Second University Clinic of Internal Medicine, Kommunehospitalet, 8000 Aarhus C, Denmark

SERUM TSH AND SERUM T₃ LEVELS DURING NORMAL MENSTRUAL CYCLES AND DURING CYCLES ON ORAL CONTRACEPTIVES

By

Jørgen Weeke and Aage Prange Hansen

ABSTRACT

Serum TSH and serum T₃ levels were studied in 35 normally menstruating women and in 35 women on oral contraceptives in order to find out whether the levels of TSH and T₃ were related to the menstrual cycle or changed by oral contraceptives. Serum TSH and T₃ were found to be unchanged throughout the normal menstrual cycles and during the cycles of oral contraceptives. The TSH level was higher in the women on oral contraceptives than in the normally menstruating women. This might be due to a direct thyroid inhibitory effect of oestrogen. As could be expected the levels of T₃ and T₄ were higher in women on oral contraceptives than in normally menstruating women.

The development of sensitive immunoassays of TSH and T₃ has made it possible to study physiological variations in these two parameters. The influence of oestrogen on thyroid function and thyroid regulation has long been recognized. We have therefore studied serum TSH and serum T₃ levels daily throughout the normal menstrual cycle, and also the effect on these two parameters of long-term use of oral contraceptives of combination type. Such a study is of importance for the clinical evaluation of the two thyroid parameters in women of fertile age.

MATERIAL AND METHODS

Seventy healthy young non-obese women were examined in the study. Thirty five of the women had normal menstrual cycles. These cycles had been regular at least during
Table 1.

<table>
<thead>
<tr>
<th>Composition and doses of the contraceptives</th>
<th>Number of subjects</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Serum TSH and T&lt;sub&gt;3&lt;/sub&gt; levels during a menstrual cycle</td>
</tr>
<tr>
<td>Progestin (mg)</td>
<td>Oestrogen (mg)</td>
</tr>
<tr>
<td>Ethinyloestrenol 2.5</td>
<td>Mestranol 0.075</td>
</tr>
<tr>
<td>Norgestrel 0.5</td>
<td>Ethinyloestradiol 0.05</td>
</tr>
<tr>
<td>Norgestrel 0.25</td>
<td>Ethinyloestradiol 0.05</td>
</tr>
<tr>
<td>Norethindrone acetate 3</td>
<td>Ethinyloestradiol 0.05</td>
</tr>
<tr>
<td>Megestrol acetate 4</td>
<td>Ethinyloestradiol 0.05</td>
</tr>
</tbody>
</table>

The last six months before the experiment. Thirty five of the women used contraceptive steroids of combination type. The composition and doses of the contraceptives are shown in Table 1. The contraceptives had been used in at least three cycles before the experiment. Except for oral contraceptives no medication was used by any of the subjects. They were all euthyroid by clinical examination, and none of them had goitres. Two experimental procedures were used.

Table 2 shows the number of subjects studied with each procedure and the distribution of age and ideal body weight.

Table 2.

<table>
<thead>
<tr>
<th>Experimental procedure</th>
<th>Use of oral contraceptives (−, +)</th>
<th>Number of subjects</th>
<th>Age (years) mean (range)</th>
<th>Ideal body weight per cent* mean (range)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Serum TSH and T&lt;sub&gt;3&lt;/sub&gt; levels daily during a menstrual cycle</td>
<td>−</td>
<td>11</td>
<td>30 (24–41)</td>
<td>89 (78–99)</td>
</tr>
<tr>
<td>Fasting serum TSH, T&lt;sub&gt;3&lt;/sub&gt; and T&lt;sub&gt;4&lt;/sub&gt; levels, single samples</td>
<td>−</td>
<td>24</td>
<td>24 (19–41)</td>
<td>93 (79–114)</td>
</tr>
<tr>
<td></td>
<td>+</td>
<td>24</td>
<td>24 (20–34)</td>
<td>93 (76–108)</td>
</tr>
</tbody>
</table>

* Calculated from the tables by Natvig (1956).
Serum TSH and serum T₃ levels daily during a menstrual cycle

A blood sample was obtained by venepuncture between 7 and 11 a.m. every day during the menstrual cycle. In the individual subject the time for blood sampling varied by less than an hour from day to day. The subjects had rested in a chair for 15 min after arrival to the hospital. In the normally menstruating women sampling was started on the second day of menstrual bleeding, and terminated on the first day of the next menstrual bleeding. In the women taking oral contraceptives sampling was started on randomly chosen days and terminated 28 days later.

Fasting serum TSH, T₃ and T₄ levels, single samples

A fasting blood sample was drawn between 7.30 and 8.30 a.m. The subjects had rested in a chair for 15 min before the venepuncture. The women taking oral contraceptives were all studied within the period of the menstrual cycle during which contraceptives were taken; otherwise the women were studied on randomly chosen days in the cycle.

The blood samples were centrifuged at +4°C and serum was stored at −20°C until analysis. The serum TSH was determined by a radioimmunoassay method previously described (Weeke & Ørskov 1973). Results were expressed in terms of μU of human TSH research standard A. The limit of sensitivity was 0.2 μU/ml in the assay involved. Serum T₃ was determined by a single antibody wick-chromatographic radioimmunoassay method (Weeke & Ørskov, to be published). T₃ acid form (Sigma Chemical Company, St. Louis) was used as standard. The inter-assay coefficient of variation was 7% in one serum sample with an average T₃ value of 2.06 ng/ml determined in triplicate in 40 consecutive assays. The intra-assay coefficient of variation determined on single determinations was 8, 5 and 5% respectively in three different serum samples with average T₃ values of 0.75 ng/ml, 1.14 ng/ml and 1.62 ng/ml respectively. Serum luteinizing hormone (LH) was determined by a double antibody method using ¹²⁵I labelled human chorionic gonadotrophin as tracer (BIO-RIA, LH radioimmunoassay Kit). The LH values were expressed as mU/ml and the intra-assay coefficient of variation was 3% at an LH level of 2.6 mU/ml. All samples from one subject were determined within the same TSH, T₃ and LH assay respectively. The serum thyroxine was determined by a modification of the method described by Murphy & Jachan (1965).

Student's t-test was used for the statistical calculations. In order to meet the assumptions of the analyses the TSH values were transformed to common logarithms before the statistical calculations. For TSH values below 0.2 μU/ml this value was used in the calculations.

RESULTS

Fig. 1 shows the mean serum LH, TSH and T₃ values in the 11 women with normal menstrual cycles. The values are centered according to the day of the midcycle LH peak (day 0). An LH peak occurred in all 11 subjects. The mean serum TSH level was stable at the time of the LH peak, and the mean pre- and post-ovulatory serum TSH levels were not different from each other. Moreover the values around menstruation were stable. The mean and individual serum T₃ levels were also stable throughout the menstrual cycle. There was no
Serum LH, TSH and T₃ in 11 women studied during a normal menstrual cycle. The values are centered according to the day of midcycle LH peak (day 0). Mean ± 1 SEM. For TSH the mean values and their standard errors were calculated from their logarithmic transformed data. In the figure these values were transformed back into linear scale by looking up their antilogarithms.

correlation between the serum TSH and serum T₃ levels in any of the individual subjects.

Fig. 2 shows the mean serum TSH and T₃ values in the 11 women on oral contraceptives. There was no difference in the serum TSH or T₃ levels during the 21 days of therapy and during the 7 days without therapy. Comparing the TSH and T₃ levels in Figs. 1 and 2 the values in the women taking oral contraceptives were slightly higher than the values in the normally menstruating women. This difference was, however, not significant. There was no correlation between serum TSH and serum T₃ levels in any of the individual subjects.

Fig. 3 gives a comparison of fasting serum TSH, T₃ and T₄ values between normally menstruating women and women on oral contraceptives. For all the parameters the values were slightly but significantly higher in the women on oral contraceptives. For serum TSH: 1.1 as compared with 0.7 µU/ml
(P < 0.05). For serum T₃: 1.41 as compared with 1.21 ng/ml (P < 0.02). For serum T₄: 10.8 as compared with 7.9 μg/100 ml (P < 0.001). The serum TSH level was not correlated to the serum T₃ or serum T₄. A significant correlation, however, could be shown between serum T₃ and serum T₄ (r = 0.549, P < 0.01, in normally menstruating women, and r = 0.736, P < 0.001, in women on oral contraceptives).

**DISCUSSION**

In the present study, in which the day to day variations in TSH and T₃ were followed over an entire menstrual period, and in which the time of ovulation was determined by serum LH analysis, there was no indication whatsoever of a relationship between the ovarian cycle and the function of the thyroid gland or the pituitary TSH producing cell system. This is in agreement with results obtained by Lemarchand-Béraud & Vannotti (1969) who studied fasting serum TSH and PBI once weekly during a normal menstrual cycle in 5 women. They found a non-significant increase in both parameters in the midcycle samples, and a slight also non-significant fall during the luteal phase. Chan et al. (1972) studied urinary excretion of T₃ daily during a normal menstrual cycle in one woman and found that the excretion remained within narrow limits except for a transient increase during menstruation.

![Fig. 2.](image)

Serum TSH and T₃ values in 11 women on oral contraceptives of combination type studied during a 28 day period. Mean ± sem. The TSH values are expressed as in Fig. 1.
Fasting serum TSH, T4 and T3 values in 24 normally menstruating women, left column, and 24 women on oral contraceptives, right column. The serum TSH values are given on a log scale. The broken lines indicate the limit of sensitivity of the TSH assay. The solid lines indicate the mean levels of TSH, T4 and T3. The mean TSH values were calculated from the logarithmic transformed data.

It was found that total T4 and T3 values were higher in the women taking oral contraceptives than in the normally menstruating women. This is due to the well known fact that oestrogen increases the binding capacity of TBG, which is the primary thyroid hormone binding protein in blood. The absolute concentrations of the free hormones, however, are probably unchanged, since the increase in thyroid hormone concentration is followed by a proportional fall in the per cent of unbound T4 and T3 (Oppenheimer et al. 1963; Sterling & Brenner 1966; Dussault et al. 1973). It has been found that the progestin component in oral contraceptives is without any effect on the thyroid hormone concentration in blood (Winikoff 1968).

The effects of oestrogen on thyroid function and thyroid regulation are less well understood. Gross et al. (1971) showed that administration of a single dose of ethinyloestradiol (0.1 to 3 mg) to males suppressed thyroid iodine release, whereas smaller doses (0.05 mg or less) were without such effect. The reports
on the effect of single doses of oestrogen on the serum TSH level are conflicting. Adams & Maloof (1970) showed that a single dose of ethinyloestradiol (0.5–1 mg) given to males resulted in a 3–10 fold increase in the serum TSH levels. Hall et al. (1971) showed that a single dose of ethinyloestradiol (1 mg) increased the serum TSH in only one of five male subjects studied. Other investigators (Carlson et al. 1973; Faglia et al. 1973; Woolf et al. 1973) have found no effect on the serum TSH level of single doses of oestrogen. In the present study we found a slight but significant higher TSH level in women who had used oral contraceptives of combination type for at least 6 months than in normally menstruating women. As the absolute concentrations of the free hormones are probably unchanged, the higher level of TSH induced by the contraceptives is likely to be caused by a direct thyroid inhibitory effect of oestrogen. Such an effect has been demonstrated by Gross et al. (1971) who studied the effect of ethinyloestradiol on thyroid iodine release after injection of bovine TSH to subjects whose endogenous TSH secretion had been blocked by T<sub>3</sub>.

ACKNOWLEDGMENTS

Human TSH and anti-TSH serum were kindly supplied by the Pituitary Agency of the National Institute of Health, Bethesda, Maryland, USA, and human TSH standard A by the National Institute of Medical Research, London. The study was supported by grants from Statens lægevidenskabelige Forskningsråd and P. Carl Petersens Fond. We are most grateful to Ketty Jensen and Jenifer Martin for conscientious technical assistance. The serum thyroxine determinations were kindly performed in the Department of Clinical Chemistry, Kommunehospitalet, Aarhus, Denmark (Professor Rud. Keiding).

REFERENCES


Received on September 3rd, 1974.