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Iron is not responsible for *Didymosphenia geminata* bloom formation in phosphorus-poor rivers

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Abstract: Blooms of the river benthic diatom *Didymosphenia geminata* are an enigma because they occur under phosphorus-poor conditions. A recent proposal that ferric–ferrous iron redox shifts sequester the additional phosphorus needed to stimulate and sustain *D. geminata* blooms does not agree with published experimental data showing that blooms only occur when cells are phosphorus-limited. The "iron hypothesis" also infers that blooms would be favoured in rivers with elevated iron, and management should target iron. Surveys of rivers around the world affected by *D. geminata* show that blooms most often occur in iron-poor rivers. Phosphorus uptake experiments conducted under realistic environmental conditions with living *D. geminata* colonies showed no effect of iron enrichment on phosphorus uptake. Iron does not solve the mystery of *D. geminata* growth causing nuisance blooms worldwide.

Résumé : Les proliférations de la diatomée benthique de rivière *Didymosphenia geminata* constituent une énigme puisqu'elles se produisent dans des conditions d'appauvrissement en phosphore. Il a récemment été proposé que des changements d'oxydoréduction du fer ferrique au fer ferreux retiennent le phosphore supplémentaire nécessaire pour stimuler et soutenir les proliférations de *D. geminata*. Ce postulat ne concorde toutefois pas avec les données expérimentales publiées qui démontrent que les proliférations ne se produisent que quand le phosphore est restreint. Il décellsle également de cette « hypothèse du fer » que les rivières présentant de fortes teneurs en fer favoriseraient ces proliférations et que les mesures d'aménagement devraient cibler le fer. Les études de rivières affectées par *D. geminata* aux quatre coins du monde montrent que les proliférations se produisent le plus souvent dans des rivières pauvres en fer. Des expériences de mobilisation du phosphore menées dans des conditions ambiantes réalistes avec des colonies de *D. geminata* vivantes n'ont montré aucun effet d'un enrichissement en fer sur la mobilisation du phosphore. Le fer ne résout pas l'énigme des formes de croissance de *D. geminata* qui entraînent des proliférations néfastes partout sur la planète.

[Traduit par la Rédaction]

Sundareshwar et al. (2011) have proposed that complexes between iron (Fe) and phosphorus (P) that occur on the polysaccharide stalks of the benthic diatom *Didymosphenia geminata* play a central role in the development and maintenance of blooms in oligotrophic streams. In this view, ferric–ferrous redox shifts within mats result in P accumulation and recycling, providing *D. geminata* with an advantage not available to competing species. The importance of ferric–ferrous Fe redox shifts in the cycling of P in lakes has been known since the 1940s (Mortimer 1941) and is one of the most important

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biogeochemical processes influencing productivity of many freshwater lakes and some streams. However, an association between Fe and *D. geminata* dominance of river benthic communities is not supported by water chemistry data from multiple regions experiencing *D. geminata* blooms, and survey data show that elevated P concentrations that can occur in interstitial waters of *D. geminata* mats do not influence the rate of division of *D. geminata* cells on the periphery of colonies. Further, sorption of Fe and other metallic cations onto algal exopolysaccharide is well known and often occurs in high P environments (Lawrence et al. 1998). Here we outline evidence that Fe–P complexes that occur on *D. geminata* stalks are not responsible for the development or the maintenance of blooms in oligotrophic rivers.

Published research from New Zealand

Experiments in New Zealand show that *D. geminata* maintains its dominance of river benthic algal communities while remaining in a highly P-limited state (Bothwell and Kilroy 2011; Kilroy and Bothwell 2012). P-limited cell division rates of *D. geminata* were not influenced by attachment to thick mats of polysaccharide stalk material (Bothwell and Kilroy 2011). Experiments document that the elongation of *D. geminata* stalks, associated with blooms, preferentially occurs under P-limited conditions (Kilroy and Bothwell 2011), and blooms did not form when supplemental P was available (Kilroy and Bothwell 2012). Surveys around the world find that *D. geminata* blooms are usually associated with very low P conditions (Bothwell et al. 2009). While this could be interpreted as an indication that *D. geminata* has access to P sources not available to other taxa, the application of the frequency of dividing cells technique to *D. geminata* in situ across a spectrum of dissolved reactive phosphorus (DRP) concentrations in rivers on South Island, New Zealand, indicated that this was not the case (Kilroy and Bothwell 2012). Kilroy and Bothwell (2012) found that cell division rates of *D. geminata* correlated positively with DRP in the overlying water and that bloom formation was inversely related to DRP. *Didymosphenia geminata* blooms end when cell division rates are not limited by P, and blooms do not begin unless these rates are P-limited.

Fe concentrations in rivers with and without *D. geminata* blooms

In natural oxygenated waters, Fe exists primarily as a finely divided precipitate of hydrated ferric hydroxide, with particle sizes ranging from filterable (>0.45 μm) to colloidal (0.001–0.5 μm) (Wetzel 2001). For this reason the fraction of total iron (Fe-T) operationally defined by filtration as dissolved (Fe-D) varies widely depending on the size distribution of ferric hydroxide particles and the porosity characteristics of filters. In the case of rivers affected by *D. geminata* on Vancouver Island, British Columbia, the percentage of Fe-T classified as Fe-D spans the range from 3.5% to 100%, with a median of 44% (n = 216; Supplemental Table S1[1]). Because of this wide variability, caution is needed when comparing Fe data. Unfortunately, in supporting their contention that higher Fe levels in rivers might foster *D. geminata* blooms, Sundareshwar et al. (2011) compared Fe-T in affected rivers, with Fe-D in non-*D. geminata* rivers (see supplemental table S1 in Sundareshwar et al. 2011). Our comprehensive surveys show no consistent pattern between Fe-T or Fe-D in river water and the blooming behaviour of this nuisance alga. Moreover, it is unclear what, if any, temporal changes in Fe or other ions (e.g., sulphate) have occurred at the disparate sites where *D. geminata* is blooming.

Many streams in the Black Hills, South Dakota, have naturally high levels of Fe, and in those with highest Fe, ferric hydroxide precipitate significantly reduces attached algal biomass (Holcomb 2002). Fe-T levels in Rapid Creek (mean 0.31 $mg \cdot L^{-1}$, n = 3 (Sundareshwar et al. 2011) versus mean 0.14 $mg \cdot L^{-1}$, median 0.079 $mg \cdot L^{-1}$, n = 64 (our 2008–2009 surveys); Table 1*a*, Supplemental Table S2[1]), the stream affected by *D. geminata* that was the focus of the Sundareshwar et al. (2011) study, are at the lower end of the range for both mean and median Fe-T values for Black Hills streams (Supplemental Table S2[1]). Rapid Creek remains the only stream in the region affected by extensive *D. geminata* blooms.

The absence of association between *D. geminata* colonies and Fe-T was also evident within Rapid Creek. Although *D. geminata* was widespread within the stream, colonies were not visible at all sites or on all occasions. The average Fe-T concentration at sites and on occasions with colonies was not different from that when colonies were absent (Supplemental Table S2[1]). Likewise, *D. geminata* standing crop in Rapid Creek, quantified using a biovolume index, was unrelated to Fe-T concentration of the water (Supplemental Fig. S1[1]). Collectively, these results indicate no positive relationship between the concentration of Fe-T in river water and *D. geminata* colonies within or among streams in the Black Hills of South Dakota.

Didymosphenia geminata blooms commencing in 2006 in Quebec spread to numerous rivers along the Gaspe Peninsula. Those rivers eventually supporting blooms had significantly lower levels of Fe-D than those rivers remaining *D. geminata* bloom free (Table 1*b*; Supplemental Table S3[1]). In Quebec rivers, alkalinity and pH are related to the distribution of *D. geminata* blooms (Gillis et al. 2010).

Seven rivers on Vancouver Island supporting large *D. geminata* blooms had median Fe-D and Fe-T concentrations of 0.037 and 0.08 $mg \cdot L^{-1}$, respectively (Table 1*c*). Mean Fe concentrations for these rivers ranged between 0.028 and 0.058 $mg \cdot L^{-1}$ for Fe-D and between 0.08 and 0.17 $mg \cdot L^{-1}$ for Fe-T (Supplemental Table S1[1]). Fe-T data were available in two rivers both prior to and during *D. geminata* bloom years. Lower Fe-T during blooms in the Puntledge River might indicate adsorption onto stalk material. However, such a decline was not observed in the Oyster River (Table 1*c*).

Two remote, adjacent Canadian Rocky Mountain streams were monitored for 8 years as part of a long-term watershed study. *Didymosphenia geminata* was not detected in either stream until 2010, when blooms commenced in one (North York). Both streams had the same level of Fe-T for the entire period of study (Table 1*d*). Concentrations of Fe-T (median 0.01 $mg \cdot L^{-1}$) in these high elevation streams were among the lowest in our surveys and did not differ significantly between the streams affected and unaffected by *D. geminata* (Table 1*d*).

On South Island, New Zealand, Fe-D data were available from several rivers affected by *D. geminata* and their spring-

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[1] Supplementary data are available with the article through the journal Web site at http://nrcresearchpress.com/doi/suppl/10.1139/f2012-112.

Table 1. Summary of iron (Fe) and dissolved organic carbon (DOC) concentrations measured in rivers in Canada, USA and New Zealand, in relation to the occurrence of *D. geminata* blooms.

	No. of rivers or sites	Total Fe (mg·L^{-1})				Dissolved Fe (mg·L^{-1})				DOC (mg·L^{-1})			
		Sample *n*	Med.	Max.	Statistical test	Sample *n*	Med.	Max.	Statistical test	Sample *n*	Med.	Max.	Statistical test
(*a*) Black Hills, South Dakota													
D. geminata affected	1	64	0.079	1.57	Mann–Whitney *U* test; $p = 0.028$		NA				NA		
Not affected	6	57	0.15	8.01									
(*b*) Quebec													
D. geminata affected	12		NA	NA		576	0.035	1.60	Two-tailed *t* test; $t = 6.51$, $p < 0.001$	511	2.4	50.0	Two-tailed *t* test; $t = 6.17$, $p < 0.001$
Not affected	17		NA	NA		510	0.150	4.13		501	4.8	94.0	
(*c*) Vancouver Island													
D. geminata affected	7	567	0.08	2.39		271	0.037	0.370		NA	NA	NA	
Before *D. geminata*	Oyster	104	0.1	1.96	Mann–Whitney *U* test; $p = 0.460$					NA	NA	NA	
After *D. geminata*		24	0.1	0.43						NA	NA	NA	
Before *D. geminata*	Puntledge	19	0.09	0.96	Mann–Whitney *U* test; $p < 0.005$					NA	NA	NA	
After *D. geminata*		32	0.041	0.335						NA	NA	NA	
(*d*) Canadian Rockies													
D. geminata affected	North York	118	0.01	0.98	Nonparametric sign test; $p < 0.461$	NA	NA	NA		137	1.05	6.1	Nonparametric sign test; $p < 0.001$
Not affected	Star	118	0.01	0.36		NA	NA	NA		158	1.3	7.9	
(*e*) South Island, New Zealand													
D. geminata affected	10	NA	NA	NA		21	0.016	0.071	Mann–Whitney *U* test; $p = 0.463$	Note: g_{440} values available; see Supplemental Fig. S2			
Not affected	25	NA	NA	NA		34	0.011	0.67					
(*f*) Colorado Rocky Mountains													
D. geminata affected	6	24	0.079	0.315	Two-tailed *t* test; $t = 3.35$, $p < 0.009$	24	0.036	0.196	Two-tailed *t* test; $t = 3.12$, $p < 0.012$	24	6.2	10.9	Two-tailed *t* test; $t = 2.13$, $p < 0.07$
Not affected	5	20	0.017	0.079		20	0.004	0.028		20	2.6	5.9	

Note: Comparisons are between medians or single values from multiple rivers or sites. NA refers to no data available or not applicable. Statistical tests used for the comparisons were selected according to the data distribution and type (paired or independent). Complete data sets are in Supplemental Tables S1–S5[1].

fed tributaries in 2007 and in 2011 from 19 South Island river sites selected randomly from the New Zealand National Water Quality Network (Supplemental Table S4[1]). Fe-D spanned a wide range, and there was no significant difference between Fe-D concentrations in rivers affected or unaffected by *D. geminata* (Table 1*e*). None of the sites with blooms had Fe-D > 0.071 mg·L^{-1}.

The only data set in which *D. geminata* bloom formation was associated with higher levels of Fe was from a suite of 11 Rocky Mountain streams near Gunnison, Colorado. Six streams with *D. geminata* blooms in 2010 had significantly higher levels of both Fe-D and Fe-T compared with five other nearby streams without *D. geminata* blooms during the same period (Table 1*f*; Supplemental Table S5[1]). Experiments were undertaken to determine if higher Fe concentrations might stimulate P uptake by *D. geminata* colonies.

Fe enrichment experiments in the Colorado Rocky Mountains

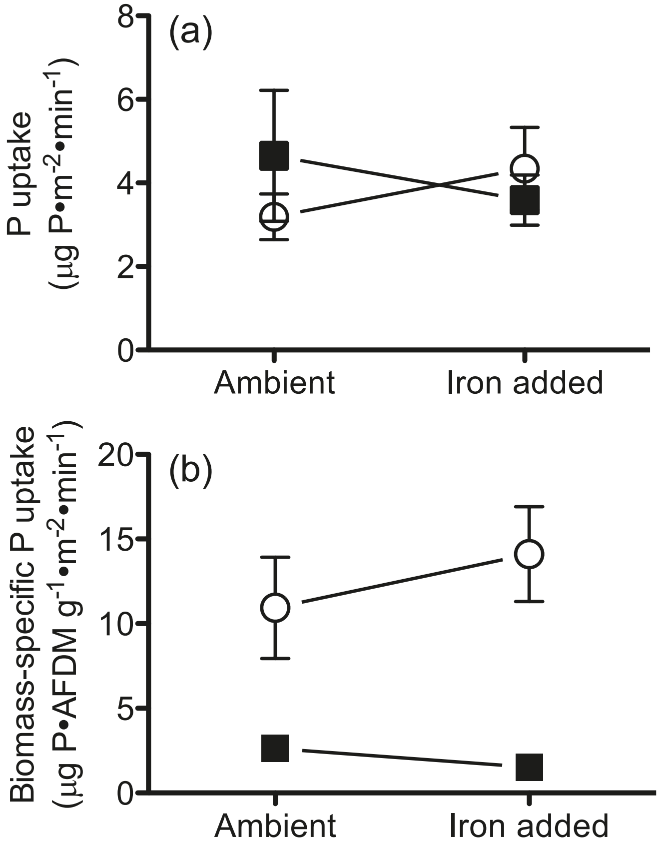
Sundareshwar et al. (2011) used poisoned, nonliving segments of *D. geminata* colonies to demonstrate that additions of Fe increase sequestration of P onto *D. geminata* stalks. They also used concentrations of Fe and P in their trials that were three to six orders of magnitude greater than dissolved levels in typical rivers dominated by *D. geminata*. Our P-uptake experiments with Fe enrichment were conducted with live material on substrates from Rocky Mountain streams incubated in containers for 2 h under in situ stream conditions with 20 µg P·L^{-1}, sodium phosphate added, and with or without Fe additions of 0.3 mg Fe·L^{-1}.

Our realistic level of Fe augmentation failed to increase the uptake of P, and rates of P uptake were similar between substrates with versus without visible colonies of *D. geminata* (Fig. 1*a*). Biomass-specific P uptake was five times higher on substrates without *D. geminata* colonies than on substrates with colonies and was also not affected by the addition of Fe (Fig. 1*b*). There was also no interaction between Fe and *D. geminata* colonies on P uptake (Fig. 1*b*). Hence, using live material under environmentally realistic conditions, the uptake of P by *D. geminata* was not stimulated by additional Fe. More importantly, P removal from the water column (adsorption or uptake) by substrates with *D. geminata* colonies was significantly lower on a per ash-free dry mass basis than P removal by substrates without *D. geminata* colonies, indicating the importance of cellular uptake rather than noncellular adsorption onto *D. geminata* polysaccharide stalks as has been proposed. In addition, these experimental results also suggest that the low P concentration typical of streams in which *D. geminata* blooms (Kilroy and Bothwell 2012) is not a result of greater uptake.

Reactivity of Fe

Fe–organic complexes are important in increasing Fe levels in surface waters but are problematic in predicting Fe reactivity because dissolved organic carbon (DOC) in natural waters is an unknown mixture of compounds with binding constants for Fe that are either unknown or unpredictable among natural waters (Schecher and McAvoy 2003). The best that can be done to evaluate the potential for DOC to hinder Fe–P complexes is to compare DOC concentrations in rivers.

Fig. 1. Effects of *D. geminata* and iron on water column phosphorus. (*a*) Removal rates (±1 standard error of the mean, SE) of phosphorus (20 µg P·L^{-1}, as sodium phosphate) by substrates covered with visible colonies of *D. geminata* (solid squares) versus substrates lacking visible colonies of *D. geminata* (open circles) at ambient iron concentrations and with iron added (0.3 mg Fe·L^{-1}, as ferrous sulfate) did not differ (two-way analysis of variance (ANOVA), $F_{[3,31]} = 1.65$, $p = 0.19$). (*b*) Phosphorus removal rates (±1 SE) normalized to ash-free dry mass (AFDM) on the substrates were significantly higher in the absence of *D. geminata* colonies (two-way ANOVA, $F_{[1,31]} = 38.63$, $p < 0.0001$) but also not affected by the addition of iron (two-way ANOVA, $F_{[1,31]} = 0.031$, $p < 0.86$). Uncovered containers with substrates from Washington Gulch and Coal Creek were incubated at ambient stream temperature and light.



Across a broad range, rivers dominated by *D. geminata* tend toward lower DOC (Bothwell et al. 2009). It is unclear if the negative relationship between *D. geminata* blooms and DOC is due to reduced light penetration in highly coloured rivers or the association with lower pH in more humate-stained waters. In the view of Sundareshwar et al. (2011), low DOC might also favour *D. geminata* blooms if DOC chelation of Fe reduces reactivity and minimizes sequestration of P. However, we found little evidence to support the latter argument.

In the Colorado Rocky Mountain streams where *D. geminata* blooms were associated with higher concentrations of Fe-T and Fe-D, DOC was also twice as high, potentially off-setting the higher Fe (Table 1*f*). Median DOC concentrations in two high elevation streams in the Canadian Rocky

Mountains were the lowest of all streams surveyed (1.1 and 1.3 mg·L^{-1}; Table 1*d*), and although the mean DOC was higher in the stream free of *D. geminata*- compared with the stream affected by *D. geminata*, the differences were small ($<$0.5 mg·L^{-1}; Table 1*d*). In New Zealand, there was no relationship between the coloured DOC content of river water and the presence or absence of *D. geminata* blooms (Supplemental Fig. S2[1]). Only the Quebec data set showed lower concentrations of DOC in rivers affected by *D. geminata*, but they were also associated with Fe-D concentrations four times lower than in rivers lacking blooms (Table 1*b*).

Conclusion

Values of Fe-T in natural surface waters typically range between 0.05 and 0.20 mg·L^{-1} (Wetzel 2001). We have found that *D. geminata* often blooms in rivers at the lower end of this spectrum, which contradicts the inference of Sundareshwar et al. (2011) that higher Fe concentrations favour *D. geminata* blooms. More importantly, quantitative measurements indicate that *D. geminata* blooms do not rely on P sourced from within the mat, and uptake rates of P by living *D. geminata* colonies are not increased by realistic levels of Fe enrichment. Rather, *D. geminata* blooms occur and are sustained because of an inadequate supply of P. This conclusion is consistent with observations that *D. geminata* blooms disappear in river reaches immediately downstream of point-source nutrient outfalls in Canadian and US Rocky Mountain rivers (M.L. Bothwell and B.W. Taylor, personal observation) and observations in South Dakota that nutrients added to a stream curtailed *D. geminata* bloom coverage up to 0.6 km downstream (James and Chipps 2012).

The global emergence and spread of *D. geminata* is widely attributed to new introductions. While the root causes of blooms in Northern Hemisphere environments where *D. geminata* is thought to be endemic are unknown, the association between *D. geminata* bloom formation and very low P suggests increasing intensity and frequency of P limitation in pristine waters, perhaps associated with global nitrogen deposition or other regional mechanisms of P depletion. While this remains speculation, the available data do not support the idea that Fe is responsible for promoting and sustaining *D. geminata* blooms in P-poor rivers.

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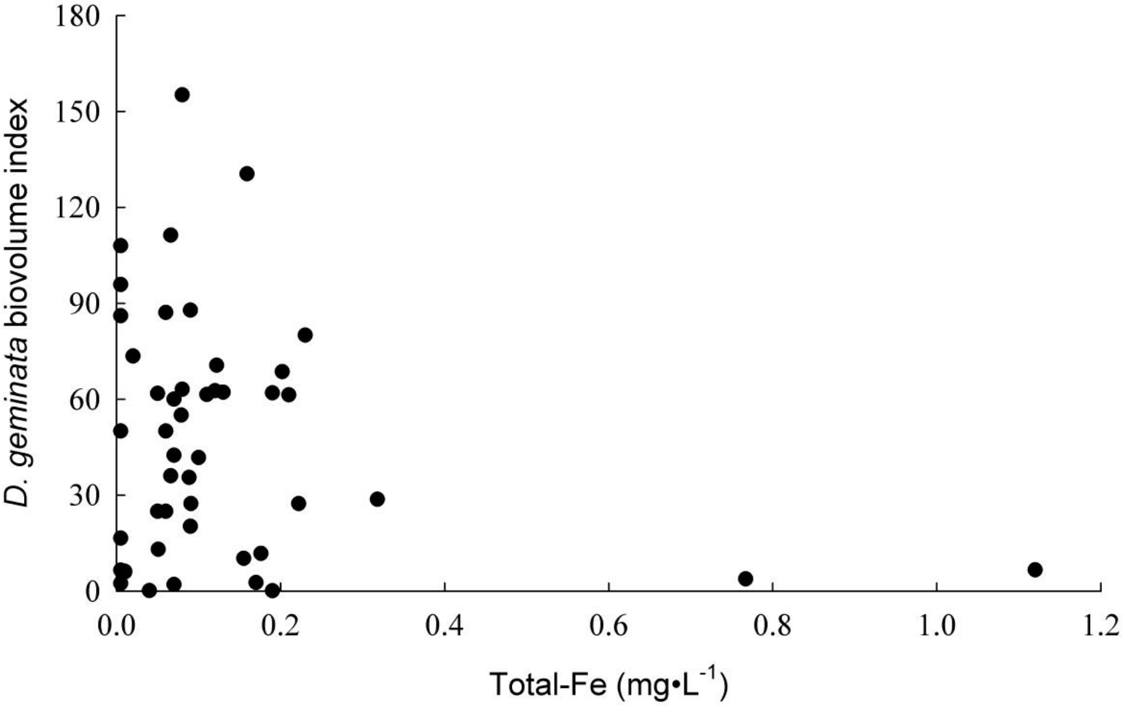
Supplemental materials for Bothwell et al. CJFAS

Supplemental Figure Captions

Figure S1. *D. geminata* biovolume index versus the total iron concentration at sites within Rapid Creek during 2008-2009. Biovolume index was computed using visually estimated percent coverage and thickness of the *D. geminata* mat (mm) from one hundred randomly selected rocks from each sampling riffle. Thickness was assigned a score from 0 to 5 based on the following: 0; 1 (< 1 mm thick); 2, (1-5 mm); 3, (6-15 mm); 4 (16-30 mm); 5, (> 30 mm). The coverage of *D. geminata* was multiplied by the thickness to yield an index ranging from 0 to 500. The biovolume index was unrelated to Fe-T concentration of the water (linear regression: $F_{1, 46}=2.09$; $p=0.155$; $r^2=0$

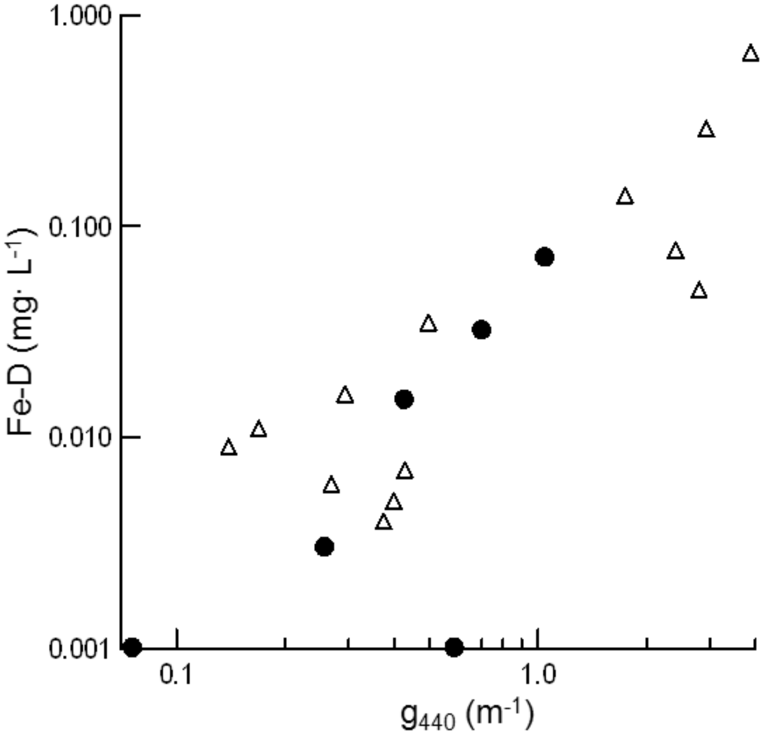
Figure S2. Mean optical density of water at 440 nm (g_{440}), a measure of colored dissolved organic matter (CDOM), plotted against dissolved iron (Fe-D) concentration measured at 19 South Island, New Zealand, river sites. Filled circles are sites with *D. geminata* blooms present, and open triangles with blooms absent. The positive relationship between CDOM and concentrations of Fe-D was significant ($r=0.822$, $p<0.001$, $n=19$) but there was no difference in CDOM between streams with and without *D. geminata* blooms.

Figure S1



180
150
120
90
60
30
0
D. geminata biovolume index
0.0
0.2
0.4
0.6
0.8
1.0
1.2
Total-Fe (mg•L^{-1})

Figure S2



Supplemental Tables

Table S1. Summary of data on dissolved and total iron concentrations in Vancouver Island rivers. All seven rivers were affected by *D. geminata* blooms from the early 1990s. Fractions of dissolved Fe were calculated from the ratios (as %) of dissolved to total iron for pairs of samples collected at the same time.

		Dissolved Fe (mg•L^{-1})				Total Fe (mg•L^{-1})				Fraction dissolved Fe (%)			
River	Yrs	Mean	s.e.m.	*n*	Med.	Mean	s.e.m.	*n*	Med.	Min.	Max.	*n*	Med.
Cowichan	1985-1993	0.046	0.005	39	0.040	0.092	0.013	53	0.06	44	100	5	50
Englishman	1996-2010	0.558	0.522	21	0.034	0.136	0.020	181	0.086	6.0	81	14	35
Nanaimo	1989-2010	0.058	0.006	74	0.044	0.115	0.012	75	0.082	15	100	73	56
Oyster	1980-1993	0.056	0.008	69	0.040	0.172	0.022	128	0.1	7.7	100	69	33
Puntledge	1980-2008					0.084	0.020	51	0.047				
Somass	1993-2004	0.031	0.004	35	0.022	0.080	0.029	47	0.031	3.5	92	27	45
Stamp	1993-1995	0.028	0.005	37	0.020	0.113	0.037	32	0.025	5.0	92	30	27

Data from the British Columbia Environmental Monitoring System (EMS).
http://www.env.gov.bc.ca/epd/wamr/ems_internet/index.html

Filter pore size 0.45µm used to separate dissolved and total Fe.

Table S2. Total iron and *D. geminata* bloom status in streams of the Black Hills, South Dakota.

River	*D. geminata* blooms	Year	Total Fe (mg•L^{-1})			
			Mean	s.e.m.	n	Median
Castle Creek*	no	2001	0.18	0.02	3	0.19
Castle Creek	no	2008	0.57	0.23	10	0.17
North Fork Castle*	no	2001	4.95	1.55	3	3.84
South Fork Rapid Creek*	no	2001	0.67	0.19	3	0.85
North Fork Rapid Creek*	no	2001	0.62	0.15	3	0.51
Spearfish Creek	no	2008-2009	0.058	0.018	25	0.04
Whitewood Creek	no	2008	0.44	0.098	10	0.42
Rapid Creek**	yes	2008-2009	0.13	0.028	47	0.08
Rapid Creek§**	no	2008	0.17	0.089	17	0.07

Total iron analyses conducted by Olson Analytical Services Laboratory at South Dakota State University
*Data from Holcomb 2002
§ sampling sites and dates when no *D. geminata* coverage was seen
**Comparison of Fe-T during bloom presence vs. absence in Rapid Creek; Mann Whitney U-test, p =0.670

Table S3. Summary of dissolved iron and dissolved organic carbon (DOC) data from Quebec Rivers.

		Fe-D (mg•L^{-1})					DOC (mg•L^{-1})				
River	*D. gem.* blooms	Years	mean	s.e.m.	*n*	med.	Years	mean	s.e.m.	*n*	med.
Bonaventure	1	1984-1986	0.05	0.12	28	0.01	1984-1986 2004-2009	1.1	0.6	122	1.0
Cascapédia	1	1984-1986	0.05	0.07	35	0.03	1984-1986	2.1	1.3	61	1.8
Nouvelle	1	1984-1986	0.03	0.05	33	0.03	1984-1986	0.9	0.7	79	0.7
Matapédia	1	1984-1992	0.07	0.08	98	0.04	1984-1990 2004-2009	3.2	0.8	198	3.2
Causapscal	1	1984-1988 1990-1992	0.08	0.07	24	0.06	1984-1988 2004-2009	4.2	3.2	42	3.1
Madawaska	1	1984-1990	0.06	0.04	69	0.05	1984-1988 1993-2009	4.4	0.5	267	4.4
Saint-Jean Gaspé	1	1984-1985	0.03	0.05	33	0.01	1984-1986	0.9	0.6	56	0.8
York	1	1984-1985	0.09	0.15	24	0.03	1984-1986 1995-1997	1.9	1.1	60	1.6
Madeleine	1	1984-1986	0.04	0.04	14	0.02	1984-1986	1.6	1.0	19	1.2
Sainte-Anne	1	1984-1986	0.05	0.06	30	0.04	1984-1986	1.5	0.8	50	1.3
Matane	1	1984-1986	0.11	0.18	33	0.04	1984-1986 1995-1997	3.4	1.6	75	3.0
Mitis	1	1984-1986	0.21	0.33	32	0.10	1984-1986 1995-1997	4.3	5.3	80	3.6
Rimouski	0	1984-1986	0.14	0.14	32	0.10	1984-1986	4.9	1.2	56	4.8
Trois-Pistoles	0	1984-1986	0.12	0.09	31	0.10	1984-1986	5.5	2.1	53	5.4
du Loup	0	1984-1986	0.40	0.12	36	0.39	1984-1986 1994-2010	14.1	8	241	11.8
Ouelle	0	1984-1986	0.38	0.56	32	0.25	1984-1986 2005-2010	11.1	4.4	106	10.5
Des escoumins	0	1984-1986	0.47	0.49	9	0.26	1984-1986 2004-2009	4.9	1.6	57	4.7
Du sault	0	1984-1986	0.49	0.11	9	0.51	1984-1986	5.3	1.6	9	5.0
Bestiamites	0	1984-1986	0.38	0.58	31	0.17	1984-1986	4.9	1	52	4.6
Outardes	0	1984-1990	0.12	0.03	93	0.12	1984-1990 1993-1996	4.7	0.6	147	4.6
Manicouagan	0	1984-1990	0.10	0.03	91	0.09	1984-1990 1993-1996	4.2	0.6	141	4.2
Godbout	0	1984-1986	0.24	0.14	9	0.20	1984-1986	5.8	1.3	9	5.4
Pentecôte	0	1984-1986	1.09	1.17	9	0.70	1984-1986	6.7	0.7	9	6.8
aux Rochers	0	1984-1986	0.20	0.05	27	0.20	1984-1986	6.4	1.3	52	6.0
Sainte-Marguerite	0	1984-1986	0.29	0.1	9	0.28	1984-1986	6.5	1.5	9	6.8
Moisie	0	1984-1990	0.28	0.47	74	0.19	1984-1990 1993-1996	4.8	1.5	130	4.6
Manitou	0	1984-1986	0.11	0.09	8	0.07	1984-1986	5	0.7	8	4.8
Magpie	0	1984-1986	0.04	0.02	7	0.04	1984-1986	3.6	0.3	7	3.4
Saint-Jean	0	1984-1985	0.73	0.17	2	0.73	1984-1986	5.4	0.3	2	5.4

Footnote for Supplemental Table S3

Data on dissolved Fe (Fe-D) and DOC for Quebec rivers from the MDDEP, 2012. Banque de données sur la qualité du milieu aquatique (BQMA), Québec, ministère du Développement durable, de l’Environnement et des Parcs, Direction du suivi de l’état de l’environnement. Filter pore size 0.45µm used for Fe-D and DOC. Fe-D data are primarily from the pre-*D. geminata* bloom period (1984 and 1986). DOC was monitored consistently from 1984 to the present at most stations. Pre- and post-incursion DOC data are combined for the Matapédia, Bonaventure, Causapscal, Madawaska, du Loup, des Escoumins and Ouelle rivers.

Table S4. Summary of dissolved iron, dissolved organic carbon (DOC) and water colour data from South Island, New Zealand rivers.

		D. gem.	Dissolved Fe (mg•L^{-1})				DOC				**g_{440}
River	*Year	blooms	mean	s.e.m.	*n*	med.	mean	s.e.m.	*n*	med.	med.
Mararoa	2007	yes	0.015	0.007	5	0.010	0.83	0.37	5	0.80	
Oreti	2007	yes	0.013	0.008	3	0.010	1.07	0.62	3	1.00	
Waitaki (d/s)	2007	yes	0.055	0.023	6	0.038	0.40	0.18	5	0.25	
Buller (u/s)	2011	yes	0.001		1						0.36
Clutha	2011	yes	0.001		1						0.06
Hurunui	2011	yes	0.003		1						0.26
Opuha	2011	yes	0.032		1						0.69
Shotover	2011	yes	0.016		1						0.27
Waiau	2011	yes	0.071		1						0.75
Waitaki (u/s)	2011	yes	0.015		1						0.35
Ashton Flats Spring	2007	no	0.010		1						
Ferry Rd Spring	2007	no	0.037	0.021	3	0.040	0.47	0.27	3	0.25	
Flaxy Creek	2007	no	0.020	0.014	2	0.020	0.53	0.37	2	0.53	
Hen & Chicken Spr.	2007	no	0.200		1						
Oreti Roadend Spr.	2007	no	0.010	0.006	3	0.010	0.25	0.18	2	0.25	
Otiake Spring	2007	no	0.046	0.026	3	0.047	0.25	0.14	3	0.25	
Wakakahi Spring	2007	no	0.200		1						
Wash Creek	2007	no	0.010	0.006	3	0.010	0.47	0.27	3	0.25	
Wilson Road Spring	2007	no	0.030		1						
Brightwater Creek	2011	no	0.001		1						
Buller (d/s)	2011	no	0.077		1						2.45
Flaxy Creek	2011	no	0.001		1						
Grey	2011	no	0.050		1						2.60
Mataura (d/s)	2011	no	0.140		1						1.47
Mataura (u/s)	2011	no	0.035		1						0.36
Monowai	2011	no	0.007		1						0.44
Motueka (Gorge)	2011	no	0.006		1						0.28
Opihi (d/s)	2011	no	0.004		1						0.38
Opihi (u/s)	2011	no	0.005		1						0.40
Sutton	2011	no	0.290		1						2.81
Taieri	2011	no	0.670		1						3.78
Upper Oreti	2011	no	0.001		1						
Waimakariri	2011	no	0.009		1						0.16
Wairau	2011	no	0.011		1						0.12
Wash Creek	2011	no	0.001		1						

d/s; u/s: downstream and upstream sites, where sampling locations on the same river were far apart.
*Analyses in 2007 and 2011 had detection limits (dissolved iron) of 0.02 and 0.002 mg•L^{-1}, respectively
**median values for g_{440} (gilvin, calculated from absorbance at 440 nm) from a long-term record.

Table S5. Total and dissolved iron concentrations and dissolved organic carbon (DOC) concentrations in Colorado Rocky Mountain streams.

River	years	*D. geminata* blooms*	Fe-T (mg•L^{-1})	Fe-D (mg•L^{-1})	DOC (mg•L^{-1})
Copper[§]	2010-2011	Absent	0.0113	0.0025	1.3
East Fork Crystal[§]	2010-2011	Absent	0.0098	0.0013	5.9
Poverty Gulch[§]	2010-2011	Absent	0.0299	0.0123	1.2
Rustler's Gulch[§]	2010-2011	Absent	0.0168	0.0038	3.1
Upper East[§]	2010-2011	Absent	0.0787	0.0284	2.6
median			*0.0168*[**]	*0.0038*[**]	*2.6*[***]
Cement	2010-2011	Present	0.0569	0.011	10.9
Coal	2010-2011	Present	0.2195	0.1054	3.6
Lower East	2010-2011	Present	0.0895	0.0332	7.7
Oh-Be-Joyful	2010-2011	Present	0.0569	0.0391	2
Washington Gulch	2010-2011	Present	0.3154	0.196	6.2
West Brush	2010-2011	Present	0.069	0.0273	6.2
median			*0.0793*[**]	*0.0362*[**]	*6.2*[***]

* Present indicates visible colonies of *D. geminata* covering > 1 m^2 of the stream bottom.

[§] *D. geminata* cells were detected in these streams but not visible colonies of *D. geminata* covering > 1 m^2 of the stream bottom.

All Fe samples and blanks were acidified with 400 µL of Optima-grade nitric acid. Concentrations of field blanks were 0.0014 mg•L^{-1} total Fe and 0.0006 mg•L^{-1} dissolved Fe.

Gelman A/E glass fiber filters, nominal pore size 1µm, were used for Fe-D and DOC samples.

[**] Dissolved and total iron concentrations were higher in streams with visible colonies of *D. geminata* (dissolved iron two-tailed t test, $t_9=3.12$, $p<0.012$; total iron two-tailed t test, $t_9=3.35$, $p<0.009$).

[***] DOC concentrations were not significantly different between streams with versus without visible colonies of *D. geminata* (two-tailed t test, $t_9=2.13$, $p<0.07$), and the trend for higher DOC in streams with visible colonies was driven by one stream (Cement Creek).