Effects of a Long-Acting Gonadotropin-Releasing Hormone Analog on the Pituitary-Ovarian-Adrenal Axis in Women With Severe Hirsutism

Camil Castelo-Branco, María J. Martínez de Osaba, Sergio Martínez, and Albert Fortuny

We evaluated modifications in the pituitary-ovarian-adrenal axis in severly hirsute women after administration of the gonadotropin-releasing hormone analog (GnRHa), D-Trp-6-luteinizing hormone–releasing hormone (LHRH) (Triptorelin) in a prospective study at a tertiary hospital. A total of 20 hirsute women aged 19 to 38 years were included. Hyperandrogenism of adrenal origin was excluded in all subjects. Patients received 3.75 mg D-Trp-6-LHRH intramuscularly (Decapeptyl 3.75; Lasa-Ipsen, Barcelona, Spain). Serum levels of follicle-stimulating hormone (FSH), LH, estradiol (E2), prolactin (PRL), testosterone (T), androstenedione (Δ4 An), dehydroepiandrosterone sulfate (DHEAS), 17-OH-progesterone (17-OHP), and sex hormone–binding globulin (SHBG) were determined before GnRHa administration, 24 and 48 hours after, and on days 7, 15, 30, and 45. GnRHa suppresses FSH, LH, and E2 in all women. Unexpectedly, adrenal steroids showed a flare-up phenomenon in the first days and subsequent decrease to lower values than before GnRHa administration. SHBG showed slight changes. After GnRHa, patients showed a significant decrease in T and Δ4 An: these hormones were reduced to half the basal levels. We conclude that GnRHa can potentially be used in the treatment of hyperandrogenism to reduce androgen levels in hirsute women.

CHRONIC HYPERANDROGENISM with varied degree of hirsutism is not uncommon in women of reproductive age, particularly in the younger age range. The causes of these phenomena lie in either an increased production of androgens or an increased sensitivity of peripheral target organs.

Actually, most treatments for hirsute women are only slightly effective. Although cosmetic measures may be helpful, they also have inconveniences such as skin intolerance, discomfort, and expense. The choice of endocrine therapy is a personal one, and traditionally the choices have included glucocorticoids, estrogens, and nonestrogenic antiandrogens.[1] Nevertheless, therapeutic results have not been totally satisfactory.

Gonadotropin-releasing hormone analogs (GnRHas) decrease ovarian steroid secretion and may be useful in the treatment of hyperandrogenism.[2] In the current study, we explored the effects of GnRHa on a variety of serum hormone levels in 20 women with hirsutism.

SUBJECTS AND METHODS

Patients

After provision of informed consent, 20 healthy women complaining of severe hirsutism were asked to participate in this study. None of the women had been on any form of drug therapy for 6 months before taking part in the study. The age of the subjects ranged from 16 to 38 years (mean ± SD, 22.4 ± 4.7). None of the women referred to the study presented with a personal or family history of heart disease, and none were hypertensive. All patients had intact uteri and ovaries. According to basal levels of steroids, hyperandrogenism of adrenal origin was unlikely since dehydroepiandrosterone sulfate (DHEAS) and 17-OH-progesterone (17-OHP) were within the normal range. Hypothyroidism and prolactinomas were excluded in all subjects. None of the patients smoked more than half a pack of cigarettes per day, and none were vegetarians. Hirsutism was evaluated according to Ferriman and Gallwey scores.[3]

Study Design

Before GnRHa administration, all patients underwent a history and physical examination, gynecologic examination, pelvic ultrasound, and hormonal assays. Patients received 3.75 mg D-Trp-6-luteinizing hormone–releasing hormone (LHRH) intramuscularly (Decapeptyl 3.75; Lasa-Ipsen, Barcelona, Spain) in the early follicular phase if they had a regular menstrual cycle. D-Trp-6-LHRH was available in a long-acting formulation[4] designed to release 100 μg of the compound per day (compound total dose, 3.75 mg). Serum levels of follicle-stimulating hormone (FSH), LH, estradiol-17β (E2), prolactin (PRL), testosterone (T), androstenedione (Δ4 An), DHEAS, 17-OHP, and sex hormone–binding globulin (SHBG) were determined before GnRHa administration, after 24 and 48 hours, and after 7, 15, 30, and 45 days. Blood samples were taken in the morning between 8:30 and 10:00 AM, after an overnight fast and tobacco abstinence.

Hormone Analysis

LH and FSH levels were measured by radioimmunoassay (RIA) (Farmos, Turku, Finland). LH and FSH standards were calibrated with IRP 68/40 and IRP 69/104. E2 was quantified by a coated-tube RIA (Medgenix, Brussels, Belgium). The E2 antibody cross-reacts 3.3% with E1, 1.0% with estriol, 2.0% with E2-3-glucuronide, and less than 0.2% with E2-17-glucuronide. PRL concentration was determined by a coated-tube RIA (Farmos). The PRL standard used was IRP 75/504. T and Δ4 An levels were measured by RIA after extraction with ether and purification by partition chromatography on a chromatolithe A column (bioMérieux Marcy l'Etoile, France). DHEAS level was measured by a coated-tube RIA (Diagnostic Products, Los Angeles, CA). The antiserum is highly specific for DHEAS, with a relatively low cross-reactivity to other steroids: 17β-E2 0.03%, E1 0.01%, E1-3-SO_4 0.5%, androsterone-SO_4 0.36%, T 0.10% and, Δ4 An 0.12%. SHBG level was measured by a two-site immunofluorometric assay (Wallac, Turku, Finland). No human serum protein is known to cross-react with the polyclonal-monoclonal antibody combination used in this assay.[5] The free androgen index (FTI) was calculated

From the Department of Gynaecology and Obstetrics and the Hormones Laboratory, Hospital Clínic i Provincial, University of Barcelona Medical School, Barcelona, Spain.

Submitted September 21, 1994; accepted April 11, 1995.

Address reprint requests to Camil Castelo-Branco, MD, PhD, Department of Gynaecology and Obstetrics, Hospital Clínic i Provincial de Barcelona, C/ Villarroel 170, 08036 Barcelona, Spain.

0026-0495/96/4501-0004$03.00/0

Table 1. Clinical Characteristics of 20 Hirsute Women Who Received GnRHa

Characteristic	Mean ± SD	Range
Age (yr)	22.48 ± 4.45	16-39
Hirsutism*	24 ± 5	16-32
Weight (kg)	64.1 ± 13.2	45-82
Body mass index (kg/m^2)	27.0 ± 2.3	25-29
Systolic blood pressure (mm Hg)	128.1 ± 19.0	
Diastolic blood pressure (mm Hg)	75.9 ± 8.3	
Menstrual rhythm (n)		
Eumenorrhea	13	
Oligomenorrhea	7	

*Ferriman-Gallwey.

with the formula (T × 3.47)/SHBG. The coefficients of variation have been previously described elsewhere.[5]

Statistical Analysis

Results are expressed as the mean ± SD. Statistical differences between baseline and treatment values were determined by one-way ANOVA. Significance was set at P less than .05. Results were analyzed using the Statistical Analysis Package (Walonick, Minneapolis, MN).

RESULTS

Clinical data of the women who participated in the study are shown in Table 1. No differences were found in clinical features among the subjects before the study, and no significant changes occurred in either body mass index or blood pressure during treatment. Ovarian volume evaluated by ultrasound using the formula for elipsoids was between 6 and 13 cm^3.

Levels of hormones at baseline and after GnRHa administration are presented in Table 2. As expected, there was an initial increase in production and secretion of gonadotropins and ovarian steroids, followed by a decrease in gonadotropin secretion with E2 at menopausal levels (< 50 pg/mL; Fig 1A). T and Δ4 An also showed an increase after GnRHa administration, and although it was of lesser degree, it persisted for a longer period than that of E2. Moreover, after 15 to 45 days, levels of T and Δ4 An were found to be reduced by 200% (Fig 2). No significant changes were detected in PRL and SHBG levels (Table 2). 17-OHP showed a pattern of stimulation-inhibition with GnRHa administration similar to that observed with E2 (Table 2 and Fig 1B): a sharp stimulation after 24 hours of treatment followed by a marked decrease after 48 hours. In addition, the 17-OHP levels were significantly lower as compared with basal levels after 30 days. Unexpectedly, DHEAS showed a pattern similar to T, with an initial increase during the first week, followed by a decrease that resulted in levels being 10% of basal at 30 to 45 days.

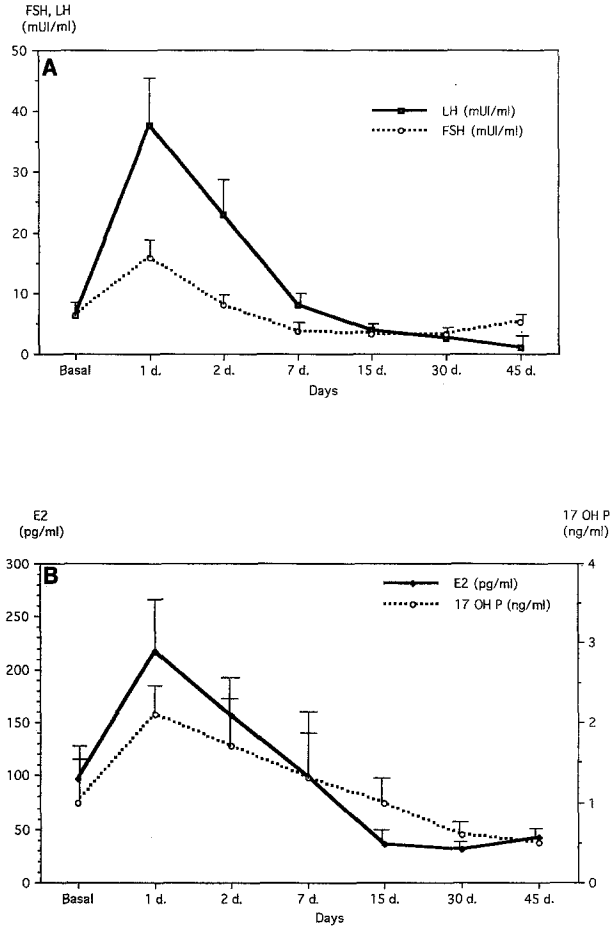

Fig 1. (A) Changes observed in FSH and LH levels following long-acting GnRHa administration. (B) Changes observed in E2 and 17-OHP levels following long-acting GnRHa administration. Values are the mean ± SD.

DISCUSSION

Manifestations of hyperandrogenism in women, such as hirsutism, acne, seborrhea, and alopecia, are most often a

Table 2. Levels of Pituitary Hormones, Sex Steroids, SHBG, and FTI

Parameter	Basal	1 d	2 d	7 d	15 d	30 d	45 d
PRL (ng/mL)	8.4 (6.6-10.2)	9.3 (7.2-11.5)	8.7 (6.3-11.2)	9.7 (6.8-12.6)	9.0 (6.6-11.5)	8.5 (6.1-10.8)	6.4 (4.9-8.0)*
LH (mUI/mL)	6.3 (3.7-8.9)	37.7 (29.6-45.9)*	22.9 (17.5-28.3)*	8.0 (6.1-9.9)	3.8 (2.0-5.6)	2.5 (0.6-4.3)	1.0 (0.5-1.5)*
FSH (mUI/mL)	6.2 (5.1-7.3)	15.9 (12.0-19.9)*	8.1 (6.5-9.7)	3.7 (3.0-4.4)*	3.3 (2.6-4.0)*	3.3 (2.5-4.1)*	5.1 (4.1-6.1)
E2 (pg/mL)	96.8 (71.1-122.4)	216.9 (161.9-271.8)*	156.1 (119.2-192.9)*	98.6 (22.0-175.2)	36.5 (29.2-43.8)*	30.4 (26.4-34.4)*	42.0 (33.9-50.1)*
T (ng/dL)	37.6 (31.7-43.5)	43.05 (33.8-52.2)	46.4 (36.3-56.5)*	40.9 (28.4-53.4)	29.1 (18.8-39.4)*	26.4 (15.5-37.4)*	22.1 (17.1-27.1)*
Δ4 An (ng/dL)	230.5 (200.5-260.5)	252 (210.4-293.6)	264 (222.1-305.9)	242.5 (175.2-309.7)	155.6 (122.7-188.5)*	148 (113.4-182.5)*	146.4 (132.9-159.8)*
DHEAS (μg/mL)	2.1 (1.8-2.5)	2.3 (1.9-2.8)	2.3 (1.9-2.7)	2.1 (1.7-2.5)	1.9 (1.6-2.2)	1.9 (1.6-2.2)*	1.9 (1.6-2.2)*
17-OHP (ng/mL)	1 (0.8-1.3)	2.1 (1.6-2.7)*	1.7 (1.4-2.1)*	1.3 (0.8-1.8)	1.0 (0.7-1.3)	0.6 (0.5-0.8)*	0.5 (0.4-0.7)*
SHBG (nmol/L)	48.7 (36.3-61)	49.2 (33.5-64.8)	51.1 (35.2-66.9)	48.5 (35.4-62.2)	49.2 (34.8-63.6)	52.3 (36.5-68.2)	54.6 (38.3-70.8)
FTI	3.1 (2.4-3.8)	3.7 (2.9-4.5)	3.8 (2.9-4.6)*	3.3 (2.4-4.3)	2.5 (1.7-3.3)*	2.2 (1.2-3.1)*	1.8 (1.2-2.4)*

NOTE. Values are expressed as the mean (95% CI).
*P < .05 v basal.

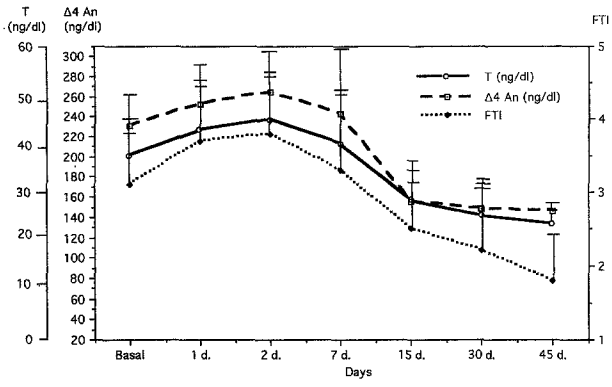

Fig 2. Changes observed in T, Δ4 An, and FTI levels after GnRHa administration. Values are the mean ± SD.

cosmetic problem that concerns the patient to varying degrees. Management of these entities is becoming increasingly important in daily gynecological practice. Therapeutic approaches to hirsutism are focused on suppression of the gland producing the hyperandrogenemia or the pilosebaceous unit. Antiandrogenic substances, such as flutamide,[6,7] cyproterone acetate,[8] and spironolactone,[6,9] presently constitute the main resources for the treatment of external signs of virilization. However, recent data suggest that the use of GnRHa may be helpful in the treatment of hirsutism.[2,10]

It is well known that the use of GnRHa has a desensitizing action on the pituitary gland, and that the action is characterized by significant decreases in gonadotropin and ovarian steroid levels.[11] Previous reports on the use of GnRHa in hirsute women demonstrate no action on adrenal steroid production.[10,12] However, in our study, triptorelin, a decapeptide GnRHa, not only signficantly reduced gonadotropins and ovarian steroids but also DHEAS.

The flare-up and subsequent decrease observed in DHEAS and 17-OHP levels could be due to a direct extrapituitary action of GnRHa or to a LH effect on the theca cells or adrenal gland. Studies in animals and humans have demonstrated this extrapituitary effect on ovarian cells,[13-15] and additionally, it is recognized that LH can stimulate adrenal production of androgens.[16]

The preferential enzymatic route for conversion of precursors to androgens in the ovary and adrenal gland is via the 5-ene-3β-hydroxysteroid pathway,[17] and the final product is DHEA, metabolized to DHEAS and Δ4 An. LH action via specific receptors present on the theca cells provides the principal stimulus for these steroidogenic activities.[18] Transiently increased production of 17α-OHP in the human at midcycle may arise in theca cells before similar pathways are fully active in granulosa cells.[19,20] The steroidogenic action of LH on theca cells increases the activities of 17α-hydroxylase:C17,20-lyase.[21,22] These enzyme activities are rate-limiting and seem to be the site at which LH stimulates C19-steroid production by theca cells.[22]

Ehrmann et al[23] have recently demonstrated the usefulness of testing with GnRHa to distinguish ovarian causes of hyperandrogenism in women. In their study, they found significant increases of 17-OHP without detecting modifications in DHEAS levels after administration of nafarelin, suggesting an ovarian cause of the androgen excess in their subjects.

The significant decrease in PRL levels observed after 30 days of GnRHa administration may be explained by the previous decrease of estrogen levels.

The overall observed effect of D-Trp-6-LHRH on androgen synthesis suggests that it rapidly and effectively suppresses ovarian function and defines its potential use in the treatment of hyperandrogenic states in which suppression of such steroids may be of therapeutic benefit. However, in such patients, suppression of other sex steroids such as estrogens and progestogens is an undesirable side effect and requires replacement therapy.[10,24] Additionally, one must keep in mind that, as with the majority of drugs used in hyperandrogenic states, treatment with GnRHa also carries some risk of side effects, and it is therefore especially important to carefully assess its use and discontinue treatment if it is ineffective.

REFERENCES

1. Jeffcoate W: The treatment of women with hirsutism. Clin Endocrinol (oxf) 39:143-150, 1993

2. Rittmaster RS: Use of gonadotropin-releasing hormone agonists in the treatment of hyperandrogenism. Clin Obstet Gynecol 36:679-689, 1993

3. Ferriman D, Gallwey JD: Clinical assessment of body hair growth in women. J Clin Endocrinol Metab 21:1440-1447, 1961

4. Schally AV, Redding TW, Coramu-Schally AM: Inhibition of prostate tumors by agonistic and antagonistic analogs of LH-RH. Prostate 4:545-552, 1983

5. Castelo-Branco C, Martínez de Osaba MJ, Vanrell JA, et al: Effects of oophorectomy and hormone replacement therapy on pituitary-gonadal function. Maturitas 17:101-111, 1993

6. Cusan L, Dupont A, Gómez JL, et al: Comparison of flutamide and spironolactone in the treatment of hirsutism: A randomized controlled trial. Fertil Steril 61:281-287, 1994

7. Cusan L, Dupont A, Bélanger A, et al: Treatment of hirsutism with the pure antiandrogen flutamide. J Am Acad Dermatol 23:462-469, 1990

8. Falsetti L, Dordoni D, Gastaldi C, et al: A new association of ethinylestradiol and cyproterone acetate in the therapy of polycystic ovary syndrome. Acta Eur Fertil 17:19-25, 1986

9. Tremblay RR: Treatment of hirsutism with spironolactone. J Clin Endocrinol Metab 15:363-371, 1986

10. Falsetti L, Pasinetti E: Treatment of moderate and severe hirsutism by gonadotropin-releasing hormone agonists in women with polycystic ovary syndrome and idiopathic hirsutism. Fertil Steril 61:817-822, 1994

11. Balasch J, Gómez F, Casamitjana R, et al: Pituitary-ovarian suppression by the standard and half-doses of D-trp-6-luteinizing hormone–releasing hormone depot. Hum Reprod 7:1230-1234, 1992

12. Falsetti L, Pasinetti E, Chioda C, et al: Treatment of moderate and severe hirsutism with a gonadotrophin-releasing hormone agonist. Hum Reprod 7:894, 1992

13. Hsueh AJW, Erickson GF: Extrapituitary action of gonadotropin-releasing hormone: Direct inhibition of ovarian steroidogenesis. Science 204:854-855, 1979

14. Pellicer A, Miró F: Steroidogenesis in vitro of human

granulosa cells pretreated in vitro with gonadotropin-releasing hormone analogs. Fertil Steril 54:590-596, 1990

15. Loumaye E, Coen G, Pampfer S, et al: Use of gonadotropin-releasing hormone agonist during ovarian stimulation leads to significant concentration of peptide in follicular fluids. Fertil Steril 52:256-263, 1989

16. Richardson GS: Hormonal physiology of the ovary adrenal cortex, in Gold JJ, Josimovitch JB (eds): Gynecologic Endocrinology. New York, NY, Plenum, 1987, p 65

17. Aakvaag A: Pathways in the biosynthesis of androstenedione in the human ovary in vitro. Acta Endocrinol (Copenh) 60:517-526, 1969

18. Dennefors BL, Janson P, Knutson F, et al: Steroid production and responsiveness to gonadotrophin in isolated stromal tissue of human menopausal ovaries. Am J Obstet Gynecol 136:997-1002, 1980

19. Florensa E, Sommerville IF, Harrison RF, et al: Plasma 20α-dihydroprogesterone, progesterone, and 17-hydroxyprogester one: Daily and four-hourly variations during the menstrual cycle. J Steroid Biochem 7:769-777, 1976

20. Holmdahl TH, Johansson EDB: Peripheral plasma levels of 17α-hydroxyprogesterone, progesterone and oestradiol during normal menstrual cycle in women. Acta Endocrinol (Copenh) 71:743-754, 1972

21. Aono T, Kitamura Y, Fukuda S, et al: Localization of 4-ene-5α-reductase, 17β-O_1-dehydrogenase and aromatase in immature rat ovary. J Steroid Biochem 14:1369-1377, 1981

22. Bogovich K, Richards JS: Androgen biosynthesis in developing ovarian follicles: Evidence that luteinizing hormone regulates thecal 17α-hydroxylase and C_{17-20}-lyase activities. Endocrinology 111:1201-1208, 1982

23. Ehrmann DA, Rosenfield RL, Barnes RB, et al: Detection of functional ovarian hyperandrogenism in women with androgen excess. N Engl J Med 327:157-162, 1992

24. Carmina E, Janni A, Lobo R: Physiological estrogen replacement may enhance the effectiveness of the gonadotropin-releasing hormone agonist in the treatment of hirsutism. J Clin Endocrinol Metab 78:126-130, 1994