

Photochemical transformation of dissolved organic matter in lakes

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Abstract

In a survey of photochemical transformation of dissolved organic matter (DOM) in lake water, we found photochemical production of dissolved inorganic carbon (DIC) and low molecular weight carboxylic acids (oxalic, malonic, formic, and acetic acid), upon simulated solar ultraviolet (UV) radiation exposure of filter-sterilized surface water from 38 investigated lakes. Given the large variability in water chemistry of the studied systems, we suggest that these photoproducts are universally produced in sunlight exposed lake water. The measured production of carboxylic acid carbon corresponded to 34.4% (median) of the produced DIC. The production of DIC and carboxylic acids during 8 h of mild UV irradiation corresponded to an average of 4.7% (SD 2.9) of the total dissolved organic carbon pool. Hence, photochemical degradation could be a major sink for DOM in the surface layer of lakes. All photoreaction rates were strongly correlated to the total radiation absorbed during UV exposure ($r^2 > 0.8$), which depends mainly on amount of colored DOM in the water. The variability in DIC production after normalizing for absorbed radiation energy could partly be attributed to general water chemistry parameters (e.g., pH, iron concentration, conductivity).

Dissolved organic matter (DOM) affects the character and function of aquatic ecosystems and is a major reservoir of organic carbon. There is a continuous supply of DOM to freshwaters, both from terrestrial input (allochthonous material) as well as from indigenous primary production (autochthonous material). The organic material is not conservative in freshwater systems, but rather undergoes various biological and photochemical transformations before a residual of the DOM is eventually discharged into the oceans (Miller and Zepp 1995; Amon and Benner 1996; Opsahl and Benner 1998; Tranvik 1998).

Heterogeneous, high molecular weight polymeric compounds (e.g., humic substances) constitute the major part of terrestrially derived DOM (Thurman 1985). There is increasing evidence that these substances are susceptible both to microbial and photochemical degradation (see reviews by Moran and Zepp 1997; Tranvik 1998). Solar radiation, par-

ticularly in the ultraviolet (UV)-A–UV-B range, promotes transformation of both the structure, molecular weight, and optical properties of humic substances (Strome and Miller 1978; DeHaan and DeBoer 1991; Allard et al. 1994; Kulo-vaara et al. 1996). In addition, DOM is oxidized into dissolved inorganic carbon (DIC) (Salonen and Vähätalo 1994; Miller and Zepp 1995), carbon monoxide (Conrad and Seiler 1980; Miller and Zepp 1995), and transformed into specific low molecular weight organic compounds, such as aldehydes and carboxylic acids (reviewed by Moran and Zepp 1997). Bioassay experiments demonstrate that irradiation of humic substances and humic surface waters can significantly increase the pool of bioavailable carbon substrates (Strome and Miller 1978; Geller 1986; Lindell et al. 1995, 1996; Bertilsson and Allard 1996). Thus, solar radiation has the potential to alter the spectral and molecular properties of DOM, and to promote its degradation, either directly through complete photooxidation or indirectly by increased bioavailability.

Most of the information on the photoreactivity of DOM comes from systems with abundant allochthonous, mainly humic DOM, and minor influence of autochthonous DOM. The few published studies that do focus on autochthonous DOM are distinctly different in terms of experimental design and obtained results. Thomas and Lara (1995) reported that DOM derived from a diatom culture was rather recalcitrant towards photoreactions, since no production of DIC from

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^{14}C -labeled DOM could be detected during a 15 h exposure to UV-A radiation. They suggested that this resistance to photoreactions could be a general feature of algal-derived DOM. This view is contrasted by a recent study, where DOM photobleaching was observed in a wide variety of lakes, irrespective of the indigenous production of organic matter (Reche et al. 1999). In addition, the combined presence of DOM originating from two principal sources (i.e., indigenous phytoplankton and terrestrial vascular plants), may cause interactive and less obvious effects of solar radiation on DOM quality (Keil and Kirchman 1994; Tranvik and Kokalj 1998). There is a lack of systematic studies addressing the photochemical production of DIC and various organic photoproducts in lakes of varying quality, including eutrophic and oligotrophic as well as clear and humic lakes. We hypothesize that photochemical processes (formation of DIC and carboxylic acids as well as bleaching) occur ubiquitously in temperate lakes and that the quality of the total DOM, and in particular the ability of this material to directly absorb radiation energy, is a major factor governing the rates of the various photoreactions. We surveyed the photochemical reactivity of organic matter in water from 38 different Swedish lakes and assessed it in relation to chemical characteristics of the water samples.

Materials and methods

Sampling and incubation—To obtain lake waters covering a wide range of organic matter quality, water chemistry, and geographic position (Table 1), we sampled the surface water (0–0.5 m) of 38 selected lakes during June to August 1996. Samples (3 liters) were collected in polyethylene bottles, and were frozen within 1 h of sampling. All material in contact with the water had been soaked in 2 M HCl overnight, rinsed in hot (70°C) 0.02 M HCl, and subsequently rinsed at least four times in ultrapure water (Milli-Q, Millipore) prior to use.

Immediately prior to experiments, water samples were thawed at room temperature and were subsequently filtered through Milli-Q rinsed 0.2 μm membrane filters (Filtropur-L, Sarstedt) into autoclaved glass bottles. Thereafter, subsamples were transferred to quartz tubes (approximate volume 70 ml, diameter 3 cm) with ground glass stoppers. Duplicate tubes were set up for each lake. One of the duplicates was exposed to artificial UV radiation while the other was wrapped in aluminum foil for use as a dark control. The quartz tubes were exposed during 12 h under mild rotary shaking (60 rpm) at 20°C, placed horizontally at 30 cm distance from 8 UVA-340[®] fluorescent tubes (Q-panel), with an emission maximum in the UV-A band (340 nm; Fig. 1). A radiometer (IL-1400, International Light) equipped with broad-band sensors for UV-B (280–320 nm), UV-A (320–400 nm), and photosynthetically active radiation (PAR) (400–750) was used to monitor the total irradiation dose during the incubation. In addition, an Optronics Model 752 spectroradiometer was used for calibration and for obtaining a detailed emission spectrum for the lamps. The irradiance was about 1.5 W m^{-2} of UV-B, 17.4 W m^{-2} of UV-A, and 15.4 $\mu\text{E m}^{-2} \text{s}^{-1}$ of PAR (equal to about 5 W m^{-2}).

Dissolved carbon characterization—Immediately after incubation, the concentration of DIC was analyzed by direct injection of samples from quartz-tubes into a Shimadzu TOC-5000 total carbon analyzer (Granéli et al. 1996). At least three injections were made resulting in a coefficient of variation of less than 2%. Instrument calibration and performance was checked by repeated injection of various standards (0, 2.5, 5, 10, and 20 mg DIC L^{-1}) prepared from Na_2CO_3 and NaHCO_3 .

We analyzed four low molecular weight (LMW) carboxylic acids (oxalic, malonic, formic, and acetic acid) initially in the sterile filtered surface waters, as well as after the experimental incubations. Analytes were separated on a capillary electrophoresis system (Quanta 4000, Millipore), equipped with an 80-cm fused silica capillary (75 μm inner diameter). A 5 mM 1,2,4-benzene tri-carboxylic acid buffer (pH 7.9) containing 0.5 mM of an electroosmotic flow modifier (OFM-BT, Waters) was used as the mobile phase (Dahlén et al. 1996; Bertilsson and Tranvik 1998). A separation voltage of 15 kV was applied, and injection was accomplished using electrokinetic preconcentration (45 s at 5 kV). The separated analytes were indirectly detected by their lower absorption at 254 nm compared to the highly-absorbing mobile phase used. Molybdate (200 $\mu\text{g L}^{-1}$) was added to all samples as an internal standard. Identification and quantification of the analytes were achieved by a range of carboxylic-acid standard additions to the initial samples from each individual surface water. The added concentrations always covered the range of observed concentrations in the experimental treatments. Standard curves for concentration estimates were obtained by linear regression ($r^2 > 0.8$ for all acids and water samples, $N = 4$).

Aliquots of filtered lake water were sampled prior to experimental incubations for initial characterization of dissolved organic carbon (DOC) and humic substance fluorescence, and after the experimental incubations for observations of treatment effects (absorbance). Aliquots for DOC analysis (5 ml) were acidified with 20 μl of 2 M HCl and purged with CO_2 -free air for 5 min to remove inorganic carbon. DOC was measured with the high temperature catalytic oxidation method (HTCO) and infra-red detection using a Shimadzu TOC-5000 total carbon analyzer. Samples of 33 μl were injected in triplicate with a resulting coefficient of variation of 2% or less. Efficient removal of inorganic carbon was checked by analysis of IC in purged samples. Carbon concentration was calculated using standard solutions prepared from potassium hydrogen phthalate.

Humic substance fluorescence was determined with a Shimadzu RF-1501 spectrofluorometer equipped with a 10 \times 10 mm quartz cuvette at 355 nm excitation and 455 nm emission wavelengths (10 nm bandwidth). A 0.1 M H_2SO_4 solution was used as a blank. Values were normalized to the fluorescence of quinine sulfate using standard solutions, with one quinine sulfate unit being equal to the fluorescence of a 0.01 mg L^{-1} solution in 0.1 M H_2SO_4 .

Absorbance spectra (200–500 nm) were measured at 0.5 nm intervals in a 10 mm quartz cuvette, using a Shimadzu MPS-2000 spectrophotometer. The absorption of UV-radiation in lake water typically decreases with increasing wavelength, and the absorption in the upper UV-range (280–400

Table 1. Description of lakes. The position is given in longitude (Long.) and latitude (Lat.); DOC is the dissolved organic carbon; HS-FI is the humic substance fluorescence in quinine sulphate units (QSU); Cond. is the conductivity; Alk. is the alkalinity (acid neutralizing capacity); and Tot-Fe is the total concentration of Fe in the water. Abs is the absorptivity (m^{-1}) at the indicated wavelength and $S_{280-400}$ represents the slope of exponential fits ($r > 0.98$, 120 wavelengths) for the wavelength-dependent absorption between 280 and 400 nm using absorption at 280 nm as the reference wavelength.

Nr	Lake name	Position Long. (N)/Lat. (E)	DOC mg L ⁻¹	HS-FI (QSU)	Chl- <i>a</i> μg L ⁻¹	pH	Cond. μS	Alk. mEq L ⁻¹	Tot-Fe μg L ⁻¹	$S_{280-400}$ $10^3 \times nm^{-1}$	Abs _{280nm} (m ⁻¹)
1	Vättern	58°28'0/14°55'2	2.4	1.4	0.2	7.8	139	0.57	0.9	21.3	5.6
2	Skärölen	57°10'5/14°31'2	3.1	3.4	2.6	7.5	55	0.02	2.8	17.5	9.0
3	Klaintsjön	57°07'5/14°42'4	3.2	5.4	0.8	5.2	48	0.00	12	16.4	10.9
4	Yxningö	58°16'5/16°13'0	5.0	17.4	2.2	7.8	99	0.42	5.0	19.9	15.9
5	Ännsjön	63°17'5/12°33'0	2.0	4.5	0.3	7.4	27	0.16	7.9	15.8	12.5
6	Fiolen	57°04'7/14°32'5	4.4	7.6	2.1	6.4	61	0.04	6.9	18.4	17.5
7	Vallentunasjön	59°30'8/18°02'5	8.7	18.0	50.0	8.2	341	1.93	2.3	21.1	26.5
8	Västra Ringsjön	55°54'7/13°27'0	7.1	15.6	21.4	9.4	204	1.10	17	21.0	27.5
9	Jämlunden	58°11'5/15°35'5	7.4	18.2	1.8	8.2	145	0.58	9.4	19.0	34.5
10	Östra Ringsjön	55°52'1/13°30'0	7.3	23.1	48.3	8.6	208	0.85	80	18.2	36.9
11	Finjasjön	56°07'4/13°44'0	7.7	22.7	4.9	9.0	240	0.87	14	18.3	37.6
12	Hålsjön (Jul)	59°49'0/17°15'0	8.9	17.8	4.6	8.5	38	0.94	14	19.2	41.9
13	Risten	58°17'0/16°04'0	8.5	19.5	4.5	9.1	155	0.62	18.0	18.8	40.8
14	Siholmatjärn	71°14'0/16°42'0	4.4	10.4	1.4	6.5	13	0.01	21	15.2	32.4
15	Hålsjön (May)	59°49'0/17°15'0	8.4	19.4	13.8	7.8	136	0.90	19	18.4	41.4
16	Vombsjön littoral	55°42'3/13°35'1	12.0	42.1	164.2	8.9	285	1.37	12	15.8	35.4
17	Sätroftasjön	55°54'0/13°33'0	10.1	26.8	36.0	9.2	201	0.90	NA	18.4	41.7
18	Norrgårdstjärn	61°55'0/14°44'0	4.2	12.1	0.2	6.8	22	0.10	44	14.7	33.0
19	Stråken	57°07'0/14°34'5	8.1	23.6	4.5	8.1	74	0.09	22	17.1	45.2
20	Dagstorpssjön	55°59'2/13°30'0	8.1	25.7	7.7	8.4	132	0.43	25	17.6	48.9
21	Vombsjön	55°42'3/13°35'1	21.8	67.3	503.5	8.5	322	1.67	7.2	14.2	39.4
22	Svinstadsjön	58°22'0/15°52'0	13.6	35.5	170.3	9.5	161	1.16	8.1	19.7	59.9
23	Gruvsjön	58°16'0/16°01'5	10.3	31.4	9.0	5.5	247	0.00	220	16.5	54.9
24	Siggeforasjön	59°58'0/17°09'0	10.2	24.9	1.4	7.2	53	0.18	41	17.4	60.1
25	Örträsk	64°10'0/18°55'0	7.3	28.8	1.1	6.8	31	0.12	66	15.5	58.3
26	Åspuss	59°43'0/17°06'0	18.9	43.3	157.4	8.8	277	0.96	400	16.0	85.8
27	Skärshultsjön (Jul)	57°10'1/14°31'2	11.2	45.6	2.4	5.4	62	0.00	330	15.3	84.8
28	Skärshultsjön (Jun)	57°10'1/14°31'2	10.9	46.1	1.7	5.4	63	0.00	240	15.3	88.3
29	Drommatjärn	71°10'0/16°55'0	9.1	37.0	1.5	5.4	17	0.00	270	14.2	86.8
30	Hultsjön	58°34'4/15°35'5	12.8	37.8	28.3	7.3	75	0.16	150	15.8	98.6
31	Bjån	58°34'4/15°30'5	12.6	45.0	7.5	7.0	67	0.18	170	15.1	102.8
32	Lilla Björntjärn	71°16'0/16°44'0	9.9	34.3	2.9	5.6	16	0.00	310	14.2	100.0
33	Lindhultsgölen	57°08'7/14°28'4	14.1	46.3	4.2	8.0	100	0.47	95	14.9	120.2
34	Nedre Björntjärn	71°16'0/16°45'0	12.0	44.6	1.1	5.6	21	0.00	500	14.1	119.3
35	Grytsjön	58°35'9/15°31'5	15.1	53.0	3.8	6.5	64	0.06	90	15.3	131.2
36	Byxriverhåden	71°16'0/16°43'0	14.7	49.6	0.6	5.5	27	0.01	490	14.3	131.2
37	Tvigölingen	60°05'0/17°24'0	22.0	66.8	50.7	7.7	92	0.39	180	16.1	168.4
38	Bergkärrsgölen	58°35'7/15°33'0	21.3	71.0	1.4	4.5	40	0.00	480	14.3	203.4

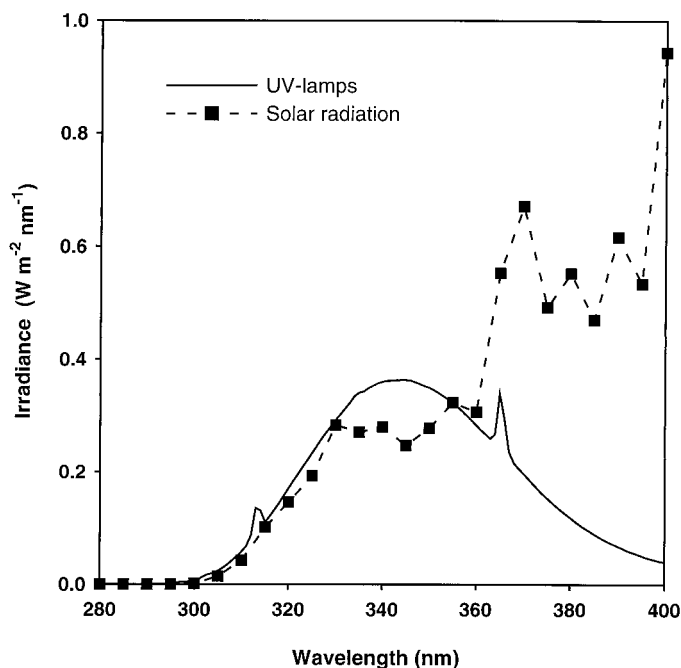


Fig. 1. Irradiance spectra of natural solar radiation (solid squares, dotted line) and the artificial UV-lamps (solid line) used in this study.

nm) can be approximated by an exponential function (Eq. 1; Davies-Colley and Vant 1987):

$$\text{Abs}_\lambda = \text{Abs}_{\text{ref}} \cdot e^{S \cdot (\lambda_{\text{ref}} - \lambda)} \quad (1)$$

Abs_λ is the absorbance at a specific wavelength (λ), Abs_{ref} is the absorbance at the reference wavelength (λ_{ref}). The constant S represents the slope of the exponential curve. Exponential regressions were forced through the chosen reference-wavelength absorbance, which was set at 280 nm (λ_{ref}).

Unfiltered water samples were collected for pigment analysis immediately after thawing. Between 100 and 300 ml of water sample was filtered onto Whatman GF-C filters (25 mm diameter), and these were immediately frozen to avoid deterioration of pigments. Pigments retained on the filters were extracted with ethanol, and chlorophyll *a* (Chl *a*) concentration was measured spectrophotometrically at 665 nm (Jespersen and Christoffersen 1987).

Inorganic water chemistry—Samples (20 ml) for metal analysis were added to polypropylene bottles and were immediately acidified by adding 10 μl of HCl (Suprapur, Sigma). Total iron was analyzed on a Perkin-Elmer 1100 atomic absorption spectrophotometer equipped with an HGA 500 graphite-furnace. Flame or furnace techniques were chosen according to requirements set by the metal concentration in the respective samples. Standard procedures were followed for measuring acid neutralizing capacity (ISO 9963-2), and conductivity (ISO 7888:1985), while pH was measured at room temperature without stirring.

Absorbed radiation energy—Photoreaction rates (DIC and carboxylic acid production) were normalized for the total

amount of absorbed radiation energy during the irradiation. The procedures presented by Scully et al. (1996) were followed, with the exception that formation was normalized to absorbed energy rather than absorbed moles of photons. For each of the irradiated water samples, the absorption of radiation per flat irradiated surface area of the tubes was calculated for the total incubation period (E_w , total absorbed energy in water samples per m^2 , Eq. 2). The calculation was performed for the entire UV-B and UV-A wavelength range (280–400 nm), i.e., throughout the principal radiation spectrum of the lamps (see Fig. 1).

$$E_w = T_{\text{UV}} \cdot \sum_{\lambda=280}^{\lambda=400} (I_{0\lambda} \cdot (1 - 10^{-\text{Abs}})_\lambda) \quad (2)$$

E_w is the total absorbed energy (J m^{-2}), T_{UV} is the irradiation time in seconds, $(1 - 10^{-\text{Abs}})_\lambda$ is the average fraction of penetrating radiation that is absorbed in a tube at a specific wavelength (λ). $I_{0\lambda}$ is the lamp irradiance in W m^{-2} for each corresponding 1 nm wavelength band. Spectral irradiance of the lamps was derived from an emission spectrum measured with an Optronic Model 752 spectroradiometer. To calculate the absorbed fraction of the radiation within each wavelength band $(1 - 10^{-\text{Abs}})_\lambda$ in each irradiated sample, the average radiation pathlengths in various parts of the cylindrical quartz tubes were taken into account (Eq. 3). This operation was performed to account for the impact of self-shading within the irradiated tubes.

$$(1 - 10^{-\text{Abs}})_\lambda \quad (\text{for an entire cylinder}) = \sum_1^{10} (1 - 10^{-\text{Abs} \cdot L}) \cdot 1/10 \quad (3)$$

We longitudinally divided each cylindrical quartz tube in 10 parallel subvolumes, each having a fixed width of 3 mm. L is the average irradiation path length for each of the 10 individual subvolumes (cm) and “Abs” is the absorbance (cm^{-1}) at the respective wavelength λ , obtained from absorption scans.

Statistical methods—The Unscrambler 6.11[®] software (Camo AS) was used for partial least squares regression analysis (PLS; Wold et al. 1984) in an effort to use background water chemical parameters to explain the residual variance in the two response parameters, DIC and carboxylic acid production (Table 2), after normalizing for total absorbed energy. This multivariate projection method is based on iterative fitting of partial bilinear models and involves replacement of the original response, and predictor variables (presented in Table 1) with two sets of orthogonal latent variables (LVs). Due to the orthogonality, consecutive LVs describe the variation in the data set independently of each other. Additional LVs are calculated as long as they improve the total model according to cross- or test set validation. The influence that the different predictor variables exert on each LV can be represented by weight vectors, where a large positive or negative value indicates a strong influence on the respective LV included in the model. Weight vectors can also be used to illustrate the relation between each LV and the response variables, hence allowing a direct comparison of the various predictor variables and the modeled responses.

Table 2. Total absorption of radiation energy in irradiated incubations (E_w) and photochemical response parameters. The contribution of UV-B to total absorbed radiation energy is given in percentage of total values. Photobleaching represents the difference in absorptivity between dark controls and irradiated samples, while photoproduction rates of DIC and the combined photoproduction rates of oxalic, malonic, formic, and acetic acids are based on concentrations in irradiated samples with dark controls subtracted. These photoproduction rates are also given as rates normalized for total absorbed energy (E_w). ND = not detected.

Lake	E_w		Photobleaching		Photoproduction rates		E_w -normalized Photoproduction	
	Total (kJ m ⁻²)	UV-B % of total	Abs _{250 nm}	Abs _{365 nm}	DIC prod.	C.A. prod.	DIC prod.	C.A. prod.
			(m ⁻¹)		(μg C L ⁻¹ h ⁻¹)		(μg C L ⁻¹ h ⁻¹ kJ ⁻¹ m ²)	
1	26	13.7	0.6	0.2	5.1	3.4	0.197	0.130
2	54	12.6	0.7	0.2	10.5	2.5	0.194	0.046
3	71	12.4	2.4	0.8	14.5	3.3	0.204	0.046
4	79	13.2	0.8	0.1	2.2	5.0	0.028	0.064
5	86	12.1	1.3	0.4	10.4	4.2	0.121	0.049
6	97	12.8	2.5	0.3	9.5	6.1	0.098	0.063
7	119	13.3	1.7	0.2	1.3	4.4	0.011	0.037
8	128	13.4	1.5	-0.7	17.3	9.3	0.135	0.073
9	176	12.6	4.7	1.0	15.4	5.9	0.087	0.034
10	194	12.3	5.7	1.4	7.5	13.1	0.039	0.067
11	198	12.4	5.9	1.5	21.4	7.4	0.108	0.037
12	204	12.5	4.8	1.2	17.5	6.0	0.086	0.029
13	205	12.4	5.8	1.4	24.3	12.4	0.118	0.061
14	210	11.5	4.6	1.2	20.1	6.7	0.096	0.032
15	212	12.2	4.5	1.3	21.3	17.2	0.100	0.081
16	214	11.2	6.0	2.5	13.8	13.6	0.064	0.064
17	215	12.3	5.7	1.6	28.6	9.4	0.133	0.044
18	219	11.4	4.6	1.4	28.8	12.1	0.131	0.055
19	249	11.7	6.7	1.4	34.8	11.5	0.140	0.046
20	257	11.8	5.8	1.6	31.0	14.6	0.121	0.057
21	262	10.6	4.4	3.0	ND	12.5	ND	0.017
22	264	12.0	9.9	2.9	37.5	22.1	0.142	0.084
23	299	11.4	7.4	0.7	40.1	12.3	0.134	0.041
24	305	11.5	8.2	1.8	36.3	11.7	0.119	0.038
25	331	11.0	8.1	1.8	43.4	20.4	0.131	0.062
26	416	10.5	10.9	3.6	0.5	10.2	0.001	0.025
27	433	10.4	20.6	3.9	114.5	28.1	0.264	0.065
28	445	10.4	19.4	4.0	92.5	23.3	0.208	0.052
29	460	10.1	10.9	2.6	69.9	21.3	0.152	0.046
30	462	10.3	11.2	2.6	67.9	24.1	0.147	0.052
31	490	10.1	11.6	2.9	69.3	23.6	0.141	0.048
32	501	9.9	15.1	3.2	93.8	22.1	0.187	0.044
33	537	9.7	13.9	4.7	69.2	33.0	0.129	0.061
34	550	9.6	21.6	4.7	125.6	40.8	0.228	0.074
35	556	9.7	14.7	3.5	85.3	26.6	0.153	0.048
36	575	9.5	22.8	4.5	158.5	44.2	0.276	0.077
37	605	9.4	12.4	2.8	86.3	29.3	0.142	0.048
38	688	8.8	27.9	6.3	140.0	37.1	0.203	0.054

This method does not rely on some of the more critical assumptions made when classical multivariate methods (e.g., analysis of variance, multiple linear regression) are used—in particular, the assumption of negligible analytical error in the measured variables and the absence of covariance between the predictor variables used in the model (Eriksson et al. 1995). All parameters (predictors and response variables) were autoscaled before use in the model (the geometric means centered at zero and all data normalized for standard deviation). Full cross validation was used in the modeling procedure, one sample being excluded at the time. The response parameters for each excluded sample was then pre-

dicted from a PLS model created from the remaining samples. The predictive strength of the model was assessed by linear regressions of measured values in the response variables versus the predicted values obtained in the cross validation procedure. Q^2 is the coefficient of determination for this regression, and is the proportion of variance in the response parameter being explained by the model, and RMSEP is the root mean square error of prediction (i.e., the average difference between the measured and predicted values of the response variable). For a more detailed description of PLS, see Wold et al. (1984), Geladi and Kowalski (1986), and Eriksson et al. (1995).

Results

Initial character of DOM in lakes—The total DOC in lakes ranged from 2 to 22 mg C L⁻¹ (median 9 mg C L⁻¹; Table 1). The humic substance fluorescence and the concentration of Chl *a* in the various water samples ranged between 2 and 3 orders of magnitude, respectively (Table 1). The ratio between these two parameters covered almost 3 orders of magnitude (range 0.1–87 QSU (μg Chl *a* L⁻¹)⁻¹).

DOC-specific absorbance at 250 nm (range 0.01–0.06 cm⁻¹ (mg C L⁻¹)⁻¹), DOC-specific humic substance fluorescence (range 0.6–4.2 QSU (mg C L⁻¹)⁻¹), and DOC-specific Chl *a* concentration (range 0.04–23.2 μg Chl (mg C L⁻¹)⁻¹) varied considerably. The observed variability in these DOM-related chemical parameters indicates substantial differences in the amount and quality of total DOM in the studied systems.

Spectral absorbance and absorption of radiation energy—The absorbance at the reference wavelength (280 nm), as well as the exponential slope of the spectra in the upper UV-range (*S*; see Eq. 1), varied substantially between the different lakewater samples (Table 1). The coefficients of determination for the exponential regressions of individual spectra (*r*²) were always higher than 0.99 (120 data points), except for the extremely clearwater lake 1 (*r*² = 0.98). The total absorbed radiation energy in irradiated water samples, calculated according to Eqs. 2–3, ranged from 26 to 688 kJ m⁻² (Table 2) and was linearly correlated to the absorbance at 250, 280, and 365 nm (*P* < 0.001, *r* = 0.97, 0.97, and 0.99 respectively). The UV-absorbance decreased upon irradiation, as exemplified by two fixed wavelengths (250 and 365 nm) in Table 2, while there was no significant impact of the irradiation on spectral slope (*P* > 0.05 paired *t*-test, *n* = 38).

Photoproduction of DIC and carboxylic acids—We observed a photochemical production of DIC in all but one of the studied lake waters (Table 2). The very high background DIC in this lake (17 mg C L⁻¹; lake 21) may have obscured the detection of photoproduction of DIC.

A number of low molecular weight carboxylic acids (i.e., formic, acetic, malonic, and oxalic acid) were photochemically produced at high rates. The combined photoproduction of these four compounds ranged from 0.5 to 158.5 μg C L⁻¹ h⁻¹ in the studied waters (Table 2). Production of all four compounds could be detected in most waters (no oxalic acid production in lakes 23 and 36; no acetic acid formation in lakes 12, 21, and 26). Formic acid was the major photoproduct constituting on average 42.2% of the total photoproduction of carbon retrieved in the four quantified carboxylic acids (SD ± 11.4%). The relative production of acetic acid was slightly lower and more variable (28.1 ± 19.4%). Oxalic and malonic acid were both produced in lower amounts (15.6 ± 11.7 and 14.1 ± 6.6% respectively). Several additional photoproducts were also observed using the described capillary electrophoresis method, but the produced amounts appeared to be at least an order of magnitude lower compared to any of the four carboxylic acids quantified in the present study. Some of these products were tentatively

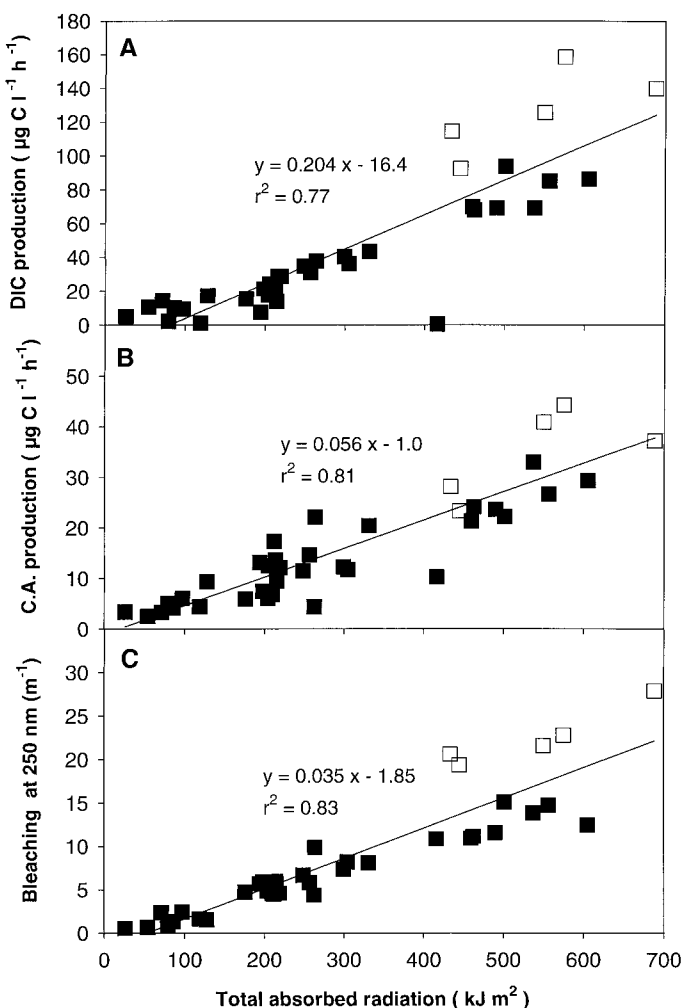


Fig. 2. Total absorbed radiation energy in irradiated samples and the observed photochemical production of DIC (A), the sum of photochemically produced oxalic, malonic, formic, and acetic acid (B), and photobleaching at 250 nm (C). Photoproduction of DIC and carboxylic acid carbon are hourly rates while photobleaching and the total absorbed radiation (kJ m⁻²) is for the entire 8 h incubation period. The lines represent linear regressions (*N* = 38), and the open symbols represent 6 humic-rich, iron rich, and acidic waters that appeared to have a substantially higher photochemical DIC production and photobleaching compared to the rest of the data set when normalized for total absorbed radiation.

identified as succinic acid, pyruvic acid, butyric acid, and lactic acid. The photoproduction rates both for DIC and the total carboxylic acid carbon (sum of the four compounds identified above) were strongly correlated to total absorbed radiation energy (Fig. 2A,B). There was a strong linear correlation between the photoproduction rates for DIC and total carboxylic acids (*P* < 0.001, *r* = 0.87), and the produced carboxylic acid carbon corresponded to 34.4% (median) of the measured DIC production. The rates for both of these photoreactions were also linearly related to photobleaching at 250 nm (*P* < 0.001, *r* = 0.87 and 0.91 respectively) and 365 nm (*P* < 0.001, *r* = 0.77 and 0.82 respectively), as well as to DOC (*P* < 0.001, *r* = 0.63 and 0.69) and initial UV

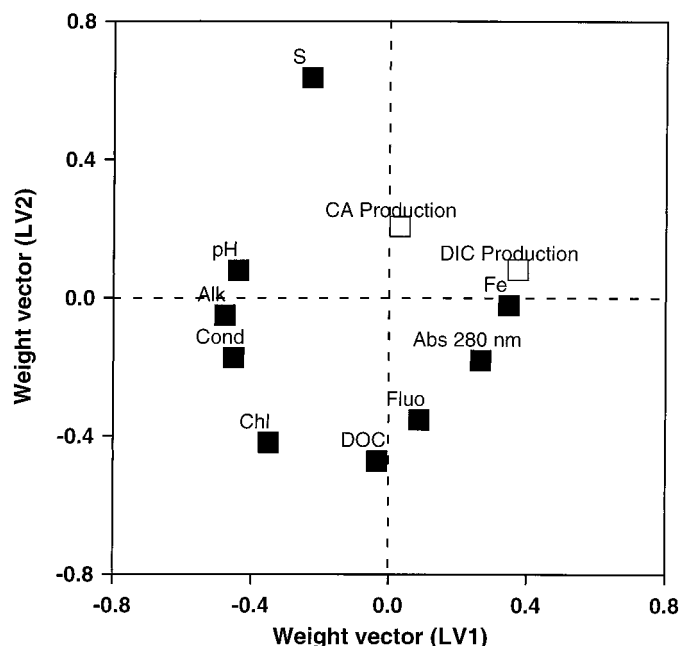


Fig. 3. PLS-model weight-vector plot for predictor variables (solid squares) and the response variables (open squares), for the first (LV1) and second latent variable (LV2). DIC- and carboxylic acid production indicate the total photoproduction of these compounds after normalizing for the total absorbed radiation energy in the experiment. Alk. indicates alkalinity; Cond. is the conductivity; Chl. is the chlorophyll *a* concentration; Fluo. is the humic substance fluorescence; Fe. is the total iron concentration; Abs 280 nm is the absorbance at the reference wavelength 280 nm; and *S* indicates the exponential regression slope of the spectral absorbance in the upper UV-range (280–400 nm) for the irradiated water samples.

absorbance at 250 nm ($P < 0.001$, $r = 0.86$ and 0.87) and 365 nm ($P < 0.001$, $r = 0.88$ and 0.87).

Influence of water chemistry on DIC and carboxylic acid formation—The total absorption of radiation energy explained a major part of the observed variability in photoproduction of DIC and carboxylic acids (Table 2; Fig. 2A,B). However, considerable residual variability in reaction rates remained even after this normalization. PLS was used to investigate the relation between these normalized reaction rates and nine water-chemistry variables (Table 1). All nine of these predictor variables had a significant impact on either or both of the two LVs that were included in the PLS model (Fig. 3). The energy-normalized DIC production could be modeled with some success (Fig. 4A). For DIC production efficiency, almost all of the explained variance was due to the first latent variable (LV1) with a negligible influence from the second latent variable (LV2) (1.1% of the total explained variance). On the contrary, the model could not explain any of the residual variance in energy-normalized carboxylic acid production ($Q^2 < 0.001$; Fig. 4B). Total iron and UV-absorbance (280 nm) exhibited a strong positive relation with DIC production, while alkalinity, pH and conductivity were negatively related to this response parameter for the LV1 (Fig. 3; LV1). Consequently, the use of only two parameters (pH and total Fe) was adequate to model

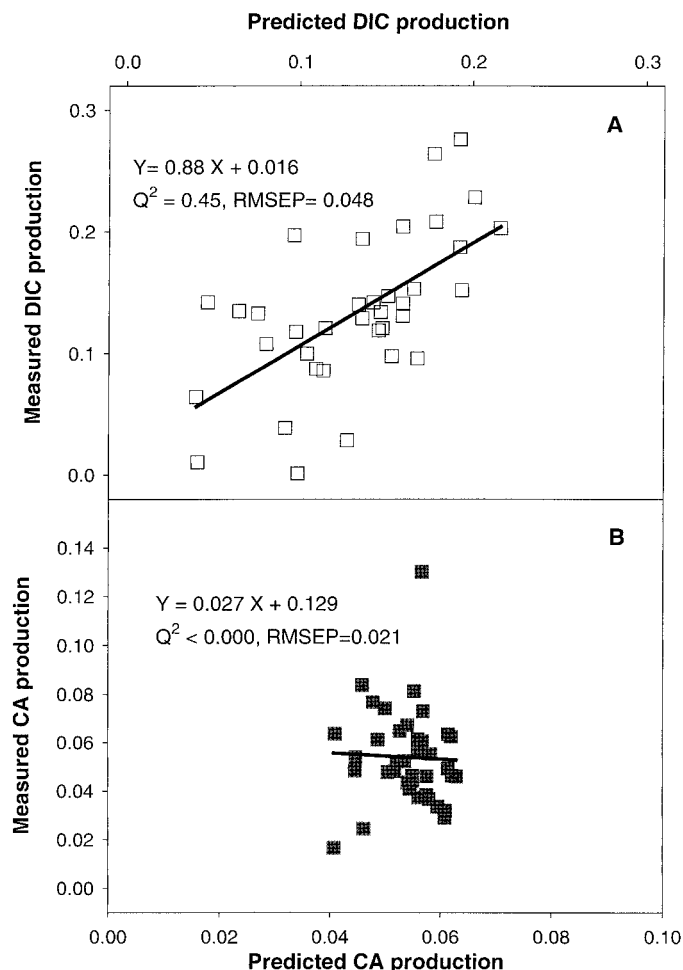


Fig. 4. PLS-modelling correlation plots of (A) predicted versus measured DIC- and (B) carboxylic acid production rates after normalization for total absorbed radiation energy. Predicted values are from leave-one-out cross validations and lines indicate linear regressions ($N = 38$). Q^2 is the coefficient of determination for cross-validated predictions and RMSEP is the root mean square error of prediction.

DIC production efficiency with only a slightly lower precision than the complete model with all nine predictor variables (slope 0.88, $Q^2 = 0.30$, RMSEP = 0.43). These results agree with the observation that 5 acidic waters with a high content of humic substances and iron (lakes 27, 28, 34, 36, and 38) exhibited a substantially higher photoreactivity (DIC production and photobleaching) compared to systems with similar absorption of total radiation energy (Fig. 2A,C; Table 2). This pattern was not apparent for the combined photoproduction of the four carboxylic acids (Fig. 2B)

Discussion

Photochemical production of both DIC and carboxylic acids has been previously described, separately or in combination, from about two dozen of different oligotrophic lakes, most of which have been characterized by high concentrations of terrestrially derived, highly colored organic matter

(i.e., humic substances: Salonen and Vähätalo 1994; Miller and Zepp 1995; Wetzel et al. 1995; Bertilsson and Allard 1996; Granéli et al. 1996, 1998; Kulovaara 1996; Bertilsson and Tranvik 1998; Bertilsson et al. 1999). Notably, there are no studies reporting the occurrence of these photochemical processes in highly productive surface waters with a large algal contribution to the total DOM pool.

We surveyed the photochemical production of DIC and carboxylic acids in water from a wide range of lakes, varying extensively, in humic content and primary productivity. We observed a photochemical formation of these products in virtually all of the 38 studied waters (Fig. 2A,B). Our results imply that DIC and carboxylic acids are photochemically produced by solar radiation in a wide range of lake waters that are dramatically different regarding water chemistry parameters, including those related to organic matter composition. Thus, we show that not only waters heavily influenced by terrestrial DOM, but most lakes in the temperate climate region, are likely to be influenced by a solar driven photochemical production of DIC and carboxylic acids. It is reasonable to assume that these results could apply also to water bodies in other climate zones.

Photons must be absorbed to produce a photochemical reaction in water. However, the quantum yield for the photoreactions (i.e., number of photoreactions per absorbed photon), will most likely vary with the wavelength of the absorbed radiation (Miller 1998). Few studies have addressed the wavelength-dependency of quantum yields for photoreactions involving natural organic matter, and those that do exist imply a strong negative correlation between irradiation wavelength and quantum yield, at least in the upper UV-range (Kieber et al. 1990; Moore et al. 1993; Valentine and Zepp 1993; Moran and Zepp 1997). So far, no detailed studies have been presented of the wavelength-dependent quantum yields for the photochemical production of carboxylic acids or DIC from natural organic matter in lakes. However, in a recent study by Gao and Zepp (1998), quantum yields for DIC production for water from a strongly coloured river appeared to decrease exponentially with increasing wavelength between 290 and 390 nm.

In the present study, a normalization for the total absorbed radiation energy explained about 80% of the variance in all of the measured photoreaction parameters (DIC production, carboxylic acid photoproduction, and bleaching of UV-absorbance; Fig. 2A,C). The remaining variance in photoproduction rates after this normalization could have several causes, for example (1) differences in total concentration of substrate for the photochemical reaction that were not represented by absorbance data, (2) dependence of photochemical quantum yield on DOM quality and (3) hydrochemical influences on reaction rates, e.g., iron catalyzed photooxidation of DOM and impact of pH and ionic strength on both intermolecular interactions and molecular configuration.

We used a multivariate projection method (PLS) in an effort to correlate the variance in photochemical response parameters normalized for absorbed radiation energy to (1)–(3) above. DOC was used as a measure of the total available substrate for the measured photoreactions (issue [1] above), and Chl *a* concentration and humic substance fluorescence were indicators of the origin and quality of DOM (autoch-

thonous vs. allochthonous [2]). Alkalinity, conductivity, and pH were included since these parameters indicate major differences in water chemistry that could influence photoreaction rates as well as the structure and speciation of the organic matter (3) (Miles and Brezonik 1981; Reche et al. 1999).

All of these parameters had a significant impact on one or both of the two latent variables in the PLS model (LV1 and LV2; Fig. 3). The model was able to explain 45% of the residual variance in DIC production efficiency (Fig. 4A), while it was inadequate for explaining the variance in carboxylic acid production efficiency (Fig. 4B). Hence, it appears that the complete oxidation of DOC to DIC, but not the partial degradation into carboxylic acids, is affected by the measured parameters. Only total iron content, and to some extent absorbance and humic substance fluorescence (indicators of highly humic waters), were positively related to energy-normalized DIC production. We suggest that a high concentration of iron in the water promotes a more complete oxidation of DOM to DIC. Iron both absorbs UV-radiation and is a catalyst for photochemical humic substance oxidation (Stumm and Morgan 1995). Thus, high iron concentration may result in an increased quantum yield for photochemical DIC production through a facilitated, iron-redox-cycle catalyzed oxidation reaction. Such a catalytic effect has previously been observed for natural humic waters, where photochemical oxygen consumption (Miles and Brezonik 1981) and DIC-production rate (Gao and Zepp 1998), is positively related to free Fe(III) concentration. The elevated photochemical production of DIC after normalizing for absorbed energy, which we observed in waters with a high content of total iron, was accompanied by a similar, enhanced photochemical bleaching efficiency at 250 nm (Fig. 2A,C). This response may be a mixed effect of degradation of chromophoric organic matter and photoreduction of colored Fe(III) to transparent Fe(II).

Several parameters, including pH, alkalinity, conductivity, and chlorophyll, were negatively correlated to energy-normalized DIC production. This pattern is indicative of a less efficient DIC production in highly productive lakes. Hence, our results suggest that the DOM is slightly less photochemically reactive in such waters, possibly due to the larger component of DOC that is likely to originate from indigenous primary production. Alternatively, this feature of the model could be a direct consequence of ionic-strength or pH-induced changes in the solution chemistry of iron and organic matter in the studied waters. This explanation is in accordance with results by Gao and Zepp (1998), who found that the photoproduction of DIC in a river water sample increased significantly with decreasing pH.

The total production of carboxylic acids carbon (sum of the four analyzed acids) was on average 34.4% of the total DIC production in the studied waters. This is similar to what we observed previously for a number of oligotrophic waters of varying humic content using similar light sources (on average 25%; Bertilsson et al. 1999) and during a seasonal study (on average 30%; Lindell et al. 1999). It is also close to the relationship between photoproduction of DIC and carboxylic acids that we found in water from humic lake Skärshultsjön in southern Sweden that was exposed to natural

sunlight. In early July 1995, Granéli et al. (1998) found an average photoproduction of $63 \mu\text{g DIC L}^{-1} \text{h}^{-1}$ (6 h of solar irradiation, approximate UV-A irradiance 520 kJ m^{-2}), while Bertilsson and Tranvik (1998) found an average photoproduction of $19 \mu\text{g}$ carboxylic acid carbon $\text{L}^{-1} \text{h}^{-1}$ (8 h of solar irradiation, approximate UV-A irradiance 710 kJ m^{-2}). Taken together, these studies indicate that carboxylic acid carbon is photochemically produced in the surface layer at a rate that corresponds to 30% of the rate of DIC photoproduction. For comparison, carbon monoxide has been estimated to be photochemically produced in natural waters at rates representing 5–10% of concurrent DIC production (Miller and Zepp 1995; Miller and Moran 1997). The total pool of photochemically produced DIC and formic, acetic, malonic, and oxalic acid during the 8 h experimental exposures ranged from 0.2 to 11.1% of the total DOC (mean 4.7%). The formation of such photoproducts indicates either a direct removal of carbon from the DOM pool (DIC production) or an indirect removal, since the studied carboxylic acids are efficiently mineralized into CO_2 or incorporated into bacterial biomass (Bertilsson and Tranvik 1998).

Absorption of radiation explained most of the observed variance in reaction rates. After normalizing for absorbed energy, high concentrations of iron, and high UV absorption were positively correlated to photochemical DIC production. In contrast, pH, alkalinity, conductivity, and Chl *a* in the water were negatively correlated to photochemical DIC production after normalizing for absorbed energy. Taken together, these relationships suggest that the DOC is more easily photomineralized in oligotrophic humic lakes than in eutrophic lakes with high algal production. In addition, the results suggest that both inherent properties of the DOC, as indicated by UV absorbance, and extrinsic attributes of the lake water (pH, alkalinity, conductivity, iron content) are related to the complete photooxidation of DOC into DIC. The corresponding correlations were not detected for the production of carboxylic acids. Hence, the studied parameters appear to affect the complete photochemical mineralization of DOM, but not the production of organic low molecular weight compounds. We suggest that photochemical production of both DIC and low molecular weight carboxylic acids in lakes is a ubiquitous phenomenon. Based on our results obtained from temperate surface waters representing a wide range in productivity and DOM quality, we conclude that the photochemical effects of absorption of solar radiation by DOM depend on attributes of the DOM itself, but also on the inorganic matter dissolved in the water.

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