Gonadotoxic Effect of Polychlorobiphenyls

D. S. Gromenko, Sh. N. Galimov, Z. K. Amirova*, A. Z. Abdullina, Yu. Yu. Gromenko, and E. F. Galimova

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny*, Vol. 146, No. 7, pp. 76-79, July, 2008
Original article submitted May 27, 2008

The distribution of polychlorobiphenyls (environmental pollutants) in the reproductive organs and fatty tissues of rats was analyzed and selective accumulation of the toxicants in the epididymis, but not in the ovaries, was demonstrated. Reduction of the fertilizing activity of the ejaculate after chronic exposure to polychlorobiphenyls was detected. Mechanisms of spermatogenic dysfunction under conditions of high technogenic load are discussed.

Key Words: *environment; xenobiotics; epididymis; semen; fertility*

Highly toxic compounds, the most hazardous for humans, form a special group of superecotoxicants among xenobiotics. These are primarily dioxins and some polychlorinated biphenyls (PCB) [11], characterized by carcinogenic, mutagenic, and immunosuppressive effects [14]. Studies of the gonadotoxic effects of PCB were started recently and focused mainly on their effects on the female reproductive system.

We studied the sex-associated distribution and accumulation of PCB under conditions of a model experiment and evaluated the spermatotoxic effect of chronic intoxication with these technogenic pollutants.

MATERIALS AND METHODS

The study was carried out on adult rats of both sexes (220-270 g), receiving intragastrically a PCB preparation Sovtol during 3 months in a summary dose of 0.05 LD_{50}. Controls received the solvent (mineral oil).

The content of individual congeners was measured in pooled samples of the testes, epididymis, seminal fluid, ovaries, and fatty tissue, obtained by pooling the material from at least 3 animals. Quantitative analysis of PCB was carried out by gas chromatography combined with high resolution chromatographic mass-spectrometry [6].

One of the main criteria in evaluation of the effect of chemical compounds on male fertility is the capacity of spermatozoa to fertilize oocytes. As different generations of spermatogenic epithelium are characterized by different sensitivity to the toxic agent, male fertility was evaluated after chronic intoxication with sovtol. With this aim, male rats after 2.5 months of intoxication were caged with intact females (2 females per male). After 2 weeks the males were removed and females were regularly controlled. The number of pregnant females, females with progeny, and the number of newborns per female were recorded. For more objective evaluation of fertility, three main indexes were calculated for quantitative characterization of gonadotropic effects of the toxicant: fertility index=(number of pregnant females/total number of females)×100; gestation index=(number of females with newborns/number of pregnant females)×100; and productivity index=number of newborn pups/number of mothers [2].

RESULTS

The following qualitative characteristics of PCB distribution in males and females were detected: the

Bashkir State Medical University; *Bashkir National Ecological Research Center, Ufa, Bashkiria, Russia

0007-4888/08/1461-0070

TABLE 1. Experimental Study of the Content of PCB in Rat Tissues and Organs

Object of analysis	PCB, μg/g tissue	Biphenyls						SPCB, μg/g tissue	Lipids, g/g tissue	SPCB, μg/g lipids
		Cl_4	Cl_5	Cl_6	Cl_7	Cl_8	Cl_9			
Fatty tissue	33.2	184.1	3240	2654	148.9	—	—	6226.7	0.807	7716
Testes	1.5	14.5	180.2	169.1	16.7	0.9	Traces	381.4	0.247	1544
Semen	0.95	1.5	13.8	15.8	2.0	0.1	Traces	33.2	0.022	1509
Epididymis	21.1	122.4	1335	1073	104.1	Traces	—	2634.2	0.340	7748
Ovaries	41.9	17.9	191.3	211.3	20.7	Traces	—	441.2	0.651	678

entire spectrum of individual congeners from tetra- to nonachlorobiphenyls, the levels of which were within the range of sensitivity of the detection method, was present only in the testes and semen; no octa- and nonachlorobiphenyls were detected in fatty tissue, no nonachlorobiphenyls were found in the epididymis and ovaries, the ovaries containing the highest levels of less toxic nonchlorinated biphenyls (Table 1).

The concentrations of PCB in the rat tissues can be ranked in the following order, from high to low: fatty tissue>epididymis>ovaries>testes>semen. Fatty tissue ranks first by the level of toxicants in this series. Published data also indicate higher accumulation of PCB in the fat [10], which is determined by high lipophilia of these compounds.

Epididymis ranks second, PCB levels in it differing greatly from other tissues: almost 7-fold higher than in the testes and 6-fold higher than in the ovaries. This gradient of toxicant concentrations in the gonads implies that the detected differences are not accidental, but indicate selective accumulation of PCB in the epididymis. Distribution of PCB converted to lipid content in tissues is even more demonstrative: epididymis>fatty tissue>testes>semen>ovaries; in other words, PCBs are deposited virtually similarly in the fatty tissue and epididymis lipids. It is known that this organ is crucial for maturation of spermatozoa, their mobility, and fertilizing capacity.

The most toxic planar PCB with 4-6 chlorine atoms cumulate in the epididymis, similarly as in the testes, in much greater amounts than in the ovaries. By the totality of negative effects these congeners are close to dioxins, that is why toxicity coefficients for them are established in relation to the reference dioxin (2,3,7,8-tetrachlorodibenzo-para-dioxin) [3]. The data in conversion to these coefficients are presented in Table 2.

This method of expression simiplifies comparison of complex heterogeneous mixtures of dioxin-like compounds, because not merely the summary levels of compounds close by the structure and greatly differing by toxicity (by 3-4 orders of magnitude) are estimated, but easily comparable universal values, the so-called WHO toxic equivalents, are analyzed.

This approach demonstrated in fact the same trends of PCB distribution in the body with predominant accumulation in fatty tissue and epididymis with the only difference that conversion to tissue weight virtually completely leveled the differences between the testes and ovaries and the differences became even more demonstrative in conversion to lipid content.

Hence, PCB are highly tropic to fatty tissue and reproductive organs, primarily male, and this fact can serve as one of prerequisites of the fertile potential attenuation under conditions of intoxication by these anthropogenic pollutants.

According to our data, chronic intoxication with PCB was associated with a pronounced reduction of fertilizing activity of the ejaculate (Table 3).

Daily sovtol treatment in a total dose of 0.05 LD50 (for 3 month) resulted in a decrease in the number of pregnant females and females with newborns and in the number of newborn rats, which was associated with a reduction of fertility, gestation, and productivity indexes 5, 1.3, and 2.1 times, respectively. Gestation index, carrying infor-

TABLE 2. Content of PCB in Rat Tissues and Organs in Conversion to WHO Toxic Equivalents (WHO-TEF)

Object of analysis	Fatty tissue	Epididymis	Semen	Testes	Ovaries
WHO-TEF, per g tissue	16.26	8.15	0.08	0.99	1.02
WHO-TEF, per g lipids	21.41	23.97	3.64	4.01	1.57

TABLE 3. Effect of PCB on Rat Male Productivity

Parameter	Groups	
	experiment	control
Total number of females	20	20
Number of pregnant females	4	20
Number of females with newborns	3	20
Fertility index	20	100
Gestation index	75	100
Number of newborn rattlings	13	182
Rattling sex, % of males	53.8	51.1
Productivity index	4.3	9.1

mation mainly on the capacity to full-term pregnancy, changed least of all. All animals of the control group gave birth to progeny.

One of the sensitive indicators of environmental effects on the reproductive function is the progeny sex ratio. The effects of polyhalogenated aromatic compounds on the progeny, specifically, reduction of the proportion of boys, were demonstrated by the results of the analysis of 19,675 newborns, whose fathers were occupationally exposed to dioxins [9]. However, we failed to detect appreciable shifts in this parameter.

One of approaches to elimination of ecopollutants from the body (but ineffective) is excretion with reproductive secretions. It is noteworthy that according to various authors, the concentration of ecopollutants in the spermatoplasma is many-fold higher than in follicular or cervical fluid [7]. Our findings on PCB distribution, on the one hand, confirm these data and on the other, suggest regarding PCB detection in the semen as a reliable informative biological marker of gonadotoxic effect of the habitat pollutants. We previously showed the phenomenon of dioxin contamination of the semen in residents of ecologically unfavorable territories, which served as an indirect proof of relationship between environmental status and changes in the reproductive function [1].

There is no universal opinion on the probable mechanisms of destructive effect of ecopollutants on the reproductive system. The hormonal hypothesis [12] seems to be mentioned more often than others; according to this hypothesis, abnormalities of the male gonadal system are associated with intoxication with chemical compounds simulating the effects of estrogens (so-called xenoestrogens). However, many facts indicate that the xenoestrogen hypothesis lacks sufficient proofs [8].

Dioxins and PCB are also characterized by the xenoestrogen effects, but their main effects are determined by high affinity for Ah (aryl hydrocarbon) receptor (AhR), a specific cytosol protein responsible for bioreception of aromatic carbohydrates [15]. Tryptophane derivatives indigo and indirubin are the physiological ligands of AhR, normally functioning as the transcription factor of regulatory genes involved in xenobiotic metabolism, cell proliferation and differentiation processes, and maintenance of the reproductive function [13]. It was found that estrogen and progesterone receptors inhibited the reaction of the AhR-dioxin complex with xenobiotic-sensitive element of DNA, that is, the two classes of receptors were in competitive relationships, presumably responsible for reproductive disorders.

Exposure to dioxin-like compounds, including PCB, is so polymodal, that it was hypothesized that these substances can act as evolution factors. Today AhR is regarded as a chemical sensor present in all cells, one of its functions being to provide an adequate response to foreign agents of the halogen-aromatic compounds and to minimize the toxic effects [4].

REFERENCES

1. Sh. N. Galimov, Z. K. Amirova, and E. F. Galimova, *Probl. Reprod.*, No. 2, 19-22 (2005).
2. S. A. Kutsenko, *Fundamentals of Toxicology* [in Russian], St. Petersburg (2004).
3. V. I. Maistrenko and N. A. Klyuev, *Ecological Analytical Monitoring of Stable Organic Pollutants* [in Russian], Moscow (2004).
4. S. V. Sibiryak, V. A. Vakhitov, and N. N. Kurchatova, *Cytochrome P450 and Immune System: Facts, Hypotheses, Prospects* [in Russian], Ufa (2003).
5. R. Aitken and D. Sawyer, *Adv. Exp. Med. Biol.*, **518**, 85-98 (2003).
6. M. D. Erickson, *Analytical Chemistry of PCBs*, N. Y. (1997).
7. I. Figa-Talamanca, M. E. Traina, and E. Urbani, *Occup. Med.* (Lond.), **51**, No. 3, 174-188 (2001).
8. H. Greim, *Pediatrics*, **113**, No. 4, Suppl.**,** 1070-1075 (2004).
9. W. H. James, *Scand. J. Work Environ. Health*, **23**, No. 1, 69 (1997).
10. M. Oberg, A. Sjodin, H. Casabona, *et al.*, *Toxicol. Sci.*, **70**, No. 2, 171-182 (2002).
11. A. Schecter and T. Gasiewicz, *Dioxins and Health.* Hoboken: Wiley Interscience, New York (2003).
12. R. M. Sharpe and N. E. Skakkebaek, *Lancet*, **341**, 1392-1395 (1993).
13. K. Sugihara, T. Yamada, S. Kitamura, *et al.*, *Organohal. Comp.*, **59**, 453-456 (2002).
14. M. G. Wade, W. G. Foster, E. V. Jounglai, *et al.*, *Toxicol. Sci.*, **67**, No. 1, 131-143 (2002).
15. C. Wilson and S. Safe, *Toxicol. Pathol.*, **26**, No. 5, 657-671 (1998).