Clinical Meaning of Circulating Anti-thyroglobulin Antibodies in Differentiated Thyroid Cancer: A Prospective Study

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In recent studies of patients with differentiated thyroid cancer (DTC), an association between the persistence of tumor and the presence of circulating anti-thyroglobulin antibodies (TgAbs) have been described. The aim of the present study was to evaluate TgAb variations before and after total thyroid ablation and to correlate TgAb levels to the outcome of disease. Forty-three patients with DTC were studied (35 female, 8 male; 33 patients had papillary cancer and 10 follicular cancer). Tumor was intrathyroid in 20 cases, had spread to the lymph nodes in 19 and to the lungs in 4. All patients underwent total thyroidectomy and ^{131}I therapy, and were then treated by suppressive doses of L-thyroxine. After a mean follow-up of 3.55 yr, TgAbs became undetectable in 24 patients (all were considered tumor-free), whereas TgAbs remained elevated in 19 cases. In 5 of these 19 patients, disease progression or persistence was documented (to the lymph nodes in three and to the lungs in two). TgAb levels were higher in patients with persistent disease in comparison with those tumor-free. Serum thyroglobulin (S-Tg) results were only elevated in the two patients with persistent disease in the lungs. Our data suggest that TgAbs determination may give some additional information to the follow-up of patients with DTC: the disappearance of TgAbs after therapy seems to represent a favorable prognostic factor, while the persistence of circulating TgAbs, particularly at high levels and in the absence of detectable S-Tg, may be representative of disease.

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The accuracy of serum thyroglobulin (S-Tg) measurements, one of the most important tools for the follow-up of patients with differentiated thyroid cancer (DTC) (*1–3*), is limited by the presence of thyroglobulin antibodies (TgAbs). In a patient's serum, TgAbs may interfere with S-Tg determination, resulting in either over or underestimated S-Tg values, depending on the assay (*4,5*). Therefore, we included a TgAb determination in the follow-up of DTC patients to validate the S-Tg assay (*6,7*).

Additionally, recent observations suggest that TgAbs themselves may be useful in the follow-up of these patients (*8*). Authors have reported that the presence of circulating antithyroid antibodies during the follow-up of patients with DTC may be associated with the persistence or recurrence of disease (*8*).

In a previous study from this Center (*7*) on 1,457 patients with DTC, the prevalence of circulating TgAbs was 9.7%. That study reflected a retrospective examination of our general series of DTC patients admitted between 1967–1987, and the data regarding S-Tg and TgAb determinations were collected during the follow-up following their initial treatment. The systematic preoperative measurement of both S-Tg and TgAb levels at our center was begun in January 1986. In our previous study (*7*), we described TgAb variations before and after treatment in a small group of patients who underwent follow-up for a short time period. In the present prospective study, more detailed data regarding a consistent group of 43 TgAb-positive patients with DTC, who underwent prolonged medical surveillance, are evaluated. In particular, we took into consideration the possible clinical meaning of circulating TgAb variations before and after therapy in these patients.

MATERIALS AND METHODS

From January 1986 to March 1989, 384 consecutive newly diagnosed DTC patients were referred to our center. Forty-three patients (11.2%) (35 female and 8 male; age range, 18–65 yr, mean ± s.d.: 47±13 yr) had detectable circulating TgAbs at diagnosis before therapy was begun. Histologic examination revealed papillary tumor in 33 patients and follicular tumor in 10 patients. Tumor was intrathyroid in 20 patients, had spread to cervical lymph nodes in 19 and to both cervical lymph nodes and the lungs in the other 4. Tumor results were associated with autoimmune chronic thyroiditis in five patients, to a longstanding multinodular goiter in nine patients and to uninodular goiter in the remaining patients. With the aim of obtaining a follow-up period sufficient for a judgment of remission or persistence of

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disease after therapy, only patients who had entered the study before March 1989 were evaluated for this study. Follow-up ranged from 2 to 5.4 yr, mean ± s.d.: 3.55 ± 1.06 yr, median 3.5 yr.

All patients underwent total thyroidectomy. Lymphoadenectomy was done in cases with macroscopic tumor invasion. Then, within 8–10 wks after surgery, a total body scan (TBS) was performed 72 hr after a tracer dose of ^{131}I (185 MBq) was given to detect thyroid remnants or metastases. All patients received one or more therapeutic doses of ^{131}I. The therapeutic dose was 3.7 GBq in patients with remnants in the thyroid bed and 5.55 GBq in patients with cervical lymph node metastases. Dosage ranged between 5.55 and 22.2 GBq in patients with lung metastases. All patients were treated with suppressive doses of L-thyroxine (2–2.5 μg b.w.). L-thyroxine doses were periodically modulated on the basis of serum thyroid hormones and TSH measurements.

The follow-up schedule consisted of a clinical examination, chest x-rays, neck high-resolution echography, which was associated with fine-needle aspiration cytology in doubtful cases, liver echography and S-Tg and TgAb determinations every 6–12 mo. In addition, in some doubtful cases, neck-mediastinum CT scans and/or radionuclide bone scans and/or bone x-rays were performed. With regard to the total body scan, in all patients a total body scan control was performed within 2 yr of the initial treatment. In addition, in 6 of 14 patients with a follow-up longer than 4 yrs and with persistence of circulating TgAbs, another total body scan control was performed. For the four patients with proven pulmonary metastases, the first total body scan control was made within 3–6 mo from therapy, and following scans were performed at least yearly.

S-Tg was measured by the immunoradiometric (IRMA) method (HTGK-Sorin, Italy). The interassay variation coefficient (VC) was 6.5%, the intra-assay VC was 3.1%. The cut-off limit in our laboratory, to distinguish pathological from nonpathological S-Tg values, was 3 ng/ml. TgAb serum levels were measured by means of a radioimmunoassay (RIA) method (Biodata, Italy). The interassay VC was 6.7% and the intra-assay VC was 6.3%. TgAb levels below 50 U/ml were considered negative. Before 1986 we routinely used a passive hemoagglutination technique with tanned erythrocytes (Wellcome, UK) for TgAb determination. The cut-off of 50 U/ml with the RIA method was found to correspond to a titer of 1:160 with the hemoagglutination technique.

Total triiodothyronine was measured by RIA (normal values 80–200 ng/dl), total thyroxine by fluorescence polarization immunoassay (normal values 4.5–12 μg/dl), free thyroxine by RIA (normal values 0.8–2.3 ng/dl) and TSH by IRMA (normal values 0.2–4 μU/ml).

Statistical analysis was done using Student's t-test. A p value of <0.05 was considered significant. Data were expressed as mean ± s.d.

RESULTS

For the 43 DTC patients, TgAb levels before therapy and at last control, after a mean 3.55 yr follow-up period, are shown in Figure 1. Pre-therapeutic values of circulating TgAbs were not correlated to tumor extension or to disease outcome after therapy (TgAb levels were 445.0 ± 250.6 U/ml in patients with intrathyroid tumor, 418.4 ± 236.1 U/ml in cases with lymph node metastases and 473.7 ± 277.2 U/ml in cases with lung metastases).

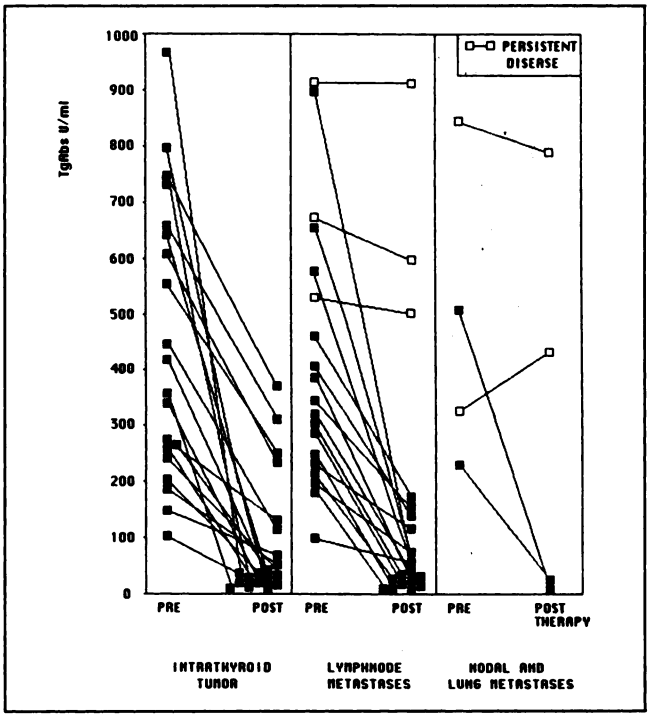

FIGURE 1. TgAb variations before and after therapy (last control) in patients with differentiated thyroid cancer.

In 23 patients, TgAbs became undetectable within 6–12 mo after therapy. These patients, including eight with a follow-up longer than 4 yrs, were considered to be tumor-free on the basis of clinical, scintigraphic, echographic, radiologic and laboratory findings. In the other 19 patients, TgAbs remained detectable, even if, after therapy, a wide range of variations were observed. In two of these patients, a persistence of disease in the lungs was found, and in three other patients a cervical lymph nodal persistence was documented. The other 14 patients from this group, including five with a follow-up longer than 4 yr, were considered to be tumor-free. TgAb levels were significantly higher in patients with persistent disease in comparison with patients considered tumor-free (653 ± 196 versus 163 ± 95 U/ml, respectively; $p < 0.01$).

S-Tg levels remained elevated during the follow-up only in the two patients with persistent disease in the lungs. In particular, during suppressive hormonal therapy, S-Tg levels were 52 and 308 ng/ml, 3.6 and 4.8 yr after therapy, respectively. During hormonal withdrawal, S-Tg levels rose in both patients up to 106 and 440 ng/ml, respectively. S-Tg levels were less than 3 ng/ml in the other 41 patients, both during and after withdrawal of hormonal therapy.

TgAb levels did not show any significant variation after hormone withdrawal in comparison with the values detected during L-thyroxine, both in patients considered to be tumor-free and in those with proven metastases.

DISCUSSION

A finding of circulating TgAbs is not uncommon in DTC patients, with a reported prevalence ranging from 2% to 15% (*8–10*). In a large series of 1,457 patients from our center (*7*), the prevalence of circulating TgAbs was 9.7%.

For many years, TgAb determination has been utilized to verify the diagnostic accuracy of the S-Tg assay. The presence of circulating TgAbs may distort S-Tg values in RIA S-Tg assays, resulting in either over or underestimation (*4,5*). More recently, by using IRMA methods for S-Tg assay, it has been demonstrated that the presence of circulating TgAbs invariably results in an underestimation of S-Tg values (*4,5,7*). Consequently, some authors have suggested that IRMA S-Tg, despite the presence of TgAbs, maintains its value as a tumoral marker even with lower sensitivity (*6,7*). It has also been recently observed that the presence of circulating antithyroid antibodies in patients previously treated for DTC may be associated with a relapse of disease, implying that TgAbs themselves could play some clinical role in DTC patients (*8*).

Our data show that the persistence of detectable circulating TgAbs, particularly if at high levels, may suggest the presence of metastatic tissue in some patients. It should be noted, however, that most of our patients with persistently circulating TgAbs after therapy were considered tumor-free on the basis of currently available diagnostic techniques. This observation appears difficult to explain. Possible hypotheses, however, may be made. It cannot be excluded, for example, that in these patients some microfoci of metastatic tissue, not shown by currently available diagnostic techniques, may produce Tg, and so provide the immune system with a continuous supply of antigen. At the same time, S-Tg measurement in these patients may be lowered because of the presence of TgAbs. If this is the case, patients with persistent circulating TgAbs should be considered at risk. An alternative hypothesis is that some lymphocytic memory cells, responsible for the TgAb synthesis, maintain their ability to supply antibodies for a prolonged period. In our series, five of six patients with a follow-up of longer than 4 yr and with persistent TgAbs in their serum were considered to be tumor-free. Nevertheless, we feel that these patients should be followed carefully. It should also be emphasized that in our series, all patients in whom circulating TgAbs became undetectable after therapy were considered tumor-free. Thus, the disappearance of circulating TgAbs after therapy seems to represent an important favorable prognostic factor.

In conclusion, while it seems appropriate to note that our observations on the clinical role of circulating TgAbs in DTC involve a relatively small number of patients, TgAb determination appears useful in the follow-up of patients with DTC, not only to validate the S-Tg assay, but because TgAbs themselves may provide additional clinical information for the follow-up of DTC patients.

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REFERENCES

1. Van Herle AJ, Uller RP. Elevated serum thyroglobulin. A marker of metastases in differentiated-thyroid carcinomas. *J Clin Invest* 1975;56:272–277.
2. Blahd WH, Drickman MV, Porter CW, Hill VA, Baumgartner WA. Serum thyroglobulin, a monitor of differentiated thyroid carcinoma in patients receiving thyroid hormone suppression therapy: concise communication. *J Nucl Med* 1984;25:673–676.
3. Pacini F, Lippi F, Formica N, et al. Therapeutic doses of iodine-131 reveal undiagnosed metastases in thyroid cancer with detectable serum thyroglobulin levels. *J Nucl Med* 1987;28:1888–1891.
4. Mariotti S, Cupini C, Giani C, et al. Evaluation of a solid-phase immunoradiometric assay (IRMA) for serum thyroglobulin: effect of anti-thyroglobulin autoantibodies. *Clin Chim Acta* 1982;123:347–355.
5. Feldt-Rasmussen U, Schlumberger M. European interlaboratory comparison of serum thyroglobulin measurement. *J Endocrinol Invest* 1988;11:175–181.
6. Schlumberger M, Fragu P, Gardet P, Lumbroso J, Violot D, Parmentier C. A new immunoradiometric assay (IRMA) system for thyroglobulin measurement in the follow-up of thyroid cancer patients. *Eur J Nucl Med* 1991;18:153–157.
7. Rubello D, Girelli ME, Casara D, Piccolo M, Perin A, Busnardo B. Usefulness of the combined antithyroglobulin antibodies and thyroglobulin assay in the follow-up of patients with differentiated thyroid cancer. *J Endocrinol Invest* 1990;13:737–742.
8. Pacini F, Mariotti S, Formica N, et al. Thyroid antibodies in thyroid cancer: incidence and relationship with tumor outcome. *Acta Endocrinol* 1988;119:373–379.
9. Amino N, Pysher Y, Cohen E, De Groot LJ. Immunological aspects of human thyroid cancer. *Cancer* 1975;36:963–970.
10. De Groot LJ, Hoye K, Refetoff S, Van Herle AJ, Asteris GT, Rochman H. Serum antigen and antibodies in the diagnosis of thyroid cancer. *J Clin Endocrinol Metab* 1976;45:1220–1226.