Environmental Pollution 157 (2009) 3218–3224

Contents lists available at ScienceDirect

Environmental Pollution

journal homepage: www.elsevier.com/locate/envpol

Development of a comprehensive analytical method for the determination of chlorinated paraffins in spruce needles applied in passive air sampling

Saverio Iozza [a,b], Peter Schmid [a], Michael Oehme [b,*]

[a] Empa, Swiss Federal Laboratories for Materials Testing and Research, Laboratory for Analytical Chemistry, Überlandstrasse 129, CH-8600 Dübendorf, Switzerland
[b] University of Basel, Department of Chemistry, St. Johanns-Ring 19, CH-4056 Basel, Switzerland

A developed method for chlorinated paraffins (CPs) provided evidence that spruce needles are a suitable passive sampling system for the monitoring of atmospheric CPs.

ARTICLE INFO

Article history:
Received 15 May 2009
Received in revised form
3 June 2009
Accepted 15 June 2009

Keywords:
Polychlorinated *n*-alkanes
Conifer needles
Total CPs
SCCPs
MCCPs
Alps

ABSTRACT

Conifer needles are used for the monitoring of atmospheric persistent organic pollutants. The objective of the present study was to develop a method for the detection of airborne chlorinated paraffins (CPs) using spruce needles as a passive sampler. The method is based on liquid extraction of the cuticular wax layer followed by chromatographic fractionation and detection of CPs using two different GCMS techniques. Total CP concentrations (sum of short (SCCP), medium (MCCP) and long chain CPs (LCCP)) were determined by EI-MS/MS. SCCP and MCCP levels as well as congener group patterns (*n*-alkane chain length, chlorine content) could be evaluated using ECNI-LRMS. For the first time, data on environmental airborne CPs on spruce needles taken within the Monitoring Network in the Alpine Region for Persistent and other Organic Pollutants (MONARPOP) are presented providing evidence that spruce needles are a suitable passive sampling system for the monitoring of atmospheric CPs.

1. Introduction

Conifer needles are suitable as passive sampler for air pollutants. A number of publications described the accumulation of airborne organic compounds in the cuticular wax layer of conifer needles such as PCBs, PCNs, DDTs, HCHs, and dioxins (Buckley, 1982; Gaggi et al., 1985; Eriksson et al., 1989; Jensen et al., 1992; Kylin et al., 1994; Strachan et al., 1994; Levy et al., 2007; Wyrzykowska et al., 2007). Air pollutants in the vapor phase are trapped in the waxy surface, while particle-associated compounds are deposited on the needle surface (Kylin et al., 1994; Strachan et al., 1994). Particularly, conifer needles are well-suited as passive samplers exhibiting a high interception capacity for rain, fog and snow (Umlauf and McLachlan, 1994). Moreover, conifers do not loose their canopy in autumn and, therefore, sampling is possible at all seasons. The amount of accumulated air pollutants generally increases with needle age (Hellström et al., 2004; Romanic and Krauthacker, 2004). Furthermore, the high prevalence of spruce forests in temperate areas allows comparisons of geographical, seasonal, and temporal variations of levels of air pollutants (Eriksson et al., 1989; Jensen et al., 1992; Levy et al., 2007; Romanic and Krauthacker, 2007; Wyrzykowska et al., 2007).

Chlorinated paraffins (CPs), also known as polychlorinated *n*-alkanes (PCAs), were introduced in the 1930s. CPs are produced by the reaction of specific *n*-alkane fractions from petroleum distillation with chlorine. They are differentiated into three main categories according to their carbon chain length: short chain CPs (SCCPs, C_{10-13}), medium chain CPs (MCCPs, C_{14-17}) and long chain CPs (LCCPs, $C_{>17}$) (Muir et al., 2000). The chlorination degree of CPs can vary between 30 and 70% (Muir et al., 2000). CPs are in use for a wide range of industrial applications such as flame retardants and plasticizers in sealants, paints and coatings, and as additives in metal working fluids (Campbell and McConnell, 1980; Tomy et al., 1998).

CPs are persistent chemicals and their physical properties (log K_{OW} 4.4–8, depending on the chlorination degree) imply a high potential of bioaccumulation as well as of global long-range atmospheric transport (Muir et al., 2000). Their presence was detected in the Canadian (Tomy et al., 1999, 2000) and European Arctic (Reth et al., 2006). Moreover, CP levels in the environment increased in the last decades. In a sediment core from Lake Thun, Switzerland, Iozza et al. observed a substantial rise of CP levels in the 1980s (Iozza et al., 2008).

The acute toxicity of CPs is low (World Health Organization, 1996). Nevertheless, SCCPs showed chronic toxicity in aquatic organisms and are carcinogenic in rats and mice (OSPAR Commission, 2001).

* Corresponding author. Tel.: +41 61 703 16 30; fax: +41 61 703 16 31.
E-mail addresses: saverio.iozza@empa.ch (S. Iozza), peter.schmid@empa.ch (P. Schmid), michael.oehme@unibas.ch (M. Oehme).

0269-7491/$ – see front matter © 2009 Elsevier Ltd. All rights reserved.
doi:10.1016/j.envpol.2009.06.033

Therefore, SCCPs were categorized in group 2B as 'possibly carcinogenic to humans' by the International Agency for Research on Cancer (IARC) (World Health Organization, 1996). Due to their higher tendency of bioaccumulation (World Health Organization, 1996) and higher toxicity of SCCPs than MCCPs and LCCPs they have also been included in the list of priority hazardous substances of the European Water Framework Directive (European Community, 2000). For the same reasons, SCCPs are now under discussion to be included in the Stockholm Convention on POPs (Persistent Organic Pollutants Review Committee (POPRC) 2004). Consequently, environmental CP levels should be monitored more comprehensively.

Technical CP mixtures contain thousands of different congeners (homologues and isomers) which cannot be resolved into single congeners by any chromatographic and mass spectrometric technique. Currently, three analytical methods for CPs are used based on gas chromatography coupled with mass spectrometry (GCMS). All methods fully exploit the specificity of mass spectrometry:

- Tandem mass spectrometry (EI-MS/MS) is based on fragment ions with low mass-to-charges ratios, which are common to all CPs. The response factors of different CP mixtures are independent from their chlorine content (Zencak et al., 2004, 2005).
- Electron capture negative ionization with low resolution mass spectrometry (ECNI-LRMS) records single ions of the $[M–Cl]^-$ isotope clusters ions (Schmid and Müller, 1985; Castells et al., 2004; Zencak et al., 2005).
- Electron capture negative ionization with high resolution mass spectrometry (ECNI-HRMS) is a very selective method eliminating interferences from CP fragments with the same nominal m/z values or from other organochlorine compounds (Tomy et al., 1997) not removed by preceding clean-up procedures. Due to the high costs of HR instruments, this method is not available at many laboratories.

LRMS methods based on ECNI or on EI-MS/MS are well-suited for routine analysis (Castells et al., 2004; Zencak et al., 2005). However, a highly efficient clean-up is mandatory to avoid interferences from both matrix and other POPs (Parera et al., 2004; Reth and Oehme, 2004; Hüttig and Oehme, 2005).

Only a few more analytical methods are described in the literature enabling quantitative routine analysis of CPs (Pellizzato et al., 2007). Moreover, only few laboratories analyze CPs worldwide (UNECE ad hoc Expert Group on POPs, 2003), although they are ubiquitously present in the environment including remote areas. Therefore, limited information is available about environmental levels, metabolic pathways and toxicokinetics of CPs compared to other polychlorinated environmental pollutants such as polychlorinated biphenyls (PCBs), dioxins, and organochlorine pesticides (Muir et al., 2000; Pellizzato et al., 2007).

The aim of the present work was to develop an integral analytical method for the determination of CPs in conifer needles. This required the development of a sample clean-up procedure handling the waxy matrix. The properties of the wax differ from lipids present in fish or mammals. Moreover, separation and quantification had to be optimized to minimize interferences from the remaining sample matrix. Besides a detailed method description, first data about CP environmental levels in spruce needles from the Alps taken within the MONARPOP project are presented including recommendations concerning the appropriate handling of spruce needles as passive sampling material.

With support from the European Union the project MONARPOP (Monitoring Network in the Alpine Region for Persistent and other Organic Pollutants) was set up in 2003 by Austria, Germany, Italy, Switzerland, and Slovenia to investigate input, pathways and fate of atmospheric anthropogenic contaminants such as POPs in the Alps (Moche et al., 2005). The monitoring of anthropogenic contaminants including CPs and using spruce needles as passive sampling material is one of the objectives within this project.

2. Experimental

2.1. Chemicals and solvents

Cyclohexane, dichloromethane (DCM) and *n*-hexane (nHex) for residue analysis were obtained from Biosolve (Vallenswaard, Netherlands). The internal standard $^{13}C_{10}$-*trans*-chlordane (100 ng μl^{-1}, solution in *n*-nonane, purity 99%) was purchased by Cambridge Isotope Laboratories (Andover, USA). Reference SCCP ($C_{10–13}$, chlorine contents of 51.5, 55.5 and 63.0%) and reference MCCP mixtures ($C_{14–17}$, chlorine contents of 52.0 and 57.0%) with concentrations of 100 ng μl^{-1} in cyclohexane as well as ε-hexachlorocyclohexane (ε-HCH, solution in cyclohexane, 10 ng μl^{-1}) were supplied from Ehrenstorfer GmbH (Augsburg, Germany). Florisil® PR (60–100 mesh) and anhydrous sodium sulfate (Pestanal®) were obtained from Fluka (Buchs, Switzerland). Silica gel for column chromatography (230–400 mesh, 0.045–0.063 mm) and sulfuric acid (98%) were purchased from Merck KGaA (Darmstadt, Germany).

2.2. Spruce needle samples

Spruce needle samples from eight selected sampling sites distributed in the Alps were taken in five countries (Austria, Germany, Italy, Slovenia, and Switzerland) in autumn 2004 (see Table 1). All sampling sites were located in alpine remote forests of Norway spruce (>80% specific purity) being 30 or more years old and of at least 0.5 ha surface. 3–5 spruce needle branches were cut from the 7th whirl (from top) of two dominant adult trees in October 2004. Six months old twigs were collected, transported on dry ice and stored at −20 °C until further processing. After immersion into liquid nitrogen needles were separated from twigs, immediately transferred to precleaned brown glass containers and stored at −20 °C. Ancillary information about the spruce needle samples will be published by Offenthaler et al. (2009).

2.3. Needle extraction

In a 250 ml Duran glass bottle (Schott®), 20–25 g of fresh needles were spiked with the internal standard (10 ng of $^{13}C_{10}$-*trans*-chlordane) and extracted by shaking at room temperature (ca. 22 °C) with 150 ml of dichloromethane/*n*-hexane 1 + 1 (DCM/nHex, v/v) for 16 h. The extract was filtered through a glass funnel of 115 mm diameter filled with glass wool (Riedel-de Haën, Seelze, Germany) directly into a Turbo Vap vessel (Zymark, Hutchinson, USA) of 200 ml. A second portion of 30 ml DCM/nHex was added to the needles and the procedure repeated. The combined extracts were concentrated to 1 ml using a Turbo Vap 500 (Zymark, Hutchinson, USA).

2.4. Extract clean-up

A 20 mm i.d. glass column was filled from bottom to top with 1 g of anhydrous sodium sulfate, 20 g of silica gel impregnated with concentrated sulfuric acid (44%) and 1 g of anhydrous sodium sulfate. It was rinsed with 20 ml of DCM/nHex 1 + 1 (v/v). The sample extract was transferred to the column, and CPs were eluted with 70 ml of DCM/nHex 1 + 1 (v/v). The eluate was evaporated to 0.5 ml with a Turbo Vap 500. After dilution with 10 ml of nHex the extract was reduced to 0.5 ml and the latter procedure was repeated.

A further clean-up step was carried out on a chromatographic column containing 16 g of Florisil® deactivated with 1.5% water and conditioned with 20 ml of nHex. After passing the extract to the column, the first fraction obtained with 75 ml of nHex and 5 ml of DCM was discarded. CPs were collected in a second fraction of 60 ml of DCM, which was concentrated to 0.5 ml. 10 ml of cyclohexane were added, the volume was reduced to 100 μl with a Turbo Vap 500 and this step was repeated. Then, 10 ng of ε-HCH in 10 μl of cyclohexane were added as recovery standard.

2.5. Instrumentation

Quantification was performed on a gas chromatograph CP-3800 coupled to a 1200L triple quadrupole mass spectrometer (Varian, Walnut Creek, USA). The gas chromatograph was equipped with a fused silica capillary column (15 m length, 0.25 mm i.d.) coated with 0.25 μm of crosslinked 5% phenyl-methylpolysiloxane (DB5-MS, J&W Scientific, Folsom, USA). Helium (99.996%, Sauerstoffwerk Lenzburg, Lenzburg, Switzerland) at a constant flow of 2 ml min^{-1} was used as carrier gas. The temperature of the split/splitless injector was set to 275 °C. Splitless injections (3.0 min) of 2.5 μl were carried out with a Combi Pal autosampler (CTC Analytics, Zwingen, Switzerland). The following temperature program was used: 100 °C (3 min), then with 50 °C min^{-1} to 300 °C (isothermal for 3 min). The temperatures of