SUPPORTING INFORMATION

Corroboration of method for estimating the photochemical attenuation of pharmaceuticals in river water under field conditions over 2 years and evaluation of toxicity changes under sunlight

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Information on WWTPs

Information on WWTPs T and K and their effluent quality is summarized in Tables S1 and S2. WWTP T treats wastewater by an anaerobic–anoxic–oxic process followed by chlorination, while WWTP K uses multistage nitrification–denitrification followed by ozonation.

Table S1. Details of the wastewater treatment plants on the River Nishitakase.

	treatment process[a]	disinfection process (chlorine/ozone dose)[a,b]	volume of treatment (m^3/d)[a,b]	travel time to Tenjin Bridge (h)[c]
WWTP T	Anaerobic–anoxic–oxic process	chlorination (0.8 mg Cl_2/L)	119,190	1.0
WWTP K	Multistage nitrification–denitrification	ozonation (4.3 mg O_3/L)	64,640	2.9

[a] reference *1*, [b] annual average value, [c] average value in dry weather

Table S2. Quality of effluents from wastewater treatment plants on the River Nishitakase.[a, b]

	Temperature (℃)	pH	BOD (mg/L)	COD_{Mn} (mg/L)	SS (mg/L)	DO (mg/L)	T-N (mg/L)	NH_4-N (mg/L)	NO_2-N (mg/L)	NO_3-N (mg/L)	T-P (mg/L)
WWTP T	21.4	7.0	3.1	6.6	2	6.7	7.8	0.3	N.D.[c]	6.6	0.5
WWTP K	21.7	6.7	3.8	6.7	2	17	6.6	0.3	N.D.[c]	5.9	0.6

[a] reference *1*, [b] annual average value, [c] not detected

Detection, Concentrations, and Source Distributions of PPCPs along the River Stretch

The measured concentrations and frequencies of detection of the PPCPs at each sampling site are shown in Table S3. The source distributions of the 28 PPCPs detected consistently at more than one of the sources were calculated using median values (Figure S1). WWTP T contributed most, mainly because it uses chlorination, whereas WWTP K uses ozonation.[2]

Table S3. Measured concentrations and frequencies of detection of PPCPs at each sampling site.

	Site 1 (n=49)				Site 2 (n=49)				Site 3 (n=49)			
	Concentration (ng/L)			Freq.[a]	Concentration (ng/L)			Freq.[a]	Concentration (ng/L)			Freq.[a]
	average	±	SD	(%)	average	±	SD	(%)	average	±	SD	(%)
Acetaminophen	3.7	±	3.9	84	19.9	±	26.6	92	16.4	±	20.4	88
Antipyrine	0.0	±	0.3	2	3.2	±	2.8	72	2.6	±	2.5	66
Atenolol[b]	20.0	±	15.5	94	77.9	±	31.9	100	57.4	±	22.7	100
Azithromycin[b]	8.3	±	10.6	68	156.7	±	54.5	100	101.9	±	35.5	100
Bezafibrate[b]	73.2	±	95.7	88	170.0	±	136.4	100	141.6	±	95.9	100
Caffeine[b]	65.4	±	273.4	100	145.2	±	423.9	100	123.7	±	243.5	100
Carbamazepine[b]	0.6	±	2.3	58	35.4	±	9.6	100	23.9	±	6.5	100
Ceftiofur	N.D.[c]			0	N.D.			0	N.D.			0
Chloramphenicol	0.8	±	2.0	21	1.2	±	3.1	18	0.4	±	1.3	11
Ciprofloxacin[b]	3.0	±	3.0	73	20.7	±	9.8	100	9.9	±	5.4	98
Clarithromycin[b]	42.6	±	71.9	100	506.9	±	167.0	100	347.2	±	115.7	100
Clenbuterol	0.0	±	0.2	4	0.0	±	0.2	6	0.1	±	0.3	4
Clofibric acid	5.8	±	4.0	94	19.5	±	8.5	98	15.8	±	7.0	98
Crotamiton[b]	16.2	±	47.9	98	576.5	±	247.7	100	389.6	±	158.3	100
Cyclophosphamide	1.6	±	2.1	66	6.7	±	4.4	92	4.8	±	2.9	90
DEET[b]	27.9	±	31.0	100	71.5	±	83.0	100	58.6	±	68.8	100
Diclofenac[b]	1.0	±	5.9	8	76.9	±	32.8	100	49.7	±	30.3	98
Diltiazem[b]	2.3	±	3.9	88	31.5	±	8.5	100	22.4	±	6.5	100
Dipyridamole	0.2	±	0.7	19	4.1	±	6.0	66	2.1	±	3.4	66
Disopyramide[b]	43.4	±	28.3	100	151.5	±	38.5	100	116.3	±	31.0	100
Enrofloxacin	1.2	±	2.7	33	1.9	±	3.2	41	1.4	±	2.4	40
Ethenzamide[b]	1.3	±	1.2	80	8.5	±	4.0	100	6.2	±	3.1	98
Fenoprofen	0.1	±	0.5	4	0.1	±	0.6	2	7.1	±	50.1	4
Furosemide[b]	1.5	±	5.3	14	142.1	±	54.5	100	62.2	±	37.3	96
Griseofulvin	0.3	±	1.0	8	0.9	±	2.1	20	0.7	±	1.7	20
Ifenprodil[b]	0.2	±	1.0	12	5.7	±	2.2	100	3.2	±	1.8	92
Indometacin[b]	2.3	±	5.6	34	65.3	±	18.6	100	43.7	±	16.0	100
Isopropylantipyrine	0.0	±	0.1	4	1.5	±	1.3	78	1.1	±	1.0	70
Ketoprofen[b]	55.3	±	38.5	100	177.3	±	67.0	100	37.2	±	39.9	98
Mefenamic acid[b]	0.6	±	1.5	40	24.5	±	11.9	100	16.6	±	10.0	100
Metoprolol[b]	1.3	±	1.4	66	8.3	±	2.4	100	5.9	±	2.0	100
Nalidixic acid[b]	2.1	±	1.7	82	5.1	±	2.3	100	3.8	±	2.0	98
Naproxen	0.2	±	1.0	6	2.0	±	3.1	38	1.4	±	2.3	34
Norfloxacin	14.1	±	90.2	23	7.4	±	6.3	76	3.3	±	4.4	52
Ofloxacin[b]	15.7	±	26.9	98	310.1	±	114.1	100	171.8	±	69.8	100
Oxytetracycline	0.3	±	1.2	8	0.3	±	0.9	18	0.6	±	1.6	24
Pirenzepine[b]	0.7	±	2.0	34	14.1	±	4.5	100	10.0	±	3.7	100
Primidone[b]	5.9	±	4.1	92	19.4	±	7.1	100	15.2	±	5.9	100
Propranolol	0.1	±	0.3	14	3.4	±	1.9	96	2.0	±	1.6	76
2_Quinoxalinecarboxylicacid	2.4	±	2.1	65	9.2	±	4.2	90	12.3	±	5.8	92
Roxithromycin[b]	4.7	±	6.0	84	79.3	±	17.9	100	56.2	±	14.9	100
Salbutamol	0.0	±	0.1	18	0.1	±	0.2	22	0.0	±	0.1	11
Sulfadimethoxine	0.1	±	0.4	19	3.8	±	3.4	96	3.2	±	3.4	96
Sulfadimidine	0.1	±	0.7	9	N.D.			0	0.0	±	0.2	2
Sulfamerazine	0.0	±	0.1	4	0.5	±	0.9	30	0.3	±	0.6	22
Sulfamethoxazole[b]	3.7	±	4.3	86	104.8	±	29.7	100	74.1	±	24.0	100
Sulfamonomethoxine	0.1	±	0.9	4	0.1	±	0.3	6	0.1	±	0.5	10
Sulfapyridine[b]	3.6	±	8.5	70	172.7	±	56.8	100	117.1	±	41.6	100
Sulfathiazole	0.0	±	0.2	2	0.3	±	1.6	8	0.2	±	1.3	4
Sulpiride[b]	72.6	±	100.7	90	670.9	±	224.3	100	480.8	±	130.8	100
Tetracycline	0.4	±	1.2	10	3.2	±	2.7	68	1.8	±	2.0	56
Theophylline	12.0	±	32.3	92	43.7	±	57.9	96	37.9	±	39.7	96
Thiamphenicol	0.5	±	2.5	4	0.4	±	3.1	2	1.0	±	5.6	4
Tiamulin	0.1	±	0.6	20	0.0	±	0.2	16	0.1	±	0.2	10
Trimethoprim[b]	1.2	±	4.4	40	70.1	±	26.0	100	46.8	±	19.2	100
Tylosin	0.5	±	1.3	20	0.5	±	1.9	14	0.4	±	1.8	16

[a] frequency of detection, [b] PPCPs which were detected consistently (i.e. Freq. = 100%) at more than one of the sources, [c] not detected

23

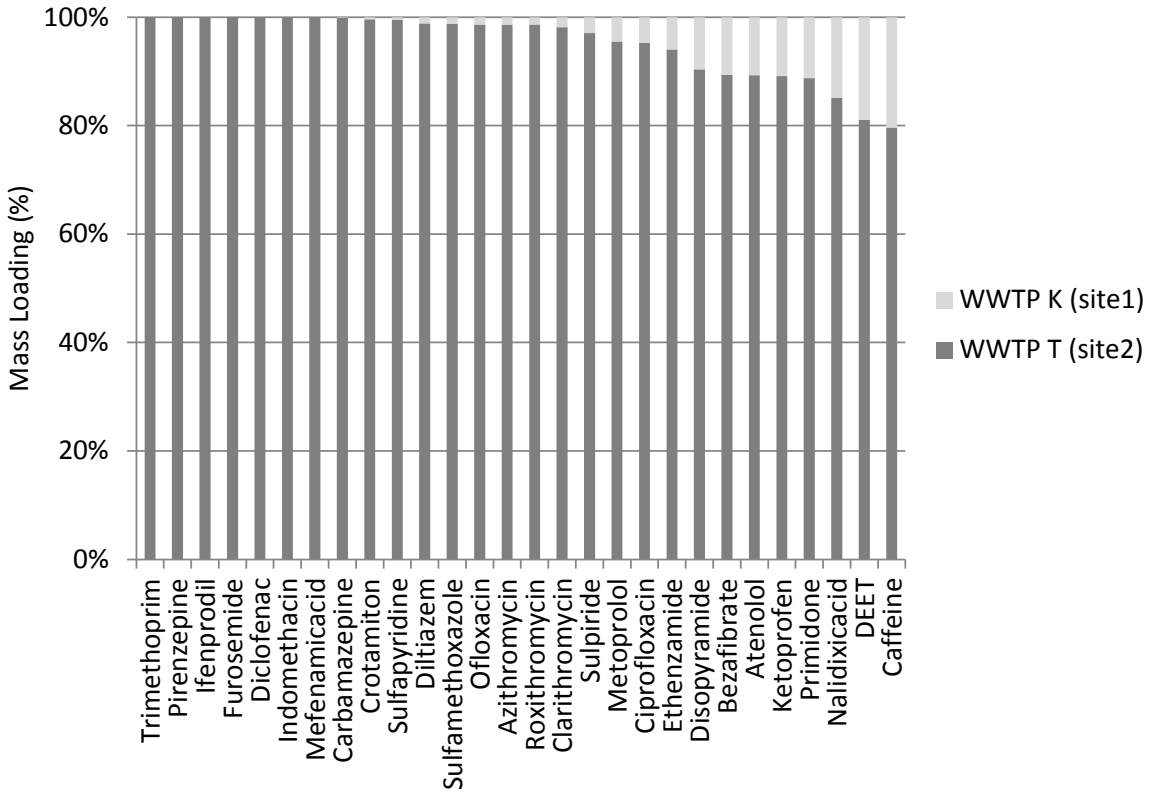

Figure S1. Source distribution of 28 PPCPs from each source at site 3. Median mass loadings were used for calculating the source distribution.

Effect of Water Temperature on Direct Photolysis

Ultrapure water was autoclaved and the pH was adjusted to 7.3 with phosphate buffer (6.67 mM). All 56 PPCPs were added to give an initial concentration of 50 μg/L each. The mixture (100 ml) was poured into a 100-ml beaker made of borosilicate glass and exposed to artificial sunlight (Ultra-Vitalux, 300 W, Osram, Munich, Germany) from directly above at around 1600 W/m^2. Water temperature was maintained at 10, 20, or 30 ± 1 °C during the experiment by a water circulator (CTP-300, Tokyo Rikakikai Co, Ltd., Tokyo, Japan). A 1-ml aliquot was collected, and concentrations of PPCPs were measured at 0, 5, 10, 15, 20, 30, 45, and 60 min after the start of exposure. The change in concentrations in darkness was negligible (data not shown). The first-order reaction constant and temperature-dependent factor obtained from the Arrhenius equation (eq. 1) are shown for PPCPs whose concentration change followed the first-order reaction ($R^2 > 0.90$) (Table S4). Although the photolysis rate constants of many PPCPs, especially quinolone and tetracycline antibiotics, were affected appreciably by water temperature, those of ketoprofen, diclofenac, furosemide, and naproxen were not.

$$k_T = k_{20} \times \theta^{T-20} \quad (1)$$

where k_T = first-order reaction constant at T °C (h^{-1}), k_{20} = first-order reaction constant at 20

°C (h^{-1}), θ = temperature-dependent factor (–), and T = temperature (°C).

Effect of pH on Direct Photolysis

Ultrapure water was autoclaved and the pH was adjusted to 5.8, 7.0, or 8.0 with phosphate buffer (100 mM). The other conditions were the same as above except that the water temperature was maintained only at 20 ± 1 °C. The first-order reaction constants are shown for PPCPs whose concentration change followed the first-order reaction ($R^2 > 0.90$) (Table S4). Although the photolysis rate constants of many PPCPs, especially quinolone and tetracycline antibiotics, were affected appreciably by pH, those of ketoprofen, diclofenac, chloramphenicol, and furosemide were not.

Table S4. Effects of water temperature and pH on direct photolysis rate constants of 14 PPCPs.

	first-order reaction constant (h^{-1})						ratio of first-order reaction constant (h^{-1})[a]						θ^b
	water temperature (°C)			pH			water temperature (°C)			pH			
PPCPs	10	20	30	5.8	7.0	8.0	20/10	30/20	30/10	7.0/5.8	8.0/7.0	8.0/5.8	
Ketoprofen	4.94	5.02	4.98	7.28	7.31	6.82	1.02	0.99	1.01	1.00	0.93	0.94	1.00
Enrofloxacin	1.95	2.82	3.36	1.65	2.81	2.91	1.45	1.19	1.73	1.71	1.03	1.77	1.03
Norfloxacin	0.97	1.67	2.50	0.53	0.85	1.28	1.72	1.50	2.58	1.60	1.51	2.40	1.05
Ciprofloxacin	0.87	1.52	2.34	-[c]	-	0.63	1.75	1.54	2.69	ND[d]	ND	ND	1.05
Diclofenac	1.26	1.45	1.55	1.69	1.91	1.70	1.15	1.07	1.23	1.13	0.89	1.01	1.01
Oxytetracycline	0.26	0.53	0.76	0.26	0.48	1.12	2.00	1.43	2.86	1.85	2.33	4.31	1.05
Chloramphenicol	0.51	0.78	0.84	0.90	1.00	1.08	1.54	1.09	1.67	1.11	1.08	1.20	1.03
Furosemide	0.70	0.85	1.01	0.97	0.77	0.75	1.21	1.18	1.43	0.80	0.97	0.77	1.02
Tetracycline	0.24	0.48	0.67	0.16	0.26	1.00	1.95	1.39	2.72	1.60	3.83	6.12	1.05
Ofloxacin	-	0.23	0.32	-	0.64	0.74	ND	1.41	ND	ND	1.16	ND	1.06
Propranolol	0.24	0.35	0.58	-	-	-	1.47	1.63	2.39	ND	ND	ND	1.04
Sulfathiazole	0.23	0.38	0.45	-	0.30	0.56	1.68	1.17	1.96	ND	1.88	ND	1.03
Ifenprodil	0.20	0.34	0.46	-	-	0.24	1.66	1.35	2.25	ND	ND	ND	1.04
Naproxen	0.27	0.32	0.40	0.26	0.19	-	1.18	1.26	1.49	0.74	ND	ND	1.02

[a] Value in red cell is within a range of 0.7-1.3, which means that photolysis rate constant does not depend on water temperature or pH so much. [b] temperature-dependent factor, [c] Concentration change during the experiment did not follow first-order reaction (i.e. R^2<0.90). [d] No data.

Indirect Photolysis

Surface water samples collected at Tenjin Bridge were brought to the laboratory, filtered through a membrane filter with a pore size of 0.45 μm (Toyo Roshi Kaisha Ltd., Tokyo, Japan), and pH adjusted to 7.3 with phosphate buffer solution (6.67 mM). The other conditions were the same as above except that the water temperature was maintained only at 20 ± 1 °C. The first-order reaction constants of the surface water samples were smaller than those of pure water for all PPCPs whose concentration change followed the first-order

reaction ($R^2 > 0.90$). Therefore, indirect photolysis was implied to be negligible in the attenuation of PPCPs in the river stretch.

Biodegradation

Surface water samples collected at Tenjin Bridge were brought to the laboratory. All 56 PPCPs were added to give an initial concentration of 1 μg/L each. The mixture was incubated at 25 ± 1 °C in the dark on a rotating shaker at 100 rpm. Samples autoclaved were incubated under the same conditions (control). Aliquots were collected from both sets, and concentrations of PPCPs were measured at 0, 1, 2, 3, 4, and 5 days after the start of incubation. The biodegradation rate constants were determined by subtracting the first-order reaction constant of the control from that of the unsterilized sample. The biodegradation rates in the river stretch were calculated in accordance with a first-order reaction from the travel time and the calculated biodegradation rate constants. The experiments were conducted 3 times in summer. The biodegradation rate constants were <0.10 day^{-1} on average for all PPCPs except dipyridamole (0.46 ± 0.05 day^{-1}). Consequently, the biodegradation rates in the river stretch were estimated as <2% on average for all PPCPs except dipyridamole. Therefore, biodegradation of the 15 PPCPs shown in Figure 2 was negligible in the river stretch.

Other Attenuation Factors

Because the values of Henry's law constant of the selected PPCPs are low (5.77×10^{-38} for tylosin to 1.53×10^{-7} for crotamiton),[3,4] the selected PPCPs had low volatility. The low first-order reaction constants obtained from the biodegradation experiment controls (<0.11 ± 0.03 day^{-1} for diltiazem) of the 15 PPCPs shown in Figure 2 suggest that the 15 PPCPs were insensitive to hydrolysis.

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