















































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Decabromodiphenyl ether in breast milk collected from Saudi mothers

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Abstract

This study presents for the first-time temporal changes of Decabromodiphenyl ether (decaBDE) in Saudi human breast milk samples. Fifty samples were taken, and then the extracted and cleaned samples were put via solid-phase extraction (SPE) before being analyzed with gas chromatography-mass spectrometry. In this research, women eat more meat (69%), followed by eggs (50%), and milk (36%). The bulk of donors (44%) and eggs (33%) eat fish and eggs twice a week. The most of the moms who took part had finished higher education (68%). Among BDEs, BDE-209, -28, -138, and -208 were the predominant congeners. BDE-209 was detected in all analyzed samples (median concentration 0.19 µg/kg lipid) suggesting recent exposure of mothers to decaBDE formulations. The levels of BDE -209 (the most Carcinogenic agent type) were the highest in breast milk were higher (2.5-8 Fold) the levels of other BDEs. Higher rates of egg consumption were positively associated with higher breast milk levels of BDE-28 (P=0.01).^[1] In conclusions, increased exposure to BDE-209 raises health concerns since its breakdown may result in lower brominated congeners and/or other products, which are more toxic than the parent compound. Furthermore, Eggs could be a way for these congeners to get into the bodies of mothers. Also, our results suggest that mothers in Saudi Arabia get BDE-28 from sources other than their jobs, like eating eggs.^[62] More research needs to be done to find out if chickens raised in Saudi Arabia have eggs with a lot of BDEs.

Keywords: DecaBDEs, Breast milk, Estimated dietary intakes, Saudi

1. Introduction

Polybrominated diphenyl ethers (PB DES) are chemical pollutants. They are added & during the manufacture of electronic equipment such as Computers and mobile phones and also in the interior parts of cars and airplanes.^[49] These pollutants leach into the environment by volatilization and are known to have a high bioaccumulation capacity through the food chain (Sjodin et al., 2000, Jakobsson et al., 2002, Jones-Otazo et al., 2005, Darnerud et al., 2006, Wang et al., 2007, Anderson et al., 2008, Lorber 2008, Fraser et al., 2009, Roosens et al., 2009, Harrad et al., 2010, Kuo et al., 2010, Sun et al., 2013, Babichuk et al., 2022, Zhao et al., 2022). PBDES are known to cause maternal toxicity at and are detected in breast milk; with potential fetal toxicity. Toxicity may affect several organs such as the thyroid, liver, kidney, and nervous Systems. These pollutants are also potentially Carcinogenic (Stewart et al., 2003, Costa and Giordano 2007, Main et al., 2007, Cetin and Odabasi 2008, Costa et al., 2008, Albina et al., 2010, Alonso et al., 2010, Schreiber et al., 2010, Sun et al., 2013). The level of exposure to PBDES are expected to be affected by two main factors: the close proximity to products containing PBDES and their level in these products (certain electronic products such as these imported from certain countries, have a relatively high levels).

PBDEs have 209 congeners and are distributed into three marketable products (penta-, octa- and deca-BDE) depending on the number of bromine atoms in their chemical structure (Birnbaum and Cohen Hubal 2006). The penta-BDE formulation, primarily consists of BDE-47 and -99; the octa-BDE formulation consists of a mixture of hexa- to nona-BDEs, while the decaformulation comprises 98% BDE-209 and various nona-BDEs (Miller et al., 2012).^[1] Usage of the penta-BDE formulation has been banned in many countries worldwide since 2004 but the deca-formulation continued to be used until 2013 (Darnerud et al., 2015, Guo et al., 2016).

^[2] Octa-BDE is used in conjunction with antimony trioxide as a flame retardant in the housings of electrical and electronic equipment (Morf et al., 2005) with about 12–15% of the weight of the final product. In the environment, "photolysis, anaerobic degradation and metabolism in biota" can cause

debromination of octaBDE, which produces PBDEs with fewer bromine atoms "which may have higher toxicity and bioaccumulation potential." (Zhang et al., 2017). In May 2009, commercial octaBDE was added to the Stockholm Convention as it meets the criteria for the so-called persistent organic pollutants of persistence, bioaccumulation and toxicity.^[28] There are currently no restrictions on the manufacture or use of PBDEs in Asian countries. Breast milk and house dust have been indicated as the possible main exposure pathways for PBDEs ingested or inhaled by breastfed infants after delivery (Jones-Otazo et al., 2005).

No Previous studies from Saudi Arabia investigated the exposure to BDEs.^[47] The aim of the current study is to investigate the levels PBDEs in breast malks of Saudi lactating mothers.

2. Materials and method

2.1. Patients and sample collection

In this non-international perspective study, ethical approval was obtained from institutional review board (IRB) of King Khalid University Hospital, Riyadh (ref, No. 21/0044/IRB).^[84] A total of 75 mothers were included in the study. These were Consecutive mothers that attended on clinic. Inclusion Criteria were: lactating mothers with overall normal health condition and who are willing to participate in the study. Exclusion criteria were mothers with underlying health problems and those who were not willing to participate.^[67] All Mathers were fully informed with the nature and objectives of the study and signed a consent form. Completed a self-reported questionnaire regarding age, height, weight, diet, residence, and parity. Single-use gloves were worn for sample collection. The breasts were washed with water (without soap or detergents). About 100 ml breast milk were collected in a clean glass. Samples were immediately Ferzan (at - 80°C).

2.2. Samples analysis

PBDES Extraction and clean up from breastmilk was don done according by pajewskaSzmytetal (2019). The level analysis of PB DES were obtained using gas chromatography equipped with mars

detector. Compound quantification will be based on the signals in the mass chromatograms and comparisons with internal standards.

2.3. ^[69] Statistical analysis

Data were presented as mean \pm SD and median (Q1-Q3) Non-parametric tests were used to investigate these values according to the characteristics of the participants (age, education level, parity, obesity status, diet, and residency). A P-value less than 0.05 was considered significant. Further analysis was done using multiple linear regression with a p value less than 0.05 also considered significant.

3. Results and Discussions

3.1. Maternal characteristics

Mothers' demographic data (age, parity, employment, smoking, diet, etc.) is shown in Table 1. The majority (about 90%) of mothers were over 26 years old with average age of 31.4 years. Except for one mother who lived in a rural area, all residence was in Riyadh (Capital city of Saudi Arabia). ^[40] People who live in cities may be more likely to be exposed to flame retardants at home or at work, which could lead to higher PBDE levels. Majority of mothers (about 90%) had a BMI less than 30. The average BMI was calculated to be $25.6 \pm 3.0 \text{ kg/m}^2$. About 90% of mothers were primiparous. In this study, the mothers ate more meat (69%), then eggs (50%), and then milk (36%). Most donors ate fish (44%) and eggs (33% of them) two times a week. Most of the mothers who took part had graduated from college (68%).

3.2. ^[2] Levels and congener profiles of DecaBDEs

The median (Q1-Q3) and average (SD) concentrations of congeners in the breast milk samples collected at three different periods from the participants are shown in Table 2. The repeated-measures ANOVA results indicated that the congener concentrations were same across the three samples. Among

PBDEs, BDE-209, -28, -138, and -208 were the predominant congeners.^[13] The levels of the deca BDE -209 (The most Carcinogenic agent type) were the highest in breast milk were higher (2.5-8 Fold) the levels of other BDES.

The level in our study is still lower to compound in other countries (Meironyte et al.,^[13] 1999, Noren and Meironyte 2000, Hites 2004) but it is alarming that the Deca BDE 209 is the highest level any other types of BDES (Table 2), Other Countries showed that BDE -209 is found at a significantly lower concentration than other brominated BDES (Jakobsson et al., 2012). As Mentioned in the introduction, BDE-209 is carcinogenic compound. BDES are commonly used in electronics as flame retardants. USA, Canada, and several Europeans countries banned the Production of several BDE (Meironyte et al., 1999, Noren and Meironyte 2000, Hites 2004). This is not the care in other countries like china (Ma et al., 2012, Pajewska-Szmyt et al., 2019).

The predominance of BDE-209 in human milk was also found in previous study (Shi et al., 2013) and many other studies from China (Zhang et al., 2014, Yang et al., 2016, Li et al., 2017).^[12] BDE-209 was also the most abundant congener in human milk from a few European countries, including Belgium and Sweden (Croes et al., 2012, Darnerud et al., 2015).^[53] After Penta-BDE and Octa-BDE were phased out, Deca-BDE has become the only commercial PBDE produced and used in China in the past several years.^[60] Because BDE-209 is the main component in commercial Deca-BDE (N95%), we infer that the wide and heavy industrial usage of commercial Deca-BDE should be the primary source for the predominance of BDE-209 in this study.

Most of the electronics in Saudi Arabia are imported from China and that is another concern. A third Concern is the recent increase of involvement of females in industry in Saudi Arabia and hence the levels of BDES are expected to rise over the next decade.^[53] All these concerns dictate further investigations in the future and our study results can be used as a base-line for future studies.^[1]

Increased exposure to BDE-209 raises health concerns since its breakdown may result in lower brominated congeners and/or other products, which are more toxic than the parent compound (Darnerud et al., 2015).^[1] In addition, higher brominated congeners less readily transfer from blood to breast milk

compared to the lower brominated congeners (Mannetje et al., 2013; Thomsen et al., 2010).^[1] Therefore, the levels of BDE-209 in breast milk may underestimate the actual body burden of this compound, since serum levels may be higher than those in breast milk.

^[89] On the other hand, because the partitioning of BDE-209 between serum and human milk is quite low (serum/milk ratio N 10), the proportion of BDE-209 in human milk would be accordingly low (Inoue et al., 2006; Mannetje et al., 2012). Thus, we infer that the predominance of BDE-209 in human milk might suggest that the Saudi population is continuously exposed to elevated Deca-BDE levels.

3.3. Association of PBDE levels with Maternal characteristics and dietary habits

Table 3 shows the correlation between maternal features and maternal nutrition and BDE congeners levels.^[48] There was no statistically significant correlation between maternal age and PBDE congeners.^[1] In contrast to other persistent organic pollutants (POPs), such as polychlorinated biphenyls (PCBs), BDEs have not been observed to accumulate with maternal age (Herbstman et al., 2007, Dimitriadou et al., 2016). This could be because PBDEs have only recently been made and used, which means that people are exposed to more of them than to other POPs whose production and use have been banned for a long time (Herbstman et al., 2007, Muller et al., 2016).^[1] Also, it was suggested that the lack of a link between PBDE levels and age could be due to a behavioral cohort effect, in which younger women may be more likely to use consumer products with PBDEs or eat more foods with PBDEs.^[1] Also, it could be because there wasn't enough control over possible confounders like parity, diet, living habits, and educational level, which can hide the age correlation (Thomsen et al., 2010).^[40] None of the congeners were shown to be significantly associated with body mass index before pregnancy (Table 3).

We studied further the potential relationship between maternal diet and BDE levels (Table 3).^[13] Ingestion of contaminated food, particularly that of animal origin such as fish, meat, milk, and eggs, has been demonstrated to be a primary source of human exposure to BDEs (Matovu et al., 2019). In the current investigation, correlations between the intake of fish, meat, and milk and BDEs were not significant. Similarly, minor and insignificant correlations between human milk BDEs and fish diet were

discovered (Mussalo-Rauhamaa et al., 1988, Mes et al., 1993, Campoy et al., 2001). Variability in the absorption and metabolism of the contaminants among individuals may account for these weak relationships. In addition, contributions from numerous sources of BDEs, such as other dietary items and indoor and outdoor exposure, may potentially account for the absence of connection. According to prior research (Jones-Otazo et al., 2005, Wilford et al., 2005, Wu et al., 2007), food consumption is not the only main source of human exposure to BDEs; nonetheless, environmental scientists and epidemiologists may need to focus on alternate pathways such as dust for the biomonitoring of BDEs.

^[32] On the other hand, our study found that higher rates of egg consumption were linked to higher levels of BDE-28 in breast milk ($P=0.01$). This shows that eggs could be a major source of BDE-28 in the diet. Some authors have found high levels of PBDEs in fatty foods that come from animals, like eggs (Polder et al., 2016, Babalola and Adeyi 2018, Huang et al., 2018). These results agree with those findings.

A recent study by Polder et al. (2016) (Polder et al., 2016) found that eggs from free-range chicken had high amounts of BDE-209 (0.2–311 ng/g lw)^[2]. The authors said that the levels were caused by the birds' behavior of scavenging, which put them in contact with particle-bound contaminants (like BDE-209) that could be broken down into lower congeners (such as BDE-28). Eggs could be a way for these congeners to get into the bodies of mothers. Also, our results suggest that mothers in Saudi Arabia get BDE-28 from sources other than their jobs, like eating eggs.^[34] More research needs to be done to find out if chickens raised in Saudi Arabia have eggs with a lot of BDEs.

Also, regression analysis showed that mothers who previously lived outside the kingdom also had significantly higher level of BDE-28 ($p=0.03$). Living outside the kingdom increases the exposure to electronics and hence the result is not surprising.

4. Conclusion

Our study is the first investigation of decaBDEs in the milk of Saudi lactating women. Among BDEs, BDE-209, -28, -138, and -208 were the predominant congeners. BDE-209 was detected in all analyzed samples (median concentration 0.19 µg/kg lipid) suggesting recent exposure of mothers to decaBDE formulations. The levels of BDE -209 (the most Carcinogenic agent type) were the highest in breast milk were higher (2.5-8 Fold) the levels of other BDEs.^[1] The levels were in the same range of data, or lower than those of mothers in other countries.^[1] Higher rates of egg consumption were positively associated with higher breast milk levels of BDE-28 (P=0.01). Several concern are raised including consumption of eggs and mothers who previously lived outside Saudi Arabia. The results may be used as base-line for future studies Since a gradual rise of BDEs levels is expected in our Country.

Author Contributions

SMY, AMI and AAE contributed to the study design. AMI did the experiments. SMY wrote the manuscript. Manuscript was reviewed by AAE. All authors (SMY, AMI and AAE) have read and approved the final manuscript.

Disclosure of any Conflict

The authors declare that they have no competing interests.

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Table 1 Patients Characteristics

Characteristic		Mean (SD) or N	%	
Mean age in years (SD)		31.4 ± 4.4		
Age, years	21–25	6	8.0%	
	26–30	27	36.0%	
	30	42	56.0%	
Mean BMI, kg/m2 (SD)		25.6 ± 3.0		
BMI, kg/m2	25	27	37.0%	
	25–29.9	41	56.2%	
	30–34.9	3	4.1%	
	35–39.9	2	2.7%	
Urban residence		74	98.0%	
Residence in years	≤5	1	1.3	
	5	74	98.7%	
Rural residence		1	1.3%	
Residence in years	≤5	-		
	5	1	100.0%	
Education				
	≤High school	24	32.0%	
	College	8	10.7%	
	≥Bachelor's degree	43	57.3%	
Birth order	First	68	90.7%	
	Second or higher	7	9.3%	
Parity	Primiparous	68	90.7%	
	Multiparous	7	9.3%	
Diet (before pregnancy)	Omnivore	74	98.7%	
	Vegetarian	0	0	
Reported weekly food consumption (persons)		Egg n(%)	Fish n(%)	Meat n(%) Milk n(%)
Never		0(0.0)	6(8.0)	0(0.0) 0(0.0)
Sometimes		4(5.6)	16(21.3)	0(0.0) 2(2.7)
Once a week		3(4.2)	19(25.3)	1(2.7) 0(0.0)
Twice a week		24(33.3)	33(44.0)	4(5.3) 2(2.7)
More than twice a week, but not every day		38(50.7)	1(1.3)	52(69.3) 27(36.0)
Every day		3(4.0)	0(0.0)	18(24.0) 44(58.7)

Table 2. Mean and Median concentration of BDEs

Mean ± SD	Median (Q1 -	Q3)	N	LOD
BDE-209	0.8 ± 0.5	0.9 (0.2 - 1.2)	63 (84.0)	
BDE-28	0.1 ± 0.1	0.1 (0.1 - 0.2)	2 (2.7)	
BDE-138	0.3 ± 0.1	0.2 (0.2 - 0.3)	62 (82.7)	
BDE-208	0.2 ± 0.1	0.2 (0.1 - 0.3)	2 (2.7)	

Note: Data presented as mean ± SD and median (Q1-Q3) ; LOD indicates limit of detection.

Table 3. Correlation between BDE and participant characteristics

BDEs	Age		BMI		Residing Years		Fish and Fish Products		Meat and Poultry		Milk and Milk products		Eggs	
	R	P-value	R	P-value	R	P-value	R	P-value	R	P-value	R	P-value	R	P-value
BDE-209	0.18	0.57	0.16	0.63	-0.16	0.61	0.18	0.57	0.03	0.92	0.39	0.21	0.17	0.6
BDE-28	0.00	0.97	0.05	0.69	-0.03	0.78	-0.14	0.23	0.03	0.81	-0.01	0.97	0.29	0.01
BDE-138	0.30	0.32	0.02	0.96	0.07	0.82	0.03	0.93	0.45	0.12	0.09	0.77	0.36	0.23
BDE-	0.1	0.13	-	0.74	0.0	0.43	0.03	0.77	0.1	0.31	0.05	0.67	-	0.39

208	8	0.0 4	9	2	0.1
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Note: Data presented as correlation coefficient (R); P-value 0.05 considered significant.