of *D. geminata*, and include this organism within the scope of nuisance and invasive species within the United States.
    - Rivers in the southern hemisphere are particularly at risk to new introduction and invasion. Appropriate agency personnel in Australia, Argentina, Chile and Peru must be notified and made aware of the potential ecological damage and urgency of implementing decontamination procedures.
- Determine if there has been a genetically based physiological change in this organism that is linked to a nuisance strain. Physiological changes may be:
    - General physiological changes, including such processes as photosynthetic capacity and ecological tolerances.
    - Specific changes in mode of production of stalk, that is, alterations in the synthesis of extra polymeric substances (EPS).
- Trace the relationships of nuisance outbreaks and those records can be compared with models of predicted global distribution using molecular markers.
- Determine the degree to which the spread of *D. geminata* is aided by specific human vectors, such as felt-soled waders, or other plausible mechanisms.
- Track the geographic distribution of *D. geminata* on a global scale using effective and proper documentation of sites and voucher samples.

- Determine the ecological conditions under which excessive biomass is produced in low nutrient streams and rivers, over short periods of time. Develop strategies to mitigate existing blooms.
- Determine the unique composition, structure, and cellular processes that produce the *D. geminata* stalk, which is responsible for its negative ecosystem impacts.
- Determine it perturbations in signal, regulatory, and/or synthetic pathways of stalk production by cymbelloid diatoms has resulted in increased production in *D. geminata*.
- Evaluate the apparent resistance of the stalk to degradation by bacteria and fungi, and determine ecosystem effects of stalk material.
- Investigate the contribution that *D. geminata* makes to nutrition of macroinvertebrates. Are macroinvertebrates able to access the cells from within the mass of stalks?
- Resolve the extent to which macroinvertebrate grazing can reduce *D. geminata* abundance.
- Determine the direct and indirect impacts of *D. geminata* and its stalks to aquatic macroinvertebrates and fish. Resolve the impacts of *D. geminata* at both high and low densities and whether there are threshold levels of nuisance growths. Testing the following hypotheses will clarify the potential impacts:
    - The impact of *D. geminata* on aquatic macroinvertebrates is directly related to temporal and spatial extent of nuisance blooms.
    - *D. geminata* masses alter the taxonomic composition and size of benthic macroinvertebrates present in the drift.
    - The presence of *D. geminata* alters the energetics of fish through altering the macroinvertebrates present in drift.
    - The reduction in food energy reduces the growth rate of trout, and favors small individuals over large individuals.

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Appendix A: Additional Resources
Available resources for more information about *Didymosphenia geminata* and aquatic invasive species.

Internet resources
Biosecurity New Zealand
www.biosecurity.govt.nz/didymo

United States Environmental Protection Agency
www.epa.gov/Region8/water/monitoring/didymosphenia.html

Federation of Fly Fishers
www.fedflyfishers.org/conInvasiveSpecies.php

Stop Aquatic Hitchhikers
www.protectyourwaters.net/

Global Invasive Species Database
www.issg.org/database/species

State of Arkansas Department of Environmental Quality
www.adeq.state.ar.us/water/didymo.htm

New Zealand Game and Fish – Video clip
www.southlandfishgame.co.nz/didymo.htm

Symposium abstracts
Abstracts of presentations at the International Symposium on *Didymosphenia* are available. The symposium was co-sponsored by the Federation of Fly Fishers and U.S. EPA Region 8, held in association with the Western Division American Fisheries Society Annual Meeting May 15-16, 2006, Bozeman, Montana, USA.
http://www.epa.gov/region8/water/events.html#agenda

Report occurrences
Report suspected growths of *Didymosphenia* by collecting a small sample (put a pinch of the material in a vial with ethanol or in a folded business card). Label samples with the date, latitude, longitude (provide detailed accurate site information). Send reports and samples to:

Dr. Sarah Spaulding
US Geological Survey
999 18th St., Suite 300
Denver, Colorado 80202 USA
Email: sarah.spaulding@usgs.gov
Tel: 303-312-6212

Appendix B: Scientific meetings

Presentations at scientific meetings are a vital form for communication of scientific findings. Issues stemming from *D. geminata* have stimulated a rapid and broad scope of research interests. Scientists have presented, or are scheduled to present, talks or posters on *D. geminata* at the following national and international scientific conferences:

CONFERENCE	DATE	LOCATION	LINK
Didymosphenia Symposium, American Fisheries Society	15-16 May 2006	Bozeman, Montana, US	http://www.epa.gov/region8/water/events.html
International Conference Aquatic on Invasive Species	15-18 May 2006	Key Biscayne, Florida, US	http://www.icais.org/
North American Benthological Society	4-9 June 2006	Anchorage, Alaska US	http://www.benthos.org/Meeting/
Phycological Society of America	7-12 July 2006	Juneau, Alaska US	http://www.psaalgae.org/ops/psa2006.shtm
Joint Conference of the NZ Ecological Society	28 Aug - 1 Sep 2006	Wellington, New Zealand	www.vuw.ac.nz/ecology06
International Diatom Symposium	28 Aug - 3 Sep 2006	Irkutsk, Russia	http://lin.irk.ru/ids2006/
International Conference on Harmful Algae	4- 8 Sep 2006	Copenhagen, Denmark	http://www.bi.ku.dk/hab/
NZ Freshwater Sciences Conference	26-30 Nov 2006	Rotorua, New Zealand	http://limsoc.rsnz.org/
American Society of Limnology and Oceanography	4- 9 Feb 2007	Santa Fe, New Mexico, US	http://www.aslo.org/meetings.html
Society for International Limnology	12- 18 Aug 2007	Montreal, Canada	http://www.sil2007.org/

Appendix C: Glossary

acid neutralizing capacity (ANC): A water chemistry measure that gives an indication of the ability of a water sample to counter the effects of acid without changing its pH. Water low in ANC may easily become acidic, while waters high in ANC are much more resilient. ANC is a measure similar to alkalinity, which is a measure of the buffering capacity of a water sample.

apical pore field: A structure that is part of the silica cell wall of diatoms. The apical porefield is an area of very fine pores, through which the mucilaginous stalk is secreted. *Didymosphenia geminata* has a single apical porefield on one end of each valve.

ash free dry mass (AFDM): A measure which indicates the amount of organic material present in a sample.

benthic or benthos: Refers to the bottom surface of a stream, river, or lake. The aquatic organisms that live in, on, or near the bottom surface are termed benthic organisms and they inhabit the benthos. Benthic organisms may include macroinvertebrates, algae, bacteria, fungi, clams, worms, and anything else that inhabits the bottom.

chlorophyll a: Chlorophyll is the primary pigment used by plants to obtain energy from the sun through photosynthesis. Chlorophyll *a* is a specific form of the chlorophyll molecule found in photosynthetic algae. The amount of chlorophyll *a* in a stream gives an indication of the amount of algal biomass present. High amounts of algal biomass are usually considered undesirable and indicative of increased nutrient loads.

chrysolaminarin (ß1,3 linked glucan): A molecule produced by some groups of algae including the diatoms. The material is composed of modified glucose, functions as a food reserve, and is stored within the cell.

cymbelloid: Referring to a group of freshwater diatoms within the Family Cymbellaceae. Cymbelloid symmetry is typically asymmetrical to both primary axes (the cells are crescent moons in shape). Although *D. geminata* is a member of this group (Kociolek & Stoermer 1993), it does not share the characteristic symmetry.

EDTA or ethylenediaminetetraacetic acid: A specific molecule that binds strongly to ions in a solution. In the example given here, EDTA is used to bind and separate fractions of the diatom stalk.

*eutrophic:*Waters that are high in nutrients, specifically phosphorus and nitrogen, are considered eutrophic. High concentrations of phosphorus and nitrogen often lead to correspondingly high algal productivity and biomass.

extracellular: Material that is located outside the boundaries of the cell wall. In the example of *D. geminata*, the stalk is produced within the cell but is then excreted outside the cell wall.

gomphonemoid: Referring to group of freshwater diatoms within the Family Gomphonemaceae. Gomphonemoid symmetry is typically symmetrical to the apical axis, and asymmetrical to the transverse axis (the cells are club shaped). Although *D. geminata* is often included in this group because it has gomphonemoid symmetry, Kociolek & Stoermer (1993) demonstrated it is a member of the cymbelloid lineage.

invasive species: Any species, including its seeds, eggs, spores, or other biological material capable of propagating that species, that is not native to that ecosystem; and whose introduction does or is likely to cause economic or environmental harm or harm to human health.

molecular markers: Specific sequences of genetic material (DNA) that are used to characterize or differentiate organisms.

mucopolysaccaride: A complex chain of molecules primarily composed of sugar molecules linked together to form a chain. The diatom stalk is composed of mucopolysaccarides.

nuisance bloom: The term “bloom” is traditionally applied to planktonic algae that form growths in lakes or oceans. It is not a “bloom” in the sense of flowering plants. Here, the term “nuisance bloom” is applied to the condition that *D. geminata* creates in streams because the growths threaten the diversity of other species, aquatic ecosystem function, or economic activities dependent on flowing waters.

oligotrophic: Waters that are very low in nutrients, specifically phosphorus and nitrogen, are considered oligotrophic. Such low nutrient waters are usually low in algal productivity and biomass.

periphyton: Although the strict definition of periphyton is “growing on or around plants”, the term is used to apply to photosynthetic organisms (mostly algae) growing on surfaces in aquatic systems. The algae in periphyton is an important source of food for organisms of higher trophic levels (e.g., macroinvertebrates, fish).

plankton: Organisms that have little or no ability to control their position within a body of water, that is, they are suspended in the water column. Plankton may be photosynthetic and plant-like (phytoplankton), heterotrophic and animal-like (zooplankton), or composed of bacteria (bacterioplankton).

raphe: A structure in the silica cell wall of some diatoms. Diatoms that possess this slit-like structure are able to move on the surface of substrates. Because these cells can move, they have some ability to select preferred habitats for growth. *Didymosphenia* cells possess a raphe which is functional before cells anchor to a substrate via a stalk.

valve: The siliceous part of the diatom cell wall is composed of two parts, termed valves. Together, the two valves are called a frustule. Diatom valves are often highly ornamented and diatom taxonomy is primarily based on the morphology of these structures.

visual biovolume index: A measure developed in New Zealand to assess the impact of *D. geminata* cells and stalks to a stream ecosystem. The index is a measure of the percent cover of algal mat in a stream transect multiplied by the thickness of the algal mat.

Appendix D: Media coverage

Magazines
Flyfisher, Fall 2006
Dealing with didymo

FlyRod and Reel, April 2006
Short Casts: getting to know didymo

South Dakota Conservation Digest, March/April 2006
Didymo and the Rapid Creek brown trout

Biosecurity, February 2006
Personal responsibility key to stopping didymo spread

High Country Angler, Winter 2006
"Didymo" What is it, and should we be worried?

Print articles
Bozeman Daily Chronicle, May 16, 2006
Algae outbreak threatens rivers around world

Bozeman Daily Chronicle, May 14, 2006
Scientists to gather and discuss slimy algae

Colorado Daily, November 6, 2005
"Rock snot" spreading: pesky algae could threaten waterway ecosystems across country

Denver Post, November 1, 2005
Slime covers streams

Rapid City Journal, April 14, 2005
Algae invader: survey tracks spread of "thug"

Online articles
Montana's News Station, May 16, 2005
Slimy algae draws scientists to Bozeman
www.kbzk.com

Vail Daily, May 18 2006.
Didymo along the Gore Creek in Vail
www.vaildaily.com

Billings Gazette, May 18, 2006
Slimy alga threatens state rivers
www.billingsgazette.net