Antithyroperoxidase and Antithyroglobulin Antibodies in a Five-Year Follow-Up Survey of Populations with Different Iodine Intakes

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Objective: In a follow-up study, we determined the prevalence, incidence, and natural course of positive antithyroperoxidase antibodies (TPOAbs) and antithyroglobulin antibodies (TgAbs) in the general population and examined the influences of iodine intake.

Design: The study was conducted in Panshan, Zhangwu, and Huanghua, regions with mildly deficient, more than adequate, and excessive iodine intake, respectively. Of the 3761 unselected subjects who were enrolled at baseline, 3018 participated in the 5-yr follow-up study. Serum TSH, TPOAb, and TgAb levels were measured.

Results: Among subjects in Panshan, Zhangwu, and Huanghua, the prevalence of positive TPOAbs was 11.23, 11.83 and 12.02%, respectively, whereas 11.23, 11.17, and 11.26% of subjects were TgAb positive, respectively. In the older population (≥45 yr), TgAb-positive individuals were more frequent in Huanghua than Panshan and Zhangwu ($P < 0.05$). The 5-yr cumulative incidence of positive TPOAb was 2.08, 3.84, and 2.84% in Panshan, Zhangwu, and Huanghua, respectively, whereas 2.91, 3.64, and 5.07% of subjects were TgAb positive, respectively ($P < 0.05$), corresponding to the increase in iodine intake. Subjects who were TPOAb and/or TgAb positive at baseline developed thyroid dysfunctions more frequently than those without antibodies (14.44 *vs.* 3.31%, $P < 0.01$); their incidence of elevated TSH levels was 1.32, 8.46, and 15.38% in Panshan, Zhangwu, and Huanghua, respectively ($P < 0.05$).

Conclusions: Subjects who were TPOAb and TgAb positive at baseline developed thyroid dysfunctions more frequently than seronegative subjects. High iodine intake was a risk factor for developing hypothyroidism in antibody-positive subjects. A constant exposure to excessive iodine intake increased the incidence of positive TgAb. **(*J Clin Endocrinol Metab* 93: 1751–1757, 2008)**

Antithyroperoxidase antibody (TPOAB) and antithyroglobulin antibody (TgAb) are two important thyroid autoantibodies that are commonly found in patients with autoimmune thyroid diseases (1, 2). Several studies have suggested that TPOAb can induce antibody-dependent cell-mediated cytotoxicity, and that TPOAb titers correlate with the severity of lymphocytic infiltration, regardless of the presence or absence of hypothyroidism (3–5). However, the function of TgAb remains uncertain (6). The two antibodies, either separately or combined, are also frequently present in the general population (7, 8). The National Health and Nutrition Examination Survey III study reported that more than 10% of disease-free populations are TPOAb or TgAb positive (9). Although there have been several cross-sectional studies on the prevalence of thyroid autoantibodies, apart from the Whickham study (10), longitudinal studies have rarely explored the

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Abbreviations: CI, Confidence interval; CV, coefficient of variation; MUI, median urinary iodine; OR, odds ratio; TgAb, thyroglobulin antibody; TPOAb, thyroperoxidase antibody.

incidence and natural courses of these antibodies in the general population.

Beginning in 1999, we initiated a project to investigate iodine-induced thyroid diseases, consisting of a cross-sectional study and a 5-yr follow-up study (11, 12). In the baseline study, the prevalence of positive TPOAb and TgAb was 9.81 and 9.09%, respectively. However, most of the antibody-positive subjects were euthyroid. Thus, the significance of thyroid autoantibodies in these subjects warranted further study. We undertook the present study to analyze the prevalence, accumulative incidence, and natural courses of TPOAb and TgAb in euthyroid subjects and to explore the factors leading to TPOAb and/or TgAb seroconversion in general subjects, including the influence iodine intake.

Subjects and Methods

Study subjects

The baseline and follow-up studies on thyroid disease in three rural communities in northern China have been described previously (11, 12). The three communities of Panshan, Zhangwu, and Huanghua were areas of mild iodine deficiency with a median urinary iodine (MUI) concentration of 83.5 μg/liter in schoolchildren, more than adequate iodine intake with a MUI concentration of 242.9 μg/liter, and excessive iodine intake with a MUI of 650.9 μg/liter, respectively. Iodine nutrition was stable in each area during the study period. Home visits were made in each of the three areas to all inhabitants who were older than 13 yr of age in 1999, which included 16,287 individuals. Among them, 3761 (934 men, 2827 women) volunteers were enrolled in the baseline study (1103 in Panshan, 1584 in Zhangwu, and 1074 in Huanghua). Of these, 3018 (80.2%) (884 in Panshan, 1270 in Zhangwu and 864 in Huanghua) participated in the 2004 follow-up study that used the same protocol. Blood and urine samples were collected from all participants. Palpation and B-mode ultrasonography of the thyroid were performed.

Research protocols were approved by the medical ethics committee of China Medical University. All subjects provided written informed consent after the research protocols were carefully explained to them.

Laboratory methods

All blood samples were tested for TSH, TPOAb, and TgAb levels using commercial kits (Immulite 2000 chemiluminescent immunoassay; Diagnostic Products Corp., Los Angeles, CA). The baseline samples were tested in 1999, and samples from the follow-up survey were examined in 2004 using the same assay methods, assay kits, laboratory, and technicians. Thirty baseline blood samples tested in 1999 were retested in 2004 for TSH and antithyroid antibodies. The TSH assay is a third-generation assay with a calibration range up to 75 mIU/liter and analytical sensitivity of 0.002 mIU/liter. The intraassay coefficient of variation (CV) was 1.23–1.38%, and the interassay CV was 1.57–4.93% for TSH. The Immulite 2000 TPOAb and TgAb assays had been calibrated against the international reference preparation Medical Research Council 66/387 (anti-TPO) and Medical Research Council 66/93 (anti-Tg) according to National Academy of Clinical Biochemistry recommendations. The TPOAb assay has a calibration range up to 1000 IU/ml and analytical sensitivity of 7 IU/ml. Data higher than 1000 IU/ml were recorded as 1000 IU/ml, and those lower than 10 IU/ml were recorded as 10 IU/ml. The TgAb assay has a calibration range up to 3000 IU/ml and analytical sensitivity of 10 IU/ml. TgAb levels higher than 3000 IU/ml were recorded as 3000 IU/ml, and those lower than 20 IU/ml were recorded as 20 IU/ml. The intraassay CV was 3.51–4.65 and 3.86–6.06%, and the interassay CV was 6.22–8.29 and 5.82–8.78% for TPOAb and TgAb, respectively.

Urinary iodine excretion was determined by the colorimetric ceric ion-arsenious acid ash method as described previously (13). The intraassay and interassay CVs were less than 6.7%.

Reference ranges

The reference range for serum TSH was determined in our laboratory according to the National Academy of Clinical Biochemistry guidelines. TSH levels from 0.3 to 4.8 mIU/liter were euthyroid. This range was derived from the 2.5th to 97.5th percentile log-transformed values for 2503 subjects without a known personal or family history of thyroid disease, without antithyroid antibodies, and without goiter or nodules on B-mode ultrasonography in our survey.

Although the manufacturer of the detection kits has provided their reference ranges for TPOAb and TgAb at 35 and 40 IU/ml, respectively, we determined the cutoff values for these antibodies in our own laboratory (14). Briefly, 2437 euthyroid subjects with normal thyroid ultrasound patterns and without a personal history of thyroid diseases were selected from the baseline survey. Based on these subjects, the upper 95th percentile values for TPOAb and TgAb were 38 and 39 IU/ml, respectively. In the total 3761 baseline population, the percentage of abnormal TSH (>4.8 or < 0.3 mIU/liter) was calculated separately in groups of subjects within certain TPOAb ranges (38–50, 50–60, 60–70, . . . , 500-1000, > 1000 IU/ml) and then compared with that in subjects with TPOAb levels less than 38 IU/ml. Percentages began to significantly increase in the 50–60 IU/ml group, and this trend persisted in the groups with increasingly higher TPOAb levels. The same method was applied for TgAb. The percentage of abnormal TSH in subjects with TgAb levels higher than 40 IU/ml was significantly different from that in subjects with TgAb levels lower than 40 IU/ml. Accordingly, we set the cutoff values at 50 and 40 IU/ml for TPOAb and TgAb, respectively. We used these cutoff values to evaluate the prevalence and incidence of antibodies throughout the survey.

Thyroid ultrasound

Thyroid ultrasonography was performed by trained observers using the same equipment (SA600 with 7.5-MHz linear transducers; Medsion America, Inc., Cypress, CA) in both studies. The volume of each thyroid was estimated by multiplying the thickness, width, length and a correction factor (0.479). A goiter was defined as a thyroid volume exceeding 19.4 ml for women and 25.6 ml for men. Thyroid echo patterns were also detected, and hypoechogenicity was defined as median level echoes not brighter than the overlying cervical strap muscles. This was either multifocal or diffuse. The observers agreed on the methods of image acquisition and interpretation.

Statistical methods

All data were entered into an Excel database and analyzed with the SPSS statistical software package (version 11.5; SPSS Inc. Chicago, IL). For continuous variables, nonparametric tests (two tailed Mann-Whitney *U* tests) were used. Group differences between the numbers of subjects were analyzed using the χ^2 test. Multiple logistic regression analysis was also performed. $P < 0.05$ was considered statistically significant.

Results

The baseline study

The prevalence of TPOAb and TgAb

At the beginning survey, 9.81 and 9.09% of the total population were TPOAb and TgAb positive, respectively. The prevalence of TPOAb- and TgAb-positive subjects was significantly higher among females than males ($P < 0.0001$) and increased with age, especially among females, although there

was a slight decrease in antibodies in participants older than 64 yr of age (Table 1). The overall prevalence of individual antibodies was similar among the three areas with different iodine intakes (Table 1). When divided into age groups, the prevalence of both TPOAb- and TgAb-positive subjects was the same among the three areas in subjects younger than 45 yr. However, in the age group 45 yr and older, TgAb was more common in the Huanghua area (16.33%), with higher iodine intake than in Panshan (9.52%) and Zhangwu (11.05%) (the significance was observed when females and males were calculated together: Huanghua *vs.* Panshan, $P = 0.016$; Huanghua *vs.* Zhangwu, $P = 0.029$, Fig. 1). The same tendency was seen for TPOAb, especially in males, but it was not significant (Fig. 1).

In the total population, 13.69% were positive for TPOAb and/or TgAb. Of the TPOAb-positive subjects, 53.12% were also TgAb positive, and of the TgAb-positive subjects, 57.31% were also TPOAb positive. TPOAb alone was positive in 4.6%, and TgAb alone was positive in 3.88%. Both TPOAb and TgAb were positive in 5.21% of the total population.

Relationship between thyroid autoantibodies and TSH

The TSH values were significantly higher in patients positive for both TPOAb and TgAb than in populations with negative antibodies or only one positive antibody ($P < 0.05$); however, there were no significant differences among these three groups (Fig. 2). The percentage of elevated TSH (TSH > 4.8 mIU/liter) in either TPOAb- or TgAb-positive subjects was 14.76, and 2.65% in the antibody negative group ($P < 0.0001$). The percentage of subnormal TSH (TSH < 0.3 mIU/liter) in either TPOAb- or TgAb-positive subjects was 15.34, and 3.3% in the antibody-negative group ($P < 0.0001$). The percentage of elevated TSH in either TPOAb- or TgAb-positive subjects was 5.26, 14.29, and 23.42% in Panshan, Zhangwu, and Huanghua, respectively ($P < 0.0001$), and the increase was associated with higher iodine intake. The percentage of subnormal TSH in either TPOAb- or TgAb-positive subjects was 19.55, 15.18, and 12.03% in Panshan, Zhangwu, and Huanghua, respectively, with no significant difference among the three regions.

The follow-up study

The 5-yr cumulative incidence of TPOAb and TgAb-positive subjects (Table 2)

There were 3047 subjects (922 in Panshan, 1265 in Zhangwu, and 860 in Huanghua) who had normal TSH values and were not positive for TPOAb and/or TgAb in the baseline study. Among them, 2381 (78.14%, 721 in Panshan, 990 in Zhangwu, and 670 in Huanghua) participated in the follow-up study. The 5-yr cumulative incidence of TPOAb-positive subjects was 2.81% in this population, with no differences between males and females. The cumulative incidence of TgAb-positive subjects was 3.82% and was significantly higher in females than males (4.41 *vs.* 2.21%, $P = 0.014$). The cumulative incidence of TgAb-positive subjects was 2.91, 3.64, and 5.07%, in Panshan, Zhangwu, and Huanghua, respectively, and was significantly higher in the iodine excessive Huanghua community than in the mildly iodine-deficient Panshan community ($P = 0.039$). Among females, the incidence of TgAb was significantly higher than that of TPOAb ($P = 0.019$), and this difference was also clear in Huanghua community ($P = 0.019$). In males, the incidence of TPOAb-positive subjects in the Zhangwu community was much higher than that in the Panshan community ($P = 0.029$).

By logistic regression, our survey indicated that euthyroid individuals with normal TPOAb concentrations at the higher end of the normal range were at risk for becoming TPOAb positive in 5 yr [odds ratio (OR) 4.3, 95% confidence interval (CI) 2.82–6.57, $P < 0.0001$]. Age, gender, and iodine nutri-

TABLE 1. Prevalence of positive TPOAb and TgAb in three counties with different iodine intakes at baseline study

	Panshan			Zhangwu			Huanghua			All		
Age (yr)	n	TPOAb n (%)	TgAb n (%)	n	TPOAb n (%)	TgAb n (%)	n	TPOAb n (%)	TgAb n (%)	n[a]	TPOAb n (%)	TgAb n (%)
Male												
14–24	76	1 (1.32)	1 (1.32)	70	4 (5.71)	1 (1.43)	63	2 (3.17)	2 (3.17)	209	7 (3.35)	4 (1.91)
25–34	66	2 (3.03)	2 (3.03)	79	2 (2.53)	1 (1.27)	65	1 (1.54)	0 (0)	210	5 (2.38)	3 (1.43)
35–44	66	3 (4.55)	3 (4.55)	80	0 (0)	0 (0)	64	5 (7.81)	1 (1.56)	210	8 (3.81)	4 (1.9)
45–54	48	1 (2.08)	0 (0)	72	2 (2.78)	1 (1.39)	42	4 (9.52)	5 (11.9)	162	7 (4.32)	6 (3.7)
55–64	17	1 (5.88)	1 (5.88)	47	2 (4.26)	3 (6.38)	26	3 (11.54)	1 (3.85)	90	6 (6.67)	5 (5.56)
65+	11	1 (9.09)	0 (0)	27	2 (7.41)	1 (3.7)	15	2 (13.33)	2 (13.33)	53	5 (9.43)	3 (5.66)
Overall	284	9 (3.17)	7 (2.46)	375	12 (3.2)	7 (1.87)	275	17 (6.18)	11 (4)	934	38 (4.07)	25 (2.68)
Female												
14–24	114	8 (7.02)	9 (7.89)	105	4 (3.81)	7 (6.67)	117	5 (4.27)	6 (5.13)	336	17 (5.06)	22 (6.55)
25–34	280	34 (12.14)	38 (13.57)	351	41 (11.68)	38 (10.83)	230	18 (7.83)	23 (10)	861	93 (10.8)	99 (11.5)
35–44	228	23 (10.09)	20 (8.77)	356	40 (11.24)	35 (9.83)	235	37 (15.74)	20 (8.51)	819	100 (12.21)	75 (9.16)
45–54	135	19 (14.07)	17 (12.59)	252	37 (14.68)	34 (13.49)	158	23 (14.56)	33 (20.89)	545	79 (14.5)	84 (15.41)
55–64	47	6 (12.77)	7 (14.89)	92	15 (16.30)	17 (18.48)	50	10 (20)	6 (12)	189	31 (16.4)	30 (15.87)
65+	15	2 (13.33)	1 (6.67)	53	6 (11.32)	4 (7.55)	9	3 (33.33)	2 (22.22)	77	11 (14.29)	7 (9.09)
Overall	819	92 (11.23)	92 (11.23)	1209	143 (11.83)	135 (11.17)	799	96 (12.02)	90 (11.26)	2827	331 (11.71)	317 (11.21)

n, Number of cases.

[a] Number of the total population at baseline study. Total 3761 volunteers were sampled at baseline.

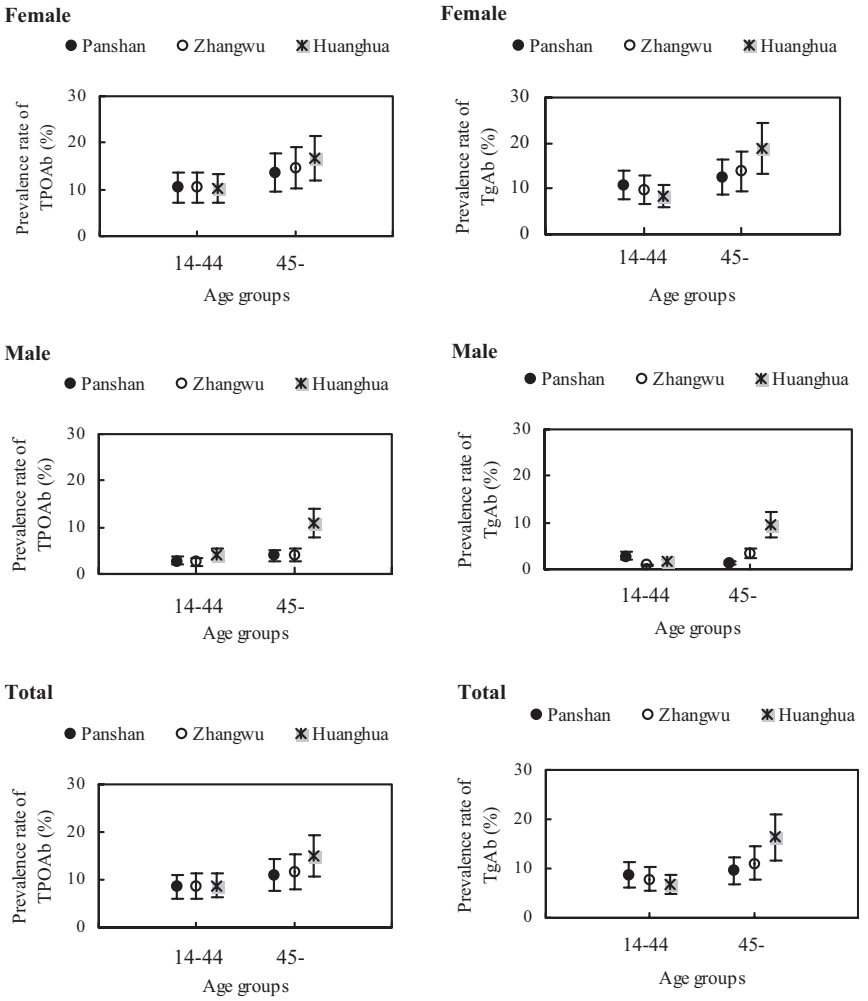

FIG. 1. Comparison of the prevalence of TPOAb- and TgAb-positive subjects from different iodine intake areas by different age groups. Panshan, Zhangwu, and Huanghua were areas with mild iodine deficiency, more than adequate iodine intake, and excessive iodine intake, respectively. *Vertical bars* represent 95% confidence intervals for prevalence.

tion status were not indicated as risk factors for becoming TPOAb positive. Logistic regression also showed that females with TPOAb and TgAb levels at the higher end of the normal range were at increased risk for becoming TgAb positive in 5 yr (female gender OR 1.9, 95% CI 1.06–3.4, $P = 0.03$; baseline TPOAb concentration OR 2.13, 95% CI 1.27–3.56, $P = 0.004$; baseline TgAb concentration OR 5.17, 95% CI 1.68–15.9, $P = 0.004$). Age and iodine nutrition were not risk factors for becoming TgAb positive.

The outcomes of subjects who were TPOAb and/or TgAb positive at the baseline survey

In our follow-up survey in the Panshan, Zhangwu, and Huanghua communities, 56, 79, and 49 euthyroid subjects who were TPOAb positive in the first survey were resampled. As described before, our survey indicated that 79.35% remained TPOAb positive, and the higher the original TPOAb levels, the more likely the individuals remained TPOAb positive (11). The percent of subjects remaining TPOAb positive was higher in females than males (81.33 *vs.* 61.11%) but failed to reach statistical significance ($P = 0.088$). In addition, the original thyroid volume in subjects that were persistently TPOAb positive was significantly larger than those who became TPOAb negative (male: 16.30 ± 4.68 ml *vs.* 11.66 ± 4.23 ml, female: 17.7 ± 6.93 ml *vs.*14.33 ± 5.38 ml, $P = 0.001$).

In 2004 in the Panshan, Zhangwu, and Huanghua communities, 61, 85, and 50 euthyroid subjects who were TgAb positive in the original survey were resampled. In these subjects, 73.47% remained TgAb positive. Similar to the TPOAb results, the higher the TgAb level, the higher the percentage of subjects who remained TgAb positive (52.63% of subjects with levels from 40 to < 100 IU/ml, 84.15% of subjects with levels from 100 to < 500 IU/ml, and 92.11% with levels of 500 IU/ml or higher). Neither thyroid volume nor gender influenced the TgAb outcome.

Our survey indicated that TgAb serostatus became negative more frequently than TPOAb serostatus when both were originally positive (28.12 *vs.* 15.63%, $P = 0.001$). As for subjects positive for both antibodies at the baseline survey, only 7.29% were negative for these antibodies in the follow-up study.

People who were TPOAb positive at the first survey were more likely to become TgAb positive than those who were TPOAb negative (cumulative incidence 11/88). Similarly, people who were initially TgAb positive were more likely to become TPOAb positive (cumulative incidence 21/100).

The outcomes of thyroid function in euthyroid, TPOAb and/or TgAb-positive subjects

After 5 yr, the incidence of abnormal TSH (TSH > 4.8 mIU/liter or TSH < 0.3 mIU/liter) in euthyroid subjects with antibodies at baseline was much higher than euthyroid subjects that were antibody negative ($P < 0.0001$). Positive antibodies, whether TPOAb alone, TgAb alone, or both, increased the incidence of TSH abnormalities; there were no obvious differences among these positive groups (Table 3).

A higher iodine intake and higher incidence of supranormal TSH (TSH > 4.8 mIU/liter) was found in euthyroid subjects that were TPOAb and/or TgAb positive at baseline (among the three cohorts of Panshan, Zhangwu, and Huanghua, $P = 0.010$) (Fig. 3). Meanwhile, after 5 yr, the incidence of subnormal TSH (TSH < 0.3 mIU/liter) in initially antibody-positive euthyroid subjects was not significantly different among the three cohorts (Fig. 3).

Among the 184 euthyroid individuals that were either TPOAb positive or TgAb positive in 1999, those developing

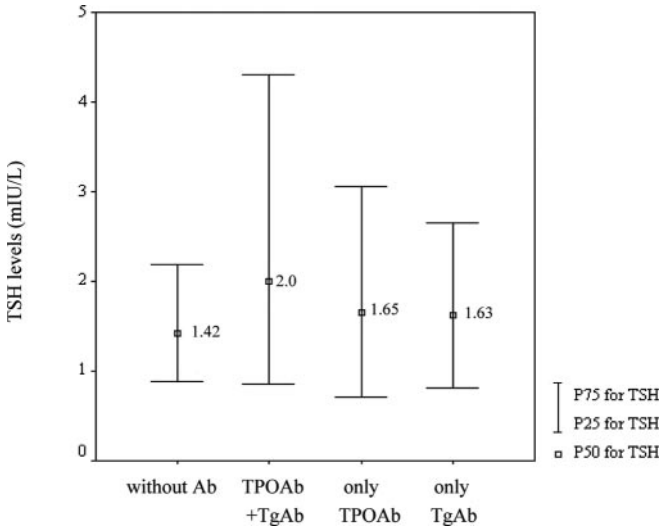

FIG. 2. Median TSH levels in subjects with or without thyroid antibodies at baseline. P50, P25, and P75 represented TSH levels at the median, 25th, and 75th percentile of the groups, respectively. The median TSH level was significantly higher in subjects positive for both antibodies than in seronegative subjects ($P < 0.0001$) or subjects positive for one antibody ($P < 0.05$).

hypothyroidism in 2004 had higher baseline TSH levels than those remaining euthyroid (median: 3.14 *vs.* 1.74 mIU/liter, $P < 0.0001$). Subjects who were antibody positive with TSH 2 mIU/liter or greater in 1999 were more likely to develop supranormal TSH levels than those who were antibody positive with TSH less than 2 mIU/liter (16.28 *vs.* 2.04%, $P = 0.001$).

Discussion

In the present study, we used a sensitive method to detect TPOAb and TgAb in the general subjects from three rural Chinese communities with a mild iodine deficiency, more than adequate iodine intake, and excessive iodine intake. At baseline, a higher prevalence of thyroid antibodies in the older age group, especially TgAb, presented in the community with excessive iodine intake. This differs from previous, independent reports (8, 15). In the older age group, subjects from Jutland with long-standing low iodine intake (MUI 38 μg/liter) were twice as likely to have thyroid antibodies than individuals in Iceland with relatively high iodine intake (MUI 150 μg/liter) (15). In another study, thyroid antibodies were more common in a moderate iodine deficiency area (MUI 45 μg/liter) than an area with mild iodine deficiency (MUI 61 μg/liter) (8). This inconsistency may be related to the discrepancy of iodine intake in the areas involved because our result was from an area with long-standing iodine excess (MUI > 500 μg/liter) as described previously (12, 16). Together, these studies might indicate that both extremely low and high iodine intake levels correlate with a high tendency for thyroid autoimmune abnormalities, with a U-shaped distribution curve. In fact, as seen in several animal strains, mild iodine deficiency might partially protect against autoimmune thyroid disease (17).

To our knowledge, the incidence of thyroid autoantibodies in the general population has only been previously reported by the 20-yr Whickham survey (10). In that survey, antibodies were more frequent in women than men, and there was no age-related distribution. However, in the present study, TgAb, but not TPOAb, developed more frequently in women than men. These differences might be due to the relatively short interval between the baseline and follow-up studies and the long period needed for development of positive antibodies. Consistent with the baseline study, the higher incidence of TgAb was associated with higher iodine intake, indicating that chronically high iodine intake may increase the likelihood

TABLE 2. Five-year cumulative incidences of thyroid autoantibody-positive individuals

		Panshan, mild ID	Zhangwu, iodine more than adequate	Huanghua, iodine excess	All
Male	Follow-up, n	192	261	180	633
	Incidence of positive TPOAb, n (%)	1 (0.25)	10 (3.83)	5 (2.78)	16 (2.53)
	Incidence of positive TgAb, n (%)	2 (1.04)	7 (2.68)	5 (2.78)	14 (2.21)[a]
	Incidence of positive TPOAb and TgAb, n (%)	0	3 (0.38)	3 (1.67)	6 (0.95)
	Incidence of positive TPOAb and/or TgAb, n (%)	3 (1.56)	14 (5.36)	7 (3.89)	24 (3.79)
Female	Follow-up, n	529	729	490	1748
	Incidence of positive TPOAb, n (%)	14 (2.65)	23 (3.16)	14 (2.86)[b]	51 (2.92)
	Incidence of positive TgAb, n (%)	19 (3.59)	29 (3.99)	29 (5.86)[b]	77 (4.41)[a]
	Incidence of positive TPOAb and TgAb, n (%)	4 (0.76)	9 (1.23)	6 (3.33)	19 (1.09)
	Incidence of positive TPOAb and/or TgAb, n (%)	29 (5.48)	43 (5.9)	37 (7.55)	109 (6.24)
Total	Follow-up, n	721	990	670	2381
	Incidence of positive TPOAb, n (%)	15 (2.08)	33 (3.84)	19 (2.84)	67 (2.81)
	Incidence of positive TgAb, n (%)	21 (2.91)[c]	36 (3.64)	34 (5.07)[c]	91 (3.82)
	Incidence of positive TPOAb and TgAb, n (%)	4 (0.55)	12 (1.21)	9 (1.34)	25 (1.05)
	Incidence of positive TPOAb and/or TgAb, n (%)	32 (4.44)	57 (5.76)	44 (6.57)	133 (5.59)

n, Number of cases; ID, iodine deficiency.

[a] The overall incidence of TgAb-positive individual was significantly higher in females than males ($P = 0.036$).

[b] In the community of iodine excess, the incidence of TgAb-positive females was significantly higher than TPOAb-positive females ($P = 0.019$).

[c] Incidence of TgAb-positive subjects in a region with iodine excess was higher than that in a region with mild ID ($P = 0.038$).

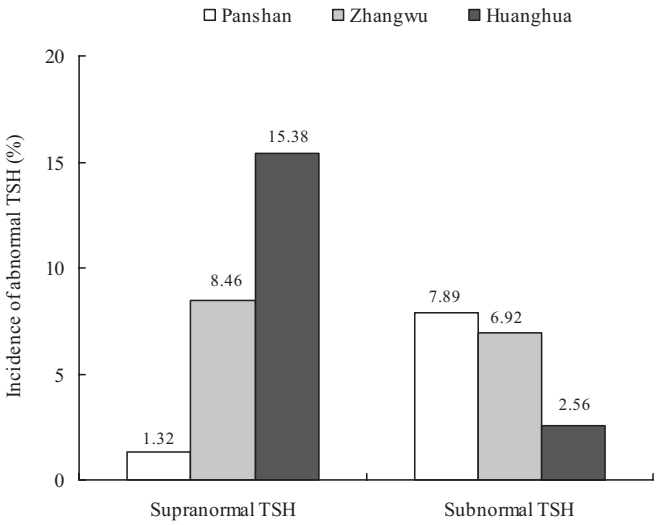

FIG. 3. Five-year cumulative incidences of abnormal TSH levels in euthyroid subjects that were TPOAb and/or TgAb positive at the baseline study in three cohorts. Panshan, Zhangwu, and Huanghua were areas with mild iodine deficiency, more than adequate iodine intake, and excessive iodine intake, respectively. The cumulative incidence of supranormal TSH increased with greater iodine intake ($P = 0.010$ among the three cohorts). Although the incidence of subnormal TSH decreased with increasing iodine intake, no significant difference was observed among the three cohorts.

of becoming TgAb positive. Interestingly, increased positive TgAb was also reported in an area after iodine prophylaxis (18). The mechanism behind this phenomenon may be explained, to a partial extent, by some experimental studies. These studies have shown that thyroglobulin combined with high iodine enhances its antigenicity and promotes lymphocyte proliferation (19–21). Furthermore, nearly 100% of genetically susceptible animals fed a high-iodine diet became TgAb positive (22–25).

Nearly three of four euthyroid subjects with positive antibodies remained antibody positive, especially subjects with high antibody levels. Carrying positive thyroid antibodies related to developing hypothyroidism. In the present study, 8% of euthyroid subjects with positive antibodies at baseline developed elevated TSH 5 yr later, more frequently than those without antibodies. Comparable data were also shown in the Whickham surveys and the 10-yr Zoetermeer study (10, 26, 27). Iodine seemed to play an important role in the positive antibody-related hypothyroidism. In the baseline study, the percentage of elevated TSH level in subjects that were either TPOAb or TgAb positive was much higher in the more than adequate iodine intake Zhangwu area and excessive iodine intake Huanghua area than in the mild iodine-deficient Panshan area. This was confirmed in the follow-up study; the incidence of hypothyroidism in antibody-positive subjects in the Huanghua and Zhangwu communities was nearly 11- and 6-fold higher than that of the Panshan community, respectively. All the above data confirmed that increased iodine intake was a risk factor for autoimmune prone subjects to develop hypothyroidism.

In line with other studies in which sensitive assay methods were applied to measure thyroid antibodies (8, 9, 28), TPOAb and TgAb were measured with the same frequency in our cross-sectional study and appeared in parallel; subjects with one antibody easily developed the other. Accordingly, a common mechanism might be implicated behind the generation of the two antibodies. As in previous population-based studies (9), this survey showed that TPOAb was associated with abnormally low and high TSH concentrations. However, the significance and function of TgAb remains controversial (9, 18). Several cross-sectional studies have shown that elevated TgAb alone is not strongly associated with elevated TSH levels (9, 28). However, our follow-up study showed that positive TgAb and positive TPOAb, either alone or combined, could significantly increase the cumulative incidence of thyroid dysfunction, indicating both of them should be considered as risk factors for thyroid dysfunction. We also discovered that only TgAb-positive subjects from areas with relatively high iodine intake were more likely to develop hypothyroidism. Thus, we supposed that positive TgAb was a powerful supplement for TPOAb in its roles of promoting and predicting autoimmune thyroiditis, at least for the population exposed to more than adequate or excessive iodine intake. Given that more than 50% of TPOAb-positive subjects were also TgAb positive and that the link between TPOAb and thyroid dysfunction is much clearer, including TgAb in the routine autoimmune thyroid diseases diagnosis, will require further investigation and a cost-effectiveness analysis.

In conclusion, we studied TPOAb and TgAb in the general population with different iodine intakes. We found that the two antibodies presented frequently in parallel and seldom disappeared; presence of these antibodies was associated with the development of thyroid dysfunction, particularly hypothyroidism. Our study also indicated that excessive iodine intake might promote thyroid autoimmunity and that it was a risk factor for autoimmune-prone subjects to develop hypothyroidism.

TABLE 3. The incidence of abnormal TSH in euthyroid subjects with or without thyroid autoantibodies in 1999

	Numbers in 1999	TSH greater than 4.8 mIU/liter in 2004, n (%)	TSH less than 0.3 mIU/liter in 2004, n (%)
Without antibodies	2381	35 (1.47)	44 (1.85)
Only TPOAb positive	88	6 (6.82)[a]	6 (6.82)[a]
Only TgAb positive	100	8 (8)[a]	5 (5)[a]
Both TPOAb and TgAb positive	96	10 (10.42)[a]	6 (6.25)[a]
Either TPOAb or TgAb positive	284	24 (8.45)[a]	17 (5.99)[a]

[a] Compared with subjects without antibodies, $P < 0.0001$.

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