Impaired Steroidogenic Function of Corpora Lutea from Hyperprolactinemic Baboons Induced by Sulpiride

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Abstract

It has been noticed that hyperprolactinemia can cause luteal insufficiency as well as anovulation in women. In order to explore the mechanism underlying this disorder, hyperprolactinemia was induced in baboons (Papio cynocephalus) by daily administration of sulpiride during follicular and early luteal phases. In hyperprolactinemic baboons, the plasma progesterone level was suppressed without notable changes in plasma estradiol, LH and FSH levels. When corpora lutea from these baboons were examined in vitro to investigate their ability to convert 14C-pregnenolone into various steroids, there was progressive inhibition of steroid metabolism related to the plasma levels of prolactin. These findings strongly suggest, although do not actually prove, that an elevated level of prolactin could directly impair luteal function by adversely affecting 3β-hydroxysteroid dehydrogenase activity.

Material and Methods

Although hyperprolactinemia is known to cause luteal insufficiency (LI) (Seppala et al., 1976) as well as anovulation (Bohnet et al., 1976) in women, the precise site of prolactin (PRL) action and the mechanism by which PRL exerts its inhibitory effects on ovarian function have been poorly understood. To further elucidate the mechanism involved in the pathogenesis of hyperprolactinemic LI, we have developed sulpiride-induced LI in the baboon (Aso et al., 1982). Using this experimental model, steroidogenic function of corpora lutea exposed to high levels of PRL in vivo was studied.

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was estimated on the bases of sex skin swelling (start of lessening of sex skin swelling=postovulatory day 0).

Hyperprolactinemia was induced by daily administration of sulpiride 100 mg i.m. at 1000 h, starting on the first day of the cycle and continuing until the 25th day (the seventh day postovulation). Corpora lutea which had been exposed to high PRL level in vivo (40 and 70 ng/ml compared to 18 ng/ml in control) were excised on the seventh day postovulation under general anesthesia with 50 mg ketamine i.m.. Whole CL was cut into small pieces, and after thorough mixing, approximately 50 mg (w.w.) of minced tissue was incubated in 3 ml of Hank’s BSS with 1 μCi 14C-pregnenolone (57 mCi/mmol) for three hours at 37°C under 5% CO2/95% O2. Separation and identification of the radiolabeled steroids formed were performed as described elsewhere (Suzuki et al., 1977; Fujita et al., 1981). The results are expressed as disintegrations per minute (dpm) per 100 mg (w.w.) tissue after correction for recovery and counting efficiency. The rate of conversion of 14C-pregnenolone into various steroids by CL of control baboons was between 15 to 20%.

Plasma levels of prolactin, gonadotropins and steroids of sulpiride-treated baboons and control baboons were determined on samples drawn immediately before surgery on the 7th postovulatory day as described earlier (Aso et al., 1982; Suzuki et al., 1980; Aso et al., 1975).

Results

Plasma levels of PRL, gonadotropins and steroids in those baboons used for the experiments are shown in Table 1. As seen plasma PRL level of sulpiride-treated baboons were elevated whereas LH and FSH levels were slightly decreased. While the estradiol (E2) level in the treated baboons appeared to be essentially unaffected, plasma progesterone (P4) was markedly suppressed in treated baboons indicating that these baboons had LI. As shown in Figure 1, the major metabolites formed following in vitro incubation with 14C-pregnenolone of CL from these baboons were P4, 17OHP4, androstenedione (A) and a lesser amount of E2. The amounts of steroids formed with the exception of E2 were inversely correlated to the plasma PRL level: at a plasma PRL level of 40 ng/ml, P4, 17OHP4, A and E2 production was inhibited by 62%, 83%, 62% and 85%, respectively, and at 70 ng/ml, 96%, 96%, 97% and 75% inhibition was attained.

Table 1 PLASMA LEVELS OF PROLACTIN, GONADOTROPINS AND STEROIDS IN SULPIRIDE-TREATED AND UNTREATED BABOONS.

<table>
<thead>
<tr>
<th></th>
<th>SULPIRIDE-TREATED</th>
<th>UNTREATED</th>
<th>CONTROL (mean±SEM,n=10)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>BABOON 1</td>
<td>BABOON 2</td>
<td></td>
</tr>
<tr>
<td>PRL (ng/ml)</td>
<td>40.0</td>
<td>70.0</td>
<td>18.0</td>
</tr>
<tr>
<td>LH (μg/ml)</td>
<td>1.2</td>
<td>1.3</td>
<td>1.5</td>
</tr>
<tr>
<td>FSH (μg/ml)</td>
<td>4.1</td>
<td>3.9</td>
<td>5.0</td>
</tr>
<tr>
<td>E2 (pg/ml)</td>
<td>115.4</td>
<td>98.6</td>
<td>121.3</td>
</tr>
<tr>
<td>P4 (ng/ml)</td>
<td>3.5</td>
<td>1.8</td>
<td>5.8</td>
</tr>
</tbody>
</table>

The data presented are the means of duplicate determinations which agreed within ±10%.
Figure 1: CONVERSION OF $^{14}$C-PREGNENOLONE BY CORPORA LUTEA FROM SULPIRIDE-TREATED AND UNTREATED BABOONS.

The data presented are the means of duplicate determinations which agreed within ±10%, except for $^{14}$C-estradiol (E$_2$) formation by CL of sulpiride-treated baboons. In treated baboons, values for E$_2$ were 622 and 708 dpm per 100 mg tissue (PRL 40 ng/ml), and 656, 1102 dpm (PRL 70 ng/ml).

**Discussion**

In the CL from the control baboon, pregnenolone was metabolized into P$_4$, 17OHP$_4$, A and E$_2$. This steroidogenic profile is essentially similar to that in the human CL (Fujita et al., 1981) suggesting that the D'-pathway is the major route of steroid biosynthesis by baboon CL as well. Since all steroids metabolized from $^{14}$C-pregnenolone by CL of the two hyperprolactinemic baboons with the exception of E$_2$ were decreased in a parallel manner, the principal site of inhibition may be at the step of 3β-hydroxysteroid dehydrogenase (3β-HSD) activity. The significance of higher E$_2$ production in the baboon with a plasma PRL level of 70 ng/ml compared to E$_2$ in the baboon with 40 ng/ml PRL could not be ascertained since the method used in this study does not allow a detailed assessment of each metabolic step. Inhibitory action of PRL on the development of 3β-HSD activity in CL was also envisioned in a histochemical study of these CL (data not shown). This marked inhibitory effect of hyperprolactinemia on the induction of 3β-HSD activity can well account for the lower progesterone level in hyperprolactinemic ba-
boons.

It has been reported that sulpiride-induced hyperprolactinemia impaired luteal function in vivo of normally cycling women when the drug was administered during the follicular phase of the corresponding cycle, but not when the drug was administered during the luteal phase only (Delvoye et al., 1973; Kauppila et al., 1982). In addition, hyperprolactinemic LI was normalized by administration of the dopaminergic agonist, bromocriptine, throughout the whole cycle (Muhlenstedt et al., 1978; Andersen et al., 1979) or during the follicular phase only (Kano and Nishikawa, 1983). The present results suggest that the inhibitory effect of hyperprolactinemia on luteal function was exerted mainly at the level of 3β-HSD induction during follicular maturation and an early stage of CL formation.

Since circulating levels of gonadotropins in sulpiride-treated baboons remained comparable to those of the control baboon, it would appear that elevated PRL acted directly at the ovarian level. However, direct action of PRL on steroidogenic function of baboon luteal cells remains to be further clarified.

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**References**


