

# Distribution of pesticides and bisphenol A in sediments collected from rivers adjacent to coral reefs

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## Abstract

To investigate the deteriorating health of coral reefs in Okinawa, Japan, natural sediment samples were analyzed for diuron, Irgarol 1051, chlorpyrifos, and bisphenol A (BPA) which are hazardous to corals. Samples were analyzed by solid-phase extraction (SPE) followed by high-performance liquid chromatography with tandem mass spectrometry (LC–MS–MS). Although diuron and chlorpyrifos usage is only well recorded for farms and not for cities, these chemicals were detected in both rural and urban areas. Additionally, diuron concentration in urban areas was in some cases higher than in rural areas, which might be caused by greater consumption of these chemicals in home gardens in city areas. Irgarol 1051 was detected in downstream river areas, which are situated far from the source sites such as pier or fishery harbor (0.6–3.2 km). This result suggested that Irgarol 1051 could be transported from the river mouths to the sampling sites during flood tides. High BPA concentrations were associated with urban areas ( $<1.2\text{--}22.0\ \mu\text{g kg}^{-1}$ ), while low concentrations were associated with rural areas ( $\text{nd}\text{--}6.8\ \mu\text{g kg}^{-1}$ ). The river sediments under study are delivered to coral reefs in large quantity through runoff caused by typhoons and other heavy rains. The highly hazardous chemicals are carried into coral reefs on these sediments. Therefore, these hazardous chemical substances may already be influencing the coral reefs.

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**Keywords:** Bisphenol A (BPA); Diuron; Irgarol 1051; Chlorpyrifos; LC–MS–MS

## 1. Introduction

Coral reef ecosystems are characterized by high biological diversity and are important ecological components. Coral reefs have been seriously deteriorating worldwide during recent years, mainly due to outbreaks of crown-of-thorns starfish, terrestrial runoff and bleaching events caused by high seawater temperature (Hughes et al., 2003). Terrestrial runoff of soil is intensified by human

land-based activities, and reduces corals' photosynthetic efficiency and increases its respiration, resulting in bleaching and necrosis (Fabricius, 2005). Hazardous chemical substances are contributing to the destruction of coral reef ecosystems, but little attention has been paid to their distribution (Harrington et al., 2005; Carbery et al., 2006).

A variety of hazardous chemical substances are released into the environment in Okinawa by humans through domestic activities, agriculture, and industrial waste discharge (West and Van Woosik, 2001; Kawahata et al., 2004). In urban areas of Okinawa with high population density, bisphenol A (BPA) were detected at  $\text{nd}\text{--}11\ \mu\text{g kg}^{-1}$  (Kawahata et al., 2004). BPA is classified as an endocrine disrupting chemical (EDC) and is released into the environment as a result of its use in polycarbonate (PC) plastics and

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epoxy resins (Krishnan et al., 1993). BPA leaching from PC tubes into water increases with higher temperature and time (Sajiki and Yonekubo, 2003), though BPA in river water rapidly degrades under aerobic conditions (Kang and Kondo, 2002; Kang and Kondo, 2005). Investigations on BPA degradation in marine sediments show that BPA degrades under aerobic conditions but not under anaerobic conditions (Ying and Kookana 2003).

Recently herbicides released into environment have also been problems, since the discovery of diuron [*N'*-(3,4-dichlorophenyl)-*N,N*-dimethylurea] and Irgarol 1051 [2-(methylthio)-4-(*tert*-butylamino)-6-(cyclopropylamino)-s-triazine] in tropical marine environments (Haynes et al., 2000; McMahon et al., 2005; Carbery et al., 2006; Lam et al., 2006). The concentration of diuron in sediments adjacent to the Great Barrier Reef Marine Park (Australia) has been reported to range from 0.1 to 10.1  $\mu\text{g kg}^{-1}$  (Haynes et al., 2000; McMahon et al., 2005). It is difficult to study the impact of urban diuron use because private uses are poorly accounted for. Irgarol 1051 is used in antifouling paints as a biocide. Chlorpyrifos [*O,O*-diethyl *O*-3,5,6-trichloro-2-pyridyl phosphorothioate] is a broad-spectrum organophosphate insecticide that is rapidly gaining importance in worldwide agriculture, especially as a replacement for other organochlorine insecticides that have been restricted or banned. In Okinawa, prior to a ban, chlorpyrifos was applied in houses as a termiticide. Corals contain steroidal estrogens, and 17 $\beta$ -estradiol is released during coral spawning events (Atkinson and Atkinson, 1992; Tarrant et al., 2004). Tarrant et al. (2004) reported that continuous exposure to 17 $\beta$ -estradiol caused abnormal reproduction in *Montipora capitata*, and that continuous exposure to dilute estrone reduced the growth rate of *Porites compressa*. Exposure to EDCs can cause sterility or decreased fertility, impaired development, birth defects of the reproductive tract, and metabolic disorders in corals (Tarrant et al., 2004). Diuron and Irgarol 1051 inhibit photosynthesis by preventing oxygen production and block the electron transfer at the level of photosystem II (PSII) in the chloroplasts of algae. These toxicological effects were studied in symbiotic conditions (Jones and Kerswell, 2003; Jones, 2005). These hazardous chemical substances pose a potential risk for corals and coral reef ecosystems.

In order to evaluate the possible contamination of coral reefs by the chemical substances discussed earlier, we analyzed their levels in sediments collected from rivers adjacent to coral reefs in Okinawa, Japan. The results indicated that the liquid chromatography–electrospray tandem mass spectrometry (LC–MS–MS) method was suitable for analysis of diuron, Irgarol 1051, chlorpyrifos and BPA in aquatic sediment samples.

## 2. Study area

Okinawa Island (26°25'N, 128°00'E) is one of the Ryukyu Islands of Japan. Most of the coral reefs around Okinawa

are fringing or patch reefs; they are adjacent or close to coastal urban areas and farmland. Human activities such as farming and road building, together with modifications of river drainage systems, have accelerated runoff of the red clay soil, which has negatively affected many Okinawa reefs (Omija, 2004).

Sediment sampling sites were chosen to represent different pollution conditions (Fig. 1). In northern Okinawa, two sites at the mouths of the Manna and Kouchi rivers are situated in areas characterized by a variety of farmland types and low population density. The two sites at the mouths of the Hija, Makiminato, and Aja rivers in central Okinawa are very close to city areas. The Kokuba, Nagado, Noha, and Youhi rivers are situated in southern Okinawa, and the Kokuba, Nagado, and Noha rivers flow through Naha City, which has a high population density. The upstream section of Kokuba River, the location of sampling site K-1, flows through a residential area and receives mostly domestic wastes. Although these sites do not reflect any major industrial activity, both agricultural and residential uses may affect water quality (Tashiro et al., 2003), and are both represented.

## 3. Material and methods

### 3.1. Chemicals

The standards of bisphenol A (BPA), stable isotope-labeled surrogate standards (BPA- $d_{16}$ ) and pesticides (diuron, Irgarol 1051, chlorpyrifos) were obtained from Kanto Chemical Co. Inc. (Tokyo, Japan). In Fig. 2, the chemical structures of diuron, Irgarol 1051, chlorpyrifos and BPA are represented. The physical and chemical properties of these chemicals were given in Table 1. HPLC analysis grade methanol (MeOH), acetonitrile (ACN), methyl-*t*-butylether (MTBE) and acetone (Kanto Chemical Co. Inc., Tokyo, Japan) were used in this study. Water purified with a Milli-Q<sup>®</sup> water-purification system was used (see Table 2).

### 3.2. Sampling

For the analysis of pesticides and BPA, sediments were collected with a stainless-steel spatula. In each case, the uppermost 5-cm-thick layer of sediment was collected at the water/sediment interface near the bank. The samples were stored at 4 °C in brown glass bottles that had been first washed with Milli-Q<sup>®</sup> water and then pre-rinsed with acetone. The sediments for pesticides analysis were collected in July 2004 and September 2005 in Okinawa (Fig. 1). Analysis for BPA was carried out on ten suites of samples except Tairawan (Fig. 1). To study temporal variation of BPA, several sampling campaigns covering different seasons were carried out over a two-year (2003–2004) period. Sampling was conducted in September and November 2003 and in February, May and July 2004. All sampling started at rising tide and ended at ebb tide.

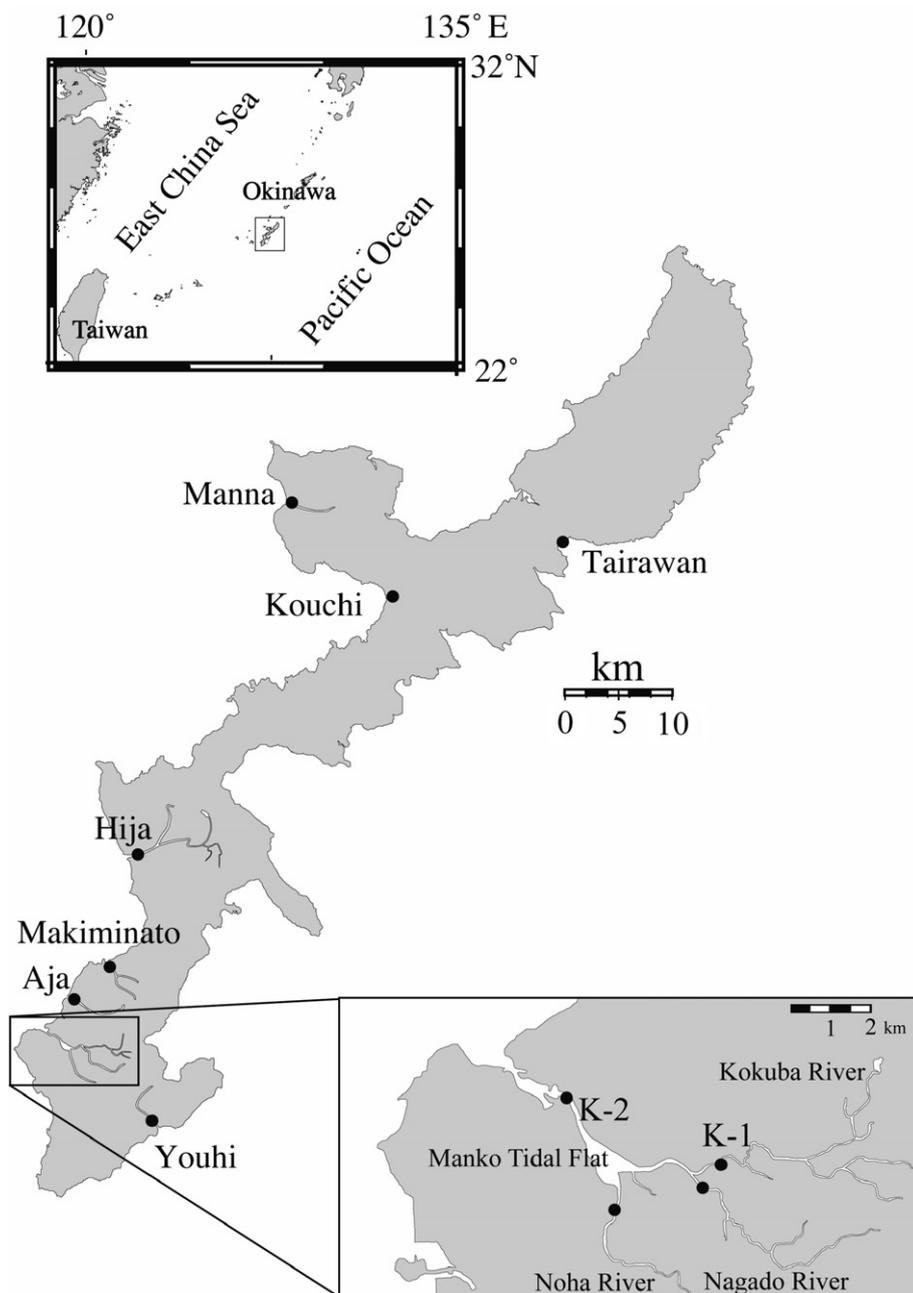


Fig. 1. Locations of sampling sites. The study site is shown as a solid circle on the inset map of Okinawa Island.

### 3.3. Analysis of hazardous chemical substances

#### 3.3.1. Sample treatment of solid-phase extraction

Subsamples of exactly 5 g with surrogate materials (BPA- $d_{16}$ ) was shaken for 30 min and extracted under ultra sonic vibration for 15 min with 25 mL of ACN. A 5-mL aliquot of the ACN extract was diluted to 100 mL with water and the solution was passed through an Oasis<sup>®</sup> HLB cartridge (60  $\mu$ m *N*-vinylpyrrolidone/divinyl benzene copolymer, 250 mg) (Waters, Tokyo, Japan) that has been conditioned prior to use with MTBE (3 mL), MeOH (3 mL) and water (3 mL). The cartridge was then rinsed with 3 mL of water and the pesticides and BPA retained

in the cartridge was eluted with 6 mL of MeOH/MTBE (10/90, v/v) solution. The extract was evaporated under a gentle stream of nitrogen to near dryness and reconstituted to 1 mL with MeOH/water (80/20, v/v) for pesticide analysis or ACN/water (70/30, v/v) for BPA analysis.

#### 3.3.2. LC-MS-MS analysis

The liquid chromatography-tandem mass spectrometry (LC-MS-MS) system consisted of a Waters 2695 Alliance separation module, coupled to a Quattro Ultima pt triple-stage quadrupole mass spectrometer with a pneumatically assisted electrospray ionization (Z-spray source), from Micromass (Manchester, UK). Chromatographic

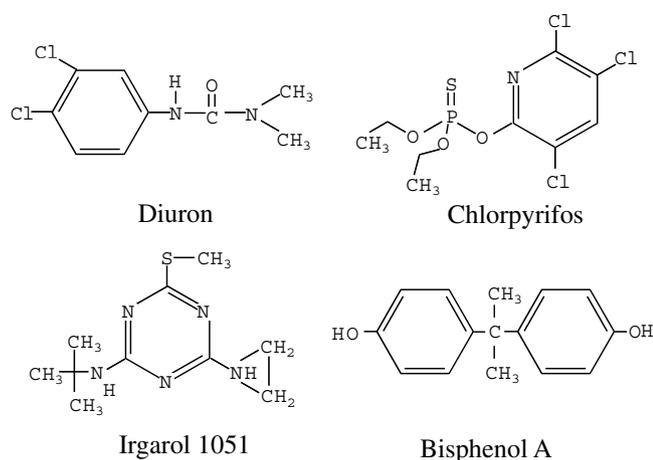


Fig. 2. Structures of selected hazardous chemical substances.

Table 1  
The physical and chemical properties of hazardous chemical substances

Name	Sorption coefficient		Half life (days)	
	Log $K_{ow}$	Log $K_{oc}$	Aerobic	Anaerobic
BPA <sup>a,b</sup>	3.30	3.20	14.5	No degradation
Diuron <sup>c,d</sup>	2.68	2.40	Extream persistant	14
Irgarol 1051 <sup>e,f,g</sup>	3.38	2.16	100–200 (seawater)	No degradation
Chlorpyrifos <sup>c,h</sup>	5.27	3.70	14–24	58–223

<sup>a</sup> Staples et al. (1998).

<sup>b</sup> Ying and Kookana (2003).

<sup>c</sup> Sabljic et al. (1995).

<sup>d</sup> Thomas et al. (2002).

<sup>e</sup> Thomas et al. (2003).

<sup>f</sup> Lam et al. (2006).

<sup>g</sup> Ciba-Geigy (1988).

<sup>h</sup> Bondarenko and Gan (2004).

separation was performed at 40 °C on a 150 mm × 2.1 mm, 5- $\mu$ m particle-size, Inertsil ODS-3 (GL Science Inc. Japan) column. The injection volume was 10  $\mu$ L. Pesticide measurement was performed with an isocratic mobile phase, water/MeOH = 30/70. Run time was 25 min at a flow rate of 0.2 mL min<sup>-1</sup>. BPA measurement was performed with an isocratic mobile phase, ACN/water = 75/25. Run time was 15 min at a flow rate of 0.2 mL min<sup>-1</sup>.

Mass spectrometry interface conditions, the optimized parameters and product ions are given in Table 1. Method validation and quantitative analysis were performed by multiple reactions monitoring (MRM). For each analysis, parent-product ion transitions were chosen

and voltages and gas flows were optimized for maximum intensities.

### 3.3.3. Quantification

Primary stock solutions were prepared individually at a concentration of 10 mg g<sup>-1</sup> by placing about 10 mg in a 1-mL volumetric flask and diluting to volume with ACN. The working standards of mixtures were prepared in ACN/water (70/30, v/v) solution at concentrations ranging from 1.0 to 500 ng g<sup>-1</sup> (10 points) for BPA and in MeOH/water (80/20, v/v) solutions at concentrations ranging from 0.01 to 10 (10 points) for pesticides. A calibration curve was generated for every analysis by linear regression, which over the analyzed concentration range yielded a high  $r^2$  value of 0.997.

### 3.3.4. Recoveries and detection limits

In order to examine the quality of the data obtained by the analytical procedure, we calculated recovery rates and relative standard deviations (RSDs). Diuron and chlorpyrifos were spiked with 50 ng in 5 g of sediment, respectively. The recovery rates of diuron were in the range of 66–74% and RSD was 6.8%. The recovery rates of chlorpyrifos were in the range of 63–71% and the RSD was 5.2%. Irgarol 1051 was spiked at the level of 1 ng g<sup>-1</sup> in 5 g of sediment. The recovery rates of Irgarol 1051 were in the range of 83–88% and the RSD was 3.2%. Instrumental limits of detection (LODs, S/N = 3) and limits of quantification (LOQs, S/N = 10) were estimated from a dilution series of standard mixtures. LODs were 0.02 ng g<sup>-1</sup> for diuron, 0.005 ng g<sup>-1</sup> for Irgarol 1051 and 0.02 ng g<sup>-1</sup> for chlorpyrifos. LOQs were 0.07 ng g<sup>-1</sup> for diuron, 0.016 ng g<sup>-1</sup> for Irgarol 1051 and 0.41 ng g<sup>-1</sup> for chlorpyrifos. Based upon three spike tests (level 100 ng g<sup>-1</sup>), the recoveries of BPA were 90–95%. The BPA values in sediments were calibrated by BPA-d<sub>16</sub>. LOD and LOQ for BPA in sediment were 0.6 ng g<sup>-1</sup> and 1.2 ng g<sup>-1</sup>, respectively.

## 4. Results and discussion

### 4.1. Pesticides

The concentrations of pesticides were variable at July 2004 and at September 2005 (Table 3). However, typhoon and heavy rainfalls in Okinawa were similar between 2004 and 2005. We cannot find the major reason why pesticides released into the river sediments were the difference

Table 2  
ESI–MS–MS interface conditions, optimized parameters and product ions

Name	Polarity	Capillary voltage (V)	Precursor ion (m/z)	Product ion (m/z)	Collision energy (V)	Cone voltage (V)
BPA	ES–	–2.8	227	212	20	40
BPA-d <sub>16</sub>	ES–	–2.8	241	223	20	40
Diuron	ES+	+3.0	233	72	15	70
Irgarol 1051	ES+	+3.0	254	198	15	60
Chlorpyrifos	ES+	+3.0	352	200	15	60

Table 3  
Diuron, Irgarol 1051 and chlorpyrifos concentrations in river sediments

Sampling period	Location	Site name	Diuron ( $\mu\text{g kg}^{-1}$ )	Irgarol 1051 ( $\mu\text{g kg}^{-1}$ )	Chlorpyrifos ( $\mu\text{g kg}^{-1}$ )
2004 July	North	Tairawan	<0.07 <sup>a</sup>	nd <sup>b</sup>	nd <sup>b</sup>
		Manna River	0.09	0.12	<0.41 <sup>a</sup>
		Kochi River	<0.07 <sup>a</sup>	<0.016 <sup>a</sup>	nd <sup>b</sup>
	Midland	Hija River	<0.07 <sup>a</sup>	0.02	<0.41 <sup>a</sup>
		Makiminato River	nd <sup>b</sup>	nd <sup>b</sup>	nd <sup>b</sup>
		Aja River	0.11	0.066	<0.41 <sup>a</sup>
	South	Noha River	0.09	0.018	nd <sup>b</sup>
		Nagado River	<0.07 <sup>a</sup>	nd <sup>b</sup>	nd <sup>b</sup>
		K-2	0.09	0.029	nd <sup>b</sup>
		Youhi River	0.09	<0.016 <sup>a</sup>	nd <sup>b</sup>
2005 September	North	Tairawan	0.07	nd <sup>b</sup>	nd <sup>b</sup>
		Manna River	<0.07 <sup>a</sup>	<0.016 <sup>a</sup>	<0.41 <sup>a</sup>
	Midland	Hija River	<0.07 <sup>a</sup>	0.051	nd <sup>b</sup>
		South	Noha River	0.07	<0.016 <sup>a</sup>
	Nagado River		0.22	<0.016 <sup>a</sup>	<0.41 <sup>a</sup>
	K-2		0.58	0.034	<0.41 <sup>a</sup>
	Youhi River	0.08	0.039	<0.41 <sup>a</sup>	

<sup>a</sup> <values indicate that contents were below the quantification limit.

<sup>b</sup> nd means not detected.

between in July 2004 and in September 2005. On the other hand, the distributions of these chemicals are affected by their chemical and physical properties and/or application.

#### 4.1.1. Diuron

Diuron was detected in both rural and urban areas (Table 3). However, even the maximum level of diuron concentrations in sediment in Okinawa ( $0.22 \mu\text{g kg}^{-1}$ ) was low relative to those reported adjacent to the Great Barrier Reef World Heritage Area (Australia) ( $0.1$ – $0.5 \mu\text{g kg}^{-1}$  and  $<0.1$ – $10.1 \mu\text{g kg}^{-1}$ ) (Haynes et al., 2000; McMahan et al., 2005).

Farmlands are common in northern Okinawa with an area of  $14.7 \text{ km}^2$  around the Manna River. In contrast, there is relatively little farmland around Naha City ( $3.8 \text{ km}^2$ ). According to the Japan Plant Protection Association, about  $17000 \text{ kg y}^{-1}$  of diuron is used in Okinawa. Some authors have evaluated the transport of diuron from agricultural land into rivers and adjacent to coral reefs (Haynes et al., 2000; McMahan et al., 2005). The sorption coefficients of diuron are relatively moderate, with  $\log K_{\text{OW}}$  of 2.68 and  $\log K_{\text{OC}}$  of 2.40, suggesting that diuron will be found predominantly in the dissolved phase and will be only weakly adsorbed to soil (Sabljic et al., 1995). The occurrence of diuron in northern Okinawa is therefore likely to be a result of local agricultural usage combined with low sorption coefficient. Though, diuron was also detected at locations close to the urban areas such as Naha City. Furthermore, diuron concentrations in the urban areas were much higher than those in the rural areas in September 2005. This may be attributed to diuron consumption in urban activity.

Diuron concentration was significantly high at Site K-2 in September 2005. This site was under aerobic condition because of seawater contribution at the flood tide

(OPGDCE, 2003; Kitada et al., 2006). Diuron has been shown to be extremely persistent in seawater, while it degrades much quickly under anaerobic conditions with half-life of 14 days in marine sediments (Thomas et al., 2002; Thomas et al., 2003). This property of diuron suggests that it can persist at Site K-2 for relatively longer period. Since diuron was transported to the downstream due to its low affinity for sediments, its concentrations increased from the upstream to the downstream in September 2005.

#### 4.1.2. Irgarol 1051

Since Irgarol 1051 is used in antifouling paints, it is reasonable to assume that this compound will be detected close to ports, including fishing ports. However, Irgarol 1051 was detected in downstream river areas situated far away from the source sites, which are generally in coastal regions. An interesting characteristic of Irgarol 1051 is its relatively low  $\log K_{\text{OC}}$  of 2.16 in the field in spite of the relatively high  $\log K_{\text{OW}}$  of 3.38 (Lam et al., 2006). Biselli et al. (2000) showed that Irgarol 1051 was remobilized from sediments into water because of low partition/sorption coefficients. In fact, Irgarol 1051 was detected in water more frequently than in sediments (Biselli et al., 2000). Another characteristic of Irgarol 1051 is that it degrades slowly. Half-lives of Irgarol 1051 are 100–200 days in marine and freshwater sediments under aerobic conditions (Ciba-Geigy, 1988). The degradation of Irgarol 1051 is considerably slower in seawater under aerobic conditions ( $t_{1/2} = 350$  days) (Thomas et al., 2002). In marine sediments under anaerobic conditions, Irgarol 1051 did not degrade during a 42-day period (Thomas et al., 2003). These results show that Irgarol 1051 could be much more persistent under the influence of seawater. A third characteristic of Irgarol 1051 is that the tidal current can affect its distribution.

The sampling sites where Irgarol 1051 was detected were influenced by seawater at the flood tide, whereas Irgarol 1051 was not detected at the site close to Site K-1, where little marine influence was indicated by low salinity (Kitada et al., 2006). Biselli et al. (2000) showed that tidal currents might contribute to the dispersion of Irgarol 1051. Therefore, Irgarol 1051 could be transported from the river mouths to the sampling sites during flood tides.

#### 4.1.3. Chlorpyrifos

Chlorpyrifos was also detected in both rural (the Manna and Youhi river sites) and urban areas (Naha City) although those values were below LOQs. A previous study reported that chlorpyrifos was detected in runoff water and sediments in agricultural streams (Jergnetz et al., 2005; Bollmohr et al., 2007). The catchments of the Manna and Youhi rivers include many farmlands. Therefore, agricultural runoff from the farmlands could yield the detectable amounts of chlorpyrifos in river sediments.

Chlorpyrifos concentrations were detected around Naha City at September 2005, where there are many residential areas with high population density, and where the chemical has been used as a termiticide. Chlorpyrifos does not degrade when used as a termiticide. Racke et al. (1993) reported that application of chlorpyrifos as a termiticidal soil barrier created persistent residues; nearly 70% of the initially applied chlorpyrifos remained in soil even after 18 months. Subsequent laboratory investigations determined that chlorpyrifos degradation proceeds with half-lives from 116 to 1576 days in five kinds of soil treated with termiticide at a concentration of 1000 ppm (Racke et al., 1993). Yoshida et al. (2004) reported that summertime levels of chlorpyrifos in the indoor air of one house did not decrease over a period of seven years after initial application of the termiticide. These results suggest that chlorpyrifos remains in houses and other buildings after use as a termiticide. On the other hand, chlorpyrifos in sediments were moderately persistent under aerobic conditions ( $t_{1/2} = 14\text{--}24$  days). The persistence of chlorpyrifos increased significantly under anaerobic conditions, and  $t_{1/2}$  was prolonged to 58–223 days (Bondarenko and Gan, 2004). Although the Building Standards Law (BSL) in Japan has banned the use of chlorpyrifos as a termiticide for houses since July 2003, traces of this compound were detectable in the urban areas as shown in the present study.

#### 4.1.4. Influence of diuron and Irgarol 1051 on coral photosynthetic system

Most corals rely on symbiotic dinoflagellates (*Symbiodinium* spp.) to acquire the additional energy required for colony maintenance, growth and reproduction (Rowan, 1998). Diuron and Irgarol 1051 inhibit photosynthesis by preventing oxygen production, and block the electron transfer at the level of photosystem II (PSII) in the chloroplasts of plants and algae. *In symbio*, the diuron concentration required to reduce the effective quantum yield of

photosynthesis by 50% over a period of 10 h (i.e. 10 h-EC<sub>50</sub>) in *Acropora formosa*, *Montipora digitata*, *Porites cylindrica* and *Seria topora hystrix* ranged from 2 to 6  $\mu\text{g L}^{-1}$  (Jones and Kerswell, 2003). The LOEC was 0.25  $\mu\text{g L}^{-1}$  for dinoflagellates isolated from corals and 0.3  $\mu\text{g L}^{-1}$  for dinoflagellates within coral tissue (*in hospite*) (Jones, 2005). High diuron concentrations have also been shown to cause expulsion of symbionts from the host (bleaching) (Jones and Kerswell, 2003). *In symbio*, the photochemical efficiency of the symbiotic algae in *Seria topora hystrix* is significantly reduced following several hours' exposure to Irgarol 1051 concentrations as low as 0.05  $\text{ng L}^{-1}$  (Jones and Kerswell, 2003).

The diuron and Irgarol 1051 concentrations in seawater estimated from those concentrations in sediments reported in this study were lower than the effective concentrations. However, sedimentation combined with exposure to toxic chemicals significantly enhances stress on symbiotic algal species (Harrington et al., 2005). Flooding events caused by typhoons and other heavy rain can transport high amounts of hazardous chemical substances to near-shore reefs. Sediment loads (generally referred to as red soil or red clay runoff) due to agriculture and urbanization are considered to be a major factor in the deterioration of the coral reefs in Okinawa (Omija, 2004). These results support the view that sedimentation may cause serious damage to coral reef ecosystems when combined with the chemicals that are entrained with the sediment flow during runoff events. Therefore, the presence of diuron and Irgarol 1051 may already be affecting coral reefs in Okinawa.

## 4.2. BPA

### 4.2.1. Distribution of BPA in Okinawa Island

BPA is a widely used compound in the production of PC plastics and epoxy resins. Leaching of BPA from PC plastics into water has been reported (Sajiki and Yonekubo, 2003). The prime source of BPA in Okinawa is likely to be domestic wastewater, because PC plastics are used for domestic applications such as food packaging and plastic bottles for water, and epoxies are also used as a coating for polyvinyl chloride water drainpipe walls (Imaoka et al., 2006).

BPA concentrations in farmland-area sediments such as in northern Okinawa (nd–6.8  $\mu\text{g kg}^{-1}$ ) were lower than in urban areas such as central and southern Okinawa (<1.2–22.0  $\mu\text{g kg}^{-1}$ ) (Fig. 3). This trend suggests that BPA pollution in Okinawa is being caused by domestic and/or industrial wastes, and less by agricultural activities, coinciding with conclusions by Kawahata et al. (2004). Note that there is no major heavy chemical industrial activity in Okinawa: the amount of wastewater generated from plastics manufacturing in Okinawa was 0.003  $\text{kg y}^{-1} \text{km}^{-2}$ , according to Pollutant Release and Transfer Register (PRTR) data (MOE, 2004), much lower than in large cities such as Tokyo and Osaka (0.076 and 0.116  $\text{kg y}^{-1} \text{km}^{-2}$ , respec-

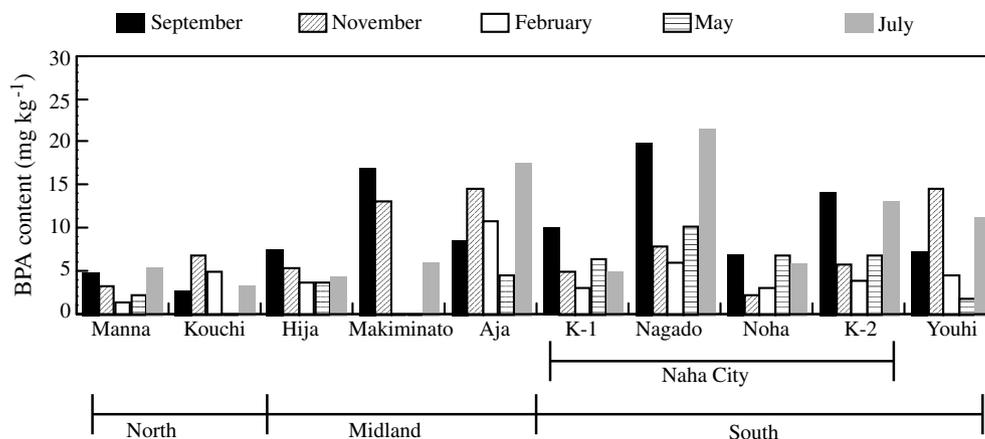


Fig. 3. Temporal variation of BPA concentrations at 10 selected sites.

tively). The PRTR data supports our results that most of the aquatic BPA in Okinawa is from domestic sources.

#### 4.2.2. Variation of BPA concentrations in sediments

BPA concentrations in sediments collected in July were higher than in February. These concentrations are determined by various factors such as adsorption, degradation and transport. Kitada et al. (2006) suggest that sediments might offer an advantage in evaluating contamination by BPA because BPA concentrations in sediments are higher than in river water in Okinawa. However, variation of BPA concentrations cannot be explained completely by local population density or adsorption. BPA in sediments generally degrades in aerobic conditions, but persists in anaerobic conditions (Ying and Kookana, 2003). BPA did not degrade at the Nagado and Noha river sites or at Site K-1 because these sites were under anaerobic conditions. Okinawa Prefecture monitoring data indicates that these sites were under anaerobic conditions because of high BOD (OPGDCE, 2003). On the other hand, even if BPA could degrade, BPA degradation was retarded because TOC contents were high at these sites (Kitada et al., 2006). Another source of variation in BPA concentration is the temperature-dependent rate of BPA leaching from PC tubes (Sajiki and Yonekubo, 2003). Rates of BPA leaching from PC tubes in one experiment were  $1.6 \text{ ng mL}^{-1} \text{ d}^{-1}$  at  $20^\circ\text{C}$  and  $11 \text{ ng mL}^{-1} \text{ d}^{-1}$  at  $37^\circ\text{C}$ . The air temperature in Okinawa was high in July and low in February (Supplementary material), which suggests strongly that the transport of BPA included in domestic wastewater in July was higher than in February due to increased leaching at higher temperatures. The BPA degradation under aerobic condition is likely dependent on temperature (Kang and Kondo, 2002). The degradation rate increases as temperature increases. Although BPA degraded at Makiminato site, which was under aerobic conditions, similar variation of BPA concentrations was observed. The seasonal variation in BPA concentration at Makiminato site supports the possibility that it is due to the degree of leaching of BPA from PC plastics rather than its degradation.

#### 4.2.3. Influence of BPA on coral endocrine systems

Endocrine systems in invertebrates are diverse and often different from those in vertebrates, although the basic endocrine strategy has been widely conserved. Recently, it has been shown that natural and synthetic estrogens, and some other estrogenic compounds, affect invertebrate development and reproduction (Segner et al., 2003; Brennan et al., 2006). For example,  $17\beta$ -estradiol was detected during a mass spawning event of scleractinian (hard) corals, although how  $17\beta$ -estradiol and estrogen-like compounds are synthesized and stored by corals, and have yet been elucidated (Atkinson and Atkinson, 1992; Tarrant et al., 2004). Meanwhile, Tarrant et al. (2004) report that chronic exposure to  $17\beta$ -estradiol causes abnormal reproduction in *Montipora capitata*, and exposure to dilute estrone ( $0.002 \mu\text{g L}^{-1}$ ) reduces growth rates in *Porites compressa*. These studies suggest that estrogenic compounds may alter steroid concentrations and rates of steroid metabolism in coral tissue, and affect physiological processes such as growth and reproduction. Although it is as much as  $10^4$  times less potent than  $17\beta$ -estradiol, BPA has been recognized as a relatively potent xenoestrogen to humans and wildlife in *in vivo* and *in vitro* studies (Krishnan et al., 1993). The adsorption coefficients of BPA are relatively high, with  $\log K_{\text{OW}}$  of 3.32 and  $\log K_{\text{OC}}$  of 3.18, suggesting that BPA will be adsorbed to sediments and little will be found in river water (Staples et al., 1998). In fact, previous studies reported low BPA concentrations in river water in Okinawa (Kawahata et al., 2004; Kitada et al., 2006). BPA concentrations in water in the area studied will be substantially lower than the effective concentration of BPA as an estrogenic stimulant for corals and coral reef organisms. Therefore, the BPA concentrations presently observed in Okinawa sediments are not likely to damage coral reef communities.

## 5. Conclusions

The hazardous chemical substances were present in river sediments and transported to coral reefs depending upon

their physico-chemical properties and applications by human activity. Diuron and chlorpyrifos were detectable in river sediments in both rural (Manna River) and urban areas (Naha City). Irgarol 1051 was frequently detected at the downstream of the river because of the low affinity to sediments and its low degradation rate in seawater. BPA was released into rivers because it leached from PC plastics used in house and contaminated the domestic wastewater.

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### Appendix A. Supplementary material

Supplementary material associated with this article can be found, in the online version, at doi:10.1016/j.chemosphere.2008.01.025.

### References

- Atkinson, S., Atkinson, M.J., 1992. Detection of estradiol-17 $\beta$  during a mass coral spawn. *Coral Reefs* 11, 33–35.
- Biselli, S., Bester, K., Huhnerfuss, H., Fent, K., 2000. Concentrations of the compound Irgarol 1051 and of organotins in water and sediments of German north and Baltic Sea marinas. *Mar. Pollut. Bull.* 40, 233–243.
- Bollmohr, S., Day, J.A., Schulz, R., 2007. Temporal variability in particle-associated pesticide exposure in a temporarily open estuary, Western Cape, South Africa. *Chemosphere* 68, 479–488.
- Bondarenko, S., Gan, J., 2004. Degradation and sorption of selected organophosphate and carbamate insecticides in urban stream sediments. *Environ. Toxicol. Chem.* 23, 1809–1814.
- Brennan, S.J., Brougham, C.A., Roche, J.J., Fogarty, A.M., 2006. Multi-generational effects of four selected environmental oestrogens on *Daphnia magna*. *Chemosphere* 64, 49–55.
- Carbery, K., Owen, R., Frickers, T., Otero, E., Readman, J., 2006. Contamination of Caribbean coastal waters by the antifouling herbicide Irgarol 1051. *Mar. Pollut. Bull.* 52, 635–644.
- Ciba-Geigy, 1988. Irgarol 1051 in antifouling paints. Technical Information Bulletin. Ciba-Geigy Ltd., Basel, Switzerland. 03/88-31.
- Fabricius, K.E., 2005. Effects of terrestrial runoff on the ecology of corals and reefs: review and synthesis. *Mar. Pollut. Bull.* 50, 125–146.
- Harrington, L., Fabricius, K., Eaglesham, G., Negri, A., 2005. Synergistic effects of diuron and sedimentation on photosynthesis and survival of crustose coralline algae. *Mar. Pollut. Bull.* 51, 415–427.
- Haynes, D., Muller, J., Carter, S., 2000. Pesticide and herbicide residues in sediments and sea grasses from the Great Barrier Reef World Heritage Area and Queensland coast. *Mar. Pollut. Bull.* 41, 287–297.
- Hughes, T.P., Baird, A.H., Bellwood, D.R., Card, M., Connolly, S.R., Folke, C., Grosberg, R., Hoegh-Guldberg, O., Jackson, J.B.C., Kleypas, J., Lough, J.M., Marshall, P., Nystrom, M., Palumbi, S.R., Pandolfi, J.M., Rosen, B., Roughgarden, J., 2003. Climate change, human impact, and the resilience of coral reefs. *Science* 301, 929–933.
- Imaoka, T., Hayashi, K., Ueda, T., Yoshimura, T., 2006. Investigation of the source and the path of bisphenol-A discharged from a housing development through domestic wastewater. *J. Environ. Syst. Eng. (JSCE)* 62, 332–339.
- Jergnetz, S., Mugni, H., Bonetto, C., Schulz, R., 2005. Assessment of insecticide contamination in runoff and stream water of small agricultural streams in the main soybean area of Argentina. *Chemosphere* 61, 817–826.
- Jones, R., 2005. The ecotoxicological effects of photosystem II herbicides on corals. *Mar. Pollut. Bull.* 51, 495–506.
- Jones, R.J., Kerswell, A.P., 2003. Phytotoxicity of photo system II (PSII) herbicides to coral. *Mar. Ecol. Prog. Ser.* 261, 149–159.
- Kang, J.H., Kondo, F., 2002. Effects of bacterial counts and temperature on the biodegradation of bisphenol A in river water. *Chemosphere* 49, 493–498.
- Kang, J.H., Kondo, F., 2005. Bisphenol A degradation in seawater is different from that in river water. *Chemosphere* 60, 1288–1292.
- Kawahata, H., Ohta, H., Inoue, M., Suzuki, A., 2004. Endocrine disrupter nonylphenol and bisphenol A contamination in Okinawa and Ishigaki Islands, Japan—within coral reefs and adjacent river mouths. *Chemosphere* 55, 1519–1527.
- Kitada, Y., Kawahata, H., Suzuki, A., Oomori, T., 2006. Concentrations of nonylphenol and bisphenol A in the Okinawa Island using liquid chromatography–electrospray tandem mass spectrometry. In: Proceedings of the 10th International Coral Reef Symposium, Okinawa, pp. 810–818.
- Krishnan, A., Stathis, P., Permceth, S., Tokes, L., Feldman, D., 1993. Bisphenol A: an estrogenic substance is released from polycarbonate flasks during autoclaving. *Endocrinology* 132, 2279–2286.
- Lam, K.H., Wai, H.Y., Leung, K.M.Y., Tsang, V.W.H., Tang, C.F., Cheung, R.Y.H., Lam, M.H.W., 2006. A study of the partitioning behavior of Irgarol-1051 and its transformation products. *Chemosphere* 64, 1177–1184.
- McMahon, K., Bengston Nash, S.M., Eagelshman, G., Muller, J.F., Duke, N., Winderrich, S., 2005. Herbicide contamination and the potential impact to seagrass meadows in Harvey Bay, Queensland, Australia. *Mar. Pollut. Bull.* 51, 325–334.
- Ministry of the Environment (MOE). Government of Japan, 2004. Pollutant Release and Transfer Register (PRTR). (in Japanese) (<<http://www.env.go.jp/chemi/prtr/risk0.html>>).
- Okinawa Prefecture Government Department of Culture and Environment (OPGDCE), 2003. Results of Water Quality Measurement in Okinawa: Public Water and Ground Water. Environmental Preservation Division Publication (in Japanese). p. 115.
- Omija, T., 2004. Terrestrial inflow of soils and nutrients. In: Tsuchiya, M., Nadaoka, K., Kayanne, H., Yamano, H. (Eds), *Coral Reefs of Japan*. Coral Reef Society, Tokyo, pp. 64–68.
- Racke, K.D., Lubinshki, R.N., Fontaine, D.D., Miller, R.J., McCall, P.J., Oliver, G.R., 1993. Comparative fate of chlorpyrifos insecticide in urban and agricultural environments. In: Racke, K.D., Leslie, A.R. (Eds), *In: Pesticides in Urban Environments: Fate and Significance*, 522. ACS Symposium Series, Washington, DC, pp. 70–85.
- Rowan, R., 1998. Diversity and ecology of zooxanthellae on coral reefs. *J. Phycol.* 34, 407–417.
- Sabljić, A., Gusten, H., Verhaar, H., Hermens, J., 1995. QSAR modelling of soil sorption. Improvements and systematics of log  $K_{oc}$  vs. log  $K_{ow}$  correlations. *Chemosphere* 31, 4489–4514.
- Sajiki, J., Yonekubo, J., 2003. Leaching of bisphenol A (BPA) to seawater from polycarbonate plastic and its degradation by reactive oxygen species. *Chemosphere* 51, 55–62.
- Segner, H., Carroll, K., Fenske, M., Janssen, C.R., Mack, G., Pascoe, D., Schafers, C., Vandenberg, G.F., Watts, M., Wenzel, A., 2003. Identification of endocrine-disrupting effects in aquatic vertebrates and invertebrates: report from the European IDEA project. *Ecotoxicol. Environ. Saf.* 54, 302–314.

- Staples, C.A., Dorn, P.B., Klecka, G.M., O'Block, S.T., 1998. A review of the environmental fate, effects, and exposures of bisphenol A. *Chemosphere* 36, 2149–2173.
- Tarrant, A.M., Atkinson, M.J., Atkinson, S., 2004. Effects of steroidal estrogens on coral growth and reproduction. *Mar. Ecol. Prog. Ser.* 269, 121–129.
- Tashiro, Y., Takemura, A., Fujii, H., Takahira, K., Nakanishi, Y., 2003. Livestock wastes as a source of estrogens and their effects on wildlife of Manko tidal flat, Okinawa. *Mar. Pollut. Bull.* 47, 143–147.
- Thomas, K.V., McHugh, M., Hilton, M., Waldock, M., 2003. Increased persistence of antifouling paint biocides when associated with paint particles. *Environ. Pollut.* 123, 153–161.
- Thomas, K.V., McHugh, M., Waldock, M., 2002. Antifouling paint booster biocides in UK coastal waters: inputs occurrence and environmental fate. *Sci. Total Environ.* 293, 117–127.
- West, K., Van Woesik, R., 2001. Spatial and temporal variance of river discharge on Okinawa (Japan): inferring the temporal impact on adjacent coral reefs. *Mar. Pollut. Bull.* 42, 864–872.
- Ying, G.G., Kookana, R.S., 2003. Degradation of five selected endocrine-disrupting chemicals in seawater and marine sediment. *Environ. Sci. Technol.* 37, 1256–1260.
- Yoshida, S., Taguchi, S., Hori, S., 2004. Chlorpyrifos and S-421 residues in indoor air and polished rice around nine years after application for termite control. *J. Soc. Indoor Environ. Japan* 7, 7–15.