Journal of Steroid Biochemistry, 1975, Vol. 6, pp. 1043–1046. Pergamon Press. Printed in Great Britain.

LH AND FSH RELEASING MECHANISM IN THE TESTICULAR FEMINIZATION SYNDROME

A. ZÁRATE, J. SORIA and E. S. CANALES
Department of Gynecologic Endocrinology, Hospital Gineco Obstetricia No. 1, Instituto Mexicano del Seguro Social, México

INTRODUCTION

It has been assumed that sexual differentiation of the brain is an androgen dependent characteristic, and that patients with the testicular feminization syndrome provide the possibility to study the sexual steroid feedback control of gonadotropin secretion when androgen effects are presumably absent. Moreover, it has been postulated that in normal males FSH secretion is regulated by the testicular production of estrogens or an unidentified substance from the germinal epithelium, and that LH secretion is regulated by testosterone [1–4]. In the syndrome of testicular feminization, if the hypothalamic center which regulates gonadotropin secretion is sensitive to testosterone, one would predict a marked elevation of LH in the blood and a normal or reduced level of FSH. Early studies have suggested that in this syndrome the gonads exert a more inhibitory action on the release and synthesis of FSH than on LH with the result of an adequate gonadal feedback control of FSH secretion and partial control of LH secretion [4]. In the present study we investigate the effect of various stimuli on FSH and LH secretion in seven patients with the complete form of the testicular feminization syndrome.

Administration of luteinizing hormone–releasing hormone (LH–RH)

Patients were studied after an overnight fast. An indwelling venous catheter was inserted and an infusion of isotonic saline solution was started. After a baseline consisting of blood samples at 15 min intervals for one hour, a dose of 50 μg of synthetic LH–RH (Hoechst) was administered as a bolus injection through the catheter. Serum samples were obtained at repeated intervals. In some cases the LH–RH test was carried out before and after gonadectomy. Serum LH and FSH were measured by a double antibody radioimmunoassay procedure according to the methods of Midgley[5–6]. The results were expressed as ng/ml using LER-907 as the reference standard. The FSH and LH assays could detect 37·5 and 18·7 ng/ml respectively in our laboratory. Within assay error, measured as the coefficient of variation, averaged 10·0% for LH and 7·5% for FSH. All sera from individual subjects were measured in duplicate in the same assay. Highly purified FSH, LH, LER-907, and antisera were generously provided by the National Pituitary Agency.

In these patients basal LH levels were found to be elevated in comparison to normal males and females; likewise FSH serum basal levels were increased in most of the patients. Although the present patients' LH and FSH levels were elevated before surgery, they rose substantially following castration. Their FSH serum levels increased two to seven fold following gonadectomy, and 10 to 15 fold two months after castration. The increase in LH after gonadectomy was of a lesser magnitude. The concentrations of LH and FSH for each sampling time before and after the administration of LH–RH are shown in Fig. 1. The administration of LH–RH increased serum LH by eight minutes in all cases. An increase in serum FSH levels occurred in all patients after LH–RH. The maximal increase for FSH occurred at 32–64 min. LH–RH released relatively more FSH in the testicular feminization syndrome than in adult normal subjects; however maximum LH increase did not differ significantly ($P < 0{\cdot}05$) between the patients with testicular feminization and normal patients. The administration of LH–RH after gonadectomy further elevated the already increased serum levels of both LH and FSH in the four patients. LH–RH released comparable amounts of LH and FSH to before castration (Fig. 2).

Estrogen and progesterone administration

Two weeks after castration 50 mg progesterone in oil was administered intramuscularly, and venous blood samples were drawn at 0, 2, 4, 6, 16, 24 and 48 h. One week later, the four patients were treated with ethinyl estradiol in a daily oral dose of 80 μg for four weeks. Then progesterone 50 mg dissolved in oil was given intramuscularly to all subjects at 8 a.m. Blood samples were obtained at repeated intervals. The estrogen administration was continued both during and after progesterone treatment.

The intramuscular administration of 50 mg progesterone in oil to the four patients not treated with estrogens did not result in any major changes either in the FSH or the LH levels. During the administration of 80 μg ethinyl estradiol there was a decrease in the serum levels of both FSH and LH in all four patients (Fig. 3). This decrease was already clear after one week. The relative decrease was much greater for FSH than for LH. The intramuscular administration of progesterone to these patients treated with estrogens resulted in an increase in both LH and FSH. These peak levels were observed 6 to

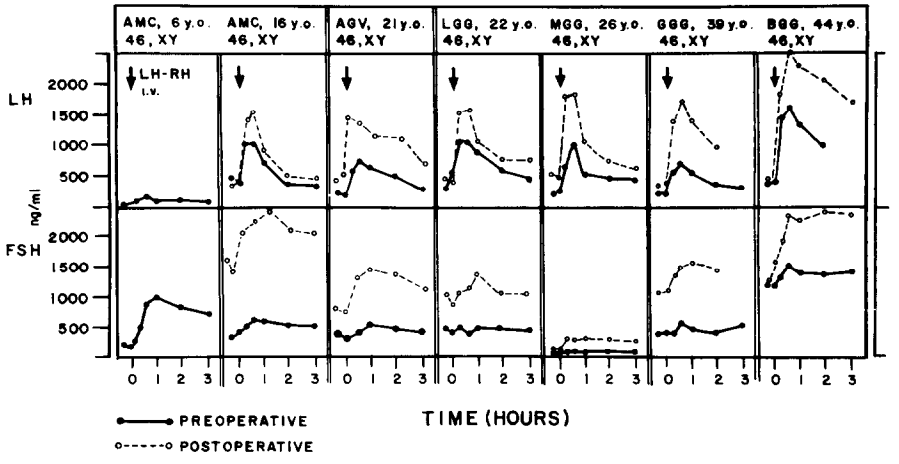

Fig. 1. Serum LH and FSH responses to the intravenous injection of synthetic LH–RH (Hoechst) in 7 patients with the testicular feminization syndrome. The gonadotropin secretory response to LH–RH after gonadectomy in some of these patients is also shown. Note the diminished LH section after LH–RH in a patient 6 years old; contrast the hyper-responsiveness to LH–RH in a patient 44 years old.

12 h after progesterone administration. The rise in the serum levels of FSH was only slightly smaller than the rise in the LH levels.

Three months after gonadectomy, three patients were stimulated with estradiol benzoate to assess the positive feedback effect of estrogen in patients with testicular feminization. Estradiol benzoate in a dose of 50 μg per kg of body weight was administered by single i.m. injection. Circulating levels of FSH and LH were measured at two, four, or six hour intervals, up to 72 h after the estradiol benzoate injection. No positive gonadotropin response was observed (Fig. 4).

Clomiphene administration

Two weeks before gonadectomy was performed, four patients received orally 100 mg of clomiphene citrate twice daily for seven days. Blood samples were obtained the day before clomiphene treatment, and then the 3rd, 5th, and 7th day of drug administration. None of the 4 patients exhibited significant increments in serum FSH and LH levels (Fig. 5).

DISCUSSION

Our results demonstrate the presence of a partial gonadal feedback control of both FSH and LH secretion suggesting that this gonadotropic feedback control is exerted through gonadal factors, probably estrogens or via formation of estrogens from circulating testosterone by the brain [7]. Therefore the present data are unable to confirm the disparity in the gonadal feedback control of LH and FSH secretion

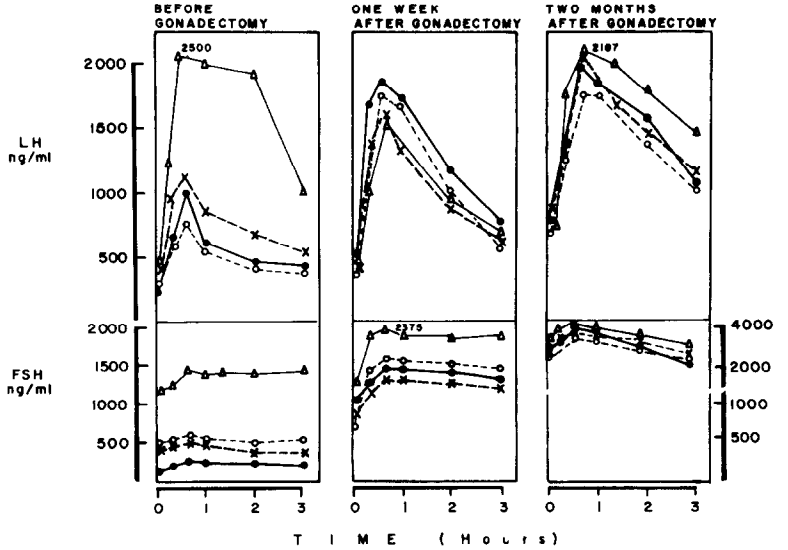

Fig. 2. The administration of synthetic LH–RH after one week and two months after gonadectomy in 4 patients further elevated the increased serum levels of both LH and FSH. LH–RH released comparable amounts of gonadotropins to those seen before castration. (From Zarate *et al. Am. J. Obstet. Gynec.* (1974) 119–971. Courtesy of C. B. Mosby Co.)

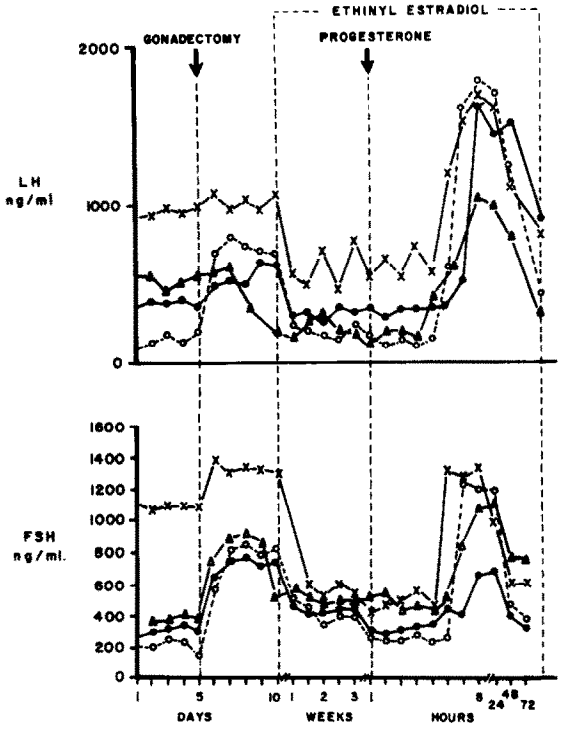

Fig. 3. Serum concentrations of LH and FSH in four patients tteated with 80 μg per day of ethinyl estradiol. and the intramuscular injection of 50 mg of progesterone. (From Zarate *et al. Am. J. Obstet. Gynec.* (1974) 119–971. Courtesy of C. B. Mosby Co.)

as has been suggested by others[3]. It has also been postulated that FSH levels are reciprocally related to spermatogenic activity [1–2]; however, in these patients in spite of a presumed lack of androgen effect

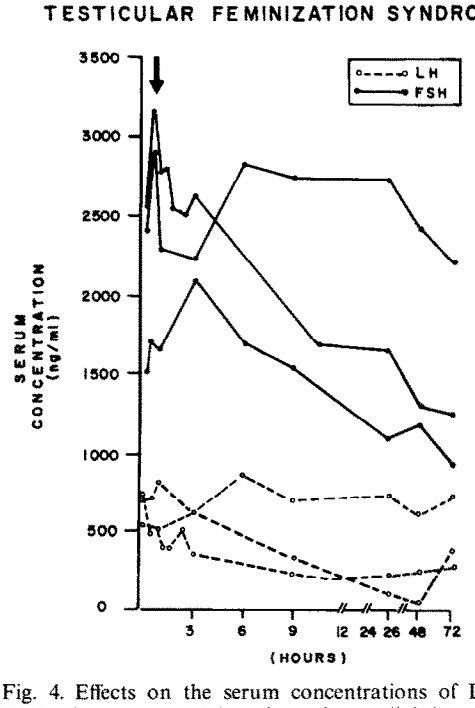

Fig. 4. Effects on the serum concentrations of LH and FSH after i.m. administration of estradiol benzoate to three patients with testicular feminization syndrome.

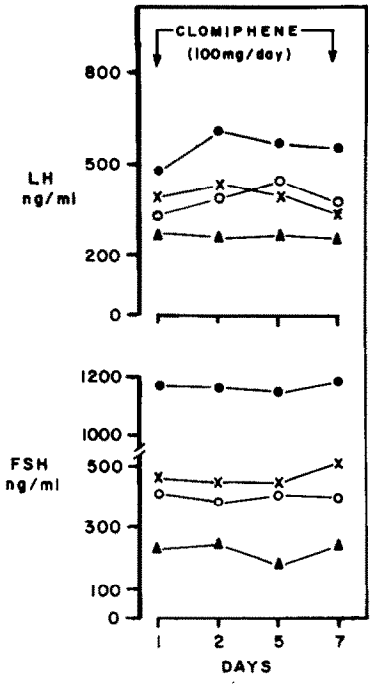

Fig. 5. Effect of clomiphene administration on serum concentration of LH and FSH.

and a lack of total maturation of germinal cells, these gonads were able to maintain some feedback control on the FSH levels in some of the subjects. The data reported here demonstrate that Leydig cell steroids partially modulate gonadotropin secretion. The observed rise in LH and FSH after gonadectomy would indicate that testicular sources of estrogens play a role in the regulation of gonadotropin secretion. Likewise FSH and LH levels were markedly reduced during administration of estrogens [8]. It has been reported previously that the administration of testosterone to patients with testicular feminization has been followed by a decrease in both FSH and LH serum levels suggesting either that the hypothalamus is not entirely insensitive to testosterone in patients with this syndrome or that the effects of the administered or endogenous androgens were produced by partial conversion to active estrogens. Clomiphene has been shown to promote gonadotropin release presumably by competing for estrogen binding sites in the hypothalamus and anterior pituitary gland; however, prior to puberty or in presence of hypoestrogenic states this drug does not release FSH and LH. In present study, the four patients who were studied and already had high gonadotropin levels failed to increase FSH and LH after clomiphene therapy. The absent response to clomiphene could be due to either the low estrogen concentration present in these four patients or to some abnormality at the hypothalamic receptors.

The sensitivity of pituitary responsiveness to LH–RH is modulated by gonadal steroids; thus in the absence of gonadal feedback as in cases of gonadal failure (Fig. 6) the pituitary responsiveness to LH–RH

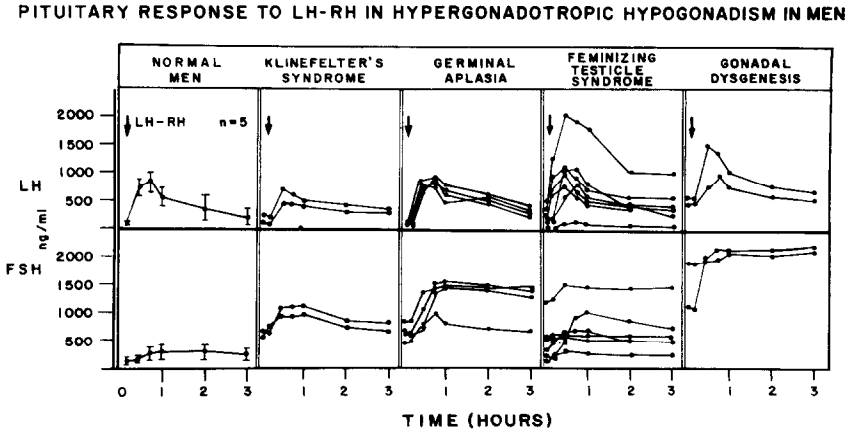

Fig. 6. Pituitary gonadotropin responsiveness to synthetic LH–RH in patients with hypergonadotropic hypogonadism of various etiologies.

is significantly increased for both LH and FSH compared with normal patients. In patients with testicular feminization, synthetic LH–RH elicited an augmented FSH response and normal LH response when compared to that observed in normal male and females. Similar responses have been observed in patients with germinal aplasia or Sertoli cell only syndrome and in patients with azoospermia. The augmented secretion of FSH to synthetic LH–RH observed in these patients could be due to a modified LH–RH effect on the FSH gonadotropes by the absence of a hormonal factor from the germinal epithelium.

Studies by Odell and Swerdloff[9] in man and by Taleisnik *et al.*[10] in the rat demonstrating a positive gonadotropin feedback effect of progesterone in estrogen-primed castrate females but not in similarly treated males suggested the hypothesis of a sex-specific difference in hypothalamic function possibly related to differing perinatal exposure to endogenous androgens. In contrast, the observations of a positive feedback effect of progestin upon serum gonadotropins in estrogen primed castrate men by Stearns[11] and a positive feedback effect of estrogens in both male and female castrate rhesus monkeys by Karsch *et al.*[12] refute the existence of a sex specific difference in hypothalamic hypophyseal response. Our results showing a similar response in castrate men with the testicular feminization syndrome demonstrate that this positive feedback phenomenon is not sex-specific and cannot be used as evidence for a hypothalamic sex-related difference in primates.

CONCLUSION

It is concluded from this study that in testicular feminization: (1) there is a partial gonadal feedback control of both FSH and LH secretion, (2) hypothalamic receptors are not sensitive to the stimulatory effect of clomiphene, (3) there is an unusually large response of FSH release by the pituitary after intravenous LH–RH, (4) FSH and LH secretion is readily suppressed by estrogen administration, and (5) there is a positive feedback effect of progesterone upon serum gonadotropins in estrogen primed castrate patients with testicular feminization, (6) positive feedback effect of estradiol upon FSH and LH serum levels in castrated subjects could not be demonstrated.

REFERENCES

1. Swerdloff R. S. and Odell W. D.: *Lancet* **2** (1968) 633.
2. Shering R. J. and Loriaux D. L.: *J. clin. Endocr. Metab.* **36** (1973) 886.
3. Judd H. L., Hamilton C. R., Barlow J. J., Yen S. S. C. and Kliman B.: *J. clin. Endocr. Metab.* **34** (1972) 229.
4. Naftolin F. and Judd H. L.: In *Obstetrics and Gynecology Annual* (Edited by R. M. Wynn). Appleton-Century-Crofts, New York, 1973, p. 25.
5. Midgley A. R. Jr.: *Endocrinology* **79** (1966) 10.
6. Midgley A. R. Jr.: *J. clin. Endocr. Metab.* **27** (1967) 295.
7. Naftolin F., Ryan K. J. and Petro Z.: *J. clin. Endocr. Metab.* **33** (1971) 377.
8. Friedman S. and Goldfien A.: 53rd Meeting, Endocrine Society, 1971, A-509.
9. Odell W. D. and Swedloff R. S.: *Proc. natn. Acad. Sci., U.S.A.* **61** (1968) 529.
10. Talcisnik S., Cagliaris L. and Astrada J. J.: *J. Endocr.* **44** (1969) 313.
11. Stearns E. L., Winter J. S. D. and Faiman C.: *J. clin. Endocr. Metab.* **37** (1973) 635.
12. Karsch F. J., Dierschke D. J. and Knobil E.: *Science* **179** (1973) 484.