

# Assessment and Investigation on the Fate of Organochlorine Pesticides in Water and Sediments of International Amir-kalaye Wetland in North of Iran

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**Abstract** A total of 48 water samples and 24 sediment samples were collected at four sampling stations along the wetland during four seasons from 2009 to 2010 and analyzed by gas chromatograph–electron capture detector (GC–ECD). In water the total concentration of OCPs was 0.33, 0.01, 0.1 and 0.07 mg/L in summer, autumn, winter and spring, respectively. The most frequent OCP compounds detected were endrin and chlordane (0.08 and 0.07 mg/L), heaxachlorobenzene and chlordane (0.06, 0.02 mg/L), and chlordane (0.07 mg/L) in summer, winter and spring, respectively. The maximum concentration of  $\Sigma$ OCPs was found in samples collected from station 1 in summer (0.26 mg/L). In sediments the total concentrations of OCPs were 15.84 and 2.62 mg/g-dry weight (dw) in summer and winter, respectively. Chlordane was the most frequently found OCP compound, followed by lindane, 9.92 and 2.47 mg/g-dry weight (dw), respectively, in summer. While, lindane (2.52 mg/g-dw) and endosulfan I (0.1 mg/g-dw) were the highest OCP compounds detected in winter.

The results obtained in this study show that there still exist a variety of organochlorine pesticide residues in the water and sediments from the Amir-kalaye wetland in Iran.

**Keywords** Organochlorine pesticides · Water · Sediment · Amir-kalaye wetland · North of Iran

During the past two decades, the rapid economic boom and growing agricultural, industrial and municipal development led to substantial accumulation of toxic organic compounds and a significant environmental impact has been imposed on the ambient conditions (Chau and Sin 1992).

Since the 1950s, persistent organic pollutants (POPs) have been produced in large volumes. During production, use and disposal, these POPs have entered the environment. The so called ‘dirty dozen’ are POPs that are toxic, bioaccumulate in fatty tissues of animals and humans and do not easily degrade (Stockholm Convention on Persistent Organic Pollutants 2001). These pollutants are officially registered by the United Nations Environmental Program (UNEP) under the Stockholm Convention on the 23rd of May 2001. In this convention, 90 signatory countries have agreed to reduce and/or eliminate the production, use, and release of the 12 POPs of greatest concern to the global community. They can be sub-divided as (1) eight chlorinated pesticides (dieldrin, endrin, aldrin, chlordane, heptachlor, DDT, mirex and toxaphene), (2) two industrial chemicals (hexachlorobenzene (HCB) and polychlorinated biphenyls (PCBs) and (3) two unintentionally produced compounds (polychlorinated dibenzo-*p*-dioxins (PCDDs), also abbreviated as dioxins, and polychlorinated dibenzofurans (PCDFs), also abbreviated as furans). Although production of most POPs has ceased for over 20 years, we are still facing considerable POP levels in the environment

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(USEPA 2002a, b). The Iranian government signed the Stockholm Convention on Persistent Organic Pollutants (hereinafter referred to as the Stockholm Convention or the Convention) on 23rd May 2001 and it was ratified on 6th February 2006 (NIP 2008).

Several types of wetlands have been described in the north of Iran located in the West part of Southern Caspian Sea. There are two important marshes (Anzali and Amir-Kalaye) in Gilan Province in the north of Iran. Amir-Kalaye is one of the most important wetlands for water supply and irrigation. It has a catchment basin of 1,230 ha. There are some agricultural activities around this wetland. Despite the potential for direct discharge of organic contaminants into the wetland, no measurement has been made for the content and composition of POPs in this wetland. In this study, the available data on chlorinated pesticides of POPs (dieldrin, endrin, aldrin, chlordane, heptachlor, DDT, heaxachlorobenzene) + (lindane and endosulfan) in water and sediments within the wetland are gleaned and compiled. Few concentration ranges for organochlorine pesticides have been reported in marine sediments and water of Iran so far, but several studies have been carried out in China (Zhou et al. 2001; Liu et al. 2003; Zhang et al. 2003; Wang et al. 2008; Chen et al. 2011), Hong Kong (Richardson and Zheng 1999), Japan (Iwata et al. 1994), Vietnam (Nhan et al. 1999), Korea (Hong et al. 2003), Egypt (Said et al. 2008) and Taiwan (Doong et al. 2002) showed extensively contamination with POPs. The primary objectives of most monitoring surveys can be summarized comparisons of spatial and temporal changes, checks on compliance (with reference, for instance, to governmental standards and established guidelines) and assessment of possible adverse effects (e.g. ecological risks).

## Materials and Methods

A total of 72 samples, including 48 water [in August (summer), November (autumn), February (winter) and May (spring)] and 24 sediment (in August, February) samples, were collected from the wetland from 2009 to 2010, which means that each station contained three water and three sediment samples. To avoid site deviations in each seasonal sampling, sampling sites were chosen based on their distinguishable locations and were positioned by a Global Positioning System (GPS) based on the main entrance and area wetland. One liter of wetland water from 20 to 30 cm below the water surface was sampled at 4 locations near the site of sediment sampling. The upper 15 cm of the surface sediments were sampled using a boat with a Birge-Ekman sediment grab sampler. The sampling stations are illustrated in Fig. 1. Water characteristics at the different sampling sites are summarized in Table 1.



**Fig. 1** The study areas and sampling locations in the selected wetland

The water samples were directly collected from the wetland using 1 L pre-cleaned glass jars with polytetrafluoroethylene (PTFE) screw caps. The glass jar was cleaned with double distilled water first, and then rinsed with methanol, acetone and hexane in sequence to remove trace amount of organic contaminants. All the samples were transferred to the laboratory under cold condition (4°C) directly. The water samples were stored at 4°C and analyzed within 2 weeks, while sediment samples were air-dried in a clean laboratory condition and ground into fine powder using a stainless steel mortar and pestle. Water and sediments were brought to Iranian Research Institute of Plant Protection, Tehran, for further chemical extraction and quantification.

For extraction at first, 100  $\mu$ L nitrobenzene was added to sediment for internal standard. About 10 g of sediments were transferred into a soxhlet apparatus and extracted with 100 mL of petroleum benzene in 40°C for 4 h at a rate of 4–6 cycles/h. The sediment extracts were then concentrated to 1 mL in a rotary evaporator with 30–40°C, and then transferred to 10 mL glass tubes with 1 mL of hexane and analyzed with a gas chromatograph–electron capture detector (GC–ECD) system. Briefly, all water samplers were filtered on-line, onboard over a precleaned GF/A filter. 1,000 mL water samples were mixed with 30 g NaCl and 50 mL dichloromethane, and shaken twice, and then was shaken in mechanical shaker for 30 min. The mixture was kept in decanter until two phases were separated. The organic phase

**Table 1** Station description of Amir-kalaye wetland

Parameters	Season	Geographic coordinates of station			
		St <sub>1</sub> (428698, 4131471)	St <sub>2</sub> (428699, 4132346)	St <sub>3</sub> (428545, 4134173)	St <sub>4</sub> (428238, 4135541)
Temperature (°C)air	Summer	25.5	25	24.5	24.5
	Autumn	11	10	11	11
	Winter	13.5	12	12.5	12.5
	Spring	29	28.5	28	29
Temperature (°C)water	Summer	24	23	24	24
	Autumn	10	9	10	10
	Winter	12	11	11	11
	Spring	28	27	27.5	28
PH	Summer	7.9	7.45	7.3	7.7
	Autumn	8.06	8.43	8.48	8.41
	Winter	7.36	8.27	8.19	8.28
	Spring	9.30	9.65	10.17	10.40
EC(μs/cm)	Summer	1,821	1,905	1,954	1,630
	Autumn	1,812	1,781	1,792	1,922
	Winter	1,115	984	960	964
	Spring	478	496	455	483
Depth(cm)	Summer	150	150	130	140
	Autumn	170	190	180	180
	Winter	200	220	200	210
	Spring	140	185	170	180

(dichloromethane extract) was kept in beaker, and then the supernatant was transferred into glass jar again and mixed with 50 mL dichloromethane for 5 min to separate organic phase. The latter was added to dichloromethane extract. The organic phase was dried over 15 g anhydrous sodium sulfate and passed through separator Buchner funnel by vacuum pump. The residue concentrated in rotary evaporator below 40°C, and then transferred to 10 mL glass tubes with 1 mL of hexane and analyzed with a gas chromatograph–electron capture detector (GC–ECD) system (Hans 1982). The recovery test was in range of 75%–90%.

The concentrations of OCPs in the extracts were monitored with a gas chromatograph (GC) equipped with 63Ni electron capture detector (ECD). The GC system was operated in a split/splitless mode. A fused silica capillary column DB1 (30 m length, 0.25 mm inner diameter, 0.25 μm film thickness) was used for separating OCPs. For OCPs, column oven temperature increased from 150 to 250°C at 5°C/min, held for 3 min, and then programmed 250 to 290°C, held for 10 and 20 min respectively. The temperatures of injector and detector were 250 and 280°C, respectively. Ultra-high purity N<sub>2</sub> was used as carrier gas. One μL of each sample was injected into the GC system for detection of organochlorine pesticides. GC peaks were identified with the accurate assignment of retention times of each standard (1%).

Comparisons between the concentrations of specific pollutants detected in the sediments and their corresponding sediment quality values (i.e. concentrations below which adverse effects in the marine ecosystem were unlikely) were performed in the present investigation. The levels of risks posed by certain chemicals in the sediments were characterized by risk quotients, which were calculated (Eq. (1)).

In most cases, sediment quality values are not single numbers, but are often represented in ranges of values, which have lower and upper limits. These two values could be used to calculate risk quotients under the best-case (RQ<sub>bcs</sub>) and worst-case (RQ<sub>wcs</sub>) scenarios (Eqs. (2), (3)).

$$\text{Risk quotient(RQ)} = \frac{\text{Concentration of chemical X in sediment}}{\text{Sediment quality value}} \quad (1)$$

$$\text{RQ}_{\text{bcs}} = \frac{\text{lowest measured concentration of chemical X in sediment}}{\text{Upper limit of sediment quality value}} \quad (2)$$

$$\text{RQ}_{\text{wcs}} = \frac{\text{Highest measured concentration of chemical X in sediment}}{\text{Lower limit of sediment quality value}} \quad (3)$$

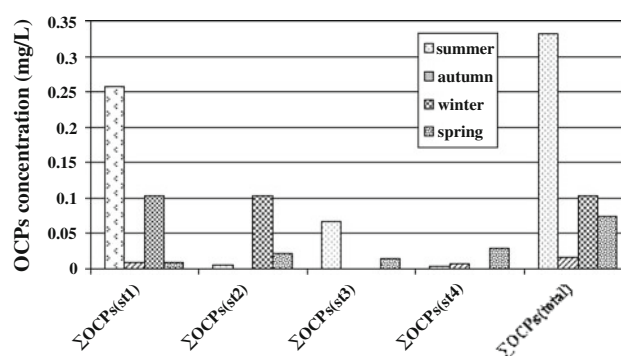
The calculation of  $RQ_{bcs}$  and  $RQ_{wcs}$  provides a simple way to distinguish chemicals, which may or may not require further analysis. In principle,  $RQ_{bcs} > 1$  would indicate that the chemical in question would require attention, and probably some control measure or remedial action is needed. In contrast, if  $RQ_{wcs} < 1$ , the chemical is probably of little concern, and thus should be accorded a lower priority in terms of management actions. In situations where  $RQ_{bcs} < 1$  or  $RQ_{wcs} > 1$ , a more refined risk assessment should be undertaken to ascertain the risks due to the specific chemicals. Here, a precautionary approach may be needed as the sediment quality values used may not be specific or directly applicable to the system under investigation (in this case, the wetland) (Fung et al. 2005).

## Results and Discussion

This study reports on results obtained from a comprehensive survey of Amir-kalaye wetland for the levels of organic contaminants, and represents an attempt to understand the effects of the agricultural pollution and current contamination status in this area. As a result, the following interpretation and discussion will be focused on the nine organochlorine pesticides (lindane, heptachlorepoxyde, chlordane, endosulfan I, endosulfan II, dieldrin, endrin, DDT, heaxachlorobenzene) in water and sediment.

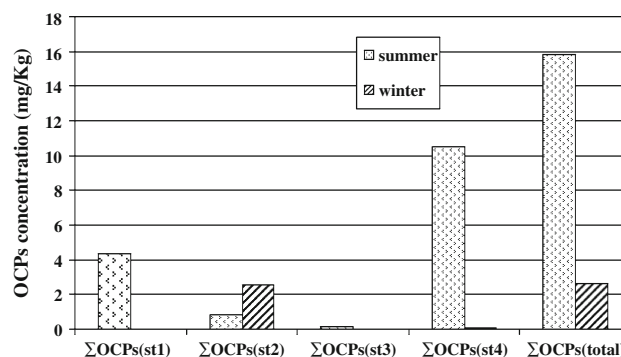
In water  $\Sigma OCPs$  and total concentration of  $\Sigma OCPs$  in the water from four sampling sites in Amir-kalaye wetland among all sampling seasons are shown in Fig. 2. There were no significant differences observed neither in total  $\Sigma OCPs$  concentration among sampling seasons nor in stations when the ANOVA-test was applied. The total concentration of  $\Sigma OCPs$  was 0.33, 0.01, 0.1 and 0.07 mg/L in summer, autumn, winter and spring, respectively. The samples collected in summer have the highest amount of total  $\Sigma OCPs$  concentration, most likely due to the higher level of agricultural activity (growing season of rice) and use of pesticides in this season. On the other hand, the lowest amount of total  $\Sigma OCPs$  concentration was met in autumn. The dilution effect during the flood season is one of the possible reasons for the low concentration of OCPs in autumn. The most frequent OCPs compounds were endrin and chlordane (0.08 and 0.07 mg/L), heaxachlorobenzene and chlordane (0.06, 0.02 mg/L), and chlordane (0.07 mg/L) in summer, winter and spring, respectively, (not shown in the Fig).

The spatial distribution of OCPs was site-specific in water. The maximum concentration of  $\Sigma OCPs$  was found in samples collected from station 1 in summer (0.26 mg/L). It can be explained that, station 1 is the main entrance of wetland. Therefore, the  $\Sigma OCPs$  might be seen in this station more than the other stations.



**Fig. 2** The  $\Sigma OCPs$  and total concentration of  $\Sigma OCPs$  in water collected from four sampling sites Amir-kalaye wetland among all sampling seasons

Figure 3 illustrates the  $\Sigma OCPs$  and total concentration of  $\Sigma OCPs$  in surface sediments from four sampling sites in wetland between two sampling seasons (summer, winter). Any significant difference in total  $\Sigma OCPs$  concentrations either between sampling seasons or in stations was not observed when the t-test and one way ANOVA was applied, respectively. The total concentrations of  $\Sigma OCPs$  were 15.84 and 2.62 mg/g-dry weight (dw) in summer and winter, respectively. Such variations can be explained by using pesticides in late spring and early summer, also the relative seasonal and regional changes in runoff of fresh-water in winter (Simpson et al. 1996; Hong et al. 1999). As a result of temperature changes, water circulation occurs in winter and the OCPs residue in sediment are wash out; it is possible, also, that the OCPs residue migrate to lower levels of sediment. So the surface sediment shows the lowest amount of OCPs in winter. Chlordane was the most frequently found OCP compound, followed by lindane, 9.92 and 2.47 mg/g-dry weight (dw), respectively, in summer. While, lindane (2.52 mg/g-dw) and endosulfanI (0.1 mg/g-dw) were the highest OCP compound in winter (not shown in the Fig).



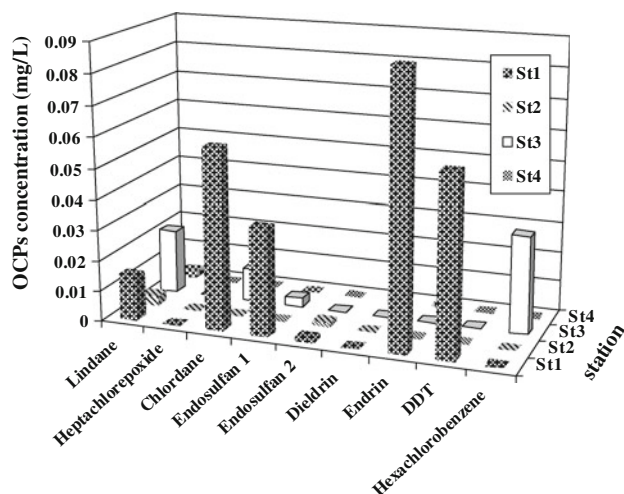
**Fig. 3** The  $\Sigma OCPs$  and total concentration of  $\Sigma OCPs$  in sediments collected from four sampling sites Amir-kalaye wetland between two sampling seasons

In summer, the highest concentration of  $\Sigma$ OCPs was detected in samples collected from station 4, while the lowest concentrations of  $\Sigma$ OCPs was found in sediments collected from station 3. It can be explained that, there is a gentle slope in wetland towards station 4. Hence it can be expected that, sediments tend to accumulate at the exit mouth of the wetland and form a bank which shows the OCPs compound accumulate in large amounts. Using two way-ANOVA test, shows a significant difference between interaction effects of seasons and stations. This difference was limited to seasons. Contamination levels of chlorinated hydrocarbons in the wetland sediments were higher than those in the water. This phenomenon is largely related on the particle size of sediment as is evident from the fact that sediments with large quantity of clay minerals can retain large amounts of pesticide residues than water. Also, Organochlorine pesticides are typically hydrophobic organic compounds and preferentially retain on the organic phase of sediment and organisms (Sarkar et al. 1997).

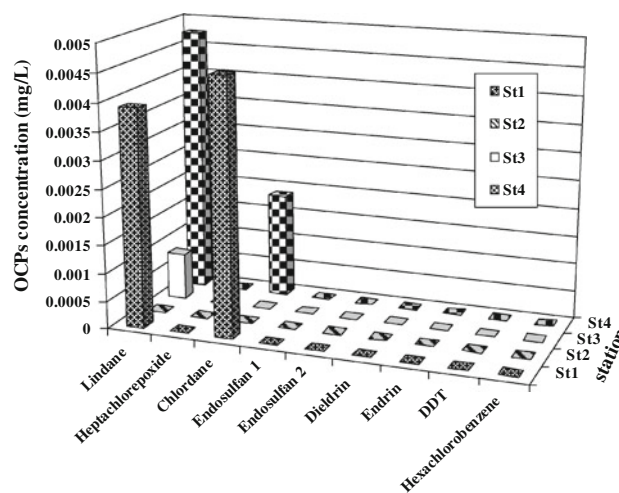
In water Figs. 4, 5, 6, and 7 clarify the profiles of OCPs concentration in water collected from four sampling site in Amir-kalaye wetland in summer, autumn, winter and spring, respectively. At first we considered the comparison of all pesticides in each station among seasons based on one way ANOVA Test, and we found that there were no significant differences in OCP compound concentrations unless for lindane in station 3 ( $p < 0.05$ ), but this difference based on a Post Hoc Test is so negligible that is not detectable between seasons.

Then based on one way ANOVA Test, there was no significant differences in OCP compound concentrations among stations in each season.

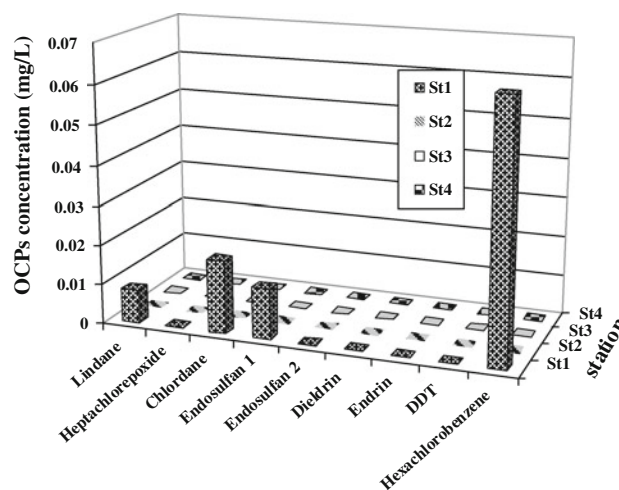
We measured OCP compound concentrations among station without limiting season. The results showed that no



**Fig. 4** The OCPs concentration profiles in water collected from 4 sampling sites Amir-kalaye wetland, Iran in summer



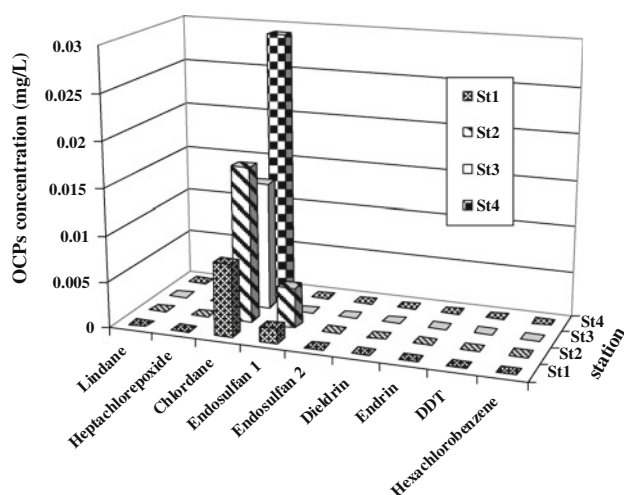
**Fig. 5** The OCPs concentration profiles in water collected from 4 sampling sites Amir-kalaye wetland, Iran in autumn



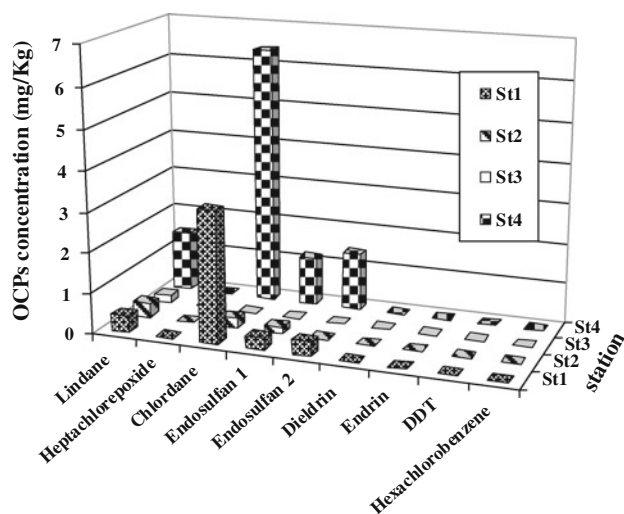
**Fig. 6** The OCP concentration profiles in water collected from 4 sampling sites Amir-kalaye wetland, Iran in winter

significant differences can be seen among stations. We measured OCP compound concentrations among season without limiting station. The results showed that no significant differences can be seen among seasons unless for lindane. Based on Post Hoc test we notice this difference between spring and summer and according to mean value of lindane, it differs in spring with another season which can be related to time of sampling (2009–2010), cultured products and time of spraying pesticides in fields and farms.

In sediment Figs. 8 and 9 show the OCPs concentration profile in sediments collected from four sampling sites in Amir-kalaye wetland in summer and winter, respectively. At first we considered the comparison of all pesticides in each station among seasons based on T-Test. The results show that no significant differences in OCP compound

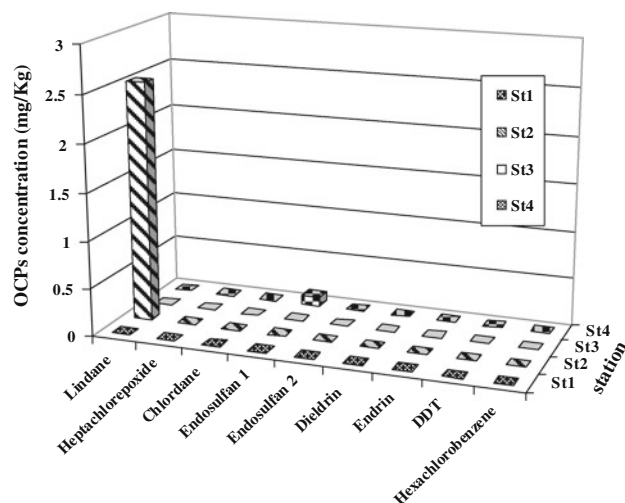


**Fig. 7** The OCPs concentration profiles in water collected from 4 sampling sites Amir-kalaye wetland, Iran in spring



**Fig. 8** The OCP concentration profiles in sediments collected from 4 sampling sites Amir-kalaye wetland, Iran in summer

concentrations in stations 1, 2, 3 were seen among seasons, but Only significant effect of season on OCP compound concentrations was shown for chlordane ( $p < 0.05$ ) between summer and winter. According to mean value of chlordane, it was more in summer than winter. Then based on one way ANOVA Test, there was no significant differences in OCP compound concentrations among stations in each season. We measured OCP compound concentrations among station without limiting season. The results showed that no significant differences can be seen among stations. We measured OCP compound concentrations between season without limiting station The results showed that no significant differences can be seen between seasons unless for chlordane. According to mean value of chlordane, its level was more in summer than winter. This can



**Fig. 9** The OCP concentration profiles in sediments collected from 4 sampling sites Amir-kalaye wetland, Iran in winter

be related to time of sampling (2009–2010), cultured products, kind of pesticide used and time of spraying pesticides in fields and farms.

The freshwater criterion maximum concentration is the highest concentration of a pollutant that freshwater aquatic organisms can be exposed to for a short period (1 h) without deleterious effects. Excursions above the criterion maximum concentration are permitted occasionally (once in 3 years) because it is believed that most aquatic ecosystems can recover from such excursions within 3 years. Freshwater chronic criteria depend on continuous exposure testing over 4 days and lead to a final chronic value, which is an estimate of the concentration of a chemical that is lower than chronic toxicity values for 95% of the genera that have been chronic-toxicity tested. Excursions above the final chronic value are treated in the same way as excursions above the criterion maximum concentration.

According to Table 2, some of the concentrations of selected organochlorine pesticides exceed the acute level of

**Table 2** EPA ambient water-quality criteria for aquatic organisms in Fresh water (Hamilton et al. 2003)

Organochlorine pesticides	Acute ( $\mu\text{g/L}$ )	Chronic ( $\mu\text{g/L}$ )
Lindane	2	0.08
Heptachlorepoide	0.52	0.0038
Chlordane	2.4	0.0043
Endosulfan I	0.22	0.056
Endosulfan II	0.22	0.056
Dieldrin	0.3595	0.0651
Endrin	0.19	0.061
p,p'-DDT	1.1	0.001
Heaxachlorobenzene	6	3.68

**Table 3** Drinking water standards and guidelines (All standard are expressed as mg/L) (EPA 2002, WHO 2003)

Standards	Lindane	Heptachlorepoxide	Chlordane	Endosulfan I	Endosulfan II	Dieldrin	Endrin	DDT	Heaxachlorobenzene
USEPA DWQS	0.0002	0.0002	0.002	NG	NG	NG	0.002	NG	0.001
WHO DWQS	0.002	NG	0.0002	NG	NG	0.00003	NG	0.002	NG

NG no guideline

EPA guideline for water quality criteria for aquatic organisms in both seasons and stations.

In summer, lindane in all stations, chlordane in station 1 and 3, endosulfan I in station 1 and 3, endosulfan II in station 1 and 2, endrin in station 1, DDT in station 1, and heaxachlorobenzene in station 3 were higher than EPA acute level. In autumn, amounts of lindane in station 1 and 4, and chlordane in station 1; and in winter lindane, chlordane, endosulfan and heaxachlorobenzene in station 1; in spring chlordane in all stations and endosulfan I in station 1 and 2 were more than EPA acute levels. Therefore, the water quality in station 1 presents risk for aquatic organisms.

According to Table 3, in summer lindane in all stations, chlordane in station 1 and 3, DDT and endrin in station 1, heaxachlorobenzene in station 3; in autumn lindane in station 1 and 4, chlordane in station 1; in winter lindane, chlordane and heaxachlorobenzene in station 1; and chlordane in station 1, 2, 3 and 4 have exceeded the standard limits of drinking water. It can be inferred from assessing the water quality by USEPA and WHO DWQS that water of wetland is not suitable and it poses a high risk for human to drink.

The sediment quality guidelines (SQG) specified by the USEPA (1997) was used to assess the potential eco-toxicological impacts of organic contaminants measured in the surface sediments. Effects range-low (ER-L) and effects range-median (ER-M) values are used to predict potential impacts of contaminants in sediments, whereas ERL values correspond to the lower 10 percentile and ERM values to median values, when the chemical concentration of a contaminant in sediments are sorted according to the degree of their effects levels. ER-L represents the value at which toxicity may begin to be observed in sensitive aquatic species, whereas ER-M represents the concentration below which adverse effects are expected to occur only rarely. Beside the threshold effect level (TEL), the probable effect level (PEL) is used as the criterion for the prediction of toxicity, and corresponds to a level above which adverse effects are frequently expected (Wurl and Obbard 2005).

Sediment quality criteria for Organochlorine pesticides in freshwater ecosystems are summarized in Table 4. According to the risk assessment, the  $RQ_{WCS}$  for lindane in summer and winter, chlordane in summer were above 10,

**Table 4** Sediment quality guidelines for Organochlorine pesticides in freshwater ecosystems (MacDonald et al. 2000)

Organochlorine pesticides (in $\mu\text{g/kg DW}$ )	TEL	ERL	PEL	ERM
Lindane	0.94	NG	1.38	NG
Heptachlorepoxide	0.6	NG	2.74	NG
Chlordane	4.5	0.5	8.9	6
Endosulfan I	NG	NG	NG	NG
Endosulfan II	NG	NG	NG	NG
Dieldrin	2.85	0.02	6.67	8
Endrin	2.67	0.02	62.4	45
Total DDTs	7	3	4,450	350
Heaxachlorobenzene	NG	NG	NG	NG

TEL threshold effect level; dry weight (Smith et al. 1996), ERL effect range low; dry weight (Long and Morgan 1991), PEL probable effect level; dry weight (Smith et al. 1996), ERM effect range median; dry weight (Long and Morgan 1991), NG no guideline

suggesting that these contaminants may be of concern to the integrity of the Amir-kalaye wetland. Despite the fact that endosulfan I, II were in high level, we could not assess the risk for these contaminants due to the lack of threshold levels for them. From these assessments, it can be stated that the levels of OCP compounds in Amir-kalaye wetland environment are alarming. It may cause toxicological effects in aquatic species inhabiting the wetland.

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