ECOLOGY

Effect of Iodine Isotope on the Pituitary-Thyroid and Immune Systems of Children Living on the Territories Polluted by Radionuclides

V. P. Chernyshov, E. V. Vykhovanets, I. I. Slukvin, Yu. A. Antipkin, A. N. Vasyuk, and E. F. Klimenko

Translated from *Byulleten' Eksperimental'noi Biologii i Meditsiny,* Vol. 126, No. 8, pp. 216-220, August, 1998
Original article submitted April 30, 1997

Examination of thyroid function and immune status of children living on the territories polluted by radionuclides in 1993-1994 revealed ^{131}I-dependent thyroid autoimmune reactions. These data indicate a possible effect of ^{131}I on the pituitary-thyroid and immune systems of children living on the radiation controlled territories.

Key Words: *thyroid; iodine isotope; thyroid hormones; lymphocyte subpopulations; children*

Epidemiological studies demonstrated that increased incidence of thyroid cancer among subjects who were children during Chernobyl disaster may directly depend on radioactive exposure, specifically, to ^{131}I [9]. About 160,000 children aged up to 7 years were exposed in 1986 to ^{131}I in a dose of about 0.3×10^{18} Bq. The mean individual dose on the thyroid of children living on the territories contaminated by radionuclides was estimated as 0.6-1.9 Gy [6]. The incidence of ultrasonic changes in the thyroid (nodes and cysts) and changes in echogenic compactness of the thyroid in children living on polluted territories of Russia, Ukraine, and Belarus increased in comparison with the control [7]. Fine-needle biopsy of the thyroid in children with altered echostructure and echocompactness of the thyroid revealed chronic thyroiditis in 31% cases [7].

We assessed the probable role of dosed exposure to ^{131}I (dose-effect) in dysfunction of the pituitary-thyroid system and involvement of autoimmune reactions in its development by analysis of thyroid hormones, thyrotropic hormone (TTH), autoantibodies to thyroglobulin (AAT), ultrasonic examination of the thyroid, and analysis of lymphocytic subpopulations in children living on territories polluted by radionuclides.

Institute of Pediatrics, Obstetrics, and Gynecology, Academy of Medical Sciences of Ukraine, Kiev

MATERIALS AND METHODS

Peripheral blood specimens were collected in 1993-1994 from 53 somatically healthy children (24 boys and 29 girls) aged 7-14 years (mean age 9.8 years), rural residents of the Chernigov and Kiev regions of Ukraine, endemic for goiter [10]. Estimated mean total equivalent dose of exposure to $^{137(134)}$Cs and ^{90}Sr for the population of the studied villages was 0.57-3.09 mSv (milli Sievert, a dose unit for assessing the radiation risk) [1]. Individual absorbed doses of ^{131}I were calculated by Dr. I. A. Likhtarev *et al.* (Research Center of Radiation Medicine, Academy of Medical Sciences of Ukraine) on the basis of direct dose measurements in children in May-June, 1986. Mean estimated age-specific ^{131}I doses for the thyroid in the children living in the Chernigov and Kiev regions [3,4] were used in cases when individual absorbed dose of ^{131}I was unknown. The ratio of children with individual absorbed dose and the

0007-4888/98/0008-0835$20.00

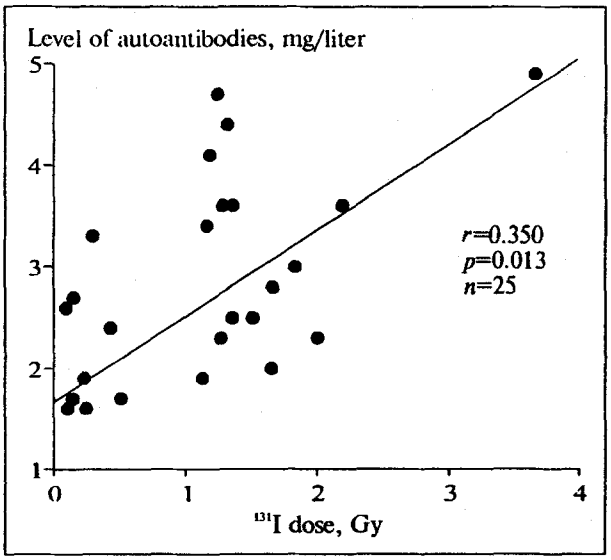

Fig. 1. Relationship between thyroid ^{131}I dose and level of serum autoantibodies to thyroglobulin in children living on the territories polluted by radionuclides.

mean estimated age-specific dose of ^{131}I for the thyroid was 38:18. The reference group consisted of 45 healthy children (21 boys and 24 girls) aged 6-13 years (mean age 7 years) living in the Poltava region on the territories without radionuclide pollution. Doses of ^{131}I for the thyroid, mean total equivalent dose of $^{137(134)}$Cs+^{90}Sr, and the number of children in dose subgroups are listed in Table 1.

Free triiodothyronine (sT_3) and thyroxin (sT_4), TTH, and AAT were measured in the serum by enzyme immunoassay using diagnostic kits (Hoffmann-La Roche and I. I. Metchnikoff Institute of Vaccines and Sera, Russian Academy of Medical Sciences). Ultrasonic examination was carried out using an Aloka SSD-500 device in the real time mode with 7.5 Mhz transducer. Peripheral blood lymphocyte subpopulations were studied with a panel of monoclonal antibodies Simultest IMK-Lymphocyte (Becton Dickinson): CD45/CD14 (HLe-1/Leu-M3), CD3/CD19 (Leu-4/12), CD3/CD4 (Leu-4/3a), CD3/CD8 (Leu-4/2a), CD3/CD16,CD56 (Leu-4/11c+19), conjugated with fluorescein isothiocyanate or ficoerythrin. Whole heparinized blood (100 μl) was mixed with 20 ml of each reagent in the panel of monoclonal antibodies and incubated for 15-20 min. After adding 2 μl lysing solution (Becton Dickinson) to each specimen and 10-min incubation, the mixture was centrifuged to remove lyzed erythrocytes and washed twice in a Cell Wash solution (Becton Dickinson). Cells were then fixed in 0.5 ml 1% paraformaldehyde and analyzed in a FACScan flow cytometer (Becton Dickinson) with the discrimination window imposed on lymphocytes. Nonspecific staining was analyzed using Simultest Control (murine IgG1-FITC+IgG2a-PE, Becton Dickinson). Results were statistically processed using χ^2 test, Mann-Whitney's *U* test, and analysis of correlations.

RESULTS

The number of AAT-positive sera in children with thyroid ^{131}I dose was significantly higher than in the reference group: 25 out of 31 (80.6%) *vs.* 7 out of 42 (16.7%) $p<0.001$. AAT levels in positive sera correlated with ^{131}I dose for the thyroid (Fig. 1). TTH levels did not depend on thyroid ^{131}I dose and were significantly higher than in the reference group (Table 2). On the other hand, the levels of sT_3 and sT_4 in children with thyroid ^{131}I dose were the same as in the control (Table 2). Ultrasonic examinations of 41 children with thyroid ^{131}I dose revealed 13 (31.7%) children with changes in the thyroid echostructure and size. The thyroid was markedly enlarged in 6 out of 41 (14.6%) children, decreased echocompactness of thyroid parenchyma was observed in 6 (14.6%), and thyroid nodules were detected in 4 (9.8%) (Table 3). In 3 children the enlargement of the gland was combined either with decreased echogenicity of the parenchyma or with a node (Table 3). Flow cytofluorometry revealed a significant positive correlation between $CD3^+CD4^+$/$CD3^+CD8^+$ cell ratio and thyroid ^{131}I dose (Fig. 2, *a*). Thyroid ^{131}I dose and the count of both $CD3^+CD8^+$ (Fig. 2, *b*) and $CD3^-$/$CD16,CD56^+$ cells (Fig.

TABLE 1. Thyroid ^{131}I Doses and Mean Total Equivalent Dose of $^{137(134)}$Cs and ^{90}Sr in Children Living on the Territories Polluted by Radionuclides (*M±m*)

Thyroid ^{131}I Dose in subgroup, Gy	Number of children	Mean thyroid ^{131}I dose in subgroup, Gy	Mean total equivalent dose of ^{137}Cs and ^{90}Sr in subgroup, mSv
Less than 1.0	28	0.4±0.07*	1.9±0.17
1.0-2.0	18	1.4±0.09**	1.4±0.22
More than 2.0	7	3.2±0.15	1.4±0.34

Note. $p<0.005$: *in comparison with children with thyroid ^{131}I dose of 1.0-2.0 and more than 2.0 Gy; **in comparison with children with thyroid ^{131}I dose of more than 2.0 Gy.

2, *c*) in peripheral blood of children were in a significant negative correlation.

High level of TTH can explain the fact that territories polluted by radionuclides as a result of Chernobyl disaster are endemic for goiter and the soil in this region contains low levels of natural iodine [10]. Normal levels of sT_3 and sT_4 do not depend on the presence of autoimmune thyroiditis in children with essentially enlarged thyroid and circulating AAT [11]. On the other hand, a persistent high level of serum TTH induces prolonged proliferation of thyroid parenchyma and tumor development in it [13]. It is noteworthy that high content of TTH in children with thyroid ^{131}I dose was associated with autoimmune disorders. The residents of goiter-endemic regions with radiation involvement of the thyroid are at risk of autoimmune disorders caused by death of thyroid tissue and liberation of antigens [2]. In our study such a probability is supported by a positive correlation be-

TABLE 2. Serum Levels of sT_3, sT_4 (pmol/liter), and TTH in (mIU/liter) in children with Thyroid ^{131}I Dose (Gy, $M \pm m$)

Thyroid hormones and TTH	Reference group (n=20)	Children with thyroid ^{131}I dose (n=29)
sT_3	9.6±0.29	10.3±0.44
sT_4	14.5±0.46	18.7±3.0
TTH	1.6±0.18	6.1±1.23*

Note. $p<0.01$ *vs.* reference group.

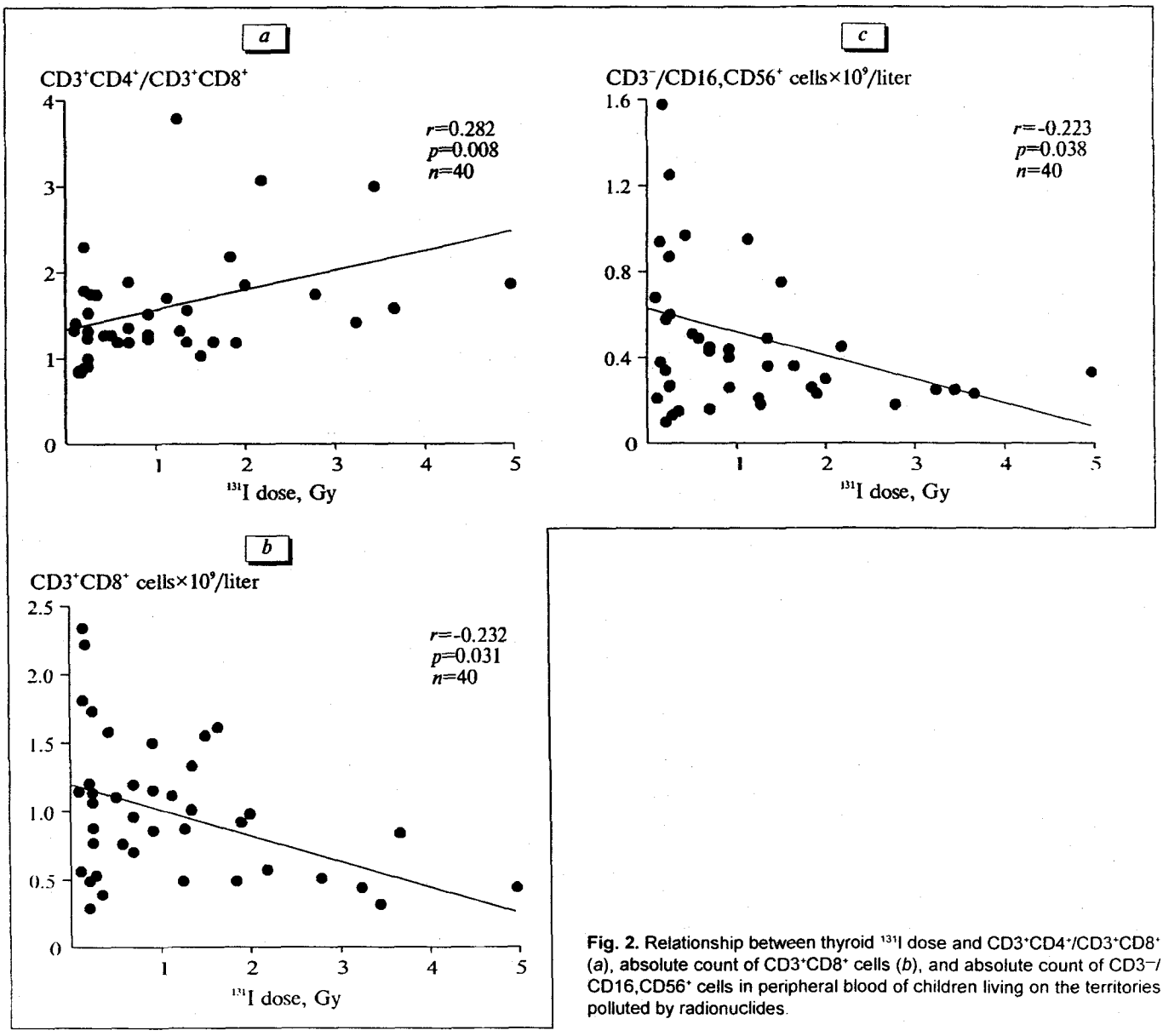

Fig. 2. Relationship between thyroid ^{131}I dose and $CD3^+CD4^+/CD3^+CD8^+$ (*a*), absolute count of $CD3^+CD8^+$ cells (*b*), and absolute count of $CD3^-/CD16,CD56^+$ cells in peripheral blood of children living on the territories polluted by radionuclides.

TABLE 3. Ultrasonic Characteristics of the Thyroid in Children with Thyroid ^{131}I Doses

Group	Norm			Enlargement[1]		
	thyroid echocompactness					
	norm	decrease[2]	node[3]	norm	decrease[2]	node[3]
With thyroid ^{131}I dose (n=41)	28/68.3	4/9.8	3/7.3	3/7.3	2/4.9	1/2.4
Control (n=30)	30/100	0	0	0	0	0

Note. [1]The thyroid was larger than normal for age+2 standard deviations. [2]Diffuse decrease in echocompactness in this subgroup was classified as weak, medium, or expressed. [3]All children in this subgroup had hyperechogenic nodes of more than 5 mm in diameter. Numerator: number of children, denominator: %.

tween thyroid ^{131}I dose and AAT level and the T-helper/inductor and T-suppressor/killer ($CD3^+CD4^+$/$CD3^+CD8^+$) ratios, and the negative correlation between thyroid ^{131}I dose and absolute count of T-suppressors/killers ($CD3^+CD8^+$). A decrease in the count of $CD3^+CD8^+$ cells and inadequate suppressor function of T-cells are typical of an autoimmune thyroid disease [8,15]. Recent ultrasonic examinations of the thyroid demonstrated that diffuse decrease in echocompactness of this organ is typical of patients with autoimmune thyroid diseases [11]. In 15% children in the main group echocompactness of the thyroid decreased and the levels of circulating AAT increased. Therefore, our data point to ^{131}I-dose-dependent autoimmune disorders in children living at goiter-endemic territories polluted with radionuclides.

$CD3^-/CD16,CD56^+$ natural killer cells mediate cytotoxicity irrespective of the major histocompatibility complex and participating in the defense from viral infections and destruction of tumor cells [14]. Therefore, the correlation between the decrease in the count of natural killer cells and higher thyroid doses of ^{131}I may indicate reduced elimination of tumor cells in children with higher ^{131}I doses.

Our data revealed the signs of autoimmune thyroid disease in at least part of children exposed to ^{131}I in 1986 as a result of the Chernobyl disaster and a dose-dependent type of autoimmune reactions in these children. Reports about the relationship between autoimmune disease and thyroid cancer [5,12] prompt further studies of thyroid autoimmune reactions and the pituitary-thyroid status of children living on the territories polluted by radionuclides.

REFERENCES

1. *Dosimetric Passport of Settlements of Ukraine Exposed to Radiation Contamination after Chernobyl Accident* [in Russian], No. 4, Kiev (1994), pp. 230-235.
2. A. M. Lyaginskaya and I. Ya. Vasilenko, *Med. Radiol.*, No. 6, 57-63 (1996).
3. *Thyrodosimetric Passportization of Settlements in the Kiev Region* [in Russian], Kiev (1993), pp. 25-26.
4. *Thyrodosimetric Passportization of Settlements in the Chernobyl Region* [in Russian], Kiev (1993), pp. 47-50.
5. J. G. Desjardins, J. Bass, G. Leboeuf, *et al.*, *J. Pediatr. Surg.*, **23**, 709-713 (1988).
6. *International Chernobyl Project. Technical Report of an International Advisory Committee, International Atomic Energy Agency.* Vienna (1991).
7. M. Ito, B. Yamashita, K. Ashizawa, *et al.*, *Thyroid*, **5**, 365-368 (1995).
8. Y. Iwatani, N. Amino, Y. Hidaka, *et al.*, *Clin. Exp. Immunol.*, **87**, 444-449 (1992).
9. I. A. Likhtarev, B. G. Sobolev, I. A. Kairo, *et al.*, *Nature*, **375**, 365 (1995).
10. V. V. Markov and N. Takeichi, in: *The Chernobyl Accident*, ed. N. Takeichi, Hiroshima (1996), pp. 25-30.
11. C. Marcocci, P. Vitti, F. Cetani, *et al.*, *J. Clin. Endocrinol. Metab.*, **72**, 209-213 (1991).
12. N. Mauras. D. Zimmerman, and J. R. Geollner, *J. Pediatr.*, **106**, 895-898 (1985).
13. J. Robbins and W. Adams, in: *Radiation and the Thyroid*, ed. S. Nagataki, Amsterdam (1989), pp. 11-24.
14. M. J. Robertson and J. Rits, *Blood*, **76**, 2421-2438 (1990).
15. R. Volpe, *Autoimmunity*, **2**, 55-72 (1988).