EFFECT OF ENDOGENOUS THYROID STIMULATING HORMONE LEVELS ON THE SECRETION OF THYROID HORMONES IN MAN

By

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ABSTRACT

The effect of endogenous thyroid stimulating hormone (TSH) on the thyroid secretion of triiodothyronine (T₃) and thyroxine (T₄) was evaluated by serial determinations of serum T₃, T₄ and TSH concentrations in the following groups of patients: a) three patients submitted to surgical removal of a solitary, autonomous thyroid nodule which had completely inhibited the extranodular tissue; b) five subjects, with the same disease, in whom functional recovery of the extranodular tissue was induced by increased circulating TSH levels, produced by treatment with methimazole; c) one patient submitted to hemithyroidectomy for multinodular goitre; d) two hyperthyroid patients who had been treated with methimazole. In all these patients serum T₃ and T₄ levels progressively decreased, with a consequent progressive increase in serum TSH concentrations, leading to stimulation of the thyroid gland. During this TSH-induced stimulation of thyroid tissue, a significant positive correlation was found between the serum TSH concentrations and the corresponding ratio between the serum levels of T₃ and T₄ (T₃/T₄), both within each patient group (P < 0.001) and among all patients (P < 0.001). The same correlation also governs the relationship between the TSH and the T₃/T₄ values of 34 euthyroid control subjects and one patient with incipient hypothyroidism. These data strongly suggest that endogenous TSH can induce a preferential secretion of T₃ over T₄ by the human thyroid.

A relative hypersecretion of triiodothyronine (T₃) over thyroxine (T₄) may be observed both in mild, or incipient, hypothyroidism and in endemic goitre, as indicated by the observation of the ratio between circulating T₃ and T₄ (T₃/T₄) being above the normal range (Ingbar & Woeber 1974). The increased T₃/T₄
ratio in these conditions has been explained on the basis of enhanced thyroid stimulation by thyroid stimulating hormone (TSH) (Editorial 1971; Patel et al. 1973; Ingbar & Woerber 1974; Haibach & Avioli 1976; Schimmel & Utiger 1977), an adaptation of the thyroid to reduced iodine availability (Patel et al. 1973; Ingbar & Woerber 1974) or a combination of both mechanisms. Indeed, preferential secretion of T₃ over T₄, induced by pharmacologic amounts of TSH, has been shown both in man and animals (Matsuda & Greer 1965; Chopra et al. 1972; Vigneri et al. 1975; Lauberg 1977). Conversely, a decreased T₃/T₄ ratio in serum has been reported to be associated with reduced circulating levels of TSH (Elewant & Vermeulen 1971). Furthermore, hypophysectomy in iodine depleted animals is followed by disappearance of T₃ within the thyroid gland, while T₄ synthesis is maintained (Studer & Greer 1965).

The aim of this study was to verify the hypothesis that a preferential secretion of T₃ over T₄ in man can be related to changes in the circulating levels of endogenous TSH, within either the physiological or the pathological range. For this purpose, the reciprocal changes of TSH and of the serum thyroid hormone concentrations were monitored in patients with various thyroid diseases associated with progressively increasing endogenous TSH levels. Thus, different pathophysiological conditions were studied in order to establish whether a general pattern of the stimulating effect of endogenous TSH on thyroid hormonal secretion could be identified in man.

**MATERIAL AND METHODS**

**Patients**

A total of 12 patients with various thyroid diseases were selected for this study, including 8 patients with a solitary autonomous nodule, 2 patients with toxic goitre, 1 euthyroid patient with multinodular goitre and 1 patient with low T₄ (2.8 µg/100 ml), slightly elevated TSH (8.2 ng/ml) and normal T₃ (150 ng/100 ml) serum concentrations, but without clinical features of hypothyroidism. In this latter patient, a diagnosis of T₃ euthyroidism was postulated, as proposed by Ingbar & Woerber (1974). All patients, except one (the subject with T₃ euthyroidism), were women, aged 23 to 70 years.

The diagnosis of autonomous thyroid nodule had been made on the basis of the usual scintigraphic and laboratory findings (Ramsey et al. 1972; Carpi et al. 1977). Seven patients in this group were euthyroid at the time of the study, whereas one showed features of hyperthyroidism (patient H).

In the patients with an autonomous thyroid nodule, toxic goitre and multinodular goitre, a progressive increase in the serum level of endogenous TSH had been induced either by surgical or pharmacologic treatment, as described below.

**Surgical treatment**

Patients with an autonomous functioning thyroid nodule, even those in whom the circulating levels of T₄ and T₃ remain within the normal range, have suppression of TSH secretion with consequent inhibition of the function of the normal, extranodular,
parenchyma (Ridgway et al. 1973; Carpi et al. 1977). Following surgical excision of
the functioning nodules, and after the fall of thyroid hormone serum levels, recovery
of pituitary TSH secretion would be expected (Ridgway et al. 1973). Thus, useful
information on the effect of endogenous TSH on thyroid hormone secretion pattern
may be gathered by monitoring the phase of TSH secretion recovery with its consequent
stimulation of extranodular tissue hormonogenesis.

Three of the patients with a single "hot" nodule of the thyroid (patients A, B and
C, all clinically euthyroid) underwent surgical removal of the nodule, and their serum
concentrations of T3, T4 and TSH were measured in blood samples drawn approxi-
mately every 3 h in the first two days, then at approximately daily intervals, for a
total of more than 30 days after surgery. In order to correct for the possible effects
of body fluid changes on serum hormone concentrations the haematocrit values were
determined before surgery, 8-hourly during the first day and twice during the
following day. Histology of the excised nodules revealed follicular adenoma in all
cases.

A similar protocol for blood sampling and hormone assays was used in patient I,
who underwent hemithyroidectomy for multinodular goitre, after which we would
also have expected an important fall in circulating T3 and T4, followed by increased
TSH secretion with stimulation of the residual thyroid tissue.

Non-specific changes in thyroid hormonal pattern, induced by the surgical trauma
per se, were evaluated in 2 control patients submitted to other surgical procedures
(cholecystectomy in one case and excision of a perianal abscess in the other). In both
these patients serum levels of TSH, T3 and T4 were monitored in the same manner
as in the above patients.

**Pharmacologic treatment**

Pharmacologic blockade of nodular or thyroid hormonogenesis, induced by pro-
longed treatment with methimazole, was achieved in the remaining 5 subjects with a
solitary autonomous nodule (patients D, E, F, G and H) and in the 2 patients with
toxic goitre (patients L and M). This treatment was designed to reduce the circulating
levels of thyroid hormones to such an extent as to increase pituitary TSH secretion.
Methimazole was chosen because it does not affect the peripheral conversion of T4
into T3.

The serum T3, T4 and TSH concentrations were monitored at frequent intervals
(every 2 or 3 days) in patient D (autonomous thyroid nodule) and in patient M (diffuse
toxic goitre). The re-appearance of the extranodular thyroid tissue uptake was assessed
in patient D by repeated 99mTc-pertechnetate thyroid scans (every 10 days until the
40th day on treatment, at which time the functional recovery of the extranodular
tissue had occurred and was confirmed by a 131I scan). In all the patients with autonomous
thyroid nodules the 131I scan demonstrated the functional recovery of the extranodular
tissue. In one of these patients (patient G) methimazole administration was continued
for ten more days, and then an additional serum sample was obtained for hormonal
assay.

**Laboratory methods and analysis of the hormonal data**

Serum triiodothyronine and TSH concentrations were measured in all the samples
collected during the study by specific radioimmunoassay, using commercial kits
supplied by Sorin Biomedica (Saluggia, Vercelli, Italy). A competitive protein-binding
technique was utilized to measure the serum concentrations of T4 (Tetralute kits,
supplied by Ames, Co., Research Product Division of Miles Laboratories, Inc., Emkart,
Table 1.
The most significant values of $T_3$, $T_4$ and TSH serum concentrations before and after surgical excision of autonomous thyroid nodule.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Basal $T_3$ ng/100 ml</th>
<th>$T_3$ lowest value ng/100 ml (time after surgery)</th>
<th>Basal $T_4$ µg/100 ml</th>
<th>$T_4$ lowest value µg/100 ml (time after surgery)</th>
<th>Basal TSH ng/ml</th>
<th>Starting of TSH increase (days after surgery)</th>
<th>TSH max ng/ml (days after surgery)</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>235</td>
<td>67 (38 h)</td>
<td>6.7</td>
<td>3.2 (4 days)</td>
<td>0.84</td>
<td>5</td>
<td>2.45 (10)</td>
</tr>
<tr>
<td>B</td>
<td>200</td>
<td>85 (105 h)</td>
<td>9.0</td>
<td>3.5 (9.5 days)</td>
<td>1.65</td>
<td>4.5</td>
<td>3.2 (13)</td>
</tr>
<tr>
<td>C</td>
<td>160</td>
<td>67 (80 h)</td>
<td>5.8</td>
<td>1.3 (11 days)</td>
<td>1.10</td>
<td>10</td>
<td>7.0 (18)</td>
</tr>
</tbody>
</table>

Indiana). The normal range for serum concentrations of $T_3$, $T_4$ and TSH, determined in a group of 34 healthy euthyroid control subjects (19 women and 15 men, aged 25 to 65 years), were: 100–200 ng/100 ml, 3.5–12.2 µg/100 ml, and 1–4.5 ng/ml, respectively.

The $T_3/T_4$ ratio values utilized for the present study were those obtained for all the blood samples taken in each patient during the phase of increased serum concentration of TSH; these figures (multiplied by $10^3$ for easier handling) were then correlated with the values of the serum concentrations of TSH at corresponding sampling times. Thus, the regression lines between TSH serum levels and $T_3/T_4$ ratio values, as well as the corresponding correlation coefficients and statistical significances, were calculated for the different sets of data (individual patients and whole groups).

RESULTS

Serum TSH, $T_3$ and $T_4$ levels after surgical or pharmacologic treatment

Table 1 summarizes the main hormonal data of patients A, B and C, submitted to surgical removal of "hot" thyroid nodules, and one typical case of this group is diagrammatically shown in the upper left panel of Fig. 1.

Fig. 1.

Serum $T_3$, $T_4$ and TSH concentrations monitored in: patient A, submitted to surgical excision of solitary autonomous thyroid nodule (upper left panel); patient I submitted to hemithyroidectomy for multinodular goitre (upper right panel; the $^{131}$I thyroid scans before, on the left, and on the 28th post-operative day, on the right); patient D affected by solitary autonomous thyroid nodule treated with methimazole (lower left panel; the $^{131}$I thyroid nodule scans before treatment, on the left and on the 41st day of treatment, on the right; open circles correspond to measurements performed after withdrawal of the drug); patient M affected by toxic goitre treated with methimazole (lower right panel; in this patient the time course of the $T_3/T_4$ ratio is also shown).
Table 2.
T₃, T₄ and TSH serum concentrations before and during methimazole treatment.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Diagnosis</th>
<th>Before methimazole</th>
<th>During methimazole (20 mg/day)</th>
<th>Time on treatment</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>T₃ ng/100 ml</td>
<td>T₄ µg/100 ml</td>
<td>TSH ng/ml</td>
</tr>
<tr>
<td>E</td>
<td>Hot nodule</td>
<td>105</td>
<td>11.6</td>
<td>2.6</td>
</tr>
<tr>
<td>F</td>
<td>Hot nodule</td>
<td>225</td>
<td>10.6</td>
<td>2.2</td>
</tr>
<tr>
<td>G</td>
<td>Hot nodule</td>
<td>170</td>
<td>4.5</td>
<td>1.4</td>
</tr>
<tr>
<td>“</td>
<td>“</td>
<td>170</td>
<td>4.5</td>
<td>1.4</td>
</tr>
<tr>
<td>H</td>
<td>Hot nodule</td>
<td>400</td>
<td>14.0</td>
<td>1.0</td>
</tr>
<tr>
<td>L</td>
<td>Toxic goitre</td>
<td>550</td>
<td>19.0</td>
<td>0.9</td>
</tr>
</tbody>
</table>
The serum levels of TSH, T₃ and T₄ of these patients showed a fairly consistent pattern. In fact, in all 3 patients, both T₃ and T₄ serum concentrations decreased markedly after surgery and, depending on their different blood clearance rates, they reached hypothyroid values over an average interval of 2 days (for T₃) and 6 days (for T₄). The serum TSH concentrations remained at the basal, pre-operative levels over an average interval of 6 days after surgery and then started to rise significantly in all 3 patients. The maximum level reached by serum TSH was within the normal range in patients A and B, and above normal in patient C. In all cases the rise of serum TSH levels was followed by a progressive increase in the serum T₃ and T₄ concentrations to the normal levels.

The general pattern of the hormonal changes in the patient submitted to hemithyroidectomy (patient I, upper right panel of Fig. 1) was approximately the same as in the above patients, apart from a slower rate of decline of both T₃ and T₄. However, in this patient also, the serum TSH concentrations started to rise on the 5th day after surgery, followed by a progressive return to normal of the serum T₃ and T₄ levels.

Table 3.
Correlation coefficients and statistical significance of the correlations between TSH and T₃/T₄ ratio within each monitored patient, within the various groups of patients and among all patients.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Condition</th>
<th>r</th>
<th>P</th>
</tr>
</thead>
<tbody>
<tr>
<td>A</td>
<td>Excised hot nodule</td>
<td>0.900</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>B</td>
<td>Excised hot nodule</td>
<td>0.655</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td>C</td>
<td>Excised hot nodule</td>
<td>0.717</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td>A + B + C</td>
<td>Excised hot nodules</td>
<td>0.789</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>D</td>
<td>Hot nodules monitored during methimazole</td>
<td>0.850</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td>I</td>
<td>Hemithyroidectomy for multinodular goitre</td>
<td>0.626</td>
<td>&lt; 0.01</td>
</tr>
<tr>
<td>M</td>
<td>Methimazole treated toxic goitre</td>
<td>0.842</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>D + E + F + G + H</td>
<td>Methimazole treated hot nodules</td>
<td>0.887</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>D + E + F + G + H + M</td>
<td>Methimazole treated patients</td>
<td>0.823</td>
<td>&lt; 0.001</td>
</tr>
<tr>
<td>All patients</td>
<td></td>
<td>0.842</td>
<td>&lt; 0.001</td>
</tr>
</tbody>
</table>
The changes observed in the serum hormonal concentrations were totally unrelated to the post-operative haematocrit changes, which never exceeded 8% of the basal value. Furthermore, the two control operated patients did not show any sustained significant changes in the serum T₃, T₄ and TSH levels following surgery.

The results of the hormonal assays performed in the 7 patients receiving methimazole (5 subjects with autonomous thyroid nodules and 2 patients with diffuse toxic goitre) are shown in Table 2 (the patients not submitted to hormonal monitoring) and in Fig. 1 (the 2 subjects with hormonal monitoring: patient D, lower left panel, and patient M, lower right panel, affected respectively by a solitary autonomous thyroid nodule and by toxic goitre). The rise of the serum TSH levels in this group was considerably delayed relative to the surgically treated patients, not being detectable until the 40th and 45th day of treatment, respectively in the 2 monitored patients receiving methimazole, compared to 6 days in the surgical group (see Fig. 1 and Table 1). In the patient with autonomous thyroid nodule, the increase in serum TSH level was followed by a slight increase in the T₃ circulating level, while serum T₄ concentration reached a plateau until discontinuation of drug administration. However, in the patient with toxic goitre the increased TSH circulating levels reduced the rate of decline of serum T₃ more than that of T₄, as demonstrated by the progressive increase of the T₃/T₄ ratio, a behaviour parallel to that of serum TSH.

**Correlation between serum TSH levels and T₃/T₄ ratios**

The hormonal data utilized for this type of analysis were those obtained for all the blood samples taken in each patient during the phase of increased serum TSH concentration; the T₃/T₄ ratio values calculated for these samples were then correlated with the serum TSH concentration values at the corresponding times. The pooled data obtained in all the subjects studied showed the existence of a significant positive correlation between serum TSH concentrations and the corresponding T₃/T₄ ratio values (r = 0.842, P < 0.001). The correlation held both when the data were considered as a single group, and when the results in the individual patients were considered (see detailed data in Table 3). Fig. 2 shows the regression line calculated for all the TSH and T₃/T₄ data obtained during the phase of increased serum TSH levels in all patients submitted to the study, i.e. the 8 patients with a solitary autonomous thyroid nodule (treated either surgically or pharmacologically), one case of diffuse toxic goitre (patient M; the other patient with toxic goitre is not included in this representation because of the very high TSH and T₃/T₄ ratio values), the patient submitted to hemithyroidectomy for multinodular goitre, and the patient with T₃ euthyroidism. The mean value ± 1 sd of the
Correlation between the serum TSH concentrations and the $T_3/T_4$ ratio in all patients studied. The cross represents the mean values and the standard deviations of TSH and $T_3/T_4$ values of 34 euthyroid, control subjects. The values of patient L are not reported because of the very low T$_4$ levels (0.5 μg/100 ml) and of the very high levels of TSH (30 ng/ml).

The main finding of this study is the consistent effect produced by TSH stimulation on the thyroid hormonal secretion pattern. In fact, the close positive correlation observed between the raised endogenous serum TSH concentrations and the corresponding $T_3/T_4$ ratio values indicates that, in man, endogenously increased TSH levels are able to stimulate preferential secretion of T$_3$ over T$_4$. 

**Fig. 2.**
by the thyroid gland. Moreover, the variety of thyroid diseases in which this
effect was observed (autonomous thyroid nodules, toxic goitre, multinodular
goitre, T₃ euthyroidism) suggests that this preferential stimulation of T₃ secre-
tion over T₄ is a general effect of TSH activity. This conclusion is further
supported by the similar quantitative effects observed in the T₃/T₄ ratio values,
despite two important pathophysiologic differences: 1) the mechanism by which
endogenous TSH secretion was increased in the various patients (by surgical
operation or by pharmacologic treatment) and 2) the variable basal condition
of the thyroid tissue prior to stimulation by TSH. Preferential secretion of T₃
over T₄ was induced either when TSH activated a tissue which had been
previously inhibited (in the patients with autonomous thyroid nodules), or when
it stimulated a tissue which was already functioning (patient submitted to hemi-
thyroidectomy, and patients with diffuse toxic goitre). Furthermore, this pre-
ferential stimulation of thyroid hormonogenesis by TSH was observed to
operate, at the same level, even during the thyrostatic effect of methimazole,
which presumably acted also on the extranodular parenchyma (patients affected
by autonomous nodules) or on the whole of the thyroid tissue (diffuse toxic
goitre).

The increased T₃/T₄ ratio values observed during methimazole administra-
tion can hardly be attributed to the effect of methimazole per se, as this drug
does not seem to have a preferential thyrostatic effect on T₄ relative to T₃
(Abuid & Larsen 1974). Moreover, in the treated patients, the increased values
of T₃/T₄ ratio were observed only after the rise of serum TSH (see Fig. 1 lower
right panel), while a possible pharmacologic effect should have occurred earlier
in the course of methimazole administration.

The direct causal relationship between increased serum TSH levels and
preferential secretion of T₃ over T₄ by the thyroid tissue is further confirmed
by the data obtained in the patient with T₃ euthyroidism, whose spontaneous
hormonal pattern was in close agreement with the correlation found for all
the patients studied (see Fig. 2). It is remarkable that a unique correlation links
the TSH and the T₃/T₄ values both within each patient submitted to hormonal
monitoring and within all the different groups of patients. Furthermore, the
same correlation is valid among the operated patients, regardless of the possible
individual variability of the residual tissue response to TSH stimulation (Fig. 2
and Table 3). Thus, all the data obtained in the present study point to the
conclusion that endogenous TSH operates in man through a general mechanism
which induces a preferential secretion of T₃ over T₄, directly proportional to
its own serum concentrations and that this is a general pattern of stimulating
activity.

Considering the possible mechanism by which circulating TSH leads to this
preferential secretion of T₃ by the thyroid, the following observations should
be noted:
a) TSH stimulates the activity of an intrathyroid deiodinase which converts diiodothyrosine (DIT) into monoiodothyrosine (MIT) (Maayan & Rosenberg 1963).

b) During iodine depletion, the activity of an intrathyroidal deiodination mechanism, different from that in a), and which converts T$_4$ into T$_3$ is increased (Haibach 1971). Since TSH is considered to be the main factor responsible for the thyroid adaptation to iodine deficiency (Halmi 1954; Studer & Greer 1965; Greer & Haibach 1974; Fukuda et al. 1975) it can be inferred that TSH also activates intrathyroid conversion of T$_4$ into T$_3$ (Greer & Haibach 1974).

Thus, the activation of intrathyroid conversion of DIT into MIT and of T$_4$ into T$_3$, induced by elevated serum TSH levels, may represent the events leading to the preferential thyroid synthesis and secretion of T$_3$ over T$_4$ in man.

Finally, the present results, showing a preferential thyroid secretion of T$_3$ induced by TSH, together with the evidence that T$_3$ is much more potent than T$_4$ to inhibit TSH secretion (Shenkman et al. 1973; Sawin et al. 1977), suggest the occurrence of a pathophysiologic short loop, in which TSH is a potent stimulator of T$_3$, which, in turn, is a very sensitive and effective determinant of TSH secretion (Evered et al. 1974; Saberi et al. 1975; Vignieri et al. 1975).

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