TSH Receptor Antibody Measurements and Prediction of Remission in Graves’ Disease Patients Treated with Minimum Maintenance Doses of Antithyroid Drugs

YASUYUKI OKAMOTO, SYUN-ICHI TANIGAWA*, KAZUYUKI ISHIKAWA** AND NOBORU HAMADA

Sumire Hospital, Osaka 536-0001, Japan
*Yamasa Corporation, Chiba 288-0056, Japan
**Cosmic Corporation, Tokyo 112-0002, Japan

Abstract. Prediction of remission is one of the main problems of antithyroid drug (ATD) therapy for Graves’ disease especially in patients who are treated with a minimum maintenance dose of ATD. We evaluated the ability of new sensitive TSH receptor antibody (TRAb) assays to predict remission in Graves’ patients using two commercially available kits (TRAb-CT from Cosmic Corporation and TRAb-Dyno from Yamasa Corporation), compared to the original PEG assay. When a euthyroid state was achieved for more than 6 months with methimazole 5 mg/day or propylthiouracil 50 mg/day and thereafter for three months with 5 mg every other day or 50 mg every other day, respectively, we discontinued ATD medication. One year of observation after discontinuation of ATD was completed in 71 patients (60 females, median age 43 years, range 18–71), and TRAb values from these patients were analyzed in relation to prognosis. Twenty-six (37%) of the 71 patients had relapse of thyrotoxicosis and 45 remained euthyroid. The median TRAb levels in the relapse group were significantly higher than those in the remission group ($P<0.05$). Relapse occurred in 15/51 patients negative by TRAb-CT, in 11/20 patients positive by TRAb-CT ($\chi^2 = 4.1; P<0.05$), in 11/42 patients negative by TRAb-Dyno and in 15/29 patients positive by TRAb-Dyno ($\chi^2 = 4.8; P<0.05$). By contrast, relapse occurred in 23/64 patients with negative TRAb by PEG assay and in 3/7 patients with PEG assay positive values (n.s.). All patients with TRAb-CT values of 30% inhibition or greater, or TRAb-Dyno values of 3.0 U/L or greater relapsed during the observation period. Thus, measurement of TRAb by the new sensitive assays is useful for prediction of remission in our patients.

Key words: TSH receptor, Graves’ disease, Antithyroid drugs, Thyroid, Autoantibody

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Graves’ disease [11, 12]. Recent studies from Europe suggest that the new TRAb assays are also useful for the prediction of remission of Graves’ disease treated with ATD, but that their predictive abilities are not necessarily improved compared to the original PEG assay [13–17]. Treatment regimes of Graves’ disease in Europe and the United States generally involve treatment with ATD for a fixed period (1 to 2 years) and then treatment is discontinued regardless of dosage required to maintain euthyroid status [18, 19]. The European studies cited above used this fixed period regimen. In Japan, however, patients are often treated with ATD until a euthyroid state is attained and then the dose of ATD is reduced to the minimum amount required to maintain a euthyroid state (minimum maintenance dose) [18, 20]. With this regimen, ATD doses do not reach the minimum maintenance dose in 20 percent of patients for up to 2 years after start of treatment [21]. The value of TRAb measurements in predicting remission might be different between the minimum maintenance therapy regime used in Japan and the fixed period regimes used in Europe. A recent study from Japan reported that the measurement of TRAb using the PEG assay or bioassay was not particularly helpful for predicting remission during minimum maintenance ATD therapy in Graves’ disease [22].

The aim of the current study was to evaluate the ability of the new more sensitive TRAb assays to predict remission of Graves’ disease at the end of a course of treatment with a minimum maintenance dose of ATD.

**Methods**

**Patients**

Diagnosis of Graves’ disease was based on clinical and laboratory findings (including TSH, FT3, FT4, TRAb and ultrasound), and consecutive patients were studied who had been treated with ATD in our hospital and met the criteria of: serum FT4 levels at or below normal and serum TSH levels at or above normal for more than 6 months with MMI 5 mg/day or PTU 50 mg/day, and ATD dose being further decreased to 5 mg every other day or 50 mg every other day, respectively. After three months, ATD medication was discontinued if serum FT4 levels were at or below normal and serum TSH levels were at or above normal. Seventy-five consecutive patients (63 females, 12 males; median age 45 years, range 18–71) with Graves’ disease fulfilled this criteria during the period from January to June, 2003. Serum FT4, TSH, and TRAb levels were measured at the time of discontinuation of ATD and serum FT4 and TSH levels were measured every 3 months after discontinuation of ATD for the following year. A one-year observation period was completed for 71 (60 females, 11 males; median age 43 years, range 18–71) out of the 75 patients, and the data from these patients (Table 1) were analyzed.

**Thyroid function tests**

The serum concentrations of TSH (reference range: 0.4–4.0 mU/L) and FT4 (reference range: 0.8–1.9 ng/ml) were measured by commercially available kits (TSH: ADVIA Centaur TSH-3, FT4: ADVIA Centaur FrT4, from Bayer HealthCare LLC, USA).

<table>
<thead>
<tr>
<th>Table 1.</th>
<th>Characteristics of the group of Graves’ disease patients at the time of ATD discontinuation. Median and (ranges) are shown</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Remission group</td>
</tr>
<tr>
<td>Number of patients</td>
<td>45</td>
</tr>
<tr>
<td>Male/Female</td>
<td>7/38</td>
</tr>
<tr>
<td>Age (years)</td>
<td>48 (22–71)</td>
</tr>
<tr>
<td>Duration of treatment in months</td>
<td>50 (10–180)</td>
</tr>
<tr>
<td>FT4 (ng/dl)</td>
<td>1.3 (0.9–1.8)</td>
</tr>
<tr>
<td>TSH (mU/L)</td>
<td>1.6 (0.4–4.3)</td>
</tr>
<tr>
<td>Time in months until relapse after discontinuing ATD in months</td>
<td>6 (3–12)</td>
</tr>
</tbody>
</table>

* P<0.01 for remission vs relapse group
TRAb assays

TRAb was measured by 3 different assays: the PEG assay (TRAb III from Cosmic Corporation, Tokyo, Japan); a TSH receptor autoantibody coated-tube kit from Cosmic (TRAb-CT) [23]; and the DYNOtest TRAb human kit from Yamasa Corporation, Tokyo, Japan (TRAb-Dyno) [11]. TRAb results measured by the PEG and TRAb-CT kits were expressed as percentage inhibition of 125I-TSH binding to the TSH receptor, and a cut-off value of 10% inhibition was used as recommended by the manufacturers. TRAb results with the TRAb-Dyno were expressed in units per liter of the international reference preparation NIBSC 90/672. Values below 1.0 U/L were considered to be TRAb negative as recommended by the kit manufacture.

Statistical analysis

Data were expressed as median values and were analyzed non-parametrically using the Mann-Whitney test for group comparisons. The χ² test or Fisher’s exact probability test was used for comparison of the prevalence of TRAb in the remission and relapse groups. P-values below 0.05 were considered as statistically significant.

Results

Twenty-six (37%) of the 71 Graves’ disease patients relapsed within a year of stopping ATD treatment and 45 remained euthyroid. A comparison of patient characteristics and clinical parameters indicated that the relapse group had significantly lower TSH values than the remission group at the time of stopping ATD (Table 1). Patient age, duration of ATD treatment, and FT4 were not different between the two groups.

The median TRAb levels measured in the three different assays in the relapse group were all significantly higher (P<0.05 for each assay) than those in the remission group at the time of ATD discontinuation (4.3% vs 1.9% inhibition for the PEG assay, 5.7% vs 2.0% inhibition for the TRAb-CT and 1.0 U/L vs <1.0 U/L for the TRAb-Dyno). Detailed results are shown in Figure 1. Although there was considerable overlap of TRAb values between the remission and relapse groups, all patients with TRAb-CT values of 30% inhibition or greater, or TRAb-Dyno values 3.0 U/L or greater relapsed within 12 months of stopping ATD. Patient characteristics and clinical parameters of these patients were no different from those of other TRAb positive patients.

Relapse of hyperthyroidism occurred in 23/64 (36%) patients negative for TRAb by PEG assay when ATD were stopped and in 3/7 (43%) of patients with PEG assay positive values at this time (not significantly different by Fisher’s exact probability test) (Table 2). Relapse of hyperthyroidism occurred in 15/51 patients with negative TRAb-CT values and in 11/20 patients with positive values (χ² = 4.1; P<0.05). In the case of measurement by TRAb-Dyno relapse occurred in 11/42 patients with negative values and in 15/29 patients with positive values (χ² = 4.8; P<0.05). TRAb values determined by the 2 more sensitive assays exhibited higher positive predictive values (probability of relapse among patients with a positive test: 55% for TRAb-CT and 52% for TRAb-Dyno) and negative predictive values (probability of remission among patients with a negative test: 71% for TRAb-CT and 74% for TRAb-Dyno) than TRAb measured by the PEG assay (43% and 64% positive and negative predictive values respectively), although differences in predictive values did not reach statistical significance.

As shown in Table 1, TSH values in the relapse group were lower than those in the remission group at the time of ATD discontinuation and consequently we accessed the value of a combination of TRAb and TSH...
measurements for prediction of remission. In patients with TRAb-CT values less than 10% or TRAb-Dyno below 1.0 U/L, overall remissions were 71% and 74%, respectively. Patients with TSH of 1.0 mU/L or greater showed similar remission rates, 73% and 74%, respectively, in this TRAb negative group. Therefore in patients with negative TRAb values a combination of TRAb and TSH measurement did not improve predictive values of remission. In contrast, in patients with TRAb-CT values between 10 and 30% inhibition or TRAb-Dyno values between 1.0 and 3.0 U/L, the overall remission rate was 56%. Patients with TSH of 1.0 mU/L or greater exhibited a higher remission rate in this TRAb positive group (71%) close to that in the TRAb-CT-negative or TRAb-Dyno-negative patients. However, the relationship between TSH values and remission or relapse did not reach statistical significance.

### Discussion

In this prospective study, we compared 3 different TRAb assays (the original PEG method and two new more sensitive methods), as predictors of remission in Graves’ patients who were euthyroid on a minimum maintenance dose of ATD. The relationship between TRAb negativity or positivity and remission or relapse was statistically significant for TRAb measurements with the new more sensitive methods but not with the PEG assay. Positive and negative predictive values for relapse or remission tended to be higher with the new assays than with the original assay.

Absence of TRAb measured by PEG assay has already been shown by meta-analysis to be a significant indicator of remission of Graves’ disease after ATD treatment is stopped [9]. However, measurement of TRAb using this assay was not helpful for prediction of remission in the present study. Kashiwai et al. [22] have also reported that measurement of TRAb by PEG assay or bioassay provides little additional information for predicting remission in patients being treated with minimum maintenance doses of ATD. This discrepancy in the predictive value of the original TRAb assay may be due to differences in treatment regimes. In particular in Europe and the United States fixed periods of ATD treatment for 1 to 2 yrs are generally used, whereas our patients were treated with minimum maintenance doses of ATD. Durations of ATD treatment were approximately 4 yrs (median) in our patients, and were 73.5 months (mean value) for remission group and 60.3 months for relapse group in the study from Kashiwai et al. [22]. Treatment with minimum maintenance doses of ATD for longer durations could eliminate active patients with often high TRAb values as candidates for stopping ATD treatment. This elimination of active patients might lead to less usefulness of TRAb by PEG assay for prediction of remission in Japan. Ninety percent (64/71) of our patients were TRAb negative by PEG assay at the end of a course of ATD, whereas 20% (13/64) of these PEG assay negative patients were positive by TRAb-CT and 34% (22/64) positive by TRAb-Dyno. This improved sensitivity of the new more sensitive TRAb assays appeared to be useful for prediction of remission in our patients.

### Table 2. Prognostic value of measurements by 3 different TRAb assays

<table>
<thead>
<tr>
<th>Number of patients</th>
<th>Remission group</th>
<th>Relapse group</th>
<th>Total</th>
<th>Significance</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>45</td>
<td>26</td>
<td>71</td>
<td></td>
</tr>
<tr>
<td>PEG assay 10% inhibition or less</td>
<td>41</td>
<td>23</td>
<td>64</td>
<td></td>
</tr>
<tr>
<td>PEG assay more than 10% inhibition</td>
<td>4</td>
<td>3</td>
<td>7</td>
<td>N.S.</td>
</tr>
<tr>
<td>TRAb-CT 10% inhibition or less</td>
<td>36</td>
<td>15</td>
<td>51</td>
<td></td>
</tr>
<tr>
<td>TRAb-CT more than 10% inhibition</td>
<td>9</td>
<td>11</td>
<td>20</td>
<td>$P&lt;0.05$</td>
</tr>
<tr>
<td>TRAb-Dyno less than 1.0 U/L</td>
<td>31</td>
<td>11</td>
<td>42</td>
<td></td>
</tr>
<tr>
<td>TRAb-Dyno 1.0 U/L or greater</td>
<td>14</td>
<td>15</td>
<td>29</td>
<td>$P&lt;0.05$</td>
</tr>
<tr>
<td>Negative predictive value = 41/64 (64%)</td>
<td>Positive predictive value = 3/7 (43%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Negative predictive value = 36/51 (71%)</td>
<td>Positive predictive value = 11/20 (55%)</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Negative predictive value = 31/42 (74%)</td>
<td>Positive predictive value = 15/29 (52%)</td>
<td></td>
<td></td>
<td></td>
</tr>
</tbody>
</table>

N.S.: not significant
treated by minimum maintenance therapy.

All patients with TRAb-CT values of 30% or greater, or TRAb-Dyno values of 3.0 U/L or greater relapsed within 12-months of stopping ATD. We suggest therefore that ATD treatment should be continued in these patients, or that other treatment modalities, such as surgery or radioactive iodine therapy, be considered even if the euthyroid status could be maintained with a minimum maintenance dose of ATD.

The 2 new sensitive TRAb assays we used both employed TSH receptor coated tubes and $^{125}$I-labelled TSH but the TSH receptor and TSH preparations were different. The TRAb-CT assay uses TSH and TSH receptor of porcine origin whereas, the TRAb-Dyno uses bovine TSH and recombinant human TSH receptor. Kamijo [24] reported that there were no significant differences in the sensitivity and specificity between assays based on the two types of TSH receptor preparations in the differentiation of thyrotoxicosis, and we also found that there were no differences between the value of the assays in prediction of remission at the time of ATD discontinuation.

In conclusion, measurement of TRAb, detected by new sensitive assays, but not by the original PEG assay, is useful for prediction of remission in patients with Graves’ disease who are treated with a minimum maintenance dose of ATD. The probability of relapse tends to be high in patients with highly positive-TRAb values.

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References


