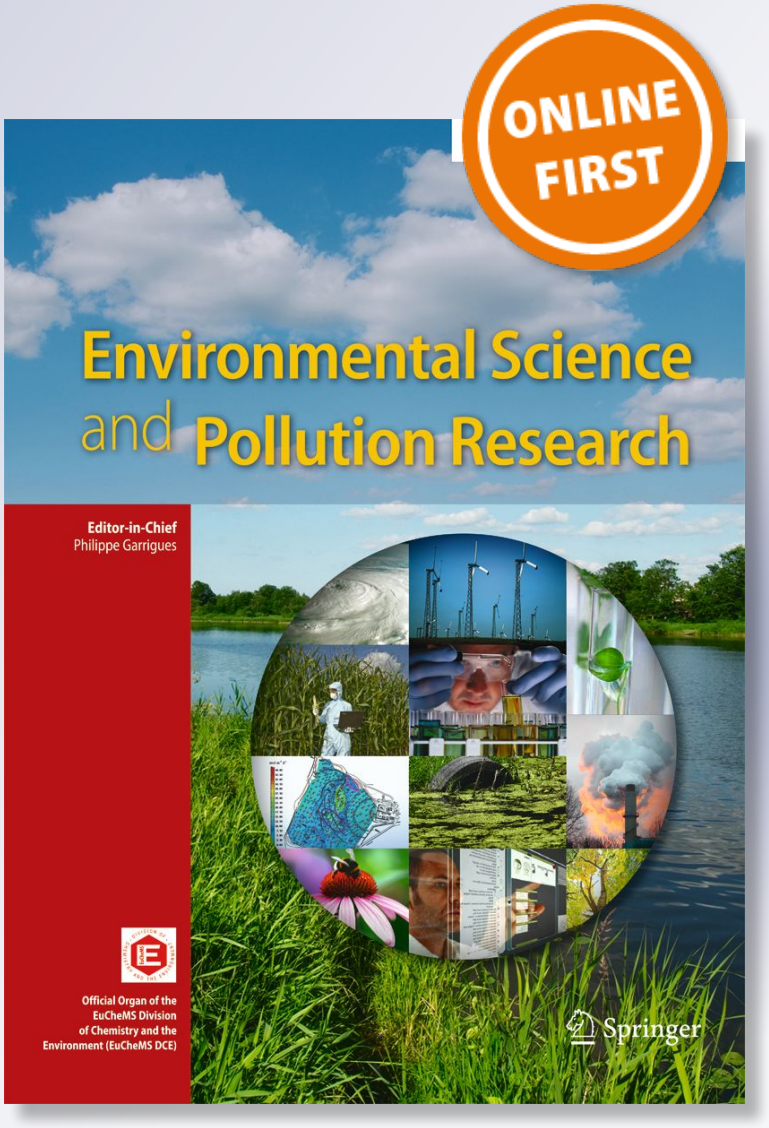
Investigation of environmental contamination of mono-isopropylnaphthalene, di-isopropylnaphthalene and tri-isopropylnaphthalene in Hyogo in Japan

Motoharu Suzuki, Chisato Matsumura, Takeshi Nakano & Hiromasa Imaishi

Environmental Science and Pollution Research

ISSN 0944-1344

Environ Sci Pollut Res
DOI 10.1007/s11356-012-0987-9



Springer

Environ Sci Pollut Res
DOI 10.1007/s11356-012-0987-9

RESEARCH ARTICLE

Investigation of environmental contamination of mono-isopropylnaphthalene, di-isopropylnaphthalene and tri-isopropylnaphthalene in Hyogo in Japan

Motoharu Suzuki · Chisato Matsumura · Takeshi Nakano · Hiromasa Imaishi

Received: 29 December 2011 / Accepted: 15 May 2012

Abstract Di-isopropylnaphthalene (DIPN) has highly persistent and bioaccumulative properties, and a large amount of DIPN is used as a PCB substitute in Japan. However, DIPN in the environment has not been thoroughly investigated. In addition, mono-isopropylnaphthalene (MIPN) and tri-isopropylnaphthalene (TIPN), which are the homologues of DIPN, have similar properties to DIPN. In this study, simultaneous analytical methods for MIPN, DIPN, and TIPN for air, environmental water, sediment, and biological samples were developed, and the resultant contamination caused by each in the environment was investigated. DIPN was detected at 1.1±0.38 ng/m^3 in air and between <1.9 and 9.8 ng/L in river water, but MIPN and TIPN were not. In *Lateolabrax japonicas* (Japanese sea perch), TIPN was detected from only females at between 0.65 and 1.4 ng/g-wet. DIPN was detected from all perches at between 1.2 and 3.4 ng/g-wet. DIPN and TIPN isomer fingerprints in females were different from those in the reference standard stock solution ones. In sediments, MIPN, DIPN, and TIPN were detected at between <0.16 and 8.6 ng/g-dry, between <1.1 and 4400 ng/g-dry, and between <0.83 and 500 ng/g-dry, respectively. The contamination trend of DIPN in the sediments was similar to that of PCBs.

Keywords Mono-isopropylnaphthalene · Di-isopropylnaphthalene · Tri-isopropylnaphthalene · PCB substitute · Alkylnaphthalene

Responsible editor: Hongwen Sun

M. Suzuki (✉) · H. Imaishi
Graduate School of Agricultural Science, Kobe University,
1-1 Rokkodai-cho, Nada-ku,
Kobe, Hyogo 657-8501, Japan
e-mail: Motoharu_Suzuki@pref.hyogo.lg.jp

C. Matsumura · T. Nakano
Hyogo Prefectural Institute of Environmental Sciences,
3-1-27 Yukihiracho, Suma-ku,
Kobe, Hyogo 654-0037, Japan

T. Nakano
Graduate School of Engineering, Osaka University,
1-1 Yamadaoka, Suita,
Osaka 566-0871, Japan

H. Imaishi
Research Center for Environmental Genomics, Kobe University,
1-1 Rokkodai-cho, Nada-ku,
Kobe, Hyogo 657-8501, Japan

Introduction

Di-isopropylnaphthalene (DIPN) has been increasingly used as a PCBs substitute since the 1970s in Japan (Peterman and Delfimo 1990). By the 1990s, Japan produced and imported DIPN at a rate of about 6000 tons annually (data provided by the Ministry of the Environment (MOE)). DIPN had been considered to be a safe chemical as a result of testing the biodegradation property of DIPN (Yoshida and Kojima 1978a, b). The alkyl side chains of DIPN protect the aromatic ring during metabolism (Kojima et al. 1982). Therefore, the effect of DIPN is dissimilar to that of the parent compound and those with small alkyl side chains (Hoke and Zellerhoff 1998). Recently, the toxicity of DIPN to marine mussels was reported to be considerably less than that of smaller alkyl-substituted naphthalenes (Hoke and Zellerhoff 1998; Scarlett et al. 2011). Based on test results on ecological effect by MOE in 2006, several biological influences of DIPN have been reported, for example acute inhibition of swimming for *Daphnia magna* (48-h EC50=0.035 mg/L) and toxicity for fish designed as OECD classification standard (48-h LC50 of *Oryzias latipes*=4.5 mg/L). A

Published online: 26 July 2012

carcinogenic potential of DIPN has not been clear, and it later became clear that DIPN has highly persistent and bioaccumulative properties (data provided by the National Institute of Technology and Evaluation (NITE)). As a result, it was designated as a type I Monitoring Chemical Substance by the Chemical Substances Control Law of Japan in 2004. The Monitoring Chemical Substances are expected to assess environmental contaminations in Japan.

Since 2004 DIPN usage has decreased, but the compound is still used at the rate of about 600 tons annually (data provided by the MOE). In 2007, a total of 813 tons of DIPN was used in Japan: 431 tons of DIPN was used for the solvent of the carbonless duplicating paper, 206 tons as a heat carrier, 147 tons as a solvent for the pesticides and others, 17 tons for other uses, and 12 tons for export.

In addition to DIPN, mono-isopropylnaphthalene (MIPN) and tri-isopropylnaphthalene (TIPN), which are homologues of DIPN, are also used as a heat carrier and contaminated in the solvent. MIPN and TIPN were also designated as type III and type I Monitoring Chemical Substances in 2004.

Because MIPN, DIPN, and TIPN are used as solvents, there is a concern that these compounds widely contaminate the environment. Food contamination of DIPN from wrapping with recycled paper has been reported by Sturaro et al. (1994) and Mariani et al. (1999). MIPN and DIPN in environmental water, sediment, and biological samples have been measured in Japan (Kodama 1981; MOE1980; 1983; 1985; 1997); however, only 2,6-DIPN and 2,7-DIPN were assigned. Though nonpolar column was used for gas chromatography in these reviews, it could not help separate DIPN isomers enough (Brzozowski et al. 2002). Therefore, other isomer peaks might overlap with 2,6-DIPN or 2,7-DIPN. Not only DIPN but also MIPN and TIPN are usually used as isomer mixtures; therefore, it is important to assign the isomer fingerprints of MIPN, DIPN, and TIPN to reveal their fates in the environment. Recently, in the field of chemosynthesis, studies into DIPN isomerization have now succeeded in the isolation and identification of DIPN isomers (Brzozowski et al. 2001, 2002). On the other hand, DIPN and TIPN isomers in the environmental field have not been sufficiently assigned. Sturaro et al. reported that the paper recycling and waste disposal process is a source of DIPN pollution (1994). We investigated the MIPN, DIPN, and TIPN contaminations in water, air, sediment, and algae samples obtained in the vicinity of a paper recycling plant (Suzuki et al. 2007). Though the analytical method had some problems in DIPN contamination at pre-treatment and recovery ratios of MIPN, DIPN, and TIPN, high concentrations of DIPN were detected in the wastewater and the exhaust from the plant recycling plant, but concentrations of MIPN and TIPN were relatively low in comparison. In 2007 and 2008, simultaneous analytical methods with GC/MS for MIPN, DIPN, and TIPN in water, sediment, air, and biological samples were improved. As the result, more detail of contaminations of MIPN, DIPN, and TIPN in the environment could be analysed, and the methods for sediment, air, and biological samples were adopted as official methods by MOE (2009). The method for water was also adapted but only for DIPN (MOE 2008a).

In recent years, the environmental contamination of DIPN in Japan has been researched by MOE using these methods. MIPN, DIPN, and TIPN are hydrophobic and persistent like PCBs; therefore, they are likely to accumulate in the environment, as replacements for PCBs. They will likely follow similar patterns of contamination as the priority pollutants. The concentration of residual PCBs in sediment was known to be higher than air and water (Nakano et al. 2004).

In this study, the concentrations and isomer fingerprints of MIPN, DIPN, and TIPN in sediments were investigated with PCBs in Hyogo Prefecture in Japan in 2010. Furthermore, some water, air, and biological samples were investigated to verify the distribution of these compounds in the environment.

Methodology section

Reagents and cartridges

2, 6-DIPN (> 99 %), 2-MIPN (> 95 %), and 1-MIPN (> 85 %) were obtained from Kanto Chemical Co., Inc (Tokyo, Japan). Technical stock solutions of MIPN, DIPN, and TIPN (used as heat carriers in industry) were provided by Soken Tecnic Co., Ltd. (Tokyo, Japan). They were used as reference standards, to identify the isomer retention time or to perform the accuracy control test. The individual isomer reference standard stock solutions of DIPN were provided by Dr. Brzozowski (Industrial Chemistry Research Institute, Poland). MBP-CG, which was a PCB isotope mixture ($^{13}C_{12}$-MoCB–DeCB), and MBP-70 ($^{13}C_{12}$-2,3′,4′,5-tetrachlorobiphenyl) were obtained from Wellington Laboratories Japan Inc. (Tokyo, Japan) for use as a PCB surrogate and syringe spike, respectively. Hexachlorobenzene-$_{13}C^6$ (HCB-$^{13}C_6$) for use as a syringe spike and other solvents was obtained from Kanto Chemical Co., Inc. The Supelclean ENVI™-Carb and LC-Silica cartridges were obtained from Sigma-Aldrich Japan K.K. (Tokyo, Japan) for use in

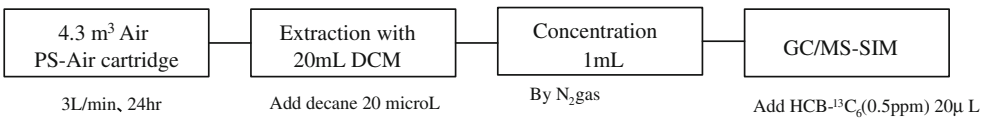


Fig. 1 The analytical procedure of MIPN, DIPN, and TIPN for air

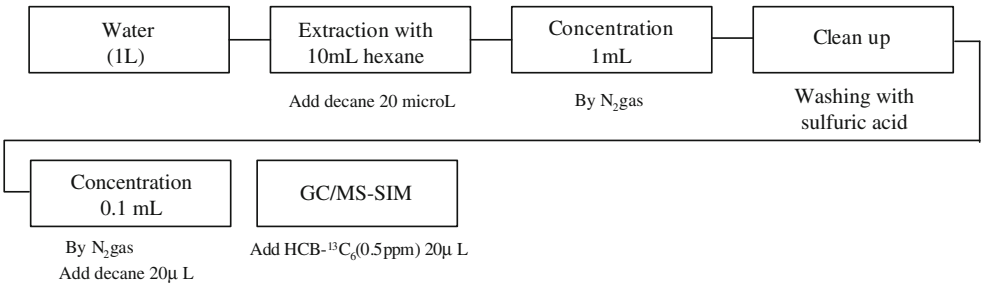


Fig. 2 The analytical procedure of MIPN, DIPN, and TIPN for environmental water

cleanup. The Sep-Pak Plus PS-Air cartridge was obtained from Nihon Waters K.K. (Tokyo, Japan) for air sampling.

Analytical procedure

The simultaneous analytical methods with GC/MS for DIPN, MIPN, and TIPN in air, environmental water, sediment, and biological samples are shown in Figs. 1, 2, 3, and 4, respectively.

A Supelclean ENVI™-Carb cartridge was chosen as a cleanup cartridge for sediments, because compared with silica gel cartridges, the cartridge had less DIPN contamination and DIPN as a remaining pigment inside was easily washed by hexane. DIPN contained in the environmental water might be acquired on the surface of a sampling bottle, because of its hydrophobic property; Log P was 4.90 by Hazardous Substances Data Bank (HSDS). Therefore, the environmental water was directly sampled in a 1-L glass bottle, and hexane extraction had to be done inside the bottle. It also had efficacy for preventing DIPN contamination, as DIPN was notably ubiquitous.

All the measurements were performed using an Agilent GC/MS system consisting of a 6890 N gas chromatograph equipped with a SPELCOWAX-10 capillary column (30-m length, 0.32-mm i.d., 0.25-μm film thickness) and Q1000GC mass spectrometer. The injection volume was 2 μm, and the temperature was 270°C. The carrier gas was helium at 1.2 mL/min. The oven temperature was 50°C with 1-min hold at first; it was elevated to 100°C by 20°C/min, 210°C by 5°C/min, and 270°C with 1-min hold by 20°C/min. The ionization method was EI (200 μA, 70 eV). The temperatures of the interface and ion source were 240 and 210°C, respectively. The *m/z* for the quantification of MIPN, DIPN, and TIPN were 170, 212, and 254, respectively. The SIM chromatograms of the mixture of MIPN, DIPN, and TIPN stock solutions are shown in Fig. 5. MIPN, DIPN, and TIPN in the amount of 0.2 ng were injected to GC/MS. Their isomers were successfully separated using a polar column. DIPN isomers have been reported not to be sufficiently separated when using intermediate or nonpolar columns (Brzozowski 2004). The eight major isomer peaks of DIPN were assigned by checking the retention times of individual isomer stock solutions of DIPN provided by Brozozouski. In the case of TIPN, the six isomer peaks of TIPN were detected, although they were not assigned. The concentrations of MIPN, DIPN, and TIPN in the samples were calculated with standard curves made from 2-IPN, 2,6-DIPN, and a stock solution of TIPN.

The method detection limits (MDLs), the quantification limits (MQLs), and the recovery ratios for MIPN, DIPN, and TIPN are shown in Table 1. These were calculated using stock solutions and according to the MOE methods. The detection limit of a single isomer peak (2,6-DIPN) for water and sediment was 0.71 ng/L and 0.17 ng/g-dry, respectively, with sensitivities about ten times higher than those reported by MOE (1997).

The PCBs measurement for the sediments was according to the MOE method using an Agilent GC/MS system consisting of a 6890 N gas chromatograph equipped with an HT-8PCB capillary column (60-m length, 0.25-mm i.d.) and

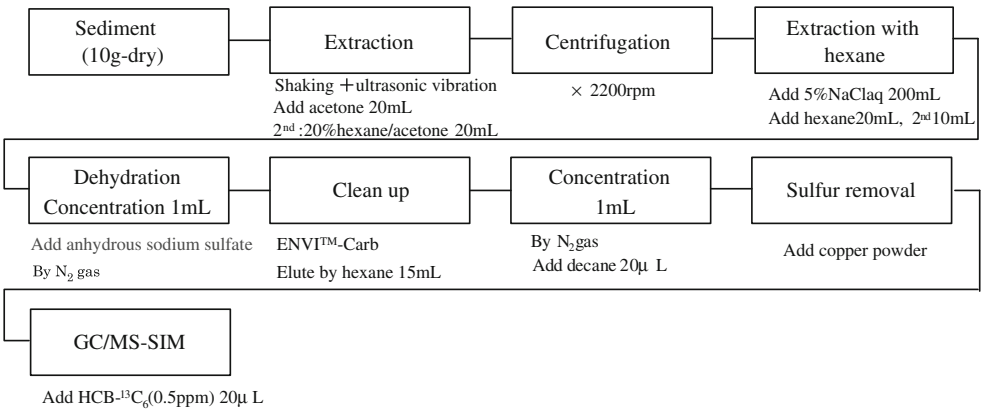


Fig. 3 The analytical procedure of MIPN, DIPN, and TIPN for sediment

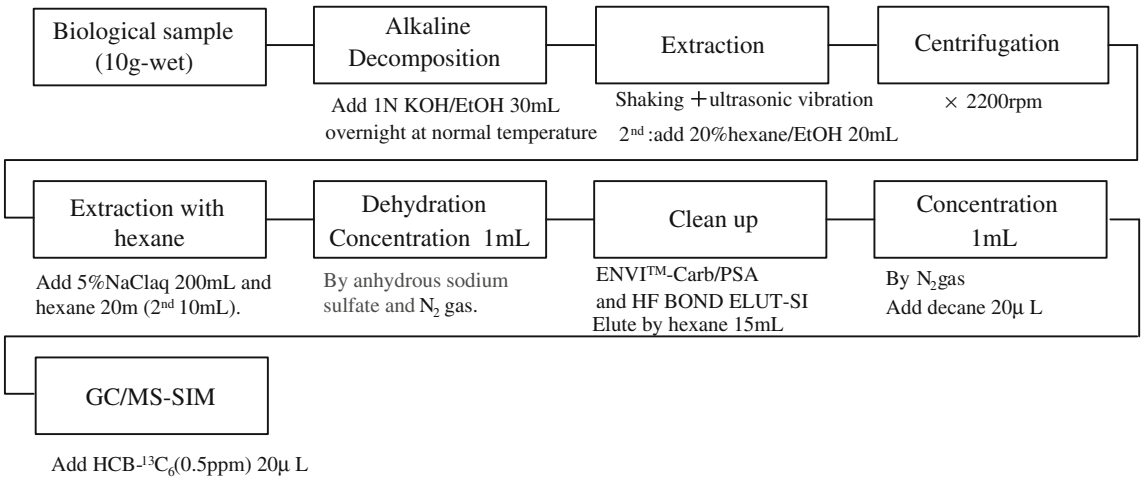


Fig. 4 The analytical procedure of MIPN, DIPN, and TIPN for biological sample

an 800D high-resolution mass spectrometer. The PCBs concentrations were calculated by a surrogated method.

Air sampling method

Figure 6 shows the relationship of the air sampling time (1, 2, 3, 16, and 24 h) to the trapping amount and the air concentration of DIPN. The test was conducted in a temperature-controlled room. The trapping amounts of DIPN increased with the sampling time. The air concentration of DIPN was constant at the level of approximately 15 ng/m^3. Although it was possible to assess the air concentration of DIPN with an hour of sampling time, it was better to use a long sampling time to eliminate the DIPN contamination in the PS-Air cartridge and during pre-treatment. Sampling with a circuit of four cartridges revealed that there was no breakthrough from the first cartridge after 24 h of sampling. Storage of MIPN, DIPN, and TIPN in a PS-Air cartridge was stable for at least a week.

Sampling site

The sampling sites studied are shown in Fig. 7. The air was sampled at the floor and the roof of the institute in Kobe City at an urban site near point S6. The waters were sampled at all river points and at sea point S27 in 2009. The sea sediments were sampled using an Ekman-Birge-type bottom sampler at 41 points located in the Seto Inland Sea and Osaka Bay in southern Hyogo Prefecture in 2009 and 2010. The river sediments were sampled with a dredge at eight rivers that empty into the sea except for points R7 and R8 in 2010. *Lateolabrax japonicas* (Japanese sea perch) as biological samples were sampled in the vicinity of point S27. Five samples were made up from a filleted perch or by mixing fillet of two or three perches for each.

Results and discussion

Air, water, and fish concentrations of MIPN, DIPN, and TIPN

Table 2 shows the air concentrations of MIPN, DIPN, and TIPN at the floor and the roof of the institute in Kobe City at an urban site near point S6. There was very little MIPN and TIPN contamination outdoors and indoors. The outdoor air concentration of DIPN was 1.1±0.38 ng/m^3. No differences

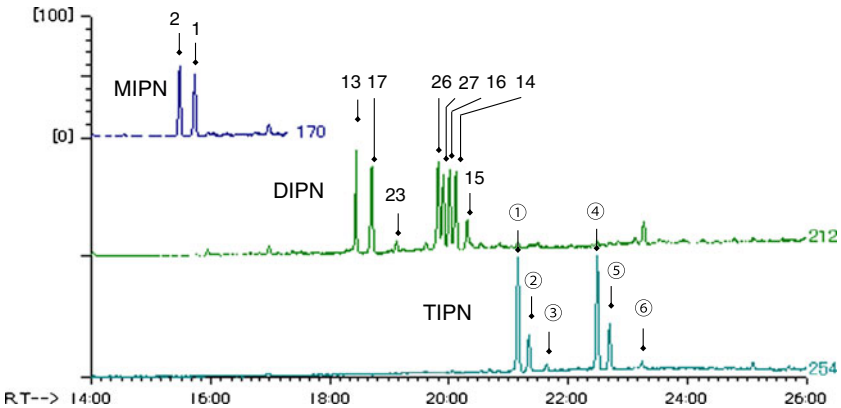


Fig. 5 The SIM chromatogram of a mixture of MIPN, DIPN, and TIPN stock solutions. Their concentrations were 100 μg/mL each

Table 1 Recovery ratios, method detection limits (MDLs), and quantification limits (MQLs) for MIPN, DIPN, and TIPN calculated by the MOE method

	Water			Air			Sediment			Biological sample		
	Recovery (n=3) (%)	MDL (ng/L)	MQL (ng/L)	Recovery (n=3) (%)	MDL (ng/m^3)	MQL (ng/m^3)	Recovery (n=5) (%)	MDL (ng/g-dry)	MQL (ng/g-dry)	Recovery (n=4) (%)	MDL (ng/g-wet)	MQL (ng/g-wet)
MIPN	75±4.5	1.8	4.6	97±5.5	0.46	1.2	64±3.4	0.16	0.41	73±8.6	0.15	0.38
DIPN	77±4.0	1.9	5.0	102±8.6	0.48	1.2	86±3.8	1.1	2.9	79±11	0.60	1.6
TIPN	76±5.9	5.6	14	102±18	0.23	0.59	90±1.7	0.82	2.1	79±13	0.52	1.3

in concentrations were observed for air sampled in warm and cold periods. The indoor contamination level was almost ten times higher than outdoors. This was attributed to the fact that DIPN was volatilized from the ink solvent used in a room at the location. The isomer fingerprints of DIPN in the air samples had almost the same pattern as that in the reference standard stock solution. In our past study (Suzuki et al. 2007), the air contamination level of DIPN around point R7, in the vicinity of the location of a paper recycling plant, was 16 ng/m^3, which was almost ten times higher than that obtained this time. In 2009, the air contamination levels of DIPN in Japan were investigated at 20 points, including the point of this study, with this method by MOE (2011). With the exclusion of that at the highest point (22 ng/m^3), the contamination level of DIPN was between 1 and 10 ng/m^3.

Table 3 shows the water concentrations of MIPN, DIPN, and TIPN in the river and the sea sampled in 2009. MIPN and TIPN were not detected at all points. DIPN was detected between <1.9 and 9.8 ng/L. The isomer fingerprints of DIPN in the water samples had almost the same pattern as that in the reference standard stock solution. In 2006 and 2007 by MOE (2008; 2009), the water contamination levels researched at 20 points in Japan, including the point S27 and R7, were between <0.83 and 3.2 ng/L. The contamination levels

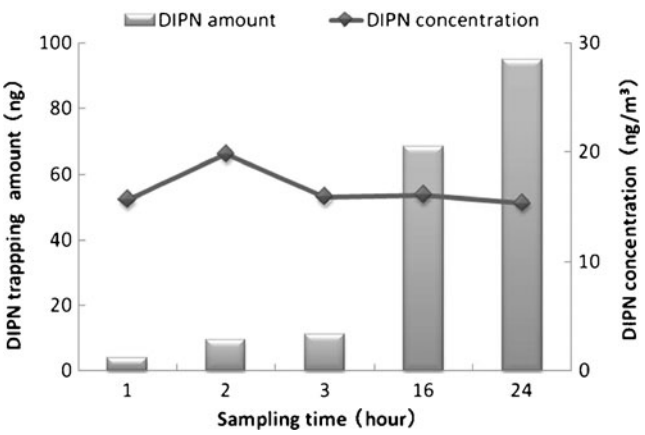


Fig. 6 The relation of air sampling time to air trapping amount and air concentration of DIPN

of DIPN at points R1, R3, and R4 were higher than those at others. It was speculated that there were contamination sources of DIPN in their upper stream.

Table 4 shows the concentrations of MIPN, DIPN, and TIPN in the perches sampled around point S27. Samples 1 and 2 were extracted from two and three male perches. Sample 3, 4, and 5 were made from three, two, and one female perches, respectively. Although MIPN was detected in almost all samples, it was below the quantification limit. TIPN was detected only in female samples at concentrations between 0.65 and 1.4 ng/g-wet. Hasegawa et al. (1982) reported that DIPN was accumulated in mainly body fat and subcutaneous fat. The average of fat contents of female perches was 1.2±0.22 %, which was higher than that of male perches (0.60±0.12 %). TIPN was detected only in female perches. This may be due to the high hydrophobicity of TIPN (Log P=7.54; KOWWIN v1.68) and therefore more likely to partition into the lipids of the females which possess relatively higher fat content than males. The TIPN concentrations on per-lipid basis were 1.1 ng/g lipid.

The detected isomers of TIPN were peak Nos. 2, 4, and 5 shown in Fig. 5, and the pattern was the same as that in the reference standard stock solution one. DIPN was detected from all samples between 1.2 and 3.4 ng/g-wet. There was no difference in the concentration of DIPN in males and females. Although the metabolism of 2,6-DIPN in carp (*Cyprinus carpio*) had been identified by Kojima et al. (1982), other isomers had not been researched yet. In this study, the isomer patterns of DIPN in female samples were different from those in male samples. In male samples, the DIPN isomer patterns were almost the same as those in the reference standard stock solution one. On the other hand, 1, 3-DIPN and 1, 4-DIPN were mainly detected in the female samples. The reason that the isomer fingerprints of DIPN and TIPN detected in the perches were different from those in the reference standard stock solutions was that the degradation or accumulation of each isomer was different in parch.

Figure 8 shows the sediment concentrations of MIPN, DIPN, and TIPN in the sediment samples in 2010. MIPN was detected at between <0.16 and 8.6 ng/g-dry, with a

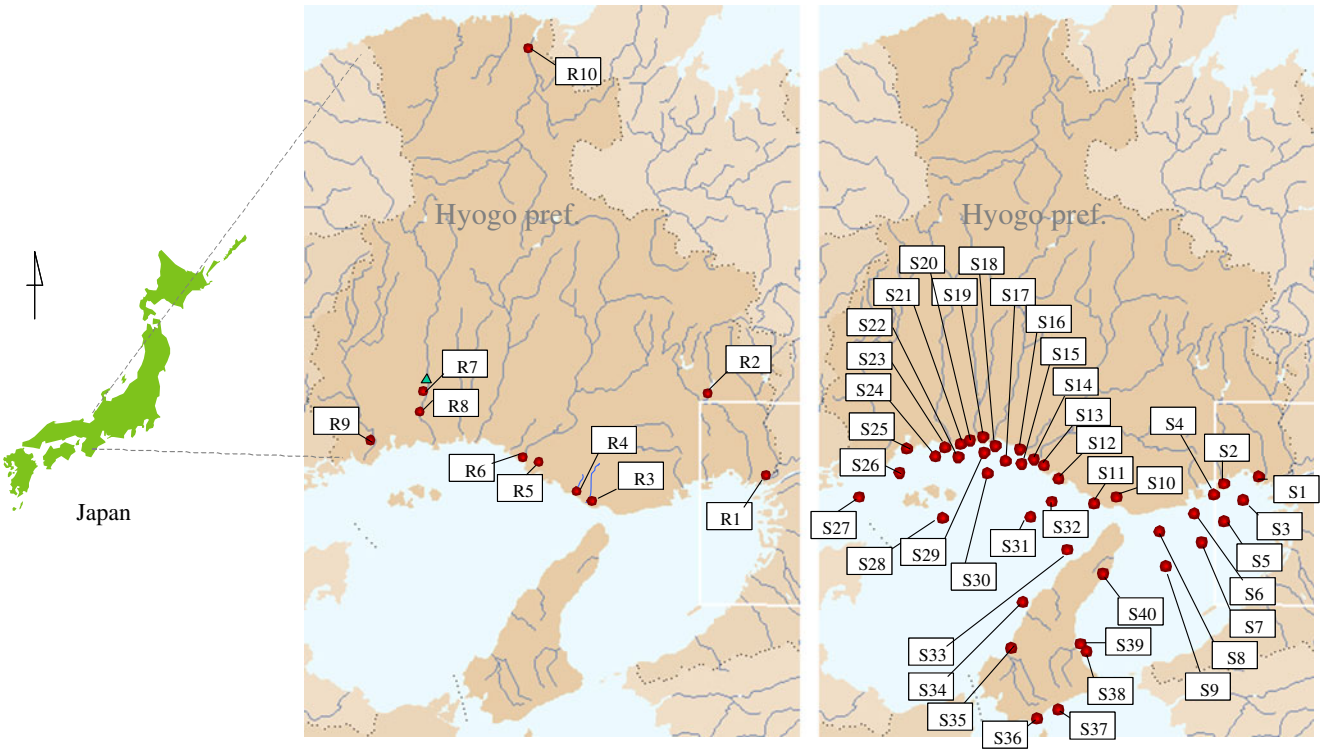


Fig. 7 The sampling points of air, water, sediment, and biological samples in Hyogo Prefecture. A *triangle point* which is upper stream of point S7 indicates a paper recycling plant

median concentration of 0.27 ng/g-dry; DIPN, between < 1.1 and 4400 ng/g-dry (median=1.9 ng/g-dry); and TIPN, between <0.83 and 500 ng/g-dry (median=0.20 ng/g-dry). In the sea area, the concentrations in the coast were higher than those offshore. The highest concentrations of MIPN, DIPN, and TIPN were 1.7, 100, and 9.0 ng/g-dry at point S21, respectively. Point S21 is the bay where the Ibo River empties. The effluent from a paper recycling plant flows into the Ibo River, and DIPN and TIPN were detected at 290 and 3.3 ng/g-dry in the sediment at point R7 in our previous study (Suzuki et al. 2007). In the river, the highest concentrations of MIPN, DIPN, and TIPN were detected: 8.6, 4400, and 500 ng/g-dry at point R1, respectively. In Germany, DIPN had been detected at between <1 and 83 ng/g in two river sediments in 1999–2001 by Franke et al. (2007). In Japan, the sediment contamination levels of DIPN were also investigated with this method at 30 points in 2009 by MOE (2011), and the highest concentration was 230 ng/g-dry. Therefore, point R1 was the highest polluted area of DIPN in Japan. Given that the isomer pattern matched that of the technical mixtures of MIPN, DIPN, and TIPN, it is likely that the source of the sediment contamination was industrial. However, no paper recycling industry exists in the vicinity of this sampling station.

Figure 9 shows the concentration ratio of MIPN, DIPN, and TIPN at each sampling point where DIPN was detected above MDL. The main contaminant was DIPN at every point in this study, and its average ratio was stable at 79±

Table 2 Air concentrations of MIPN, DIPN, and TIPN at the floor and the roof of the institute in Kobe City at an urban site near point S6

Sampling location	Date	Air concentration (ng/m^3)		
		MIPN	DIPN	TIPN
Floor of the institute	7–8 Feb 2008	0.84	10	< 0.23
	5–6 May 2008	< 0.46	16	< 0.23
Roof of the institute	8–9 May 2008	< 0.46	1.2	< 0.23
	6–7 May 2008	< 0.46	0.76	< 0.23
	25–26 Nov 2009	< 0.46	0.95	< 0.23
	26–27 Nov 2009	< 0.46	1.7	< 0.23
	27–28 Nov 2009	< 0.46	1.0	< 0.23

Table 3 Water concentrations of MIPN, DIPN, and TIPN at river sampling points

Sampling point	Water concentration (ng/L)		
	MIPN	DIPN	TIPN
R1	< 1.8	9.8	< 5.6
R2	< 1.8	2.5	< 5.6
R3	< 1.8	5.1	< 5.6
R4	< 1.8	7.6	< 5.6
R5	< 1.8	3.6	< 5.6
R6	< 1.8	3.5	< 5.6
R7	< 1.8	2.6	< 5.6
R8	< 1.8	1.9	< 5.6
R9	< 1.8	< 1.9	< 5.6
R10	< 1.8	< 1.9	< 5.6
S27	< 1.8	< 1.9	< 5.6

11 %. Although MIPN and TIPN were used as a PCB substitute, just like DIPN, the primary cause was speculated to be contamination to DIPN products.

According to the prediction of the environmental media partition of DIPN by level III Fugacity Model, Estimation Programs Interface (EPI) Suite v3.20, 56 % of DIPN discharged into the environment is distributed to sediment, 41 % to soil, 3.4 % to water, and 0.1 % to air.

Comparing the median concentrations of DIPN detected from air, environmental water and sediment samples, which were 0.0010, 3.1, and 1900 ppt, the median concentration of sediments was 620 times higher than that of waters. It indicates that DIPN in the environment was accumulated more to sediments than calculated. The median concentration of DIPN detected from perches was the same level as that of the sediment. The bioaccumulation tests by NITE show that DIPN accumulated 6100 times higher to carp in 5 μg/L of water and 2400 times in 0.5 μg/L of water. In this study, the DIPN concentration of sea water sampled at the point where the perches were sampled was below MDL; therefore, by contrast with all water samples, the medium concentration of DIPN from perches was 550 times higher than the waters. The cause that the accumulation value was lower than NITE test might result from the lower water concentration (0.0031 μg/L) compared with the tests. It indicates that in the environment DIPN was also accumulated to perches.

The water concentrations of DIPN would be safety levels for livings because these were below the concentration that caused acute inhibition of swimming for *D. magna* (48-h EC50=0.035 mg/L). However, the sediment concentrations detected from points S21 and R1 were 100 ng/g-dry and 4400 ng/g-dry, which might cause the acute inhibition for livings in the sediment. Moreover, the concentration of point R1 was over 4.5 mg/L, which was 48-h LC50 of *O. latipes*, and might cause adverse effects on fishes. The sediment concentration of TIPN at point R1 was also much higher than that at other points; however, toxicity of TIPN was little revealed. As a result, especially at point R1, there might be some negative impacts for livings.

Correlation between the DIPN and PCBs concentration in the sediments

The sediment samples were also analysed for the PCBs concentrations. Figure 10 shows the correlation between the DIPN and PCBs concentration in the sediments. PCBs were detected to be between 0.16 and 1700 ng/g-dry. The concentrations of DIPN tended to increase as those of PCBs increased. In addition, the highest contamination point for DIPN was also the highest for PCBs in both the river and the sea.

The coefficient of determination of correlation between the sediment concentrations of DIPN and PCBs at all points except for points S1, S21, and R1 was 0.619. The concentrations of DIPN at points S1, S21, and R1 were much higher than that of PCBs compared with other points; therefore, there would be some pollution sources of DIPN around the points S1, S21, and R1.

A concentration of 3, 3′-dichlorobiphenyl (CB11) is considered to be an indicator of the influence of the paper recycling plant, because CB11 is reported to derive from 3,3′-dichlorobenzidine salts which are used for printing ink (King et al. 2002). A ratio of CB11 in total dichlorobiphenyls (CB11/ΣDiCBs) at point S21, where the highest concentration of DIPN was detected in the sea, was 21 %, slightly higher than that of other sea samples (average 17 %), and the concentration of CB11 was 0.30 ng/g-dry higher than that of others (average 0.17 ng/g-dry). At point R1, where the highest concentration of DIPN was detected, the concentration of CB11 was 3.8 ng/g-dry higher, but that of CB11/ΣDiCBs was 2.5 % lower in all samples. Although

Table 4 MIPN, DIPN, and TIPN concentrations of filleted perches *(Lateolabrax japonicas*) sampled at point S27. Samples 1 and 2 were made from female perches, and samples 3, 4, and 5 were made from males

	Perch concentration (ng/g-wet)		
	MIPN	DIPN	TIPN
Sample 1	0.23	1.7	< 0.52
Sample 2	0.37	1.8	< 0.52
Sample 3	0.19	1.3	0.67
Sample 4	0.19	3.4	1.4
Sample 5	< 0.15	1.2	0.65

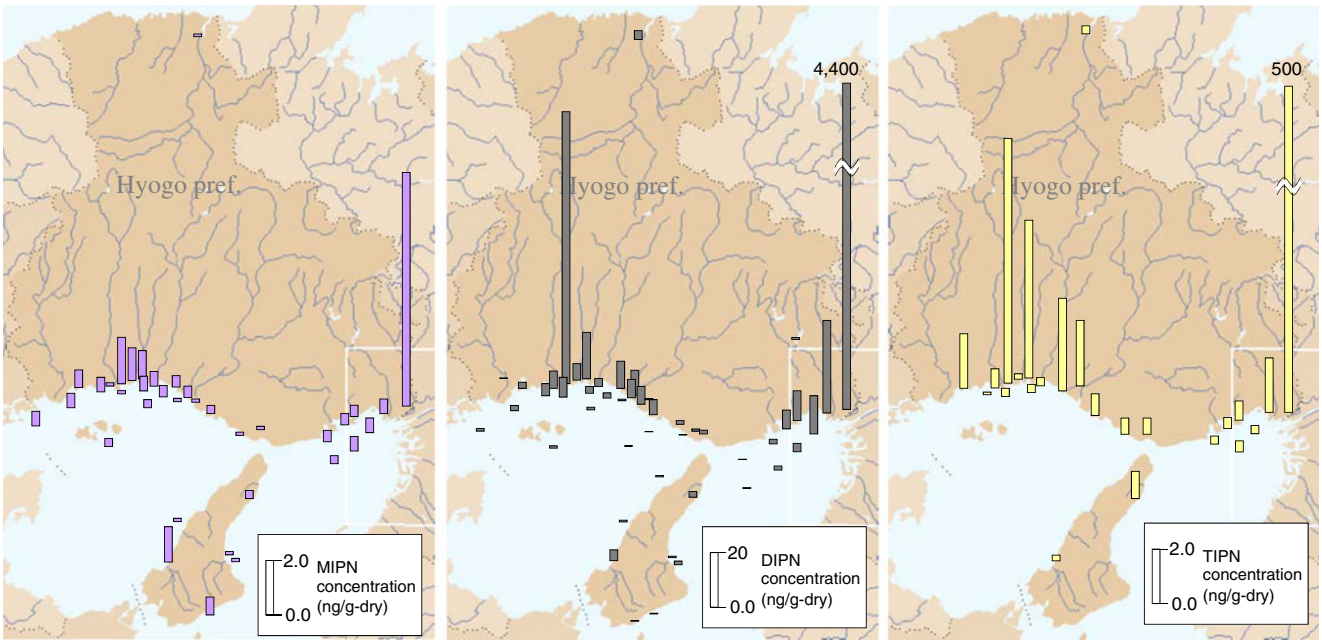


Fig. 8 The sediment concentration of MIPN, DIPN, and TIPN in sediment samples in 2010

point R1 was one of the largest urban rivers, where many kinds of industries are located, there was no paper recycling plant upstream of point R1. The contamination source of DIPN in this river needs to be clarified.

Conclusions

Simultaneously analytical methods of MIPN, DIPN, and TIPN in air, water, sediment, and biological samples satisfying the standard to be official methods of Ministry of Environment (MOE) were developed, and the environmental contaminations in Hyogo Prefecture were quantified. In air and river water, MIPN and TIPN were not detected in all samples. DIPN was detected outdoors at 1.1±0.38 ng/m^3, a value which was almost ten times lower than that indoors. DIPN between <1.9 and 9.8 ng/L was detected in river water. In *L. japonicas* (Japanese sea perch), TIPN was detected from only females at between 0.65 and 1.4 ng/ g-wet. DIPN was detected from all perches at between 1.2 and 3.4 ng/g-wet. DIPN and TIPN isomer fingerprints in females were different from those in the reference standard stock solution ones.

In the sediment, MIPN was detected at between <0.16 and 8.6 ng/g-dry, DIPN between <1.1 and 4400 ng/g-dry, and TIPN between <0.83 and 500 ng/g-dry. The main

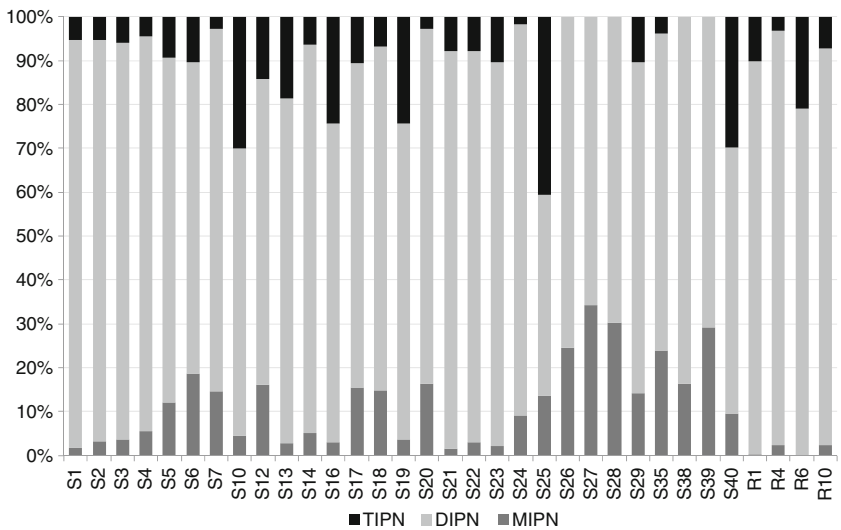


Fig. 9 The concentration ratio of MIPN, DIPN, and TIPN at each sampling point where DIPN was detected over MDL

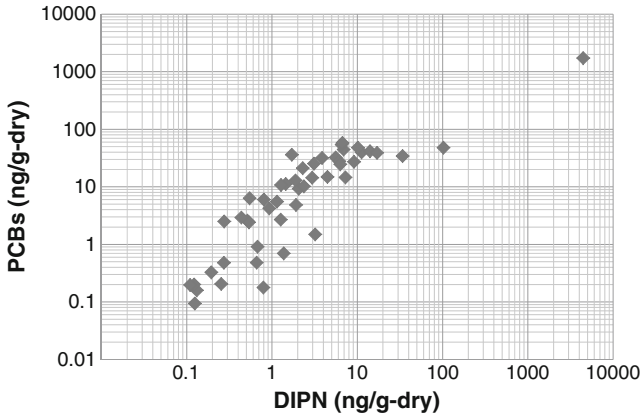


Fig. 10 The correlation between DIPN and PCBs concentration in sediments

contaminant was DIPN in the sediments, and the contamination by MIPN and TIPN was speculated to be related to their presence as minor components within DIPN technical mixtures used in industry. The contamination source of the second-highest polluted point of DIPN might be the wastewater from the paper recycling plant. However, the sampling point with the highest contamination did not have a paper recycling plant. Other potential sources of contamination should be investigated. As expected for hydrophobic chemicals, DIPN was mainly associated with the sediment and fish tissues. The concentrations of DIPN in the sediments tended to increase as those of PCBs increased. DIPN is still being used in Japan at the annual rate of 600 tons. If DIPN continues to be used as a common industrial chemical, for example as a replacement for PCBs, it is likely that it will further accumulate in the environment. It is therefore important to monitor DIPN concentrations, especially in sediment and tissues, to ensure that they do not reach levels that may cause adverse effects in humans or biota. However, due to the paucity of toxicity studies of these chemicals, defining what concentrations are safe is currently not possible. Simultaneous analyses of DIPN, MIPN, and TIPN by the methods reported herein should be carried out in other regions to ascertain how widespread this contamination has become. Appropriate toxicity tests should be carried out so that the risks posed by these chemicals can be quantified.

Acknowledgments The development of the analytical methods in this study was supported by the Ministry of Environment.

References

Boccacci Mariani M, Chiacchierini E, Gesumundo C (1999) Potential migration of diisopropyl naphthalenes from recycled paperboard packaging into dry foods. Food Addit Contam 16(5):207–213

Brzozowski R (2004) Shape-selectivity in diisopropylnaphthalene synthesis or analytical error? Appl Catal A Gen (272) 1/2, 215-218

Brzozowski R, Dobrowolski J, Jamroz MH, Skupinski W (2001) Studies on diisopropylnaphthalene substitutional isomerism. J Mol Catal Chemical 170:95–99

Brzozowski R, Skupinski W, Jamroz HM, Skarzynski M, Otwinowska H (2002) Isolation and identification of diisopropylnaphthalene isomers in the alkylation products of naphthalene. J Chromatogr A 946:221–227

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (1980) Report on environmental survey and wildlife monitoring of chemicals. Ministry of the Environment, Tokyo

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (1983) Report on environmental survey and wildlife monitoring of chemicals. Ministry of the Environment, Tokyo

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (1985) Report on environmental survey and wildlife monitoring of chemicals. Ministry of the Environment, Tokyo

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (1997) Report on environmental survey and wildlife monitoring of chemicals. Ministry of the Environment, Tokyo

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (1997) Chemicals in the environment. Report on Analytical Methods of Chemicals, 176-227

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (2008) Chemicals in the environment. Report on Analytical Methods of Chemicals, 389-410

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (2008) Chemicals in the environment. Report on Analytical Methods of Chemicals, 762-778

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (2008) Chemicals in the environment.

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (2009) Chemicals in the environment.

Environmental Health and Safety Division, Environmental Health Department, Ministry of the Environment Government of Japan (2011) Chemicals in the environment.

Franke S, Grunenberg J, Schwarzbauer J (2007) The isomer-specific analysis of di-iso-propylnaphthalenes. Int J Environ Anal Chem 87(6):437–448

Hasegawa H, Sato M, Tsuruta H (1982) A toxicological study of diisopropylnaphthalene (KMC) and 1-phenyl-1-xylyl-ethanes (SAS). Ind Heal 20:237–246

Hoke H, Zellerhoff R (1998) Metabolism and toxicity of diisopropylnaphthalene as compared to naphthalene and monoalkyl naphthalenes: a minireview. Toxicology 126:1–7

King TL, Yeats P, Hellou J, Niven S (2002) Tracing the source of 3,3′-dichlorobiphenyl found in samples collected in and around Halifax Harbour. Mar Pollut Bull 44(7):590–596

Kodama T (1981) Determination of diisopropylnaphthalene in aqueous solution. Bull Aichi Environ Res Cent 9:34–39

Kojima H, Saito H, Yoshida T (1982) Identification of 2,6-diisopropylnaphthalene metabolites in carp Part 1. In vivo experiment. Chemosphere 11(10):1003–1010

Nakano T, Fukushima M, Shibata Y, Suzuki N, Takazawa Y, Yoshida Y, Nakajima N, Enomoto Y, Tanabe S, Morita M (2004) POPs monitoring in Japan—fate and behavior of POPs. Organohalogen Compd 66:1464–1469

Peterman HP, Delfimo JJ (1990) Identification of isopropylbiphenyl, alkyl diphenylmethanes, diisopropylnaphthalene, linear alkyl benzenes and other polychlorinated biphenyl replacement compounds in effluents, sediments and fish in the Fox River system, Wisconsin. Biol Mass Spectrom 19:755–770

Scarlett AG, Clough R, West C, Lewis CA, Booth AM, Rowland SJ (2011) Alkylnaphthalenes: priority pollutants or minor contributors to the poor health of marine mussels? Environ Sci Technol 45:6160–6166

Sturaro A, Parvoli G, Rella R, Bardati S, Doretti L (1994) Gas chromatographic–mass spectrometric, high-performance liquid chromatographic–UV and gas chromatographic–Fourier transform IR responses to an industrial mixture of diisoporpylnaphthalenes. J Chromatogr A 688:211–219

Suzuki M, Matsumura C, Moriguchi Y, Nakano T (2007) Investigation of mono-isopropylnaphthalene, di-isopropylnaphthalene and tri-isopropylnaphthalene in the environment around the paper recycling plant. Organohalogen Compd 69:2910–2913

Yoshida T, Kojima H (1978a) Studies on environmental safety of di-isopropylnaphthalene (DIPN) part-I bioconcentration of 14C-DIPN in carp. Chemosphere 7(6):491–496

Yoshida T, Kojima H (1978b) Studies on environmental safety of di-isopropylnaphthalene (DIPN) part-II biodegradation of 14C-DIPN with activated sludge. Chemosphere 7(6):497–501