SURVEY ON THE ORGANOCHLORINE COMPOUNDS IN HUMAN BREAST MILK COLLECTED FROM BEIJING, CHINA

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Introduction

Organochlorine compounds (OCs) may enter human body as contaminants of dietary animal products. They store in adipose tissue, serum, or breast milk at similar levels on a fat weight basis and persist for a relatively long period. Monitoring of organochlorine pesticides in human milk is very important because infants do not have a fully developed detoxification mechanism and their immune systems and other organs are immature. On the other hand, during lactation process, organochlorine compounds in the body of the mother are mobilized and excreted with the milk. In fact, several studies (1-4) have shown that milk secretion is the most important means of excretion of OCs in women.

In China, the production and usage of technical HCHs and DDTs have been banned in 1983; however, they had been found in all environmental matrixes, such as land, water, and air, as a result of past production and subsequent distribution from polluted sites. In this study we have attempted to estimate the levels of organochlorine pollutants in human milk samples collected in Beijing (China) during 2000-2001. Levels of the residues of these compounds are compared with data from other countries.

Methods and Materials

Sampling. Human milk samples were collected from lactating mothers who resided in Beijing, China. Each donor volunteered ~50mL of her breast milk by manual expression. Samples were collected into glass containers that had been thoroughly cleaned and rinsed in acetone and then carefully dried. Basic information of mothers, such as ages, occupation, possible pesticides exposure, dietary habits, long-term resident region, birth weights of the children were by questionnaire. Milk samples were frozen in a refrigerator at -20 ºC until analysis.

Extraction and Clean-up. The milk fat was dissolved by 20mL *n*-hexane and acetone (1:1, v/v) mixture, and then the sample tube was placed in an ultrasonic bath for 20min. The tubes were centrifuged at 2000 rpm for 10min at room temperature. The total supernatant was transferred into a 100mL flask. The lower aqueous phase was re-extracted by the same procedure and combined organic phases. The organic phases were evaporated to about 2mL through rotary evaporator. After the initial solvent extraction, the column clean-up procedure was performed to further clean and concentrate samples. 2mL of extract was added to a pre-filled glass column contained 5g Florisil with a little sodium sulfate on the top. The column was eluted with 25mL mixed *n*-hexane and petroleum ether (80:20, v/v). The elute was concentrated and adjusted to 1mL by *n*-hexane. Aldrin was added as internal standard before analysis.

Chemical Analysis. 1mL of aliquot of the extracted organochlorine compounds was injected into a Hewlett-Packard 6890A gas chromatograph (GC) equipped with a ^{63}Ni micro-cell electron capture detector (m-ECD) (Agilent, USA). The system was controlled by computer with Chemstation software

obtained from Agilent. A HP-1 fused-silica column (30m×0.25mm I.D.) with 100 % dimethylsiloxane (film thickness 0.25mm) was used. A Shimadzu 17A GC and 5050A mass spectrometer (Shimadzu, Japan) was used to identify the organochlorine compounds with column of DB-1 (30 m × 0.25 mm, 0.25 mm film thickness, J&W Scientific, USA)

The injector temperature was kept at 230°, and the detector temperature was maintained at 250°. The column oven temperature was programmed as follows: initial temperature of 60° for 1min, then rising to 150° at a rate of 20°/min for 1min, and then to a final temperature of 270° (10°/min). High purity nitrogen was used as carrier gas (1.0 mL/min) and make-up gas (40 mL/min).

The gas chromatographic condition of GC-MS was same as GC-mECD, except that the carrier gas was He. Effluents from the GC column were transferred via a transfer line kept at 230° and fed into a 70eV electron impact source held at 250°. The MS were operated under full-scan acquisition mode with mass range 50-500.

Results and Discussion

Basic information

Because of the family planning policy of Chinese government, most of the mothers were feeding their first child. The age of the mothers ranged from 23 to 34 years (mean age: 28.2 years). Eight donors (18 % of the total) were living in the rural region of Beijing city or came from countryside of other provinces in the recent years. Only one donor had contacted with pesticides directly for agricultural purpose.

Residue levels of HCHs and DDTs

We performed the quantitative determinations of organochlorine compounds by internal standard procedure. The aldrin was used as internal standard. Recoveries of spiked cow's milk of different organochlorine compounds ranged from 80 % to 95 %. The detection limit of the method ranged from 0.1 ng/g whole milk for p,p-DDE to 1.0 ng/g whole milk for p,p-DDT. But the final results are not corrected according to the recoveries. The mean value, range, and frequency of occurrence of organochlorine compounds in human milk are determined both in whole milk and in fat weight.

The major organochlorine compounds found in human milk are α-HCH and p,p-DDT, which were detected in about 35 % of samples, but some pesticides (γ-HCH and p,p-DDD) showed only in about 2-4 % of the samples. Others (i.e. dieldrin, endrin, heptachlor and heptachlor epoxide) were not detected in all of the samples. α-HCH was found in 38.6 % of the samples and b-HCH in 22.7 %, with a mean concentration of 63.9 ng/g fat weight (range not detected to 303.7 ng/g) and 49.5 ng/g fat weight (range not detected to 259.1ng/g) respectively. The g-isomer, which is the most toxic HCH, was found in only 4.4 % of the samples with a mean concentration of 55.6 ng/g fat weight (range not detected to 86.9 ng/g). The highest mean concentrations of pesticides found in our study was p,p-DDE, which maximum value reached 1333.9 ng/g fat weight, with a mean of 604 ng/g fat weight; meanwhile their original form, p,p'-DDT, varied from not detected to 188.8 ng/g fat weight, with a mean of 83.5 ng/g. No significant differences between residual levels and the donors' age, dietary habits were observed.

Composition of HCHs and DDTs

The total global HCH usage between 1948 and 1997 has been estimated to be approximately 10 million tons (5). In general, technical HCHs contains the isomers in the following percentages: α: 55-80 %, ß: 5-14 %; γ: 8-15 %, δ: 2-16 %. As previous experimental tests data showed, α-HCH is a more persistent contaminant. In our research, the ß-HCH contains 37 % of total HCHs, which was higher than its percentage in technical HCHs. Because lindane (which contains more than 90 % of γ-HCH)

was not used in large scales in China, the results are reasonable. By comparison with published data from other countries , total HCH residue levels in this study are lower than developing countries, such as India, Jordan, Turkey and Uganda, but higher than most of industrialized countries.

It is noteworthy that the concentrations of DDT and its metabolites found in developed countries are normally higher than in industrialized countries. The maximum values of DDTs in human milk reported in 1990's are in Zimbabwe (6), where the total DDTs value ranged from 1607 to 25,259 ng/g milk fat. The total DDT residues in human milk in Beijing is lower than most of developing countries compared in Table 3 in the same period, and similar with that of Sweden (1992), Canada (1992) and Japan (1998).

Conclusions

The present report only shows results of a small-scale survey of organochlorine contaminations in human milk collected from Beijing, China. The concentrations of organochlorine compounds found in this research are relatively lower than our expectation. It is difficult to compare with data of other countries because most of the investigations were focused on rural individuals, while the mothers in this research are living in a modern city and have seldom chance to contact these compounds directly. Investigations including a greater samples and related organochlorine compounds (such as PCBs, PCDD/PCDFs) may give more complete information of contamination status of mothers and their infants.

Acknowledgments

This work was supported by the National Natural Science Foundation of China and the Chinese Academy of Sciences (20137010, KZCX2-414)

References

1. Jensen, A. A. Chemical contaminants in human milk. *Residue Rev.* 1983, 89, 1-128.

2. Hemandez, L. M.; Fernandez, M. A.; Hoyas, E.; Gonzalez, M. J.; Garcia, J. F. Organochlorine insecticide and polychlorinated biphenyl residues in human breast milk in Madrid. *Bull. Environ. Contam. Toxicol.* 1993, 50, 308-315.

3. Newsome, W. H.; Davies, D.; Docet, J. PCB and organochlorine pesticides in Canadian human milk-1992. *Chemosphere* 1995, 30(11), 2143-2153.

4. Ejobi, F.; Kanja, L. W.; Kyule, M. N.; Muller, P.; Kruger, J.; Latigo, A. A. R. Organochlorine pesticide residues in mother's milk in Uganda. *Bull. Environ. Contam. Toxicol.* 1996, 56, 873-880.

5. Li, Y. F. Global technical hexachlorocyclohexane usage and its contamination consequences in the environment: from 1988 to 1997. *Sci. Total Environ.*1999, 232, 121-158.

6. Chikuni, O.; Nhachi, C. F.; Nyazema, N. Z.; Polder, A.; Nafstad, I.; Skaare, J. U. Assessment of environmental pollution by PCBs, DDT and its metabolites using human milk of mothers in Zimbabwe. *Sci. Total Environ.* 1997, 199, 183-190.