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RESEARCH ARTICLE

Seston fatty acid composition and copepod RNA:DNA ratio with respect to the underwater light climate in fluvial Lac Saint-Pierre

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Abstract The relationship between the underwater light availability at different wavelengths (from 351 to 700 nm) and the fatty acid (FA) composition of seston, as well as the trophic transfer of fatty acids from producers to consumers and its influence on copepod growth condition, were investigated throughout fluvial Lac Saint-Pierre (Québec, Canada). Seston and zooplankton were collected at 11 sampling sites located within distinct water masses discriminated according to their underwater spectral characteristics. Diffuse light attenuation coefficients ($K_{d(\lambda)}$) varied among sampling sites and wavelengths (λ) and were negatively correlated to seston composition in some essential fatty acids. Particularly, the relationships between $K_{d(\lambda)}$ and the seston concentration in 20:5n3 and 22:6n3 differed and were wavelength dependent, being stronger for λ close to the absorption maxima of chlorophyll *a*, suggesting a potential link with photosynthetic processes. The concentrations of 16:1n7, 18:3n3 and 20:5n3 in copepods were strongly correlated to those in the seston, which points towards the trophic transfer of these fatty acids between primary producers and herbivorous consumers. Moreover, the growth condition of copepods, as expressed by their RNA:DNA ratio, was correlated to the concentrations of 16:1n7, 18:3n3 and 20:5n3 in the seston and in copepods. Our field study sheds light on the potential importance, yet to be precised, of specific wavelengths as a driver of Lac Saint-Pierre's productivity through their influence on fatty acids composition of seston and its nutritional quality for primary consumers.

Keywords Fatty acids · RNA/DNA ratio · Underwater light climate · Seston · Copepod

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Introduction

Among all levels of aquatic food webs, the plant/animal interface is the most variable and least predictable link (Müller-Navarra et al. 2000). It deserves peculiar attention as the factors governing the growth and reproduction of herbivorous primary consumers can ultimately translate up or down the food chain and influence the productivity and biodiversity of aquatic ecosystems. Zooplankton growth responds to various factors, among which food quality has proven to be of more relevance than food quantity. Indeed, while a sufficient supply of poor-quality food may sustain the biomass of non-growing individuals, high growth rate and reproduction rely on a high-quality diet that meets the consumer requirements for elements and macromolecules (Andersen et al. 2007). Particularly, the importance of food quality as a key factor governing zooplankton growth prevails for herbivorous copepods which are able to selectively feed on nutritionally rich phytoplankton despite its low contribution to total POM in fluvial ecosystems (Martineau et al. 2004).

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Fatty acids are useful indicators of the nutritional quality of primary producers for planktonic grazers. Saturated and monounsaturated fatty acids (SAFA and MUFA, respectively) are of poorer nutritional quality than polyunsaturated fatty acids (PUFA)-rich organic matter, which constitutes a highly valuable food item for consumers (Brett and Müller-Navarra 1997; Parrish 2009). MUFA and SAFA are major components of the neutral lipids fraction, which mainly serves as energy storage reserves and generally increases in phytoplankton as a result of exposure to stressful environmental conditions (Huggins et al. 2004). PUFA, such as eicosapentaenoic acid (EPA; 20:5n3) and docosahexaenoic acid (DHA; 22:6n3), are usually associated with the polar lipids fraction and recognized as important structural components of cell membranes influencing many physiological processes (Guschina and Harwood 2009). From a consumer's perspective, many PUFA are also essential fatty acids considering that they cannot be synthesized by the consumer and should therefore be provided through its diet. PUFA are thus recognized as key factors of food quality, influencing the growth and reproduction of zooplankton as well as the efficiency of trophic transfer in aquatic food webs (Brett and Müller-Navarra 1997; Müller-Navarra et al. 2000; Kainz et al. 2004).

The fatty acid composition of algae is influenced by various physical and chemical factors such as temperature, salinity, light and nutrient supply (Guschina and Harwood 2009). In large river ecosystems, which are usually nutrient-replete, the underwater light climate is thought to be one of the most important environmental factors affecting the production and fatty acid content of photosynthetic organisms (Reynolds and Descy 1996; Wainman et al. 1999; Guschina and Harwood 2009). Particularly, fatty acid synthesis in primary producers relies on the energy provided by NADPH and generated during the light reactions of photosynthesis (Wainman et al. 1999), so that lipid production tends to increase with the intensity of photosynthetically active radiations (PAR, 400–700 nm). Conversely, excessively high PAR irradiance increase oxidative damage of PUFA (Guschina and Harwood 2009). Triacylglycerols, which are abundant storage products and can be easily catabolised to provide metabolic energy, are mostly synthesized during the light phase of photosynthesis, stored in cytosolic lipid bodies, and then reutilized for polar lipid synthesis in the dark (Thompson 1996). Ultraviolet radiations (UVR, 280–400 nm) are also known to alter the lipid composition of phytoplankton by reducing their content in polyunsaturated fatty acids (Wang and Chai 1994). Moreover, the effect of light on seston fatty acid composition may be modulated by its influence on the taxonomic composition of primary producers. For instance, high UVR irradiance tends to promote the abundance of chlorophycae (Wellnitz and Ward 1998) which, based on its fatty acid composition, are generally of poorer nutritional value than most diatom species.

Lac Saint-Pierre (Québec, Canada) is the largest fluvial lake and last enlargement of the St. Lawrence River before the estuary. It is strongly connected with the terrestrial ecosystem at various spatial and temporal scales. Its extensive floodplain (180 km^2) and the confluence of seven tributaries (Frenette et al. 2006; Lapierre and Frenette 2009) drain waters characteristic of the Canadian Shield, Laurentian Great Lakes and rivers from farmlands, all which contribute to the formation of a mosaic of distinct water masses. The large width/depth ratio of Lac Saint-Pierre (13.1 km wide and 3.17 m deep at mean discharge) reduces lateral mixing between the lake's water masses. Because of contrasting land use in their respective catchments, water masses of Lac Saint-Pierre differ in their concentrations of dissolved and particulate organic and inorganic suspended matter, which translates into a strong longitudinal and lateral heterogeneity of the underwater spectral characteristics, both in UVR and PAR spectra (Frenette et al. 2003, 2006). Considering the dependency of fatty acid composition in primary producers upon the underwater light climate, the contrasted underwater spectral characteristics across Lac Saint-Pierre's water masses are likely to influence the FA composition of seston and thus its nutritional quality for herbivorous zooplankton. In turn, this may translate into varying zooplankton growth.

Within this framework, our study aims to (1) investigate the relationship between the underwater spectral characteristics within Lac Saint-Pierre and the nutritional quality of the seston, evaluated for its fatty acid composition, (2) assess the transfer of essential fatty acids from producer to consumer, by relating the fatty acid composition of seston to that of copepods, and (3) investigate the relationship between the nutritional quality of seston and the growth condition of copepods, using the RNA:DNA ratio as a biochemical indicator of zooplankton growth.

Materials and methods

Eleven stations, located within six distinct water masses of Lac Saint-Pierre, were sampled on 13 July 2007 aboard the RV "Lampsilis" from the University of Quebec, Trois-Rivières (Fig. 1). The exact station locations were fixed by a digital Global Positioning System (±2 m accuracy).

Identification of Lac Saint-Pierre water masses

The location of the sampling sites relative to each water mass was identified based on Landsat-5 Enhanced Thematic Mapper (ETM) satellite images of Lac Saint-Pierre taken on 7 July 2007 (no data available for 13 July 2007) and by using

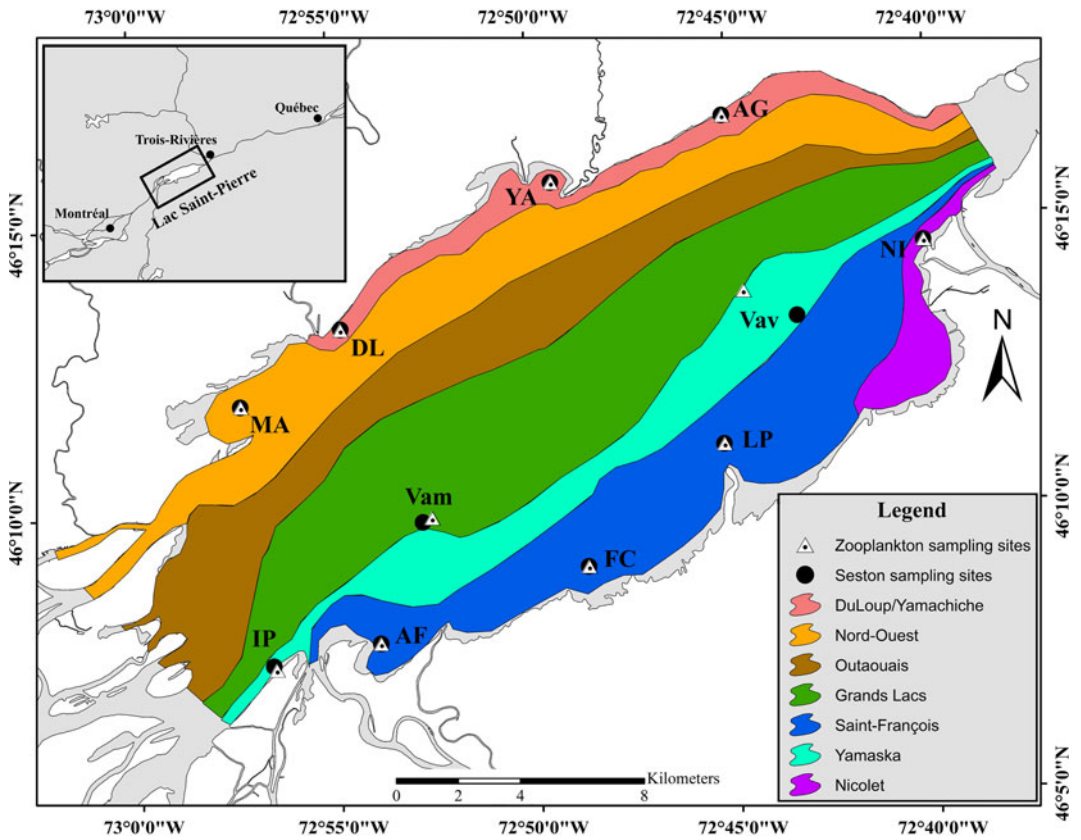

Fig. 1 Location of seston and zooplankton sampling sites within Lac Saint-Pierre (LSP). The distinct water masses of the LSP, discriminated by their spectral characteristics (see "Materials and methods" for details), are also indicated

the approach described in Frenette et al. (2006). Briefly, inland waters were discriminated according to their spectral properties, which are mainly determined by chromophoric dissolved organic matter (CDOM) and suspended particulate inorganic matter (SPIM). Optical satellite sensors allow the characterization of CDOM and SPIM since they have distinct spectral responses. The green band (520–600 nm) is particularly sensitive to the absorption by CDOM while the red band (630–690 nm) shows the reflection characteristics of SPIM. Based on a similarity matrix of the per-pixel red to green surface reflection ratio, a clustering analysis, processed by an unsupervised clustering algorithm (ISODATA/PCI-Geomatica 10), was used to identify the specific lateral limits of each water mass and the relative location of each sampling site. Inspection of the St. Lawrence River and major tributaries showed little variation in flow and water level between the dates of satellite image acquisition and the sampling campaign, supporting the assumption of the relative stability of water masses during this period.

Underwater light climate

At each sampling station, vertical profiles of underwater downwelling irradiance (E_d) were measured every 1 nm throughout the UVA (ultraviolet-A radiations, from 351 to 400 nm) and PAR (photosynthetically active radiations, from 400 to 700 nm) spectra with a spectroradiometer (HyperPro, Satlantic Instruments). Though the instrument did not allow for the measurement of the entire UVA spectrum (i.e., 315–400 nm), the downwelling irradiance profiles from 351 to 400 nm were used as a proxy for UVA radiation. The spectroradiometer was slowly lowered through the water column and measurements were taken every 0.02 m from surface to $\sim$30 cm above the bottom. Light data were corrected automatically for "dark irradiance" values by the shutter dark correction method. These are continuously recorded during the measurements by occulting the input fiber with an optical shutter, typically after every five light samples. Further details are available in *Prosoft* software user manual (Satlantic, Document SAT-DN-00228-Rev. C). Downward diffuse vertical attenuation coefficients ($K_{d(\lambda)}$) were calculated at a given wavelength (λ) taken each and every 10 nm from 351 to 700 nm by linear regression of the natural logarithm of E_d versus depth. These $K_{d(\lambda)}$ values were also integrated from 351 to 400 nm and from 400 to 700 nm to calculate the diffuse vertical attenuation coefficients for the whole UVA ($K_{d(UVA)}$) and PAR ($K_{d(PAR)}$) spectra, respectively.

Sampling

At each station, temperature and conductivity were measured using a YSI 6600 EDS-M sensor array (Yellow Spring Instruments, Inc.) connected to a pumping system collecting water at a depth of ~0.5 m. Given the shallow depth and the advective forcing that generally prevails in fluvial ecosystems, a point-source sampling is representative of these variables in the entire water column. The water drawn from the pumping system was filtered onto precombusted (4.5 h at 500°C) and preweighed 45 mm Millipore GF/F filters (nominal pore size 0.7 µm). The filtrate was collected into acid washed polycarbonate cryovials for later analyses of dissolved inorganic nutrients. The filters were used for the determination of seston FA composition. Both the filtrates and the filters were immediately frozen onboard at −80°C until analysis in the laboratory. Zooplankton were collected using a conical net (1 m mouth opening, 150-µm mesh size) hauled horizontally for 5 min at 3.5 knots and at a depth close to that of the water sample collected. Zooplankton samples were collected at the same locations as that of the water samples except at station Vav due to low water levels which constrained the deployment of the zooplankton net (Fig. 1). Samples collected with the net were filtered onto a 500 µm mesh. The material retained on that mesh was immediately sorted on ice according to the calanoida order. No distinction was made between individuals of different development stage and sex (copepodites, males, egg-producing females) while sorting our samples. We focused on copepods, in general, and calanoids, in particular, as they are amongst the most abundant zooplankters in Lac Saint-Pierre and play a key role in the trophic dynamics of this freshwater ecosystem (Langlois et al. 1992; Basu et al. 2000; Pinel-Alloul et al. 2011). From 5 to 10 subsamples of three copepods were placed into Eppendorf vials (acid washed and autoclaved) for RNA and DNA quantification. Subsamples of 50 copepods were collected and placed into acid-washed and autoclaved Eppendorf vials for FA analyses. Due to the variability in copepod abundance between sampling sites and the minimal biomass constraints for FA analyses, data for the quantification of copepod FA were only available at stations MA, DL, YA, AG, IP, Vam, FC and NI. Upon collection of the appropriate number of copepods for RNA, DNA and FA quantification, the Eppendorf vials were immediately frozen with liquid nitrogen and stored at −80°C until analysis.

Analyses

Dissolved inorganic nutrient concentrations ($NO_2 + NO_3$ and SRP, soluble reactive phosphorus) were determined following the American Public Health Association protocols (1998). Briefly, nitrate (NO_3) were first reduced into nitrites (NO_2) by cadmium, then nitrite concentrations (that was originally present plus reduced nitrate) were determined by the sulphanilamide colorimetric method. $NO_2 + NO_3$ concentrations at station NI were not determined due to analytical problems. SRP was analysed using the ascorbic acid-molybdate blue technique.

Lipid extractions and fatty acid analyses were performed on pools of 50 copepods and on duplicate filtered seston samples following the protocol described in Nordin et al. (2008). Briefly, zooplankton samples and GF/F filters were freeze dried and weighed prior to fatty acid (FA) analysis. Fatty acid methyl esters (FAME) were obtained by lipid extraction in chloroform:methanol (2:1 v/v), transesterification, and quantification using a capillary gas chromatograph (HP Agilent 6890N) coupled with a flame ionization detector. A 37-component FAME standard (Supelco no. 47885-U) was used to identify and quantify (4-point calibration curve) FAME in the samples by comparing their retention times to those of the FAME standard. An internal standard (5α-cholestane; Sigma-Aldrich; #C8003) was added to each sample prior to extraction to provide an estimate of extraction efficiency. Moreover, accurate amounts of unmethylated 20:2, 20:5n3 and 22:6n3 fatty acids (Sigma-Aldrich; #E3127, E2011 and D2534, respectively; ≥98% purity) were run through the same procedure as the sample extracts. A comparison between the known amounts of the aforementioned unmethylated FA added and the amounts computed (using the 4-point calibration curve) from the GC chromatograms gave an estimate of esterification efficiency close to 100% which was used in the computation of FA concentrations in the sample extracts. Results are reported as absolute (µg FAME/mg dry weight of tissue extracted) and relative (percent of total FA) concentrations of identified total fatty acids.

The ratio of ribonucleic acid (RNA) to deoxyribonucleic acid (DNA) of copepods was determined according to Kyle et al. (2003). Briefly, nucleic acids were extracted from replicated pools of 3 copepods homogenized in a solution of *N*-lauroylsarcosine (0.1% final concentration; Sigma-Aldrich®) in Tris buffer (10 mM Tris, 1 mM Na EDTA, pH 7.5; Molecular Probes®). Aliquots of the extract were distributed onto three 96-well microplates. Nuclease-free water was added to plate 1, whereas nucleases were added to plates 2 (RNase) and 3 (RNase + DNase). Nucleic acids were stained using the cyanine base fluorescent dye RiboGreenTM (Molecular Probes, Inc.), which binds non-specifically to nitrogenous bases of nucleic acids. RNA and DNA concentrations were determined from fluorescence readings (530 nm excitation, 590 nm emission filters) using a VarianTM Cary Eclipse Fluorescence spectrophotometer. Readings of plates 1 (DNA + RNA concentrations) and 2 (DNA concentration) were corrected for background

fluorescence (plate 3). RNA concentrations were then determined by subtracting the corrected fluorescence of plate 2 from that of plate 1.

Data analyses

Data analyses were performed using the [R] graphical and statistical computing environment (http://www.r-project.org). Data were checked for normality and, when needed, normalization of the original data was applied using the Box–Cox transformation technique. Briefly, this consists of a power transformation optimized in order to reduce anomalies such as non-additivity, non-normality and heteroscedasticity (for more information, see Sakia 1992). Results are presented as means ± standard deviation when applicable. A one-way analysis of variance (ANOVA) was used when three or more sets of data were compared and was followed by the Tukey's post hoc test when a significant difference was detected. Correlation matrixes (Pearson's product moment correlation) were calculated in order to relate the variability in copepod mean RNA:DNA ratio to abiotic (water depth, temperature, conductivity, $K_{d(UVA)}$, $K_{d(PAR)}$, dissolved inorganic nutrient concentrations and stoichiometry) and biotic (fatty acid composition of seston and zooplankton) variables. Data from station Vav were not used in the calculation of the correlation between copepod RNA:DNA ratio and the other abiotic and biotic variables, given that copepod and seston were not collected at the exact same location at that particular sampling site (Fig. 1). Linear correlation analyses were also used to investigate the relationships between seston FA composition and the underwater physical (water depth, temperature, conductivity, $K_{d(UVA)}$, $K_{d(PAR)}$) and chemical (dissolved inorganic nutrients concentration and stoichiometry) characteristics, as well as the trophic transfer of FA from seston to zooplankton.

Results

Sampling site location relative to Lac Saint-Pierre's water masses

Seven distinct water masses were identified within Lac Saint-Pierre of which six were sampled (Fig. 1; Table 1). On the north shore of Lac Saint-Pierre, stations DL, YA and AG were under the influence of the Du Loup and Yamachiche Rivers, while the MA station was located in the northwest tributaries' water mass. Station Vam was sampled within the St. Lawrence central channel draining waters originating from the Great Lakes. On the south shore, stations IP and Vav were under the influence of the Richelieu and Yamaska Rivers, while stations AF, FC and LP were located in waters influenced by the Saint-François River. Finally, station NI was under the influence of the Nicolet River.

Underwater light climate and nutrient concentrations

The variability in physical and chemical characteristics between the different sampling sites of Lac Saint-Pierre is reported in Table 1. Throughout the lake, summertime temperatures were fairly constant. $K_{d(PAR)}$ and $K_{d(UVA)}$ were the most variable environmental factors with coefficient of variation of 38 and 28%, respectively. Throughout Lac Saint-Pierre, $K_{d(UVA)}$ was positively correlated to $K_{d(PAR)}$ ($R^2 = 0.77$, $p < 0.01$, df = 9), and $K_{d(UVA)}$ values were on average two times higher than those of $K_{d(PAR)}$. The highest $K_{d(PAR)}$ and $K_{d(UVA)}$ values were generally measured in embayments close to river mouths (e.g., at stations MA, YA and AF).

The variation of the diffuse vertical attenuation coefficient ($K_{d(\lambda)}$) measured every 10 nm from 400 to 700 nm throughout Lac Saint-Pierre is presented in Fig. 2. The average $K_{d(\lambda)}$ decreased progressively from 3.8 m^{-1} at 400 nm to 1.3 m^{-1} at 550 nm and then remains fairly stable from 550 to 700 nm.

The concentrations of $NO_2 + NO_3$ and SRP ranged from 12.86 to 29.29 μM and from 0.10 to 0.58 μM, respectively. The concentration of SRP throughout Lac Saint-Pierre was about twice as variable as that of $NO_2 + NO_3$, with coefficients of variation of 51 and 23%, respectively. The highest $NO_2 + NO_3$ concentrations were measured at stations IP and AF, i.e., close to the mouth of Yamaska and Saint-Francois rivers.

Seston fatty acid composition

The absolute (mg FAME/μg dry weight) and relative (percent of total FA) concentrations of identified fatty acids (FA) in the seston are presented in Table 2. At all sampling sites, saturated fatty acids (SAFA) dominate the seston FA composition, contributing from 51 to 67% of total identified FA. Particularly, 16:0 and 14:0 were the most abundant FA in the seston at all sampling sites, contributing 27 and 13% of total identified FA (average values for all sampling sites), respectively. Mono- and polyunsaturated fatty acids (MUFA and PUFA, respectively) shared a similar contribution to total identified FA in the seston (average value for all sampling sites of 22 and 21%, respectively). Essential fatty acids (EFA) such as 18:3n3 (alpha-linoleic acid, ALA), 20:5n3 (eicosapentaenoic acid, EPA) and 22:6n3 (docosahexaenoic acid, DHA) contributed less than 13, 11 and 3%, respectively, of total identified FA in the seston (Table 2).

Table 1 Physical and chemical characteristics at the sampling sites in Lac Saint-Pierre

Station	Water mass	Depth (m)	Temp (°C)	Cond (mS cm^{-1})	$K_{d(PAR)}$ (m^{-1})	$K_{d(UVA)}$ (m^{-1})	$NO_2 + NO_3$ (μM)	SRP (μM)
MA (Maskinongé)	Nord-Ouest	1.62	22.40	0.22	1.67	3.69	18.57	0.39
DL (Du Loup)	Du Loup/Yamachiche	1.86	21.91	0.22	1.45	3.22	19.29	0.48
YA (Yamachiche)		2.00	21.93	0.22	2.81	4.69	20.00	0.39
AG (Aux Glaises)		1.88	21.85	0.21	2.02	3.57	15.71	0.58
Vam (Vers l'Amont)	Grands Lacs	2.94	21.75	0.28	1.34	2.13	16.43	0.10
IP (Île Plate)	Yamaska	4.48	21.73	0.28	0.99	1.55	23.57	0.26
Vav (Vers l'Aval)		1.94	21.25	0.28	0.88	2.46	12.86	0.13
AF (Anse du fort)	Saint-François	1.94	22.37	0.28	1.96	3.69	29.29	0.45
FC (Fer à cheval)		2.58	22.40	0.28	1.07	2.68	20.71	0.42
LP (Longue pointe)		1.80	22.41	0.27	0.98	2.86	18.57	0.23
NI (Nicolet)	Nicolet	1.48	21.30	0.28	1.59	3.04	NA	0.13

Temp temperature, *Cond* conductivity, $K_{d(PAR)}$ and $K_{d(UVA)}$ diffuse vertical attenuation coefficients integrated over the PAR (photosynthetically active radiation; 400–700 nm) and UVA (351–400 nm) spectra, respectively, $NO_2 + NO_3$ nitrite + nitrate concentrations, *SRP* soluble reactive phosphorus concentrations, *NA* not analysed

The concentrations (both absolute and relative) of identified fatty acids within the seston were neither related to $K_{d(UVA)}$ nor to $K_{d(PAR)}$ ($p > 0.05$) except for EPA, the absolute concentration of which was negatively correlated to $K_{d(PAR)}$ ($R^2 = 0.41$, $p < 0.05$, df = 9). Moreover, the absolute and relative concentrations of identified FA within the seston were neither correlated to other physical (water depth, temperature, conductivity) and chemical (dissolved inorganic nutrients concentration and stoichiometry) characteristics ($p > 0.05$). We also calculated the correlations between seston concentrations (both absolute and relative) in identified fatty acids and $K_{d(\lambda)}$ values at every 10 nm from 351 to 700 nm. Varying and significant negative correlations were found between $K_{d(\lambda)}$ within the PAR (400–700 nm) spectrum and seston absolute concentrations in EPA and DHA (Table 3). The coefficients of determination (R^2) of the significant correlations ($p < 0.05$) at a given wavelength are presented in Fig. 3 for both fatty acids. Throughout Lac Saint-Pierre, the seston absolute concentration in EPA was negatively correlated to $K_{d(\lambda)}$ from 410 to 700 nm ($p < 0.05$, df = 9), with the strongest correlations (i.e., highest R^2 values) at 430, 460 and 690 nm and from 520 to 590 nm (Fig. 3). The seston absolute concentration in DHA was negatively correlated to $K_{d(\lambda)}$ from 490 to 700 nm ($p < 0.05$, df = 9), with R^2 values progressively increasing from a minimum value of 0.36 at 490 nm to a maximum of 0.48 at 690 nm.

Copepod fatty acid composition

The absolute and relative concentrations of identified fatty acids (FA) in copepods are presented in Table 4. At all sampling sites, the most abundant FA in copepods were 16:0, EPA and DHA, which contributed 24, 13 and 11% of total identified FA (average values for all sampling sites), respectively. The absolute concentrations of 16:1n7 (palmitoleic acid, PAL), ALA and EPA in copepods were significantly correlated to those in the seston ($R^2 = 0.99$, 0.72, and 0.78, respectively, $p < 0.01$, df = 6; Fig. 4). The relative concentrations of identified fatty acids in copepods were not significantly correlated to those in the seston ($p > 0.05$).

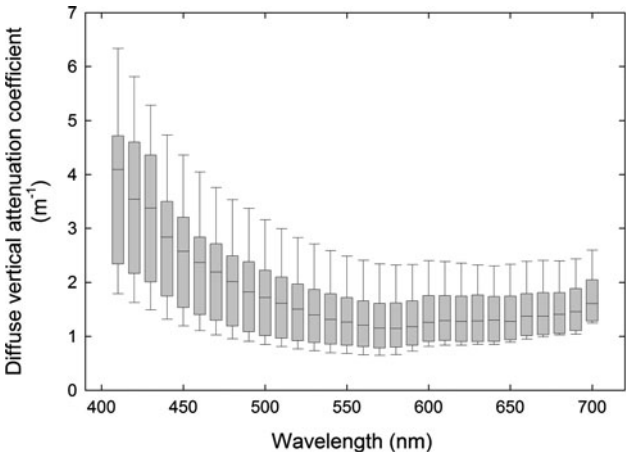

Fig. 2 Boxplots of the diffuse vertical attenuation coefficient at every 10 nm from 400 to 700 nm throughout Lac Saint-Pierre. The *vertical bar* within the *box* is the median ($n = 11$)

Copepod RNA:DNA ratio

The spatial variability in copepod RNA:DNA ratio is presented in Fig. 5. Throughout Lac Saint-Pierre, copepod RNA:DNA ranged from 7.32 (St. Vav) to 21.34 (St. Vam) with an average value (±SD) of 11.9 ± 4.2. At all sampling sites, the average RNA:DNA value was associated

Table 2 Absolute (mean values ± SD when applicable, in μg FAME/mg dry weight) and relative (percent values; in brackets) concentrations of identified fatty acids in the seston at various locations in Lac Saint-Pierre

Station	SAFA				MUFA		
	14:0	15:0	16:0	18:0	16:1n7 (PAL)	18:1n7	18:1n9 (OLE)
MA	0.56 ± 0.25 (10)	0.41 ± 0.23 (6)	1.19 ± 0.52 (21)	0.70 ± 0.37 (11)	0.22 ± 0.05 (6)	0.12 ± 0.06 (3)	0.40 ± 0.16 (8)
DL	0.34 (9)	0.03 (1)	1.29 (34)	0.21 (6)	0.37 (10)	0.18 (5)	0.34 (9)
YA	0.85 ± 0.54 (15)	0.43 ± 0.25 (6)	1.45 ± 0.75 (26)	0.85 ± 0.32 (14)	0.30 ± 0.02 (6)	0.13 ± 0.04 (3)	0.44 ± 0.27 (8)
AG	0.51 ± 0.23 (15)	0.38 ± 0.25 (7)	0.93 ± 0.43 (25)	0.60 ± 0.32 (14)	0.12 ± 0.03 (5)	0.07 ± 0.04 (3)	0.24 ± 0.11 (6)
Vam	0.60 ± 0.49 (9)	0.08 ± 0.04 (1)	1.94 ± 0.01 (31)	0.38 ± 0.14 (6)	0.60 ± 0.03 (10)	0.22 ± 0.01 (3)	0.61 ± 0.02 (10)
IP	0.33 ± 0.18 (13)	0.03 ± 0.01 (1)	0.59 ± 0.01 (24)	0.14 ± 0.01 (6)	0.27 ± 0.02 (11)	0.10 ± 0.02 (4)	0.17 ± 0.02 (7)
Vav	0.48 ± 0.27 (12)	0.09 ± 0.04 (2)	0.98 ± 0.01 (27)	0.43 ± 0.03 (12)	0.29 ± 0.02 (8)	0.12 ± 0.03 (3)	0.26 ± 0.07 (7)
AF	0.56 ± 0.24 (23)	0.06 ± 0.01 (3)	0.48 ± 0.02 (20)	0.10 ± 0.01 (4)	0.26 ± 0.05 (11)	0.07 ± 0.03 (3)	0.14 ± 0.01 (6)
FC	1.03 ± 0.08 (10)	0.10 ± 0.01 (1)	3.38 ± 0.25 (33)	0.36 ± 0.04 (3)	0.96 ± 0.09 (9)	0.40 ± 0.05 (4)	1.15 ± 0.09 (11)
LP	1.78 ± 0.14 (14)	0.12 ± 0.01 (1)	4.08 ± 0.08 (32)	0.42 ± 0.02 (3)	1.48 ± 0.08 (12)	0.40 ± 0.05 (3)	1.25 ± 0.02 (10)
NI	0.60 (13)	0.12 (2)	1.08 (23)	0.89 (19)	0.29 (6)	0.10 (2)	0.69 (15)

Station	PUFA				∑n3	∑n6	∑SAFA	∑MUFA	∑PUFA
	18:2n6 (LIN)	18:3n3 (ALA)	20:5n3 (EPA)	22:6n3 (DHA)					
MA	0.15 ± 0.02 (4)	0.21 ± 0.01 (6)	0.16 ± 0.01 (5)	0.04 ± 0.02 (1)	0.54 ± 0.12 (15)	0.22 ± 0.10 (5)	3.73 ± 1.96 (60)	0.94 ± 0.32 (20)	0.80 ± 0.27 (20)
DL	0.17 (4)	0.48 (13)	0.21 (6)	0.05 (1)	0.75 (20)	0.17 (4)	1.96 (51)	0.94 (25)	0.92 (24)
YA	0.15 ± 0.03 (3)	0.34 ± 0.01 (7)	0.20 ± 0.03 (4)	0.07 ± 0.03 (1)	0.76 ± 0.28 (14)	0.17 ± 0.06 (3)	2.79 ± 1.34 (64)	1.00 ± 0.52 (18)	0.94 ± 0.36 (18)
AG	0.06 ± 0.02 (2)	0.10 ± 0.01 (6)	0.08 ± 0.01 (5)	0.03 ± 0.01 (1)	0.33 ± 0.16 (13)	0.09 ± 0.04 (3)	2.60 ± 1.33 (67)	0.57 ± 0.25 (17)	0.42 ± 0.24 (16)
Vam	0.31 ± 0.01 (5)	0.71 ± 0.01 (11)	0.35 ± 0.01 (6)	0.09 ± 0.01 (1)	1.15 ± 0.01 (19)	0.31 ± 0.01 (5)	3.32 ± 0.85 (53)	1.43 ± 0.02 (23)	1.45 ± 0.01 (24)
IP	0.10 ± 0.00 (4)	0.22 ± 0.01 (9)	0.27 ± 0.01 (11)	0.05 ± 0.01 (2)	0.54 ± 0.01 (22)	0.10 ± 0.01 (4)	1.28 ± 0.19 (52)	0.54 ± 0.02 (22)	0.63 ± 0.01 (26)
Vav	0.14 ± 0.05 (4)	0.29 ± 0.03 (8)	0.31 ± 0.03 (8)	0.10 ± 0.01 (3)	0.70 ± 0.07 (19)	0.15 ± 0.03 (4)	2.18 ± 0.40 (59)	0.67 ± 0.11 (18)	0.85 ± 0.10 (23)
AF	0.09 ± 0.01 (4)	0.17 ± 0.01 (7)	0.24 ± 0.01 (10)	0.04 ± 0.01 (1)	0.45 ± 0.01 (19)	0.10 ± 0.01 (4)	1.39 ± 0.24 (58)	0.47 ± 0.08 (19)	0.55 ± 0.01 (23)
FC	0.38 ± 0.01 (4)	1.30 ± 0.07 (13)	0.50 ± 0.03 (5)	0.18 ± 0.02 (2)	1.98 ± 0.11 (19)	0.40 ± 0.03 (4)	5.33 ± 0.47 (52)	2.52 ± 0.15 (25)	2.38 ± 0.14 (23)
LP	0.47 ± 0.01 (4)	1.24 ± 0.01 (10)	0.65 ± 0.04 (5)	0.19 ± 0.02 (1)	2.08 ± 0.06 (17)	0.50 ± 0.05 (4)	6.85 ± 0.03 (54)	3.12 ± 0.12 (25)	2.58 ± 0.11 (21)
NI	0.07 (2)	0.17 (4)	0.12 (2)	0.06 (1)	0.39 (8)	0.09 (2)	2.89 (61)	1.33 (28)	0.48 (21)

PAL palmitoleic acid, *OLE* oleic acid, *LIN* linoleic acid, *ALA* alpha-linolenic acid, *EPA* eicosapentaenoic acid, *DHA* docosahexaenoic acid, *SAFA* saturated fatty acids, *MUFA* monounsaturated fatty acids, *PUFA* polyunsaturated fatty acids, *n3* omega-3 fatty acids, *n6* omega-6 fatty acids

Table 3 Pearson's product moment correlation coefficient (r) and associated probability (p) of the correlation between the diffuse vertical attenuation coefficient at specific wavelengths (λ) from 400 to 700 nm (i.e., within the PAR spectrum) and the seston absolute concentrations of 20:5n3 (eicosapentaenoic acid, *EPA*) and 22:6n3 (docosahexaenoic acid, *DHA*)

λ (nm)	20:5n3 (EPA)		22:6n3 (DHA)	
	r	p	r	p
400	−0.56	0.072*	−0.46	0.150
410	−0.63	0.038*	−0.52	0.104
420	−0.68	0.021*	−0.54	0.089
430	−0.69	0.018*	−0.55	0.079
440	−0.67	0.024*	−0.56	0.075
450	−0.67	0.025*	−0.57	0.066
460	−0.69	0.020*	−0.58	0.060
470	−0.68	0.023*	−0.58	0.060
480	−0.68	0.022*	−0.59	0.057
490	−0.68	0.021*	−0.60	0.049*
500	−0.68	0.021*	−0.61	0.046*
510	−0.68	0.022*	−0.61	0.047*
520	−0.69	0.020*	−0.62	0.041*
530	−0.69	0.019*	−0.63	0.039*
540	−0.69	0.019*	−0.63	0.037*
550	−0.69	0.020*	−0.64	0.035*
560	−0.69	0.019*	−0.64	0.033*
570	−0.69	0.019*	−0.65	0.031*
580	−0.69	0.020*	−0.65	0.030*
590	−0.69	0.020*	−0.66	0.028*
600	−0.68	0.021*	−0.66	0.026*
610	−0.68	0.021*	−0.66	0.026*
620	−0.68	0.021*	−0.67	0.025*
630	−0.67	0.023*	−0.67	0.023*
640	−0.67	0.024*	−0.67	0.025*
650	−0.66	0.026*	−0.67	0.023*
660	−0.65	0.029*	−0.67	0.025*
670	−0.65	0.031*	−0.67	0.024*
680	−0.65	0.032*	−0.66	0.028*
690	−0.69	0.018*	−0.69	0.020*
700	−0.66	0.026*	−0.68	0.022*

* Significant correlation at $p < 0.05$

with a large variability, with coefficients of variation ranging from 43 to 98% emphasizing the large spectrum of variation in sampled habitats. No significant differences in the average copepod RNA:DNA ratio were found between sampling sites (one-way ANOVA; $F_{10,94} = 1.83$, $p = 0.24$).

The RNA:DNA ratio of copepods was significantly correlated to the absolute concentration of PAL, ALA, EPA and DHA in the seston ($R^2 = 0.82$, 0.84, 0.74 and 0.76, respectively, $p < 0.01$, df = 8; Fig. 6). Significant correlations were also found between the copepod RNA:DNA ratio and the absolute concentration of PAL, ALA and EPA in copepods ($R^2 = 0.81$, 0.72, and 0.76, respectively, $p < 0.01$, df = 6). The RNA:DNA ratio of copepods was neither correlated to physical (water depth, temperature, conductivity, $K_{d(UVA)}$, $K_{d(PAR)}$) and chemical (dissolved inorganic nutrients concentration and stoichiometry) characteristics nor to the relative concentration of identified FA in seston and copepods ($p > 0.05$).

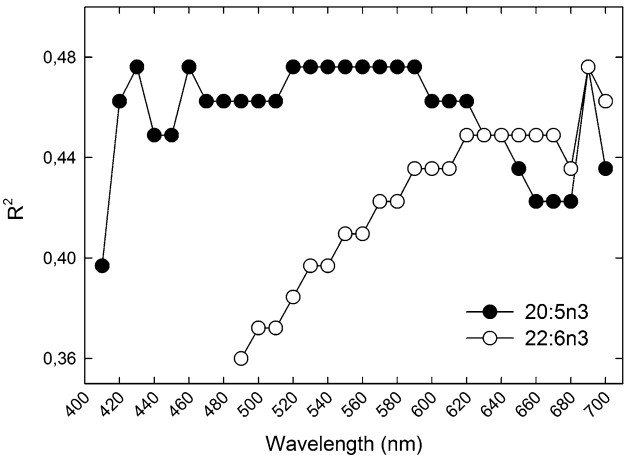

Fig. 3 Coefficient of determination (R^2) of the correlation between seston absolute concentrations in 20:5n3 (*black circle*) or 22:6n3 (*white circle*) fatty acids and the diffuse vertical attenuation coefficient of photosynthetically active radiations calculated every 10 nm from 400 to 700 nm. Each *circle* indicates a significant correlation ($p < 0.05$, df = 9) at a given radiation wavelength

Discussion

Underwater light climate and seston FA composition

The underwater light climate throughout Lac Saint-Pierre displayed strong spatial heterogeneity given both lateral and longitudinal variability of $K_{d(UVA)}$ and $K_{d(PAR)}$ (Fig. 1; Table 1), as previously described by Frenette et al. (2006). Huggins et al. (2004) reported that the lateral variability in the underwater light climate within Lac Saint-Pierre results in greater exposure to UV and PAR in the southern (sampling site close to station FC) than in the northern (sampling site close to station DL) water masses, which in turn influenced the nutritional quality of biofilms. On the south shore, biofilms had greater proportion of low nutritional SAFA while biofilms on the north shore had greater proportions of EPA and DHA (Huggins et al. 2004). Accordingly, we measured lower $K_{d(UVA)}$ and $K_{d(PAR)}$ values at station FC than DL (Table 1), which points towards a greater exposure to UVA and PAR in the

Table 4 Absolute (in µg FAME/mg dry weight) and relative (percent values; in brackets) concentrations of identified fatty acids in the copepods at various locations in Lac Saint-Pierre

Station	SAFA				MUFA			PUFA				∑n3	∑n6	∑SAFA	∑MUFA	∑PUFA
	14:0	15:0	16:0	18:0	16:1n7 (PAL)	18:1n7	18:1n9 (OLE)	18:2n6 (LIN)	18:3n3 (ALA)	20:5n3 (EPA)	22:6n3 (DHA)					
MA	0.87 (2)	0.45 (1)	10.19 (18)	7.47 (13)	2.67 (5)	2.27 (4)	1.48 (3)	1.31 (2)	2.92 (5)	10.72 (19)	10.59 (19)	25.35 (44)	3.10 (5)	20.83 (36)	7.44 (13)	28.80 (50)
DL	21.98 (13)	9.14 (5)	49.50 (29)	9.27 (5)	3.99 (2)	3.45 (2)	8.03 (5)	2.28 (1)	3.32 (2)	10.56 (6)	8.25 (5)	31.29 (19)	4.59 (3)	105.91 (63)	26.71 (16)	36.34 (22)
YA	1.16 (2)	0.39 (1)	11.49 (23)	3.40 (7)	3.67 (7)	2.09 (4)	2.38 (5)	1.11 (2)	3.30 (7)	9.60 (19)	7.02 (14)	20.92 (41)	2.38 (5)	18.05 (36)	8.94 (18)	23.50 (47)
AG	7.41 (12)	2.89 (5)	13.96 (22)	8.19 (13)	1.76 (3)	1.58 (3)	2.16 (3)	1.49 (2)	2.70 (4)	6.20 (10)	4.83 (8)	14.60 (23)	2.52 (4)	38.47 (62)	6.74 (11)	17.12 (27)
Vam	1.60 (2)	0.58 (1)	18.40 (22)	3.96 (5)	6.87 (8)	7.07 (9)	4.97 (6)	3.09 (4)	8.16 (10)	13.29 (11)	9.50 (8)	30.94 (37)	6.63 (8)	26.11 (32)	19.12 (23)	37.59 (45)
IP	1.14 (2)	0.47 (1)	12.15 (2)	3.92 (7)	3.83 (7)	3.32 (6)	3.03 (5)	1.48 (3)	3.52 (6)	10.61 (19)	8.52 (15)	23.36 (42)	2.46 (4)	18.94 (34)	10.79 (19)	26.09 (47)
Vav	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
AF	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
FC	2.46 (2)	0.62 (1)	24.32 (24)	6.26 (6)	10.13 (10)	3.42 (3)	4.33 (4)	2.77 (3)	7.93 (3)	16.47 (16)	13.59 (14)	39.33 (39)	4.52 (5)	36.15 (36)	19.87 (20)	44.30 (44)
LP	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND	ND
NI	45.65 (19)	10.30 (4)	75.91 (31)	27.61 (11)	3.73 (2)	5.32 (2)	13.82 (6)	3.15 (1)	2.27 (1)	4.80 (2)	6.49 (3)	19.89 (8)	6.16 (3)	175.96 (71)	44.19 (18)	26.16 (11)

PAL palmitoleic acid, *OLE* oleic acid, *LIN* linoleic acid, *ALA* alpha-linolenic acid, *EPA* eicosapentaenoic acid, *DHA* docosahexaenoic acid, *SAFA* saturated fatty acids, *MUFA* monounsaturated fatty acids, *PUFA* polyunsaturated fatty acids, *n3* omega-3 fatty acids, *n6* omega-6 fatty acids, *ND* no data

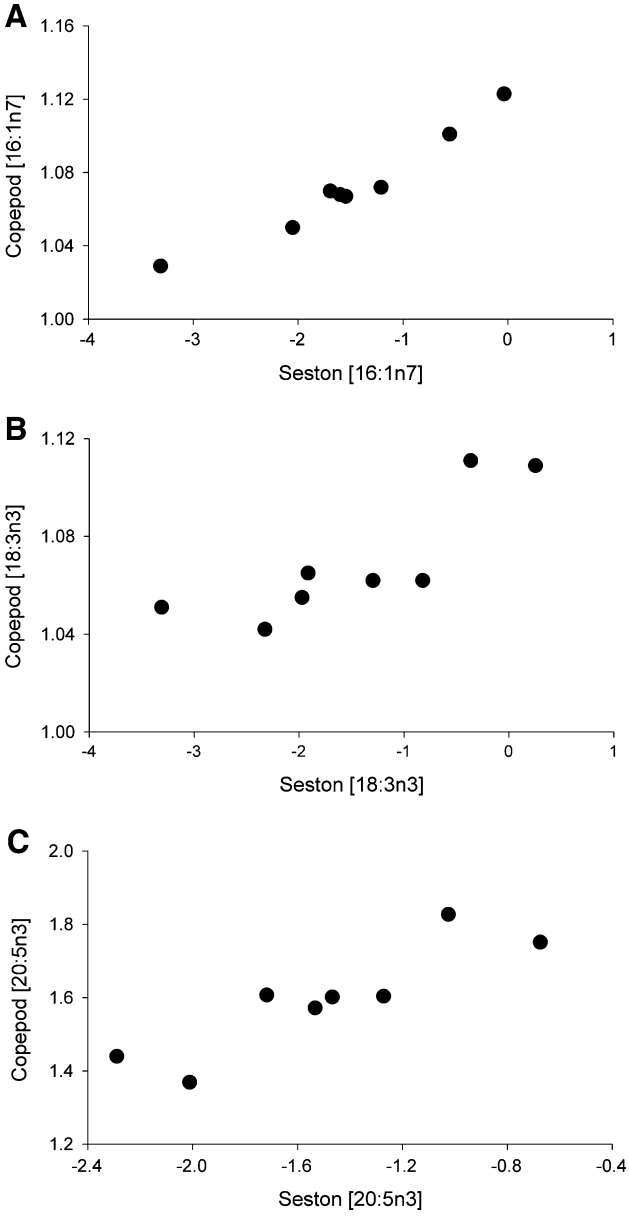

Fig. 4 Relationship between the concentrations (µg/mg dry weight) of **a** palmitoleic acid ([16:1n7]), **b** linolenic acid ([18:3n3]) and **c** eicosapentaenoic acid ([20:5n3]) in seston and copepods. Data are transformed values (see "Materials and methods" for details)

southern than in the northern water masses. In contrast to Huggins et al. (2004), the different light climate at stations DL and FC did not translate into varying nutritional quality of seston which had similar relative FA concentrations (Table 2). This exemplifies that static autotrophic organisms such as periphyton experience a drastically different light history than free-moving phytoplankton in the water column, which can impact the fatty acid composition accordingly.

So far, most studies that focused on the relationship between PAR and the FA composition of primary

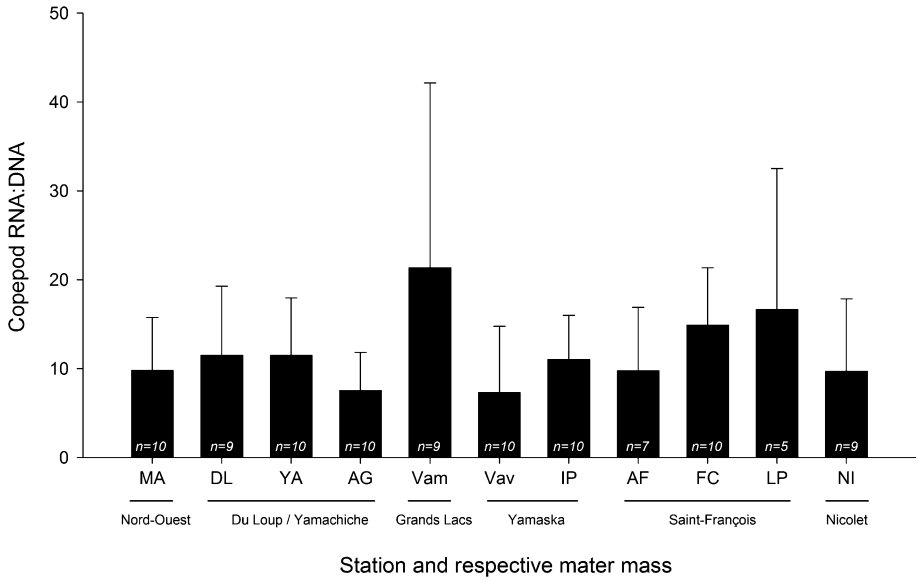

Fig. 5 Spatial variability in copepod RNA:DNA ratio (mean ± SD) within Lac Saint-Pierre. Stations are grouped according to the respective water mass in which they were sampled. The value within each *bar* is the number (n) of replicates of three copepods used in the computation of the mean RNA/DNA ratio

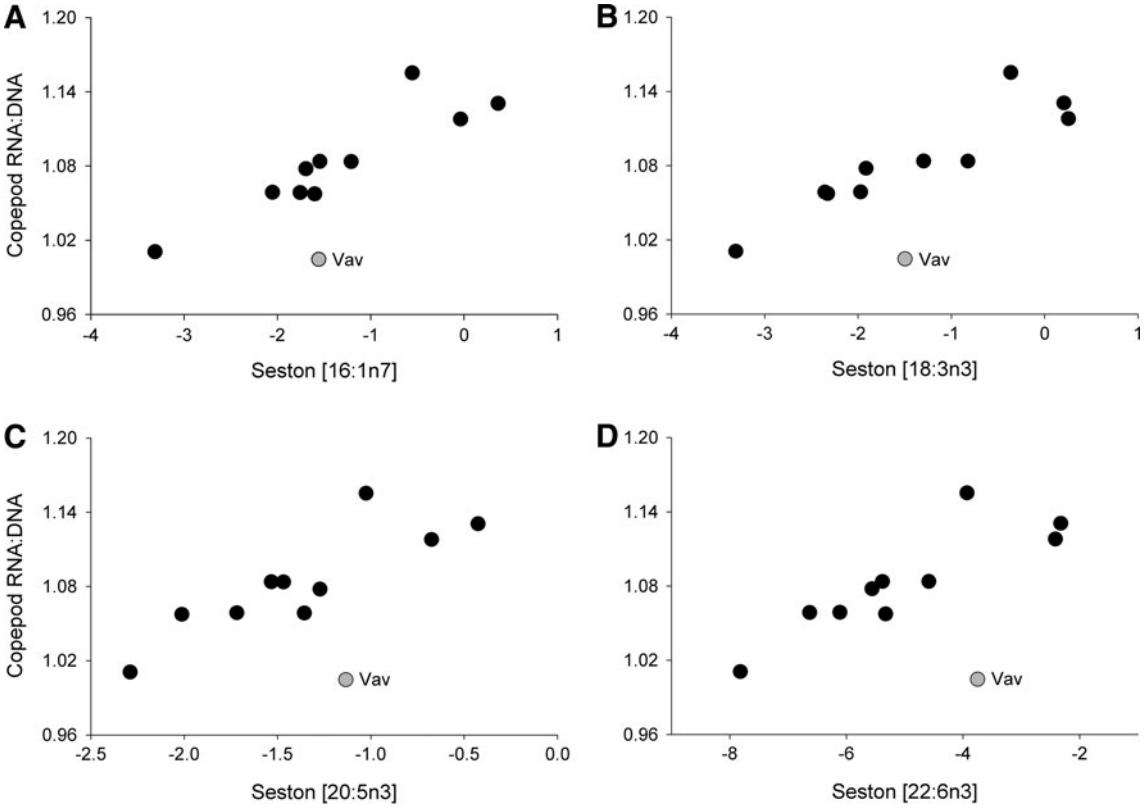

Fig. 6 Relationship between the RNA:DNA ratio of copepods and the concentration (μg/mg dry weight) of **a** palmitoleic acid ([16:1n7]), **b** linolenic acid ([18:3n3]), **c** eicosapentaenoic acid ([20:5n3]) and **d** docosahexaenoic acid ([22:6n3]) in the seston. Data are transformed values (see "Materials and methods" for details). Station Vav was differentiated from the other sampling site by a *grey circle* and was not used in the calculation of the regression between copepod RNA:DNA ratio and seston fatty acid concentration (see "Materials and methods" for details)

producers have considered the effect of light intensity (Mortensen et al. 1988; Thompson et al. 1990; Wainman et al. 1999) without considering the potential influence of specific wavelengths. Rather than involving the whole PAR spectrum, our results (Fig. 3; Table 3) suggest that the effects of light on the concentrations of some essential PUFA within the seston may occur at a finer scale. Indeed, the concentration of EPA in the seston was related to light

availability over almost the entire PAR spectrum (i.e., from 410 to 700 nm), while DHA responded to light availability over a narrower range of wavelengths (i.e., from 490 to 700 nm) (Fig. 3). Interestingly, the absolute concentration of DHA in the seston showed the greatest response to light availability from 620 to 700 nm (Fig. 3), a range of wavelengths close to the absorption maxima of chlorophyll *a* and *b* at 660 and 645 nm, respectively, at which high photosynthesis rates are known to occur (Kirk 1994). Over that range of wavelengths (i.e., 620–700 nm), the diffuse vertical attenuation coefficient ($K_{d(\lambda)}$) values were relatively low (Fig. 2) indicating that light penetrated deeper in the lake with potentially greater impact on aquatic organisms and photosynthesis. Similarly, the absolute concentration of EPA in the seston showed relatively high responses to light availability at 690 nm (close to the absorption maxima of chlorophyll *a* at 660 nm) and at 430 and 460 nm. The two latter wavelengths are close to the absorption maxima of chlorophyll *a* and *b* at 430 and 450 nm, respectively, also associated with optimal photosynthesis rates (Kirk 1994). These results suggest a potential biochemical link between photosynthesis and the phytoplankton content in essential fatty acids such as EPA and DHA in Lac Saint-Pierre. In support of this idea, Barsanti et al. (2000) reported that the synthesis of PUFA is correlated to the activity of the photosynthetic apparatus in the flagellate *Euglena gracilis*. Similarly, Harwood (1998) reported that qualitative changes in lipids, as a result of various light conditions, are associated with alterations in chloroplast development. It has also been suggested that some fatty acids may associate with pigments to accommodate the phytol side chain of chlorophyll *a* (Thompson et al. 1990 and references therein).

Previous studies have reported a close correspondence between the absorption spectra of phototrophic microrganisms and the prevailing underwater light spectrum (Kirk 1994; Stomp et al. 2007), which may play a key role in structuring phytoplankton communities (Rocap et al. 2003). Frenette et al. (2012) demonstrated that the distribution of pico and nano-phototrophic prokaryotes and eukaryotes is related to the availability of the green and red wavelenghts of the underwater light spectrum in the St. Lawrence river (including Lac Saint-Pierre). Since the algal fatty acid composition varies among phytoplankton species or functional groups (Brett et al. 2009 and references therein), the underwater light climate may influence the seston fatty acid composition by modulating the phytoplankton community structure. Our study did not focus on the taxonomic composition of sestonic primary producers, but rather uses fatty acids as markers of the trophic transfer efficiency between producers and consumers. Still, our results based on seston fatty acids could be indicative of the presence of specific taxonomic groups. We compared the mean percent fatty acid composition of seston from Lac Saint-Pierre (A) with that compiled by Brett et al. (2009) for freshwater phytoplankton (B). By computing the ratio A/B as a proxy for such a comparison (Table 5), we found that seston from Lac Saint-Pierre sampled during our study most closely match the FA composition of cryophytes, diatoms and cyanophytes, with average A/B ratio of 0.8, 1.2 and 2.7, respectively. Most studies about the fatty acid content of phytoplankton and the trophic transfer of fatty acids throughout food webs have been based on laboratory experiments conducted with monoalgal cultures (Reuss and Poulsen 2002). Caution should therefore be taken when comparing our field data with monospecific laboratory values considering that microalgal diversity within our seston samples may have buffered the FA signatures of specific phytoplankton groups. Yet, our data are coherent with Langlois et al. (1992) who reported that the summertime phytoplankton community in Lac Saint-Pierre is dominated by cyanophytes with some occurrence of diatoms. Hudon et al. (1996) also reported the dominance of cyanophytes and cryptophytes within the summertime phytoplankton population on the north shore of Lac Saint-Pierre.

Copepod RNA:DNA ratio

The RNA:DNA ratio has proven to be a useful tool for the determination of the short-term growth of various aquatic organisms (Chícharo and Chícharo 2008). On an individual basis, DNA per somatic cell is assumed to be constant in sexually mature adults so that the RNA:DNA ratio can be related to the magnitude of RNA transcription, protein synthesis and hence growth condition. Each RNA:DNA value reported in our study is relative to a replicated pool of three individuals and consequently represents an average value for the copepod population sampled at a given location. Moreover, the RNA:DNA ratio of copepods was significantly correlated to their RNA concentration ($R^2 = 0.88$, $p < 0.01$, $df = 8$) but not to that of DNA ($p > 0.05$), which suggests that the spatial variability in RNA:DNA ratio throughout Lac Saint-Pierre was likely related to variable growth conditions of the copepod population. So far, measurements of RNA:DNA ratios in freshwater zooplankton have mostly been reported for cladocerans (Gorokhova and Kyle 2002; Vrede et al. 2002; Persson 2007). There are very few data reporting RNA:DNA ratios in freshwater copepods (Ventura 2006) despite their importance as one of the most abundant zooplankton class and a major prey item for fish in aquatic ecosystems (Langlois et al. 1992). The average RNA:DNA ratio of 11.9 ± 4.2 reported in our study is ~ 8 times higher than the values of 1.4 reported for freshwater calanoid copepods (Ventura 2006). Yet, our RNA:DNA

Table 5 Comparison between the mean fatty acid composition (expressed as a percent of total FA) of seston from Lac Saint-Pierre (A) and that determined by Brett et al. (2009) for freshwater phytoplankton functional groups (B)

	This study	Chlorophytes		Cryptophytes		Diatoms		Cyanophytes	
	A	B	A/B	B	A/B	B	A/B	B	A/B
ΣSAFA	57.4	32.5	1.8	28.4	2.0	23.8	2.4	58.6	1.0
ΣMUFA	21.9	27.3	0.8	9.9	2.2	40.3	0.5	24.8	0.9
ΣC_{16} PUFA	0.002	0.001	1.5	0.1	0.02	9.1	0.0002	0.001	1.5
18:2n6 (LIN)	3.6	14.4	0.2	3.3	1.1	2.0	1.8	7.2	0.5
18:3n3 (ALA)	8.5	25.5	0.3	39.7	0.2	2.9	2.9	7.0	1.2
20:5n3 (EPA)	6.1	0.1	61.0	15.1	0.4	16.9	0.4	0.6	10.1
22:6n3 (DHA)	1.6	0.001	1,600.0	2.9	0.5	2.5	0.6	0.7	2.2
Σn3/Σn6	4.3	1.9	2.3	16.8	0.3	7.6	0.6	1.0	4.3
Average A/B ratio			204		0.8		1.2		2.7

SAFA saturated fatty acids, *MUFA* monounsaturated fatty acids, *PUFA* polyunsaturated fatty acids, *LIN* linoleic acid, *ALA* alpha-linolenic acid, *EPA* eicosapentaenoic acid, *DHA* docosahexaenoic acid, *n3* omega-3 fatty acids, *n6* omega-6 fatty acids

average value is in good agreement with previous study reporting RNA:DNA ratio for marine copepod under optimal nutritional and growth conditions. In laboratory experiments, Speekman et al. (2007) reported RNA:DNA ratio for *Acartia tonsa* of up to 14 when fed on the EFAs-rich *Thalassiosira* sp. Similarly, RNA:DNA values of up to 8 were reported for *Calanus finmarchicus* under excess-food conditions (Wagner et al. 2001).

Due to time constraints aiming to prevent the degradation of nucleic and fatty acids upon collection of our zooplankton samples, a basic screening and sorting of the samples was performed so that we were not able to determine precisely which calanoid copepod species were collected for biochemical analyses. Based on the essential fatty acid composition of various groups of freshwater copepods, Smyntek et al. (2008) demonstrated that calanoids contain nearly equal amounts of EPA and DHA while cyclopoids accumulate over twice as more DHA than EPA. Comparatively, throughout Lac Saint-Pierre, copepods contained about the same amount of EPA than DHA (Table 4), with EPA:DHA ratios of 0.7 to 1.4. These results are coherent with our preferential selection of calanoids during sorting. Moreover, Pinel-Alloul et al. (2011) reported that the copepod population, sampled in August 2006 within the fluvial section zone of the St. Lawrence River (including Lac Saint-Pierre), was mostly represented by calanoids (48% of total copepod abundance), with cyclopoids and harpaticoids contributing 39 and 13% of total copepod abundance, respectively. Hence, our a priori selection of calanoids during sorting may be considered as representative of a significant fraction of the summertime zooplankton community of Lac Saint-Pierre. Since no precise distinction was made between individuals of different development stages during zooplankton sorting, the spatial variability in RNA/DNA ratio in Lac Saint-Pierre reported in our study is potentially biased considering that different copepod stages differ in their growth pattern. Yet, Pinel-Alloul et al. (2011) reported that unidentified copepodites dominate (94% of total abundance) the summertime calanoid population of Lac Saint-Pierre. Therefore, copepodites may have shared a significant contribution to the total calanoid copepod population sampled in summertime during our study so that the variability in RNA/DNA values (Fig. 5) may be associated with various growth conditions of this dominant copepodite fraction.

Relationship between copepod growth condition and seston nutritional quality

Previous studies have related copepod growth to temperature (Katona 1970; Lee et al. 2003) as well as food quantity and quality (Koski et al. 1998; Andersen et al. 2007). Temperature is recognized as the dominant factor influencing copepod growth under adequate food supplies; conversely, when the temperature range is narrow, food becomes the predominant determining factor for growth (Chícharo and Chícharo 2008 and references therein). Accordingly, throughout Lac Saint-Pierre, temperature was fairly constant (Table 1) and the copepod RNA:DNA ratio was significantly correlated to the seston absolute concentration in PAL, ALA, EPA and DHA (Fig. 6). Our results are also consistent with those of Pommier et al. (2010) who reported that the growth condition of *Eurytemora affinis* within the St. Lawrence Estuarine Transition Zone, downstream of Lac Saint-Pierre, is associated with the relative abundance of EPA-rich seston advected from upstream waters. Fatty acids such as PAL and EPA are potential biomarkers for diatoms which are recognized as highly valuable food items for herbivorous primary consumers

(Dalsgaard et al. 2003; Brett et al. 2009). Moreover, zooplankton can elongate dietary ALA into EPA (Brett and Müller-Navarra 1997) which, along with DHA, is a key essential fatty acid in aquatic ecosystems (Brett and Müller-Navarra 1997; Müller-Navarra et al. 2000; Kainz et al. 2004).

The significant correlation between copepod RNA:DNA ratio and the seston absolute concentrations in PAL, ALA, EPA and DHA (Fig. 6) suggest that in systems with organic matter from a variety of sources, nutritional factors associated with phytoplankton can have a dominant influence on copepod growth, as previously reported for cladocerans (Müller-Solger et al. 2002). Moreover, the significant relationships between PAL, ALA and EPA absolute concentrations in the seston and those in copepods (Fig. 4) point towards the trophic transfer of these fatty acids from primary producers to herbivorous consumers. Our results are consistent with those of Müller-Navarra et al. (2000), who report that EPA is a good predictor for carbon transfer between planktonic primary producers and consumers in freshwater ecosystems. More recently, Kainz et al. (2010) demonstrated that dietary access to EPA and DHA improve the somatic growth potential (RNA:DNA ratio) of the benthic amphipod *Diporeia*. Our results also support those of Brett and Brett and Müller-Navarra (1997) who report that EFA are key nutritional constituents of zooplankton diets, determining the energetic efficiency across the plant–animal interface, secondary production and strength of trophic coupling in aquatic pelagic food webs.

The relationship between copepod RNA:DNA ratio and the seston concentrations in essential fatty acids (such as EPA and DHA) suggest that FA, considered as trophic markers in aquatic food webs (Dalsgaard et al. 2003), may also be viewed as limiting factor for zooplankton growth. Indeed, some PUFA, required for biochemical and physiological processes, are considered essential fatty acids as they cannot be synthesized by the consumer itself and should therefore be provided through its diet. However, from a conceptual point of view, Müller-Navarra (2006) reported that the suspected limiting character of certain PUFA could contradict their use as trophic markers. Indeed, according to Liebig's Law of the minimum (Liebig 1940), if seston PUFA are to limit zooplankton growth, these fatty acids within copepod should remain constant to preserve homeostasis of the consumer. Contrastingly, our results revealed that the FA in the seston whose concentrations are related to copepod growth (PAL, ALA and EPA) also vary concomitantly in copepods. Yet, Liebig's Law of the minimum only applies at steady state, a condition which was likely not met during our study as it just represents a "snapshot" in the annual productivity cycle of Lac Saint-Pierre. Moreover, essential PUFA are usually not directly converted into growth but rather accumulated and stored in the neutral lipid fraction, from which recent trophic history can be inferred (Dalsgaard et al. 2003; Müller-Navarra 2008). This may explain the concomitant variability in the concentrations of some PUFA in seston and copepods observed on a short timescale during our field campaign. The theoretical contradiction in the use of fatty acids as both a trophic marker and a limiting factor for growth (Müller-Navarra 2006) may hold true on a longer time scale than that encountered during our summertime sampling of Lac Saint-Pierre.

Light as controlling factor of food web productivity

The influence of light on primary producers nutritional quality have been restricted, so far, to the UV and PAR components of the light spectrum. It now appears that a better understanding of food web processes may be obtained by studying PAR at a finer scale through the influence of specific wavelengths on the biochemical composition of photosynthetic organisms. The spatial heterogeneity (both lateral and longitudinal) of the underwater light climate throughout Lac Saint-Pierre is therefore likely to influence food web productivity by setting the initial conditions for primary producers and consumers abundance which could further be modulated by biotic interactions. However, light contributed only 48%, at the most, of the variability in seston concentrations in EPA and DHA throughout Lac Saint-Pierre (Fig. 3) and we cannot preclude that other abiotic factors may have covaried with light in their potential influence on seston nutritional quality. For instance, dissolved inorganic nutrients have been reported to impact the taxonomic, and hence the FA composition, of the seston (Mayzaud et al. 1990). Yet, neither the concentrations of dissolved inorganic nutrients, nor their stoichiometry, explained the variability in seston FA composition in our study. Coherently, Reynolds and Descy (1996) reported that in large river systems, which are usually nutrient-replete, primary production is mainly controlled by physical factors, such as light, that are in turn linked to hydrology. Temperature and conductivity have also been reported to influence the fatty acid content of phytoplankton (Guschina and Harwood 2009) but these two variables were not related to seston FA in Lac Saint-Pierre during our study. Actually, though phytoplankton diversity within our samples may have buffered and mitigated the seston fatty acid signature in response to the underwater light climate, our field campaign did not account for all the environmental variables that may have influenced the FA composition of the seston in Lac Saint-Pierre. For instance, due to analytical problems we did not consider the influence of phytoplankton biomass within our seston samples. Nonetheless, our field study sheds light on

the potential importance, yet to be precised, of specific wavelengths as a driver of Lac Saint-Pierre's productivity through their influence on the fatty acids composition of seston and its nutritional quality for primary consumers. Further laboratory research should be conducted to elucidate the physiological processes underlying the suspected influence of specific PAR wavelengths on the concentration of essential PUFA in planktonic primary producers, and to discriminate the direct (i.e., through photosynthetic processes) and indirect (i.e., through changes in the phytoplankton community) impacts of light on seston nutritional quality. In addition, complementary field studies should also be conducted, covering a wider range of conditions over the seasonal cycle of production of Lac Saint-Pierre during which the underwater spectral characteristics, seston nutritional quality as well as zooplankton taxonomic composition (Pinel-Alloul et al. 2011) and growth are likely to vary.

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References

American Public Health Association (1998) Standard methods for the examination of water and wastewater, 20th edn. APHA, Washington, DC

Andersen T, Fœrøvig PJ, Hessen DO (2007) Growth rate versus biomass accumulation: different roles of food quality and quantity for consumers. Limnol Oceanogr 52:2128–2134

Barsanti L, Bastianini A, Passarelli V, Tredici MR, Gualtieri P (2000) Fatty acid content in wild type and WZSL mutant of *Euglena gracilis*. J Appl Phycol 12:512–520

Basu BK, Kalff J, Pinel-Alloul B (2000) The influence of macrophyte beds on plankton communities and their export from fluvial lakes in the St Lawrence River. Freshw Biol 45:373–382

Brett MT, Müller-Navarra D (1997) The role of highly unsaturated fatty acids in aquatic foodweb processes. Freshw Biol 38:483–499

Brett MT, Müller-Navarra DC, Persson J (2009) Crustacean zooplankton fatty acid composition. In: Arts MT, Brett MT, Kainz MJ (eds) Lipids in aquatic ecosystems. Springer, New York

Chícharo MA, Chícharo L (2008) RNA:DNA ratio and other nucleic acid derived indices in marine ecology. Int J Mol Sci 9:1453–1471

Dalsgaard J, St. John M, Kattner G, Müller-Navara D, Hagen W (2003) Fatty acid trophic markers in the pelagic marine environment. Adv Mar Biol 46:225–340

Frenette J-J, Arts MT, Morin J (2003) Spectral gradients of downwelling light in a fluvial lake (Lake Saint-Pierre, St-Lawrence River). Aquat Ecol 37:77–85

Frenette J-J, Arts MT, Morin J, Gratton D, Martin C (2006) Hydrodynamic control of the underwater light climate in fluvial Lac Saint-Pierre. Limnol Oceanogr 51:2632–2645

Frenette J-J, Massicotte P, Lapierre J-F (2012) Colorful niches of phytoplankton shaped by the spatial connectivity in a large river ecosystem: a riverscape perspective. PLoS ONE (accepted)

Gorokhova E, Kyle M (2002) Analysis of nucleic acids in *Daphnia*: development of methods and ontogenetic variations in RNA-DNA content. J Plankton Res 24:511–522

Guschina IA, Harwood JL (2009) Algal lipids and effect of the environment on their biochemistry. In: Arts MT, Brett MT, Kainz MJ (eds) Lipids in aquatic ecosystems. Springer, New York

Harwood JL (1998) Involvement of chloroplast lipids in the reaction of plants submitted to stress. In: Siegenthaler P-A, Murata N (eds) Lipids in photosynthesis: structure. Funct Genet, Kluwer

Hudon C, Paquet S, Jarry V (1996) Downstream variations of phytoplankton in the St. Lawrence river (Quebec, Canada). Hydrobiol 337:11–26

Huggins K, Frenette J-J, Arts MT (2004) Nutritional quality of biofilms with respect to light regime in Lake Saint-Pierre (Québec, Canada). Freshw Biol 49:945–959

Kainz M, Arts MT, Mazumder A (2004) Essential fatty acids in the planktonic food web and their ecological role for higher trophic levels. Limnol Oceanogr 49:1784–1793

Kainz MJ, Johannsson OE, Arts MT (2010) Diet effects on lipid composition, somatic growth potential, and survival of the benthic amphipod *Diporeia* spp. J Great Lakes Res 36:351–356

Katona SK (1970) Growth characteristics of the copepods *Eurytemora affinis* and *E. herdmani* in laboratory cultures. Helgoländer wiss Meeresunters 20:373–384

Kirk JTO (1994) Light and photosynthesis in aquatic ecosystems. Cambrige University Press, Cambridge

Koski M, Klein Breteler W, Schogt N (1998) Effect of food quality on rate of growth and development of the pelagic copepod *Pseudocalanus elongatus* (Copepoda, Calanoida). Mar Ecol Prog Ser 170:169–187

Kyle M, Watts T, Schade J, Elser JJ (2003) A microfluorometric method for quantifying RNA and DNA in terrestrial insects. J Insect Sci 3:1–7

Langlois C, Lapierre L, Léveillé M, Turgeon P, Ménard C (1992) Synthèse des connaisances sur les communautés biologiques du Lac Saint-Pierre. Rapport technique. Zone d'intéret prioritaire. Centre Saint-Laurent, Conservation et Protection, Environment Canada

Lapierre J-F, Frenette J-J (2009) Effects of macrophytes and terrestrial inputs on fluorescent dissolved organic matter in a large river system. Aquat Sci 71:15–24. doi:10.1007/s00027-009-9133-2

Lee H-W, Ban S, Ikeda T, Matsuishi T (2003) Effect of temperature on development, growth and reproduction in the marine copepod *Pseudocalanus newmani* at satiating food condition. J Plankton Res 25:261–271

Liebig J (1940) Chemistry in its application to agriculture and physiology. Taylor and Walton, London

Martineau C, Vincent WF, Frenette JJ, Dodson JJ (2004) Primary consumers and particulate organic matter: isotopic evidence of strong selectivity in the estuarine transition zone. Limnol Oceanogr 49:1679–1686

Mayzaud P, Claustre H, Augier P (1990) Effect of variable nutrient supply on fatty acid composition of phytoplankton grown in an enclosed experimental ecosystem. Mar Ecol Prog Ser 60:123–140

Mortensen SH, Borsheim KY, Rainuzzo JR, Knutsen G (1988) Fatty acid and elemental composition of the marine diatom *Chaetoceros gracilis* Schütt. Effects of silicate deprivation, temperature and light intensity. J Exp Mar Biol Ecol 122:173–195

Müller-Navarra DC (2006) The nutritional importance of polyunsaturated fatty acids and their use as trophic markers for herbivorous zooplankton: does it contradict? Arch Hydrobiol 167:501–513

Müller-Navarra DC (2008) Food web paradigms: the biochemical view on trophic interaction. Int Rev Hydrobiol 93:489–505

Müller-Navarra DC, Brett MT, Liston AM, Goldman CR (2000) A highly unsaturated fatty acid predicts carbon transfer between primary producers and consumers. Nature 403:74–77

Müller-Solger AB, Jassby AD, Müller-Navarra DC (2002) Nutritional quality of food resources for zooplankton (*Daphnia*) in a tidal freshwater system (Sacramento–San Joaquin River Delta). Limnol Oceanogr 47:1468–1476

Nordin LJ, Arts MT, Johannsson OE, Taylor WD (2008) An evaluation of the diet of *Mysis relicta* using gut contents and fatty acid profiles in lakes with and without the invader *Bythotrephes longimanus* (Onychopoda, Cercopagidae). Aquat Ecol 42:421–436

Parrish CC (2009) Essential fatty acids in aquatic food webs. In: Arts MT, Brett MT, Kainz MJ (eds) Lipids in aquatic ecosystems. Springer, New York

Persson J (2007) Food quality effects on zooplankton growth and energy transfer in pelagic freshwater food webs. Ph.D. thesis, Upsalla University

Pinel-Alloul B, Cusson E, Aldamman L (2011) Diversity and spatial distribution of copepods in the St. Lawrence River. In: Defaye D, Suárez-Morales E, Carel von Vaupel Klein J (eds) Studies on freshwater copepoda: a volume in honour of Bernard Dussart, Crustac Monogr 16. Brill Academic Publishers, Leiden

Pommier J, Frenette J-J, Glémet H (2010) Relating RNA:DNA ratio in *Eurytemora affinis* to seston fatty acids in a highly dynamic environment. Mar Ecol Prog Ser 400:143–154

Reuss N, Poulsen LK (2002) Evaluation of fatty acids as biomarkers for a natural plankton community. A field study of a spring bloom and a post-bloom period off West Greenland. Mar Biol 141:423–434

Reynolds CS, Descy JP (1996) The production, biomass and structure of phytoplankton in large rivers. Arch Hydrobiol 113:161–187

Rocap G, Larimer FW, Lamerdin J, Malfatti S, Chain P, Ahlgren NA (2003) Genome divergence in two *Prochlorococcus* ecotypes reflects oceanic niche differentiation. Nature 424:1042–1047

Sakia RM (1992) The Box-Cox transformation technique: a review. Statistician 41:169–178

Smyntek P, Teece M, Schulz K, Storch A (2008) Taxonomic differences in the essential fatty acid composition of groups of freshwater zooplankton relate to reproductive demands and generation time. Freshw Biol 53:1768–1782. doi:10.1111/j.1365-2427.2008.02001.x

Speekman CL, Nunez BS, Buskey EJ (2007) Measuring RNA:DNA ratios in individual *Acartia tonsa* (Copepoda). Mar Biol 151:759–766

Stomp M, Huisman J, Vörös L, Pick FR, Laamanen M, Haverkamp T, Stal LJ (2007) Colourful coexistence of red and green picocyanobacteria in lakes and seas. Ecol Lett 10:290–298

Thompson GA Jr (1996) Lipids and membrane function in green algae. Biochim Biophys Acta 1302:17–45

Thompson PA, Harrison PJ, Whyte JNC (1990) Influence of irradiance on the fatty acid composition of phytoplankton. J Phycol 26:278–288

Ventura M (2006) Linking biochemical and elemental composition in freshwater and marine crustacean zooplankton. Mar Ecol Prog Ser 327:233–246

Vrede T, Persson J, Aronsen G (2002) The influence of food quality (P:C ratio) on RNA:DNA ratio and somatic growth rate of *Daphnia*. Limnol Oceanogr 47:487–494

Wagner MM, Campbell RG, Boudreau CA, Durbin EG (2001) Nucleic acids and growth of *Calanus finmarchicus* in the laboratory under different food and temperature conditions. Mar Ecol Prog Ser 211:185–197

Wainman BC, Smith REH, Rai H, Furgal JA (1999) Irradiance and lipid production in natural algal populations. In: Arts MT, Wainman BC (eds) Lipids in freshwater ecosystems. Springer, New York

Wang KS, Chai TJ (1994) Reduction in omega-3-fatty-acids by UV-B radiation in microalgae. J Appl Phycol 6:415–421

Wellnitz TA, Ward JV (1998) Does light intensity modify the effect mayfly grazers have on periphyton? Freshw Biol 39:135–149