The Effect of Prednisolone and Metyrapone on FSH Release Induced by the Administration of LRH

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Synopsis

The response of follicle-stimulating hormone (FSH) to a single injection of synthetic LRH was established in 7 and 6 women following an intramuscular dose of 0.2 mg and 0.1 mg. The secretion of FSH was greater in the group injected with 0.2 mg LRH than in the group injected with 0.1 mg. On the other hand, the response of FSH to a single injection of LRH (0.1 mg/subject) was established in 7 men before and after the pretreatment with metyrapone for one day (4.5 g/subject). Pretreatment with metyrapone provoked a hypersecretion of FSH following a single injection of synthetic LRH. Seven women, 21-48 years of age who were treated with prednisolone for at least 1.5 months were examined for the responsiveness of the anterior pituitary to a single injection of synthetic LRH (0.2 mg). The secretion of FSH was not suppressed and the maximal serum level of FSH was observed 60 min after LRH injection.

A few reports (Baldwin and Sawyer, 1974; Sakakura et al., 1975) are published on the effect of glucocorticoids on the release of LH and only report (Suda et al., 1974) is observed as to the glucocorticoid inhibition of FSH release. On the other hand, we reported previously (Sakakura et al., 1975) that pretreatment with metyrapone provoked a hypersecretion of LH by the administration of synthetic LRH, but no reports are observed about FSH. It is consequently important to make sure whether or not FSH secretion induced by the synthetic LRH was increased by the administration of metyrapone and also FSH secretion induced by LRH was suppressed by the long-term pretreatment with prednisolone.

Materials and Methods

Seven healthy male students, 19-21 years of age, with normal sexual function as determined by history, were studied in Hokkaido University Hospital at Sapporo. Each received a single injection of synthetic LRH (0.1 mg) intramuscularly. Two weeks after the first injection, they were treated with 750 mg of metyrapone by mouth every 4 hr for one day. They received a single injection of synthetic LRH (0.1 mg) intramuscularly 24 hr after the first dose of metyrapone. Two ml of blood was withdrawn from the cephalic vein at 0, 15, 30, 45, 60, 90 and 120 min after LRH injection for the determination of the concentration of FSH. All urine was collected for 24 hr during treatment with metyrapone and the concentration of 17-OHCS was determined by the method developed by Porter and Silber (1950) with minor modifications. The datum of LH in this experiment was reported previously in another journal (Sakakura et al., 1975). Thirteen healthy female nurses 22-26 years of age, with normal sexual function as determined by history, were studied. Seven nurses received a single injection of 0.1 mg of synthetic LRH intramuscularly and 6 nurses received 0.2 mg of synthetic LRH in the luteal phase of their menstrual cycle. No side effects induced by administra-
tion of LRH were observed other than a 1 or 2 day delay in menstruation. Seven women, 21-48 years of age (average: 31 years old) were studied. They had been treated with prednisolone for more than 1.5 months as shown in Table 2. Prednisolone was continued during the experimental studies. A single injection of 0.2 mg of synthetic LRH was injected intramuscularly in the morning between 9:00 and 11:30 hr and 2-ml blood samples were obtained from them at 0, 15, 45, 60, 90 and 120 min with heparinized syringes. Serum FSH was measured by a specific radioimmunoassay (Berson et al., 1964; Midgley, 1966), using HFSH-Radioimmunoassay kit Daiichi Radioisotope Lab, Japan (Hashimoto et al., 1972) and the anti-HFSH serum of this kit (Hashimoto et al., 1972) was provided by Calbiochem. All samples were measured in duplicate. The coefficient of variation for FSH in the assay was 11.5% at the logitransformed dose-response curve (Rodbard, 1968). The results of FSH assays were expressed as mIU/ml of serum using the 2nd-IRP-HMG as a standard. The minimal detectable dose in this assay was 2.0 mIU/ml for FSH. The synthetic LRH utilized in the present study was produced without pyrogen, by Daiichi Seiyaku Co., Ltd. All data were treated by the method of analysis of variance by two-way classification.

Results

The response of serum FSH to synthetic LRH of normal women is shown in Fig. 1. A single injection of 0.2 mg of LRH provoked a significant increase (p<0.05) of FSH (8.9±2.2 mIU/ml) 15 min after the injection and the gradual increase of plasma FSH was observed during this experiment. A dose of 0.1 mg of synthetic LRH provoked a maximal increase of plasma FSH (11.3±5.3 mIU/ml) at 60 min and the concentration of plasma FSH was maintained for 120 min. There was a significant difference (p<0.05) between 0.2 and 0.1 mg LRH administration groups as shown in Fig. 1. The administration of 0.1 mg of synthetic LRH to normal men provoked a peak value of plasma FSH (9.1±5.0 mIU/ml) 45 min after the injection as shown in Fig. 2. On the other hand, pretreatment with 4.5 g of metyrapone for one day resulted in an increase of FSH secretion after a single injection of 0.1 mg of synthetic LRH and the peak value of plasma FSH (16.3±5.9 mIU/ml) was observed 45 min after the injection. There was a significant difference (p<0.01) between metyrapone pretreated and control groups in the response to LRH. The high secretion of 17-OHCS in urine which was collected from the men pretreated with metyrapone are observed in Table 1. The result suggests us that the level of plasma cortisol should be low.

![Fig. 1. The responsiveness of normal people to a single intramuscular injection of synthetic LRH (0.2 or 0.1 mg) is shown in this figure; ——— = the plasma FSH of normal women who received synthetic LRH (0.1 mg); ——— = the plasma FSH of normal women who received synthetic LRH (0.2 mg). All normal women were given a single injection of synthetic LRH in the luteal phase of their menstrual cycle; Figures in parentheses=numbers of subjects; vertical lines in curves indicate SE of the mean.](image1)

![Fig. 2. The responsiveness of plasma FSH to a single injection of synthetic LRH (0.1 mg) in normal men who were pretreated with metyrapone (4.5 g) for 1 day is shown in this figure; ——— = control group; ——— = the group pretreated with metyrapone; figures in parentheses=the numbers of subjects; vertical lines in curves indicate SE of the mean.](image2)
Meanwhile, the prolonged uninterrupted treatment of women with prednisolone did not cause a significant inhibition of FSH secretion induced by the administration of 0.2 mg of synthetic LRH even though LH secretion was inhibited (Sakakura et al., 1975) by prolonged treatment with prednisolone as shown in Fig. 3.

**Discussion**

It is assumed that the site of corticoid feedback action in ACTH release both at the anterior pituitary and suprahypophyseal level in the hypothalamo-pituitary-adrenal axis (Chowers et al., 1971; Sakakura and Brodish 1972; Takebe et al., 1974). Recent studies (Nicoloff et al., 1970; Wilber and Utiger 1963) showed that glucocorticoids suppressed the plasma TSH level in patients with primary hypothyroidism and normal subjects. They suggested that glucocorticoids reduced TSH secretion at the suprahypophyseal level. Haigler et al. (1971) also found that the response to TRH was not blocked by glucocorticoids. Meanwhile, Otsuki et al. (1973) reported that the TSH secretion induced by TRH appeared to be inhibited not only at the suprahypophyseal level but also at the pituitary level after the long term and high dose treatment with glucocorticoids.

**Table 1.** The daily 17-OHCS urinary excretion from 6 male students treated with 4.5 g of metyrapone for 1 day. Following the administration of metyrapone, subjects were injected with a single dose of synthetic LRH (0.1 mg): see Fig. 2.

<table>
<thead>
<tr>
<th>Sex</th>
<th>Urine (ml)</th>
<th>17-OHCS (mg/day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Y</td>
<td>900</td>
<td>19.1</td>
</tr>
<tr>
<td>I</td>
<td>900</td>
<td>17.3</td>
</tr>
<tr>
<td>K</td>
<td>1,100</td>
<td>16.9</td>
</tr>
<tr>
<td>U</td>
<td>800</td>
<td>18.8</td>
</tr>
<tr>
<td>S</td>
<td>1,000</td>
<td>22.7</td>
</tr>
<tr>
<td>A</td>
<td>600</td>
<td>19.0</td>
</tr>
<tr>
<td>S</td>
<td>1,200</td>
<td>16.1</td>
</tr>
<tr>
<td>Baseline</td>
<td>3-11 (mg/day)</td>
<td></td>
</tr>
</tbody>
</table>

**Table 2.** The dose and duration of treatment with prednisolone. These patients were injected with a single dose of synthetic LRH (0.2 mg) intramuscularly after the long-term treatment with prednisolone: see Fig. 3.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Age</th>
<th>Diagnosis</th>
<th>Duration of treatment</th>
<th>Prednisolone (mg/day)</th>
</tr>
</thead>
<tbody>
<tr>
<td>I</td>
<td>21</td>
<td>Aortitis</td>
<td>5.0 (months)</td>
<td>30-20</td>
</tr>
<tr>
<td>T</td>
<td>24</td>
<td>Lupuserythematous</td>
<td>3.0 (months)</td>
<td>30-20</td>
</tr>
<tr>
<td>S</td>
<td>48</td>
<td>Rheumatoid arthritis</td>
<td>2.5 (months)</td>
<td>40-20</td>
</tr>
<tr>
<td>K</td>
<td>32</td>
<td>Rheumatic fever</td>
<td>2.0 (months)</td>
<td>10</td>
</tr>
<tr>
<td>G</td>
<td>25</td>
<td>Aortitis</td>
<td>4.0 (months)</td>
<td>30-10</td>
</tr>
<tr>
<td>S</td>
<td>24</td>
<td>Rheumatoid arthritis</td>
<td>1.5 (months)</td>
<td>30</td>
</tr>
<tr>
<td>N</td>
<td>43</td>
<td>Rheumatoid arthritis</td>
<td>2.0 (months)</td>
<td>30-20</td>
</tr>
</tbody>
</table>
Glucocorticoid suppression of GH release induced by hypoglycemia also has been reported in normal subjects (Frantz and Rabkin 1964; Nakagawa et al., 1969), but dexamethasone did not inhibit the GH release induced by arginine (Nakagawa et al., 1969). In rats, glucocorticoid may inhibit the GRH release (Pecile and Müller, 1966) by the hypothalamus. The mechanism of glucocorticoid suppression of GH release in human beings is still not clear at present.

As to the effect of corticoids on the release of gonadotropin, Baldwin and Sawyer (1974) studied the effects of dexamethasone on ovulation and LH release in 4-day cycling rats, and concluded that dexamethasone was most likely acting by inhibiting the synthesis and releasing of pituitary LH in response to exogenous LRH. In our experiments, the secretion of LH following LRH injection was significantly suppressed in the patients who had been continuously treated with prednisolone for a long term (Sakakura et al., 1975). Thus, we suggested that the main site of glucocorticoid inhibition in LH release might be at the anterior pituitary level. However, it still remains to be determined whether or not glucocorticoids act directly at the supra-hypophyseal or ovarian level also. Antopalm (1950) and Winter et al. (1950) reported that the ovary was virtually unaffected by glucocorticoids. This datum suggests that the acting site of glucocorticoid inhibition is scarcely possible at the ovary.

As to the glucocorticoid inhibition of FSH release, Suda et al. (1974) reported that pretreatment with glucocorticoids (8 mg dexamethasone for one day) in human beings resulted in inhibition. In contrast, Boccuzzi et al. (1975) reported that the plasma FSH response to the injection of LRH in Cushing's disease was similar to that recorded in normal subjects in spite of the decreased response in plasma LH.

In our experiments, pretreatment with prednisolone for a long term resulted in the failure of inhibition of FSH release from the anterior pituitary induced by a single injection of 0.2 mg of synthetic LRH. This datum shows that FSH release by the decapeptide will not be inhibited by glucocorticoid and confirms the datum of Boccuzzi et al. (1975). A dissociation observed between Suda's datum (Suda et al., 1974) and ours may depend on the difference of the period and dose of glucocorticoids treated.

On the other hand, the LH release induced by LRH was inhibited by the long-term treatment with glucocorticoids but the FSH release induced by LRH was not inhibited by glucocorticoid pretreatment. The reasons are not clear at present. It may relate to the dissociation of sensitivity to glucocorticoids of secretion cells of FSH and LH. But we have no data to support this explanation.

It is well known that metyrapone stimulates ACTH release (Liddle et al., 1959; Gold et al., 1960) through the inhibition of cortisol secretion. Previously, we (Sakakura et al., 1975) also reported that pretreatment with metyrapone provoked more secretion of LH than that in control men given LRH. In this experiment, pretreatment with 4.5 g of metyrapone also provoked more secretion of FSH than that in control men given a single injection of 0.1 mg of LRH. Our data showed that metyrapone enhanced the responsiveness of the pituitary gland to LRH. The reason for this phenomenon is not obvious at present but at least two explanations may be proposed; 1) the increment of pituitary responsiveness to LRH for the release of FSH may be related to the decrement of the concentration of plasma cortisol, 2) metyrapone itself may activate directly the pituitary responsiveness for FSH release to LRH irrespectively of the decrement of plasma cortisol. The precise mechanism remains obscure and must await further
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investigations.

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References