PERSISTENT ORGANIC POLLUTANTS IN HUMAN MILK FROM MOSCOW REGION

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Introduction

Breast milk is a viable biological matrix for assessing human exposure from environmental contamination to POPs in a specific area. With all existing uncertainties analyses of breast milk allow the comparison of levels of contamination for different regions. The purpose of the work is to study levels of contamination with POPs in breast milk from Moscow region.

Materials and methods

The breast milk samples have been collected in hospitals of four towns: Schelkovo, Serpukhov, Sergiev Posad and Solnechnogorsk. Inventarization of industrial and agricultural enterprises in Moscow region showed that first three towns could be attributed to heavily contaminated areas having on the sites potential sources of POPs. Sampling, storage and transportation of the breast milk samples for POPs analyses were done according to WHO protocol[1].

Fig. Location of the towns under study in Moscow region.

The following pollutants have been determined in the samples of breast milk.
Organochlorine pesticides and their metabolite:
Hexachlorobenzene, α-HCH, β-HCH, γ-HCH, 4,4'-DDE, 4,4'-DDD, 4,4'-DDT, 2,4'-DDE, 2,4'-DDD, 2,4'-DDT, toxaphene (Parlar-26, Parlar-50, Parlar-62), heptachlor, cis-chlordane, trans-chlordane, oxychlordane.
Polychlorinated biphenyls (PCBs):
Congeners #28, #31, #52, #99, #101, #105, #118, #128, #138, #153, #156, #170, #180, #183, #187.
PCDD/F:
Seventeen most toxic 2,3,7,8-substituted congeners.

The sample of 15-30 ml of breast milk was placed in Erlenmeyer flask and then isotope-labeled surrogate standard solution was added. After the sample is mixed a blend of acetone-hexane solvents in the ratio 9:1 is added and mixed thoroughly for 5 minutes. Extraction is conducted with 250 ml of hexane using saturated solution of ammonium sulfate for breaking generated emulsion. Extraction performed twice, hexane layers are combined and then washed by twice distilled water from acetone remains. Further the sample is concentrated in a rotary evaporator, cleaned from lipids using gel-filtration on the Bio-Bead SX-3 column and impurities were separated using activated aluminum oxide by column chromatography using columns of silica gel, Florisil and carbon AX-21. For determination of lipids in breast milk the aliquot of primary extract prepared for POPs analysis was used. Determination of lipids was done by the gravimetric method. For PCBs and OCPs extracts have been analyzed using GC/MS (VARIAN SATURN-2200T in the mode of electron impact ionization). For PCDD/Fs and Toxaphenes - GC/MS (VARIAN SATURN 1200MS/MS using chemical ionization with detection of negative ions (NCI) characteristic of these compounds). As part of QA/QC program the samples were analyzed in series. Each series included not more than 12 samples, a procedural blank and a control sample, containing known amounts of analytes. The validity and accuracy of measurements was ensured by using isotope labeled surrogate standards: analogues of all analytes introduced to the samples prior to extraction.

Results and Discussions
Geometric means of concentrations of selected POPs in breast milk from four towns under the study in Moscow region are presented in the Table.
Hexachlorobenzene (HCB)
Higher HCB levels in mean values as well as in concentration range were found in samples from Solnechogorsk (with geometric mean value of 41.1 ng/g of lipids). Close levels of HCB in breast milk with geometric mean of 43.1 ng/g of lipids and ranged from 31.0 ng/g to 108 ng/g[2], were observed in St. Petersburg. HCB content in the breast milk samples from other three towns under the study, varied little with geometric means in towns Serpukhov, Schelkovo and Sergiev Posad 25,4 ng/g, 26.0 ng/g, 26.9 ng/g of lipids correspondingly.
HCH
Concentrations of β-HCH determined in all samples are much higher than those values for other isomers α-HCH и γ-HCH. There is slight difference in mean values for β-HCH concentrations in the breast milk samples from different areas of Moscow region and they are within the range from 1.7 μg/l (Schelkovo) to 3.56 μg/l (Solnechogorsk) or from 65.0 ng/g of lipids (Schelkovo to 101.8 ng/g of lipids (Solnechnogorsk). HCH levels in breast milk from four towns under the study turned out to be close to HCH concentration in breast milk from St. Petersburg, but greatly lower than HCH levels in breast milk of indigenous peoples from Chukotka[2]. Comparison of obtained data with twenty years back results[3] has shown that considerable reduction of HCH levels in breast milk from Moscow region took place during that time.
DDT
Among isomers and metabolites DDT in breast milk 4.4'-DDT and especially 4.4'-DDE are notable for high concentrations. The highest mean concentration of 4.4'-DDT was found in breast milk from Serpukhov which was 12.0 μg/l (range of individual measurements 1.5-86.0 μg/l) or 407 ng/g of lipids (range 137-1900 ng/g of lipids). DDE:DDT ratio is ~15. In other towns under the study mean values range is 3.75-6.55 μg/l (range of individual measurements is 1.17-29.2 μg/l). DDE:DDT ratio is ~10. Comparison of determined data with twenty years back results for DDT concentrations in Moscow region has shown that fivefold reduction of the contamination levels took place during that period[3]. Close levels of DDT in breast milk from St. Petersburg were found[2]. In St. Petersburg samples of breast milk 4.4'-DDE also made up the principle part of DDT group (DDE:DDT ratio is ~10) and its concentrations in individual measurements varied from 2.15 to 23.3 μg/l with

mean value 10.2 µg/l. DDT levels found in breast milk of indigenous peoples of Russian North were noticeably higher (5.1-13.6 µg/l or 246-235 ng/g of lipids) which are comparable or slightly lower than Canadian Arctic levels[4].

Table. Concentrations of selected POPs in breast milk from four towns under the study in Moscow region (geometric means and ranges, ng/g lipids)

Compound	Serpukhov	Schelkovo	Sergiev Posad	Solnechnogorsk
HCB	**25,4**(n.d.- 133,8)	**26,0(**12,73- 50,05)	**26,9 (**17,91- 46,18)	**41,1 (**27,21- 69,14)
α-HCH	**0,90** (n.d.- 1,79)	**0,59** (n.d.- 3,88)	**0,45** (n.d.- 2,99)	**0,93** (n.d.- 2,18)
β-HCH	**82,0 (**37,9 -257,8)	**65,0 (**28,9- 144,8)	**66,0 (**31,5- 211,5)	**102 (**39,4- 488,6)
γ-HCH	**0,49** (n.d.- 12,34)	**0,14** (n.d.- 3,17)	**0,19** (n.d.- 0,62)	**0,24** (n.d.- 1,20)
Oxychlordane	**0,59** (n.d.- 2,66)	**2,38** (n.d.- 5,71)	**3,04** (n.d.- 7,71)	**1,04** (n.d.- 12,96)
***trans*-Chlordane**	**0,02** (n.d.- 0,07)	**0,04** (n.d.- 0,37)	**0,06** (n.d.- 0,11)	**0,05** (n.d.- 1,12)
***cis*-Chlordane**	**0,04** (n.d.- 0,15)	**0,05** (n.d.- 3,16)	**0,06** (n.d.- 0,12)	**0,14** (n.d.- 1,11)
2,4`-DDE	**0,03** (n.d.- 0,29)	**0,08** (n.d.- 0,98)	**0,07** (n.d.- 0,37)	**0,11** (n.d.- 1,23)
4,4`-DDE	**406,76 (**136,5-1897)	**142 (**70,7- 392,3)	**141 (**68,6- 273,1)	**187 (**65,4- 826,2)
2,4`-DDD	**0,07** (n.d.- 0,93)	**0,09** (n.d.- 0,48)	**n.d.**	**0,24** (n.d.- 0,93)
4,4`-DDD	**0,56** (n.d.- 4,28)	**1,22 (**0,05- 8,51)	**1,53 (**0,80- 4,06)	**1,33 (**0,58- 3,11)
2,4`-DDT	**0,38** (n.d.- 3,11)	**0,10** (n.d.- 0,10)	**n.d.**	**n.d.**
4,4`-DDT	**26,4 (**6,66- 95,82)	**14,6 (**7,39- 33,10)	**15,8 (**10,93- 24,92)	**16,1 (**6,23- 30,53)
Σ Toxaphenes	**4,23 (**1,47- 0,43)	**4,81 (**1,20- 11,04)	**3,99 (**1,32- 7,86)	**2,97 (**1,06- 6,52)
Sum of PCBs (as Aroclor 1260)	**363 (**248,6- 534,1)	**260 (**157,8- 391,3)	**308 (**121,5- 668,1)	**243 (**144,8- 494,0)
PCDD/F (TEQ pg/g lipid)	**2,66 (**0,46- 5,06)	**3,63 (**1,81- 7,89)	**3,85 (**2,44- 4,06)	**2,83 (**1,85- 5,67)
Lipid, %	**2,96 (**1,09- 6,66)	**2,62 (**1,80- 4,91)	**3,60 (**2,94- 4,91)	**3,49 (**1,18- 6,71)

n.d. – not detected

PCB

Concentrations of PCBs (geometric mean values) in breast milk from the towns under the study differ from each other not more than 1.5 times. But at the same time there are individual samples with high concentration of PCBs approaching to the levels observed in breast milk from large industrial cities. Geometrical means of PCB concentrations (as Aroclor 1260) are decreasing in a row: Sergiev Posad (11.1 µg/l), Serpukhov (10.8 µg/l), Solnechogorsk (8.50 µg/l) and Schelkovo (6.83 µg/l). Among PCB congeners the prevailing are #99, #118, #138 и #153. Proportion of the congeners in breast milk from several towns differs slightly. Geometrical means of PCB concentrations (as Aroclor 1260) normalized to lipid content in the breast milk samples in decreasing order are: Serpukhov 364 ng/g of lipids (range 249-534 ng/g of lipids), Sergiev Posad 308 ng/g of lipids (122-668 ng/g of lipids), Schelkovo 260 ng/g lipid (158-391 ng/g of lipids), Solnechogorsk 243 ng/g of lipids (145-494 ng/g of lipids). The mean concentration for Moscow shown in 15 years back research[5] is 492 ng/g of lipids. So there was no considerable decreasing of concentration of PCB levels in breast milk from Moscow region during that period of time. Therefore observed values of PCB concentrations in breast milk from the four towns under the study in Moscow region are comparable with their concentrations in breast milk from St. Petersburg and indigenous peoples of Russian North and Canada.

Chlordanes

Oxychlordane makes up the main part of total sum of chlordanes found in breast milk. Its mean concentration in breast milk from Sergiev Posad is 3.04 ng/g of lipids that exceeds its level in breast milk from Serpukhov (0.59 ng/g of lipids) and Solnechnogorsk (1.04 ng/g lipids) in 5 and 3 times accordingly. Levels of oxychlordane in breast milk from Schelkovo (2.38 ng/g of lipids) are lower than in breast milk from Sergiev Posad but comparable. On the whole concentrations of oxychlordane in beast milk from Moscow region are comparable with its concentrations in breast milk from St. Petersburg (2.5 ng/g of lipids)[2] and greatly lower than in breast milk of indigenous peoples of Russian North (5.3 ng/g of lipids)[2] and Canada (81 ng/g of lipids)[4].

Toxaphenes
Only two - #26 and #50 out of three determined congeners of toxaphenes were found in breast milk samples. There are even distribution of mean concentrations of their total in the breast milk samples by regions, though maximum values in individual samples refer to samples from Schelkovo (11.0 ng/g of lipids) and Serpukhov (10.4 ng/g of lipids). Observed levels of toxaphenes are close to concentrations in breast milk from St. Petersburg[2] and greatly lower than levels in samples from Russian North (mean value 45.1 ng/g of lipids with range 7.9–213 ng/g of lipids)[2] and Canada[4].

PCDD/F
Largest mean of PCDD/F concentrations were marked in breast milk samples from Sergiev Posad (3.85 pg/g of lipids) and Schelkovo (3.63 pg/g of lipids) though their concentration in breast milk from other two regions aren't noticeably lower: Serpukhov - 2.66 pg/g of lipids, Solnechogorsk - 2.83 pg/g of lipids. At the same time the highest PCDD/F concentrations in individual samples refer to Schelkovo. Measured levels are comparable with concentrations of dioxins in breast milk from St. Petersburg (2.29 – 5.24 pg/g TEQ of lipids) and of indigenous peoples of Russian North (2.70 -7.66 pg/g TEQ of lipids)[2], and greatly lower than PCDD/F concentrations in breast milk samples from industrialized countries such as England, Germany, Switzerland, France, Belgium (20-50 pg/g lipids)[6]. For Russia mean dioxin concentrations in breast milk (2001-2002) are 8.88 pg/g TEQ of lipids with range 7.46-12.9 pg/g TEQ of lipids[6]. The highest levels were found in breast milk from towns with chlorine industry. So in Chapaevsk breast milk is contaminated with dioxin up to levels of 43.3 pg/g TEQ of lipids[6].

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