Chemosphere 215 (2019) 206–213

Contents lists available at ScienceDirect

Chemosphere

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

Identification of halogenated polycyclic aromatic hydrocarbons in biological samples from Alberta Oil-Sands Region

Zhe Xia [a], Ifeoluwa Idowu [a], Chris Marvin [b], Philippe J. Thomas [c], Wesley Johnson [a], Olga Francisco [a], Jorg Stetefeld [a], Bernard Crimmins [d, e], Mark Fry [f], Gregg T. Tomy [a, *]

[a] University of Manitoba, Centre for Oil and Gas Research and Development, Department of Chemistry, Winnipeg, Manitoba, Canada
[b] Environment and Climate Change Canada, National Water Research Institute, Burlington, ON, Canada
[c] Environment and Climate Change Canada, National Wildlife Research Centre, Ottawa, ON, Canada
[d] Clarkson University, Department of Civil and Environmental Engineering, Potsdam, NY, USA
[e] Academic Environmental/Analytical Consulting Services, LLC, Alliance, OH, USA
[f] University of Manitoba, Department of Biological Sciences, Winnipeg, Manitoba, Canada

HIGHLIGHTS

- GC-HR-TOF/MS method was used to identify halogenated compounds in biota.
- Three halogenated compounds were positively identified in biological samples.
- These compounds bioaccumulate in biota from the Alberta Oil Sands Region.

ARTICLE INFO

Article history:
Received 7 August 2018
Received in revised form
26 September 2018
Accepted 7 October 2018
Available online 8 October 2018

Handling Editor: Keith Maruya

Keywords:
Halogenated polycyclic aromatic hydrocarbons
Bioaccumulation
Polycyclic aromatic hydrocarbons
Alkylated polycyclic aromatic hydrocarbons
Biomagnification

ABSTRACT

Halogenated polycyclic aromatic hydrocarbons (HPAHs) were identified in biological samples from the Alberta Oil-Sands Region (AOSR) using gas chromatography coupled with high-resolution time-of-flight mass spectrometry (GC-HRTOF-MS) at a resolving power of 25,000. Knowledge of the electron ionization (EI) fragmentation behavior of individual HPAH isomers, achieved by injecting authentic standards in full-scan MS mode, was paramount in identifying a suite of HPAHs in samples from the AOSR. Confirmation of compounds in biological samples was based on the measured mass accuracy (±3 ppm) of 2 characteristic ions prominent in the EI mass spectra of each compound. Numerous compounds were detected in the high resolution total ion chromatogram in liver extracts of 4 biological species from the AOSR: river otter (*Lontra Canadensis*), northern pike (*Esox lucius*), lake whitefish (*Coregonus clupeaformis*) and snails (*Gastropod* sp.) many of which remain unidentified. Careful examination of the high-resolution accurate mass data suggests that dichloro-anthracene/phenanthrene, bromo-anthracene/phenanthrene and dibromo-fluorene were present in the biological samples. Lipid corrected concentrations of dichloro-PAHs were estimated to be 16.3 ± 11.4 ($n = 4$) and 5.5 ($n = 1$) ng/g in lake whitefish and river otter, respectively. Concentrations of mono-bromo-PAHs were an order of magnitude greater in snails (170.5 ng/g) than in northern pike (12.5 ng/g) while concentrations of dibromo-PAHs were 4 times greater in snails than in northern pike. The detection of these compounds in biota implies that these compounds are bioaccumulative. The liver-based biomagnification factor of the dichloro-PAH congener in the river otter/lake whitefish feeding relationship is much smaller than 1 implying that this compound does not biomagnify.

1. Introduction

Polycyclic aromatic compounds (PACs) are a complex class of compounds derived from incomplete combustion or diagenesis of plant matter forming petroleum oils. The most common PACs are the polycyclic aromatic hydrocarbons (PAHs) of which 16 have been

* Corresponding author.
E-mail address: Gregg.tomy@umanitoba.ca (G.T. Tomy).

https://doi.org/10.1016/j.chemosphere.2018.10.050
0045-6535/

identified as priority pollutants by the United States Environmental Protection Agency (US EPA) and are also listed under Schedule 1 of the Canadian Environmental Protection Act (Achten and Andersson, 2015). However, there are other important PACs that to date have received less attention. These include halogenated polycyclic aromatic hydrocarbons (HPAHs), non-halogenated alkylated PAHs and heterocyclic aromatic compounds that contain S-, O- and N- atoms (Achten and Andersson, 2015; Lee et al., 2015). Like some of the other PACs, HPAHs are likely to be persistent in the environment (Lee et al., 2015). In addition, the toxicity of some HPAHs have been found to be similar to dibenzo-p-dioxins and dibenzofurans (PCDD/Fs) (Ma et al., 2009; Ohura et al., 2007, 2009).

HPAHs have been detected and quantified in environmental matrices and abiotic samples, such as waste incinerators, electronic wastes, atmosphere, and sediment (Horii et al., 2008, 2009a; Jin et al., 2017; Ma et al., 2009; Ohura et al., 2009). HPAHs have also been detected in biological organisms, such as seafood from south China, fish from the Great Lakes and blue mussels (*Mytilus edulis*) from Massachusetts, USA (Horii et al., 2009a; Kannan et al., 2000; Ni and Guo, 2013). In general, however, there is a paucity of reports of HPAHs in biological organisms and is due, in part, to the lack of validated analytical methods for the identification and quantification of these compounds and a lack of analytical standards for HPAHs (Ieda et al., 2011).

The Canadian oil sands are naturally occurring mixtures of crude bitumen (thick, heavy crude oil), sand, clay, ultrafine mineral solids and water that are rich in PACs. PACs are released naturally from oil sands deposits (*i.e.,* due to erosion of the bitumen deposits) and from bitumen extraction and upgrading (*i.e.,* oil sands activity) (Kelly et al., 2009; Kurek et al., 2013; Thienpont et al., 2017). However, several studies have clearly shown that increases in the concentrations of C_1-C_4 alkylated PAHs and dibenzothiophenes (prominent components of Athabasca Oil Sands Region (AOSR) bitumen (Strausz and Lown, 2003) in the atmosphere, water, soil and sediments, plants, wildlife and fish in the AOSR are a result of proximity to oil sands activity (Boutin and Carpenter, 2018; Fernie et al., 2018; Kelly et al., 2009; Kurek et al., 2013; Lundin et al., 2015; Ohiozebau et al., 2016, 2017; Parrott et al., 2018; Thienpont et al., 2017). Historically, the AOSR was covered by sea water millions of years ago, which would have contained high concentrations of chloride and bromide ions (Musial et al., 2012; Ranger and Pemberton, 1998). During the oil sand formation, the elevated pressures and temperatures of diagenesis and catagenesis combined with the catalytic activation of halides formed with abundant crustal elements Al and Fe (AlX_3 and FeX_3, X = Cl or Br) likely created an environment conducive to organohalogen formation. HPAH formation has also been associated with the presence of halogen ions and strong sunlight (Sankoda et al., 2012). Based on these two potential formation pathways, it is hypothesized that HPAHs resulting from non-combustion sources are present in biological organisms from the AOSR. Having higher log K_{ow} than their parent compounds, HPAHs have a higher propensity to partition into lipids, and as a result, bioaccumulate and biomagnify in biological organisms (Strausz and Lown, 2003; Sun et al., 2013). This could in turn induce toxicity due to the activation of the AhR pathway (Horii et al., 2009b).

Our study hypothesis, therefore, is that HPAHs are present in biological samples from the AOSR. Before testing these hypotheses, it was first necessary to establish an analytical method for identification and quantification of these compounds. We chose to use gas chromatography coupled with high resolution time-of-flight mass spectrometry (GC/HRTOF-MS). The mass spectral behavior of 16 authentic standards under electron ionization conditions was first elucidated. Our GC/HRTOF-MS system operating at a resolving power (RP) of 25,000 acquired HR mass spectra and the exact masses of two prominent ions characteristic to each HPAHs were then used to identify HPAHs in biological samples from AOSR. The results from this study provide preliminary identification of a new class of compounds in the AOSR that to date have gone undetected.

2. Materials and methods

2.1. Chemicals

All high-purity (Optima grade) organic solvents, silica gel (923 grade, 100–200 mesh), alumina (60–325 mesh), Ottawa sand and anhydrous sodium sulfate, diatomaceous earth (DE) were purchased from Fisher Chemicals (Ottawa, Canada). The analytical standards and their acronyms are provided in Table SI1.

2.2. Sample preparation

All samples were collected in 2014 and 2015 from the sites shown in Fig. 1. Individual liver samples of otters ($n = 6$), northern pike ($n = 4$) and lake whitefish ($n = 4$) were extracted separately. Snails collected in the field were placed in clean water for 24 h to depurate the contents of their guts. Shells were then removed and tissues were pooled and homogenized. A sub-sample of this pooled material was then extracted ($n = 3$). Analyte extractions were performed using an accelerated solvent extractor (ASE, Thermo Scientific, Waltham, MA). Approximately 1.5 g of each sample (wet weight) was weighed and mixed with DE and transferred to an ASE extraction cell size and spiked with the recovery internal standard (RIS, 100 ng) prior to extraction. Details of the extraction and clean-up steps can be found in Idowu et al. (2018).

2.3. GC-HRTOF-MS analysis

Analysis of the HPAH extracts and standard solutions were performed in one-dimensional mode using a 7890A GC (Agilent Technologies, Wilmington, DE, USA) fitted with a split/splitless injector, coupled with a Pegasus HRT 4D (LECO, St Joseph, MI, USA) operated in electron ionization (EI) mode and calibrated with perfluorotributylamine, PFTBA, as the mass calibrant. The stationary phase was Rxi -5Sil MS (60 m × 0.25 mm × 0.25 μm) (Restek, Bellefonte, PA, USA). Standards and samples (2 μL) were injected at 250 °C in splitless mode. The GC program started at 80 °C (held for 1 min), heated to 210 °C at 35 °C/min, further to 260 °C at 3 °C/min and finally to 315 °C at 10 °C/min (held for 5 min). The MS transfer line temperature was at 300 °C, ion source temperature at 250 °C and He as carrier gas at a flow-rate of 1.4 mL/min. The HRTOF-MS was operated at a mass range of *m/z* 50–500 with an acquisition rate of 13 spectra/second at 70 eV. The RP (full width half height) of the system was typically greater than 25 000 based on the peak width of the lock-masses *m/z* 218.9851 (C_4F_9) and 263.9871 ($C_5F_{10}N$) of PFTBA. During the chromatographic run, PFTBA was continuously bled into the ion source to serve as an internal standard for instrument mass calibration. Run to run repeatability, measured as the mean mass accuracy (Δm) of all sample and standard injections were smaller than ±0.5 ppm for both lock-masses.

2.4. GC/MS/MS analysis

Instrumental details of the analysis and quantitation of PAHs and alkyl-PAHs (APAHs) are given in Idowu et al. (2018).