PERSISTENT ORGANIC POLLUTANTS IN AMBIENT AIR, HUMAN BREAST MILK AND EDIBLE FISH – A POTENTIAL FOR HEALTH RISK IN GHANA

Adu-Kumi S[1]*, Asante KA[2], Klanova J[3]

[1]Organization A, 24 Research Drive, Sydney, Australia; [2]Organization B, 300 University Road, Tokyo, Japan; [3]Organization C, 505 Laboratory Street, Liège, Belgium

[1]Environmental Protection Agency, P. O. Box MB 326, Accra, Ghana/Graduate School of Nuclear and Allied Sciences, University of Ghana, Legon, Accra; [2] Center for Marine Environmental Studies (CMES), EHIME, Japan; [3]Research Centre for Toxic Compounds in the Environment (RECETOX), Faculty of Science, Masaryk University, Kamenice Brno, Czech.

Introduction:

Organochlorine pesticides (OCPs), polychlorinated biphenyls (PCBs), polychlorinated dibenzo-p-dioxins and polychlorinated dibenzo furans (PCDD/Fs), and brominated flame retardants (BFRs) constitute major persistent organic pollutants (POPs) of global and national concern. In Ghana intentional use of POPs involve several sectors including agriculture; manufacturing industry (chemical, electronic, electrical, food, etc.); household, livestock production; mining; and illegal activities (e.g. fishing). Uncontrolled combustion processes; waste incineration; power generation and heating; transport; and mining constitute the main sources of nintentional releases of POPs[1]. Very little scientific investigations however exist on exposure levels and human health implications of POPs in Ghana. This paper discusses a number of contaminants found to be of human health concern to the general population. The overall objective of the various studies carried out were to (1) estimate levels of POPs in environmental media (air, water), biota (fish) and human breast milk; and (2) assess the associated health risks for the general population of Ghana (including infants).

Materials and methods:

Two urban/suburban background air sampling sites (East Legon & Kwabenya, suburbs of Accra) and one rural background air sampling site (Lake Bosumtwi, located in the western central part of Ghana) were investigated using polyurethane foam (PUF) disk passive air samplers (PAS) in 2008[2]. All PUF disks were prewashed, cleaned (8 hours extraction in acetone and 8 hours in dichloromethane), wrapped in two layers of aluminium foil, placed into zip-lock polyethylene bags and kept in the freezer prior to deployment. Exposed disks were wrapped in two layers of aluminium foil, labelled, placed into zip-lock polyethylene bags and transported in a cooler to the laboratory. Field blanks were obtained by installing and removing the PUF disks at the sampling site. The sampling campaign lasted from January to December, 2008, and two different sampling schemes were applied: sampling period of 4 weeks (to stay away from equilibrium) for determination of volatile pesticides as HCHs, HCB or PeCB, and 12 weeks for remaining compounds of interest. All air samples were extracted with dichloromethane in a Büchi System B-811 automatic extractor together with laboratory and field blanks. Surrogate recovery standards (PCB 30 and PCB 185, 50 µL at concentrations of 50 ng mL^{-1}) were spiked on each PUF disk prior to extraction. PUF extracts for analysis of HCHs, HCB and PeCB (shorter deployment time) were cleaned-up using a sulphuric acid modified silica gel column. PUFs for analysis of less stabile pesticides as dieldrin or endosulfan (longer deployment time) were cleaned-up using an activated Florisil column (60-100 mesh, Supelco). Samples were reduced under the gentle nitrogen stream to 1ml, transferred to vials, and spiked with 50 µL of 10 ng mL^{-1} of PCB 121 as an internal standard. OCPs were analyzed in 12-week samples using GC-MS (Quattro Micro GC, Micromass) in GC-EI-MSMS mode. Chromatographic separation was achieved on a DB-5MS fused-silica capillary column (J&W Scientific, 60 m, 0.25 mm i.d., 0.25 µm film thickness) with helium as a carrier gas (flow rate 30 cm s^{-1}). The column temperature was programmed as follows: initial temperature 80°C for 1 min, 20°C min^{-1} to 200°C, 1.5°C min^{-1} to 260°C, 15°C min^{-1} to 300°C, final hold for 10 min). Samples (1 µl) were injected in the splitless mode. Injector, transfer line, and ion source were kept at 250°C, 275°C, 200°C, respectively. Quantification was performed using the seven-point calibration curve. All 4-week samples were analyzed for HCHs, DDTs, HCB and PeCB using GC-MS (Agilent 6890 GC - Agilent 5975 MS) in the selected ion monitoring mode (EI-SIM). Recoveries were determined for all samples by spiking with the surrogate standards prior to extraction. Amounts were similar to detected quantities of analytes in the samples. Recoveries were higher than 76 % for all samples, and recovery factors were not applied

to any of data. Recovery of native analytes measured for the reference material varied from 75 to 98 % for all OCPs. Laboratory blanks were under the detection limits for selected compounds. Field blanks consisted of pre-extracted PUF disks and they were taken on each sampling site. They were extracted and analyzed in the same way as the samples, and the levels in field blanks never exceeded 1% of quantities detected in samples for all OCPs, indicating minimal contamination during the transport, storage and analysis.

Forty-two samples were collected in 2009 from three geographical locations in Ghana; Accra (coastal) (n = 16); Kumasi (forest zone) (n = 14) and Tamale (savannah) (n = 12). In each city, samples were taken from both urban and rural areas. Prior to this, 25 samples collected from Accra in 2004 were analyzed for assessing temporal variation[3]. All donors were from the general population who were non-smokers and appeared healthy. Breast milk was expressed by the donors themselves or with the help of midwives into solvent- precleaned glass containers with Teflon-lined screw caps prepared for every individual. The samples were kept frozen and airlifted to the Center for Marine Environmental Studies (CMES), Ehime University, Japan on dry ice and stored in the Environmental Specimen Bank (*es*-BANK) of Ehime University at -25 °C (Tanabe, 2006) until extraction and chemical analyses. The samples were used for analyzing PCBs, PBDEs and HBCDs. Approximately 50g of the human milk sample was lyophilized and extracted with a high speed solvent extractor (SE-100, Mitsubishi Chemical Analytech) using 50% acetone in hexane. Fat content was determined gravimetrically from an aliquot of the extract. The remaining extract was spiked with $^{13}C_{12}$-labeled PCBs, $^{13}C_{12}$-labeled PBDEs (5 ng each) and 10 ng of $^{13}C_{12}$-labeled HBCDs as surrogates and then subjected to gel permeation chromatography (GPC) for fat removal and eluted with a mixture of hexane/dichloromethane (1:1). The lipid-removed GPC fraction containing organohalogen compounds was concentrated and passed through 4 g of activated silica gel packed in a glass column. The first fraction containing PBDEs and PCBs was eluted with 5% dichloromethane in hexane and the second fraction containing HBCDs with 25% dichloromethane in hexane. $^{13}C_{12}$-labeled BDE-126, -139 and -205 were added to the PCBs/PBDEs fraction and deuterated HBCDs added to the HBCDs fraction as internal standards. Quantification of PBDEs and PCBs was performed using a gas chromatograph coupled with a mass spectrometer (GC-MS). All the congeners were quantified using the isotope dilution method to the corresponding $^{13}C_{12}$-labeled congeners. Forty-two BDE congeners from mono- to deca-BDE and sixty-two PCB congeners were analyzed in this study based on the methods published elsewhere (Malarvannan et al., 2009; Tue et al., 2010) with slight modification. HBCD isomers (α-, β-, γ-HBCD) were quantified using a liquid chromatograph coupled with a tandem mass spectrometer (LC-MS/MS) (Isobe et al., 2007). Concentrations of all the targeted BDE and PCB congeners, and HBCD isomers were summed to obtain the values of ∑PBDEs, ∑PCBs and ∑HBCDs, respectively. Concentrations of PBDEs, PCBs and HBCDs were expressed as nanogram per gram lipid weight (ng/g lw). For quality assurance/quality control (QA/QC), procedural blanks were analyzed simultaneously with every batch of 7 samples to check for any interference or contamination from solvents and glass wares during sample processing. Detection limits for the target compounds were calculated as three times the procedural blank and the mean value was 0.01 ng/g lw for each compound. Recoveries of ^{13}C-labelled surrogates were in the range of 65-110% for PCBs, 78-110% for PBDEs and 103-112% for HBCDs.

Two species of fish samples (redbelly tilapia and catfish) were randomly collected from three locations in Ghana, namely, Lake Volta, Lake Bosumtwi and the Weija Lake in September 2008[4]. Catfish from Lake Volta was however purchased from a local market at Madina, a suburb of Accra, the capital of Ghana. Fish samples were wrapped in aluminium foil, placed in polyethylene bags, stored under ice and transported to Japan where they were stored frozen at -20 °C until they were subjected to chemical analyses. Approximately 40 g of edible portions of all fish samples (n = 13) were separately homogenized with anhydrous sodium sulphate, and subsequently extracted in Soxhlet apparatus with dichloromethane. Each extract was then concentrated and an aliquot of the extract dissolved in hexane. Prior to clean-up, each extract was spiked with known amounts of ^{13}C-labeled surrogates (Wellington Laboratories, Guelph, ON, Canada) for all 2,3,7,8-substituted PCDD/Fs congeners and all dl-PCBs as internal standards for the validation of the clean-up procedure. Each of the spiked solutions was then treated with concentrated sulphuric acid (H_2SO_4). The H_2SO_4 clean-up step was repeated until the H_2SO_4 fraction remained clear. The hexane layer was separated and washed with water. Each solution was purified (eluted) on multi-layer columns, containing sodium sulphate (2 g), 10% (w/w) silver nitrate-silica (2 g), silica gel (2 g), 22% (w/w) potassium hydroxide-silica gel (0.5 g), with hexane (100ml). Each eluate was concentrated and separated into two fractions. The first fraction was eluted with 10% dichloromethane in hexane whilst the second fraction was eluted with toluene on an activated carbon/silica gel column. The first fraction

contained HCB, HCHs (α-, β-, γ- and δ-isomers), DDTs (*o,p′*- and *p,p′*-DDD, *o,p′*- and *p,p′*-DDE, *o,p′*- and *p,p′*-DDT), CHLs (*cis*- and *trans*-chlordane, *cis*- and *trans*-nonachlor), oxychlordane, heptachlor, octachlorostyrene and mono-*ortho* PCBs, whilst the second fraction contained non-*ortho* PCBs, and PCDD/F congeners.

Results and discussion:

The levels of OCPs in ambient air ranged for the individual pesticides from below limit of quantification (LOQ) to 750 pg m^{-3} (for α-endosulfan) and current agricultural application seemed to be the main primary source of most abundant pesticides. Re-volatilization of previously used pesticides from contaminated soils could not be ruled out either as potential secondary source of contamination, especially in warm and dry seasons and periods of intensive agricultural activities. Higher atmospheric concentrations were observed in November and December during the dry season compared to lower concentrations observed in June, July and August when the country experiences heavy rains. The highest seasonal variation was observed for currently used pesticides as α-endosulfan. A *p,p′*-DDT/*p,p′*-DDE ratio suggested recent inputs of fresh technical DDT.

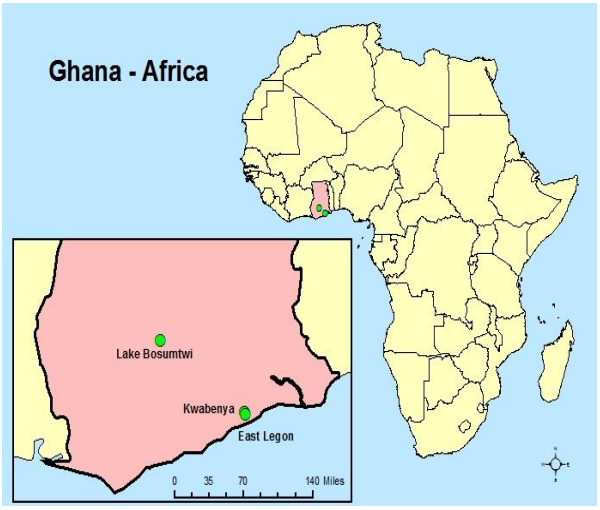

Figure 1: Map of Ghana showing 3 passive air sampling sites

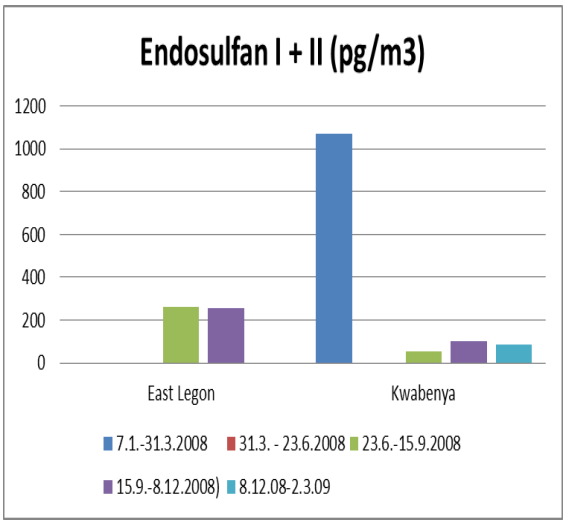

Figure 2: Ave. seasonal concentrations (pg m^{-3}) of endosulfans at two sampling sites in Ghana

Mean levels and ranges of PBDEs (4.5; 0.86-18 ng/g lw) and PCBs (62; 15-160 ng/g lw) observed in the present study were unexpectedly high, in spite of the fact that Ghana is a non-industrialized country when compared with many of the Asian and European countries. Significant increases were found in the concentrations of PCBs and PBDEs over the years, while no significant increase was observed for HBCDs. Estimated hazard quotient (HQ) showed that all the mothers had HQ values exceeding the threshold of 1 for PCBs and OCPs, indicating potential health risk for their children. PCBs in dirty oils and obsolete equipment should be of concern as potential sources in Ghana, and e-waste recycling with little or no experience in safe handling could be a threat to this sub-region noted for unregulated disposal and incessant burning of e-waste. The results may point towards an increase in trends in human milk in Ghana, especially in the larger cities but further analysis would be required to confirm this upward trend in levels. This is the first study to report BFRs in human breast milk from Africa, and undoubtedly from Ghana.

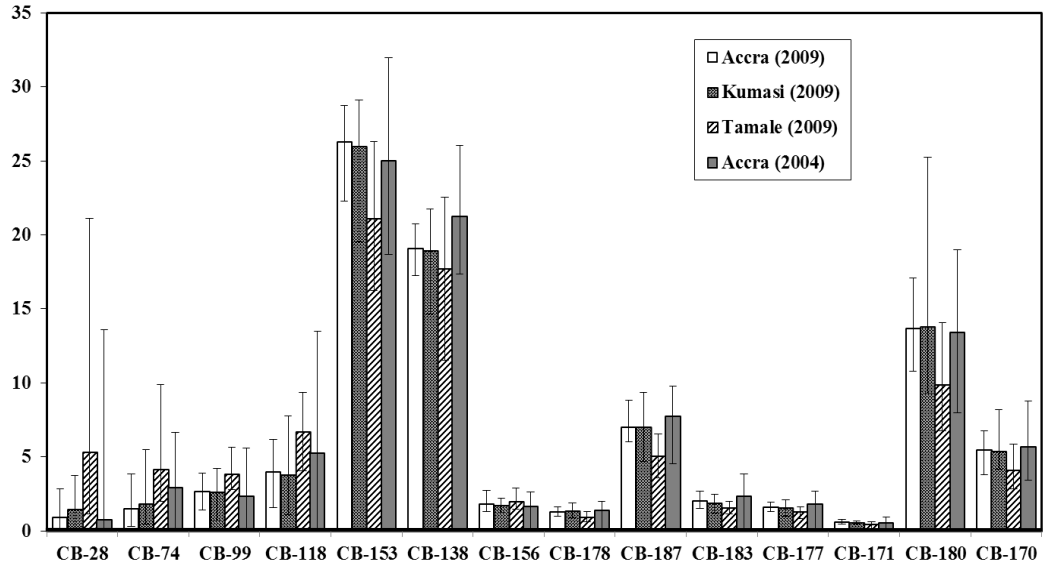

Figure 3: PCB congener profiles (%) in human breast milk from various locations in Ghana (vertical bars show the range and mean of each congener to total PCB concentration).

The levels of organochlorine pesticides measured in this study were generally low. The highest concentration of OCPs was measured for dichlorodiphenyltrichloroethane compounds (DDTs) (*p,p′*-DDT, *o,p′*-DDT, *p,p′*-DDE, o,*p′*-DDE, *p,p′*-DDD and *o,p′*-DDD), followed by chlordane compounds (CHLs) (*trans*-chlordane, *cis*-chlordane, *trans*-nonachlor and *cis*-nonachlor), hexachlorobenzene (HCB) and gamma-hexachlorocyclohexane (γ-HCH). The relatively high ratio of *p,p′*-DDT/*p,p′*-DDE in tilapia and catfish with an extremely high value in catfish purchased from a local market at Madina, a suburb of Accra, however, suggests the fresh contamination of technical DDT in Ghana. Although PCDD/Fs and dl-PCBs showed relatively low levels, the concentrations are, however, comparable with recent data of some developed countries. There is a potential health risk from DDTs, PCDD/Fs and dl-PCBs for the general population of Ghana because fish is one of their important protein sources. It is therefore necessary to estimate the total intake of DDTs, PCDD/Fs and dl-PCBs, and to assess the health risks for the general population of Ghana.

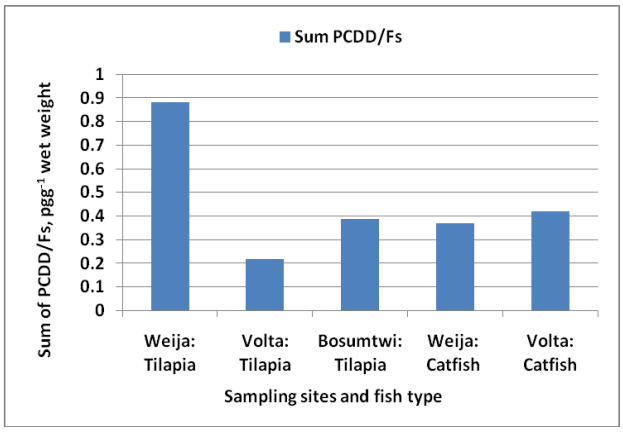

Figure 4: Ave. concentrations of PCDD/Fs in tilapia and catfish according to sampling sites

References:

1. Ghana National Implementation Plan (2007): <http://www.pops.int/>.
2. Adu-Kumi S. *et al* (2012) *Env. Sci. & Poll. Res* (in print)
3. Asante K. A. *et al* (2011) *Environ Int* 37**:** 921-928
4 Adu-Kumi S *et al* (2010) *Chemosphere* 81: 675-684