HUMAN THYROID IN THE POPULATION EXPOSED TO HIGH ENVIRONMENTAL POLLUTION BY ORGANOCHLORINATED POLLUTANTS FOR SEVERAL DECADES

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Objective. To study possible effects of long-time exposure of chemical factory employees and population of surrounding polluted area to polychlorinated biphenyls and pesticides on the thyroid volume and function as compared to the population from the area of background pollution.

Methods. A total of 461 adults consisting of 239 men and 222 women was examined and divided into four groups according to their permanent domicile as related to the level of environmental pollution, e.g. SR (area of background pollution, n=207), SI (slightly polluted area, n=59), MI (polluted city of Michalovce, n=94) and CH (employees of chemical factory subjected to high PCB exposure, n=101), combined first three groups being also called LPA (less polluted areas, n=360). Thyroid volume (ThV) and echogenicity were measured by real time sonography. The level of polychlorinated biphenyls (PCB) and pesticides (hexachlorbenzene – HCB, DDE (2,2'-2-*bis*(4-chlorobiphenyl)-1,1-dichloroethylene), *p,p'*-DDT (2,2'-*bis*(4-chlorophenyl)-1,1,1-trichloroethane) and α-, β- and γ-hexachlorcyclohexane – HCH) was estimated by congener specific analysis using HP 5890 gas chromatograph with a ^{63}Ni electron capture detector. Serum levels of thyrotropin (TSH) and thyroid peroxidase antibodies (anti-TPO) were measured by specific sensitive immunoassays.

Results. The association of very high PCB level (e.g. 7300±871 ng/g lipid; mean±S.E.) with increased ThV (e.g. 16.3±0.73 ml) in CH has been found, the values being significantly higher than these of 360 subjects in LPA (e.g. 2045±147 ng/g, $p<0.001$ for PCB and 14.0±0.32 ml, $p<0.001$ for ThV). In 23 subjects from CH with PCB level >10000 ng/g the ThV was 18.7±2.32 ml, while that in 251 subjects from LPA with PCB level of <2000 ng/g was 13.8±0.35 ml ($p<0.05$). In addition, ThV as well as PCB levels were strikingly increasing with age. In parallel with PCB levels, also the levels of other organochlorines estimated (namely these of DDE) were increasing. Although the participation of these substances in the development of adverse effects cannot yet be defined, it cannot be excluded. The association of increased levels of episodic congener PCB 101 with increased ThV appeared to be more pronounced than that of stable congeners PCB 153 and 180. Finally, significant increase in the frequency of thyroid hypoechogenicity by ultrasound, ThV >20.0 ml and thyroperoxidase antibodies in CH area was observed as compared to LPA.

Conclusions. Several associations of high PCB and pesticides level with characteristics of thyroid disorders (e.g. increased thyroid volume, frequency of hypoechogenicity and frequency of positive thyroperoxidase antibodies level in blood) were observed in the area with heavy industrial pollution by PCB.

Key Words: Polychlorinated biphenyls – Pesticides – Industrial pollution – PCB congeners – Hexachlorobenzene – DDE - Thyroid volume - Thyroid disorders

Several reports on the effects of polychlorinated biphenyls and various organochlorinated substances on possible thyroid disruption in wildlife and experimental animals were recently reviewed (Brucker-Davis 1998; Langer 1998a; Karmaus 1998; Brouwer et al. 1998; Hagmar 2003). However, so far only few studies have been conducted in humans and thus several interrelations still remain to be elucidated.

In certain chemical factory in East Slovakia 22,000 tons of PCBs resembling the well known Arochlor 1242 were produced in 1955-85. Considerable amount of solid waste still exists around the factory, while liquid waste was continuously dumped into the nearby Laborec river and Sirava lake without any protective treatment. This resulted in still persisting high contamination of underground and superficial waters, agricultural soil and food chain. During the previous regime the population was kept uninformed about the high risk of such pollution and any scientific or even public information about that has been strictly banned. The same negligence and disregard were characteristic even in respect to longterm use of bulky doses of various pesticides in agricultural production.

As previously (Kocan et al. 1994a) and recently found (Kocan et al. 2001) high environmental pollution is still persisting, since very high levels of PCBs were found in the air (100-1700 pg/m^3), superficial or drinking waters (6-92 ng/l), river, lake and channel sediments (up to 4100 mg/g dry matter), agricultural soil (2-28 ng/g dry matter), fish from polluted resort lake Sirava (average of 375,000 ng/g lipids with upper range value of 933,770 ng/g which is nearly 1 mg/g lipids or 1:1000)), wildlife (34-331 ng/g), cow milk (average 52 ng/g), lard (average 49 ng/g) and pork and beef (range 5-50 ng/g). In addition, the average PCB concentration of 12,300 ng/g was previously found in human adipose tissue and that of 1360 ng/g in human milk.

In one of the previous pilot surveys (Langer et al. 1998b) we found increased thyroid volume, increased frequency of thyroid disorders and thyroid antibodies (e.g. anti thyroperoxidase, anti thyroglobulin and anti thyrotropin receptor) in the employees of chemical factory as compared to a double number of referents from the areas of background pollution. However, in that survey the blood levels of PCBs and other organochlorinated substances (e.g. pesticides) were not measured. From such reason the aim of this survey was to examine additional groups of chemical factory employees and referent subjects and namely, in addition to the preliminary survey, to estimate also the levels of several other organochlorinated substances in all subjects examined and to correlate them with the thyroid volume and some measures of thyroid function.

The levels of PCB in serum of chemical factory employees as found in this survey (average about 7000 ng/g lipids) were 15-20 times higher than these currently reported from the US such as average level of 300-500 ng/g as found in numerous breast cancer cases and controls (Hunter et al. 1997; Helzlsouer et al. 1999; Stellman et al. 2000) and were also higher than, for instance, these of 1000-2000 ng/g (upper range limit of 5900 ng/g) found in Scandinavian fishermen consuming sea food from polluted Baltic (Johansen et al. 1996; Kiviranta et al. 2002; Hagmar 2003), these of about 4000 ng/g in exposed Inuits in arctic Quebec (Iota et al. 1997) or the median value of 5900 ng/g in sea food consuming pregnant women from Faroe Islands (Fangstrom et al. 2002).

Subjects and Methods

Subjects. A total of 239 men (age range 23-74 years, median 47) and 222 women (age range 22-73, median 47) was examined and divided into four groups according to the permanent domicile: 1. SR: 207 subjects from the low pollution district of Stropkov, 2. SI: 59 subjects from 6 villages located in the less polluted area south of resort lake Sirava, 3. MI: 94 subjects from the polluted city of Michalovce and suburban area, 4. CH: 101 long-term employees of formerly PCB producing chemical factory subjected to long-term high exposure by PCB or subjects living in a close vicinity. From each subject several data were obtained by questionnaires concerning namely their health status and professional history, smoking habits and alcohol abuse. Moreover, their thyroid volume and some morphological changes of the thyroid (echostructure, nodules and cysts) were examined by ultrasound and blood sample was obtained for the estimation of several organochlorinated substances (e.g. polychlorinated biphenyls – PCB, hexachlorobenzene – HCB, DDE (2,2'-2-*bis*(4-chlorobiphenyl)-1,1-dichloroethylene), *p,p'*-DDT (2,2'-*bis*(4-chlorophenyl)-1,1,1-trichloroethane) and α-, β- and γ-hexachlorocyclohexane – HCH), thyrotropic hormone (TSH) and thyroid peroxidase antibodies (anti-TPO).

Thyroid volume. Thyroid examination was performed by a real-time instrument (Siemens Sonoline SI-400, Germany) using 7.5 mHz linear transducer. The thyroid volume for each lobe (ml) was calculated according to the ellipsoid formula: width (cm) x length (cm) x thickness (cm) x correction factor 0.479 (Brunn et al. 1982). The measurements were made in each subject lying supine with the neck hyperextended and by the same observer (M.T.) with long-term experience in field surveys and clinical ultrasound diagnostics (Tajtakova et al. 1988, 1998). The intra-observer variation as estimated by 3 subsequent measurements of 50 ThV ranging between 3.0 and 20.5 ml (median 6.2 ml) was 3.9±3.5 % (mean ± S.D.). Since with a large number of measurements the ratio of plus and minus measurement errors approaches 1:1, the final error in estimating the mean values approaches zero. Therefore the effect of intra-observer variability on the final data was considered negligible. Special attention was paid to thyroid hypoechogenicity which was evaluated by comparing the echogenicity of the gland with that of surrounding muscles, both homogeneous and inhomogeneous hypoechogenicity being considered as the same category. The routine procedure was to evaluate the echopattern first and to determine the three dimensions of each lobe and obtain the volume only after this first step.

Blood sampling and laboratory methods. All subjects were instructed in advance to be in a fasting state. The blood was withdrawn with S/Monovette vacutainer (Sarstedt, Austria) and after centrifugation several aliquots of serum were transported frozen on dry ice to the laboratory. They were kept frozen at -20 °C until assayed.

Organochlorinated substances. As described in more detail previously (Kocan et al. 1994b), 5 ml serum were extracted with hexane:ether (1:1 v/v). After the evaporation of solvents the lipids obtained were then dissolved in hexane and applied to a small glass column packed with Florisil, sulfuric acid on silicagel and anhydrous sodium sulfate. The substances were eluted with 25 ml hexane which was then evaporated. The residue was transferred to a roundbottom flask containing hexane which was then evaporated with nitrogen immediately before analysis with an HP 5890 gas chromatograph with a ^{63}Ni electron capture detector. For separation the capillary column DB5JW, 60 m (0.25 mm i.d., 0.25 mm film thickness) made by J&W Scientific (Folsom, CA) was used. The peaks were integrated and quantified with an Apex 2.0 chromatographic computer integrator. Four standard solutions containing PCBs IUPAC No. 28, 52, 101, 138, 153, 180 , hexachlorocyclohexane at concentrations of 10, 50, 200 and 400 ng/ml (Group I), PCBs IUPAC No. 77, 114, 118, 123, 126, 169 and 189 at concentrations of 5, 25, 100 and 200 ng/ml (Group II), HCB and DDT at concentrations of 40, 200, 800 and 1600 ng/ml (Group III) were used for construction of calibration curves. The limit for quantitation for the respective groups was 0.5, 1 and 4 ppb. The results were expressed as ng PCBs (IUPAC No. 28, 52, 101, 138, 153, 180, 105, 118 and 156) per g lipids. After each set of 6-8 samples one solvent blank and one recovery sample (olive oil spiked with a known amount of the analytes) were run. The recovery was 90-110 % except for PCB No. 28 which was 80 %.

The accuracy of the method was regularly validated using certified reference material CRM 349 (PCBs in cod liver oil) and CRM 350 (PCBs in mackerel oil; Community Bureau of Reference - BCR, Commision of the European Communities, Brussels) and also passed testing by the German Society for Labor and Environmental Medicine (University of Erlangen, FRG, 1995) and GEMS/Food-Euro Proficiency Testing by Ministry of Agriculture, Fisheries and Food (Norfolk, UK, 1994).

TSH and anti-TPO. The level of TSH was estimated with the aid of supersensitive immunoradiometric (IRMA) method using the kit by Immunotech (Marseille, France) and anti-TPO was estimated by specific RIA using the kit made by Biocode (Belgium). From epidemiological view, the levels of TSH <0.15 mU/l and >3.0 mU/l were considered abnormal, the levels <0.15 mU/l indicating hyperthyroidism and the range of 3.1-4.5 mU/l indicating high normal values, while the level >4.5 mU/L was considered to indicate subclinical or overt hypothyroidism. In the case of anti-TPO the levels >20 U/ml were considered positive.

Statistical evaluation. After testing the normality of ThV distribution, the differences between individual groups were evaluated with non-parametric Mann-Whitney test and t-test or with Dunn and Bonferroni multiple range test. The differences in the prevalence of individual thyroid antibodies, TSH level and thyroid disorders were evaluated with the aid of Yates chi-square test in respect to the above indicated appropriate cut/off levels. The level of significance used was $p<0.05$.

Results

Table 1 shows a schematic overview on the interrelations between PCB levels as presented by medians, 90th percentiles and ranges in individual age decades of all examined subjects. As shown, the values

Table 1
Levels of polychlorinated biphenyls and selected measures of thyroid morphology and function

Age (years)	No.	Polychlorinated biphenyls (ng/g lipid)			Thyroid volume (ml)	TSH (>4.5 mU/l)	TPO posit	HEC posit	NOD (No.)
		Median	90 %	Range					
21-30	43	925	3 161	351 - 29 781	11.79 ± 0.70	0	2	6	1
31-40	93	1 510	4 552	436 - 58 667	13.76 ± 0.43	1	8	13	4
41-50	160	1 601	6 591	290 - 39 951	13.95 ± 0.42	6	23	24	13
51-60	76	1 907	9 008	328 - 32 010	14.82 ± 0.63	1	10	32	10
61-70	61	3 419	6 990	430 - 23 558	15.35 ± 0.81	2	12	32	2
> 70	27	2 584	13 610	590 - 29 425	17.30 ± 1.66	0	4	12	3

TSH = thyrotropin (number of cases >4.5 mU/l); TPO = thyroperoxidase antibodies (number of cases with the level >20 U/ml); HEC = hypoechogenicity (number of positive cases); NOD = number of cases with thyroid nodules as detected by ultrasound

Table 2
Thyroid volume and levels of organochlorinated substances in the examined areas

Area	No.	Thyroid volume (ml; mean ± SE)	Organochlorinated substances (mean ± SE; ng/g lipid)				
			PCB	DDE	HCB	HCH	DDT
1. SR	207	13.8±0.39^{B}	1 205±53^{BC}	2 620±156*	1 631±136	36.5±1.9^{B}	97± 5
2. SI	51	13.7±0.64^{B}	2 291±167^{B}	3 773±308	2 079±284	36.1±2.7^{B}	43±10
3. MI	94	14.4±0.78^{B}	3 717±304^{B}	3 968±324	1 757±133	28.9±1.2^{C}	137±10
4. CH	101	16.3±0.73^{A}	7 300±871^{A}	3 834±279	1 890±156	23.8±1.7^{A}	143±14
1+2+3	360	14.0±0.32	2 045±147	3 164±137	1738± 95	34.4±1.2	115 ± 4
p (vs. CH)	–	<0.001$^{\#}$	<0.001$^{\#}$	<0.02$^{\#}$	N.S.	<0.001$^{\#}$	<0.02$^{\#\#}$

SR = district of Stropkov with background pollution; SI = area southeast of lake Sirava; MI = city of Michalovce and suburbs; CH = employees of chemical factory and those living in the vicinity;
p = shows the difference between SR+SI+MI and CH;
^{A,B,C} = significant difference (p<0.05) according to Dunn's and Bonferroni's multiple range test between groups with different letters; * = significant difference as compared to Groups 2, 3 and 4 in the same column; $^{\#}$ = significant difference (as indicated in the same line) according to Mann-Whitney's test or $^{\#\#}$ according to Student's t-test

of 90th percentiles increased much more abruptly than these of medians thus showing a nonparametric distribution. The values of ranges in all age groups show very large span which means that in each age group several individuals with extremely high levels may be found.. The ThV is also increasing with age. As concerns the frequency of increased TSH level, positive thyroperoxidase antibodies (TPO), hypoechogenicity (HEC) and nodules (NOD), different absolute numbers for individual age decades represent approximately the same frequencies as related to absolute numbers of cases in the appropriate age groups, the only striking exception being the increased frequency of HEC in age groups >51 years (e.g. 40-53 %) as compared to about 14 % in younger age groups.

Table 2 shows the differences in ThV and the levels of individual organochlorinated pollutants in four different geographic areas. First, the levels of HCH and DDT were negligibly low, while the levels of two most abundant pollutants (e.g. these of PCB and DDE) were relatively the lowest in the area SR of background pollution (although in absolute terms they were still considerably high). In moderately polluted SI such levels were somewhat higher, but in MI namely the levels of PCB were almost tripled as compared to these in SR. However, in CH the average PCB level was twice as high as in MI and three or six times higher as that in SI or SR, respectively. The levels of HCB were approximately similar in all areas and not statistically different. However, the highest levels of all pollutants were found in employees of chemical factory and subjects living in the vicinity (area CH) which were significantly different (with the exception of HCB) from the pooled remaining three areas (e.g. SR+SI+MI) as shown in the last two lines of Table 2. These lines also show that the differences of PCB, DDE, HCH and DDT between 360 subjects from LPA with average PCB level of 2045 ng/g and these of CH with almost four times higher PCB level were highly significant and the same was true for the ThV.

As shown in Table 3, ThV was further compared with the levels of selected PCB congeners, among them namely with these of PCB 101 which has been recently defined by Hansen (1998) as one of "episodic" congeners considered as markers of previous "pulsatile exposures" which are rapidly metabolized and thus generally present only transiently in lower concentrations and lower frequencies in human samples. Actually, in our subjects PCB 101 was still found in 459/461 cases (99.6 %).

Table 3 shows significantly increased ThV (18.5±2.01 ml) in a group of 32 subjects with high PCB 101 level (20.1-336.0 ng/g) as compared to ThV (e.g. 14.2 ±0.28 ml) of 427 subjects from all areas which were defined exclusively by PCB 101 level <20 ng/g. In addition, Table 3 shows also the comparison of ThV with the levels of most abundant "steady state" congeners such as PCB 153 and PCB 180. Although ThV in subjects with the level of these congeners >2000 ng/g was higher as compared to the pooled subjects with the levels <2000 ng/g, the difference was not significant. However, if the cut/off level was arbitrarily increased to 6000 ng/g (see the last line in Table 3), that for PCB 153 in 9 subjects increased to 20.3±5.07 ml) and that for PCB 180 in 6 subjects appeared as high as 22.4±7.39 ml. Although even in this case the difference for individual congeners was not significant possibly due to a low number of cases and broad variation of values, a general positive interrelation between the ThV and PCB level appeared which was supported by the finding of highly significant correlation for all three congeners (p<0.001) by Spearman rank correlation.

Table 4 shows the frequency of several characteristics of thyroid disorders, e.g. that of ThV>20.0 ml, hypoechogenicity by ultrasound, solitary nodules, positive anti-thyroperoxidase antibodies and abnormal TSH level, e.g. sum of the levels >3.00 plus <0.15 mU/l. As shown, significantly increased frequency of ThV <20 ml (e.g. 23.7 % vs. 8.3 %; p<0.001), hypoechogenicity (44.4 % vs. 28.9 %; p<0.01) and positive thyroperoxidase antibodies (18.9 % vs. 11.4 %; p<0.05) was found in chemical factory employees (CH) as compared to pooled areas with lower or background pollution.

Discussion

Extraordinary high exposure of employees of chemical factory and population of certain parts of surrounding area, namely of that living closely to polluted river Laborec and lake Sirava may be strongly supported by very high PCB levels in blood as well as in foods and environmental samples in this area as compared to some other countries. Although the increased frequency of goiters as detected by semiquantitative palpation has been found in so called Yucheng patients who were subjected to rice oil poisoned by PCBs and polychlorinated dioxyfurans (Guo et al. 1999), in this study we found increased thyroid volume in the population exposed to high contamination by PCB by precise ultrasound volumetry which has been used for the first time for similar purpose in our previous study (Langer et al 1998b).

Similarly, even the high serum levels of some pesticides as found in this study apparently showed the ha-

Table 3
Thyroid volume (mean ± SE) in the subjects irrespective of their age, sex and domicile as stratified according to the levels of PCB congeners IUPAC No. 101, 153 and 180

PCB 101			PCB 153			PCB 180		
ng/g lipid	N	ThV (ml)	ng/g	N	ThV (ml)	ng/g	N	ThV (ml)
<10.0	392	14.3 ± 0.29[A]	<1000	343	14.2 ± 0.32[A]	<1000	362	14.3 ±0.32[A]
10.1-20.0	35	13.3 ± 0.91[A]	1001-2000	74	14.9 ± 0.79[A]	1001-2000	60	14.3 ±0.85[A]
20.1-336.0	32	18.5 ± 2.01[B]	2001-17696	44	16.6 ± 1.33[B]	2001-19839	39	16.9 ±0.85[B]
p[1)]		<0.02			0.064			0.061
Line for PCBs >6000			6000-17696	9	20.3 ± 5.07	6000-19839	6	22.4 ± 7.39

[1)] - for each appropriate column of ThV the line p shows the difference between the values labelled by [B] and pooled values labelled by [A] as obtained by Mann-Whitney's rank sum test

Table 4
Frequency of thyroid characteristics in individual areas

Area	No.	Thyroid characteristics (numbers of cases)				
		>20.0 ml	HEC	NOD	TPO	TSH
1. SR	207	17	66	14	24	15
2. SI	59	4	13	3	7	7
3. MI	94	9	25	7	10	9
4. CH	101	24	44	11	19	11
SR+SI+MI (%)		8.3	28.9	6.7	11.4	8.6
CH (%)		23.7^{A}	44.4^{B}	10.9	18.9^{C}	10.9

>20.0 ml = number of ThV >20.0 ml; HEC = hypoechogenicity;
NOD = thyroid nodules detected by ultrasound; TPO = positive thyroperoxidase antibodies; TSH = abnormal thyrotropin levela (e.g. <0.15 plus >3.0 mU/l)
Statistical significance of the difference between the area CH and three pooled areas (e.g. SR+SI+MI) : ^{A} = p<0.001; ^{B} = p<0.01; ^{C} = p<0.05

zardous approach of previous regime to the problem of environmental pollution and protection of the population. Thus, the high levels of DDE and HCB apparently result from uncontrolled massive use of pesticides for several previous decades. Actually, the participation of these toxicants in the observed changes of thyroid characteristics cannot be excluded, but according to the present status of knowledge, the dissociation of the impact of individual substances and the level of their participation cannot be satisfactorily clarified and defined.

When evaluating the findings obtained by thyroid examination, it should be pointed out that the iodine intake in Slovakia is based on well monitored mandatory consumption of iodized salt since early fifties and on the embargo of importing any other salt. Thus, it should be considered optimal which is supported by optimal urinary iodine as found in our previous studies (Tajtakova et al. 1988, 1998). This was also supported by European Thyromobil Study (De-Lange et al. 1997) which evaluated Slovakia as a country with one of the highest urinary iodine and lowest thyroid volume in schoolchildren among several European countries. Our opinion is that such long-term sufficient iodine intake in the whole population gives the unique opportunity to evaluate the antithyroid effects different from iodine deficiency such as environmental and genetic ones. Thus, it is very likely that the observed differences do not appear to be caused by regional, sporadic or individual differences in iodine intake, but rather by the different intake of organochlorinated compounds. Since the levels of PCB in polluted area were 3-4 times higher than these in less polluted, PCB apparently played a leading role among the variety of such substances. Nevertheless, this survey showed that even some other organochlorines, mainly hexachlorbenzene and DDE might participate in the harmful effects and also some assumed participation of dioxins and furans cannot be excluded.

This survey also supported our previous findings on the interrelations between increased organochlorine pollution and thyroid volume, echostructure and frequency of positive anti-TPO antibodies. In addition, some interrelations of PCB level with the frequency of solitary nodules, anti-TPO and abnormal TSH were observed. Our findings are also in agreement with some earlier observations by others. Thus, in workers from PCB producing factory several cases of hypothyroidism were found with elevated levels of autoantibodies against thyroid microsomes (Bahn et al. 1988) which are now considered identical with thyroperoxidase antibodies. Thirty years after acute industrial dioxin exposition of 158 men significantly higher percentage of thyroid disease was found (Zober et al. 1994) in the exposed group (e.g. 7 %) vs. matched controls (e.g. 1.2 %). In a Spanish village located in a vicinity of organochlorine factory 3 new cases of thyroid cancer were found among 5003 inhabitants within 10 years (Grimalt et al. 1994). However, no attention has been surprisingly paid to the thyroid in a number of epidemiological studies of industry workers exposed to PCBs and TCDD as reviewed by Nicholson and Landrigan (1994) and Swanson (1995).

The mechanism by which PCB might produce an increased thyroid volume and increased prevalence of thyroid disorders still remains to be explained. Such changes presumably resulted from long-term multiple effects on the thyroid hormone level in blood, their metabolism in target tissues and on fluctuations in TSH production resulting either in thyroid stimulation or inhibition. Some possible intrathyroid events include the deterioration of cell membrane fluidity by lipophilic PCB (Lopez-Aparicio et al. 1997) thus facilitating the cross-talk between cellular membrane located antigens and lymphocytes of modulated immune system resulting in autoimmune impairment of the thyroid tissue and also possible estrogen like effects of organochlorines (Hansen 1998; Kester et al. 2000; Shiraishi et al. 2003) on the thyroid via estrogen receptors (MANOLE et al. 2001) should be taken in account. Finally, the increased frequency of thyroid antibodies possibly resulting from well known general immunomodulatory effects of organochlorines (RAO and RICHARDSON 1999; MICHIELSON et al. 1999) may also contribute to the development of autoimmune thyroid disorders. Such immunomodulatory effect may be supported by our previous finding of considerably increased frequency of glutamic acid decarboxylase antibodies (anti-GAD) in the employees of chemical factory (LANGER et al. 2002). Finally, even a direct effect of organochlorines on the structure of thyroid follicular cells cannot be excluded (CAPEN and MARTIN 1989).

Finally, it should be stressed that organochlorinated substances do not belong to classic antithyroid drugs directly inhibiting the synthesis of thyroid hormones which results in a decrease of their blood level and following increase of TSH level. It may be suggested that after four decades of exposure we are facing a new and intriguing steady state and we can only find the outlasting and possibly irreversible remnants or sequels of previous long-term and possibly oscillating specific antithyroid actions of those substances such as increased thyroid volume frequently showing impaired echostructure and nodules and also increased frequency of autoantibodies in blood.

Our opinion is that long-term high levels of organochlorines are needed to develop any definitely detectable changes. From this follows that the attention should be focused to the problem of possible threshold level. Several data suggest the possibility of threshold level of organochlorine action as related to the actual and long-time blood level, time of exposure, total body burden etc. Although such level may show wide range, may be individual and may either exist or not, this problem apparently deserves attention and should be further elucidated, since even several recent studies reported relatively low organochlorine levels in serum of case patients which must not reach the critical threshold and thus must not bring conclusive data. However, it should be added that Slovakia belongs to iodine replete countries which means that, in general, any antithyroid effect is being at least partly counteracted by iodine. Thus, in iodine deficient population the threshold level for antithyroid action of PCB might be somewhat lower.

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