Distribution, seasonal pattern, flux and contamination source of pesticides and nonylphenol residues in Kurose River water, Higashi-Hiroshima, Japan

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Distributions, seasonal variation, flux and contamination source of pesticides and nonylphenol were examined during the period from April 2001 to January 2002 at 5 sites (Namatikiji, Tokumasa-Kami, Izumi, Ochiai and Hinotsume) in Kurose River water, Higashi-Hiroshima, Japan. Water samples were collected monthly and subjected to solid phase extraction followed by GC-MS analysis. Nineteen pesticide compounds were detected and isoprothiolane (37 ng/l), fenitrothion (35) and iprodione (34) showed the highest mean concentrations (as 5 site averages), while pencycuron (5), diazinon (6), tolclofos-methyl (7) and dithiopyr (8) were the compounds of lowest mean concentrations. Nonylphenol, which is a degradation product of a nonylphenolpolyethoxylates that are used in a wide range of industrial and household products, had an average concentration of 305 ng/l. At each of the five sites, its concentration was greater than individual pesticides. The concentration levels of pesticides and nonylphenol were highest at Izumi (the downtown site of Higashi-Hiroshima) and were lowest at Namatikiji and Tokumasa-Kami (upstream sites). Concentrations and fluxes of both compounds tended to be higher during spring to summer and low during fall to winter at most of the sites. The total pesticides concentration was poorly correlated with nonylphenol concentration. A statistical analysis of water quality data suggested that their main sources appear to be different such as agricultural activity for pesticides and waste water discharge for nonylphenol.

INTRODUCTION

Agricultural pesticides significantly improve crop yields and pesticides protect against pests. Japan is one of the main users of pesticides worldwide, with more than 450 active products distributed among 5400 commercial products (Japan Plant Protection Association, 2000). On the other hand, pesticides have become universal contaminants found in all segments of the environment and create many hazards to the environment and human health. Pesticide contamination in fresh water, which has emerged as an important environmental problem in the last few decades, is causing concern with respect to long-term and low-dose effects of pesticides on public health as well

as non-target species (Sudo *et al.*, 2002). Pesticides with different type such as insecticides including fenitrothion, diazinon, chlorpyrifos, isophenphos and pyridaphenthion, herbicides including simazine, propyzamide, pendimethalin and butamifos, and fungicides including flutolanil, mepronil, tolclofos-methyl, mepronil, iprodione, isoxathion, chlorothalonil, isoprothiolane, and pencycuron were considered to be common pesticides found in the environment, Japan (Numabe *et al.*, 1992; Odanaka *et al.*, 1994; Okumura and Nishikawa, 1995; Itagaki *et al.*, 2000; Kondoh *et al.*, 2001; Tanabe *et al.*, 2001; Sudo *et al.*, 2002). Some pesticides such as fenitrothion, iprodione, pendimethalin and simazine are also suspected as endocrine disrupters (Crisp *et al.*, 1998; Tamura

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et al., 2001).

Pesticides are primarily moved from agricultural fields to streams by surface runoff (Richards and Baker, 1993). The amount and kind of pesticides in water of a given area depends largely on the production intensity and kind of crops. However, the transport of pesticides out of their application area results in the presence and accumulation of these compounds in many parts of hydrosphere. For example, atmospheric precipitation is an important route of transport of pesticides, resulting in contamination of environmental waters far from agricultural areas (Majewski *et al.*, 2000). The amount of pesticides lost from fields and transported to streams depends on several factors, including soil characteristics, topography, weather, agricultural management practices and the chemical and environmental properties of individual pesticides (Leonard, 1990). Site selection is important for pesticides monitoring because the types of pesticides used are directly related to the land use and the type of agricultural system. Thus, understanding the relation between land use and pesticide use is the key to evaluating the causes of pesticides occurrence in streams (Larson *et al.*, 1999).

Nonylphenol polyethoxylates (NPEOs) is a major class of non-ionic surfactants used in household, paints, and pesticides and in industrial processes such as paper and petroleum production (Fairchild *et al.*, 1999). Nonylphenol is the most toxic and refractory metabolite of NPEOs and can cause several hazards to aquatic ecosystem (McLeese *et al.*, 1981; Ahel *et al.*, 1987). Pesticides are thought to be a secondary source of nonylphenol contamination in natural water because NPEOs are used as adjuvants in pesticide formulations (Sundaram *et al.*, 1980; Fairchild *et al.*, 1999; Scott and Jones, 2000; Thomas *et al.*, 2001). Nonylphenol is known to be an endocrine disrupter (Fairchild *et al.*, 1999; Scott and Jones, 2000). In order to assess the risk of nonylphenol, its presence and fate in the environment must be known (Mol *et al.*, 2000).

Water quality parameters may have a strong effect on the occurrence of pesticides in natural

water (Samia El-Kabbany *et al.*, 2000). Water temperature has a strong influence on the acclimation period of microorganisms and its biodegradation rate of NPEOs to nonylphenol; the biodegradation process takes 15–20 days at 7°C and 2–3 days at 21°C (Manzano *et al.*, 1999). The stability of pesticides in water is strongly dependent on pH and is highest in slightly acidic water (pH 4–6) (Petroff, 2000). Thus, because of these environmental factors, some heavily used compounds end up with low concentration while other slightly used compounds end up with high concentrations. Also dissolved organic carbon (DOC) may be an indicator of water pollution by organic compounds such as our target compounds (pesticides and nonylphenol) (Castilho *et al.*, 2000).

In this study, we attempted (1) to develop a practical method for monitoring pesticides and nonylphenol, (2) to evaluate the distribution and seasonal patterns of pesticides and nonylphenol occurrence in Kurose River, (3) to determine the discharging amount of these contaminants at each site in Kurose River, (4) and to elucidate the contamination sources of several common pesticides (pencycuron, simazine, propyzamide, diazinon, chlorothalonil, terbucarb, tolclofos-methyl, metalaxyl, bensulide, dithiopyr, fenitrothion, chloropyrifos, pendimethalin, methyldymron, captan, isophenphos, naporamide, butamifos, flutolanil, isoprothiolane, triclopyr-ester, isoxathion mepronil, iprodione and pyridaphenthion) and nonylphenol residues in Hiroshima prefecture.

MATERIALS AND METHODS

Chemicals

A mixture of 25 pesticide standard compounds (pencycuron, simazine, propyzamide, diazinon, chlorothalonil, terbucarb, tolclofos-methyl, metalaxyl, bensulide, dithiopyr, fenitrothion, chloropyrifos, pendimethalin, methyldymron, captan, isophenphos, naporamide, butamifos, flutolanil, isoprothiolane, triclopyr-ester, isoxathion mepronil, iprodione and pyridaphenthion) was obtained from Kanto Com-

pany, Japan. A nonylphenol, technical mixture of ring and chain isomers was obtained from Aldrich Company, U.S.A. Working standard solutions of these compounds were prepared by making appropriate dilutions in acetone and stored in a refrigerator at 4°C. Pesticide analytical grade solvents, such as methanol, acetone, and dichloromethane were obtained from Wako Company, Japan. The internal standard phenanthrene- d_{10} (97.5% purity) was obtained from Supelco Company U.S.A. Acetic acid (99.7% purity) was obtained from Katayama Company, Japan.

Sites and target compounds selection

Kurose River is located in Hiroshima prefecture, and is about 43 km long. It runs urban and agricultural areas of the Kamo plateau, including Higashi-Hiroshima City and Kurose Town, and flows into the Seto inland Sea. It has a water collection area of about 250 km². The runoff from agricultural fields and wastewater containing industrial and household pollutants flows into this River. Higashi-Hiroshima city is considered the granary of Hiroshima prefecture. For example, it has the largest rice production of 86 cities, towns and villages in the prefecture (Hiroshima Prefecture Office, 2002). Also there are many crops cultivated in the city such as wheat, bean potato, sweet potato and other vegetables as well as ornamental plants. In order to monitor the pesticides and nonylphenol levels in the present study, 5 sites (Namitakiji, Tokumasa-Kami, Izumi, Ochiai and Hinotsume) were established at Kurose River in Higashi-Hiroshima City as shown in Fig. 1. Sampling sites were selected in different agricultural and urban basins along the river. The Tokumasa-Kami, Ochiai and Hinotsume sites were located near agricultural areas especially near rice field and low resident population area. The Ochiai site is next to water treatment facility that serves the City of Higashi Hiroshima. The Izumi site is located near a high density residential and industrial area. The Namitakiji site is the most upstream site and is located away from agricultural and urban areas. Three classes of pesticides were investigated: insecticides, herbicides and fungicides.

Fig. 1. Locations of sampling sites in Kurose River.

Representatives of each class were selected based on amount of their use in Hiroshima prefecture from October 1999 to September 2000 (JPPA, 2000), and on their potential environmental significance. Monthly precipitation data in Higashi-Hiroshima City were obtained from Hiroshima University Meteorological Data Acquisition System.

Estimated annual use of pesticides in Higashi-Hiroshima was calculated by using the ratio of agricultural area in Higashi-Hiroshima (4560 ha.) to agricultural area of Hiroshima prefecture (65200 ha.), which data were taken from the report of Hiroshima prefecture office (2002) as well as the data of pesticides agricultural use in Hiroshima prefecture (JPPA, 2000). Terbutcarb was excluded for calculating the flux because no information about the annual use of this compound in Hiroshima prefecture.

Sampling and extraction method

Water samples were collected from the river at monthly intervals with 1 liter precleaned amber glass bottles. The samples were protected from exposure to light. They were kept at 4°C in ice cooler until delivered to analytical laboratory. Water flow rate was measured using a Hiroi-type water flow meter (Mikasa, Japan). At the sampling sites, conductivity and pH were measured with a D-24 pH/Conductivity meter, (Horiba, Ltd.) Japan. Water temperature was measured with a mercury thermometer. Water samples were filtered through glass fiber filters (GC-50, diameter: 47 mm; pore size: 0.5 μ m, Advantec). The glass fiber filters were ultrasonically extracted with acetone and combined with filtered samples. Samples were acidified with acetic acid to pH 3.0 to attaining a

high recovery of the target compounds.

Target compounds were extracted from the acidified samples using an automated solid phase extraction (SPE) apparatus according to the method of Itagaki *et al.* (2000) with some modifications. The cartridges (OASIS 60 mg Waters, Milford Ma, U.S.A.) were washed sequentially with 5 ml dichloromethane, 5 ml methanol and 5 ml Milli-Q water (Millipore. Co. Ltd. U.S.A.) under vacuum before use. A one-liter aliquot of the acidified samples was passed through the cartridges at a flow rate 10–20 ml/min and then the cartridges were washed with 10 ml Milli-Q water. Air was the sucked through the cartridges for about 30 min with a vacuum to remove the water. The target compounds were eluted with 5 ml dichloromethane. The solvent was evaporated

Table 1. Type, retention time, quantitation and confirmation ions for target compounds in SIM mode of GC-MS analysis

	Compound name	Type	Retention time (min)	Quantitation ion (m/z)	Confirmation ion (m/z)
1	Pencycuron	Fungicide	18.60	125	180
2	Simazine	Herbicide	19.45	201	186
3	Propyzamide	Herbicide	20.08	173	175
4	Diazinon	Insecticide	20.33	137	179
5	Chlorothalonil	Fungicide	20.66	266	264
6	Terbucarb	Herbicide	21.57	205	220
7	Tolclofos-methyl	Fungicide	21.72	265	267
8	Metalaxyl	Fungicide	21.94	206	132
9	Bensulide	Fungicide	21.94	77	131
10	Dithiopyr	Herbicide	22.23	354	306
11	Fenitrothion	Insecticide	22.32	125	109
12	Chloropyrifos	Insecticide	22.88	199	97
13	Pendimethalin	Herbicide	23.71	252	253
14	Methyldymron	Herbicide	23.87	107	106
15	Captan	Fungicide	23.91	79	149
16	Isophenphos	Insecticide	23.95	58	121
17	Napropamide	Herbicide	24.94	72	100
18	Butamifos	Herbicide	24.94	286	200
19	Flutolanil	Fungicide	24.98	173	145
20	Isoprothiolane	Fungicide	25.10	118	162
21	Triclopyr-ester	Herbicide	25.57	57	85
22	Isoxathion	Fungicide	25.71	105	177
23	Mepronil	Fungicide	26.56	119	91
24	Iprodione	Fungicide	28.10	314	316
25	Pyridaphenthion	Insecticide	28.23	97	77
26	Nonylphenol isomer-1	—	18.89	135	107
27	Nonylphenol isomer-2	—	19.00	135	107
28	Nonylphenol isomer-3	—	19.56	135	107

under a stream of nitrogen until almost complete dryness and the residue was re-dissolved in 1 ml acetone. An internal standard (0.5 $\mu\text{g/ml}$) was added into the solution and final volume was adjusted to 100 μl . Then 5 μl of the solution was analyzed by GC-MS.

GC-MS analysis

Samples were analyzed with a Hewlett-Packard HP 5973 series gas chromatograph equipped with HP 5973 mass selective detector, and an Agilent 6890 series injector was used for an injection of the sample. The column was a re-fused silica capillary HP-5MS chromatographic column (30 m long $\times$ 0.25 mm I.D., film thickness 0.25 μm). The oven temperature was programmed to hold for 2 min at 60°C and then increased to 280°C at the rate of 8°C min⁻¹ and hold at 280°C for 8 min. The injector and transfer line temperature were 250 and 280°C, respectively. The carrier gas (helium) was used a constant flow rate (1 ml/min). Standard and extracted solutions were injected in the splitless mode.

Target compounds were quantitatively determined in the selective ion mode (SIM) using the peak area ratio of target compounds to internal standard. The main ions of the target compounds in mass spectrometry are shown in Table 1. The total pesticide concentration was calculated by summing the concentration of individual compounds. The highest three isomer peaks of the nonylphenol mixture were selected for nonylphenol determination and the concentration of nonylphenol was calculated by summing the concentration of these isomers.

The target compounds were spiked into 200 ml Milli-Q water at levels of 1 $\mu\text{g/l}$ and 0.1 $\mu\text{g/l}$ for recovery and detection limit calculation, respectively. Another recovery test was carried out using 1 L spike volume at the same concentration level. Subsequently the water was acidified to pH 3.0 immediately after this operation; water samples were extracted with an automated solid phase extraction (SPE) apparatus. The detection limit was taken as concentration giving a signal-to-noise ratio of three, according to the method of Azevedo

et al. (2000). The test determining the recovery efficiency and the detection limit were repeated five times to obtain accurate values.

Water quality parameters

Water quality parameters, such as, DOC, anions and cations were analyzed at all the sampling sites. Anions (Cl^- , NO_2^- , NO_3^- and SO_4^{2-}) and cations (Na^+ , K^+ , NH_4^+ , Ca^{2+} and Mg^{2+}) were measured by ion chromatography (Dionex, DX500) with the column of Ion Pac AS11 for anions and Ion Pac CS12A for cations. DOC was measured by TOC 5000-A (Shimadzu Comp. Japan). DOC, anions and cations were measured after filtration of water samples.

Flux calculations

The flux is defined as the mass of the compound transported in stream in specific time period. The flux of target compounds at each sampling point was estimated as the product of the compound concentration and the site discharge (water flow rate) for each month. The monthly fluxes were summed to give the total flux over the study period according to the method of Larson *et al.* (1995). The Hiroshima prefecture office's continuous monitoring data of water flow rates collected at near Ochiai site was used for calculating the flux of individual pesticides.

Statistical analysis

A principle component analysis (PCA) was conducted with the SPSS Base 7.5 (SPSS Inc, U.S.A.) to identify underlying factors that explain the patterns of correlations within a set of observed variables and to generate hypotheses regarding to the sources of nonylphenol and pesticide contamination. The method of factors extraction was principle component and the method of factors rotation was the orthogonal varimax. Coefficient values lower than 0.5 were excluded.

RESULTS AND DISCUSSION

Water quality parameters

The main pH at all sampling sites ranged from

Table 2. Chemical composition of the Kurose River waters

Sampling site	Water temperature (°C)	pH	Conductivity (μS)	DOC (mg Cl)	NO ₂ ⁻ (μM)	NO ₃ ⁻ (μM)	Cl ⁻ (μM)	SO ₄ ²⁻ (μM)	Na ⁺ (μM)	NH ₄ ⁺ (μM)	K ⁺ (μM)	Mg ²⁺ (μM)	Ca ²⁺ (μM)
Namitakiji	19 (4.5–29)	7.00 (5.86–7.8)	23 (21–82)	2.24 (1.2–2.68)	ND	0.5 (0–4)	141 (119–197)	33 (13–31)	352 (178–1359)	35 (2.67–167)	31 (11–138)	33 (4–97)	74 (37–275)
Tokumasa-Kami	18 (6–24)	7.00 (5.54–8.39)	56 (64–66)	2.08 (1.18–2.45)	5 (2.56–9)	38 (36–39)	206 (170–254)	63 (33–83)	382 (300–4423)	28 (1–116)	39 (29–53)	44 (23–56)	127 (79–175)
Izumi	20 (6–26)	7.67 (5.36–7.95)	81 (78–86)	3.46 (2.43–4.86)	17 (5.46–27)	67 (49–81)	378 (222–509)	104 (88–123)	626 (473–783)	22 (1.5–57)	73 (45–107)	62 (44–100)	206 (145–266)
Ochiai	20 (8–27)	7.63 (5.5–8.34)	144 (48–179)	3.72 (2.64–4.98)	72 (8.28–173)	58 (30–89)	935 (844–1089)	239 (152–415)	1048 (183–1604)	177 (126–473)	70 (14–163)	70 (40–92)	296 (54–425)
Hinotsume	17 (5–27)	6.37 (5.55–7.5)	117 (106–141)	3.44 (2.17–5.87)	96 (9.26–240)	96 (75–143)	794 (575–1119)	231 (14–415)	911 (591–1310)	144 (28–368)	88 (70–137)	147 (42–683)	236 (48–422)

ND = Not detected.

Water quality data (n = 8) were obtained in the period of June 2001 to January 2002.

6.37 to 7.00 (Table 2), which is slightly higher than the ideal pH for pesticides stability in water (Petroff, 2000). Water temperature ranged from 19–29°C at all sites during the period from May to October 2001, which may accelerate biodegradation of NPEOs to nonylphenol (Manzano *et al.*, 1999). The Izumi, Ochiai and Hinotsume sites appeared to be the most heavily contaminated sites with high concentrations of DOC. They also had high concentrations of various anions (Cl⁻, NO₂⁻, NO₃⁻ and SO₄²⁻) and cations (Na⁺, K⁺, NH₄⁺, Ca²⁺ and Mg²⁺). Among cations and anions, Na⁺ and Cl⁻ were detected with higher concentration comparing with other anions and cations. NO₃⁻ and NO₂⁻, which are considered to be one of the main sources of OH radical in natural water (Mopper and Zhou, 1990), were detected with higher concentrations at these heavily contaminated sites. Therefore NO₃⁻ and NO₂⁻ may have roles the photodegradation of organic compounds and may affect their lifetimes and concentrations in the stream. NH₄⁺ concentrations were high at Ochiai and Hinotsume sites, apparently as a result of their use as fertilizers in agricultural fields. Conductivity was associated with anion and cation concentrations at all sites.

Efficiency of the method

All 25 authentic pesticides and nonylphenol isomers were separated by GC column and eluted in a reasonable time. Using solid phase extraction with a 200 ml spike volume, recoveries of the most authentic samples were in the range 70–107% (Table 3). Lower recoveries were obtained for mepronil and iprodione (63 and 61%, respectively). When the spike volume increased to 1 L, recoveries of most samples remained in the same range (70 to 109%) although the recoveries of pendimethalin and isophenphos dropped to 65 and 64%, respectively. Because the recoveries of all the targeted compounds were high, the recovery efficiencies were not used to correct the analytical concentrations.

The relative standard deviation was around 10% but it was always below 30% in accordance with the US Environmental Protection Agency

Table 3. Recovery percentage, standard deviation, relative standard deviation and limits of detection of target compounds in this study

Compound name	R (%)	SD (ng/l)	RSD (%)	DL (ng/l)
Pencycuron	82	10.6	3.5	5
Simazine	91	5.7	2.5	24
Propyzamide	82	4.6	4.2	12
Diazinon	85	7.0	6.8	2
Chlorothalonil	96	11.6	2.9	5
Terbucarb	83	5.6	4.7	20
Tolclofos-methyl	79	7.8	4.4	6
Metaxyl	70	6.9	4.5	20
Dithiopyr	77	6.4	9.1	9
Fenitrothion	101	14	9.8	21
Bensulide	97	20.0	1.9	2
Chloropyrifos	78	3.6	5.5	9
Pendimethalin	84	8.6	10.4	8
Methyldymron	94	17.4	2.6	6
Captan	70	4.9	3.5	9
Isophenphos	76	5.0	2.2	2
Napropamide	96	3.4	12.9	18
Butamifos	107	19.6	1.6	7
Flutolanil	83	3.4	5.7	4
Isoprothiolane	74	9.4	1.8	8
Triclopyr-ester	70	2.7	3.4	15
Isoxathion	76	4.9	3.1	9
Mepronil	61	4.7	3.1	12
Iprodione	63	3.7	3.0	30
Pyridaphenthion	70	6.7	4.8	9
NP-isomer1	82	3.5	2.1	10
NP-isomer2	84	15.5	9.1	18
NP-isomer3	97	15.6	8.1	15

R = Recovery; SD = Standard deviation; RSD = Relative standard deviation; DL = Detection limits.
Number of replicates = 5.

method (Azevedo *et al.*, 2000). Detection limits of the 25 pesticide compounds and nonylphenol ranged from 2 to 30 ng/l.

Pesticides

A total of nineteen pesticides were detected at the five sites (Fig. 2). Among the detected pesticides, isoprothiolane (44%), flutolanil (38%), fenitrothion (34%), terbucarb (28%) and methyldymron (26%) were detected with relatively high frequency comparing with the others pesticides. In the most crops in agricultural fields, isoprothiolane, flutolanil, are used to control a wide range of fungal diseases and methyldymron and terbucarb are used for weed control. Fenitrothion is a powerful insecticide that is

widely used against many insects and crustaceans. Fenitrothion and isoprothiolane have also been detected with high frequency by other investigators in Japan (Numabe *et al.*, 1992; Itagaki *et al.*, 2000; Kondoh *et al.*, 2001; Tanabe *et al.*, 2001; Sudo *et al.*, 2002). The pesticides with highest mean concentrations were isoprothiolane, fenitrothion and iprodione (37, 35 and 34 ng/l, respectively) (as 5 sites average), while those with lowest mean concentrations were pencycuron, diazinon, tolclofos-methyl and dithiopyr (5, 6, 7 and 8 ng/l, respectively) (Table 4). This implies that, for many of these pesticides, factors such as soil characteristics, weather and agricultural management practices, may be more influential than their chemical and physical properties (e.g., wa-

Fig. 2. Detection frequency of pesticides and nonylphenol during the study period in Kurose River. Detection frequency (%) = (number of samples detecting the compound)/(number of samples analyzed).

Fig. 3. Monthly variation of pesticides and nonylphenol concentrations at sampling sites in Kurose River.

ter solubility) in their movement from agricultural fields to the river. The types of detected pesticides in this study, and their concentrations are generally the same as those reported in other river waters in Japan. The concentration of fenitrothion, diazinon and isoprothiolane in Seta river water were 0.006, 0.023 and 0.331 $\mu\text{g/l}$, respectively

(Sudo *et al.*, 2002). Itagaki *et al.* (2000) reported that fenitrothion was detected with 0.008 $\mu\text{g/l}$ of mean concentration in Minaga river water, while Numabe *et al.* (1992) reported that fenitrothion concentration was 0.025 $\mu\text{g/l}$ in Kozakura river water. The concentration levels of simazine (0.024 $\mu\text{g/l}$), diazinon (0.024 $\mu\text{g/l}$), fenitrothion (0.036

Table 4. Mean, minimum and maximum concentrations of pesticides and nonylphenol detected at various sites of Kurose River during April 2001–January 2002

Compound name	Site name					Average
	Namitakiji	Tokumasa-Kami	Izumi	Ochiai	Hinotsume	
Pencycuron	<5	<5	<5	<5	6 (<5–12)	5
Simazine	<24	<24	25 (<24–37)	25 (<24–35)	<24	24
Propyzamide	<12	<12	14 (<12–27)	<12	<12	12
Diazinon	<2	3 (<2–10)	7 (<2–4)	16 (<2–89)	<2	6
Terbucarb	<20	24 (<20–61)	22 (<20–41)	21 (<20–31)	28 (<20–97)	23
Tolclofos-methyl	<6	<6	<6	<6	9 (<6–25)	7
Metalaxyl	<20	<20	32 (<20–99)	35 (<20–101)	27 (<20–80)	27
Dithiopyr	<9	<9	11 (<9–25)	10 (<9–25)	13 (<9–49)	8
Fenitrothion	30 (<21–65)	39 (<21–86)	54 (<21–304)	<21	30 (<21–56)	35
Pendimethalin	<8	<8	11 (<8–35)	<8	<8	9
Methyldymron	19 (<6–98)	13 (<6–52)	28 (<6–96)	23 (<6–142)	17 (<6–110)	20
Butamifos	<7	<7	15 (<7–82)	<7	<7	9
Flutolanil	6 (<4–25)	10 (<4–38)	26 (<4–113)	21 (<4–56)	19 (<4–41)	16
Isoprothiolane	8 (<8–9)	39 (<8–244)	79 (<8–567)	17 (<8–84)	44 (<8–299)	37
Triclopyr-ester	<15	<15	16 (<15–24)	<15	<15	15
Isoxathion	<9	<9	13 (<9–20)	10 (<9–16)	<9	10
Mepronil	<12	18 (<12–56)	15 (<12–26)	13 (<12–23)	13 (<12–21)	14
Iprodione	51 (<30–244)	<30	<30	<30	<30	34
Pyridaphenthion	<9	<9	<9	23 (<9–98)	10 (<9–23)	12
Total pesticides	272	300	418	318	323	326
Nonylphenol	207 (<14–700)	136 (<14–250)	629 (<14–2170)	285 (92–995)	269 (28–858)	305

Unit = ng/l.

$\mu\text{g/l}$), pyridaphenthion (0.022 $\mu\text{g/l}$), flutolanil (0.021 $\mu\text{g/l}$), and mepronil (0.063 $\mu\text{g/l}$) in Shinano river water were reported by Tanabe *et al.* (2001). The concentration level of diazinon (0.016 $\mu\text{g/l}$), fenitrothion (0.031 $\mu\text{g/l}$), isoprothiolane (0.027 $\mu\text{g/l}$), flutolanil (0.002 $\mu\text{g/l}$), tolclofos methyl (0.003 $\mu\text{g/l}$), mepronil (0.006 $\mu\text{g/l}$), iprodione (0.011 $\mu\text{g/l}$) and isoxathion (0.003 $\mu\text{g/l}$) in Ishikari river water was reported by Kondoh *et al.* (2001).

Seasonal variations of pesticide concentrations were apparent at all the sampling sites (Fig. 3a). Pesticide concentrations were generally higher during growing season (April to September) and gradually decreased starting in October. The high concentrations in the growing season were probably due to the fact that the most of pesticides are used this period (Tanabe *et al.*, 2001). Of the five sites, the Izumi, Hinotsume and Ochiai sites, had the highest concentrations throughout seasons. These three sites are located in agricultural and

urban areas where various sources of pesticides are present. The main source of pesticides in Kurose River appears to be agricultural fields. This is based on the finding of a good correlation between monthly concentration of total pesticides (as 5 sites average) and corresponding monthly precipitation (Fig. 4), and the finding that there is an involvement of pesticides with soil mineral components such as Na^+ , K^+ and Ca^{2+} in the principal component analysis (Table 5). Several studies have shown that precipitation events increase the concentration levels of pesticides in stream in agricultural areas. After carbofuran was applied to an oilseed rape crop, its concentrations increased to biologically active levels during rainstorms (Matthiessen *et al.*, 1995). In west England, relatively high concentrations of herbicides and insecticides were observed after heavy rainfall (Brooke and Matthiessen, 1991; Williams *et al.*, 1991a, b; Matthiessen *et al.*, 1992). Thus

Fig. 4. Correlation between monthly concentration of total pesticides (average of 5 sites) and corresponding monthly atmospheric precipitation of Higashi-Hiroshima during the study period.

Table 5. Principal component analysis for 5 sites average of water quality data obtained during June 2001–January 2002 in Kurose River

	Factor 1		Factor 2		Factor 3	
Total variance (%)	38.4		19.2		17.8	
Component ($r \geq 0.5$)	K ⁺	0.92	Cl ⁻	-0.88	Nonylphenol	0.82
	Precipitation	0.91	NO ₂ ⁻ + NO ₃ ⁻	0.88	NH ₄ ⁺	-0.77
	Na ⁺	0.90	Mg ²⁺	0.77	SO ₄ ²⁻	0.70
	Pesticides	0.82			WT	0.52
	DOC	0.81				
	Ca ²⁺	0.78				
	H ⁺	-0.56				

WT: water temperature; DOC: dissolved organic carbon.

we conclude that pesticide concentrations in Kurose River are determined by two processes: the time that the pesticides are applied to agricultural fields and the amount of precipitation, which washes the pesticides into the streams.

Simazine that was detected in this study is designated as a substance of environmental quality standards for the human health, and diazinon, propyzamide, fenitrothion, isoprothiolane and isoxathion are designated as monitoring needed substances by the Ministry of Environment, Japan. However, the standard values or guideline values of these substances in natural water (3 to 40 µg/l) are much higher than the concentrations that we determined. Thus, these pesticides should have no adverse health effects in the Kurose River.

The total pesticide flux in Kurose River was considerably higher during the period of April to August and decreased after September (Fig. 5b). This is due to high water flow rates (Fig. 5a) and relatively high concentrations of detected pesticides during the spring and summer seasons. At Namitakiji site, the water flow was almost zero because of a pond structure at this site. Thus the flux of pesticides at this site was assumed to be zero. The flux of total fungicides over the study period was much greater than that of herbicides and insecticides at all sites (Fig. 6). This was because fungicides were detected at higher concentrations and higher frequencies than herbicides and insecticides. These fungicides were relatively widespread; possibly due to their uses to control

Fig. 5. Monthly water flow rate and fluxes of pesticides and nonylphenol at each sampling site in Kurose River.

wide range of fungal diseases for most crops, especially for the rice the common crop in this area. The flux of individual pesticides in the Kurose River near Ochiai site ranged from 0.00002–0.013% of estimated annual use in Higashi-Hiroshima (Table 6). The large variability in these percentages is not surprising in view of the considerable variability in (1) physical properties of pesticides (2) the application practices (Larson *et*

al., 1995), and (3) the effective dissipation processes during the transit time in the surface water (Sudo *et al.*, 2002). These percentages were low compared with the previously published values (Larson *et al.*, 1995). This can be due to many reasons, such as differences in the chemical and environmental properties of the pesticides, application practices, soil characteristics, topography and weather. The flux is also influenced by low

Fig. 6. The flux of total insecticides, herbicides and fungicides during April 2001–January 2002 in Kurose River.

Table 6. Amount of agricultural use in Hiroshima prefecture, estimated annual use in Higashi-Hiroshima (a), annual flux (b) and the percentage of (b) to (a), for detected pesticides in Kurose River

	Annual use in Hiroshima (1999–2000) (kg)	Estimated annual use in Higashi-Hiroshima (a) (kg)	Annual flux in Kurose River (b) (kg)	(b)/(a) × 100 % of amount use
Pencycuron	9300	650.35	0.00014	0.00002
Simazine	1200	83.92	0.00068	0.0008
Propyzamide	900	62.94	0.00034	0.0005
Diazinon	1700	1209.7	0.00017	0.00001
Tolclofos-methyl	600	41.96	0.00020	0.00047
Metalaxyl	2600	181.8	0.00076	0.00042
Dithiopyr	700	48.95	0.00023	0.00046
Fenitrothion	1300	90.91	0.00099	0.001
Pendimethalin	5200	363.64	0.00025	0.00007
Methyldymron	4100	286.71	0.00057	0.00019
Butamifos	300	20.98	0.00025	0.0012
Flutolanil	3200	223.78	0.00045	0.0002
Isoprothiolane	4000	279.72	0.00105	0.00037
Triclopyr-ester	1900	132.87	0.00042	0.00031
Isoxathion	15900	1111.8	0.00028	0.00002
Mepronil	100	6.99	0.00040	0.005
Iprodione	100	6.99	0.00096	0.013
Pyridaphenthion	100	6.99	0.00034	0.0048

Estimated annual use of pesticides in Higashi-Hiroshima was calculated by using the ratio of agricultural area in Higashi-Hiroshima to that of Hiroshima prefecture.

water flow rate of Kurose River (estimated at 0.91 m³/sec as the average rate during the study period, based on data obtained by Hiroshima prefecture office). For comparison, the flow rate in the Seta River in Shiga prefecture in Japan is 90 m³/sec (Sudo *et al.*, 2002).

Nonylphenol

Of the compounds investigated in this study, nonylphenol was detected with highest frequency (91.3%) (Fig. 2). This may be because nonylphenol has many sources, such as detergents, emulsifiers, wetting and dispersing agents in pes-

Fig. 7. Correlation between the concentration of total pesticides and nonylphenol (as 5 site averages) in Kurose River.

ticide formulations, and household products. Other studies also reported detecting nonylphenol at high frequencies in river water in Japan (Kojima and Watanabe, 1998; Isobe *et al.*, 1999). Nonylphenol also showed higher mean concentration (305 ng/l) than that of individual pesticides at all sites (Table 4) due to the wide range of contamination sources and the heavy uses as household products. The Izumi, Ochiai and Hinotsume sites had the highest nonylphenol concentrations, followed by Tokumasa-Kami and Namitakiji sites. The higher concentrations of nonylphenol at Izumi site appear to be due to the wide range of potential nonylphenol sources at this site, which is near a densely populated urban and industrial area. Very high concentrations of nonylphenol were detected in May and August 2001 at Izumi site (Fig. 3b), which suggests the occurrence of one or more local specific sources of nonylphenol. However, the locations of these sources are unknown. The nonylphenol concentrations in the Kurose River are similar to those reported in other rivers in Japan (Isobe *et al.*, 1999; Tsuda *et al.*, 2000; Tabata *et al.*, 2001).

The mean concentration of nonylphenol in the natural water including river, lake and sea waters in Japan (170 ng/l, $n = 1574$, range <30–21000 ng/l) (Ministry of Environment Japan, 2001) is lower than the mean concentration (305 ng/l) in Kurose River. Also according to this same report the maximum concentration of nonylphenol that

has no effects as an endocrine disrupter in fish is 608 ng/l. The 10-month average concentration of nonylphenol at the Izumi site and spring and summer concentrations at some of the other sites, were higher than this level (Fig. 3b). This indicates that nonylphenol concentrations in the Kurose River may be occasionally hazardous to aquatic organisms, especially fish.

Pesticides are considered to be one of the sources of nonylphenol contamination in natural waters. Thomas *et al.* (2001) reported the occurrence of nonylphenol, as well as herbicides such as endosulfan sulfate and pendimethalin, in stream waters in south east England, possibly resulting from the use of NPEOs in pesticides formulations. Also NPEOs has been used in the formulation of insecticides such as aminocarb. Aminocarb has been used for control the spruce budworm in Canada and, after spraying, 4-nonylphenol was found at ppb levels in a forest stream (Sundaram *et al.*, 1980; Fairchild *et al.*, 1999). However, the correlation between total pesticides and nonylphenol concentrations (as 5 site averages) measured in this study was very weak (Fig. 7), which suggests that main sources of these pollutants are different from each other. The principal component analysis indicated that the concentration of nonylphenol is well correlated with the concentration of SO_4^{2-} ion, which is a major component of wastewater. The Kurose River at Higashi-Hiroshima receives treated wastewaters

from local community and local industries, as well as from many domestic sewage purifiers set up in individual houses. Thus we believe that the main sources of nonylphenol in the Kurose River are wastewaters discharged from various facilities as a result of human activities.

The flux of nonylphenol at three downstream sites was generally high from April to August and started to decrease from September during the study period (Fig. 5c). The flux of nonylphenol was highest at the Hinotsume, Izumi and Ochiai sites and this is well associated with the high water flow rates (Fig. 5a) and concentration levels recorded at these sites (Fig. 3b). At the Tokumasa-Kami site, the flux is the lowest one and this is due to the low water flow rate and relatively low concentration of nonylphenol at this site. The flux of nonylphenol was comparable with that of the total pesticides and sometimes exceeds it at the three downstream sites, apparently because the nonylphenol concentrations were almost same as or higher than the total pesticide concentrations at these sites.

CONCLUSIONS

Nineteen pesticide compounds and nonylphenol were detected in Kurose River waters from both agricultural and urban areas during the study period. The concentration range of these compounds was at the ppt level. Many of the detected insecticides, fungicides and herbicides are common agricultural pesticides used in Hiroshima Prefecture. Particularly fungicides that are commonly used for crop protection were detected at high frequencies and at high concentration at all sites.

The detection frequency and concentration of nonylphenol were higher than individual pesticides at all sites, suggesting that the water pollution by nonylphenol is considerable and may be harmful to aquatic organisms at some sites in the Kurose River. The results obtained from the present study indicate that both agricultural and urban areas are sources of pesticides and nonylphenol contamination in the Kurose River.

Seasonal variation of pesticides and nonylphenol concentrations and fluxes indicated that spring and summer are the critical seasons for the occurrence of these pollutants in the Kurose River.

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