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PRESENCE OF ALGAE IN FRESHWATER ICE COVER OF FLUVIAL LAC SAINT-PIERRE (ST. LAWRENCE RIVER, CANADA)[1]

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Winter ice cover is a fundamental feature of north temperate aquatic systems and is associated with the least productive months of the year. Here we describe a previously unknown freshwater habitat for algal and microbial communities in the ice cover of the freshwater St. Lawrence River, Quebec, Canada. Sampling performed during winter 2005 revealed the presence of viable algal cells, such as *Aulacoseira islandica* (O. Müll.) Simonsen (Bacillariophyceae), and microbial assemblage growing in the ice and at the ice–water interface. Vertical channels (1–5 mm wide) containing algae were also observed. Concentrations of chl *a* ranged between 0.5 and 169 $\mu g \cdot L^{-1}$ of melted ice, with maximal concentrations found in the lower part of the ice cores. These algae have the potential to survive when ice breakup occurs and reproduce rapidly in spring/summer conditions. Freshwater ice algae can thus contribute to in situ primary production, biodiversity, and annual carbon budget in various habitats of riverine communities.

***Key index words*: Bacillariophyceae; Chlorophyta; fluvial lake; freshwater; ice algae; protist; spatial distribution; sympagic communities; temperate ecosystem; vertical channels**

***Abbreviations*: *a*CDOM, absorbance by colored dissolved organic matter at 375 nm; TP, total phosphorus**

Sympagic organisms (in association with ice) have been widely reported in sea ice from polar regions, but little information is available for freshwater ecosystems and is limited to boreal and arctic regions. Sea ice is known to be a productive habitat for microbial food webs. These communities are composed of bacteria, microalgae, and protozoa that transfer carbon and cycle nutrients (Cota et al. 1991, Horner et al. 1992), contributing substantially to primary production (Gosselin et al. 1997). They provide food for consumers within and at the ice–water interface, both during the ice period (Nozais et al. 2001, Michel et al. 2002, Kaartokallio 2004) and throughout the water column during ice breakup (Lizotte 2001).

In Canada alone, freshwater environments total ~900,000 km^2, and a large part of this area is characterized by prolonged ice cover during winter. In polar regions, freshwater ice typically forms as dense, solid congelation ice. It lacks the anastomosing brine channels found in sea ice that allow colonization by microorganisms. This difference in structure explains why freshwater ice has been thought to lack sympagic communities (Vincent 1988). Here we describe a previously unknown freshwater habitat for algal and microbial communities in the ice cover of the St. Lawrence River, Canada. Autotrophs derived from benthic and terrestrial habitats were found in permanently ice-covered lakes in Antarctica, and incorporation of pelagic microbiota into the ice was reported on subglacial Lake Vostok during accretion (Priscu et al. 1998). The alga *Aulacoseira baicalensis* growing in the interstitial water within the ice has recently been reported in Lake Baikal, Russia (Bondarenko et al. 2006), whereas microbial communities occurred in slush layers within the ice cover of a high mountain lake from Spain (Felip et al. 1999). However, this is the first evidence that diverse and abundant microbial communities reside in the ice cover and at the ice–water interface of river ice, producing a freshwater analogue to the well-known sympagic communities in sea ice.

MATERIALS AND METHODS

Sampling. Lac Saint-Pierre (46°12′ N; 72°50′ E) is the largest fluvial lake (~400 km^2 at mean discharge) of the St. Lawrence River (Fig. 1). It is composed of up to eight water masses characterized by distinct physical and chemical properties in response to the strong connectivity with inflowing tributaries and wetlands (Frenette et al. 2006). It is typically shallow (3 m on average, at mean discharge) except in the maritime channel artificially maintained at ~11 m for transport ships. Ice usually starts to form in early December and breaks up around late March/early April; the maritime channel remains unfrozen all year long. Sampling was conducted at two different spatial scales: one to characterize the horizontal and coarse vertical variation in protist biomass and diversity, and the other to

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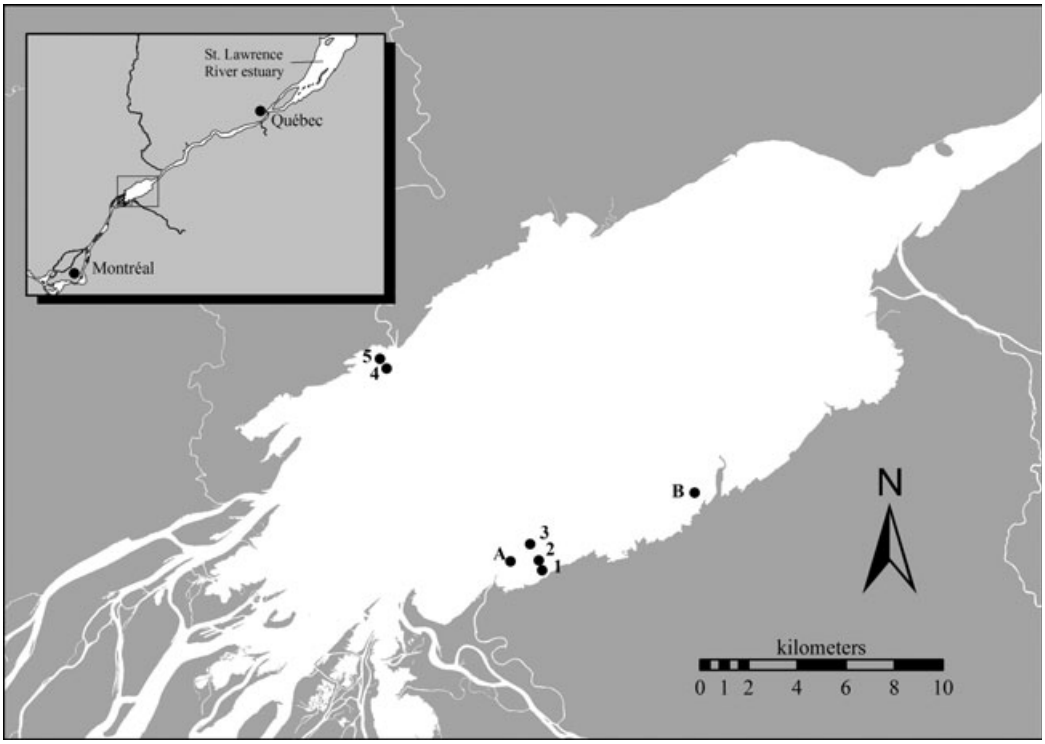

FIG. 1. Position of the sampling stations in the north and south shores of Lac Saint-Pierre, St. Lawrence River, Canada.

examine the fine-scale vertical variability. Five stations (1–5) were sampled on February 27, 2005 (Fig. 1). Three stations were located within the south shore zone, and two in the north shore zone to obtain samples from environments with contrasting physical and chemical properties. Sampling was limited to the nearshore area (<2 km) due to the thinner ice cover approaching the unfrozen maritime channel. Three ice cores (9 cm diameter) were collected at each station using a Mark II ice corer (Kovacs Enterprise, Lebanon, NH, USA). Water was sampled from the hole formed by the removal of the core using a 2 m PVC tube to obtain an integrated sample of the first 1–1.5 m of the water column. Ice cores were separated into two equal sections (upper and lower) with a stainless steel saw. Water-column and ice samples were transferred to separate 2 L acid-washed polyethylene containers and transported to the lab within 4 h. At each station, we measured snow cover, ice thickness, and irradiance above and under the ice cover using a LI-COR quantum meter (Model LI-1000-32, Lincoln, NE, USA). The sampling event on February 27 provided the first evidence of algae in the ice cover. Additional sampling was performed afterward to obtain a more detailed description of the vertical distribution of algae in the ice. On March 22, 2005, five cores were collected at station ''A.'' These cores were subsampled into five 5 cm sections and one 1 cm section called the interface, which represents the last layer of the ice cover. These sections correspond to the major community types found in sea ice (Cota et al. 1991, Horner et al. 1992): surface (one section), interior (one section), bottom (three sections), and subice (interface, one section). The ice sections were melted at 4°C, in the dark, in the cold room of the laboratory.

Community structure and biomass. Melted ice sections and water samples were preserved in 1% gluteraldehyde–paraformaldehyde for taxonomic identification and enumeration. Duplicate samples for chl *a* determination were filtered on 0.7 μm porosity Millipore (Billerica, MA, USA) glass fiber filters, and concentrations were determined by absorbance (Shimadzu spectrophotometer model UV-2401, Tokyo, Japan) after extraction in hot ethanol 95% (Marker et al. 1980).

Enumerations followed the counting protocol of Lund et al. (1958), Hasle (1978), Venrick (1978), and Hamilton et al. (2002) using ~260 μm deep settling chambers on an upright compound microscope (Leica DMR microscope, Leica Microsystems, Wetzlar, Germany). Taxa were counted from random transects scanned across the settling chamber at ×200 and ×400 magnifications. Two to seven transects were counted at each magnification, with between 300 and 1,200 cells counted using a count time per sample of 3–4 h. In the upper ice section (5–10 cm) of the core, algae were sparse, and, even with concentrated material, counts at this level were between 100 and 200 cells. Taxa were identified to the lowest possible taxonomic level. Counting was facilitated by a computerized enumeration system that calculates taxa abundance and biomass (Gosselain and Hamilton 2000). The greatest angular linear dimension was determined by the longest linear dimension of a single cell (including spines) or the largest dimension of a colony or filament (including mucilage and/or spines). Estimates of algal wet weight (wwt) biomass were acquired by converting measured cell volumes, calculated for each taxon, to a biological mass, assuming a freshwater community assemblage and water as the predominant component of each cell (Eppley et al. 1970, Smayda 1978). Compared to cell densities, wwt biomass measures are more informative about specific taxa contributions to community production.

Total phosphorus (TP) and absorbance by colored dissolved organic matter at 375 nm (aCDOM). Total phosphorus was measured using the molybdate blue-ascorbic acid reaction (Clescerl et al. 1999). Absorption by the chromophoric dissolved organic

matter (*a*CDOM) was measured at 375 nm in a 1 cm pathlength quartz cuvette using a Shimadzu spectrophotometer (UV-2401 PC).

Growth experiments. Three ice cores and 40 L of the underlying open-water samples were collected on April 3, 2005, from the south shore sampling site ''B'' (Fig. 1). Previous sampling sites (1–5 and A) were not accessible at this date because of precarious melting ice conditions. The three combined ice cores were melted in the laboratory at 4°C in the dark. Two volumes of 13 L from the underlying water were sampled as above (see ''Sampling'') and filtered in the lab using a tangential filtration flow system (Millipore Pellicon, 0.2 μm pore size); filtered-water was transferred to two 15 L aquaria. Two liters of sample was then added to each aquarium (total volume of 15 L): one from nonfiltered open water from under the ice, and the other from the melted ice. Incubations were conducted during 13 d at 20°C to mimic spring/summer conditions (maximum water temperature in Lac Saint-Pierre ≈25°C) and verify the capacity of autotrophic protists to survive the ice-free period. PAR (light available for photosynthesis) irradiance was measured with the LI-COR meter at 230 μmol photons $\cdot m^{-2} \cdot s^{-1}$ (cool-white fluorescent GE 34W) during a 12:12 light:dark (L:D) cycle. No nutrient was added. Duplicate samples were collected every day in each aquarium and measured for chl *a* concentration as above. Growth rates were calculated from the slope of the exponential relationship between chl *a* and time.

Data analysis. One-way analyses of variance (ANOVA) were used to compare chl *a*, cell densities, and wwt biomass across samples using SPSS 11 (SPSS Inc., Chicago, IL, USA). Multiple comparisons were performed using Tukey's HSD post hoc procedures when significant differences were observed; data were normalized. We tested the differences in the concentrations of chl *a* in the upper and lower ice cover and in the water for the five stations sampled on February 27. We also tested the differences in the concentrations of chl *a*, cell densities, and wwt biomass found in each layer of the five ice cores sampled on March 22.

RESULTS

Environmental variables. Physical and chemical environment is summarized in Table 1. Ice thickness varied between 62 and 97 cm, and water underneath ranged from 34 to 1.26 cm from the bottom of the lake to the lower limit of the ice. The first 10 to 30 cm of the upper ice cover was composed of white ice, much more opaque. The lower part of the ice was composed of clear ice, more dense and transparent. Snow cover was generally small (<10 cm) due to the prevailing wind pattern. Vertical channels (1–5 mm wide) were observed within 30 cm of the bottom section of the ice cores during March and April (Fig. S2 in the supplementary material). PAR under the ice varied between 16.1 and 126.5 μmol photons $\cdot m^{-2} \cdot s^{-1}$ (1.4%–9.2% of surface irradiance), indicating that there was enough light for photosynthesis to occur. Concentrations of TP in the ice and the water underneath were generally high (15.6–97.2 $\mu g \cdot L^{-1}$) but were 1.5 to 2.5 times higher in the ice cores (lower portion) than in the water underneath for 80% of the cases. The *a*CDOM values in the ice were 10 to 18 times lower (0.4–0.7 m^{-1}) than in the water underneath, which agrees with the observations of Belzile et al. (2002) for Canadian lakes.

Sympagic community structure and distribution. Algal biomass was present in every ice core and showed high variability between stations, with concentrations of chl *a* varying from 0.5 to 169 $\mu g \cdot L^{-1}$ of melted ice (Figs. 2 and 3). Algae were clearly visible at the base of the cores (Fig. S2b in the supplementary material), but chl *a* analyses were needed to detect algae in the upper part of the cores. Algae were present throughout the ice, but maximal concentrations were observed at the ice–water interface and when the channels were present (Fig. 3). For example, no channels were observed during February sampling, and no significant differences found in the chl *a* distribution from the upper, lower ice cover and underlying water, except for station 3 where the lower ice contained more biomass than the upper ice cover and underlying water ($P < 0.01$). However, on March 22, channels were observed in all samples, and chl *a* concentrations were higher in the lower ice than in the upper ice or water underneath (Fig. 3). Similar patterns were observed with cell densities and estimated wwt biomass (Fig. 4).

The distribution of sympagic organisms within complete ice cores, from the south and north shores, revealed the presence of up to 55 taxa, including diatoms, cyanobacteria, ciliates, and rotifers (Table 2). Cell densities varied from almost absent (<53,000) to 1,328,000 cells $\cdot L^{-1}$ along the examined transects, and up to 1,430,000 cells $\cdot L^{-1}$

TABLE 1. Summary of environmental variables in Lac Saint-Pierre during winter 2005.

Date	Station	Latitude (°N)	Longitude (°W)	Ice thickness (cm)	Water depth (cm)	% T	TP upper ice ($\mu g \cdot L^{-1}$)	TP lower ice ($\mu g \cdot L^{-1}$)	TP water ($\mu g \cdot L^{-1}$)	*a*CDOM upper ice (m^{-1})	*a*CDOM lower ice (m^{-1})	*a*CDOM water (m^{-1})
February 27	1	46°08′16,4′	72°50′50,2′	76	34	2.60	97.2	59.0	81.0	0.5	0.7	9.8
	2	46°08′23,8′	72°50′55,4′	77	105	3.80	32.2	50.5	19.6	0.4	0.6	9.0
	3	46 08′45,3′	72°51′09,5′	97	123	2.99	52.0	82.7	37.0	0.6	0.4	7.2
	4	46°12 44,9′	72°55′44,5′	62	126	9.22	24.0	24.2	15.6	1.1	0.6	8.3
	5	46°12′51,5′	72°55′50,0′	73	48	6.79	29.9	63.5	36.1	0.2	0.7	7.0
March 22	A	46°08′26,6′	72°51′41,4′	91	61	1.42	NA	NA	56.9	0.9	0.9	9.0
April 3	B	46°09′51,3′	72°46′09,1′	84	54	1.40	NA	NA	NA	NA	NA	NA

% T, the percentage of incident light that penetrates the ice cover; TP, the total phosphorus; and *a*CDOM, the light absorption by the colored dissolved organic matter, which is a function of its concentration. NA, data not available.

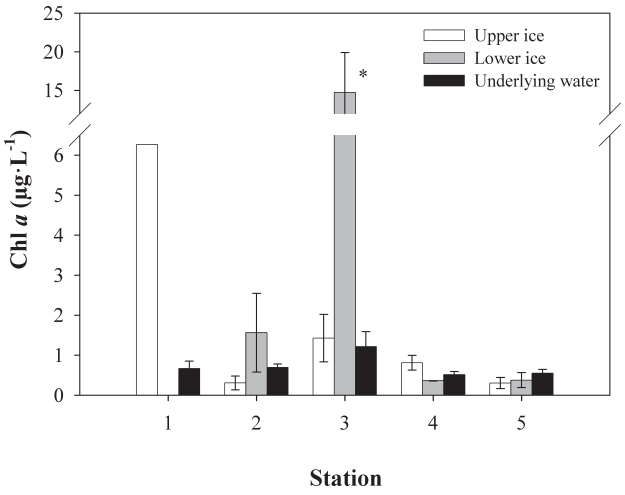

FIG. 2. Distribution of chl *a* biomass in the upper, lower half of 15 ice cores and in the underlying water sampled in the north and south zones of Lac Saint-Pierre (stations 1–5) on February 27, 2005. Data are mean values with ±1 SE. Data not available for lower ice at station 1. * refers to significant difference ($P < 0.01$) in chl *a* concentration.

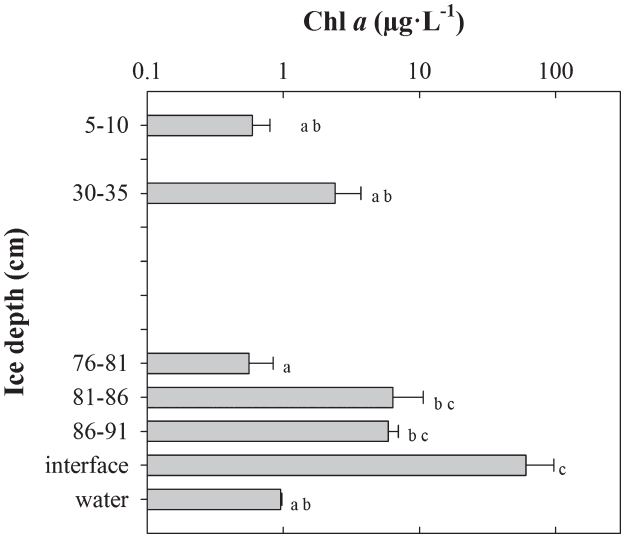

FIG. 3. Fine-scale vertical distribution of the chl *a* biomass in five ice cores from station A, on March 22, 2005. Data are mean values with ±1 SE. Bars with the same letter indicate no significant difference, $P < 0.05$.

in areas of high abundance in the cores. Likewise, estimated wwt biomass levels also varied greatly, from 0.9 to 687 $\mu g \cdot L^{-1}$ along the examined transects and up to 28,100 $\mu g \cdot L^{-1}$ in regions of high abundance (station A, ice–water interface) (Table 2). There was no obvious trend in numbers and biomass distributions between the north and south stations. Species compositions were different between north and south, with a strong Bacillariophyceae presence in the south, while along the

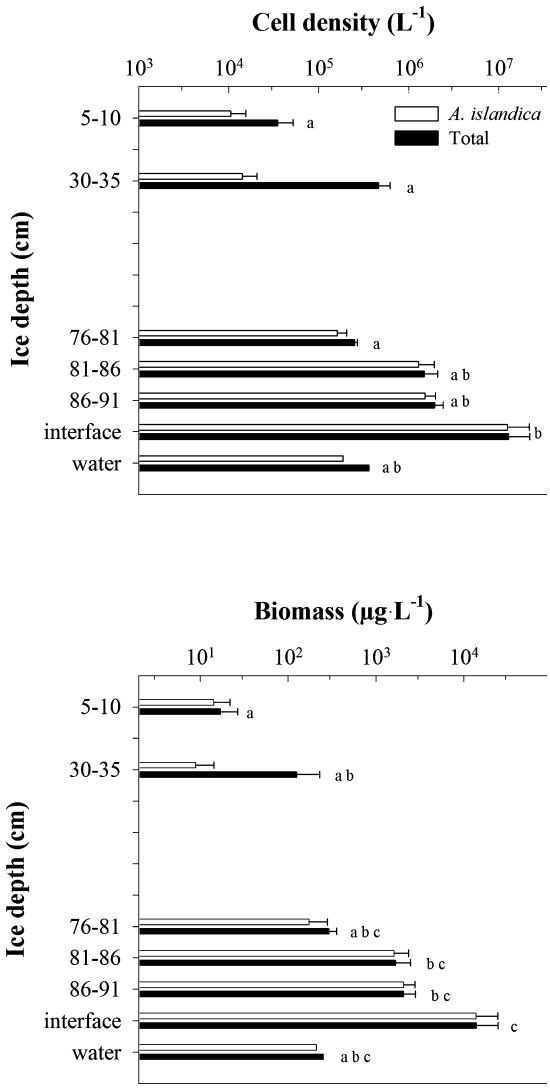

FIG. 4. Fine-scale vertical distribution of the cell densities and wwt biomass in south shore ice cores. Totals and estimates for the dominant species, *Aulacoseira islandica*, are presented. Log scale; data points presented with ±1 SE. Bars with the same letter indicate no significant difference, $P < 0.05$.

north shore, samples were predominately Chlorophyta and Bacillariophyceae. *A. islandica*, *Diatoma tenuis*, *Nitzschia dissipata*, *Tabellaria flocculosa*, and *Navicula* spp. were the more evident taxa in the south, with *Ankyra* sp., *Monoraphidium* spp., and centric diatoms more prominent in the north (see Table 2 for taxonomic authors).

Fine-scale vertical distribution of algae from the south shore (station A) revealed that ice channels at the base of the core were colonized almost exclusively by dividing filamentous chains of *A. islandica* (Fig. 4; Fig. S1, a–d, in the supplementary material). Numbers declined exponentially moving up into the channels, and 30–35 cm from the upper surface there was a diversification of the flora (Fig. 4). This zone of high diversity was characterized by 26 taxa

TABLE 2. Sympagic community structure.

Category	No. taxa	Growth type	Major taxa	Biovolume · cell^{-1} (μm^3)	Density (×1,000) (L^{-1})	Biomass (wwt) ($\mu g \cdot L^{-1}$)	Cell carbon biomass ($\mu g \cdot L^{-1}$)	% densities	% biomass
Filaments	13	P, S	*Aulacoseira islandica*	534–1,694	1.2–28,612	1.0–27,000	0.3–1,210	0.3–98	0.6–99.5
		P, B	*Limnothrix* spp.	11–13	8.1–25.5	0.1–0.3	<0.1–0.1	1.7–45	0.3–1.7
		P	*Diatoma tenuis* C. Agardh	210–468	0.8–43.3	0.2–11	0.1–4	0.3–7.6	0.1–11
		P	*Diatoma elongatum (=D. tenuis)*	210–240	0.8–24.7	0.2–6.0	<0.1–2	0.1–5.7	<0.1–1.8
		P	*Diatoma hyemalis* (Roth) Heib.	5,831–8,330	1.6–7.9	10–46	39,216	0.4–3.2	4.1–41.7
		P	*Fragilaria crotonensis* Kitton	341–1,328	0.2–1.2	0.1–1.6	<0.1–0.6	0.3–14.3	0.4–19.9
		P	*Fragilaria* spp.	36.8	2.4	<0.1	<0.1	5	0.26
		P	*Melosira varians* C. Agardh	6,775	4.2–71.7	30–490	9–140	0.1–5.5	0.8–35.8
		P, B	*Pseudoanabaena* spp.	0.6–3.8	3.8–96	<0.1–0.4	<0.1–0.15	0.9–21	<0.1
Colonies	9	P	*Scenedesmus* cf. *acutiformis* Schröd.	24–38	3.1–3.8	<0.1–0.1	<0.1–0.2	0.5–0.9	<0.1–0.1
		P	*Scenedesmus dimorphus* (Turpin) Kütz.	102–141	4.7–9.7	0.5–1	0.2–0.4	1.7–10.1	1.4–1.7
		P	*Scenedesmus* spp.	140	5.7	0.8	0.3	2.3	0.2
		P	*Woronichinia* cf. *naegeliana* (Unger) Elenkin	7	232	2	0.6	40.7	2.1
Single cells, nonmotile	27	P	*Ankyra* cf. *judayi* (G. M. Smith) Fott	50	4.1–31.8	0.2–1.9	<0.1–0.5	0.3–10.6	<0.1–8.9
		E	*Cocconeis placentula* Ehrenb.	9,952	0.4–15.7	4–160	1–44	0.8–3.7	1.6–47.9
		P	*Cosmarium* spp.	2,217	1.9	4	1.5	0.3	5.2
		P	*Stephanodiscus/Cyclotella* spp.	58.9–301	1.9–290	0.1–60	0.1–23	0.1–5.6	<0.1–14.9
		B, E	*Encyonema minuta* (Hilse ex Rabenh.) D. G. Mann	89.8–1,649	4–39.7	3–6	1–2	0.9–16.2	2–3.2
		P	*Monoraphidium contortum* (Thur.) Komárk.-Legn.	35–56	0.4–3.8	<0.1–0.2	<0.1	0.1–0.9	<0.1–0.2
		P	*Monoraphidium* cf. *griffithii* (Berk.) Komárk.-Legn.	38–57	0.3–0.8	<0.1	<0.1	0.09–0.6	<0.1–0.1
		P	*Monoraphidium* spp. (including *M.* cf. *obtusum*)	9–98	3.8–141	0.4–1	0.2–0.5	0.8–24.8	0.1–1.5
		P	*Stephanodiscus niagare* Ehrenb.	12,936	1.6	20	6.6	0.4	7.6
		B	*Ulnaria ulna* (Nitzsch) Compère	7,200–8,820	0.8–2.8	6–30	2–8	0.1–0.7	6.8–7.6
Single cells, motile	6	P	*Chlamydomonas* spp. (including *C. augustae*)	26–1,027	0.4–51	<0.1–4	<0.1–1.2	0.1–11.8	<0.1–25
		P	*Gymnodinium* spp.	333–927	2.9–17	2–16	<0.1–5	0.2–3.9	0.2–4.8
Horizontal profile (range of totals)			All taxa in a sample		52–1,328	21–687	4.7–118		
Vertical profile (range of totals)			All taxa in a sample		1– 1,430	0.9–28,100	0.4–1,255		

Summary of the photosynthetic algae enumerated. The major taxa (or taxa groups) with >5% abundance or >5% biomass are presented with growth type (P denotes planktonic, B denotes benthic, S denotes sympagic, and E denotes epiphytic), and the ranges in calculated cell biovolume, cell densities, estimated cell wet weight (wwt) biomass, and estimated carbon content (Eppley et al. 1970). The large ranges observed highlight the variations from site to site and the dramatic changes in community structure throughout the ice core.

or taxonomic groups split between the Chlorophyta and Bacillariophyceae. The chlorophytes were single-cell or small colonial forms, dominated in numbers by *Monoraphidium* cf. *obtusum*. Other chlorophytes included *Chlamydomonas augustae, Chlamydomonas* sp., *Scenedesmus spinosus, S. dimorphus, Cosmarium* spp., and *Mougeotia* sp. (Fig. S1, h, i, and m). The diatom community was a mix of large and small, colonial and solitary forms (Fig. S1, e–g). The dominant diatom by numbers was *D. tenuis* (including *D. elongatum*), with *A. islandica* and *D. tenuis* controlling the biomass. Other diatoms included *Achnanthidium minutissimum, N. dissipata, N. gracilis, Ulnaria ulna,* and *Cymbella* spp. At least one species of *Gymnodinium* (*G.* cf. *uberimuum*) was present with a sparse contingent of cyanobacteria, which included picoplankton mats, *Limnothrix* sp., and *Woronichinia naegeliana* (Fig. S1, k and l). Cryptophytes were also present in low numbers and biomass (Fig. S1).

Growth experiments. We tested the viability and growing capacity of ice algae by performing laboratory incubations of ice and water samples. Results revealed that ice algae and under-ice phytoplankton can grow efficiently without nutrient additions in warm open water (Fig. 5). The net chl *a* growth rates were similar in both cultures (0.21 and $0.23 \cdot d^{-1}$) and continuous over 13 d. Initial chl *a* concentration was about five times higher in the aquarium containing the melted ice core inoculum. Numerous heterotrophic protists (amoebae, ciliates), along with rotifers including *Trichotria tetractis* and *Tricocera* cf. *similis* were also present (Fig. S1, n–q).

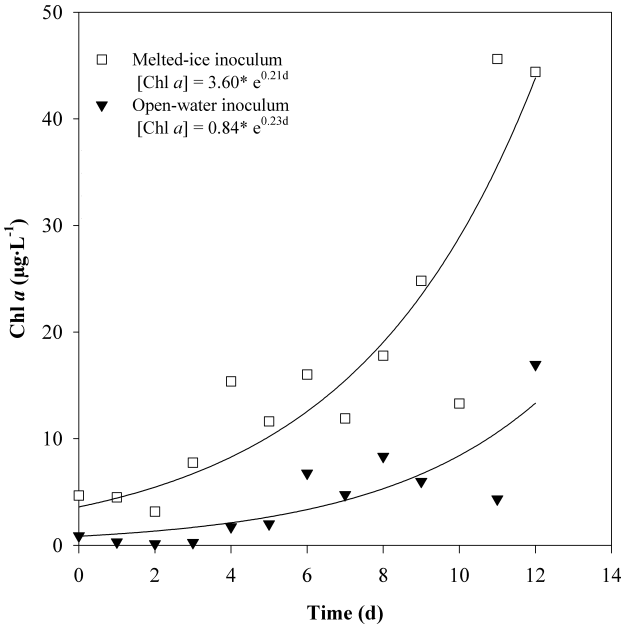

FIG. 5. Growth of ice algae and winter phytoplankton in open water under spring/summer temperature conditions.

DISCUSSION

Sympagic community structure and biodiversity. Algal communities present in freshwater ice during springtime (March and April 2005) in the St. Lawrence River (Lac Saint-Pierre) are mostly represented by planktonic and tychoplanktonic taxa. These species have discrete behaviors, allowing them to flourish in distinct spatial niches; for example, *A. islandica* passively migrates from the sediments toward the bottom ice surface in late winter (Müller 1906, Burns and Mitchell 1974, H. Kling pers. comm.). Several species of *Aulacoseira* (e.g., *A. granulata*), remain dormant as resting cells for long time periods (up to 17 months) in sediments until appropriate temperature and light conditions allow them to rapidly develop (Burns and Mitchell 1974). The shallow water environment (0.34–1.26 m) prevailing in Lac Saint-Pierre could favor both active and passive migration of algae from the sediment up through the water column, thereby facilitating colonization of bottom ice and ice cavities. Our observation of dividing cells corroborates that freshwater ice acts as an appropriate substratum offering ideal light and nutrient conditions to favor algae growth (Fig. S1, a–d). These observations combined with the increasing abundances of algae in the lower part of the ice cover and in the channels, closer to the water, demonstrate that a substantial part of the algae community colonizes the ice cover rather than being trapped during ice formation. Ice thus represents during wintertime an additional, abundant substratum for algae growth in the St. Lawrence River.

Our experiments with sympagic communities from Lac Saint-Pierre have demonstrated their rapid growth capacity and strong tolerance to rising temperatures. This finding supports the ''seeding hypothesis'' (Lizotte 2001)—algae in, or closely associated with, ice act as an inoculum to boost phytoplanktonic primary productivity during ice breakup. *A. islandica* is a typical alga in cold waters during early spring (Siver and Kling 1997, Bondarenko et al. 2006), and blooms of this species can be observed immediately after ice breakup in Lake Winnipeg, Canada (H. Kling pers. comm.). This suggests a potential for sympagic algae as important inputs of matter and energy in lake and riverine systems. Interestingly, *A. islandica* represented as much as 99.5% (median 86%) of the total algal biomass in candled ice. Large quantities of ice drifting downstream from Lac Saint-Pierre may thus act as an inoculum for primary productivity of not only the lake itself, but also of downstream portions of the St. Lawrence system and may contribute to overall microbial biodiversity.

Vertical distribution and colonization. The vertical distribution in ice cores observed at station A showed a similar pattern within each core, with the highest biomass in the lower-ice portion. The

increasing chl *a* concentrations and cell counts in the bottom section of the ice are explained by the presence of channels in the lower 30 cm combined with the aggregation of a sympagic flora at the ice–water interface. Concentrations were maximal in the bottom 5 cm of ice and included late winter/spring blooming algae, such as *A. islandica*, which colonized the channels at that time, along with the less abundant tychoplanktonic algae (e.g., *Nitzschia* spp., *D. tenuis*, *Rhoicosphenia abbreviata*, and *T. fenestrata*) growing at the ice–water interface. Vertical channels in the ice were first observed on March 22 and subsequently in April during 2005. Similar results were observed in 2006 (data not shown). The presence of candled ice channels in our samples is consistent with the warmer weather and frequent thawing events observed during March and April in Lac Saint-Pierre (Ashton 1978).

The presence of up to 55 taxa, including cyanobacteria, ciliates, and rotifers, confirms that bottom-up trophic interactions can operate within the ice and supports our contention that ice masses within shallow zones house a diversity of viable life forms. The exclusion of CDOM during ice formation could also result in solute concentration in channels and therefore favor microbial activity and food-web interactions (Belzile et al. 2002). This taxonomic diversity is within the range of that of ice algae from the Arctic Ocean and marginal seas, which varies between 33 and 251 taxa (von Quillfeldt et al. 2003 and references therein). The declining numbers of *A. islandica* coupled with the presence of a diverse community from at least five phyla, along the channel interior toward the surface, illustrates the temporal formation of these communities. A change in colonization from predominantly planktonic forms in the lower section to a mix of planktonic and benthic forms in the ice interior reflects the succession of species establishment during ice formation. The youngest communities (predominantly one species, *A. islandica*) are living at the bottom of the ice since early spring, followed by older ones presumably established earlier during the winter season.

Horizontal distribution and environmental conditions. Horizontal heterogeneity of ice algae was reported in many marine studies (Cota and Smith 1991, Gosselin et al. 1997, Rysgaard et al. 2001). Algal distribution over large surface areas (km scale) was explained by water mass properties, such as salinity and nutrients (Gosselin et al. 1986, Cota and Smith 1991, Legendre et al. 1996). In contrast, small-scale variations (10 m scale) were attributed to light-controlling factors, such as snow cover and ice texture (Gosselin et al. 1986, Legendre et al. 1991, Ambrose et al. 2005). Our limited number of sampling sites (including those on different dates), and large variations in algal numbers and biomass within ice cores, do not allow us to truly characterize the spatial distribution in Lac Saint-Pierre. However, our results clearly demonstrate that protist communities are colonizing ice under differing temperature, nutrient, and light conditions. Further, our sampling sites were located in two different water masses characterized by distinct concentrations of nutrients and dissolved organic matter, among other characteristics (Frenette et al. 2006). Additional research is needed to understand the controlling factors of ice algal distribution in freshwater ecosystems and the spatial and temporal dynamics of algal colonization.

CONCLUSION

This research is the first evidence for a dynamic, diversified sympagic environment in temperate river ice. Results show that this previously unappreciated ecosystem is likely to have a considerable impact on winter and spring primary production and contribute to the overall microbial biodiversity. However, historical trends for lake and river ice cover around the Northern Hemisphere during the last 150 years revealed consistent evidence for later freezing and earlier ice breakup (Magnuson et al. 2000). In winter 2006–2007, we observed that the ice cover formed ~1 month later than usual on Lac Saint-Pierre. Climate changes are thus likely to threaten what appears to be a key habitat for life in river ecosystems.

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Ambrose, W. G., von Quillfeldt, C., Clough, L. M., Tilney, P. V. R. & Tucker, T. 2005. The sub-ice algal community in the Chukchi sea: large- and small-scale patterns of abundance based on images from a remotely operated vehicle. *Polar Biol.* 28:784–95.

Ashton, G. D. 1978. River ice. *Annu. Rev. Fluid Mech.* 10:369–92.

Belzile, C., Gibson, J. A. E. & Vincent, W. F. 2002. Colored dissolved organic matter and dissolved organic carbon exclusion from lake ice: implications for irradiance transmission and carbon cycling. *Limnol. Oceanogr.* 47:1283–93.

Bondarenko, N. A., Timoshkin, O. A., Ropstorf, P. & Melnik, N. G. 2006. The under-ice and bottom periods in the life cycle of *Aulacoseira baicalensis* (K. Meyer) Simonsen, a principal Lake Baikal alga. *Hydrobiologia* 568:107–9.

Burns, C. W. & Mitchell, S. F. 1974. Seasonal succession and vertical distribution of phytoplankton in Lake Hayes and Lake Johnson, South Islands, New Zealand. *N. Z. J. Mar. Freshw. Res.* 8:167–209.

Clescerl, L. S., Greenberg, A. E. & Eaton, A. D. 1999. *Standard Methods for the Examination of Water and Wastewater.* 20th ed. American Public Health Association, Washington, DC, 1325 pp.

Cota, G. F., Legendre, L., Gosselin, M. & Ingram, R. G. 1991. Ecology of bottom ice algae: I. Environmental controls and variability. *J. Mar. Syst.* 2:257–77.

Cota, G. F. & Smith, R. E. H. 1991. Ecology of bottom ice algae: II. Dynamics, distributions and productivity. *J. Mar. Syst.* 2:279–95.

Eppley, R. W., Reid, F. M. & Strickland, J. D. H. 1970. The ecology of the plankton off La Jolla, California, in the period April through September 1967 – pt. III. Estimates of phytoplankton crop size, growth rate and primary production. *Bull. Scripps Inst. Oceanogr.* 17:33–42.

Felip, M., Camarero, L. & Catalan, J. 1999. Temporal changes of microbial assemblages in the ice and snow cover of a high mountain lake. *Limnol. Oceanogr.* 44:973–87.

Frenette, J. J., Arts, M. T., Morin, J., Gratton, D. & Martin, C. 2006. Hydrodynamic control of the underwater light climate in fluvial Lac Saint-Pierre. *Limnol. Oceanogr.* 51:2632–45.

Gosselain, V. & Hamilton, P. B. 2000. Algamica: revisions to a key-based computerized counting program for free-living, attached and benthic algae. *Hydrobiologia* 438:138–42.

Gosselin, M., Legendre, L., Therriault, J. C., Demers, S. & Rochet, M. 1986. Physical control of the horizontal patchiness of sea-ice microalgae. *Mar. Ecol. Prog. Ser.* 29:289–98.

Gosselin, M., Levasseur, M., Wheeler, P. A., Horner, R. A. & Booth, B. C. 1997. New measurements of phytoplankton and ice algal production in the Arctic Ocean. *Deep-Sea Res. Part II Top. Stud. Oceangr.* 44:1623–44.

Hamilton, P. B., Proulx, M. & Earle, C. 2002. Enumerating phytoplankton with an upright compound microscope using a modified settling chamber. *Hydrobiologia* 444:171–5.

Hasle, G. R. 1978. Using the inverted microscope. *In* Sournia, A. [Ed.] *Phytoplankton Manual.* United Nations Educational, Scientific and Cultural Organization, Paris, pp. 191–6.

Horner, R. S. F., Dieckmann, G. S., Gulliksen, B., Hoshiai, T., Legendre, L., Melnikov, I. A., Reeburgh, W. S., Spindler, M. & Sullivan, C. W. 1992. Ecology of sea ice biota. 1. Habitat, terminology, and methodology. *Polar Biol.* 12:417–27.

Kaartokallio, H. 2004. Food web components, and physical and chemical properties of Baltic Sea ice. *Mar. Ecol. Prog. Ser.* 273:49–63.

Legendre, L., Aota, M., Shirasawa, K., Martineau, M.-J. & Ishikawa, M. 1991. Crystallographic structure of sea ice along a salinity gradient and environmental control of microalgae in the brine cells. *J. Mar. Syst.* 2:347–57.

Legendre, L., Robineau, B., Gosselin, M., Michel, C., Ingram, R. G., Fortier, L., Therriault, J. C., Demers, S. & Monti, D. 1996. Impact of freshwater on a subarctic coastal ecosystem under seasonal sea ice (southeastern Hudson Bay, Canada). 2. Production and export of microalgae. *J. Mar. Syst.* 7:233–50.

Lizotte, M. P. 2001. The contribution of sea ice algae to Antarctic marine primary production. *Am. Zool.* 41:57–73.

Lund, J. W. G., Kipling, C. & LeCren, E. D. 1958. The inverted microscope method of estimating algal numbers and the statistical basis of estimating counting. *Hydrobiologia* 11:143–70.

Magnuson, J.-J., Robertson, D. M., Benson, B. J., Wynne, R. H., Livingstone, D. M., Arai, T., Assel, R. A., et al. 2000. Historical trends in lake and river ice cover in the Northern Hemisphere. *Science* 289:1743–6.

Marker, A. F. H., Nusch, E. A., Rai, H. & Riemann, B. 1980. The measurements of pigments in freshwaters and standardization of methods: conclusions and recommendations of the workshop. *Ergeb. Limnol.* 14:91–106.

Michel, C., Nielsen, T. C., Nozais, C. & Gosselin, M. 2002. Significance of sedimentation and grazing by ice micro- and meiofauna for carbon cycling in annual sea ice (northern Baffin Bay). *Aquat. Microb. Ecol.* 30:57–68.

Müller, O. 1906. Pleomorphismus, Auxosporen und Dauersporen bei Melosira-Arten. *Jahrb. Wiss. Bot.* 43:49–88.

Nozais, C., Gosselin, M., Michel, C. & Tita, G. 2001. Abundance, biomass, composition and grazing impact of the sea-ice meiofauna in the North Water, northern Baffin Bay. *Mar. Ecol. Prog. Ser.* 217:235–50.

Priscu, J. C., Fritsen, C. H., Adams, E. E., Giovannoni, S. J., Paerl, H. W., McKay, C. P., Doran, P. T., Gordon, D. A., Lanoil, B. D. & Pinckney, J. L. 1998. Perennial Antarctic lake ice: an oasis for life in a polar desert. *Science* 280:2095–8.

von Quillfeldt, C. H., Ambrose, W. G. & Clough, L. M. 2003. High number of diatom species in first-year ice from the Chukchi Sea. *Polar Biol.* 26:806–18.

Rysgaard, S., Kuhl, M., Glud, R. N. & Hansen, J. W. 2001. Biomass, production and horizontal patchiness of sea ice algae in a high-Arctic fjord (Young Sound, NE Greenland). *Mar. Ecol. Prog. Ser.* 223:15–26.

Siver, P. A. & Kling, H. 1997. Morphological observations of *Aulacoseira* using scanning electron microscopy. *Can. J. Bot.* 75:1807–35.

Smayda, T. J. 1978. From phytoplankton to biomass. *In* Sournia, A. [Ed.] *Phytoplankton Manual.* United Nations Educational, Scientific and Cultural Organization, Paris, pp. 273–379.

Venrick, E. L. 1978. How many cells to count? *In* Sournia, A. [Ed.] *Phytoplankton Manual.* United Nations Educational, Scientific and Cultural Organization, Paris, pp. 167–80.

Vincent, W. F. 1988. *Microbial Ecosystems of Antarctica.* Cambridge University Press, Cambridge, UK, 320 pp.

Supplementary Material

The following supplementary material is available for this article:

Figure S1. Representative algal taxa from the living flora of Lac Saint-Pierre freshwater river ice.

Figure S2. Colonization by freshwater ice algae in the channels of the candled ice.

This material is available as part of the online article from: http://www.blackwell-synergy.com/doi/abs/10.1111/j.1529-8817.2008.00481.x.

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