

Thyroid Stimulating Antibodies, Thyroglobulin Antibodies and Serum Proteins during Treatment of Graves' Disease with Radioiodine or Propylthiouracil

ULLA FELDT-RASMUSSEN,^{1,2} KARINE BECH,¹ JOHN DATE,² PER HYLTOFT PEDERSEN,² KLAUS JOHANSEN¹ and STIG NISTRUP MADSEN¹

¹Dept. of Internal Medicine and Endocrinology F, Herlev University Hospital, Copenhagen, and ²Dept. of Clinical Chemistry, Odense University Hospital, Denmark

The relation between serum concentrations of thyroglobulin antibodies (TgAb), thyroid-stimulating antibodies (TSAb) and serum immunoglobulins during treatment of Graves' disease was studied in 36 consecutive patients treated randomly with ¹³¹I-iodine ($n = 16$) or propylthiouracil ($n = 20$). The patients were investigated before treatment was started and on seven occasions within the following year.

In the entire patient group 78% were positive for TSAb and 47% for TgAb. There was a significant correlation between TSAb and TgAb in 15 patients concomitantly positive. There were no significant changes in serum immunoglobulins during treatment in either group of patients.

In the radioiodine-treated group of patients TgAb was reduced after 1 week, whereas TSAb showed insignificant variations. After 5–10 weeks both antibodies increased, for TgAb with a median peak level 3 times above the initial concentration. Of 16 patients treated with radioiodine five developed myxoedema and four of these were positive for TgAb. There was a relation between the development of myxoedema and the ratio between increases of TSAb and TgAb. Increase in TSAb was not related to serum thyroglobulin (Tg) measured in TgAb-negative patients.

Propylthiouracil showed minor effects on the studied variables, but with lower mean values of Tg, TgAb and TSAb at the end of the observation period.

The results indicate an immunological relation between TSAb and TgAb, although differences between their courses exist in some situations.

Key words: serum immunoglobulins; thyroglobulin; thyroid autoantibodies; thyrotoxicosis.

Accepted for publication 20 October 1981

Graves' disease is now well established as an autoimmune disease of the thyroid but the pathogenesis is still unknown. During the last few years it has become evident that abnormal immunoglobulins with TSH-like capacity to stimulate the thyroid gland are present in all these patients (3, 7). Observations indicate that these immunoglobulins are antibodies directed

against the TSH complex (7), and Graves' disease has consequently been called an anti-receptor autoimmune disease. The primary defect in the immune apparatus leading to formation of these thyroid-stimulating antibodies (TSAb) has not yet been established, but both humoral and cell-mediated immunity is probably involved (7, 17).

Presented in part at the XIth Congress of the European Academy of Allergy and Clinical Immunology, Vienna 1980.

Both TSAb and anti-thyroglobulin antibodies (TgAb) have been found to show different changes during various kinds of antithyroid therapy (5, 11, 14, 15, 20, 22) involving either medical antithyroid drugs, radioiodine therapy or surgery.

The purpose of the present study was, using recently developed sensitive and quantitative techniques (3, 6) to investigate the relation between these two thyroid autoantibodies during propylthiouracil (PTU) and radioiodine treatment of Graves' disease. Furthermore, the possible correlation of these autoantibodies to serum proteins was assessed.

A preliminary report has been published (12).

MATERIAL AND METHODS

Patients. Thirty-six outpatients with Graves' disease referred consecutively to an endocrine clinic were studied. The diagnosis was based on clinical examination and laboratory evaluation as earlier described (4). The patients were part of a previously performed study, which has been described elsewhere (4, 13). Two groups comparable with respect to age and sex were investigated, and treated randomly with ¹³¹iodine ($n = 16$) or PTU ($n = 20$) (Table 1). None of the patients had previously received antithyroid treatment.

Blood specimens were drawn before treatment and at 1, 2, 3, 7, 11, 24, and 48 weeks after. Measurements of serum immunoglobulins and albumin were only performed during the first 24 weeks.

Measurement of serum constituents. Serum thyroglobulin (Tg) and TgAb were determined by radioimmunological methods (6, 9). TSAb was measured by the stimulatory effects of ammonium sulphate precipitated serum immunoglobulins on adenylate cyclase activity of human thyroid homogenate (2, 3). Serum IgG, -A, -M and albumin were quantified as previously described (18, 30). All serum specimens from one patient were run in the same assay series to reduce analytical imprecision.

Statistical methods. The within-assay analytical variations of the serum protein components were expressed as coefficient of variation (CV) based on 50 duplicate determinations in several assays.

For each protein the CV was: Albumin: $CV_A = 0.02$, IgG: $CV_A = 0.06$, IgA: $CV_A = 0.08$, TSAb: $CV_A = 0.12$, TgAb: $CV_A = 0.08$, Tg: $CV_A = 0.06$.

Pooled analytical variation PCV_A was calculated as previously described (23).

Two-way analyses of variances were applied to the normalized values as described earlier

Table 1

Characteristics of 36 patients with Graves' disease before treatment with radioiodine or propylthiouracil. No difference between groups could be demonstrated with respect to any of the mentioned characteristics. Medians and ranges are given

Patient groups according to treatment	<i>n</i>	Sex F/M	Age years	TgAb positive level kU/l ¹	TgAb negative Serum Tg µg/l	TSAb positive level % ⁵
Radioiodine ³	16	15/1	54 (40–71)	9 (10–510)	7 (17.3–52)	11 (89–185)
Propylthiouracil ⁴	20	15/5	50 (42–72)	9 (10–6160)	11 (21.7–>200)	16 (91–248)

¹ Standardized according to MRC standard A 69/93.

² Indicates number of patients accessible to measurement of serum Tg (TgAb unmeasurable i.e. < 2 kU/l).

³ The amount of radioactivity given (4–15 mCi) was calculated on basis of ¹³¹I uptake in the thyroid gland and its estimated weight.

⁴ The initial dose was 600 mg daily in three divided doses and the maintenance dose was 100–300 mg daily (median 150 mg).

⁵ Results were expressed as percent of activity in a reference serum pool. 97.5 percentile (upper limit of normal) was 108 %.

(23). The variance components were calculated according to Bliss (5).

Wilcoxon's non-parametric test for paired differences, the Mann-Whitney's U-test, Spearman's Rank correlation coefficient (Rho), and Fisher's exact test were used with a level of significance of 0.05.

The ratio between the maximal simultaneous increases in TgAb and TSAb was calculated using the following formula:

$$\frac{TgAb_t/TgAb_0}{TSAb_t/TSAb_0}$$

where *t* indicates the time of increasing level, and 0 indicates pretreatment level.

RESULTS

The occurrence of TgAb and TSAb in patients with Graves' disease before treatment is shown in Table 2, where 78% (28 of 36) were positive for TSAb and 47% (17 of 36) positive for TgAb. There was a significant correlation between the quantitative measurement of TSAb and TgAb in the 15 patients concomitantly positive (Spearman's Rho = 0.65, *P* < 0.02, Fig. 1).

The serum concentrations of immunoglobulins IgG, IgA and IgM in untreated patients did not differ from values in a control donor population (Table 3), and showed no significant changes during either PTU or radioiodine treatment, except for a significantly increased serum IgA at 12 weeks in the PTU-treated

Table 2

Patients with Graves' disease before treatment. The number of patients with thyroid-stimulating immunoglobulins and thyroglobulin antibodies are indicated. No significant difference by Fisher's exact test

		Thyroglobulin antibodies		Total number
		Positive	Negative	
Thyroid-stimulating antibodies	Positive	15	13	28
	Negative	2	6	8
	Total number	17	19	36

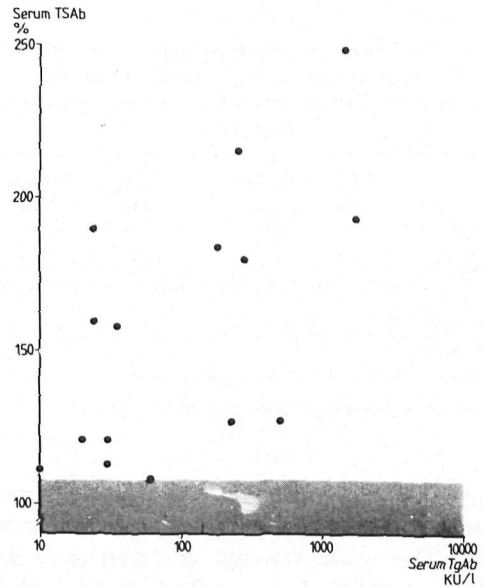


Fig. 1. Correlation between thyroid-stimulating immunoglobulins (TSAb) and thyroglobulin antibodies (TgAb) in sera of 15 patients with Graves' disease positive for both autoantibodies. Hatched area indicates 97.5-percentile of normal persons (TSAb). The detection limit of TgAb was 2 kU/L. Spearman's Rho = 0.65, *P* < 0.02.

group (*P* < 0.05 compared to start level) (Fig. 2). Serum albumin was significantly lower in patients before treatment and was normalized during treatment, with an increase to normal after 3 weeks of PTU, and 12 weeks after radioiodine.

Table 4 shows that the serum immunoglobulins and albumin followed the same pattern in the individual patients with means of CV_E from 0.06 to 0.10, of which 0.06 was accounted for by the analytical variation and only up to 0.08 was related to the individual protein concentration. The general protein variations (CV_B) ranged from 0.04 to 0.08. In contrast both TgAb and TSAb followed individual courses with higher CV_E values in all combinations, most pronounced in the radioiodine-treated group.

The sequential concentrations of TgAb, TSAb and thyroid hormones in patients positive for TgAb and treated with radioiodine (*n* = 9) and PTU (*n* = 6), respectively, are shown in Fig. 3. In the radioiodine-treated

Table 3

Serum concentrations of immunoglobulins IgG, IgA, IgM and albumin in 30 patients with Graves' disease and in a blood donor population. Medians and ranges are given for the patients

	IgG (g/l)	IgA (g/l)	IgM (g/l)	Albumin (g/l)
Patients	10.3 (6.7–17.6)	1.52 (0.42–3.48)	0.56 (0.20–1.52)	37.0 (28.0–46.5)
95%-reference* range	(6.2–13.3)	(0.40–3.43)	(0.18–1.30)	(40.0–50.0)

* Interval for non-hospitalized persons (n = 100).

group (Fig. 3b) TgAb decreased initially after treatment, in 5/9 patients to unmeasurable levels. The initial changes in TSAb were insignificant. After 5–10 weeks both antibodies gradually rose above pretreatment level, concomitantly in six of the patients. Two patients had increasing TgAb without increase in TSAb and one the opposite. Thus 8/9 patients showed at one time during the observation period a significant rise in TgAb compared to the value before treatment, peaking from 8 to 48 weeks (median 12 weeks) at a median level of 3 times (range 2–12.5 times) the start concentration.

Five out of 16 patients treated with radioiodine developed myxoedema 8 to 20 weeks after treatment, and one was negative for TgAb

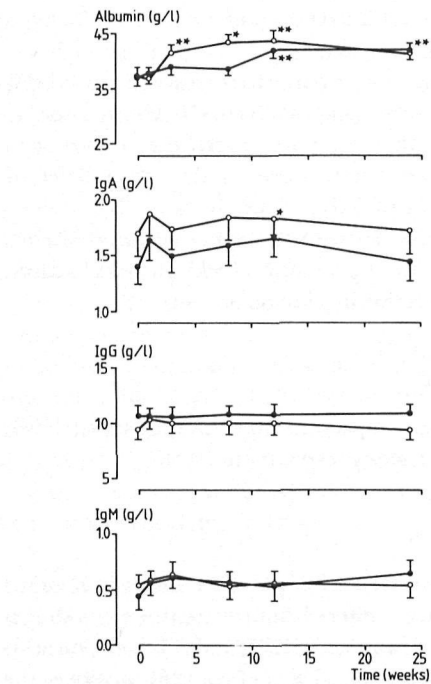


Fig. 2. Sequential changes in serum albumin and immunoglobulins A, G and M during treatment of Graves' disease with radioiodine (●, n = 16) or propylthiouracil (○, n = 20). Mean and SEM are given. * P < 0.05; ** P < 0.01 (compared to start level).

while four were positive. The latter with increasing TgAb levels prior to development of myxoedema, and in two rising TSAb levels as well. Three more patients with rising TgAb and

Table 4

Results from two-way analysis of variance of normalized combinations of serum proteins in patients with Graves' disease, giving coefficients of between-day variation (CV_B) and error term (CV_E), the latter containing both analytical variation (PCV_A) and interaction (i.e. individual variation of the proteins). CV_B and CV_E are given as mean values of group results

Patient	Treatment	n	TgAb-TSAb		TgAb-IgG		TSAb-IgG-IgA-Alb		IgG-IgA-Alb		Tg-Alb	
			CV _B	CV _E **	CV _B	CV _E **	CV _B	CV _E **	CV _B **	CV _E	CV _B	CV _E **
TgAb positive	Radioiodine*	4	0.13	0.39	0.12	0.50	0.14	0.37	0.08	0.07		
	PTU	7	0.05	0.36	0.05	0.34	0.05	0.10	0.07	0.08		
TgAb negative	Radioiodine	6							0.04	0.06	0.04	0.23
	PTU	7							0.06	0.10	0.06	0.12

* Additionally 4 patients were TgAb positive, with a transitory fall to negative values after radioiodine. The protein combination IgG-IgA-Alb gave in this group: CV_B = 0.12 and CV_E = 0.08.
** The pooled analytical variations (PCV_A) were for the combinations TgAb-TSAb: PCV_A = 0.10, TgAb-IgG: PCV_A = 0.07, TSAb-IgG-IgA-Alb: PCV_A = 0.08, IgG-IgA-Alb: PCV_A = 0.06 and Tg-Alb: PCV_A = 0.05.

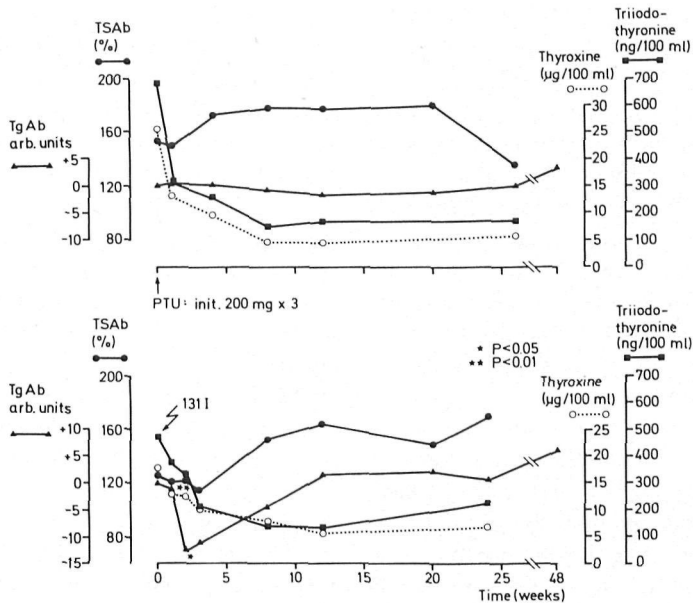


Fig. 3. Sequential changes in serum concentrations of thyroid-stimulating immunoglobulins (TSAb), thyroglobulin antibodies (TgAb) and thyroid hormones during treatment of Graves' disease with radioiodine ($n = 16$) or propylthiouracil (PTU) ($n = 20$). The arbitrary units of TgAb were calculated by dividing the increase (or decrease) by the start level.

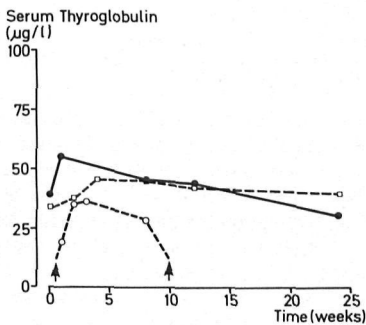


Fig. 4. Sequential changes in serum thyroglobulin during treatment of patients with Graves' disease with radioiodine (•, $n = 7$) or propylthiouracil (□---□, $n = 11$). Open circles (○---○) indicate patients treated with radioiodine ($n = 5$) with thyroglobulin antibodies (TgAb) prior to treatment, and measurable thyroglobulin in period with unmeasurable TgAb. Arrows indicate time of disappearance and reappearance of TgAb from serum (○---○).

TSAb did not develop myxoedema. The mean ratio between the factorial increases in TgAb and TSAb 8–20 weeks after radioiodine treatment was 5.3 (range 1.5–9.8, $n = 4$) in the patients who developed myxoedema, and 0.31

(0.04–1.0, $n = 4$) in those who did not ($P < 0.05$, Student's *t*-test).

In the radioiodine-treated group without TgAb, serum Tg showed a rise (Fig. 4) which was not correlated to changes in serum albumin since CV_E was high, as shown in Table 4. TSAb also increased in these patients, but at a later time than Tg, and was not correlated to serum Tg.

The patients treated with PTU showed only minor changes in serum concentrations of Tg, TgAb and TSAb during treatment, with a tendency towards lower mean values at the end of the observation period (Fig. 3a and Fig. 4).

DISCUSSION

The frequencies of TgAb and TSAb in untreated patients with Graves' disease reported in the present study are in accordance with previous reports using similar methods (3, 10, 20, 22, 31). The increasing serum concentrations following treatment of hyperthyroidism with radioiodine has been observed previously both

for TSAb (4, 20) and for TgAb (13, 16). TgAb has also been reported to rise after administration of radioiodine to euthyroid subjects (8), though in this study no increase could be seen in thyrotoxicosis (8). This difference is probably due to the use of semiquantitative methods (8, 16). Recent studies of the quantitative sequential changes in TgAb following subtotal thyroidectomy for thyrotoxicosis (11, 14) have revealed an initial decrease in the level of TgAb followed by a secondary rise within 3 months. In radioiodine-treated patients peak values were observed within 6 months.

The mechanism responsible for the rise of TgAb and TSAb, sometimes concomitant, is not known, but a number of explanations have been proposed. Destruction of thyroid tissue by surgery or radioiodine with subsequent release of antigenic products into the circulation may lead to antigenic stimulation, and radiation could also damage specific subsets of lymphocytes (20, 21). The increasing concentrations of both TgAb and TSAb after radioiodine could also be due to a reduced antigen pool in the patient, which would leave more of the antibody "free", assuming an unchanged production (7). In the majority of patients the changes have been shown to be transitory (13, 14, 16, 20, 24), which favours a reversible mechanism.

The hypothesis that the development of posttreatment myxoedema is associated with the presence of thyroid autoantibodies has been advanced (17, 24). Transient hypothyroidism following radioiodine (25) and surgery (28) appears to follow peak levels of antibodies, and an association between events is possible. In the present study no significant changes in the plasma immunoglobulins occurred. This makes a change in the general immunological response of the patients unlikely, leaving a possibility of antigen-specific actions on one or more parts of the immune system. This was not confined to a single species of thyroid antibodies since both TSAb and TgAb rose simultaneously in the majority of the patients. The rise was not correlated to the increase in serum albumin, which probably follows a return to normal protein balance (26).

A significant positive correlation between LATs and TgAb in serum has been reported (1). The patients in the present study had more frequently TSAb than TgAb, but when both were present the quantitative results showed an association.

It has been suggested that the simultaneous presence of TgAb and TSAb facilitates development of either hypo- or hyperthyroidism depending on which autoantibody is the more abundant (7). In the present study a larger factorial rise in serum TgAb compared to that of TSAb occurred in patients developing myxoedema compared to those who did not, which could appear to support the hypothesis.

An immunosuppressive action of thyrostatic drugs has recently been described (21, 27, 29). In the present study PTU did not influence concentrations of serum immunoglobulins. It therefore appears likely that the general immunosuppressive action of PTU is only moderate, or that the concentration of thyrostatic drugs in the thyroid gland (19) favours an action of antibody-producing lymphocytes present there.

ACKNOWLEDGEMENTS

The technical assistance of the staff in the Beta-laboratory of the Dept. of Clinical Chemistry, Odense University Hospital, and of Susanne Christensen is gratefully acknowledged. This work was supported by grants from P. Carl Petersen, Nordic Insulin, Novo, and Ingeman O. Buch Foundations. Ragna Riis is thanked for typing the manuscript, and Dr. Mølholm Hansen for aid in classification of the patients.

REFERENCES

1. Bastenie, P. A., Bonnyns, M. & Vanhaelst, L.: LATs and antithyroglobulin antibodies in thyrotoxicosis. In: Irvine, J. (ed.): *Thyrotoxicosis*, pp. 40–47. E. & S. Livingstone Ltd., Edinburgh and London, 1967.
2. Bech, K. & Madsen, S. N.: Human thyroid adenylate cyclase in non-toxic goitre: Sensitivity to TSH, fluoride and thyroid stimulating immunoglobulins. *Clin. Endocrinol.* 8, 457–466, 1978.
3. Bech, K. & Nistrup Madsen, S.: Thyroid adenylate cyclase stimulating immunoglobulins in thyroid diseases. *Clin. Endocrinol.* 11, 47–58, 1979.
4. Bech, K. & Madsen, S. N.: Influence of treatment with radioiodine and propylthiouracil on thyroid stimulating immunoglobulins in Graves' disease. *Clin. Endocrinol.* 13,

- 417-424, 1980.
5. Bliss, C. I.: Subsampling in relation to experimental design (Chapter 12). *Statistics in Biology*. McGraw-Hill, New York, 1967.
6. Date, J., Feldt-Rasmussen, U., Hyltoft Petersen, P. & Bech, K.: An improved co-precipitation assay for determination of thyroglobulin autoantibodies. *Scand. J. Clin. Invest.* **40**, 37-44, 1980.
7. Doniach, D. & Marshal, N. J.: Autoantibodies to the thyrotropin (TSH) receptors on thyroid epithelium and other tissues. In Talal, N. (ed.): *Autoimmunity*, pp. 621-642. Academic Press, New York, 1977.
8. Einhorn, J., Einhorn, N., Fagraeus, A. & Jonsson, J.: Hypothyroidism and humoral antibodies after radioiodine treatment of hyperthyroidism. In: Irvine, J. (ed.): *Thyrotoxicosis*, pp. 123-133. E. & S. Livingstone Ltd., Edinburgh & London, 1967.
9. Feldt-Rasmussen, U., Hyltoft-Petersen, P. & Date, J.: Sex and age correlated reference values of serum thyroglobulin measured by a modified radioimmunoassay. *Acta Endocrinol. (Kbh.)* **90**, 440-450, 1979.
10. Feldt-Rasmussen, U., Bech, K. & Date, J.: Serum thyroglobulins in patients with toxic and non-toxic goitres compared to sex- and age-matched control subjects. *Acta Endocrinol. (Kbh.)* **91**, 264-270, 1979.
11. Feldt-Rasmussen, U., Hyltoft Petersen, P., Date, J. & Madsen, C. M.: Sequential changes in serum thyroglobulin (Tg) and its autoantibodies (TgAb) following subtotal thyroidectomy of patients with preoperatively detectable TgAb. *Clin. Endocrinol.* **12**, 29-38, 1980.
12. Feldt-Rasmussen, U., Bech, K., Johansen, K., Date, J., Madsen, C. M. & Madsen, S. N.: Relation between serum thyroglobulin (Tg), its antibody (TgAb) and thyroid stimulating immunoglobulins (TSAb) in Graves' disease. *Allergol. Immunopathol.* **8**, 436, 1980.
13. Feldt-Rasmussen, U., Bech, K., Date, J., Hyltoft Petersen, P. & Johansen, K.: A prospective study of the changes in serum thyroglobulin and its autoantibody during propylthiouracil or radioiodine therapy of patients with Graves' disease. *Acta Endocrinol. (Kbh.)*. (In press).
14. Feldt-Rasmussen, U., Blichert-Toft, M., Date, J. & Christensen, C.: Serum thyroglobulin and its autoantibodies following subtotal thyroidectomy of Graves' disease. *Eur. J. Clin. Invest.* In press, 1982.
15. Gardner, D. F., Rothman, J. & Utiger, R. D.: Serum thyroglobulin in normal subjects and patients with hyperthyroidism due to Graves' disease: Effects of T₃, iodine, ¹³¹I, and antithyroid drugs. *Clin. Endocrinol.* **11**, 585-594, 1979.
16. Irvine, W. J. & Stewart, A. G.: Prognostic significance of thyroid antibodies in the management of thyrotoxicosis. In: Irvine, W. J. (ed.): *Thyrotoxicosis*, pp. 111-222. E. & S. Livingstone Ltd., Edinburgh and London, 1967.
17. Kidd, A., Okita, N., Row, V. V. & Volpé, R.: Immunological aspects of Graves' and Hashimoto's diseases. *Metabolism* **29**, 80-99, 1980.
18. Laurell, C.-B.: Quantitative estimation of proteins by electrophoresis in agarose gel containing antibodies. *Anal. Biochem.* **15**, 45-54, 1966.
19. Marchant, B., Lees, J. F. H. & Alexander, W. D.: Anti-thyroid drugs. *Pharmacol. Ther.* (B) **3**, 305-348, 1978.
20. McGregor, A. M., Petersen, M. M., Capiferri, R., Evered, D. C., Rees Smith, B. & Hall, R.: Effects of radioiodine on thyrotropin binding inhibiting immunoglobulins in Graves' disease. *Clin. Endocrinol.* **11**, 437-444, 1979.
21. McGregor, A. M., Petersen, M. M., McLachlan, S. M., Rooke, P., Rees Smith, B. & Hall, R.: Carbimazole and the autoimmune response in Graves' disease. *N. Engl. J. Med.* **303**, 302-307, 1980.
22. Mori, T. & Kriss, J. P.: Measurement by competitive binding radioassay of serum anti-microsomal and anti-thyroglobulin antibodies in Graves' disease and other thyroid disorders. *J. Clin. Endocrinol.* **33**, 688-697, 1971.
23. Nielsen, H., Hyltoft Petersen, P., Feldt-Rasmussen, U. & Binder, V.: Variations in plasma protein concentration in individuals with ulcerative colitis: analytical and biological factors. *Scand. J. Clin. Lab. Invest.* **39**, 495-502, 1979.
24. O'Gorman, P., Staffurth, J. S. & Ballentyne, M. R.: Antibody response to thyroid irradiation. *J. Clin. Endocrinol. Metab.* **24**, 1072-1075, 1964.
25. Sawers, J. S. A., Toft, A. D., Irvine, W. J., Brown, N. S. & Seth, J.: Transient hypothyroidism after iodine-131 treatment of thyrotoxicosis. *J. Clin. Endocrinol. Metab.* **50**, 226-229, 1980.
26. Shambaugh, G. E.: Chemistry and actions of thyroid hormone. Biologic and cellular effects. In: Werner, S. C. & Ingbar, S. H. (eds.): *The thyroid*, pp. 115-124. 4th ed. Harper & Row, New York, San Francisco, London, 1978.
27. Teuber, J., Maser, E., Helmke, K., Dietz, O. & Federlin, O.: Evidence of immunosuppressive effect of thyrostatic drugs in treatment of thyroid diseases. *Allergol. Immunopathol.* **8**, 437-438, 1980.
28. Toft, A. D., Irvine, W. J., McIntosh, D., Seth, J., Cameron, E. H. D. & Lidgard, G. P.: Temporary hypothyroidism after surgical treatment of thyrotoxicosis. *Lancet* **ii**, 817-818, 1976.
29. Wall, J. R., Manwar, G. L., Greenwood, D. M. & Walters, T.: The in vitro suppression of lectin induced ³H-thymidine incorporation into DNA of peripheral blood lymphocytes after the addition of propylthiouracil. *J. Clin. Endocrinol. Metab.* **43**, 1406-1409, 1976.
30. Weeke, B.: Quantitative estimation of human immunoglobulins following carbamylation by electrophoresis in antibody-containing agarose. *Scand. J. Clin. Lab. Invest.* **22**, 107-117, 1968.
31. Wilkin, T. J., Swanson Beck, J., Gunn, A., Al Moussah, M., Isles, T. E. & Crooks, J.: Autoantibodies in thyrotoxicosis: a quantitative study of their behaviour in relation to the course and outcome of treatment. *J. Endocrinol. Invest.* **3**, 5-14, 1980.

Address:

Dr. Ulla Feldt-Rasmussen
Dept. of Internal Medicine and Endocrinology F
Herlev University Hospital
DK-2730 Herlev, Copenhagen
Denmark

This document is a scanned copy of a printed document. No warranty is given about the accuracy of the copy. Users should refer to the original published version of the material.