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DYNAMIC CHANGES OF TRAb AND TPOAb AFTER RADIOIODINE THERAPY IN GRAVES' DISEASE

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Abstract

Context. To analyze the dynamic changes of serum thyrotrophin receptor antibody (TRAb) and thyroid peroxidase antibody (TPOAb) in Graves' disease (GD) patients before and after radioactive iodine (RAI) treatment and to investigate if TRAb and TPOAb play a role in the occurrence of early hypothyroidism after ^{131}I therapy for Graves' hyperthyroidism.

Subjects and Methods. A total of 240 patients newly diagnosed with GD were selected to study. A clinical and laboratory assessment was performed before and at 3, 6, and 12 months after ^{131}I therapy. Chemiluminescent immunoassays were used to detect serum free triiodothyronine (FT3), free thyroxine (FT4), sensitive thyroid-stimulating hormone (TSH) and TPOAb concentration. Radio-receptor assay was used to measure serum TRAb concentration. According to the early onset of hypothyroidism in a year after RAI therapy, patients were divided into early hypothyroidism group (group A) and non-early hypothyroidism group (group B).

Results. In both groups, serum TRAb and TPOAb increased at 3 months, reached the highest level at 6 months and returned to the baseline at 12 months after RAI therapy. TRAb showed a significant difference between the two groups at 6 months ($P<0.01$). Serum TPOAb in group A was higher than that in group B before and at 3, 6, 12 months after RAI therapy ($P<0.05$).

Conclusions. Serum TRAb and TPOAb are closely related to the occurrence of the early hypothyroidism, and play an important role in judging prognosis after ^{131}I treatment in Graves' disease.

Key words: Graves' disease, radioiodine therapy, hypothyroidism, thyrotrophin receptor antibody (TRAb), thyroid peroxidase antibody (TPOAb).

INTRODUCTION

Graves' disease (GD) is an autoimmune endocrine diseases and organ-specific disorders with a strong female-to-male predominance (1). The reported incidence of GD is up to 80/100 000 per year in women and 8/100 000 per year in men (2). GD is a clinical form of autoimmune thyroid diseases (AITD) characterized by thyroid-related autoantibodies (3). Hyperthyroidism with diffuse goiter is the most common result of GD, which is responsible for approximately 50-60% of the cases (4).

Current treatment approaches including antithyroid medications, thyroid ablation with surgery and radioactive iodine (RAI) have been widely used for more than five decades (1, 5). ^{131}I treatment is a safe and highly cost-effective therapeutic option devoid of high relapse rate (6, 7). Hypothyroidism seems an inevitable side effect of RAI for hyperthyroidism (8). Hypothyroidism after RAI therapy for GD may be influenced by many factors, such as ^{131}I dosage, age, gender, size of the thyroid gland, initial FT4, FT3, TSH levels, radioactive iodine uptake, duration of disease, administration of antithyroid drugs and the presence of thyroid antibodies (9, 10).

Thyrotrophin receptor antibody (TRAb) and thyroid peroxidase antibodies (TPOAb) are among the best characterized autoantibodies in GD (11). Thyrotrophin receptor antibody (TRAb) which stimulates and activates the TSH receptor (TSHR) plays an extremely important role in the occurrence and development of GD. The TRAb assay not only aids in diagnosis but also helps guide treatment in some patients (12). TRAb detection in GD became more sensitive with assay improvement in the last decades (13). But practical application of testing for TRAbs in clinic remains the subject of controversy. It is known that TPOAb were significantly associated with hypo- or hyperthyroidism (14). Since thyroglobulin antibody (TGAb) reflected the autoimmune impairment of thyroid gland, showing the same clinical significance like TPOAb. Both TGAb and TPOAb had a similar concentration pattern in the pathogenesis and treatment of hyperthyroidism (15, 16). Thus, due to the limitation of research funds, we merely measured and statistically analyzed TRAb and TPOAb in a group of patients with

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Graves' hyperthyroidism who developed *versus* a group of patients who did not develop early hypothyroidism after ^{131}I therapy.

MATERIALS AND METHODS

Subjects

Two hundred and forty patients with newly diagnosed Graves' hyperthyroidism aged 18-72 years were received by Endocrinology department of Linyi People's Hospital between March, 2011 and October, 2013. All study subjects gave their informed consent to the study, which was approved by the local ethical committee. All patients conformed to the diagnostic and treatment criteria of thyroid disease in China (2009). The clinical diagnosis and treatment for hyperthyroidism in the setting of disease was based on an algorithm of presenting symptoms, laboratory analysis of thyroid function, B ultrasonic imaging examination of thyroid volume and emission computed tomography (ECT) imaging of thyroid texture (17, 18).

The following exclusion criteria were applied: Firstly, patients receiving anti-thyroid drugs or surgery therapy before ^{131}I treatment were be excluded as well as patients receiving corticosteroids two months previously, were excluded. Thirdly, patients with other autoimmune diseases, co-existent gravidity or co-existent tumor were excluded. Finally, patients with severe proptosis, Graves ophthalmopathy, severe heart diseases, or poor liver and kidney function were excluded.

Radioactive iodine (RAI) therapy

Males aged 20–70 years (n = 43) and females aged 18-72 years (n = 197) were treated with radioactive iodine-131 (^{131}I) alone. The ^{131}I dose (mCi) was equal to thyroid mass (g) multiplied by ^{131}I dose of per gram thyroid tissue (μCi/g) and divided by 24h maximal ^{131}I uptake rate. Then μCi was converted into mCi. The radioactive iodine dose of per gram of thyroid tissue was determined based on the course of the disease, thyroid size, gland texture, age. Subjects were followed up for a year after ^{131}I therapy to assess if early hypothyroidism has occurred.

Serum assay

The venous blood samples were collected in the morning before ^{131}I therapy and after 3, 6, 12 months from ^{131}I therapy. Serum was kept frozen at -20˚C until measurements. Serum circulating FT3, FT4, TSH, TPOAb were detected by chemiluminescence immunoassay analyzer Elecsys 2010 using special auxiliary reagents (Roche) (6). Serum TRAb was measured by radiation receptors method with assay kit (Union-med, China). Normal ranges of these parameters were as follows: FT3 (3.5-6.5pmol/L), FT4 (11.5-22.7pmol/L), TSH (0.55-4.78 mIU/L).

Study design

According to the occurrence of early hypothyroidism after ^{131}I therapy for Graves' hyperthyroidism in a year, 240 patients were divided into two groups: early hypothyroidism group (group A, n = 134) and non-early hypothyroidism group (group B, n = 106). For Graves' disease, laboratory tests showed high values of FT4 and FT3, low levels of TSH. For hypothyroidism, FT3 and/or FT4 were below normal ranges, or only elevated TSH was higher than normal range, with or without signs and symptoms of clinical hypothyroidism.

Statistical analysis

Statistical analysis was performed with SPSS software version 13.0. Data was presented as means ± standard deviation. One-way ANOVA was used to detect multivariate significance. And t test was used to compare significant differences between two groups. P values < 0.05 were considered significant.

RESULTS

Baseline characteristics of patients after ^{131}I radiotherapy

All 240 patients who received ^{131}I therapy stratified into two groups according to the early occurrence of hypothyroidism after treatment in a year. 134 patients were in early hypothyroidism group (group A), and 106 patients were in non-early hypothyroidism group (group B). To exclude other influence factors for hypothyroidism occurrence after RAI therapy of GD's hyperthyroidism, baseline clinical data of the two groups of patients was analyzed statistically. There were no significant differences compared between the two groups in the following aspects: age, course of disease, 24h maximal ^{131}I uptake rate, thyroid weight, ^{131}I dose (P > 0.05) (Table 1).

Clinical and laboratory results of patients' serum before ^{131}I therapy

After dividing the patients into group A and group B according to hypothyroidism occurrence in a year following RAI therapy, we compared the serum

concentrations of FT3, FT4, TSH and TRAb before ^{131}I therapy between the two groups. And the level of FT3, FT4, TSH and TRAb had no obvious differences (P > 0.05) (Table 2).

TRAb analysis before and after ^{131}I

To further investigate the relationship between hypothyroidism occurrence and TRAb, we compared the serum TRAb before and at 3 months, 6 months and 12 months after ^{131}I radiotherapy. Serum TRAb levels showed dynamic changes which increased at 3 months, elevated to peak at 6 months and decreased to baseline at 12 months, especially in group A (Fig. 1). TRAb concentration in group A showed significant differences at 6 months after ^{131}I therapy *versus* before^{131}I therapy (P < 0.01), *versus* at 3 months from ^{131}I therapy (P < 0.05) and *versus* 12 months from ^{131}I therapy (P < 0.05). On the contrary, serum TRAb showed no statistical difference between the four time points in group B (P > 0.05). In addition, serum TRAb at 6 months after ^{131}I therapy in group A was significantly higher than that of group B (P < 0.01). There were no statistical differences compared between the two groups at other time points (P > 0.05) (Fig. 1).

TPOAb analysis before and after ^{131}I

Serum TPOAb before ^{131}I therapy in the group A was significantly higher than that of group B (P<0.05) (Table 2). After RAI therapy, serum TPOAb showed similar variation trend to serum TRAb. But it increased at 3 months, elevated to peak at 6 months and decreased to baseline at 12 months in both groups. Serum TPOAb was significantly higher in group A than group B before and after ^{131}I therapy (P < 0.05) (Fig. 2).

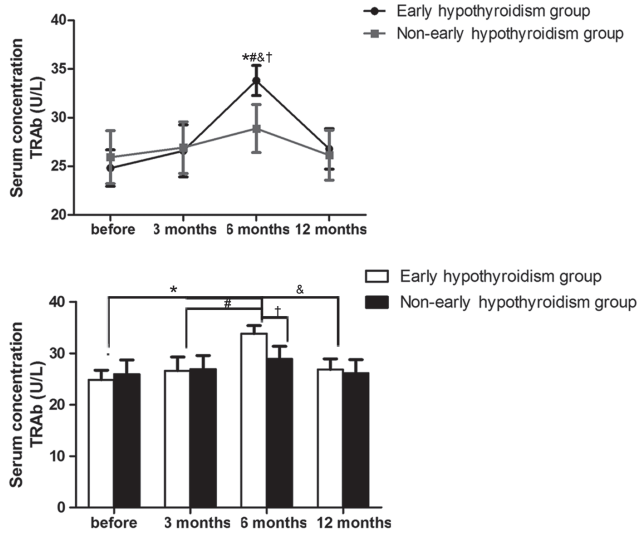

Figure 1. Serum concentration of TRAb before and after radioactive iodine therapy. In the early hypothyroidism group. *P<0.01 represents for TRAb at 6 months after ^{131}I *versus* before RAI; #P <0.05 *versus* 3 months after RAI; &P <0.05 *versus* 12 months after RAI, and †P <0.01 represents for TRAb at 6 months after RAI *versus* non-early hypothyroidism group.

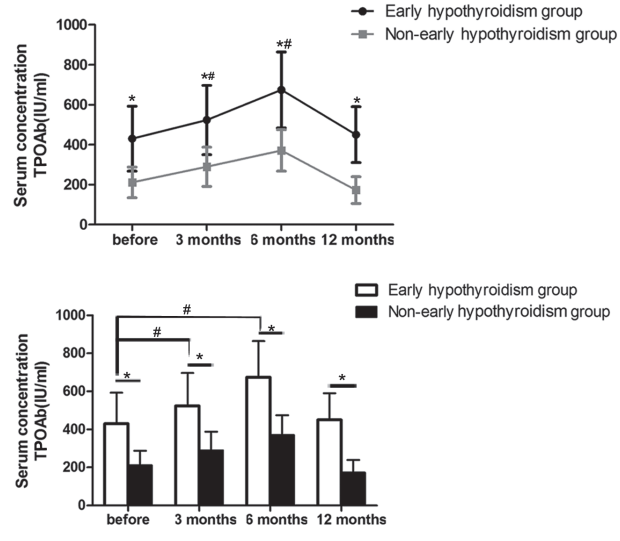

Figure 2. Serum concentration of TPOAb before and after radioactive iodine therapy. *P <0.05 represents for TPOAb in the early hypothyroidism group *versus* non-early hypothyroidism group; #P<0.05 represents for TPOAb at 3 months and 6 months after RAI *versus* before RAI.

Table 1. Comparison of baseline clinical data beweeen groups

Group	N (M/F)	Age (y)	Course of disease (m)	24h maximal ^{131}I uptake rate (%)	Thyroid weight (g)	^{131}I dose (mCi)
Early hypothyroidism group	134 (19/115)	39.01± 11.42	11.4 ± 8.9	78.4 ± 22.7	18.37 ± 14.53	7.36 ± 2.42
Non-early hypothyroidism group	106 (24/82)	37.05 ±11.57	10.5 ± 8.5	81.2 ± 23.8	22.13 ± 16.83	7.74 ± 2.94

Table 2. Comparison of throid function and throid antibodies between groups

Group	FT3 (pmol/L)	FT4 (pmol/L)	TSH (μIU/mL)	TPOAb (IU/mL)	TRAb (U/L)
Early hypothyroidism group	21.41 ± 7.96	63.41 ± 35.23	0.10 ± 0.07	430.2 ± 162.55*	24.82 ± 1.87
Non-early hypothyroidism group	18.58 ± 11.5	54.23 ± 36.95	0.11 ± 0.08	211.4 ± 76.72	25.94 ± 2.71

*P <0.05 represents early hypothyroidism group *versus* non-early hypothyroidism group

DISCUSSION

Graves' hyperthyroidism can be treated with antithyroid drugs, radioactive iodine, or subtotal thyroidectomy. Iodine-131 (^{131}I) therapy is increasingly being used as a first-line treatment of hyperthyroidism of Graves' disease (GD) (4, 19). After ^{131}I administration, patients may become hypothyroid, euthyroid, or remain hyperthyroid (20, 21). Previous studies reported that patients with Graves' hyperthyroidism were treated with a dose of ^{131}I resulting in hypothyroidism occurrence in 6-12 months in most patients (22, 23). Early hypothyroidism can be divided to transient hypothyroidism followed by thyrotoxicosis and permanent hypothyroidism according to the final outcome (23). Most of the early hypothyroidism is temporary and could recover in 6~9 months. However, clinical studies indicated that some early hypothyroidism could be aggravated into permanent which need thyroid hormone replacement therapy longtime. Thus, all patients must be closely monitored for the development of early onset of hypothyroidism after RAI treatment (9). It is of great significance to define the mechanism of early hypothyroidism after ^{131}I treatment to predict the final outcome for guiding clinical treatment.

In the present study, we treated 240 patients (197 women and 43 men) of hyperthyroidism caused by Graves' disease with ^{131}I. The age range was 18 to 72 years, and the ^{131}I dosage range in these patients was 4.8 mCi to 10.68 mCi. Most patients after RAI therapy develop early hypothyroidism within a year. 134 patients with early hypothyroidism and 106 patients with non-early hypothyroidism were compared and analyzed. The results here showed that age, course of disease, 24h maximal ^{131}I uptake rate, thyroid weight and ^{131}I dose had no statistical difference in early hypothyroidism group compared with non-early hypothyroidism group. Thus, it is suggested that these factors were not considered as involving in the occurrence of hypothyroidism after RAI therapy for GD. In addition, FT3, FT4 and TSH have already been used to assess hyperthyroidism or hypothyroidism or euthyroidism through laboratory tests (24).

Thus, we focus on the analysis of serum TRAb and TPOAb in our work. Thyrotropin receptor antibodies (TRAbs) play an important role in Graves' disease. TRAb testing can be used to aid in the clinical diagnosis and to predict a recurrence of thyroid disease (12). Less is known about whether TRAb may be responsible for the hypothyroidism. TRAb exists as thyroid stimulating antibody (TSAb) or thyroid blocking antibodies (TBAb) in the serum targeting to thyroid-stimulating hormone receptor (TSHR) (25, 26). TSAb can be combined with TSHR on thyroid follicular cells to release thyroid hormone thyroxine (T4) and triiodothyronine (T3). Conversely, TBAb could inhibit the release of thyroid hormone.

One of the currently available methods for measuring TRAb is radioreceptor assay using I^{125} labelled TSH for clinical use. Another method is with bioassays which measure cAMP production as an indicator of TSHR stimulating or inhibiting signal for research setting (26, 27). Our study assessed TRAb level by radioreceptor method before and at 3, 6, 12 months after ^{131}I therapy. It was increasing to the peak observed at 3-6 months presumably caused by the release of thyroid antigens (28), followed by a gradual decrease until 12 months. We speculated that thyroid tissue was gradually restored and auto-antigen release was gradually reduced at 12 months after ^{131}I therapy, and radioactive iodine effect gradually disappeared resulting in the decline of TRAb. TRAb levels showed a significant difference before and after ^{131}I treatment in the early hypothyroidism group. At the end of the study, the patients who became hypothyroid presented a larger increment in serum TRAb levels than patients who were hyperthyroid or euthyroid. It is suggested that serum TRAb enhancement was involved in the occurrence of early hypothyroidism after RAI therapy. Previous studies reported that early hypothyroidism during the first year after radioiodine was related to several aspects, such as thyroid destruction, radioinduced thyroiditis and thyroid antigens release (29). Our study demonstrated that a transitory increase in TRAb may be implicated in early hypothyroidism. TRAb measured in this study included thyroid stimulating antibody (TSAb) and thyroid blocking antibodies (TBAb). Further studies are required to check whether TBAb played a role or TSAb did.

Our results also showed that TPOAb was significantly higher in early hypothyroidism group than non-early hypothyroidism group before and after RAI therapy. TPOAb seems to have a predictive value for hypothyroidism after RAI therapy of GD's hyperthyroidism. TPOAb showed dynamic changes which increased at 3 months, elevated to peak at 6 months and decreased at 12 months in both groups. It is suggested that TPOAb probably play a positive role in the development of hypothyroidism.

In conclusion, radioiodine induced hypothyroidism resulted from the increase in the amount of thyroid antibody TRAb and TPOAb. It seemed to

be one of the main mechanisms responsible for the pathogenesis of early hypothyroidism. Monitoring the level of TRAb and TPOAb could be helpful to assess prognosis and guide treatment after ^{131}I therapy in Graves' disease.

Conflict of interest

The authors declare that they have no conflict of interest concerning this article.

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