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Transplacental transfer of polycyclic aromatic hydrocarbons in paired samples of maternal serum, umbilical cord serum, and placenta in Shanghai, China☆

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ABSTRACT

Prenatal exposure to polycyclic aromatic hydrocarbons (PAHs) is a high-priority public health concern. However, maternal to fetal transplacental transfer of PAHs has not been systematically studied. To investigate the transplacental transfer of PAHs from mother to fetus and determine the influence of lipophilicity (octanol-water partition coefficient, K_{OW}) on transfer process, in the present study, we measured the concentrations of 15 PAHs in 95 paired maternal and umbilical cord serum, and placenta samples (in total 285 samples) collected in Shanghai, China. The average concentration of total PAHs was the highest in maternal serums (1290 ng g^{-1} lipid), followed by umbilical cord serums (1150 ng g^{-1} lipid). The value was the lowest in placenta samples (673 ng g^{-1} lipid). Low molecular weight PAHs were the predominant compounds in the three matrices. Increases in fish and meat consumption did not lead to increases in maternal PAH levels, and no obvious gender differences in umbilical cord serums were observed. The widespread presence of PAHs in umbilical cord serums indicated the occurrence of transplacental transfer. The ratios of PAH concentrations in umbilical cord serum to those in maternal serum (F/M) and the concentrations in placenta to those in maternal serum (P/M) of paired samples were analyzed to characterize the transfer process of individual PAHs. Most F/M ratios on lipid basis were close to one (range: 0.79 to 1.36), which suggested that passive diffusion may control the transplacental transfer of PAHs from maternal serum to the fetal circulation. The P/M and F/M values calculated on lipid basis showed that PAHs with lower K_{OW} were more likely to transfer from mother to fetus via the placenta.

1. Introduction

Polycyclic aromatic hydrocarbons (PAHs), which are ubiquitous contaminants in the environment, cause great public health concern due to their carcinogenicity, teratogenicity, and genotoxicity (IARC, 2010; Kim et al., 2013). The main exposure routes of PAHs in the general population are through air inhalation, cigarette smoke inhalation, and food ingestion (Ding et al., 2012). Due to rapid urbanization, extensive motorway networks, and the popularity of car transport in China, an increase in the emission and environmental concentrations of PAHs is predicted (Wang et al., 2016). High concentrations of PAHs in road dust (14–21 mg kg^{-1}) and surface sediments (107–1710 ng g^{-1} dry) were detected in Shanghai (Liu et al., 2007, 2008). This may lead to high intake in the general population in this region.

Gestation is a sensitive period. Environmental contaminants can be transferred via the placenta and exert adverse effects on fetus (Barr et al., 2007; Bocskay et al., 2005; Pedersen et al., 2010). For example, placental benzo(a)pyrene (BaP) may disturb the differentiation of placental trophoblastic cells (Rappolee et al., 2010). A positive correlation between elevated concentrations of PAHs and an increased risk of neural tube defects has been observed (Langlosis et al., 2012; Ren et al., 2011). In addition, adverse pregnancy outcomes have been associated with exposure to PAHs (Al-Saleh et al., 2013; Edwards et al., 2010; Pedersen et al., 2013; Yi

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et al., 2015). Furthermore, placental exposure to PAHs can result in long-term adverse effects in children, including developmental delays and behavioral problems (Perera et al., 2009, 2012). It is well known that fetuses and infants are more susceptible to the adverse effects of PAHs than adults. As a result, prenatal exposure to PAHs is causing a lot of public health concern.

The measurements of contaminants in umbilical cord serums and placentas are a useful means of assessing prenatal exposure and potential toxic effects on fetuses (Sakamoto et al., 2016; Yu et al., 2013). Many organic pollutants, such as organochlorine pesticides, polychlorinated biphenyls (PCBs), polybrominated diphenyl ethers (PBDEs), perfluorooctanoate, perfluorooctanesulfonate, synthetic musks, and nanoparticles have been detected in umbilical cord serums and placentas, and their transplacental transfer has been investigated (Mori et al., 2014; Mose et al., 2007; Poulsen et al., 2015; Yu et al., 2013). Some researchers have reported the transplacental transfer of PAHs (Karttunen et al., 2010; Sexton et al., 2011; Tsang et al., 2011). However, it is difficult to compare the data, because different units and detection methods were used. For example, Tsang et al. (2011) found that the total PAH concentrations were lower in umbilical cord serums (1160 ng g^{-1} lipid) than those in maternal bloods (1460 ng g^{-1} lipid). However, Sexton et al. (2011) proposed that the PAH concentrations in umbilical cord serums generally exceeded those in paired maternal bloods, and the data were given at the ng mL^{-1}. The concentrations of hydroxylated PAHs in urine and PAH-DNA adducts in cord serums and cord tissues were also reported (Perera et al., 2012; Thai et al., 2015; Yi et al., 2015). In addition, maternal age, dietary intake, lifestyle, and residence region can have a variable influence on particular contaminants and matrices (Jakobsson et al., 2012; Lee et al., 2013; Luo et al., 2016; Pedersen et al., 2013). The extent of transplacental transfer from the mother to the fetus depends on the physical characteristics of the maternal-placental-embryonic-fetal group, and the physiochemical and structural characteristics of the contaminants (Mori et al., 2014; Needham et al., 2011; Vizcaino et al., 2014). Thus, a systematic study is needed to determine the placental transfer of PAHs.

Therefore, to comprehensively study the partition of PAHs from the mother to the fetus, 95 paired samples of maternal serum, umbilical cord serum, and placenta (in total 285 individual samples) from a mother-infant cohort in Shanghai were collected and analyzed. The main objectives of the present study were: (1) to investigate the occurrence of PAHs in maternal and umbilical cord serums, and placentas; (2) to evaluate the transplacental transfer of PAHs from mother to fetus, and investigate the possible mechanism involved.

2. Materials and methods

2.1. Sample collection

Volunteers were randomly enrolled at a hospital located in Shanghai during 2013 and 2014. Data including age, occupation, education, parity, dietary habit, and neonatal parameters were recorded by using questionnaires. Women with possibly occupational exposure to PAHs were excluded. Although samples and data were collected blindly, selection bias may not completely be excluded. Paired samples (pair number: n = 95) of maternal serum, umbilical cord serum, and placenta were collected at the time of delivery from 95 donors. Serums were obtained from blood samples by centrifugation and preserved in brown glass test tubes at −20 °C until analysis. Placenta samples were lyophilized and stored in sealed plastic bags at −20 °C. The Ethics Committee of the hospital approved the study protocol. Informed consents were obtained from all the volunteers.

2.2. Chemicals

The target PAH compounds included acenaphthylene (ACY), acenaphthene (ACE), fluorene (FLO), phenanthrene (PHE), anthracene (ANT), fluoranthene (FLA), pyrene (PYR), benz(a)anthracene (BaA), chrysene (CHR), benzo(b)fluoranthene (BbF), benzo(k)fluoranthene (BkF), indeno(l,2,3-cd)pyrene (IcdP), BaP, dibenz(a,h) anthracene (DahA), and benzo(g,h,i)perylene (BghiP). The standards of the 15 PAHs were purchased from AccuStandard Incorporated (Connecticut, USA). Mixed surrogate standards including ACY-d_8, PHE-d_{10}, FLA-d_{10}, PYR-d_{10}, BaA-d_{12}, and BghiP-d_{12} were obtained from Cambridge Isotope Laboratories, Incorporated (Andover, USA). Hexane and dichloromethane were of analytical grade and were re-distilled before use.

2.3. Sample treatment

Frozen serum samples (4–5 mL for maternal serum and 8–12 mL for umbilical cord serum) were warmed to room temperature and spiked with 2.5 ng of surrogate standards. The serum samples were treated as previously described with some modifications (Zhang et al., 2015). Briefly, hydrochloric acid and isopropanol were added before liquid-liquid extraction. Then, 30 mL of *n*-hexane was used for the extraction (3 × 10 mL). For placenta samples, 3–4 g of lyophilized sample was spiked with 2.5 ng of surrogate standards and then was Soxhlet-extracted with 200 mL of dichloromethane for 72 h. All the extracts were concentrated and their lipid contents were determined by using a gravimetric method. After the lipids were weighed and recorded, they were redissolved in *n*-hexane, and then cleaned up by passing them through a glass column (300 mm × 25 mm i.d.) packed with Biobeads S-X3 (Bio-Rad Laboratories, Hercules, CA, USA). A silica/alumina column (2:1) was used for further purification. The sequential eluents were *n*-hexane and *n*-hexane/dichloromethane (7:3, v/v). Finally, the fractions containing PAHs were collected and concentrated to 50 μL. Hexamethylbenzene was then added as the injection internal standard.

2.4. Instrument analysis

PAHs were analyzed using an Agilent 6890N gas chromatograph equipped with a 5975 mass selective detector. A DB-5MS (30 m × 0.25 mm × 0.25 μm, J & W Scientific, USA) fused-silica capillary column was used. The oven temperature was changed as follow: 80 °C–180 °C at a rate of 3 °C min^{-1}, 180 °C–240 °C (held for 1 min) at 5 °C min^{-1}, and 240 °C–290 °C (held for 2 min) at 3 °C min^{-1}. A splitless injection mode at 280 °C was used. The mass spectrometer was operated in electron impact mode with selected ion monitoring. The compounds were quantified by two diagnostic ions and the retention time as described previously (Zhang et al., 2015).

2.5. Quality assurance and quality control

A procedural blank was included in each batch of samples, and the values of the procedural blanks were subtracted from the sample concentrations. Surrogate standards were used with average recoveries of 77.3 ± 25.9% for ACY-d_8, 113 ± 19.7% for PHE-d_{10}, 108 ± 25.0% for FLA-d_{10}, 107 ± 25.9% for PYR-d_{10}, 90.7 ± 22.5% for BaA-d_{12}, and 78.5 ± 26.0% for BghiP-d_{12}. The limits of detection (LODs) were derived from six blanks spiked with 1 ng of standards, and defined as the products of 3.36 and the standard deviations of the results. The LODs were between 2 and 32 ng g^{-1} lipid. The limits