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## Spatial and Monthly Behaviour of Selective Organochlorine Pesticides in Subtropical Estuarine Ecosystems

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Additional information is available at the end of the chapter

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#### 1. Introduction

Organochlorine pesticides (OCPs) are one of the most important persistent organic pollutants (POPs) which pose threats to ecosystems and human health. The twelve so-called POPs; nine of which are organochlorine pesticides. The two organochlorine pesticides (HCH and dieldrin) were used in or arise from industry mainly for agriculture purposes. Sediments serve as both a source and a removal mechanisms for contaminants to and from rivers and streams and as a means of contaminant transport downstream. Sediment also provides habitat for benthic biota and can be in the food web around rivers and stream, and some organisms such as fish are consumed by people and birds (Brasher & Anthony, 1998; Laabs et al., 2002). Although the residue levels of the chlorinated compounds in the environments have considerable declined in the past 20 years, recent work has depicted that chlorinated pesticides could be detected in the range of 0.03-25.17 ngg<sup>-1</sup> (dry weight) (Chang & Doong, 2006; Zhou et al., 1994).

Some OCPs such as DDT and endosulfan are still used in some countries around the tropical and subtropical regions for agricultural and medicinal purposes. These compounds can be deposited into the sediments through long-range atmospheric transport, resulting in a high exposure to OCPs in the area near the pollution source (Tanabe et al., 1994; Doong et al., 2002; Fabricius, 2005). River bed sediments and fish tissues contain higher concentrations of organochlorine compounds than the surrounding water, so analysis of sediment increases the likelihood of detecting compounds that are present in the river.



#### 1.1. A brief history of organochlorine pesticides (OCPs)

A beginning of the twentieth century, early research and studies with chemical pesticides led to the widespread use of inorganic compounds within agriculture containing elements such as sulphur, arsenic, mercury, lead and other metals (Turnbull, 1998). For some natural products such as pyrethrum were also known to be effective pesticides at the time, but were considered to expensive for widespread use (Awofolu and Fatoki, 2003). Between the world wars, the development of the chlor-alkali industry provided the raw material for the mass production of synthetic chlorinated organic molecules. The first and early chlorinated phenoxy acid herbicide (2,4-D) was first discovered in 1932 (Burton and Bennett, 1987). Although this chemical rapidly breaks down in the environment, the seed fungicide hexachlorobenzene (HCB) were introduced in 1933 was found to be far more persistent (Carlsen et al., 1995). The structurally similar insecticides hexachlorocyclohexane or HCH or BHC (also known as benzene hexachloride-BHC) also emerged at this time. The outbreak of war in 1939 and the need to administer malaria and typhus amongst soldiers and civilians has led to the uncovering, unravelling and application of DDT across the world within four and half years from 19430. Related research about the nerve gas agents in Germany led to the discovery of the associated organophosphorus pesticides (Carlsen et al., 1995). Towards the end of the world war, a clear new future for the agrochemical control using these organochlorine chemicals was contemplated. After the world was, the British government considered a practical need to improve agricultural activity and increase food production by the admittance of more complex machinery creation of larger fields, use of chemical fertilizers and the new synthetic pesticides. By 1953, two insecticidal seed dressings, dieldrin and aldrin were being introduced into the UK (Burton and Bennett, 1987). In America, toxaphene was first produced in 1945 as an effective insecticide for cotton plants. This mixture of over 170 chlorinated derivatives known as camphachlor in Europe was recommended as an alternative to DDT before it was banned in the 1980s due to its environmental toxicity (Carlsen et al., 1995). Coupled with other persistent organochlorines such as chlorofluorocarbons, chlorinated biphenyls, dibenzodioxins and dibenzofurans (Doong et al., 2002) the chlorinated pesticides have the potential to cause significant damage to the natural ecosystem by interfering with reproductive processes, this influencing the biodiversity of non-target organisms (Forget et al., 1995). Some aspects of this impairment are now well researched and documented. Whether from past application in developing countries or from continuing current use, these compounds can now be detected in the most remote regions of the planet.

#### 1.2. Definition and importance of organochlorine pesticides (OCPs)

Organochlorines are carbon-based chemicals that contain bound chlorine. These compounds are hydrophobic and lipophilic to varying degrees, meaning their solubility in water is very low, whereas their solubility in fats and oils is relatively high (Cheevaporn et al., 2005). They are noted for their persistence and bioaccumulation characteristics. Some substances may be very persistent in the environment (i.e. with half-lives ( $t_{1/2}$ ) greater than 6 months). The nature of this persistence needs to be clarified- it is the length of time the compound will remain in the environment before being broken down or degraded into other and less hazardous substances (For-

get et al., 1995). The widespread use of these compounds over the past half century has led to their detection in many hydrologic systems world-wide from agricultural and non-agricultural purposes (Monirith et al., 2003). Organochlorine pesticides (OCPs) are considered to be dangerous not only for the environment but for animals and human beings as well. They are very stable substances and it has been cited that the degradation of DDT in soil in 75-100% in 4-30 years (Doong et al., 2002). Other chlorinated pesticides such as Aldrin, Dieldrin, Endrin and Isodrin remain stable in water for many years after their use (Cheevaporn et al., 2005).

#### 1.3. Chemistry

Organochlorine pesticides (OCPs) are organic compounds that highly resistant to degradation by biological, photolytic or chemical means. OCPs are mostly chlorinated. The carbon-chlorine bond is very stable towards hydrolysis and the greater number of chlorine substituted and functional groups, the greater the resistance to biological and photolytic degradation (Doong et al., 2002). Chlorine attached to an aromatic (benzene) ring is more stable to hydrolysis than chlorine in aliphatic structures (Forget et al., 1995). As a result, OCPs are typically ring structures with a chain or branched chain framework. By virtue of their solubility leading their propensity to pass readily through the phospholipids structure of biological membranes and accumulate in fat deposits.

#### 1.4. Human health

As noted for environmental effects, it is also most difficult to establish cause and effects relationships for human exposure of OCPs and incident diseases. As with wildlife species, human encounter a broad range of environmental exposures and frequently to a mixture of chemicals at any time. Much work remains to be done on the study of the human health impact of exposure to OCPs, particularly in view of the broad range of concomitant exposing experienced by humans (Vagi et al., 2005). Previous and present scientific evidences suggest that some OCPs have the potential to cause significant adverse effects to human health at the local level and at the regional and global levels through long-range transport (Doong et al., 2002). For some OCPs, occupational and accidental high-level exposure is of concern for both acute and chronic worker exposure. The risk is greatest in developing countries where the OCPs in tropical agriculture have resulted in a large number of deaths and injuries (Fu et al., 2003). In addition, to other exposure courses, workers exposure to OCPs during waste management is a significant source of high concentration of certain OCP which resulted in illness and death (Doong et al., 2002). For example, a study in the Philippines showed that in 1990, endosulfan became the number one cause of pesticide-related acute poisoning among subsistence rive-farmers and mango sprayers (Forget et al., 1995). Earliest reports of exposures to OCPs related to human health impact include an episode of HCB poisoning of food in south-east Turkey, resulting in the death of 90% of those affected and in other exposure related incidences of hepatic cirrhois, porphyria and urinary arthritic and neurological disorders (Barakat, 2004). Occupational, bystanders and near field exposure to toxic chemicals is often difficult to minimize in developing countries (WHO, 2004). Laboratory and field observations on animals as well as clinical demonstrate that over exposure to certain OCPs may be associated with a wide range of biological effects. These adverse effects may include immune dysfunction, neurological deficits, reproductive anomalies, behavioural adnormalities and carcinogenesis (Forget et al., 1995). The scientific evidence demonstrating a link between chronic exposure to sub lethal concentrations of OCPs (such as that which could occur as a result of long range-transport) and human health impacts is more difficult to establish but gives cause for serious concern (Doong et al., 2002).

#### 1.5. Organochlorine pesticides production and use

The nine OCPs out of twelve POPs compounds were used in or arise from industry, agriculture crops and disease vector control of public health (Chang and Doong, 2006). By the late 1970 all eight OCPs has been either banned or subjected to severe use restriction in the developed world but the major release of these compounds were mostly used by developing countries especially Asia (Hung and Thiemann, 2003), South/Central America (Falco et al., 2003) and Africa (Mwevura et al., 2002). Although the statistics on the use in many areas remained unclear (FAO, 1989). Previous studies revealed that some HCH remains a common compound used in large quantities in India, China, Africa and South America (Turnbull, 1995). It was also recorded that India consumed 25,000 tons of HCH annually over recent years (Davis et al., 1992) and one factory in China was thought to have an annual product of 20,000 tons (Zhang et al., 2002). In Japan, the using of these OCPs has been prohibited in the field in the 1970-1980 (Nakai et al., 2004). It was estimated that the pesticide used in the United States was 550,000 tons during 1995 (Golfinopoulos et al., 2003). In addition, Greece consumed approximately 3500 tons per year of OCPs in the form of insecticides and pesticide (Miliadis, 1993). In Vietnam, approximately 15,000 tons was used from 1957-1972 (Quyen et al., 1995) and 50 tons from the year 1999 (Hung and Thiemann, 2003). In Germany, 36,000 tons was consumed in the year 1991 (Statistisches Bundesamt, 1993, Hung and Thiemann, 2003).

#### 1.6. Characteristics of hexachlorocyclohexane (HCH)

Hexachlorocyclohexane (HCH) is an insecticide that exists in eight different forms. One of its form is known as gamma-HCH ( $\gamma$ -HCH) or commonly called Lindane is produced and used as an insecticide on fruit, vegetables and forest crops. It is a white solid that turns into a vapour when released into the air with a melting point varied with isomeric composition. Its vapour pressure at 4.2 mm Hg at 20°C. (US EPA, 2012).

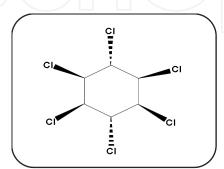


Figure 1. Chemical structure of hexachlorocyclohexane

#### 1.7. Characteristics of dieldrin

Dieldrin is an insecticide which is closely related to aldrin, which reacts further to form dieldrin. It used principally to control textile pests and insects living in agricultural soils. It is a white crystals with a melting point of 175-176°C. Its solubility in water is 140  $\mu$ gL<sup>-1</sup> at 25°C with a vapour pressure of 1.78 x 10<sup>-7</sup> mm Hg at 20°C (US EPA, 2012).

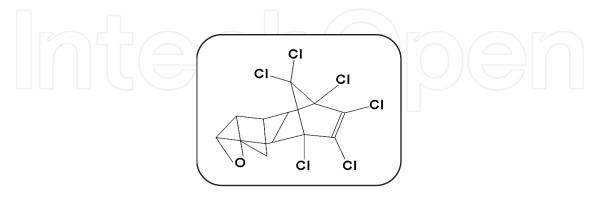


Figure 2. Chemical structure of dieldrin

#### 2. Environmental impact to estuaries

Organochlorine pesticides are carbon-based chemicals that contain bound chlorine. These compounds are hydrophobic and lipophilic to varying degrees, meaning their solubility in water is very low, whereas their solubility in fats and oils is relatively high (Cheevaporn et al., 2005). They are noted for their persistence and bioaccumulation characteristics. The widespread use of these compounds over the past half century has led to their detection in many hydrologic systems world-wide from agricultural and non-agricultural purposes (Monirith et al., 2003). The presence of HCH and dieldrin pesticides in the environment may be related to both past and present land use in a watershed. It enters the aquatic environment from a variety of sources, including the atmosphere, industrial and municipal effluents and agricultural and urban non-point source run-off. HCH and dieldrin are mostly associated with bottom sediments, which can be ingested by benthic organisms. These organisms are then eaten by fish and birds, which can result in higher concentrations through aquatic and terrestrial food chains. Due to the long residence time of these substances in the environment, it is important to examine the pollution they cause not only the environment but also for the lower invertebrates such as corals. Since the ocean is the receiving basin for terrigenous freshwater run-off and its entrained materials, some fractions of these compounds that are used in upland eventually reach the marine ecosystems.

The Manko and Okukubi estuaries are protected wetlands located in a subtropical climate on Okinawa Island. These estuaries are very famous host for migrating birds from South East Asia and mainland Japan. It also plays a great role of species conservation and it was added to the RAMSAR Convention register of wetlands. However, estrogenic activities were detected in sediment samples from these estuaries (Tashiro et al., 2007). Previous studies showed that

the coral reef ecosystems and their adjacent environments in and around the Okinawa Island are contaminated with OCPs, OTCs and PCBs (Tashiro et al., 2003; Imo et al., 2007; Sheikh et al., 2002). However, very little is known about the behaviour of HCH and dieldrin in estuarine sediments of subtropical areas. The main objective of this chapter is provide crucial information on the distribution and behaviour of HCH and dieldrin compounds in protected subtropical estuaries in the Okinawa Island.

#### 3. Experimental

#### 3.1. Sample processing

Surface sediment samples were collected with a stainless steel grab. The upper 1-3 cm of the sample were carefully removed and stored in acid rinsed polyethylene 250 mL glass bottles. Samples were transferred to the laboratory and were stored at -20°C until sample extractions. Details of sampling areas is shown in Table 1 and description of samples as shown in Table 2. Sampling location is shown in Figure 3.

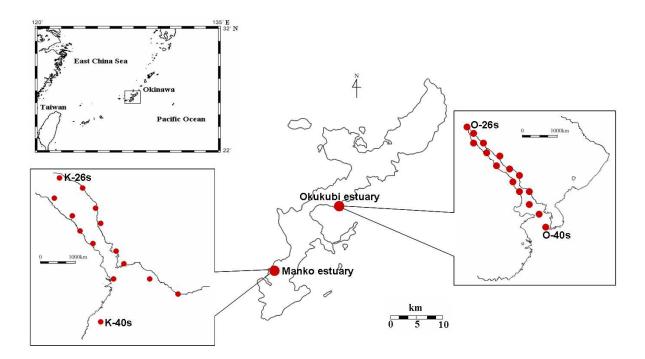


Figure 3. Sampling locations

Estuary	Transect	Sample	Location	Activities
Manko	TM1	K-26s-05		
		K-27s-05	Upstream	Residential area
		K-28s-05		
	TM2	K-29s-05		
		K-30s-05	Mid –stream	Residential area
		K-31s-05		
	TM3	K-32s-05		
		K-33s-05	Mid-stream	Residential area
		K-34s-05		
	TM4	K-35s-05		
		K-36s-05	River mouth	Residential and fishing po
		K-37s-05		
	TM5	K-38s-05		
		K-39s-05	Naha port	Commercial port
		K-40s-05		
Okukubi	TO1	O-26s-05		
		O-27s-05	Upstream	Agriculture
		O-28s-05		
	TO2	O-29s-05		
		O-30s-05	Mid-stream	Agriculture
		O-31s-05		
	TO3	O-32s-05		
	77(0)	O-33s-05	Mid-stream	Agriculture
	GU	O-34s-05		
	TO4	O-35s-05		
		O-36s-05	Mid-stream	Fishing area
		O-37s-05		
	TO5	O-38s-05		
	,	O-39s-05	River mouth	Residential, public area
		O-40s-05		

**Table 1.** Details of sampling areas.

Samuel -	Surface river sediment	
Sample	Sample sketch	
K-26s-05	mud	
K-27s-05	mud	
K-28s-05	mud	
K-29s-05	mud	
K-30s-05	mud	
K-31s-05	mud	
K-32s-05	mud	
K-33s-05	mud	
K-34s-05	mud-sandy	
K-35s-05	sandy	
K-36s-05	sandy	
K-37s-05	sandy	
K-38s-05	sandy	
K-39s-05	sandy	
K-40s-05	sandy	
O-26s-05	sandy	
O-27s-05	sandy	
O-28s-05	sandy	
O-29s-05	sandy	
O-30s-05	sandy	
O-31s-05	sandy	
O-32s-05	sandy	
O-33s-05	sandy	
O-34s-05	sandy	
O-35s-05	sandy	
O-36s-05	sandy	
O-37s-05	sandy	
O-38s-05	sandy	
O-39s-05	sandy	
O-40s-05	sandy	

Table 2. Description of surface sediment samples

#### 3.2. Sample extraction

Prior to extraction, surface sediments were freeze-dried, homogenized with a stainless spatula and passed through a 63  $\mu$ m sieve followed by mixing with anhydrous Na<sub>2</sub>SO<sub>4</sub>. The sediment samples were extracted by ultrasonication technique as described by (Vagi et al., 2005). The surrogate standard of 5  $\mu$ gL<sup>-1</sup> was added to 40 g of sediments. Portion of this amount was used for QC analysis (i.e. each batch contained 1 sample, 1 blank, 4 spiked). For the spiked samples, various concentration of Chlorinated Mix (5, 10, 50  $\mu$ gL<sup>-1</sup>) were added to each spiked sample. The samples were extracted by ultra wave sonication for 15 minutes with 10 mL of dichloromethane. The extracts were then filtered using WHATMAN filters (0.45  $\mu$ m) followed by centrifugation at 3000 rpm for 15 minutes. The clear organic supernants were removed then the combine extracts were evaporated on a rotary evaporator at 30-35°C near to dryness. A 1 mL of hexane was added to the dried residues. For further cleanup, the samples were then added to the florisil ENVI Carb and the samples were eluted with hexane. The residues were dissolved in 1 mL hexane. A 1mL of Internal Standard (Pentachloronitrobenzene, 50  $\mu$ gL<sup>-1</sup>) in the amount extracted before GC-MS analysis.

#### 3.3. Environmental parameters

#### 3.3.1. Total Organic Carbon (TOC) in sediments

Approximately 3g sediment sample was weighed ( $\pm$  0.002 g) and HCl (2M) was added to the sample and left over night to remove all carbonates. Milli Q water was added to rinse the acid from the sediments. To ensure that all the acid was removed from the samples, a 6M of HCl was added. The acid from the sample was removed by adding 2 mL of distilled water followed by centrifugation. The sediments were then dried at 60°C over night and ready for analysis (US EPA, 2012). The Total Organic Carbon (TOC) was determined using the CHNS analyser (JM 10 Model from J-Science Lab, Co. Ltd, Japan0. Calibration was performed using Antipyrine as Standard with the following compositions:  $C_8H_9NO = 135.17$  (C = 70.19%, H = 6.43%, O = 8.50% and N = 14.88%).

3.3.2. pH

The pH was measured using a portable pH meter at room temperature in the laboratory using PHM 95/ion meter, Radiometer model (± 0.001 pH).

#### 4. Results and discussion

The highest concentration of HCH was found in the sampling month of October (213 ng/g) (Manko estuary) and Dieldrin (98 ng/g) (Okukubi estuary) followed by the month of November (HCH-199 ng/g) (Manko estuary) and (Dieldrin-90 ng/g) (Okukubi estuary). The status of HCH and dieldrin in sediments in this study was compared with those in other rivers. The levels of OCPs in this study are lower than that of Er-jen river, Taiwan (80-8200 ng/g (dw))

(Zhang et al., 2002). River Mataniko, Solomon Island (140 ng/g (dw)) (Iwata et al., 1995) but higher in some rivers in Japan (2.5-12 ng/g (dw)) (Sakar et al., 1997). The basic physico-chemical parameters of sediments such as TOC were also measured. The TOC contents ranged from nd-3.96% (Table 3 and Table 4). Figure 4-Figure 5 shows a positive correlation with the concentration of HCH especially in the Manko estuary. No correlation was shown between HCH and TOC from the Okukubi estuary. It is clear that sediments from the Okukubi estuary were composed of fine particles. This observation is consistent with other studies which demonstrated that fine particles can retain large amounts of organic compound and pose a high pollution potency (Hong et al., 1995). Since HCH and dieldrin exhibit carcinogenic activities, the contamination levels detected may pose a high ecotoxicity for aquatic and marine organisms.

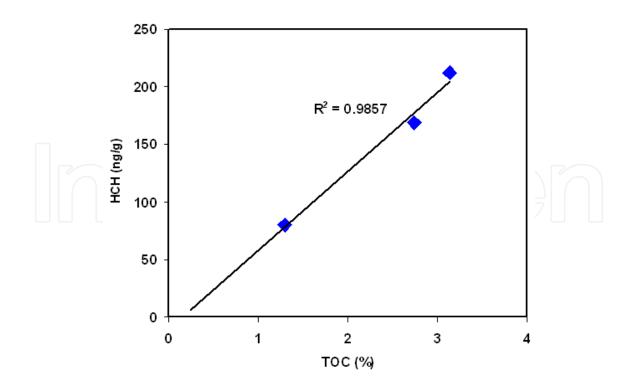
Sample	Temperature (°C)	рН	Total Organic Carbon (%)
K-26s	26.5	8.01	3.13
K-27s	27.4	7.86	2.74
K-28s	28.4	7.81	1.29
K-29s	27.9	7.43	0.24
K-30s	27.6	7.56	nd
K-31s	27.6	7.66	nd
K-32s	28.9	7.74	nd
K-33s	28.9	7.65	nd
K-34s	28.2	7.55	nd
K-35s	28.2	7.64	nd
K-36s	28.6	7.62	nd
K-37s	28.6	7.58	nd
K-38s	28.9	7.56	nd
K-39s	28.9	7.58	nd
K-40s	28.9	7.56	nd

Table 3. Summary of Environmental Parameters – Manko Estuary

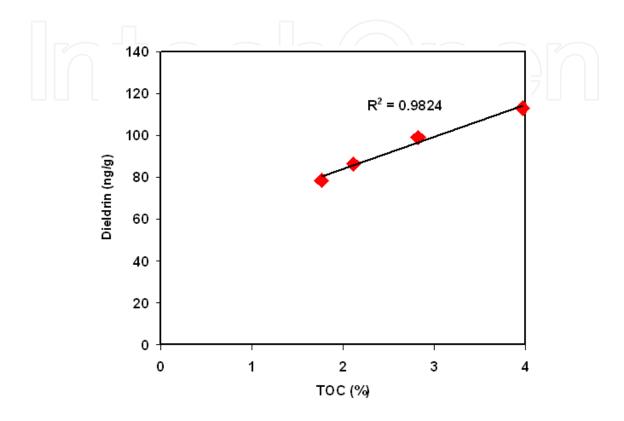
Sample	Temperature (°C)	рН	Total Organic Carbon (%)
O-26s	25.4	8.06	3.96
O-27s	25.9	8.00	2.82
O-28s	26.0	7.99	2.11
O-29s	26.2	7.99	1.76
O-30s	26.5	7.78	0.54
O-31s	26.9	7.56	nd
O-32s	27.0	7.69	nd
O-33s	27.2	7.69	nd
O-34s	27.0	7.72	nd
O-35s	27.1	7.71	nd
O-36s	27.1	7.60	nd
O-37s	26.8	7.64	nd
O-38s	26.9	7.64	nd
O-39s	26.9	7.58	nd
O-40s	26.9	7.54	nd

nd: not detected

 Table 4. Summary of Environmental Parameters – Okukubi Estuary



**Figure 4.** Correlation of HCH with TOC [Manko estuary]



**Figure 5.** Correlation of Dieldrin with TOC [Okukubi estuary]

#### 4.1. Spatial distribution of HCH and dieldrin in sediments

The highest concentrations of HCH was found in sample K-26s – 213 ng/g (dw) (Manko estuary) and the highest dieldrin concentration was found in sample O-26s – 98 ng/g (dw) (Okukubi estuary). Most samples in the Okukubi estuary had relatively low levels of HCH compared to Manko estuary, where the sediments mainly composed of sand. It may be due to the similar historical input and deposit indicating important sources of these organochlorine pesticides in these areas. The second highest concentration of HCH in the Manko estuary, 99 ng/g (dw) followed by 90 ng/g (dw). The second highest concentration of dieldrin in the Okukubi estuary was 199 ng/g (dw) followed by 95.5 ng/g (dw). The levels of HCH and dieldrin in this study are higher in those found in the sediments of the Mingjiang River Estuary, China (2.99–16.21 ng/g, with a mean value of 8.62 ng/g dw (Kennicutt et al., 1994). the Wushi Estuary, Taiwan (0.99–14.5 ng/g, with a mean value of 3.78 ng/g dw (Iwata et al., 1995) Xiamen Harbor,

China (0.14–1.12 ng/g, with a mean value of 0.45 ng/g dw, Hong et al., 1995) and Casco Bay, USA (<0.25– 0.48 ng/g) dw, but lower than the Matanico River and Solomon Islands (140 ng/g) (Walker et al., 1999).

#### 4.2. Monthly variations of HCH and dieldrin in sediments

It clearly revealed that the HCH and dieldrin pesticides residues in October were higher compared to other sampling months. This means that some organochlorine pesticides could be released from the run-off effluents to waters with much rainfall during the rainy season and typhoon season in Okinawa during the summer. In all sampling months, the highest concentration of organochlorine pesticides in the Manko estuary was 213 ng/g (dw) in October and for the Okukubi estuary, the highest concentration of organochlorine pesticides was also detected in the month of October, 213 ng/g (dw). The second highest concentration of HCH was detected in the month of November (199 ng/g (dw)) followed by the month of December (99 ng/g (dw)) for the Manko estuaries. The second highest concentration of dieldrin was detected in the month of October (90 ng/g (dw)), followed by 89.5 ng/g (dw) in the month of November for the Okukubi estuaries.

#### 4.3. Composition analyses in sediments

Composition difference of HCH in the environment could indicate contamination sources (Wu et al., 1999). Technical HCH has been used as broad spectrum pesticides for agricultural purposes, which has been banned in the 1970's in Japan. Technical-grade HCH consists principally of four isomers,  $\alpha$ -HCH,  $\beta$ -HCH,  $\gamma$ -HCH and  $\delta$ -HCH. The physiochemical properties of these HCH isomers are different. The  $\beta$ -HCH has the lowest water solubility and vapour pressure which is the most stable and relatively resistant to microbial degradation (Strandberg et al., 1998). Also it should be noted that  $\alpha$ -HCH can be converted to  $\beta$ -HCH in the environment (Lee et al., 2001). The results showed that a high percentage of HCH isomer was recorded in the sampling months December, January and February. It is possible that HCH may be re-absorbed to surface sediments. There was no strong evidence to prove the recent usage of HCH in Okinawa; however Manko estuary was contaminated with HCH.

#### 5. Conclusion

Generally the distribution of organochlorine pesticides were associated with land use practices including agriculture and urbanization and the sediments from estuary have higher contents of organic matter such as TOC and organochlorine pesticides residues. The concentration and compositions of organochlorine pesticides varied significantly with different sampling sites. The HCH in the surface sediments were well correlated with TOC content. The organochlorine pesticides residues (HCH and dieldrin) were detected due to re-absorption in sediments due to previous deposition. The possible sources of these organochlorine pesticides are still unknown but they may come from residential areas, commercial and naval ports and agriculture activities.

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