



# Differences in phytoaccumulation of organic pollutants in freshwater submerged and emergent plants

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## ABSTRACT

Plants play an important role as sinks for or indicators of semivolatile organic pollutants, however most studies have focused on terrestrial plants and insufficient information has been obtained on aquatic plants to clarify the accumulation of organic pollutants via air-to-leaf vs. water-to-leaf pathways. The presence of *p*, *p'*-dichlorodiphenyldichloroethylene (*p*, *p'*-DDE), hexachlorobenzene (HCB), 15 polycyclic aromatic hydrocarbons (PAHs), and 9 substituted PAHs (s-PAHs), including oxy-PAHs and sulfur-PAHs, in 10 submerged and emergent plants collected from Lake Dianchi was analyzed in this study. Relatively low concentrations of *p*, *p'*-DDE (ND to 2.22 ng/g wet weight [ww]) and HCB (0.24–0.84 ng/g ww) and high levels of PAHs (46–244 ng/g ww) and s-PAHs (6.0–46.8 ng/g ww) were observed in the aquatic plants. Significantly higher concentrations of most of the compounds were detected in the leaves of the submerged plants than in those of the emergent plants. The percentages of concentration difference relative to the concentrations in the submerged plants were estimated at 55%, 40%, 10%–69% and 0.5%–79% for *p*, *p'*-DDE, HCB, PAHs, and s-PAHs, respectively. The percentages were found to increase significantly with an increase in log Kow, suggesting that the high level of phytoaccumulation of pollutants in aquatic plants is due to hydrophobicity-dependent diffusion via the water-to-leaf pathway and the mesophyll morphology of submerged plants.

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## 1. Introduction

Plants have been shown to be a significant sink for semivolatile organic pollutants due to their surface area and associated lipid content (Paterson et al., 1994; Simonich and Hites, 1994, 1995). The occurrence of pollutants in plant leaves is an important indicator of local contamination (Hermanson and Hites, 1990; Meredith and Hites, 1987; Safe et al., 1992). The phytoaccumulation of organic pollutants in leaves has been widely investigated in previous studies, but attention was paid predominantly to the leaves of terrestrial plants, which accumulate organic pollutants through air-to-leaf pathway (Nakajima et al., 1995; Paterson et al., 1991; Pausch et al., 1995; Simonich and Hites, 1995; Tolls and McLachlan, 1994).

In contrast, aquatic plants were estimated to account for more than 27% of global plants species (Lacoul and Freedman, 2006), and can accumulate organic pollutants through air-to-leaf pathway and water-to-leaf pathways. Exploring the behaviors of pollutants in aquatic plants can help to determine whether aquatic plants are potential biosinks and biofilters of pollutants in aquatic environments, and clarify the accumulation of organic pollutants through air-to-leaf or water-to-leaf pathways in aquatic plant.

Aquatic plants are divided into helophytes, neustophytes and hydatophytes depending on the degree of their immersion in water (Raspopov, 1978; Ronzhina and P'yankov, 2001). Unlike those of terrestrial plants, the leaves of aquatic plants accumulate organic pollutants via both water-to-leaf and air-to-leaf pathways. However, few studies have been conducted on the occurrence of organic pollutants in aquatic plants. These studies mainly investigated emergent plants by assessing the partitioning of pollutants in the roots, stems, and leaves to explore uptake via sediment-to-plant and air-to-plant pathways (Abbassy et al., 2003; Guo et al., 2011;

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Zhou et al., 2008). The importance of air-to-plant pathways could also be identified by comparing the accumulation of organic pollutants in submerged and emergent plants. However, no studies have yet been conducted on the air-to-leaf and water-to-leaf accumulation of organic pollutants in plants, and the differences in the phytoaccumulation of organic pollutants between submerged and emergent plants are still unknown. Therefore, it is vital to analyze pollutant concentrations in the leaves of submerged and emergent plants to compare their phytoaccumulation in these two groups of aquatic plants.

Polycyclic aromatic hydrocarbons (PAHs) have been shown to accumulate in plants (Parrish et al., 2006; Simonich and Hites, 1994). In addition to parent PAHs, substituted PAHs (s-PAHs) such as oxy-PAHs and sulfur-PAHs occur widely in the environment and have various emission sources, such as the reaction of PAHs with atmospheric components and the transformation of PAHs in sediment and soil (Arey et al., 1986; Dimashki et al., 2000; Pitts, 1987). Among the limited studies concerning the environmental occurrences and fates of s-PAHs, no study has been made about their occurrences in plants, although these compounds have been reported to exhibit greater mutagenic and carcinogenic potential than PAHs (Lundstedt et al., 2007).

The present study aimed to assess the bioaccumulation potentials of organic pollutants in aquatic plants, and explore the uptake mechanisms of organic pollutants through air-to-leaf or water-to-leaf pathways. The occurrences of *p*, *p'*-dichlorodiphenyldichloroethylene (*p*, *p'*-DDE), hexachlorobenzene (HCB), PAHs, oxy-PAHs, and sulfur-PAHs in aquatic plants from Lake Dianchi were analyzed. The phytoaccumulation of the targeted pollutants was assessed in both submerged and emergent plants, and the potential mechanisms influencing phytoaccumulation in the two aquatic plant groups were clarified. The findings offer a better understanding of the phytoaccumulation of organic pollutants driven by different ecological processes (air-to-leaf vs. water-to-leaf).

## 2. Materials and methods

### 2.1. Chemicals and reagents

The studied chemicals includes *p*, *p'*-DDE, HCB, 15 PAHs (acenaphthylene (Acy), acenaphthene (Ace), fluorene (FE), phenanthrene (Ph), anthracene (An), fluoranthene (Fl), pyrene (Py), chrysene (Ch), benz[a]anthracene (BaA), benzo[b]fluoranthene (BbF), benzo[k]fluoranthene (BkF), benzo[a]pyrene (BaP), indeno [1,2,3-*cd*]pyrene (IP), benzo[ghi]perylene (BP) and dibenz[a,h]anthracene (DA)), and 9 s-PAHs (dibenzothiophene (DBT), 9-fluorenone (9-Fl), diphenyl disulfide (DD), 2,2'-bithiophene (BT), 1,4-naphthoquinone (NaP), dibenzyl sulfide (DS), benzophenone (BPH), 2-naphthalenethiol (2-NT), thianthrene (TT)). The standards and surrogate standards (PCB 121, acenaphthene-*d*<sub>10</sub>, phenanthrene-*d*<sub>10</sub>, chrysene-*d*<sub>12</sub>, and perylene-*d*<sub>14</sub>) were purchased from AccuStandard (New Haven, CT). HPLC-graded solvents (dichloromethane, acetonitrile, acetone and hexane) were obtained from Fisher Scientific (NJ). Analytical-graded sodium sulfate and aluminum oxide were purchased from CNW Technologies GmbH (Düsseldorf, Germany) and Sinopharm Chemical Reagent Co., Ltd (China), and heated at 400 °C for 4 h before use.

### 2.2. Sample collection

Lake Dianchi, a typical shallow plateau lake, is located in Yunnan Province in the middle of the Yungui Plateau of southwestern China. The lake has an elevation of 1886 m, an area of about 300 km<sup>2</sup> and an average depth of approximately 4.7 m (Hou et al., 2004). The Lake basin is in a northern sub-tropical moist

monsoon climate with average annual precipitation of approximately 1004 mm, average temperature of approximately 14.5 °C, 2470 sunshine hours per year, relative humidity of 74%, and mean wind speed of about 2.5 m/s (Sheng et al., 2012). The lake is heavily contaminated by receiving anthropogenic discharge with total nitrogen and phosphorus of 2.084 and 0.109 mg/L in the water column and 1078 and 327 mg/kg in the sediment. The samples were collected in April 2015 from four littoral zones (24°47'13"N, 102°43'8"E; 24°42'0"N, 102°40'5"E; 25°18'14"N, 102°47'2"E, and 24°51'22"N, 102°46'5"E) in Lake Dianchi. At each sampling location, most the available submerged and emergent plants were collected, and 6–8 individual samples were collected for each species (Table S1). For each species in one location, the plant samples were freeze-dried and homogenized as one sample for chemical analysis. The collected samples included four hydrotophytes (whorled watermilfoil [*Myriophyllum verticillatum* L.], potamogetonaceae [*Potamogeton maackianus* A. Bennett], curled pondweed [*Potamogeton crispus* Linn.], and hornwort [*Ceratophyllum demersum* Linn.]), five helophytes (reed [*Phragmites australis* (Cav.) Trin ex Steud.], bulrush [*Typha orientalis* C.Presl], manchurian wild rice [*Zizania latifolia* (Griseb) Stapf], alligator weed [*Alternanthera philoxeroides* (Mart.) Griseb.], and indian lotus [*Nelumbo nucifera* Gaertn.], and one neustophyte (water hyacinth [*Eichhornia crassipes* (Mart.) Solms]).

The aquatic plants were collected, enclosed in aluminum foil, and delivered in an ice-chilled box to a local laboratory. The leaves were selected, cleaned several times with tap water to remove adhering algae and insect larvae, and rinsed with distilled water. Similar steps have been widely described in previous studies investigating pollutants uptake in the plants (Gao et al., 2000; Kim and Kim, 2000; Özer et al., 2006; Waranusantigul et al., 2003). The leaves were cut into small species, homogenized, freeze-dried, and stored at −20 °C in amber glass vials before analysis.

### 2.3. Sample preparation

The chemical analysis was conducted following the method reported previously by Fan et al. (2017), with some modifications. Briefly, dry samples of approximately 4 g each, spiked with surrogate standards (PCB 121, acenaphthene-*d*<sub>10</sub>, phenanthrene-*d*<sub>10</sub>, chrysene-*d*<sub>12</sub> and perylene-*d*<sub>14</sub>), were Soxhlet extracted with 250 mL of dichloromethane and hexane (1:1) solution for 24 h. The resulting extract was treated by acetonitrile partitioning. The extract was rotary-evaporated to nearly 1 mL and then separated in a funnel containing 50 mL of hexane-saturated acetonitrile and 20 mL of hexane. After shaking for 5 min, the bottom acetonitrile layer was collected. Another 50 mL of hexane-saturated acetonitrile was added to the separator funnel and shaken for 5 min. A 100-mL sample of hexane-saturated acetonitrile containing the extract was collected. The acetonitrile extract was concentrated to about 1 mL and further to near-dryness under a gentle gas stream of nitrogen and then re-dissolved in 1 mL hexane. The extract was then passed through a glass column containing 8 g of 5% H<sub>2</sub>O-deactivated Al<sub>2</sub>O<sub>3</sub>. A solution containing 30 mL of hexane and 30 mL of hexane-dichloromethane (3:1) was used to elute the glass column. The eluent was evaporated to about 1 mL and further to near-dryness under a gentle gas stream of nitrogen and then re-dissolved in 0.1 mL hexane for analysis by gas chromatography coupled with mass spectrometry (GC-MS). The sample preparation processes have been validated by recovery experiments.

### 2.4. GC-MS analysis

The final sample extract was analyzed by GC-MS using a spectrometer (GCMS-QP2010 Plus, Shimadzu) equipped with a fused

silica capillary column (HP-5MS, 30 m × 0.25 mm × 0.1 μm film thickness, J&W Scientific, USA). To analyze *p*, *p'*-DDE, HCB, and PAHs, the injector was set to operate at 250 °C in a splitless mode. The interface temperature and ion source were maintained at 280 °C. The column oven temperature was programmed to increase from 70 °C to 220 °C (3 min) at 20 °C/min and then to 300 °C (5 min) at 20 °C/min. For the *s*-PAH analysis, the injector was set to operate at 280 °C in a splitless mode. The interface temperature and ion source temperature were held at 280 °C and 290 °C, respectively. The column oven temperature was programmed to increase from 60 °C (1 min) to 110 °C at 20 °C/min, and then to 290 °C (15 min) at 6 °C/min. Helium was the carrier gas, administered at a constant flow rate of 2 mL/min. Data acquisition was carried out in the electron impact and selected ion monitoring modes.

## 2.5. Quality assurance and quality control (QA/QC)

All equipment rinses were conducted with acetone and hexane to reduce background contamination. The surrogate standard method was used for quantification. Based on their similar analytical behaviors (e.g., GC retention time), PCB 121 was used as a surrogate for *p*, *p'*-DDE and HCB; acenaphthene-*d*<sub>10</sub> for Acy, Ace, FE, and 2-NT; phenanthrene-*d*<sub>10</sub> for Ph, An, Fl, Py, DBT, DS, BT, NaP, BPH, 9-Fl, DD, and TT; chrysene-*d*<sub>12</sub> for Ch and BaA; and perylene-*d*<sub>14</sub> for BbF, BkF, BaP, IP, BP, and DA. The procedures described above were validated for recovery experiments by analyzing six spiked biota samples. The analytical additions of unlabeled *p*, *p'*-DDE, HCB, PAHs, and *s*-PAHs were at least three times the original concentrations (Table S2). Six replicate spiked samples and one matrix blank sample were analyzed to determine the general recovery rates. The recovery rates for *p*, *p'*-DDE, HCB, PAHs, and *s*-PAHs in the spiked samples were 87% ± 8%, 86% ± 13%, 88% ± 12%–111% ± 8%, and 82% ± 11%–104% ± 10%, respectively. Detailed recovery rates for individual compounds are shown in Table S2. The recovery rates of the surrogates (PCB 121, acenaphthene-*d*<sub>10</sub>, phenanthrene-*d*<sub>10</sub>, chrysene-*d*<sub>12</sub>, and perylene-*d*<sub>14</sub>) were 86% ± 8%, 89% ± 16%, 92% ± 9%, 101% ± 13%, and 107% ± 8%, respectively.

During the sample analysis, procedural blanks, and matrix spiked samples were used for strict quality assurance and control. A procedural blank was analyzed with every set of seven samples, and all results were blank corrected. The matrix spiked samples were analyzed in six alternate batches to confirm the quantification accuracy. The accuracy attained for the target compounds ranged

from 86% to 116% (Table S2). The recovery rates for the surrogates (PCB 121, acenaphthene-*d*<sub>10</sub>, phenanthrene-*d*<sub>10</sub>, chrysene-*d*<sub>12</sub>, and perylene-*d*<sub>14</sub>) in the 38 biota samples were 96% ± 16%, 90% ± 13%, 97% ± 15%, 94% ± 15%, and 94 ± 18%, respectively. The method detection limit (MDL) was set at three times the standard deviation of the procedural blanks for chemicals with detectable blank contamination (PAHs, DBT, 9-Fl, BPH, and DS) and as the instrumental minimum detectable amount for chemicals not detected in the blank samples (*p*, *p'*-DDE, HCB, DD, BT, NaP, 2-NT, and TT). The MDL values for all compounds are shown in Table S2.

## 2.6. Data analysis

Pearson's rank correlation test and an independent-samples *t*-test were conducted, and the normality of data was tested for the pearson's rank correlation test using Shapiro–Wilk's test prior to analysis. The regression was regarded as significant for *p* values below 0.05. Various percentages of the MDL were assigned to the sample containing non-detected (ND) concentrations, and no significantly different results were obtained in the statistical analysis. Compounds with detection frequencies lower than 70% (DS, TT, 2-NT, and BPH) were not included in the statistical analysis. The SPSS 19.0 software package was used for all the tests (SPSS Inc., Chicago, IL).

## 3. Results and discussion

### 3.1. Phytoaccumulation: submerged vs emergent plants

Based on the degree of the immersion in water, the aquatic species was classified into submerged or emergent group. The submerged group consisted of whorled watermilfoil [*Myriophyllum verticillatum* L.], Potamogetonaceae [*Potamogeton maackianus* A. Bennett], curled pondweed [*Potamogeton crispus* Linn.], and hornwort [*Ceratophyllum demersum* Linn.]. The emergent one consisted of reed [*Phragmites australis* (Cav.) Trin ex Steud.], bulrush [*Typha orientalis* C.Presl], Manchurian wild rice [*Zizania latifolia* (Griseb) Stapf], alligator weed [*Alternanthera philoxeroides* (Mart.) Griseb.], Indian lotus [*Nelumbo nucifera* Gaertn.], and water hyacinth [*Eichhornia crassipes* (Mart.) Solms] (Asaeda and Siong, 2008; Center and Van, 1989; Gamage and Asaeda, 2005; Gunasekera et al., 2001; Gupta and Chandra, 1996; Inoue and Tsuchiya, 2006; JK and SQ, 2009; Keskinan et al., 2003; Pflugmacher et al., 2001; Xian et al.,

**Table 1**  
Mean biological parameters and concentrations (ng/g wet weight) of *p*, *p'*-DDE, HCB and *s*-PAHs in the leaves of submerged and emergent plants in Lake Dianchi, China.

Species	Sample numbers (n)	Water Content (%)	<i>p</i> , <i>p</i> '-DDE	HCB	<i>s</i> -PAHs				
					NaP	BT	9-Fl	DBT	DD
Submerged Plants									
Mv	3	0.91 ± 0.02	0.78 ± 0.24	0.38 ± 0.06	0.84 ± 1.14	0.22 ± 0.12	6.39 ± 3.15	12.1 ± 1.89	0.61 ± 0.19
Pm	4	0.88 ± 0.03	1.41 ± 0.62	0.55 ± 0.50	0.57 ± 0.39	0.58 ± 0.50	ND	8.50 ± 3.43	0.43 ± 0.04
Pc	5	0.95 ± 0.02	0.57 ± 0.20	0.25 ± 0.06	0.49 ± 0.37	0.33 ± 0.12	4.55 ± 3.12	5.47 ± 2.54	0.26 ± 0.12
Cd	3	0.89 ± 0.04	2.22 ± 1.51	0.84 ± 0.43	0.19 ± 0.17	0.33 ± 0.07	17.0 ± 13.2	11.4 ± 11.3	0.55 ± 0.75
Emergent Plants									
PaT	8	0.81 ± 0.05	0.86 ± 0.32	0.63 ± 0.23	1.69 ± 1.70	0.29 ± 0.27	12.3 ± 9.21	9.28 ± 7.73	0.34 ± 0.28
ToP	3	0.77 ± 0.02	ND	0.59 ± 0.13	0.51 ± 0.75	ND	32.9 ± 5.33	12.3 ± 12.8	1.12 ± 0.75
Zl	3	0.87 ± 0.05	0.82 ± 0.37	0.46 ± 0.23	0.66 ± 0.54	0.07 ± 0.12	8.62 ± 7.84	9.53 ± 7.44	0.30 ± 0.30
AP	3	0.89 ± 0.01	1.03 ± 0.55	0.35 ± 0.10	0.17 ± 0.21	ND	10.5 ± 8.56	5.28 ± 0.68	0.27 ± 0.29
Nn	3	0.87 ± 0.01	1.21 ± 0.69	0.39 ± 0.10	0.27 ± 0.38	0.07 ± 0.07	15.6 ± 5.50	6.18 ± 3.74	0.26 ± 0.14
Ec	3	0.9 ± 0.04	0.56 ± 0.10	0.24 ± 0.08	0.23 ± 0.34	0.17 ± 0.05	2.56 ± 0.14	2.90 ± 2.52	0.16 ± 0.13

Species: Mv = Whorled watermilfoil (*Myriophyllum verticillatum* L.); Pm = *Potamogeton maackianus* A. Bennett; Pc = Curled pondweed (*Potamogeton crispus* Linn.); Cd = Hornwort (*Ceratophyllum demersum* Linn.); PaT = Reed (*Phragmites australis* (Cav.) Trin ex Steud.); ToP = Bulrush (*Typha orientalis* C.Presl); Zl = Manchurian wild rice (*Zizania latifolia* (Griseb) Stapf); AP = Alligator weed (*Alternanthera philoxeroides* (Mart.) Griseb.); Nn = Indian lotus (*Nelumbo nucifera* Gaertn.); Ec = Water hyacinth (*Eichhornia crassipes* (Mart.) Solms).

Concentrations were expressed as mean ± SD.

**Table 2**  
Concentrations (ng/g wet weight) of PAHs in the leaves of submerged and emergent plants in Lake Dianchi, China.

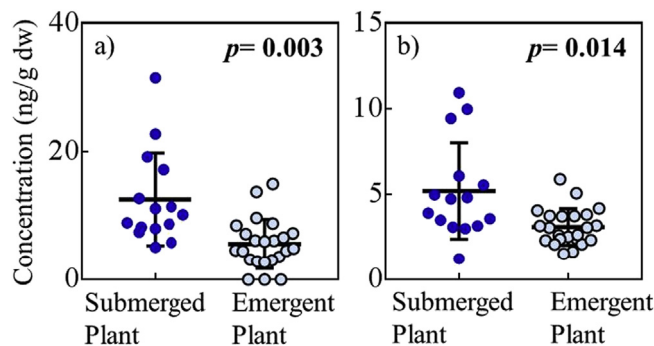
Species	Sample Numbers(n)	PAHs	Acy	Ace	FE	Ph	An	Fl	Py	Ch	BaA	BbF	BkF	BaP	IP	BP	DA
<b>Submerged Group</b>																	
Mv	3		1.26 ± 1.07	4.53 ± 1.11	13.5 ± 9.15	51.1 ± 19.5	2.22 ± 0.33	47.0 ± 39.6	2.75 ± 3.22	0.40 ± 0.26	2.43 ± 1.56	0.58 ± 0.40	0.70 ± 0.45	0.72 ± 0.54	0.54 ± 0.33	0.42 ± 0.24	0.08 ± 0.05
Pm	4		0.90 ± 0.74	5.05 ± 2.96	11.1 ± 5.65	29.8 ± 11.5	1.01 ± 0.07	49.9 ± 24.4	2.90 ± 2.26	0.90 ± 0.69	3.09 ± 2.23	1.40 ± 0.85	1.72 ± 1.07	0.75 ± 0.41	0.16 ± 0.64	0.81 ± 0.42	0.18 ± 0.12
Pc	5		0.25 ± 0.14	1.70 ± 1.34	9.23 ± 4.70	24.2 ± 11.7	0.92 ± 0.57	ND	1.48 ± 1.30	0.18 ± 0.18	1.92 ± 0.78	0.44 ± 0.23	0.36 ± 0.51	0.18 ± 0.29	0.28 ± 0.41	0.26 ± 0.40	0.06 ± 0.10
Cd	3		1.06 ± 0.51	4.69 ± 1.93	15.8 ± 13.5	54.6 ± 60.8	2.77 ± 2.97	99.9 ± 103	2.32 ± 0.45	0.72 ± 0.86	2.76 ± 2.78	0.71 ± 0.64	1.06 ± 1.22	0.38 ± 0.42	0.58 ± 0.55	0.52 ± 0.54	0.13 ± 0.14
<b>Emergent Group</b>																	
PaT	8		2.07 ± 1.46	6.50 ± 5.10	23.2 ± 21.3	40.5 ± 28.5	2.31 ± 1.34	43.2 ± 42.5	3.98 ± 4.09	0.61 ± 0.34	3.18 ± 1.64	0.71 ± 0.49	0.62 ± 0.47	0.46 ± 0.31	0.87 ± 0.82	0.46 ± 0.51	0.08 ± 0.03
ToP	3		3.93 ± 2.61	4.28 ± 3.18	37.1 ± 10.7	112 ± 64.6	7.30 ± 1.39	56.6 ± 16.5	12.3 ± 17.8	1.33 ± 0.49	5.19 ± 1.76	1.03 ± 0.27	0.82 ± 0.25	0.63 ± 0.17	0.56 ± 0.39	0.12 ± 0.1	0.05 ± 0.01
Zl	3		0.43 ± 0.33	5.67 ± 4.10	16.6 ± 9.17	30.1 ± 27.3	1.95 ± 1.28	40.3 ± 32.2	2.54 ± 2.16	0.32 ± 0.16	1.50 ± 0.91	0.31 ± 0.18	0.26 ± 0.09	0.46 ± 0.52	0.25 ± 0.14	0.13 ± 0.14	0.04 ± 0.02
AP	3		0.36 ± 0.26	ND	8.41 ± 5.73	21.9 ± 18.0	0.90 ± 0.12	40.4 ± 32.0	3.04 ± 3.24	0.31 ± 0.04	2.58 ± 1.13	0.50 ± 0.09	0.43 ± 0.05	0.20 ± 0.13	0.29 ± 0.09	0.17 ± 0.07	0.03 ± 0.01
Nh	3		0.37 ± 0.26	ND	9.13 ± 4.46	ND	ND	46.4 ± 37.3	2.06 ± 1.33	0.76 ± 0.07	2.93 ± 0.33	0.58 ± 0.06	1.08 ± 0.11	0.84 ± 0.10	0.41 ± 0.09	0.62 ± 0.35	0.08 ± 0.03
Ec	3		0.16 ± 0.09	1.14 ± 0.06	4.51 ± 0.25	9.01 ± 0.49	0.76 ± 0.04	26.3 ± 21.5	0.92 ± 0.05	0.38 ± 0.30	1.86 ± 1.77	0.38 ± 0.31	0.33 ± 0.24	0.18 ± 0.14	0.29 ± 0.22	0.17 ± 0.12	0.03 ± 0.02

Concentrations were expressed as mean ± SD.

2005; Shan et al., 2014). The detail information of each species was shown in Table 1.

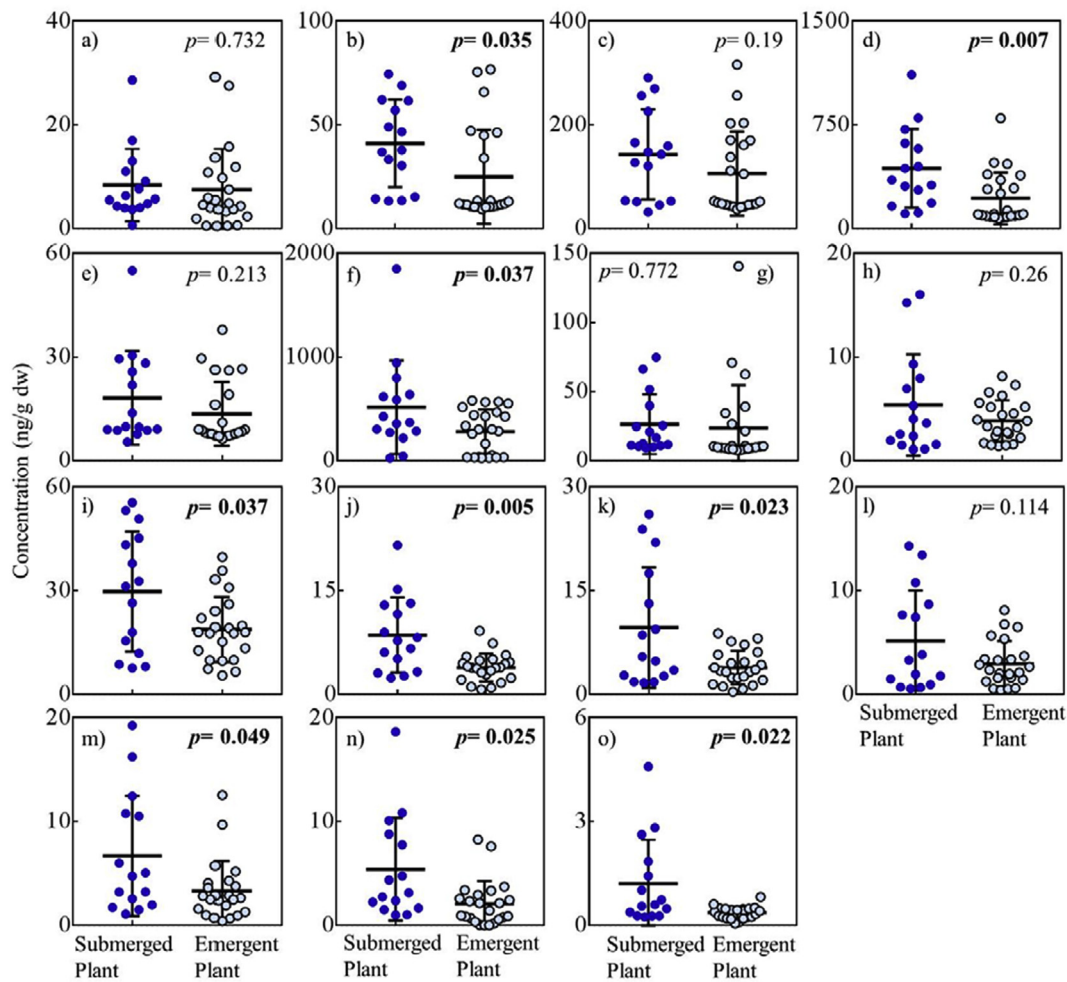
The concentrations of the compounds detected in the aquatic plants from Lake Dianchi (*p*, *p'*-DDE, HCB, 15 PAH congeners, and five s-PAH congeners) are shown in Tables 1 and 2. The concentrations of *p*, *p'*-DDE and HCB ranged from ND to 2.22 ng/g wet weight (ww) and from 0.24 to 0.84 ng/g ww, respectively, lower than those in aquatic plants from Qiantang River (*p*, *p'*-DDE: ND to 5.7 ng/g ww) (Zhou et al., 2008) and Lake Manzala (*p*, *p'*-DDE: 2.96–5.65 ng/g ww; HCB: 1.58–7.08 ng/g ww) (Abbassy et al., 2003). These relatively low concentrations of *p*, *p'*-DDE were consistent with the previous observation that the sedimentary concentrations of *p*, *p'*-DDE in Lake Dianchi (Guo et al., 2013) were lower than those in Lake Manzala (Abbassy et al., 2003). The concentrations of PAHs in the aquatic plants from Lake Dianchi ranged from 474 to 1678 ng/g dw, comparable to those of PAHs in aquatic plants from Lake Baiyangdian (ND to 2866 ng/g dw) (Guo et al., 2011). Of the PAH congeners, Fl and Ph were predominant in all of the samples, contributing 23%–58% and 16%–46% to the total concentrations of PAHs, respectively. Similar profiles dominated by Fl and Ph have also been found in plants from Lake Baiyangdian (Guo et al., 2011). To the best of our knowledge, however, this is the first report on s-PAHs in aquatic plants. NaP, BT, 9-Fl, DBT, and DD exhibited relatively high detection frequencies (>70%) in all of the samples. The total concentrations of the five s-PAHs ranged from 6.0 to 46.8 ng/g ww, lower than those of the PAHs. The ratio of PAH to s-PAH concentration ranged from 12% to 28%. Of the detected s-PAHs, 9-Fl and DBT were predominant in all samples, accounting for 25%–70% and 26%–63% of the total concentrations of s-PAHs, respectively, followed by NaP (1%–7%), DD (1%–3%), and BT (0.01%–4%).

The leaves of the submerged plants accumulated significantly larger amounts of most of the target compounds than those of the emergent plants. Fig. 1 shows that the concentrations of organochlorines in the leaves of the submerged plants (*p*, *p'*-DDE: 9.1–19.8 ng/g dw; HCB: 4.38–7.48 ng/g dw) were significantly higher than those in the emergent plants (*p*, *p'*-DDE: ND to 9.5 ng/g dw; HCB: 2.5–3.4 ng/g dw; *p* = 0.003 to 0.014). Similarly, significantly higher concentrations of aromatic compounds, including PAHs (Ace, Ph, Fl, BaA, BbF, BkF, IP, and BP) and s-PAHs (BT, DBT, and DD), were detected in the leaves of the submerged plants than in those of the emergent plants (*p* = 0.001 to 0.049; Figs. 2 and 3). Previous studies of the phytoaccumulation of organic pollutants in aquatic plants have focused on the partitioning of pollutants between plants and their environmental matrices and generally neglected differences in

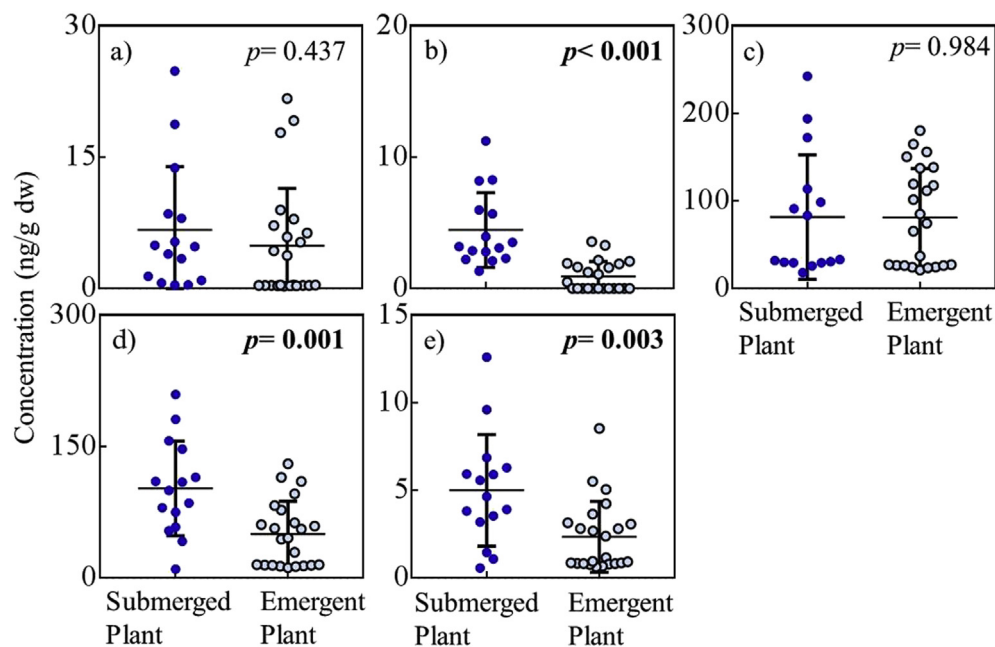


**Fig. 1.** Concentrations of (a) *p*, *p'*-DDE and (b) HCB in the leaves of submerged (*n* = 15) and emergent (*n* = 23) plants in Lake Dianchi, China. *p* values in bold represent statistically significant differences in concentration between the two groups of plants. The horizontal axis of data points shows the number of samples with the similar detected pollutant levels. The bars indicated the mean and standard deviations.





**Fig. 2.** Concentrations of (a) Acy, (b) Ace, (c) FE, (d) Ph, (e) An, (f) Fl, (g) Py, (h) Ch, (i) BaA, (j) BbF, (k) BkF, (l) BaP, (m) IP, (n) BP, and (o) DA (o) in the leaves of submerged ( $n = 15$ ) and emergent ( $n = 23$ ) plants in Lake Dianchi, China. The bars indicated the mean and standard deviations.



**Fig. 3.** Concentrations of NaP (a), BT (b), 9-Fl (c), DBT (d), and DD (e) in the leaves of submerged ( $n = 15$ ) and emergent ( $n = 23$ ) plants in Lake Dianchi, China. The bars indicated the mean and standard deviations.

phytoaccumulation between species (Abbassy et al., 2003; Guo et al., 2011; Guo et al., 2014; Miglioranza et al., 2004; Zhou et al., 2008). The phytoaccumulation of pollutants in the submerged and emergent plants actually reflected partitioning between the air-to-leaf and water-to-leaf pathways because the hydrophobic organic pollutants in the leaves could not have been transported from the root (Paterson et al., 1991; Wild and Jones, 1992). The results of this study indicate that organic pollutants are more easily accumulated by leaves via the water-to-leaf pathway than via the air-to-leaf pathway. The possible reason could be attributed to the differences in mesophyll morphology that the leaf of submerged plant has higher exposure area per unit leaf weight and smaller cuticle barrier than that of emergent plants (Ronzhina et al., 2004; Ronzhina and P'yankov, 2001). Although most previous studies in this area reported on the phytoaccumulation of pollutants in emergent plants, one study investigated the exposure of various plants to  $^{36}\text{Cl}$ -DDT in a freshwater ecosystem (Meeks, 1968). Based on the results of the exposure study, we compared the phytoaccumulation differences between submerged and emergent plants, and found the residues of  $^{36}\text{Cl}$ -DDT in five submerged plants ( $0.3\text{--}1\text{ }\mu\text{g/g}$ ) were higher than those in five emergent plants ( $0.1\text{--}0.4\text{ }\mu\text{g/g}$ ) after 14 month exposure (Meeks, 1968). That finding is consistent with the observations made in this study and was postulated to result from the greater surface area of submerged plants in contact with the water (Meeks, 1968). However, the leaves of the submerged and emergent plants in this study were found to be similar in both size and surface area, suggesting that other factors were responsible for the difference in phytoaccumulation between the submerged and emergent plants.

### 3.2. Factors influencing phytoaccumulation of pollutants in aquatic plants

Phytoaccumulation of organic pollutants in terrestrial plants has been widely studied (Nakajima et al., 1995; Paterson et al., 1991; Pausch et al., 1995; Simonich and Hites, 1995; Tolls and McLachlan, 1994). It has been reported that the lipophilic pollutants are strongly associated with the soil organic fractions, and are susceptible to plant uptake and subsequent transport (Paterson et al., 1991; Paterson et al., 1994). The major accumulation pathway of such pollutants is from the air to the leaf surface (air-to-leaf). The simplified mechanisms of pollutant uptake by plants from the air have been reported (Simonich and Hites, 1995). The gaseous compounds can directly reach the intercellular aerial medium through the open stomata (Galiulin et al., 2002). In contrast, the aquatic plants not only accumulate semivolatile organic pollutants via air-to-leaf pathway, but also through water-to-leaf pathway. The different uptake routes of aquatic plants could be clarified based on the different phytoaccumulation between submerged and emergent plants. To explore the factors that contribute to the differences in the phytoaccumulation, the differences in concentration between the two groups of aquatic plants were quantitatively assessed. As the emergent plants served as the control group for the comparison of phytoaccumulation, the percentages of concentration difference between the two groups of plants relative to the concentration in the submerged plants were estimated with the following equations.

$$\Delta = C_{\text{submerged}} - C_{\text{emergent}}, \quad (1)$$

where  $\Delta$  was the concentration difference between the two groups of aquatic plants and  $C_{\text{submerged}}$  and  $C_{\text{emergent}}$  were the average concentrations of individual compounds in the leaves of the submerged and emergent plants, respectively.

$$R_{\Delta} (\%) = \Delta / C_{\text{submerged}} \times 100, \quad (2)$$

where  $R_{\Delta}$  was the percentage of concentration difference relative to the concentration in the submerged plants. Based on the above equations,  $R_{\Delta}$  for  $p$ ,  $p'$ -DDE and HCB was estimated at 55% and 40%, respectively, and the  $R_{\Delta}$  values for PAHs and s-PAHs were 10%–69% and 0.5%–79%, respectively. Interestingly,  $R_{\Delta}$  for the high molecular weight PAHs (Acy, Ace, FE, Ph, An, and Fl) was  $56\% \pm 12\%$ , significantly ( $p = 0.031$ ) higher than that for the low molecular weight PAHs (BaP, IP, BP, and DA;  $33\% \pm 15\%$ ). Based on the concentrations reported in the abovementioned study of the exposure of freshwater plants to  $^{36}\text{Cl}$ -DDT (Meeks, 1968),  $R_{\Delta}$  for  $^{36}\text{Cl}$ -DDT was 58%. That figure is comparable to the  $R_{\Delta}$  value obtained for  $p$ ,  $p'$ -DDE in this study (55%). This similarity indicates that the calculation method described above offers an appropriate means of estimating differences in concentration between submerged and emergent freshwater plants.

Previous phytoaccumulation studies have focused on the accumulation of pollutants in terrestrial plants or vegets, reporting that organic pollutants can be absorbed by leaves via the air-to-leaf pathway, which plays an important role in removing semi-volatile organic compounds from the atmosphere (Nakajima et al., 1995; Paterson et al., 1991; Pausch et al., 1995; Simonich and Hites, 1995; Tolls and McLachlan, 1994). In contrast, only a few studies have investigated the phytoaccumulation of contaminants in aquatic plants, and these studies considered only phytoaccumulation in the leaves of emergent plants. In this study, the leaves of submerged plants were found to accumulate significantly more pollutants than the leaves of emergent plants, probably due in part to water-to-leaf uptake pathway. Since log Kow (Table S3) is related to the partition of pollutants between water and biotas, thus used to correlated to the obtained  $R_{\Delta}$ . A significantly ( $p = 0.032$ ) statistical linear regression was found between  $R_{\Delta}$  and log Kow for all the target pollutants (Fig. 4), thus the concentration differences between submerged and emergent plants increased with increasing log Kow of organic compounds. The possible mechanism could be that the uptake of organic compounds into aquatic plant was driven by diffusion between leaves and water, largely dependent on the hydrophobicity of compounds (Stottmeister et al., 2003).

## 4. Conclusion

In summary, this study detected low concentrations of  $p$ ,  $p'$ -DDE and HCB and high residue levels of PAHs and s-PAHs in 10 aquatic plants from Lake Dianchi. Significantly higher concentrations of target compounds were detected in the leaves of the submerged plants compared to those of the emergent plants, suggesting the

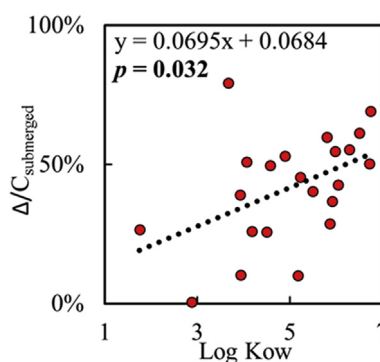


Fig. 4. Relationship between log Kow and ratio of the concentration difference to the concentration in submerged plant group of individual compounds.

water-to-leaf pathway as a more efficient uptake route than the air-to-leaf pathway. The percentages of concentration difference relative to the concentrations in the submerged plants were estimated at 55%, 40%, 10%–69% and 0.5%–79% for *p*, *p*'-DDE, HCB, PAHs, and *s*-PAHs, respectively. The percentages were found to increase significantly with an increase in log K<sub>ow</sub>. The results indicated that high level of phytoaccumulation of pollutants in aquatic plants is mainly due to hydrophobicity-dependent diffusion via the water-to-leaf pathway.

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## Appendix A. Supplementary data

Supplementary data related to this article can be found at <https://doi.org/10.1016/j.envpol.2018.05.071>.

Text, figures, and tables addressing: (1) Map of Lake Dianchi and sampling sites; (2) Number of individual samples collected from each location in Lake Dianchi; (3) spiked amounts, method detection limits, recovery rates and quantification accuracies of individual chemicals; (4) Log K<sub>ow</sub> values of individual chemicals.

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