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# Optimized cleanup method for the determination of short chain polychlorinated *n*-alkanes in sediments by high resolution gas chromatography/electron capture negative ion–low resolution mass spectrometry

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ABSTRACT

The performances of three adsorbents, *i.e.* silica gel, neutral and basic alumina, in the separation of short chain polychlorinated *n*-alkanes (sPCAs) from potential interfering substances such as polychlorinated biphenyls (PCBs) and organochlorine pesticides were evaluated. To increase the cleanup efficiency, a two-step cleanup method using silica gel column and subsequent basic alumina column was developed. All the PCB and organochlorine pesticides could be removed by this cleanup method. The very satisfying cleanup efficiency of sPCAs has been achieved and the recovery in the cleanup method reached 92.7%. The method detection limit (MDL) for sPCAs in sediments was determined to be 14 ng $g^{-1}$. Relative standard deviation (R.S.D.) of 5.3% was obtained for the mass fraction of sPCAs by analyzing four replicates of a spiked sediment sample. High resolution gas chromatography/electron capture negative ion–low resolution mass spectrometry (HRGC/ECNI–LRMS) was used for sPCAs quantification by monitoring $[M-HCl]^{\bullet -}$ ions. When applied to the sediment samples from the mouth of the Daliao River, the optimized cleanup method in conjunction with HRGC/ECNI–LRMS allowed for highly selective identifications for sPCAs. The sPCAs levels in sediment samples are reported to range from 53.6 ng $g^{-1}$ to 289.3 ng $g^{-1}$. $C_{10}$- and $C_{11}$-PCAs are the dominant residue in most of investigated sediment samples.



## 1. Introduction

Polychlorinated *n*-alkanes (PCAs) are commercial mixtures which have been synthesized for use as lubricant additives, cutting fluids, plasticizers and flame retardants since the 1930s [1,2]. They have been detected in most of the environmental compartments including air, water, sediments and biota in the past ten years [1,3]. Of particular interests are the short chain polychlorinated *n*-alkanes (sPCAs) with carbon chain lengths from 10 to 13 and a chlorine content between 30% and 72% by weight [4], which have the greatest potential for environmental release and the highest toxicity [5–7]. In 2007, European Community and its member states proposed sPCAs as persistent organic pollutants (POPs) candidate in document UNEP/POPS/COP.3/12 [8].

The environmental analysis of sPCAs is very challenging due to their complex composition, variation in congener patterns in the environment, and the lack of suitable reference standards [9]. So far, there is no standard routine analysis technique for reliable determination of sPCAs [10]. In the last two decades, numerous analytical approaches have been developed for the analysis of sPCAs in environmental matrix, which has been reviewed in detail [1,11,12]. Among them, high resolution gas chromatography (HRGC) coupled with mass spectrometry (MS) in electron capture negative ion (ECNI) mode is the most widely used approach, due to its high selectivity and sensitivity [10,13]. However, some organochlorine compounds, such as polychlorinated biphenyls (PCBs) and some organochlorine pesticides, can interfere with the quantitative analysis of sPCAs, even using high resolution mass spectrometry (HRMS) [14]. Therefore, it is very necessary to establish a highly efficient cleanup method in order to separate sPCAs from these interfering substances [10,15].

Column chromatography is mostly used for the cleanup of sPCAs in environmental matrix [3,11]. Adsorbents such as silica gel, Florisil, and alumina are frequently employed. Several researchers adopted Florisil fully activated or partially deactivated with water to separate sPCAs from other contaminants in sediments using hexane and dichloromethane (DCM) as elution solvents [14,16]. Using this method, sPCAs could be separated from all the PCBs, chlorinated benzenes, DDT and its metabolites. But more polar organochlorine compounds such as heptachlor epox-

*Abbreviations:* HRGC/ECNI–LRMS, high resolution gas chromatography/electron capture negative ion–low resolution mass spectrometry; sPCAs, short chain polychlorinated *n*-alkanes; PCBs, polychlorinated biphenyls; GPC, gel permeation chromatography; SIM, selected ion monitoring.

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ide and dieldrin are eluted with sPCAs in the same fraction [3]. Silica gel column deactivated with water (≤5%) have also been used for the separation of sPCAs from sediment, sludge and biota [17–20]. With the same elution solvents on Florisil, the sPCAs fraction contained benzenehexachloride (HCH), heptachlor epoxide, and toxaphene, etc. [3]. Steven et al. [21] proposed a combination of silica gel with 3% water and alumina in a single cleanup column in the analysis of sPCAs in sludge, but no recovery data were reported. Marvin et al. [22] proposed a successive cleanup method using Florisil with 1.2% water and activated alumina, and reported that the internal standards recoveries were generally higher than 75%. However, Rieger and Ballschmiter [23] did not recommend the use of activated alumina considering the likelihood of dehydrochlorination of chlorinated paraffins during their adsorption on activated alumina. Besides, Parera et al. [16] found it difficult to elute sPCAs on activated alumina using pure DCM. Hence, the use of activated alumina as adsorbent of column chromatography for sPCAs analysis still needs further study.

As shown from the results of the laboratory intercomparison study organized by Pellizzato et al. [10], the analysis of sPCAs is far from being satisfactory. The lack of sufficient cleanup of the extract is one of the important reasons for the discrepancy in the results of sPCAs analysis among different laboratories. It is essential for the routine and reliable determination of sPCAs to establish a critical cleanup procedure which is selective enough to avoid potential interferences from the other organochlorine compounds. In this study, two-step column chromatography using silica and alumina (neutral or basic) as adsorbents were evaluated by comparing the recoveries and selective separation of sPCAs from their potential interfering substances such as PCBs, 17 different organochlorine pesticides and toxaphene. Quality parameters for the proposed cleanup method combined with HRGC/ECNI–low resolution mass spectrometry (HRGC/ECNI–LRMS) were established. China is the largest producer of PCAs worldwide [1], however, the studies of sPCAs in China are very limited. In this study, the analytical method established was applied for the determination of sPCAs mass fractions and congener patterns in sediment samples from the mouth of the Daliao River, which run through the largest chemical industry areas in Liaoning Province, China. It is hoped to provide insight into the level of sPCAs in China.

## 2. Materials and methods

### 2.1. Reagents and materials

Three stock standard solutions of sPCAs (51%, 55.5% and 63% chlorine content, 100 ng $\mu L^{-1}$ in cyclohexane) and four stock standard solutions of chloroparaffin $C_{10}$-, $C_{11}$-, $C_{12}$- and $C_{13}$-PCAs (65.0%, 55.2%, 55.0% and 55.0% chlorine content, 10 ng $\mu L^{-1}$ in cyclohexane) were obtained from Dr. Ehrenstorfer (Augsburg, Germany). The sPCAs stock solutions with 53.5% and 59% chlorine content were achieved by 1:1 (v/v) mixing of the sPCAs stock standard solutions with 51% and 55.5% chlorine content as well as with 55.5% and 63% chlorine content, respectively. 1,2,5,6,9,10-hexachlorodecane (98% purity) which was used for the selection of monitoring ions of sPCAs in SIM mode, was synthesized by the addition reaction between 1,5,9-decatriene and trimethyl chlorosilane in our laboratory. $^{13}C_6$-α-HCH, $^{13}C_{10}$-*trans*-chlordane and $^{13}C_6$-hexachlorobenzene ($^{13}C_6$-HCB) were all purchased from Cambridge Isotope Laboratories (CIL, Andover, USA). The stock standard solution of 17 organochlorine pesticides was obtained from SUPELCO (Bellefonte, USA) and diluted to 1 mg $L^{-1}$ in hexane for each compound. The standard solution of toxaphene mixtures and dioxin-like PCBs mixtures were purchased from Chinese CRM/RM Information Center (Beijing, China) and CIL (Andover, USA), respectively.

The solvents, *i.e.* *n*-hexane, DCM and acetone, were of pesticide residue grade and obtained from J.T. Baker (Phillipsburg, USA). Anhydrous $Na_2SO_4$ (Damao, China) was cleaned with *n*-hexane in an ultrasonic bath for 30 min and was dried at 300 °C for 2 h before use. Silica gel (63–100 μm), neutral alumina (Brockmann I, activated, ~100 μm, pH 7.0 ± 0.5) and basic alumina (Activity Super I, 63–200 μm, pH 10) were purchased from Sunchrom (Friedrichshafen, Germany), Sigma–Aldrich (St. Louis, USA) and MP Biomedicals (Eschwege, Germany), respectively. Prior to use, all the adsorbents were extracted with DCM by accelerated solvent extraction (ASE 350, Dionex, USA) at the temperature of 120 °C for three cycles, and then activated at 130 °C for 10 h. Acid silica gel was prepared by mixing 200 g activated silica gel and 56.4 g concentrated sulfuric acid, and stored in desiccator.

### 2.2. Sample extraction and cleanup

An aliquot of 20 g sediment sample (dry weight) was spiked with the internal standards, and then Soxhlet extracted with 250 mL hexane/acetone (1:1, v/v) for 24 h. The extract was evaporated to ~1 mL for cleanup.

Sulfur-containing compounds were removed by gel permeation chromatography (GPC). The GPC system consisted of a high pressure pump (P230, Elite Analytical Instruments Co., Ltd, China), a glass column (600 mm × 25 mm i.d.) packed with 70 g of 200–400 mesh SX-3 Bio-beads (Bio-Rad Laboratories, Richmond, CA), and a UV detector (BT3030, Biotronik, Germany). The detection wavelength was 278 nm. DCM was used as the mobile phase at a flow rate of 5 mL $min^{-1}$. The injection volume was 1 mL. The fraction between 19 and 35 min was collected and evaporated to ~1 mL for further cleanup. The recovery of sPCAs on GPC reached 98.3%, which was calculated by sPCAs stock standard solution (55.5% chlorine content) with the concentration of 100 mg $L^{-1}$.

Three adsorbents namely silica gel, neutral and basic alumina were studied to develop an efficient cleanup procedure for the sPCAs analysis in the environmental samples. The silica gel column was packed with 5 g anhydrous $Na_2SO_4$, 2 g silica gel, 4.5 g acid silica gel and 6 g anhydrous $Na_2SO_4$ from bottom to top. The alumina column was packed with 5 g anhydrous $Na_2SO_4$, 5 g alumina and 6 g anhydrous $Na_2SO_4$ from bottom to top. Fractionation was carried out on these three differently packed columns with hexane and DCM. The fraction containing sPCAs was reduced to ~0.5 mL and transferred to a vial. The solution in the vial was further concentrated to near dryness by a gentle stream of $N_2$. Immediately prior to analysis, 0.2 ng of $^{13}C_6$-HCB in 10 μL of hexane was added in the vial as a recovery standard.

### 2.3. HRGC/ECNI–LRMS

The analysis of sPCAs was performed on Trace GC Ultra gas chromatograph (Thermo, USA) coupled with Trace DSQ II mass spectrometer (Thermo, USA) in ECNI mode. A capillary DB-5 column (15 m × 0.25 mm i.d. × 0.25 μm film thickness, J&W Scientific, USA) was used, and sample volume of 1 μL was injected in the splitless mode with an injector temperature of 260 °C. Helium was used as carrier gas at a flow rate of 0.8 mL $min^{-1}$. The temperature program was as follows: initial 100 °C, hold for 1 min, then ramp to 260 °C at 7 °C $min^{-1}$, hold for 8 min. Methane (99.995% purity) was used as ECNI reagent gas with a flow rate of 2 mL $min^{-1}$. The electron energy was 70 eV and the emission current was 100 μA. Ion source and transfer line temperatures were kept at 150 °C and 260 °C, respectively. The dwell times in the selected ion monitoring (SIM) mode was set to 75 ms for each ion, which were indicated in Table 1.

**Table 1**
Calculated *m/z* values of $[M-HCl]^{\bullet-}$ ions of sPCAs congeners used for quantification and confirmation and their relative abundances.

| sPCAs congener | *m/z* values | |
|---|---|---|
| | Quantification ion (100% abundance) | Confirmation ion (% abundance) |
| $C_{10}H_{17}Cl_5$ | 278.0 | 276.0 (75.0) |
| $C_{10}H_{16}Cl_6$ | 312.0 | 314.0 (66.7) |
| $C_{10}H_{15}Cl_7$ | 345.9 | 347.9 (83.3) |
| $C_{10}H_{14}Cl_8$ | 381.9 | 379.9 (100) |
| $C_{10}H_{13}Cl_9$ | 415.8 | 413.8 (85.7) |
| $C_{10}H_{12}Cl_{10}$ | 449.8 | 451.8 (77.8) |
| $C_{11}H_{19}Cl_5$ | 292.0 | 290.0 (75.0) |
| $C_{11}H_{18}Cl_6$ | 326.0 | 328.0 (66.7) |
| $C_{11}H_{17}Cl_7$ | 359.9 | 361.9 (83.3) |
| $C_{11}H_{16}Cl_8$ | 395.9 | 393.9 (100) |
| $C_{11}H_{15}Cl_9$ | 429.9 | 427.9 (85.7) |
| $C_{11}H_{14}Cl_{10}$ | 463.8 | 465.8 (77.8) |
| $C_{12}H_{20}Cl_6$ | 340.0 | 342.0 (83.3) |
| $C_{12}H_{19}Cl_7$ | 374.0 | 375.9 (83.3) |
| $C_{12}H_{18}Cl_8$ | 409.9 | 407.9 (100) |
| $C_{12}H_{17}Cl_9$ | 443.9 | 441.9 (85.7) |
| $C_{13}H_{21}Cl_7$ | 388.0 | 390.0 (83.3) |
| $C_{13}H_{20}Cl_8$ | 423.9 | 421.9 (100) |
| $C_{13}H_{19}Cl_9$ | 457.9 | 455.9 (85.7) |

For PCB congeners, $[M]^-$ ions were the most abundant under the instrument condition as above. Therefore, the two most abundant isotopic *m/z* values of $[M]^-$ ions of each congener [24] were monitored for quantification and confirmation. Toxaphene was also analyzed by HRGC/ECNI–LRMS in SIM mode as the same instrument condition. $[M-Cl]^-$ ions were selected as the monitoring ions. The quantification of toxaphene followed the total mass chromatogram analysis method [25,26], which quantified toxaphene by the sum of total areas of monitoring ions below the elution profile of individual congeners obtained in GC spectrum.

For the analysis of organochlorine pesticides, a GC instrument (HP 5890) equipped with the same capillary DB-5 column as above and electron-capture detector (ECD) was used. The temperature program was as follows: initial 60 °C, hold for 3 min, then ramp to 280 °C at 12 °C $min^{-1}$, hold for 20 min. The temperature of detector and injection port was 280 °C and 270 °C, respectively. The GC was run in splitless mode, and the injection volume was 1 μL. $N_2$ was used as the carrier gas with a flow rate of 1.2 mL $min^{-1}$.

### 2.4. Quantification procedure

The quantification of sPCAs in this study followed the procedure described by Reth et al. [9], which allowed the differences in the chlorine contents and patterns between reference sPCAs mixtures and the sPCAs present in environmental samples. A linear correlation between the total response factor of sPCAs standard mixture and the chlorine content was obtained by analyzing five sPCAs stock solutions with different chlorine content (51.5–63%). The total sPCAs amount and its congener composition in sediment sample were calculated by the ratio of the relative total area of the sample to the corrected total response factor according to the equations available in the Supporting Information.

## 3. Results and discussion

### 3.1. Selection of m/z values for SIM mode

Preliminary studies were carried out to select representative ion *m/z* values for quantification of sPCAs in sediment samples purified by column chromatography. The response factor of sPCAs and the patterns of generated characteristic ions in the ECNI mode were demonstrated to be sensitive to the chlorine content of individual polychloroalkanes and ion source conditions [14,27]. The ECNI mass spectra of 1,2,5,6,9,10-hexachlorodecane at the ion source temperature of 150 °C was shown in Fig. 1. Most abundant characteristic $[M-HCl]^{\bullet-}$ ion (*m/z* 312, referred to ions with the largest isotopic abundance ratio, the same below) dominated in the mass spectra, while small quantities of $[M-Cl]^-$ (*m/z* 313) and $[M+Cl]^-$ (*m/z* 383) ions were also present. The relative abundances of $[M-Cl]^-$ and $[M+Cl]^-$ ions were about 50% and 10% of that of $[M-HCl]^{\bullet-}$ ions. In addition, $[Cl_2]^{\bullet-}$ (*m/z* 70) and $[HCl_2]^-$ (*m/z* 71) ions and ions corresponding to further successive losses of HCl or $Cl^\bullet$ (*m/z* 277, 240) could also be detected with very low relative abundance. In ECNI mode, some organochlorine compounds of which the H atoms are completely substituted for Cl atoms, cannot produce $[M-HCl]^{\bullet-}$ ions. The monitoring of $[M-HCl]^{\bullet-}$ ions of sPCAs can therefore, efficiently avoid the interferences from these organochlorine compounds, especially those with molecular masses similar to sPCAs (300–500u) [14]. In view of the high relative abundance and the capacity of avoiding interferences, $[M-HCl]^{\bullet-}$ ions were chosen as the monitoring fragment ions for sPCAs quantification in our study.

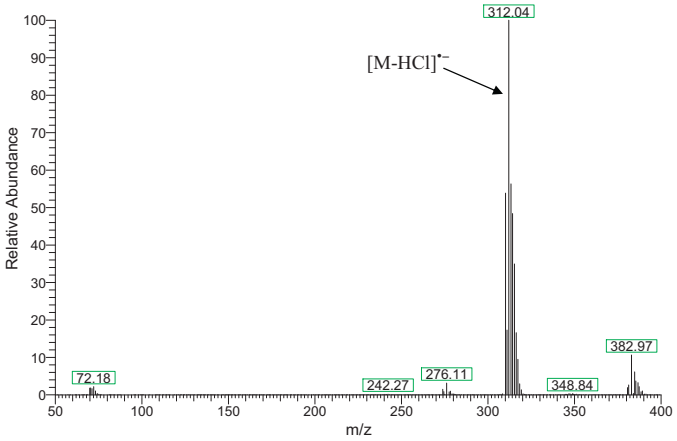


**Fig. 1.** ECNI mass spectrum of 1,2,5,6,9,10-hexachlorodecane at ion source temperature of 150 °C.

Considering the efficiency of SIM mode, it was not possible and valuable to monitor a number of ions. According to Tomy et al. [14] and the homologue patterns of sPCAs in standard solutions adopted in our study, 19 representative sPCAs homologues were chosen for monitoring. The two most abundant isotopic *m/z* values of $[M-HCl]^{\bullet-}$ ions of each congener were calculated and used for quantification and confirmation (Table 1). The relative abundances of the confirmation ions were calculated as the percent of the abundances of quantification ions. The elution profile of monitored ions in sPCAs stock standard solution with chlorine content of 55.5% was shown in Fig. S1. It was indicated co-elution of homologues became an issue. The congeners of sPCAs were identified by control of retention time, comparison of signal shape and correcting isotope ratio.

### 3.2. Optimization of cleanup procedure

Three kinds of adsorbents, *i.e.* silica gel, neutral and basic alumina (all activated), were selected for the separation of sPCAs from interfering substances. A series of standard solutions in hexane were prepared to develop a suitable cleanup procedure, including 12 PCBs congeners each at the concentration of 100 μg $L^{-1}$, 17 organochlorine pesticides each at the concentration of 1 mg $L^{-1}$, toxaphene mixture with the concentration of 10 mg $L^{-1}$, as well as the sPCAs mixture (55.5% chlorine content) at the concentration of 20 mg $L^{-1}$. 10 μL of each standard solution was transferred

**Table 2**
Recovery (%) of sPCAs (55.5% chlorine content), PCB congeners, organochlorine pesticides and toxaphene eluted by different adsorbents.

| Compound | Recovery (%) | | | | | |
|---|---|---|---|---|---|---|
| | Silica gel[a] | | Neutral alumina[b] | | Basic alumina[c] | |
| | Fraction SF1 | Fraction SF2 | Fraction NF1 | Fraction NF2 | Fraction BF1 | Fraction BF2 |
| sPCAs | –[d] | 97.6 ± 2.9 | 2.0 ± 0.1 | 97.3 ± 2.3 | 1.1 ± 0.2 | 95.5 ± 4.2 |
| PCBs | –[d] | | | | | |
| PCB-77 | 80.3 ± 8.9 | | | | | |
| PCB-81 | 83.7 ± 12.1 | | | | | |
| PCB-105 | 88.9 ± 8.2 | | | | | |
| PCB-114 | 87.5 ± 9.4 | | | | | |
| PCB-118 | 90.3 ± 7.3 | | | | | |
| PCB-123 | 102.3 ± 8.8 | | | | | |
| PCB-126 | 95.6 ± 7.9 | | | | | |
| PCB-156 | 107.4 ± 6.0 | | | | | |
| PCB-157 | 106.8 ± 6.7 | | | | | |
| PCB-167 | 98.7 ± 6.3 | | | | | |
| PCB-169 | 104.1 ± 5.7 | | | | | |
| PCB-189 | 105.2 ± 5.3 | | | | | |
| Organochlorine pesticides | | | | | | |
| DDT | 97.8 ± 10.6 | – | – | 98.0 ± 0.8 | – | 97.1 ± 0.9 |
| DDD | 86.9 ± 0.9 | – | – | 2.1 ± 0.1 | – | – |
| DDE | 86.4 ± 3.8 | – | – | 96.4 ± 4.2 | 13.2 ± 0.3 | 76.0 ± 0.9 |
| Heptachlor | 73.2 ± 8.0 | – | 2.0 ± 0.1 | 4.2 ± 0.4 | 3.7 ± 0.06 | 3.1 ± 0.04 |
| *trans*-Chlordane | – | 89.7 ± 2.1 | – | 65.5 ± 3.0 | – | – |
| Endrin aldehyde | – | 104.6 ± 0.1 | – | 108.5 ± 0.4 | – | 1.5 ± 0.3 |
| Endrin ketone | 1.2 ± 0.06 | 107.5 ± 0.8 | – | 98.1 ± 1.1 | – | – |
| α-HCH | – | 107.3 ± 6.0 | 2.5 ± 0.7 | 103.3 ± 0.1 | – | 90.1 ± 3.9 |
| β-HCH | – | 81.5 ± 4.2 | 2.6 ± 0.3 | 89.1 ± 0.5 | – | 102.4 ± 3.6 |
| γ-HCH | 0.39 ± 0.01 | 52.9 ± 2.6 | 0.7 ± 0.08 | 84.1 ± 0.4 | – | 59.6 ± 1.4 |
| δ-HCH | – | 67.8 ± 3.8 | 2.6 ± 0.1 | 69.3 ± 0.6 | – | 79.8 ± 2.4 |
| Aldrin | 0.65 ± 0.05 | – | 80.3 ± 0.5 | 1.9 ± 0.3 | 88.1 ± 0.3 | – |
| Heptachlor epoxide | – | – | – | 76.1 ± 0.6 | – | 40.2 ± 0.4 |
| Endosulfan I | – | – | – | 38.1 ± 0.2 | – | – |
| Dieldrin | 0.26 ± 0.01 | – | – | – | – | – |
| Endrin | – | – | – | 101.9 ± 0.7 | – | 1.4 ± 0.2 |
| Methoxychlor | – | – | – | 97.0 ± 1.5 | – | 19.2 ± 0.1 |
| Toxaphene | 30.5 ± 1.2 | 39.9 ± 0.3 | 1.7 ± 0.05 | 71.2 ± 1.3 | 63.3 ± 2.4 | 51.8 ± 1.8 |

[a] First eluted by 50 mL hexane (fraction SF1), then 100 mL 1:1 hexane/DCM and 50 mL 1:2 hexane/DCM (fraction SF2).
[b] First eluted by 60 mL hexane (fraction NF1), then 120 mL 1:1 hexane/DCM (fraction NF2).
[c] First eluted by 60 mL hexane (fraction BF1), then 90 mL DCM (fraction BF2).
[d] Undetected.

to the top of the column, and then eluted by *n*-hexane, DCM or their mixtures. The recoveries of sPCAs, PCB congeners, organochlorine pesticides and toxaphene were calculated by the ratio of the amount in the eluate to the added amount.

The silica gel column was first eluted by 50 mL hexane (fraction SF1), then by 100 mL 1:1 hexane/DCM and 50 mL 1:2 hexane/DCM (fraction SF2). Table 2 showed the recovery of sPCAs, toxaphene, PCBs and organochlorine pesticides, respectively. Fraction SF1 contained all the PCBs, heptachlor, DDE, DDD, DDT and part of toxaphene, which was in agreement with previous studies [14,16]. Fraction SF2 contained sPCAs, along with endrin ketone, endrin aldehyde, *trans*-chlordane, HCH and residues of toxaphene. The recovery of sPCAs congeners reached 97.6%. In addition, some more polar organochlorine pesticides such as heptachlor epoxide, dieldrin, endrin and methoxychlor could not be eluted by this elution procedure.

Due to the excellent removal efficiency of PCBs by silica gel column, the elution of PCBs from neutral and basic alumina columns was not performed. For neutral alumina column, the elution was achieved with 60 mL hexane (fraction NF1) and subsequent 120 mL 1:1 hexane/DCM (fraction NF2). For basic alumina, the elution solvents were 60 mL hexane (fraction BF1) followed by 90 mL DCM (fraction BF2).

As shown in Table 2, sPCAs could not be eluted on both alumina columns by hexane, in which the recoveries of sPCAs were only less than 2%. The recoveries of sPCAs in the second fraction were 97.3% on neutral alumina column and 95.5% on basic alumina column, respectively. The elution curves of 1,2,5,6,9,10-hexachlorodecane and sPCAs with 55.5% chlorine content on neutral and basic alumina column are shown in Fig. 2. Their recoveries in the second fraction on both alumina columns reached more than 95%, among which about 70% of mass were eluted in the first 30 mL of 1:1 hexane/DCM on neutral alumina column and 30 mL of DCM on basic alumina column. Some more polar sPCAs congeners would be eluted later.

For the elution of the organochlorine pesticides on neutral alumina column, a weak retention of aldrin was found, and its recovery in fraction F1 was above 80%. The adsorption of DDD, heptachlor and dieldrin on neutral alumina was so strong that these compounds almost could not be eluted by this procedure. The other organochlorine pesticides were mostly eluted with sPCAs in the second fraction (Table 2). For the elution of organochlorine pesticides on basic alumina column, aldrin was contained in hexane fraction, which was similar with that on neutral alumina column. DDT, DDE, HCH and part of heptachlor epoxide were contained in DCM fraction co-eluted with sPCAs (Table 2). More organochlorine pesticides exhibited such strong retentions that even DCM could not elute them from basic alumina column, including DDD, heptachlor, *trans*-chlordane, endrin aldehyde, endrin ketone, endosulfan I, dieldrin, endrin and the greatest part of methoxychlor and heptachlor epoxide. It was obvious that the separation efficiency of sPCAs from organochlorine pesticides on basic alumina column was better than that on neutral alumina column. However, either single silica gel column or alumina column could not eliminate the interferences from organochlorine pesticides completely,

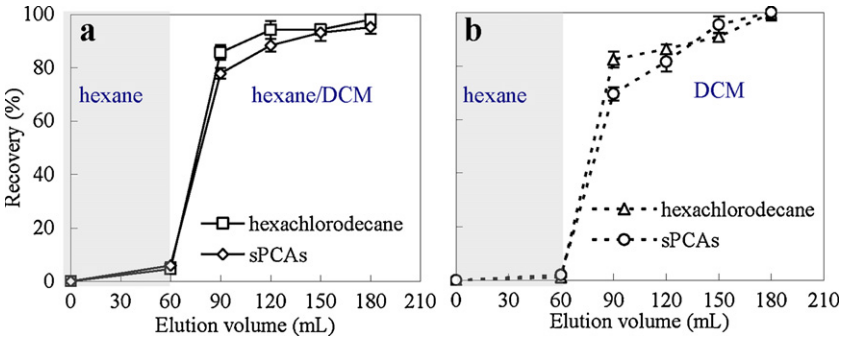


**Fig. 2.** The elution curves of 1,2,5,6,9,10-hexachlorodecane and sPCAs with 55.5% chlorine content on (a) neutral alumina column (first eluted by 60 mL hexane, then 120 mL 1:1 hexane/DCM) and (b) basic alumina column (first eluted by 60 mL hexane, then 120 mL DCM).

which was shown in Table 2. Based on the elution characters of organochlorine pesticides, a successive use of silica gel column and basic alumina column could separate them from sPCAs effectively.

Toxaphene is a mixture of approximately 200 congeners with an overall chlorine content of 67–69% by weight. They have similar molecular masses of sPCAs. Moreover, in ECNI mode, toxaphene could interfere with the monitoring of sPCAs [14,17]. As shown in Table 2, it was difficult to separate toxaphene from sPCAs completely by a single cleanup column. The successive application of silica gel column and basic alumina allowed a considerable separation between sPCAs and toxaphene. About 40% of toxaphene was eluted in the fraction SF2 on silica gel column, while on the subsequent basic alumina column, about one half of toxaphene was eluted in fraction BF2 (Table 2). These results demonstrated that about 20% of toxaphene would still be co-eluted along with sPCAs using the two-step cleanup method. However, when using GPC to remove sulfur-containing compounds in sediment samples, it is found that toxaphene could be efficiently separated from sPCAs by controlling retention time (see Fig. S2). The result was in accordance with that in the study of Coelhan [17], in which GPC was used to separate residual toxaphene from sPCAs after pretreatment by a silica gel column.

Considering the recoveries of sPCAs and their separation efficiency from PCBs, organochlorine pesticides and toxaphene, a two-step cleanup method using silica gel column and subsequent basic alumina column following Soxhlet extraction and GPC was developed and used for the analysis of sPCAs in sediment samples. As shown in Table 2, four kinds of HCH (α-HCH, β-HCH, γ-HCH, and δ-HCH) had similar elution characteristics with sPCAs on silica gel column and basic alumina. In addition, they did not interfere with the monitoring of sPCAs in ECNI mode. Therefore, the $^{13}C$-isotope labeled HCH was regarded to be a suitable internal standard in the two-step cleanup method, and the $[M-Cl]^-$ ions ($m/z$ 259) were monitored in ECNI mode.

### 3.3. Quality assurance and quality control

Precautions were taken in the use of glassware, solvents and adsorbent materials. All glassware was washed with neutralizing acid rinse lab solutions and non-foaming powder detergent (Labconco, Kansas, USA), and rinsed with MilliQ water. Before use, it was rinsed by hexane for three times. Adsorbent materials were pretreated as described in Section 2.1.

The analytical limit of detection (LOD) for sPCAs was estimated to be 50 μg $L^{-1}$ at a signal-to-noise ratio (S $N^{-1}$) of 3:1. For PCBs congeners, the response factor in ECNI mode increased with their increasing chlorine content. The LOD value of PCB-81, of which the chlorine content was the lowest, was determined to be 20 μg $L^{-1}$. The LOD value of toxaphene as a whole in ECNI mode was estimated to be 100 μg $L^{-1}$. The LOD values of organochlorine pesticides on ECD were determined to range from 5 μg $L^{-1}$ to 20 μg $L^{-1}$.

The solvent blank was carried out by concentrating 10 mL of elution solvent (hexane, DCM and acetone) to 10 μL and then analyzing sPCAs using HRGC/ECNI–LRMS. The adsorbent blank was performed by extracting 1 g of adsorbent (silica gel, neutral alumina, and basic alumina) with 10 mL DCM in an ultrasonic bath for 30 min. The extract was concentrated to 10 μL and then analyzed using HRGC/ECNI–LRMS. For all the adsorbents and solvents, no sPCAs were detected out.

The blank sediment sample was prepared by 2# sediment sample using Soxhlet extraction, and was analyzed by HRGC/ECNI–LRMS to ensure that no detectable quantities of sPCAs were present. Four replicates of 20 g blank sediment samples spiked with sPCAs (55.5% chlorine) and $^{13}C_6$-α-HCH at the mass fractions of 10 ng $g^{-1}$ and 0.01 ng $g^{-1}$ were pretreated by GPC and the two-step cleanup method, and then analyzed over a week. 0.2 ng of $^{13}C_6$-HCB in 10 μL of hexane was used as the recovery standard. The run-to-run precision was established for sPCAs in sediment samples. A relative standard deviation (R.S.D.) of 5.3% was obtained for the mass fraction of sPCAs. The average recoveries of sPCAs and $^{13}C_6$-α-HCH were calculated to be 92.7% and 92.2%. The method detection limit (MDL) of total sPCAs was determined by measuring the mean of the background signals from blank samples, and were reported as this mean plus 3 times the standard deviation. The MDL was 14 ng $g^{-1}$ ($n$ = 4 samples).

### 3.4. Analysis of sediment samples

The optimized cleanup method was applied to the analysis of sPCAs levels in sediment samples. Samples were collected from the mouth of the Daliao River, which as one of the most heavily polluted rivers in China runs though Liaoning Province and finally enters into the Bo Sea Bay [28]. The sPCAs level in the sediments from the mouth of the Daliao River is of important significance for the regional pollution survey.

As an example, the chromatogram of sPCAs in 1# sediment sample pretreated by the optimized cleanup method was shown in Fig. 3. A broad chromatographic profile occurred due to the co-elution of sPCAs congeners in sediment samples. The elution pattern of sPCAs in 1# sediment sample appeared to be sufficiently similar compared with that of sPCAs stock solution with 55.5% chlorine content. This result demonstrated definitively that the optimized cleanup method could achieve satisfying cleanup efficiency for sPCAs in sediment samples. It makes it possible for the laboratory intercomparison on the analysis of sPCAs in environmental samples without interferences from other organochlorine compounds. During the quantification procedure of sPCAs, $^{13}C_6$-α-HCH and $^{13}C_6$-HCB were used as the internal standard and the

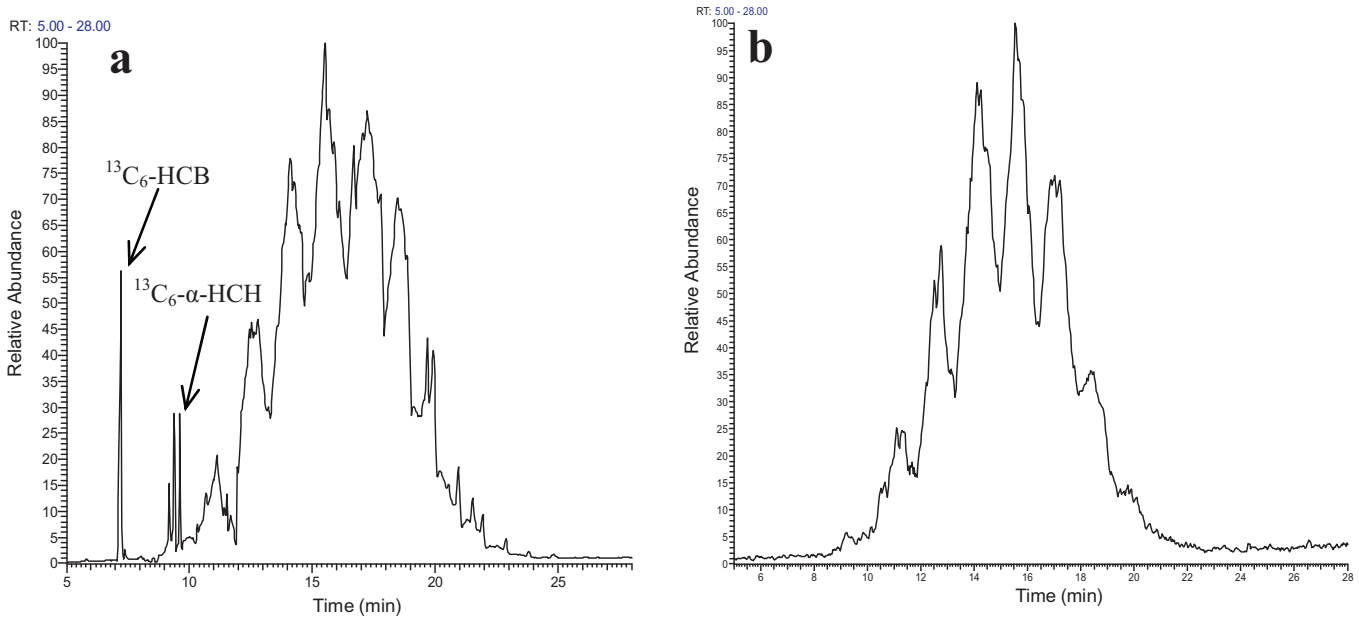


**Fig. 3.** Chromatograms of sPCAs (a) in a sediment sample pretreated by the optimized cleanup method and (b) in an sPCAs stock solution with 55.5% chlorine content.

recovery standard instead of $^{13}C_{10}$-*trans*-chlordane and ε-HCH in the study of Reth et al. [9], considering the elution characteristics on silica gel column and basic alumina in our research. A linear correlation ($R^2$ = 0.986, Fig. S3) between the total response factor of sPCAs mixture and the chlorine content was obtained.

The chlorine contents of sPCAs in the sediment samples varied between 60.2% and 61.0%. The mass fraction of sPCAs in sediment samples were determined and summarized in Table 3. In all samples, sPCAs was detected out and their mass fractions in the sediment samples range from 53.6 to 289.3 ng g$^{-1}$ dry weight (dw). It was in the same level as the sPCAs mass fractions determined in sediments of an industrial area in Czech Republic (4.58–180.75 ng g$^{-1}$ dw) [18], and in river sediments investigated in Japan (4.9–484.4 ng g$^{-1}$ dw) [29]. Zeng et al. [30] investigated the sPCAs levels in soils from wastewater irrigated farmlands in Beijing. The sPCAs mass fractions was reported to be 159.9–1450 ng g$^{-1}$ dw. The values were higher than the sPCAs level in sediments from the mouth of the Daliao River, which was due to the sampling sites in the study of Zeng et al. [30] near a large sewage treatment plant. The main sources of sPCAs in the sediment from the mouth of the Daliao River were speculated to be the discharge of chemical plants nearby and deposition of suspended particulate matters adsorbed

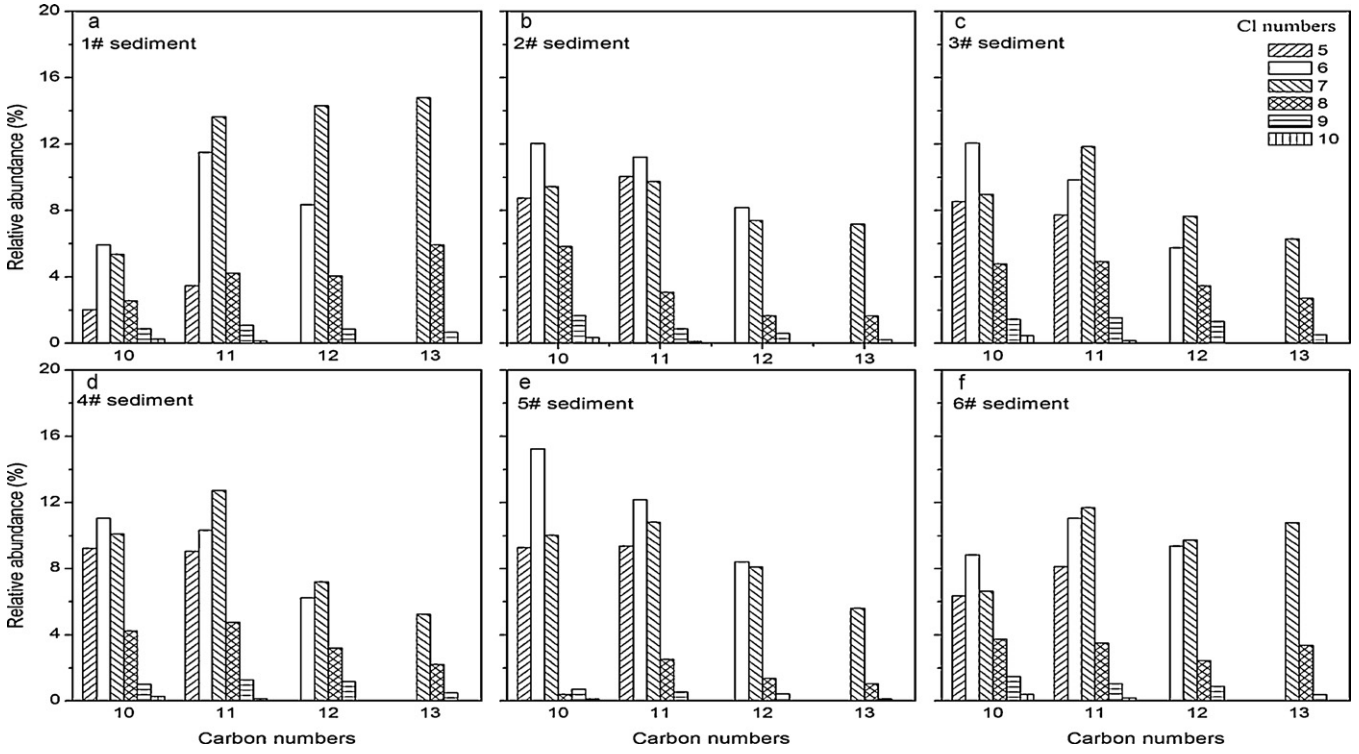


**Fig. 4.** Homologue pattern ($C_{10}$–$C_{13}$) of sPCAs in sediment samples from the mouth of the Daliao River determined by HRGC/ECNI–LRMS.

**Table 3**
Mass fractions of sPCAs and its congeners in sediment samples from the mouth of the Daliao River obtained with the optimized cleanup method.

| Sediment sample no. | $^{13}C_6$-α-HCH recovery (%) | Cl% | sPCAs mass fraction (ng g$^{-1}$) | | | | |
|---|---|---|---|---|---|---|---|
| | | | $\sum C_{10}$ | $\sum C_{11}$ | $\sum C_{12}$ | $\sum C_{13}$ | Total |
| 1# | 109.3 | 60.8 | 49.1 | 98.6 | 79.8 | 61.8 | 289.3 |
| 2# | 92.2 | 60.5 | 86.9 | 80.0 | 40.7 | 20.6 | 228.2 |
| 3# | 107.0 | 61.0 | 38.8 | 38.6 | 19.5 | 10.2 | 107.1 |
| 4# | 81.0 | 60.8 | 29.1 | 31.0 | 14.4 | 6.5 | 81.0 |
| 5# | 116.2 | 60.2 | 43.2 | 42.8 | 22.1 | 8.1 | 116.2 |
| 6# | 88.3 | 60.4 | 14.7 | 19.1 | 12.0 | 7.8 | 53.6 |

by sPCAs. Fig. 4 compared the homologue patterns of sPCAs in sediment samples. It was indicated that $C_{10}$- and $C_{11}$-PCAs predominate over the homologue pattern of sPCAs in the analyzed sediments with the exception of 1# sediment sample. The highest sPCAs level was found in 1# sediment, of which the percentages of $C_{11}$- and $C_{12}$-PCAs were a little higher that that of $C_{10}$- and $C_{13}$-PCAs. Six and seven chlorine substituted sPCAs seemed to be the major species.

## 4. Conclusion

The analysis of sPCAs is complicated by the achievement of an efficient cleanup method because of the complex composition and the strong interferences from other organochlorine compounds. The cleanup efficiencies of three adsorbents, *i.e.* silica gel, neutral and basic alumina, were compared. The results indicate that a two-step cleanup method using silica gel column and subsequent basic alumina column following the Soxhlet extraction and GPC could completely eliminate the interferences from all the PCBs, 17 organochlorine pesticides tested and toxaphene, which was of importance for the analysis of sPCAs by LRMS. In addition, GPC was used not only for the removal of sulfur-containing compounds in sediment samples, but also for the separation of sPCAs from toxaphene. The separation efficiency was pretty satisfying. The recoveries of sPCAs during the cleanup procedure reached 92.7%. The optimized cleanup method in conjunction with HRGC/ECNI–LRMS was applied to analyze sPCAs in sediment samples from the Daliao River. $[M-HCl]^{\bullet-}$ ions of 19 homologues were selected as the monitoring fragment ions. The MDL was determined to be 14 ng g$^{-1}$. The mass fractions of sPCAs in the sediment samples ranged from 53.6 ng g$^{-1}$ to 289.3 ng g$^{-1}$. With the increasing attentions to sPCAs by Chinese government, it is also hoped to be helpful in investigating the level of sPCAs inventory for China and in developing a strategy for reduction of sPCAs in the environment of China in future.

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## Appendix A. Supplementary data

Supplementary data associated with this article can be found, in the online version, at doi:10.1016/j.aca.2011.07.041.

## References

[1] S. Bayen, J.P. Obbard, G.O. Thomas, Environ. Int. 32 (2006) 915–929.
[2] S. Geiβ, J.W. Einax, S.P. Scott, Clean 38 (2010) 57–76.
[3] F.J. Santos, M.T. Galceran, Anal. Bioanal. Chem. 386 (2006) 837–857.
[4] UNECE Expert Group on POPs, Short chain chlorinated paraffins (SCCP) substance dossier, final draft II, 2003.
[5] M. Houde, D.C.G. Muir, G.T. Tomy, D.M. Whittle, C. Teixeira, S. Moore, Environ. Sci. Technol. 42 (2008) 3893–3899.
[6] G.T. Tomy, G.A. Stern, W.L. Lockhart, Environ. Sci. Technol. 33 (1999) 2858–2863.
[7] F. Wania, Environ. Sci. Technol. 37 (2003) 1344–1351.
[8] United Naitons Environment Programme, UNEP/POPS/COP.3/12, Persistent organic pollutants review committee: relevant developments for action by the conference of the parties, Dakar, 2007.
[9] M. Reth, Z. Zencak, M. Oehme, J. Chromatogr. A 1081 (2005) 225–231.
[10] F. Pellizzato, M. Ricci, A. Held, H. Emons, W. Böhmer, S. Geiss, S. Iozza, S. Mais, M. Petersen, P. Lepom, Trends Anal. Chem. 28 (2009) 1029–1035.
[11] E. Eljarrat, D. Barceló, Trends Anal. Chem. 25 (2006) 421–434.
[12] Z. Zencak, M. Oehme, Trends Anal. Chem. 25 (2006) 310–317.
[13] J. Hüttig, M. Oehme, Chemosphere 64 (2006) 1573–1581.
[14] G.T. Tomy, G.A. Stern, D.C.G. Muir, A.T. Fisk, C.D. Cymbalisty, J.B. Westmore, Anal. Chem. 69 (1997) 2762–2771.
[15] S. Iozza, P. Schmid, M. Oehme, Environ. Pollut. 157 (2009) 3218–3224.
[16] J. Parera, F.J. Santos, M.T. Galceran, J. Chromatogr. A 1046 (2004) 19–26.
[17] M. Coelhan, Anal. Chem. 71 (1999) 4498–4505.
[18] P. Stejnarová, M. Coelhan, R. Kostrhounová, H. Parlar, I. Holoubek, Chemosphere 58 (2005) 253–262.
[19] C.R. Nicholls, C.R. Allchin, R.J. Law, Environ. Pollut. 114 (2001) 415–430.
[20] A. Maulshagen, S. Hamm, M. Petersen, O. Elsholz, S. Fengler, P. Seel, Organohalogen Compd. 62 (2003) 371–374.
[21] J.L. Stevens, G.L. Northcott, G.A. Stern, G.T. Tomy, K.C. Kones, Environ. Sci. Technol. 37 (2003) 462–467.
[22] C.H. Marvin, S. Painter, G.T. Tomy, G.A. Stern, E. Braekevelt, D.C.G. Muir, Environ. Sci. Technol. 37 (2003) 4561–4568.
[23] R. Reiger, K. Ballschmiter, Fresenius J. Anal. Chem. 352 (1995) 715–724.
[24] P.S. Haglund, K.I. Wiberg, D.R. Zook, Environ. Sci. Technol. 29 (1995) 2801–2805.
[25] L. Kimmel, D. Angerhöfer, U. Gill, M. Coelhan, H. Parlar, Chemosphere 37 (1998) 549–558.
[26] E. Braekevelt, G.T. Tomy, G.A. Stern, Environ. Sci. Technol. 35 (2001) 3513–3518.
[27] P. Castells, F.J. Santos, M.T. Galceran, Rapid Commun. Mass Spectrom. 18 (2004) 529–536.
[28] H.J. Zhang, X.F. Zhao, Y.W. Ni, X.B. Lu, J.P. Chen, F. Su, L. Zhao, N. Zhang, X.P. Zhang, Chemosphere 79 (2010) 754–762.
[29] F. Iino, T. Takasuga, K. Senthilkumar, N. Nakamura, J. Nakanishi, Environ. Sci. Technol. 39 (2005) 859–866.
[30] L. Zeng, T. Wang, W. Han, B. Yuan, Q. Liu, Y. Wang, G. Jiang, Environ. Sci. Technol. 45 (2011) 2100–2106.