Accessory publication

Spatial and temporal variations and factors controlling the concentrations of hydrogen peroxide and organic peroxides in rivers

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Chemicals

To examine the peroxide production in water from these rivers, we used Suwannee River Fulvic Acid (SRFA) (ref. No 1S101H), Suwannee River Humic Acid (SRHA) (ref No 1S101F) (International Humic Substances Society, USA), tryptophan (Nakalai Tesque Inc., Kyoto, Japan), FWAs standards, such as distyryl biphenyl, DSBP: 4,4′-bis[(2-sulfostyryl)biphenyl (Tinopal CBS-X, LOT: 112013R2EY) and diaminostilbene type, DAS1: 4,4′-bis[(4-anilino-6-morphilino-s-triazine-2-yl)amino] 2,2′-stilbenedisulfonate (Tinopal AMS-GX, LOT 001288BOEK), 2-sulfonic acid benzaldehyde (2SAB) and 4-biphenyl carboxaldehyde (4BCA) (Kanto Chemicals company, Japan). One milligram per litre of each standard solution was prepared by dissolving into MQ water. 30% H_2O_2 (Wako Chemical Ltd, Japan) and peracetic acid (Aldrich, Japan) were used as the standards for H_2O_2 and ROOH respectively. Catalase and peroxidase were purchased from Sigma, Japan. All the chemicals were of analytical grade.

Experimental design

The irradiation experiment was conducted using a solar simulator (Oriel, Model 81160-1000) equipped with the 150 W Xenon lamp (Ozone free, Oriel Model 81160) and special glass filters restricting the transmission of wavelengths below 300 nm. Few experiments were conducted with the more intense radiation using the 300 W Xenon lamp by replacing the previous one. The light intensity of the lamp was calculated by measuring the degradation rate of a 8-μM standard 2-nitrobenzaldehyde (2-NB) solution in a 60 mL quartz cell. The degradation rates of 2-NB for a Xenon lamp employed in this study were in the range of 0.00196–0.00214 s^{-1} and 0.00331–0.00338 s^{-1} whilst the degradation rate for natural sunlight on 6 July 2004 at Hiroshima University Campus (at noon under clear sky conditions) was 0.00783 s^{-1}. To examine

photoproduction potential of H_2O_2 and ROOH, each FDOM standard solution (1, 3 or 5 mg L^{-1} in Milli-Q water) and river samples were prepared for light irradiation experiments. The exposure time was 10 h for 1 mg L^{-1} samples and 1 h for 3 and 5 mg L^{-1} samples. All river samples were pre-filtered and were exposed to total irradiation period of 10 h with aliquots taken for peroxide determination at 0, 30, 60, 180, 360 and 600 min. The amounts of H_2O_2 and ROOH in the standard solutions and samples from the rivers were normalised as a function of natural sunlight using the following equation (Eqn 5):

$$R_{(H_2O_2, Is)} = \frac{D_{(2-NB, Is)} R_{(H_2O_2, Ixe)}}{D_{(2-NB, Ixe)}} \quad (A1)$$

where $R_{(H_2O_2, Is)}$ is the rate of H_2O_2 production corrected for the intensity of natural sunlight (at noon under clear sky conditions on 6 July 2004 at Hiroshima University Campus) in water samples from the river and standard DOM materials, $D_{(2-NB, Is)}$ and $D_{(2-NB, Ixe)}$ are the degradation rates of 2-NB estimated using the intensity of natural sunlight and the Xe lamp respectively and $R_{(H_2O_2, Ixe)}$ is the observed H_2O_2 production rate produced under the conditions of Xe lamp.

Other analytical methods

Dissolved organic carbon (DOC) concentration in water samples was measured using a high temperature catalytic oxidation method (TOC 5000A, Shimadzu, Kyoto, Japan). The standard potassium hydrogen phthalate was used as a reference organic substance to determine DOC concentration. After removing dissolved inorganic carbon (DIC) by bubbling pure air, 106 μL of each sample was injected into TOC analyser. DOC measurements were conducted for each sample 3 to 5 times under conditions of <2% coefficient of variance or with the standard deviation being the area counted for <200 (equivalent to 1.3 μM C). Triplicate measurements were performed for each sample. The three-dimensional (3-D) excitation emission matrix (EEM) spectra of water samples were obtained using a fluorescence spectrophotometer (F-4500, Hitachi, Japan). The EEM spectra were constructed by scanning emission spectra from 225 to 500 nm as a function of excitation wavelength from 225 to 400 nm. Readings were collected at intervals of 5 nm for excitation with 1 nm emission wavelengths using a scanning speed of 1200 nm min^{-1}. The wavelength accuracy was within ± 2 nm. The fluorescence spectra were measured in triplicate for each sample and were averaged. Fluorescence readings (peaks C, W and T) were calibrated using the fluorescence intensity (Ex/Em = 350/450 nm) of a quinine sulfate standard. Quinine sulfate solution (4 μg L^{-1}) was prepared in 0.01 N H_2SO_4 for fluorescence measurements. The fluorescence intensity (FI) for 1 μg L^{-1} of quinine sulfate solution was equal to 1 QSU (quinine sulfate unit) in this study. The concentrations of the NO_3^-

and NO_2^- ions were determined using a suppressor type ion chromatograph with the column Ion Pac AS11 (Yokogawa Analytical Systems, IC-7000II and Dionex, DX500). Dissolved Fe^{II} and total Fe content were measured using the 1,10-phenanthroline method. In this method, Fe^{III} was estimated as the difference between Fe^{II} and total Fe after reduction of Fe^{III} by 5% hydroxyl amine. Ferrous ammonium sulfate was used as a standard. The river and standard samples were processed followed by the earlier method.[1] The absorbance of the samples then measured at wavelength ranges of 450–550 nm using UV-VIS Spectrophotometer (Shimadzu UV-2401, Shimadzu, Japan). The maximum absorbance at a specific wavelength was used for determination of Fe^{II} and total Fe concentrations in samples. The pH was measured using a portable pH meter (Horiba, Japan). The solar intensity (SI) was measured using a pyranometer (MS62, Eikoseiki Inc., Japan) located on the roof of the Faculty of Integrated Arts and Science in Hiroshima University (HU), in proximity to KR5 site. The data from this instrument provided meteorological data for the Kurose river. SI data at Misasa Primary School, Nishi-ku Hiroshima City (Air Pollution Monitoring Center, Hiroshima prefecture, Japan), located close to OR6 site, was used for the Ohta river meteorological data.

H_2O_2 photoproduction rates and source contribution

The rate of production of H_2O_2 in irradiated standard DOM solution and in water samples from the river was determined from the net production of H_2O_2 (final concentration minus initial concentration) measured for the initial 60 min of the irradiation period. The rate of generating H_2O_2 was then normalised to sunlight intensity at noon under clear sky conditions on 6 July 2004 at Hiroshima University Campus.[2] The normalised rate of production of H_2O_2 of an identified fluorescent substance is estimated on the basis of its fluorescence intensity observed in natural waters and can be determined using the following equation:

$$R_{Fi(river)} = \frac{FI_{Fi(river)}R_{RS}}{FI_{RS}} \quad \text{(A2)}$$

where $R_{Fi\,(river)}$ is the normalised production rate of H_2O_2 of an identified fluorescent substance in natural waters, $FI_{Fi\,(river)}$ is the fluorescence intensity of the identified fluorescent substance in natural waters, FI_{RS} is the fluorescence intensity of the relevant standard substance in the aqueous solutions, and R_{RS} is the normalised production rate of H_2O_2 of the relevant standard substance in the aqueous solution. Finally, percentages of each identified fluorescent substance contributing to the rate of production of H_2O_2 are calculated using the following equation:

$$F_{i(river)} = \frac{R_{Fi(river)} \times 100}{R_{net(river)}} \quad \text{(A3)}$$

where $F_{i(river)}$ is the contribution percentage of the normalised net H_2O_2 production rate in the water (%) for each identified fluorescent substance, $R_{Fi(river)}$ is the normalised H_2O_2 production rate generated by each identified fluorescent substance in water from these rivers, and $R_{net(river)}$ is the normalised net H_2O_2 production rate of all FDOM in water from these rivers. The percent contributions of unknown sources of H_2O_2 in water samples from the river were estimated using a simple formula: $F_{unknown} = 100 - (F_{FA} + F_{Tryptophan} + F_{FWAs})$, where the sum of the normalised H_2O_2 production rate of FA-like substances, tryptophan-like substances and FWA-like substances is subtracted from the normalised net H_2O_2 production rate of 100%.

Statistical analysis

Statistical analysis of the data obtained for peroxide measurements and other relevant analytical data was conducted using a SPSS program (SPSS Inc., USA). Significances of the differences in average values among seasonal peroxide concentrations were evaluated by one-way ANOVA and Fisher's l.s.d. analysis ($P < 0.05$). The Pearson correlation coefficients between peroxides concentrations and other water quality variables were estimated using the same program.

PARAFAC modelling

Recently, parallel factor (PARAFAC) analysis has effectively applied on EEM data to isolate the different components of DOM compositions into the organic compounds.[3] The PARAFAC model was performed in MATLAB using the 'N-way toolbox for MATLAB ver. 3.1' with methods described in earlier studies.[3] The data EEMs of the samples were modelled with excitation wavelength ranging from 220 to 380 nm by every 5 nm and emission wavelength from 280 to 480 nm by every 1 nm in this study. Milli-Q water blank was subtracted from every sample before running in the PARAFAC model. PARAFAC is a three-way multivariate method that can be applied on mathematical data of sample's EEM, which is capable of isolating the specific organic components in DOM compositions in aqueous media and then quantifying those organic components. PARAFAC can often identify the major fluorescent components in DOM compositions, but cannot isolate the minor fluorescent components in natural waters.[3]

References

[1]   Y. Zuo, Kinetics of photochemical/chemical cycling of iron coupled with organic substances in cloud and fog droplets. *Geochim Cosmochem Acta* **1995**, *59*, 3123.

[2]   K. Takeda, H. Takedoi, S. Yamaji, K. Ohta, H. Sakugawa, Determination of hydroxyl radical photoproduction rates in natural waters. *Anal. Sci.* **2004**, *20,* 153.

[3]   C. A. Stedmon, S. Markager, R. Bro, Tracing dissolved organic matter in aquatic environments using a new approach to fluorescence spectroscopy. *Mar. Chem.* **2003**, *82*, 239.
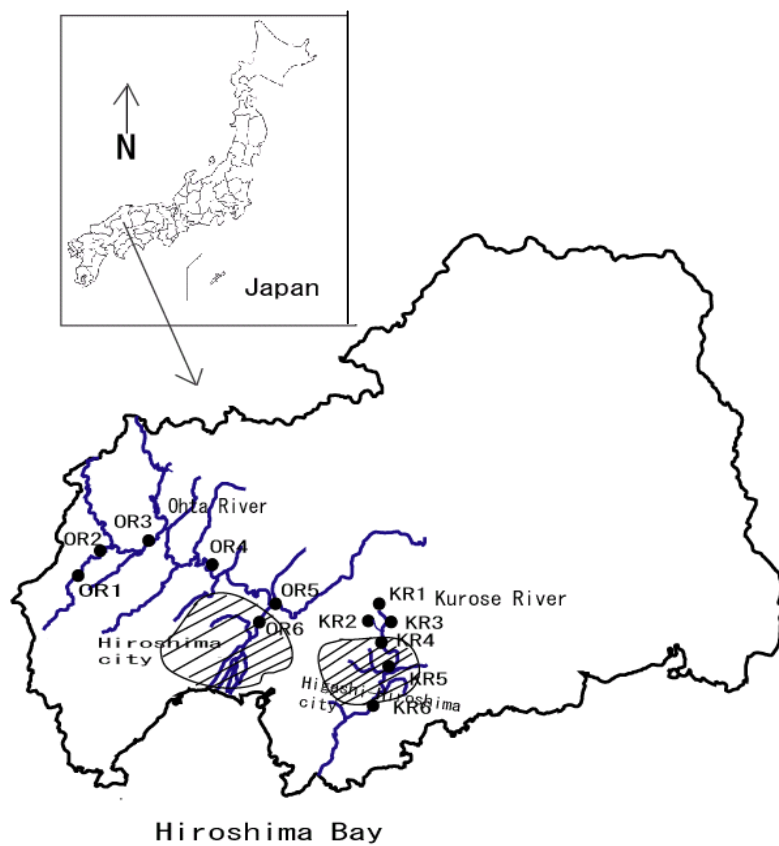
Fig. A1.   Water sampling sites in the Kurose River (sites KR1 to KR6) and the Ohta River (sites OR1 to OR6) in Hiroshima prefecture, Japan. Oblique lined areas indicate urban areas, Hiroshima or Higashi-Hiroshima.
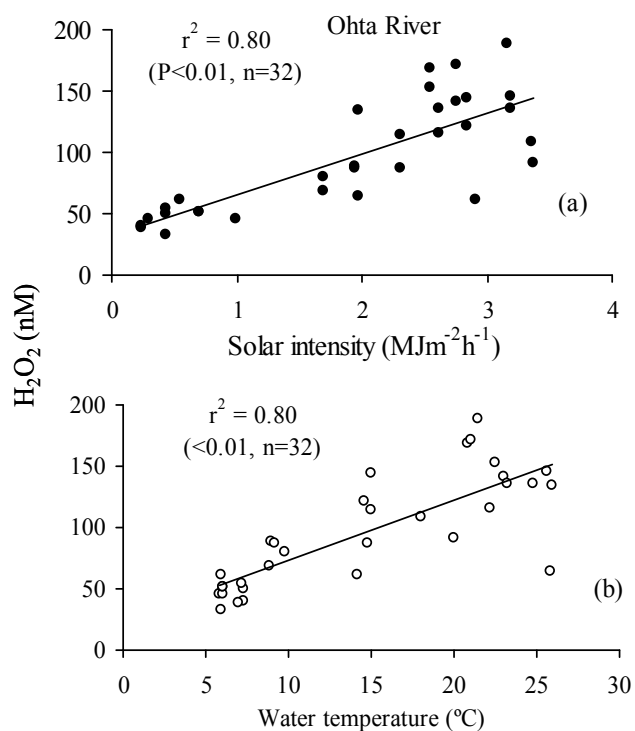
Fig. A2. Relationship between hydrogen peroxide and solar intensity estimated as MJ m^{-2} h^{-2} (Fig. 3a) or water temperature (Fig. 3b) in the waters of Ohta River.

a. DSBP: unirradiated sample

b. DSBP: 20 h of irradiated sample
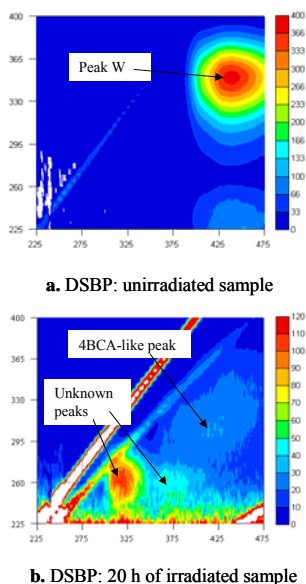
Fig. A3. The EEM of the standard DSBP before irradiation (a) and after 20-h irradiation using solar simulator (b). Part (b) shows a fluorescence peak for 4BCA-like substances and also unknown peaks in course of decomposition of DSBP.
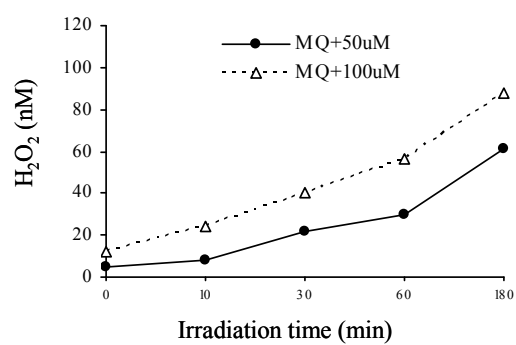
Fig. A4. Production of H_2O_2 as a result of light irradiation on the aqueous solutions of NO_2^- using solar simulator.

Table A1. Monthly variations of pH, water temperature (WT), solar intensity (SI), dissolved organic carbon (DOC), fluorescence peak intensity (FI) of fulvic acid-like substances (peak C), fluorescent whitening agents (peak W) and tryptophan-like substances (peak T), total Fe, Fe^{2+}, NO_3^-, NO_2^- H_2O_2 and ROOH at six smpling sites in the Kurose and the Ohta river waters in Hiroshima prefecture, Japan.

Sampling site and time	pH	WT	SI	DOC	FI Peaks (C or W)	Peak T	total Fe	Fe^{2+}	NO_3^-	NO_2^-	H_2O_2	ROOH
		(°C)	($MJm^{-2}h^{-1}$)	(µM C)	(QSU)			(µM)			(nM)	
Namitakiji (KR1)												
May 2002.	5.9	15.8	2.88	116±2.5	27	np	nd	nd	nd	nd	14±1.1	73±6.2
June 2002.	6.0	15.1	2.38	104±1.5	55	39	nd	nd	nd	nd	17±9.5	59±12.6
July 2002.	7.1	25.8	1.33	nd	68	67	nd	nd	nd	nd	21±4.2	44±8.4
August 2002.	7.4	25.4	1.12	146±3.9	55	np	54	nd	1.6	bd	33±9.8	nd
September 2002.	7.2	18.0	1.91	105±3.5	54	np	2	nd	8.8	bd	26±3.9	17±4.0
October 2002.	7.2	10.5	1.84	77±1.7	48	np	40	nd	5.5	bd	9±1.1	21±3.1
November 2002.	7.1	7.0	1.01	105±5.5	50	np	54	2.5	4.4	bd	6±0.6	25±2.0
December 2002.	6.9	6.0	0.76	87±6.4	52	np	20	0.2	6.4	bd	15±1.0	29±5.1
January 2003.	7.1	3.0	0.50	115±8.2	39	np	36	1.2	7.5	bd	11±0.5	9±1.0
February 2003.	7.3	5.6	1.01	47*	41	np	28	0.7	4.6	bd	9±1.1	20±5.8
March 2003.	7.0	8.7	1.91	51±3.9	51	np	8	2.8	14.2	bd	20±2.2	11±1.7
April 2003.	7.3	15.5	2.20	117±4.6	61	76	60	0.8	4.0	bd	16±4.9	30±12.6
December 2004.	8.0	9.0	0.53	nd	42	np	nd	nd	nd	bd	6±1.5	47±1.8
Mean	7.0	12.7±7.3	1.57±0.72	97±31	50±11	61±19	34±21	1±1	6±3		16±8	31±20
Shouriki (KR2)		12.7										
May 2002.	7.2	15.6	2.88	125±2.3	nd	np	nd	nd	nd	nd	29±1.6	41±1.8
June 2002.	7.1	16.1	2.38	88±5.9	59	np	nd	nd	nd	nd	68±9.0	65±5.3
July 2002.	7.1	22.3	1.33	nd	62	75	nd	nd	nd	nd	27±2.8	30±1.3
August 2002.	7.6	22.0	1.12	75±7.9	58	np	64	nd	7.4	bd	50±3.6	nd
September 2002.	7.3	19.0	1.91	66±2.0	45	np	20	nd	7.0	bd	37±1.4	19±0.6
October 2002.	7.7	11.5	1.84	124±2.5	39	np	48	nd	5.1	bd	19±1.6	17±2.4
November 2002.	7.7	7.5	0.76	146±2.6	44	34	48	2	4.3	bd	18±6.1	26±3.1
December 2002.	7.1	5.8	0.76	78±25	43	np	nd	nd	5.1	bd	21±3.3	35±7.0
January 2003.	7.3	2.0	0.50	53	32	19	10	1.3	7.7	bd	16±1.5	11±1.8
February 2003.	7.3	5.6	1.01	48±3	34	37	28	0.5	8.0	bd	21±7.7	26±7.1
March 2003.	7.2	8.9	1.91	43±3.9	40	np	40	3.2	8.7	bd	39±9.7	12±7.5
April 2003.	7.1	12.9	2.20	49±9.4	68	54	96.7	8.5	9.8	bd	19±3.4	22±10.9
December 2004.	7.5	9.0	0.53	nd	27	np	nd	nd	nd	bd	31±8.0	41±9.2
Mean	7.3	12.2±6.5	1.57±0.72	81±36	48±12	44±22	44±28	3±3	7±2		30±16	28±15
Sasa (KR3)												
June 2002.	6.5	18.0	2.74	123±7.8	145	np	nd	nd	nd	nd	49±8.6	66±9.4
December 2002.	7.3	7.0	0.97	143±9.5	117	np	6	0.0	28.7	nd	18±3.7	15±5.4
Tokumasa (KR4)												
June 2002.	7.3	19.5	2.27	154±1.3	245	86	nd	nd	nd	nd	29±3.1	28±0.7
December 2002.	7.2	7.0	1.22	146±25.8	380	93	30	0.0	71.4	1.7	31±2.4	9±2.7
Izumi (KR5)												
May 2002.	7.1	22.9	2.12	383±8.8	608	np	nd	nd	nd	nd	142±2.0	25±2.4
June 2002.	7.1	18.0	2.27	344±3.6	581	np	nd	nd	nd	nd	62±7.7	39±9.6
July 2002.	7.2	26.5	1.91	nd	666	np	nd	nd	nd	nd	76±6.6	32±11.1
August 2002.	7.6	27.2	0.94	349±0.7	659	np	176	nd	58.7	5.1	91±7.9	nd
September 2002.	7.4	22.2	2.16	215±2.1	606	207	88	nd	107.4	8.0	135±4.2	7±1.2
October 2002.	7.7	13.8	2.66	269±10.0	494	np	80	nd	84.3	6.1	77±1.2	1±1.0
November 2002.	7.4	8.2	1.3	244±6.0	548	182	60	3.2	131.9	4.7	16±2.5	14±1.1
December 2002.	8.4	7.0	1.22	319	589	np	180	0	110.9	1.4	14±3.2	3±1.7
January 2003.	7.2	2.0	0.76	212±23.9	504	240	158	6.5	127.9	2.5	9±1.5	3*
February 2003.	7.2	7.5	1.08	191±19.1	469	229	156	5.8	112.4	3.3	10±7.4	0
March 2003.	7.2	12.8	2.41	212±23.1	487	np	140	7.3	63.5	3.6	47±6.5	3±2.7
April 2003.	7.3	16.4	3.13	238±4.1	454	np	210	4.7	71.8	4.1	66±2.4	1±0.9
December 2004.	7.4	10.6	0.53	nd	263	np					47±3.2	5±3.5
Mean	7.4	15.0±8.0	1.83±0.75	271±67	555±73	215±25	139±51	5±3	97±28	4±2	62±46	12±14

Table A2. Diurnal variations of pH, water temperature (WT), air temperature (AT), solar intensity (SI) and H_2O_2 in the

upstream and downstream waters of the Kurose river.

Sampling	Upstream waters (site KR2)					Downstream waters (site KR6)				
	pH	WT (°C)	AT	SI (MJm^{-2})	H_2O_2 (nM)	pH	WT (°C)	AT	SI (MJm^{-2})	H_2O_2 (nM)
5:30 a.m.	7.0	19.2	22.5	0.00	9±1.3	7.1	19.5	18.5	0.00	4±1.3
8:00 a.m.	7.5	19.2	22.5	0.68	14±2.5	7.1	20.2	22	0.50	16±1.5
10:00 a.m.	7.4	20.0	24.8	2.09	23±1.9	7.0	22.0	19	2.16	31±4.5
11:00 a.m.	7.4	20.5	28.7	1.91	37±2.2	7.2	22.0	30.9	2.38	59±3.8
12:00 p.m.	7.4	20.8	28.5	2.74	43±4.1	7.1	23.0	30.5	2.52	63±3.1
13:00 p.m.	7.6	21.0	28.0	2.66	40±2.6	7.2	24.0	29.5	2.84	69±2.7
14:00 p.m.	7.5	21.0	28.0	1.80	34±4.5	7.2	24.5	29.5	2.63	69±5.5
15:00 p.m.	7.4	21.0	26.5	1.33	113±1.7	7.3	25.0	29	2.30	62±7.2
17:00 p.m.	7.5	21.0	26.5	1.30	27±2.6	7.3	24.5	27	1.04	62±12.5
19:00 p.m.	7.2	20.5	25.0	0.00	9±1.3	7.2	21.5	23.5	0.00	20±2.7