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Original Article

Assessment of polychlorinated biphenyls (PCBs) in maternal blood serum from selected districts of Punjab, Pakistan

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Authors' Contribution
AN, AQ, AM: conceived and planned the present research work. **GZ:** provided the facilities for samples analysis and supervised the analytical process.

Key words
Dioxin like PCBs
Maternal blood serum
Congener specific analysis,
Spatial distribution

Abstract
The current study was conducted to screen the PCB residues in blood serum of mothers residing in five districts of Punjab Province, Pakistan. The mean concentration of $\sum_{34}$ PCB congeners was recorded 232.3 ng/g l.w. PCB homologs profile showed highest levels of Tetra-CBs (53%) followed by Penta-CBs (23%) and Hexa-CBs(11%), respectively. Spatial distribution of PCB homolog reflected the higher prevalence of lower chlorinated PCBs in urban population descending towards rural population. The mean TEQ concentration of eight dioxins like PCBs was recorded as 0.03 ng/g l.w. Analysis of $\sum_{34}$PCBs and socio-demographic parameters reflected significant results for milk intake. The study findings suggested a need for regulating the continuous intentional or unintentional releases of PCB into the environment to prevent human exposure.

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INTRODUCTION

Polychlorinated Biphenyls (PCBs) are the toxic, bio-accumulative substances that can persist in the environment for a long time and lipophilic in nature having more tendency of bioaccumulation and bio-magnification in organisms (Howard and Muir, 2010; Antonio *et al.,* 2008; Fiedler *et al.,* 2002). PCBs have toxicological implications, deleterious impacts on the environment, wildlife and human and have attained the attention of global and regional scientific community during the last decades (Mahmood *et al.,* 2014; Breivik *et al.,* 2004; Cui and Forssberg 2003; Zhang *et al.,* 2005). PCBs are consistent and stable in the environment; therefore, the United States Environmental Protection Agency has declared them as probable human carcinogens (Judd *et al.,* 2003). The use of PCBs has been started on the large scale since the 1930s for commercial purposes such as plastic, lubricating oils, inks, carbonless copy paper, impregnating, paints, sealing liquids, adhesives, additives, waxes, immersion oils, fire retardants, plaster and casting in different industries (Wittsiepe *et al.,* 2015; Afghan and Chau 1989). There are several other sources of PCBs pollution from waste material such as incineration of municipal waste, volatilization from the contaminated water bodies and landfill sites having transformers and

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coolants as waste components (Mahmood *et al.,* 2014; Wittsiepe *et al.,* 2015; Mumtaz *et al.,* 2016). Up till now, 209 congeners of PCB has been identified in the environment resulted from different anthropogenic activities (WHO, 2003).

Despite from a worldwide ban on PCBs production, some countries, particularly developing countries from Asia, Africa, Europe and Latin America continued their use for industrial and commercial purposes (Mamun *et al.,* 2007). Therefore, PCBs are still detected in the environment and pose great threats to the health of the environment and human (Hernik *et al.,* 2016; Thomas *et al.,* 2006). The human population is generally exposed to PCBs through the air, water, and food (WHO 2003; Jensen *et al.,* 1987; Sharma *et al.,* 2014). Dietary intake of contaminated fish, meat, and dairy products is considered the primary exposure route of PCBs to human which pose Neuro-developmental risks (Eguchi *et al.,* 2012; Ginsberg *et al.,* 2015; Hernik *et al.,* 2016; Sharma *et al.,* 2014). Certain biomarkers have been recommended by the World Health Organization (WHO) for the assessment of pollution caused by PCBs (Osman *et al.,*, 2017). A positive association between PCBs levels in human serum and consumption of meat and dairy fat has also been reported in previous studies (Govart *et al.,*2010). The predominant target organs/tissues in human with possible higher concentrations of PCBs are liver, brain, adipose tissues and skin, blood, milk, placenta and cord blood (WHO 2003; Esteban and Castano 2009; Dirtu *et al.,* 2009). PCBs can cause adverse health impacts to human health e.g. immunotoxicity, developmental, physiological and structural disorders and even it also causes reproductive and genetic disorders such as low birth weight, vision and hearing disorder etc. (Kodavanti *et al.,* 2008; Tyagi *et al.,* 2015; Darnerud, 2003; Larsen, 2006).

Limited information regarding human exposure to POPs is available in developing Asian countries (Tanabe and Kunisue, 2007). Screening of PCBs levels in the human body is the least addressed research area in Pakistan and a few studies on PCBs assessment in male human serum (Ali *et al.,* 2014) and in maternal blood serum (Ali *et al.,* 2013) are available from Pakistan. The negative effects of rapid urbanization and industrialization have been observed in environmental matrices of Punjab Pakistan (Azmat *et al.,* 2016). Women are more vulnerable to PCB exposure as they have a relatively higher proportion of fat and PCBs being lipophilic tend to accumulate in their fatty tissues. Consumption of these contaminants not only causes adverse health effects to exposed women, but pre and post natal exposure causes developmental, reproductive, behavioral disorders and neurological effects in the fetus (Daglioglu *et al.,* 2010; De Rosa and Hicks 2001; Rice, 2012). The present study was designed to focus on the evaluation of PCBs in maternal blood serum from different environmental settings of Punjab Province, Pakistan.

MATERIALS AND METHODS

Study area and sampling strategy

This study was conducted in urban and rural areas of Lahore, Sialkot, Chakwal, Okara and Khanewal district of Punjab province, Pakistan. Punjab is the most populous province with an area of 205,344 km^2 and the population of 100 million (population density = 353 persons/ km^2). A higher proportion of the population of the province is living in rural areas and involved in agricultural activities, whereas, an urban population of the province is normally engaged in industrial and other service activities. These areas were selected by considering the fact that both the rural and urban environments are exposed to PCBs pollution. A map of study area representing the location of selected sampling sites is given in Figure1. The female volunteers with age between18 years to 45 years were selected from each sampling site. The maternal blood samples (n=44) were collected from women with the help of paramedical staff by visiting the hospitals (Thomas *et al.,* 2006). After collection, the serum was separated by centrifugation and placed in the refrigerator (-20ºC) until final analysis (Inoue *et al.,* 2006; Covaci and Schepens, 2001).

Sample preparation

The blood serum samples were thawed and homogenized at room temperature. One milliliter (1 ml) of maternal blood serum was taken, spiked with the surrogate standards (5ppb) of PCB 209 and 2,4,5,6-tetrachloro-m-xylene TcmX and kept overnight at 4^0C (Covaci and Schepens 2001; Covaci and Voorspoels 2005). Extraction of PCBs was done by adding n-hexane (6 ml) and acetone (3 ml) into serum samples followed by homogenization through ultrasonic treatment for one hour at 3ºC. The clear supernatant was separated into glass tubes by centrifugation of samples at 2000 revolution per minute (RPM) (Dewan *et al.,* 2013). The same procedure was repeated twice and the supernatant was collected (Tyagi *et al.,* 2014). The empty Agilent Bond Elut polypropylene cartridge was pre-washed with DCM (5 mL), activated through MeOH (5mL), water (5 mL) and finally dried by centrifugation for 15 minutes (Covaci and Schepens, 2001; Covaci and Schepens 2001). A column for sample cleanup was prepared by the method (Jaraczewska *et al.,* 2006). The sample was loaded on the cartridge and analytes were eluted with hexane (4 mL) and Dichloromethane (2 mL; Jaraczewska *et al.,* 2006). Samples were concentrated through nitrogen streaming (Inoue *et al.,* 2006) and equilibrated with fifty microliters (50 µl) of iso-octane (Covaci and Schepens 2001). Internal standards of ^{13}C-PCB 141 (10µL) were added to the mixture followed by vortexing and transferred to the septa vials (1.5 mL). The samples were transferred to the State Key Laboratory of Organic Geochemistry, Guangzhou Institute of Geochemistry, Chinese Academy of Sciences, Guangzhou, China for further analysis on GC/MS (Gas Chromatograph Mass Spectrometer).

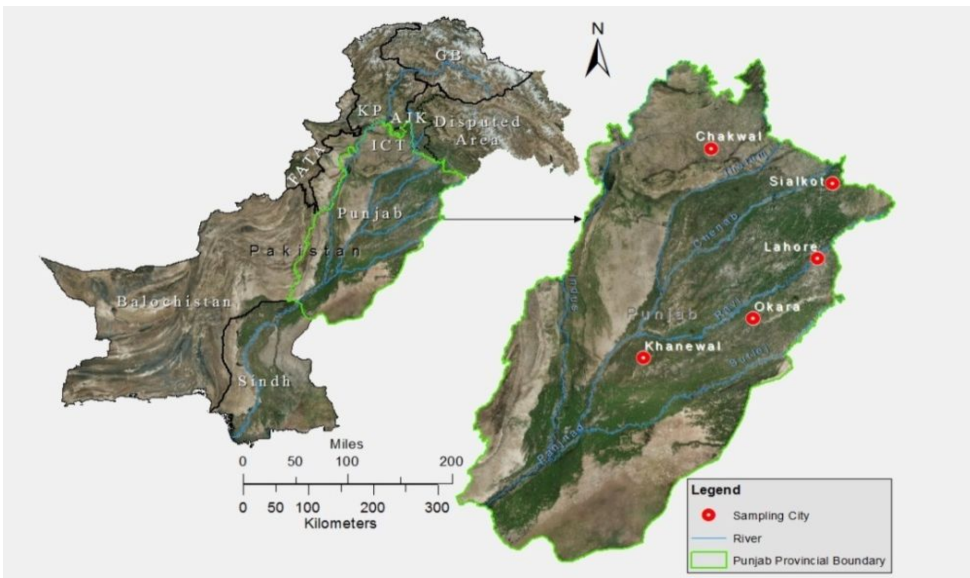

Figure 1: Map of study area of the showing different sampling districts in Punjab, Pakistan

Instrumental Analysis

An Agilent (7890A) gas chromatograph (GC) coupled with an Agilent (7000A) triple quad mass spectrometer (MS) detector and Agilent 7693 auto-sampler was used for the quantification of PCBs. The operational mode of GCMS was electron-capture negative ionization (ECNI). For PCBs analysis, a CP- Sil 8 capillary column from Netherland was used. The helium as a carrier gas was applied with a fixed flow rate of 1.2ml/min with an initial pressure of 20.9 psi. The operational mode of mass spectrometer was electron ionization (EI) with Electron Multiplier Voltage (EVM) of 2947 V and 230°C was set at the ion source temperature. Employing the pulsed splitless mode, one microliter (1µl) of the final extract was injected with the initial temperature at 100°C, pulse pressure of 25 psi kept for 0.5min. The length of total rum per sample was 40 ms and 1.5 min was total splitless time. Concentrations of 34 PCB congeners including eight (8) dioxin like PCBs (PCB70, PCB126, PCB169, PCB105, PCB114, PCB118, PCB156 and PCB189) and six indicator PCBs (PCB52, PCB101, PCB118, PCB138, PCB153 and PCB180) were determined. The total lipid in the serum samples was determined as demonstrated by Covaci *et al.,* 2005). Total cholesterol (CHOL) and triglycerides (TG) were measured enzymatically in separate subsets of the serum samples. The total lipids were determined by putting the CHOL and TG values in the formula as follows: TL (g/l) = 1.12×CHOL+ 1.33×TG + 1.48 (Covaci *et al.,* 2006). Final concentrations of PCBs in each sample were expressed in ng/g lipid weight.

Demographic Status

In order to collect the demographic information of the sampling group, a questionnaire was developed and filled by getting information from the each volunteer woman during the blood sampling regarding the age, location, parity, body mass index (BMI), eating habits, fish and milk intake and infant's birth weight.

Quality control and assurance

The surrogate standards; PCB 209 and TCmX were obtained from CPA Chem. Ltd. Stara Zagora, Bulgaria, whereas, the internal standard (^{13}C PCB 141) was obtained from Cambridge Isotope Laboratories, Inc., USA. DCM, methanol, n-hexane (Hex), acetone, iso-octane, concentrated H_2SO_4, Anhydrous sodium sulphate, and silica gel of analytical grade was purchased from Merck. WHO guidelines were followed during sample collection, transportation, and storage. Glassware was washed with distilled water, oven dried at 115 ^oC and baked for six hours (at 450 ^oC). A blank was run prior to each sample set during analysis on GCMS. The average surrogate standards recovery was 69 ± 6% for TCmX and 76 ± % for PCB 209. PCB calibration standards of 2 ppb, 10 ppb, 20 ppb, 50 ppb, 100 ppb and 200 ppb were used for evaluation of the instrument and method accuracy.

Toxicity Equivalent Factor (TEQ)

The TEQ of Some dioxin like PCBs exhibiting similar properties and toxicity as of tetrachlorodibenzo-p-dioxin (TCDD) was calculated (Chovancova*et al.,* 2012). Occurrence of dioxin like PCB congeners (non-*ortho* PCB 77, -126, -169 and mono-*ortho* PCB 105, -114,-118, - 156, -189) were also detected in the sampling population (Ahlborg *et al.,* 1994; De Vito and Birnbaum 1995; Van den Berg *et al.,*, 2006).

Statistical Analysis

PCBs concentrations in maternal serum were analyzed statistically using Microsoft excel and statistical software SPSS (version 16). The concentration of PCBs in serum samples was represented as mean, standard deviation and range. Maps were prepared using Arc GIS 10.0. Box whisker plots were prepared using Statistica 10.0 to represent spatial distribution patterns of PCBs.

RESULTS AND DISCUSSION

PCB profile and congener specific analysis

The maternal blood serum samples were analyzed to determine the concentration of 34 PCB congeners. The concentrations of 34 PCB congeners were grouped on the basis of different classes/ PCB homolog and their results in the form of mean, standard deviation and range are summarized in Table-I. The mean values of total lipids in serum samples were 4.4 g/L and ranged between 4.3 to 5.2 g/L. The mean value of all PCBs in maternal serum was 232.30 ng/g (l.w.). The concentration of PCBs in sampling population ranged from 47.5 ng/g l.w.to 522.1 ng/g l.w. The congener specific analysis showed that the priority PCB congeners were PCB 70, PCB 52, PCB 37, PCB 60 and PCB 66 in a decreasing order. However, the highest concentration of PCB 70 (mean; 18.7 ng/g l.w.) and ranged between 0.6 ng/g l.w. to 44.3 ng/g l.w., whereas, the lowest level of PCB 166 (mean value 0.4 ng/g l.w., range 0.00 ng/g l.w. to 2.4 ng/g l.w.) were recorded from study area. Six indicator PCBs were also detected in the present study and their mean concentrations were PCB 52 (17.46 ng/g l.w.), PCB 101 (12.10 ng/g l.w.), PCB 118 (5.99 ng/g l.w.), PCB 138 (4.87 ng/g l.w.), PCB 153 (6.64 ng/g l.w.) and PCB 180 (2.46 ng/g l.w.). The occurrence of persistent congeners was observed in the PCB profile with mean values as PCB 153 (6.64 ng/g l.w.), PCB 138 (4.9 ng/g l.w.) and PCB 180 (2.5 ng/g l.w.). The concentrations of these congeners were found lower than Bolivia, where the concentrations of PCBs were recorded as PCB 153 (59 ng/g l.w.), PCB 138 (1.80 ng/g l.w.) and PCB 180 (1.00 ng/g l.w.; Arrebola *et al.,* 2012). Ali *et al.,* (2013) reported the mean values of PCBs from Pakistan as PCB 153 (2.63 ng/g l.w.), PCB 138 (2.04 ng/g l.w.) and PCB 180 (1.3 ng/g l.w.) which were lower than the reported in the present study. Garcia *et al.,* (2014) reported the PCB congeners in the blood of the Mexican population as PCB 153 (1.90 ng/g l.w.), PCB 138 (1.80 ng/g l.w.) and PCB 180 (1.00 ng/g l.w.) which was higher than the reported in the present study. Moreover, non-persistent PCB congeners (PCB 52, PCB 66, PCB 74, PCB 101, PCB 105, PCB 128 and PCB 149) were detected in the blood serum samples, indicating that there is continuous and recent exposure to these PCBs from multiple sources (Covaci *et al.,* 2001).

The PCBs congeners were grouped as PCB homologs and their basic descriptive statistics was summarized in Table I. The percentage of PCB homologs on the basis of abundance was recorded as Tetra-CBs (53%) ≥ Penta-CBs (23%) ≥ Hexa-CBs (11%) ≥ Tri-CBs (8%) ≥ Hepta CBs (4%) ≥ Deca-CB (0.7%) ≥ Octa-CB (0.5%).

Table-I: Descriptive Statistics of PCB Congeners in maternal serum samples

District	**Lahore**		**Khanewal**		**Okara**		**Sialkot**		**Chakwal**	
PCBs Homolog	**Mean ± SD**	**Range**	**Mean ± SD**	**Range**	**Mean ± SD**	**Range**	**Mean ± SD**	**Range**	**Mean ± SD**	**Range**
TriCBs	25 ±11	7-54	16 ± 6	8.4-29	14 ± 5	6-23	21 ± 11	4 -38	18 ± 5	11-26
Tetra CBs	169 ± 48	55 - 299	103 ± 38	26.7-190	81 ± 36	13-190	143 ± 82	16-330	139 ± 43	89-205
Penta-CBs	66 ± 29	22-1448	48 ± 23	14 -108	29 ± 27	2.7-103	57 ± 34	9.4 - 148	76 ± 27	12-160
Hexa-CBs	21 ± 9.5	1.4-50	15 ± 10	1.1-39.6	13 ±17	1-60	21 ±14	1.9-59	56 ± 31	20-108
Hepta CBs	10 ± 8	0.8-40	4.2 ± 2	0.7-12.4	8 ± 5	0.7-28	6.5 ± 7	0.4-30	17 ±9	3.8-33
Octa CBS	2.2 ± 2.9	0.1-10	1.1 ±1	0-2.9	0.6 ±0.6	0-2	0.7 ± 0.5	0-1.4	3 ± 3.6	0.4-9
Deca CBs	1.3 ± 1.5	0.1-4.4	2.6 ± 2.4	0.3-8.9	0.8 ±1.6	0-5	2.7 ±3.5	0.1-12	2 ± 0.9	0.4-3
∑PCBs	295 ± 111	87-1905	190 ± 82	51-392	146 ± 92	24-411	252 ± 153	45-619	311 ± 120	137-544

Table II: Toxic Equivalent (TEQs) calculated for maternal blood serum samples using WHO's 2005 Toxicity Equivalency Factors (TEFs)

Compound	**WHO 2005 TEF**	**TEQs**
Non-*ortho* substituted PCB		
PCB 77	0.0001	1.2×10^{-3}
PCB 126	0.1	0.202
PCB 169	0.03	0.042
Mono-*ortho* substituted PCB		
PCB 105	0.00003	1.46×10^{-4}
PCB 114	0.00003	1.17×10^{-4}
PCB 118	0.00003	1.79×10^{-4}
PCB 156	0.00003	4.22×10^{-5}
PCB 189	0.00003	3.88×10^{-5}
$\sum_8$PCBs		**0.245**

Toxicity Equivalent Fluxes of Dioxin like PCBs

The toxicity equivalent fluxes of dioxin like PCBs are given in Table-II. The mean TEQ concentration of dioxin like PCBs was recorded as 0.03 ng/g l.w. (ranged between 3.88×10^{-5} ng/g l.w. to 0.202 ng/g l.w). The results for TEQ values in present study were higher than those reported from Taiwan (0.0045ng/g l.w; Wang *et al.,* 2004).

Spatial Distribution patterns of Homolog PCBs with potential sources

Spatial trend of PCB homologs in sampling matrix from selected sampling zones is represented in Figure 2. The overall prevalence of tetra-CBs followed by Penta-CBs and Hexa-CBs was observed in all sampling populations. This trend was different from the previous studies on serum samples from Pakistan, Bangladesh and China (Ali *et al.,* 2013; Mamun *et al.,* 2007; Bi *et al.,* 2007), where higher chlorinated PCBs were predominant. Levels of Tetra-CBs in the present study were predominant in maternal serum from Lahore. This trend is consistent with the results of a previous study on PCB concentrations reported in food commodities, particularly in rice plant and grains from different areas of Punjab province (Mahmood *et al.,* 2014; Mumtaz *et al.,* 2016). The concentration of Tetra-CBs was observed as highest in all the serum samples of the present study might be due to their higher volatilization into the environment and higher concentration in the food chain supplements as reported in previous studies. Lahore and Sialkot showed an increasing trend of urbanization and the hub of many industrial activities including chemical manufacturing, paints, and dyes manufacturing units. Therefore, exposure of lower chlorinated biphenyls was higher in these urban areas than those in rural areas. Levels of PCBs contamination were also reported in other environmental matrices such as freshwater resources, sediments, fish, indoor dust, food commodities and in human body matrices *viz.* hair, serum and milk in recent studies (Eqani *et al*, 2012; Eqani *et al*, 2013; Ali *et al,* 2014; *Ali et al*, 2013; Mahmood *et al*, 2014; Khawaja *et al*, 2010). Generally, lower chlorinated PCBs were dominated in urban air and the probability of human exposure to these contaminants resulted accumulation in the human body. Among all the homolog groups, highest levels of Tetra-CBs (18.7 ng/g l.w.) was detected in the samples collected from Lahore, whereas, Deca-CB with least concentrations of 0.76 ng/g l.w. were found in Okara. The urban population showed higher concentrations of low chlorinated biphenyls as compared to the rural population in blood samples. A similar trend was observed by Turci *et al.,* (2007) in women populations living in different environments of Italy.

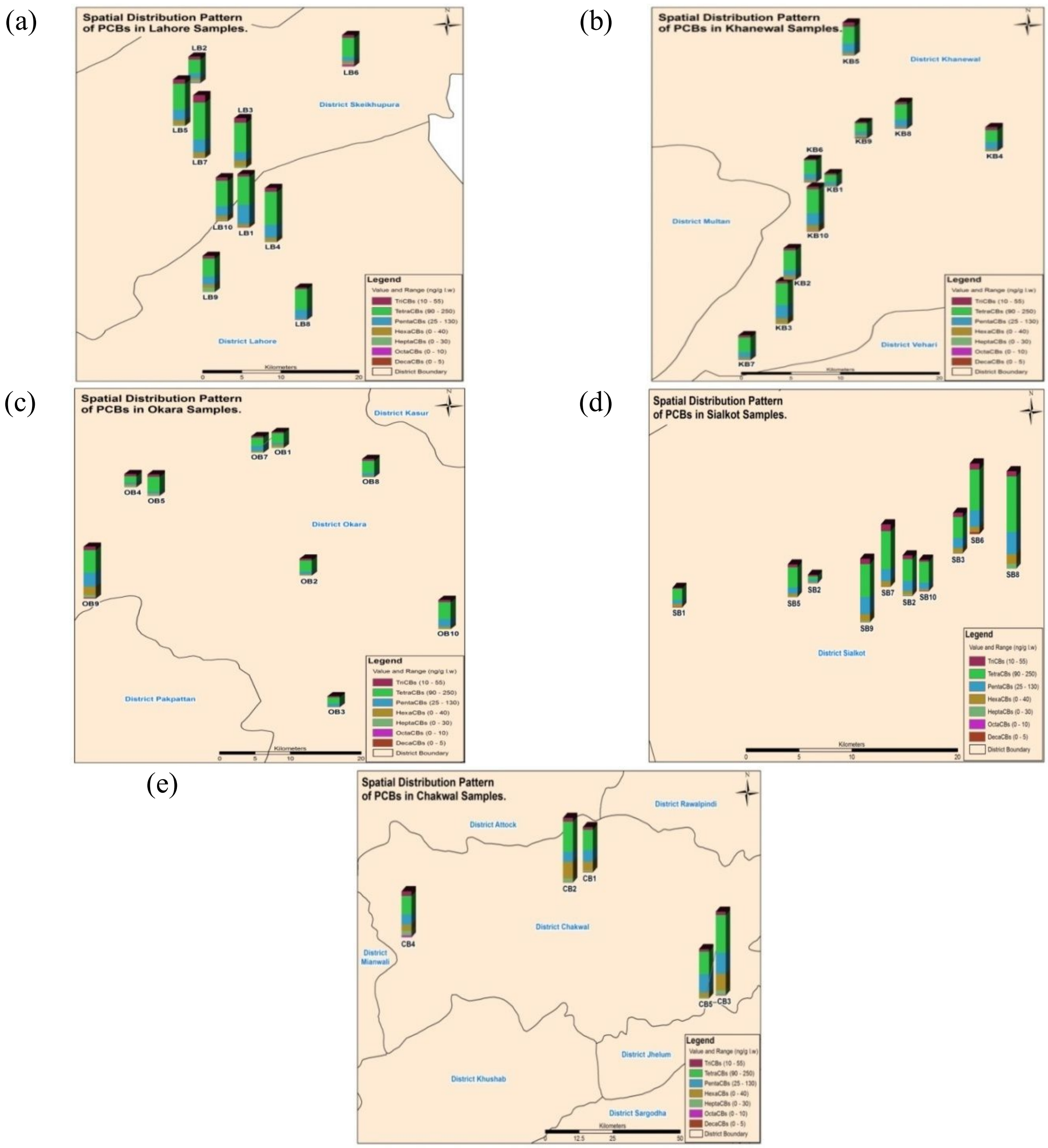

Figure 2: Spatial distribution patterns of homolog PCBs in maternal serum (a).Lahore, (b). Khanewal, (c). Okara, (d). Sialkot and (e). Chakwal.

The lower chlorinated biphenyls are unintentionally formed as byproducts in paints and pigments industry. The disposal and decomposition of PCB containing materials is one of the sources and it enters the human body through inhalation (Rodriguez, 2016). These semi volatile lower chlorinated biphenyls have been detected in substantial quantities in outdoor and indoor air in cities, and older buildings having sealants containing PCBs (Turci *et al.,* 2007; Herrick *et al.,* 2004). Levels of high chlorinated PCBs were higher in Sialkot and Chakwal in comparison to other sampling districts. Consumption of food, particularly meat, fish, oil, dairy products and water contaminated with PCBs and cement industry exhaust are main exposure routes of high chlorination biphenyls (Rodriguez, 2016; Xing *et al.,* 2009). Increasing chlorine atoms in biphenyls rings tend to increase the lipophilicity of these PCB congeners (Xing *et al.,* 2009). Therefore, higher concentrations in Chakwal may be linked with multiple sources viz; food, contaminated air, burning of solid waste and indoor PCBs exposure.

Demographic Characteristics

The mother and infant demographic characteristics are represented in Table III. The age of the volunteers ranged between 19 to 45 years and their BMI was 25.8 kg/m^2.

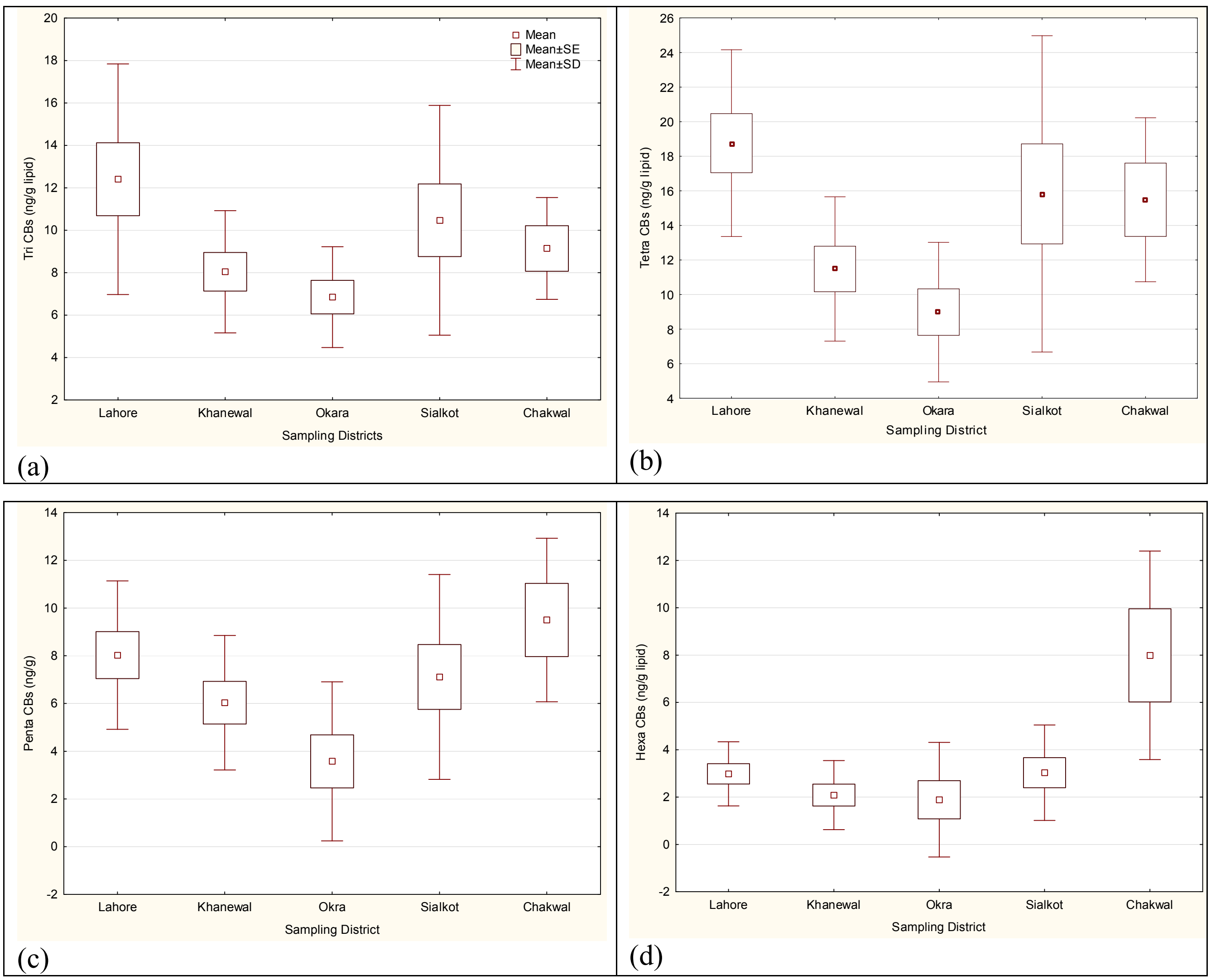

Figure-3: Box whisker plots showing Homolog PCBs patterns in maternal serum (a).TriCBs, (b).Tetra CBs, (c). Penta-CBs, (d). Hexa-CBs

Most of the women population was comprised of multiparae (65 %) with the baby birth weight of 2.21 kg, belonged to rural areas (61 %) and preferred to eat dairy products (34 %) and take milk daily (68 %). The results of one way ANOVA on the sum of PCBs, for the socio-demographic parameters *viz*; locality, age, BMI, the number of children, eating habits and fish intake are given in Table **III**. PCBs showed a significant relationship ($P < 0.05$) for milk intake, indicating the affinities of PCBs with lipophilic substances. A comparison of the present study with other studies on PCBs residues in maternal serum is given in Table IV. The mean concentration of $\sum_{34}$ PCBs in the present study was 232.30 ng/g l.w. There exists limited literature about the PCBs residues in human serum in India (Sharma *et al.,* 2014) where most of the studies were focused on DDT, HCHs, and OCPs, however, Eguchi *et al.,* (2012) reported PCB s concentration of 187 pg/g wet wt. which is less than the results of the present study. The concentration in the present study is comparable to the study reported from Japan (292 ng/g l.w.) and Korea (127.0 ng/g l.w.). The mean serum concentration of PCBs in the current study is lower than the reported from Belgium (595 ng/g l.w.) and Romania (680 ng/g l.w.; Table IV) indicating the indiscriminate past exposure of PCBs in European countries (Dirtu *et al,* 2009). The mean values of Tri-CBs were higher in Lahore (12.4 ng/g l.w) followed by Sialkot (10.47 ng/g l.w), Chakwal (9.1 ng/g l.w), Khanewal (8.04 ng/g l.w) and Okara (6.84 ng/g l.w).

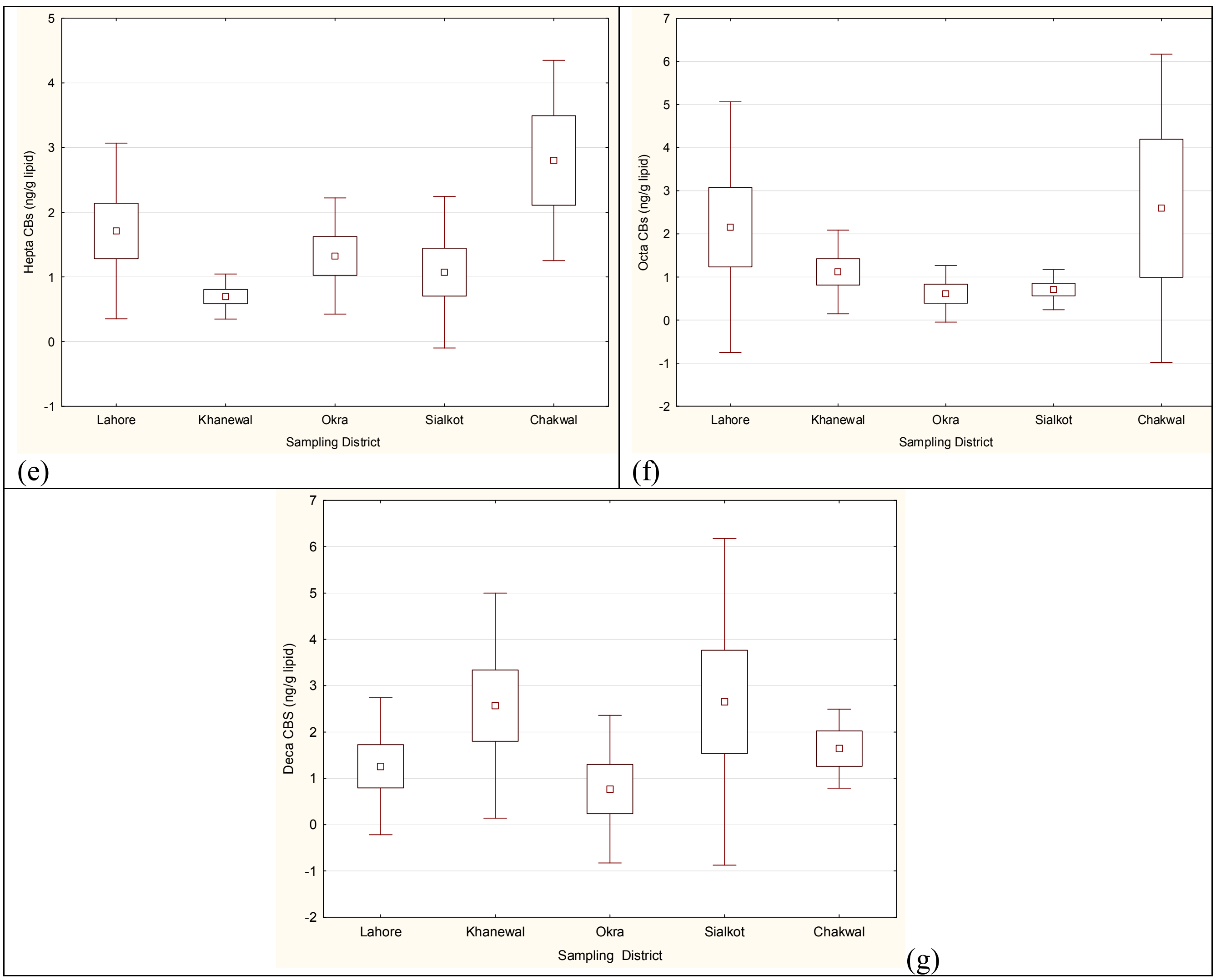

Figure-4: Box whisker plots showing Homolog PCBs patterns in maternal serum (e). Hepta CBs, (f). Octa CBs and (g). Deca CBs

The highest levels of Tetra-CBs were detected in maternal serum from Lahore followed by Penta-CBs and Hexa-CBs in district Chakwal (Figure 3). The box whiskers showed the higher prevalence of lower PCBs in urban areas decreasing towards rural population. A similar trend was observed in a previous study conducted in urban air and soil in Punjab Province (Syed *et al.,* 2013).

CONCLUSION

This study was among the first detailed efforts to report the concentrations of PCB in maternal serum from Punjab province of Pakistan. The mean concentration of PCB in the present study was 232.30 ng/g l.w. Among different PCB homolog, Tetra-CBs were the major contaminants in blood serum of sampling population. The levels of PCBs were laid down at the lower end of the concentration range when compared with some of the developed countries. Sources of PCBs in the environment may include industrial leakage and volatilization from municipal dumping sites; therefore, levels of PCB in the present study were higher in the urban environment as in comparison to rural areas. The results of this study are the potential database for future studies and highlight the need for large scale assessment of PCB residues in mothers from Pakistan and their carryover to the fetus.

Table III: Socio-demographic characteristics and their relationship with PCBs concentrations

Subject Characteristics	Mean± SD	F value	p Value
Age (years)	29.32 ± 5.77	0.563	0.901
Body Mass Index (kg/m^2)	25.77 ± 5.3	0.594	0.848
Infant Birth weight (kg)	2.21 ±1.02	0.563	0.864
Total Lipids g/L	4.4±1.18		
Parity		0.565	0.456
Primiparae	15 (34%)		
Multiparae	29 (65%)		
Location		1.257	0.269
Urban	27 (39%)		
Rural	17 (61%)		
Fish Intake		0.547	0.702
Never	9 (20%)		
Once in a week	4 (9%)		
Once in a month	14 (32%)		
Quarterly	17 (39%)		
Milk Intake		3.218	**0.050***
Never	3 (7%)		
Daily	30 (68%)		
Weekly	11 (25%)		
Eating Habits		0.176	0.982
Meat	2 (4%)		
Vegetable	13 (30%)		
Dairy	15 (34%)		
All of them	3 (7%)		
Vegetable and Dairy	9 (21%)		
Meat and Dairy	1 (2%)		
Meat and Vegetable	1 (2%)		

* Correlation is significant at the 0.05 level (one way ANOVA).

Table IV: Comparison of PCBs concentrations (ng/g l.w.) in maternal blood serum with other studies from developed and developing countries

Country Name	Sample Size	PCB congeners studied	PCB Concentrations		References
			Median	Mean	
Pakistan	44	30, 37, 44, 49, 52, 54, 60, 66, 70, 74, 77, 82, 87, 99, 101, 105 114, 118,126, 128,138, 153, 156, 158, 166, 169, 170, 179, 180, 183 187, 189, 198, and 209	-	232.3	Present study
Sweden	201	105, 118, 138, 153, 156, 167, and 180	552		Glynn *et al.,* 2003
Japan	89	74, 99, 118, 138, 146,153, 156, 163/164, 170, 180, 182/187, 194,199, 206, and 209	38	232	Inoue *et al.,* 2006
Romania	53	101, 99, 118, 146, 153, 105, 138, 187, 184, 156, and180	-	680	Dirtu *et al.,* 2009
Belgium	20	101, 99, 118, 146, 153, 105, 138, 187, 184, 156, and 180	-	595	Dirtu *et al.,* 2009
Poland	22	28, 52, 74, 99,101, 105, 118, 138, 153, 156, 170, 180, 183, 187, 194, 196/203 and 199	79.4	83.0	Jaraczewska *et al.,* 2006
Pakistan	34	118, 153, 180 and 170	13	34	Ali *et al.,* 2013
Bangladesh	24	138, 146, 153 and180	26	-	Mamun *et al.,* 2007
India	-	-	-	0.187*	Eguchi *et al.,* 2012
China	47	-	115	134	Bi *et al.,* 2007
Taiwan	20	28, 52, 101, 138, 153, 180, 77, 81, 126, 169, 105, 114, 118, 123, 156, 157, 167, 189	36.41	28.2	Wang *et al.,* 2004
Korea	40	18, 20, 28, 52, 66, 70, 74, 99, 101, 105, 111, 118, 138, 146, 153, 156, 164, 167, 170, 178, 180, 183 and 187		127	Kang *et al.,* 2008

*Concentration in wet weight

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