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An Overview of LHRH and Its Analogues: Clinical Uses

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BACKGROUND

Two experimental observations underlie all of the apparently diverse clinical applications of LHRH and its analogues. The first of these experiments is the now classic studies of Knobil's group in which arcuate nucleus-lesioned and pre-pubertal (1,20) Rhesus monkeys (both LHRH deficient) were replaced with exogenous LHRH. In this ablation and replacement model, continous administration of natural sequence LHRH was unable to restore physiologic levels of gonadotropin secretion. Episodic delivery of 1 μg/kg of LHRH, however, at a frequency designed to mimic the endogenous gonadotropin pulsations observed in castrate animals, was capable of restoring appropriate gonadotropin secretion to these animals. Moreover, increasing the duration of exposure of the gonadotroph to LHRH resulted in decreasing levels of gonadotropin secretion (14) and thus fit the general criteria of pharmacologic desensitization of pituitary gonadotropin secretion, i.e. decreasing responsiveness with increasing doses of stimulation.

The second experimental observation was made by our group in LHRH deficient adult males with hypogonadotropic hypogonadism employing a long-acting LHRH analogue, D-Trp6-Pro9-Net-LHRH (LHRHa) (9). Alternate day administration of 50 mcg of this potent LHRH agonist was capable of producing a stable pulse of gonadotropin elevation for 8 hours, whereas daily administration of the same dose totally ablated pituitary responsiveness to LHRHa administration only to have it restored by either returning the frequency of LHRHa administration to alternate days or lowering the daily dosage. By coupling these two observations, it thus became clear that intermittency of pituitary gonadotroph stimulation by LHRH was absolutely essential for physiologic secretion whereas continuous receptor occupancy by either LHRH or its long-acting analogues could produce an effective and selective blockade of gonadotropin secretion. The selectivity of this blockade is imparted to the system by the specificity of the releasing factor and occurs independently of alterations of any other anterior pituitary function.

Subsequently, numerous corollaries of these central observations have appeared and several clinical applications have stemmed from these two series of experiments. For example, it would appear that the episodic secretion of gonadotropins, previously documented during various developmental stages (3,19), in fact mirrors an intermittency of hypothalamic stimulation to the pituitary and thus can be taken as the first derivative of hypothalamic function. This observation is especially true since direct measurements of peripheral LHRH levels are plagued by numerous methodologic difficulties by virtue of the peculiarities of the hypophyseal portal blood supply, rapid cleavage of the peptide, and low circulating serum levels of this hypothalamic releasing factor. Secondly, not only is it evident that intermittency of hypothalamic stimulation is required for physiologic functioning of the gonadotroph, it also appears that such pulsatility is most likely achieved by intermittent administration of natural sequence LHRH since the rapid cleavage and metabolism of this hypothalamic releasing factor is an essential physiologic property of the neuroendocrine control of reproduction. Thirdly, continous receptor occupancy can be achieved by administration of either a pure LHRH antagonist or by the paradoxical use of long acting LHRH agonists which assure continuous receptor occupancy for periods of 24 hours or greater. It would thus seem that a good working model of the hypothalamic-pituitary-gonadal axis can be represented by the analogy of a computer. The hardware of this system is represented by the hypothalamic neurons which synthesize and secrete LHRH, the gonadotrophs which are responsible for LH and FSH production and release, and the gonads which respond when appropriately recruited with gametogenesis and steroid secretion. However, this elegantly integrated system will not function if the "software" component, i.e. the hypothalamic secretory program of LHRH, is not functioning properly. Furthermore, physiologic secretion of gonadotropins and fertility are critically dependent upon such a precise program of episodic hypothalamic secretion.

With these observations as a background, two broad categories of application of LHRH and its analogues readily emerge. The first of these clinical uses are those circumstances in which the "fertility program" of the reproductive system is to be called up by intermittent administration of the natural sequence LHRH in a physiologic dosage and frequency. The second broad category of clinical applications is the antifertility effects of LHRH and these can be evoked by less frequent administration of LHRH analogues either agonistic or antagonistic. Since the chemistry of the antagonists has lagged considerably behind the agonists, for most practical purposes the LHRH agonists have been used in this paradoxical manner to desensitize the pituitary selectively and ablate the functioning of the pituitary-gonadal axis, thus producing a reversible "chemical castration".

PROFERTILITY APPLICATIONS OF LHRH

General

Using portable infusion pumps which are small and lightweight and can be programed to any desired frequency of intermittent stimulation (Ferring), it has been possible to institute "hypothalamic replacement therapy" in both men and women with LHRH deficiency (8,12). In each of these therapeutic circumstances the selection of the frequency of LHRH administration has been based upon physiologic data available in the literature or normative data acquired by our own group. The dosage of LHRH has been selected to achieve circulating LHRH levels similar to those achieved in the hypophyseal circulation and has been adjusted by determining the amplitude of pituitary gonadotropin pulsations as well as the degree of gonadal responsiveness.

Hypogonadotropic Males

Men with idiopathic hypogonadotropic hypogonadism (IHH) have proven to be excellent models in which to investigate the physiology of LHRH in the human male and to initiate hypothalamic replacement therapy for the purpose of restoring normal pituitary and gonadal physiology (12). Each of these males was carefully selected for study after having failed to undergo puberty by age 18 and demonstrated a serum testosterone in the prepubertal range when all replacement therapy was removed for a minimum of 3 months. Normal or low levels of serum gonadotropins were documented in the face of these prepubertal testosterone levels and each subject had no evidence of other neuroendocrine or pituitary disease by insulin tolerance and TRF testing as well as a normal CT scan of the hypothalamic-pituitary area. A gonad capable of responding was documented by both hCG stimulation and testicular biopsy prior to therapy. Following an extensive evaluation of their baseline characteristics, each male was treated with natural sequence LHRH (generously supplied by Drs. Jean Rivier and Wylie Vale of the Salk Institute) at a 2 hour frequency via a subcutaneous route. The subcutaneous route was chosen in view of the long-term nature of the replacement therapy required to produce normalization of the pituitary-gonadal axis and spermatogenesis. The frequency of LHRH administration was based upon previous studies of adult males in which endogenous LH pulses occurred at approximately 2 hour frequency (18). The dose of LHRH was empirically adjusted utilizing the serum testosterone level obtained during bi-weekly 8 hour admissions to our Clinical Research Center. These doses spanned a range of 25 to 300 ng/kg.

The clinical results of this replacement therapy in the first 17 consecutive patients were quite striking as all of the signs and symptoms of puberty were encountered as listed on Table I. A uniform response of an ejaculate was

Table I

Clinical Responses to Pulsatile LHRH Therapy

1. Increased Libido	(17/17)	100 %
2. Increased Erections	(17/17)	100 %
3. Appearance of Ejaculate	(17/17)	100 %
4. Increased Facial Hair	(15/17)	88 %
5. Deepened Voice	(14/17)	82 %
6. Transient Gynecomastia	(8/17)	47 %
7. Sperm in Ejaculate	(8/12)*	67 %

* Includes only the 12 subjects who remained on therapy long enough to sustain a normal testosterone level for at least 3 months.

noted in all subjects. A normalization of the serum testosterone level (Figure 1) occurred in all but 2 patients in whom an LHRH antibody developed during the course of treatment prior to normalization of his pituitarygonadal axis. In addition, a tripling of testicular size occurred by the 3-6 month period and appearance of mature sperm in the ejaculate was noted in two-thirds of the patients in whom normalization of pituitary gonadotropin secretion and testo-

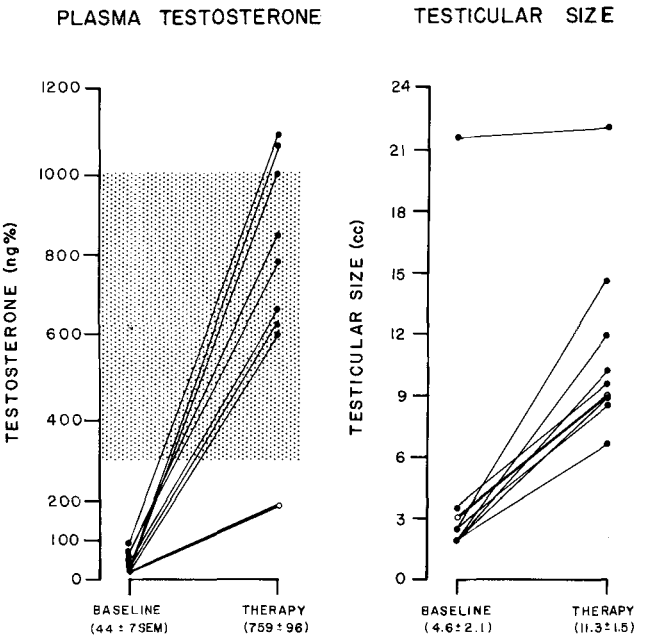

Fig. 1 Plasma testosterone levels and testicular size before and during LHRH replacement therapy of 9 men with hypogonadotropic hypogonadism. Data from Hoffman & Crowley (12).

sterone had been maintained for a minimum of 6 months. The gonadotropin dynamics previously reported in these subjects (12) are most interesting in that it appears that supraphysiologic gonadotropin levels are required to initiate gonadal steroidogenesis in these men with essentially prepubertal gonads prior to LHRH treatment. Castrate levels of gonadotropins have been observed in these patients during the 5th to 7th week despite the fact that normal circulating levels of testosterone and estradiol were achieved, suggesting that the absence of a hypothalamic locus of feedback occurring in these men was responsible for this "overshooot" of gonadotropin levels. Moreover, these patients are capable of impregnating their wives and achieving a total normal fertility with this treatment (12).

PULSATILE LHRH FOR INDUCTION OF OVULATION

Derangements of endogenous LHRH secretion are considerably more common in females than in males and manifest themselves as two basic clinical diagnoses. Amenorrhea, which occurs in 3 % of the female population aged 18-35, has a predominantly "hypothalamic" etiology. More recently it has been demonstrated by our group that the common hypothalamic defect in this heterogeneous group of disorders is one of faulty endogenous LHRH secretion. Chronic anovulation, yet another manifestation of aberrant endogenous LHRH secretion, represents 10-15 % of infertile women. Thus, in theory, both of theses diagnoses might prove amenable to replacement therapy with exogenous LHRH. This potential mode of therapy would thus be important since the existing treatments of theses conditions have substantial limitations. Clomiphene citrate, an anti-estrogen which is capable of stimulating endogenous LHRH secretion, is the agent of choice in normoprolactinemic hypothalamic amenorrhea as well as anovulatory infertility. Its advantages are that it is relatively inexpensive, orally active, and requires little in the way of monitoring. On the other hand, it is associated with an increased frequency of multiple gestation and mild ovarian hyperstimulation (11). Most importantly, this agent works in only 50-80 % of subjects with this disorder (11). Human gonadotropin injections (Pergonal) represent a direct approach to ovarian stimulation which requires very expensive parental injections, often results in multiple gestations as well as the ovarian hyperstimulation syndrome, and necessitates extensive monitoring by ultrasound and estrogen levels (4). Thus a new treatment of hypothalamic amenorrhea and anovulatory infertility would be most welcome.

In attempting to understand the pathophysiology of the disorder in hypothalamic amenorrhea and anovulatory infertility and its difference from normal, we have performed detailed studies of ovulatory women in the menstrual cycle

in which frequent gonadotropin samples were obtained over the course of a 24 hour period to understand the normal sequence of amplitude and frequency changes of LHRH-induced gonadotropin pulsations across the normal menstrual cycle and thus be able to define more precisely the differences between these changes and those aberrancies of hypothalamic LHRH secretion seen in amenorrheic and anovulatory women. Utilizing this information, we have determined that the vast majority of patients with hypothalamic amenorrhea have either disorders of the frequency and/or amplitude of LHRH-induced gonadotropin pulsations. Consequently these patients represent the female counterpart of hypogonadotropic males and as such should prove equally amenable to treatment with pulsatile LHRH. However, the precise dosage, frequency, and route of LHRH administration is considerably more complicated in the female than in the male. The reasons for this difficulty are manyfold. First, the frequency and amplitude of normal LHRH-induced gonadotropin pulsations varies strikingly across the ovulatory menstrual cycle. Secondly, the heterogeneity of baseline abnormalities of LHRH-induced gonadotropin pulsations in female patients with hypothalamic amenorrhea and anovulatory infertility is considerably more evident than in hypogonadotropic males. Consequently these women have varying degrees of "ovarian readiness" which in turn translates to a variability of clinical responsiveness. These factors have made the task of inducing ovulation in females with pulsatile LHRH considerably more complex than inducing puberty in males. In fact, the remainder of this conference will address the variability of dosage regimens and routes of LHRH administration in anovulatory women. These factors notwithstanding, it appears that a remarkably diverse group of hypothalamic replacement regimens of LHRH have proven capable of inducing ovulation and certain generalizations regarding this mode of therapy appear justified. It appears that intravenous LHRH administration is more effective than the subcutaneous route in these women and that a dosage of LHRH of 1-5 μg intravenuously or 5-10 μg subcutaneously represents the most "physiologic" replacement dosages. Similarly, the various frequencies of LHRH administration to be reported herein demonstrate a convergence upon the 60-90 minute frequency and the results of these studies confirm virtually uniform success rates in hypothalamic amenorrhea when all of these factors are taken into consideration. Figure 2 represents the first published case of an LHRH deficient female (Kallmann's syndrome) in whom LHRH replacement was capable of simulating all of the events of a normal menstrual cycle (6). It is clear from studies such as these that in LHRH deficient patients with an intact pituitary and ovaries that the entire process of folliculogenesis, ovulation, and formation and demise of the corpus luteum can be modulated with a fixes input of exogenously administered LHRH. Since this treatment will largely be the focus of other

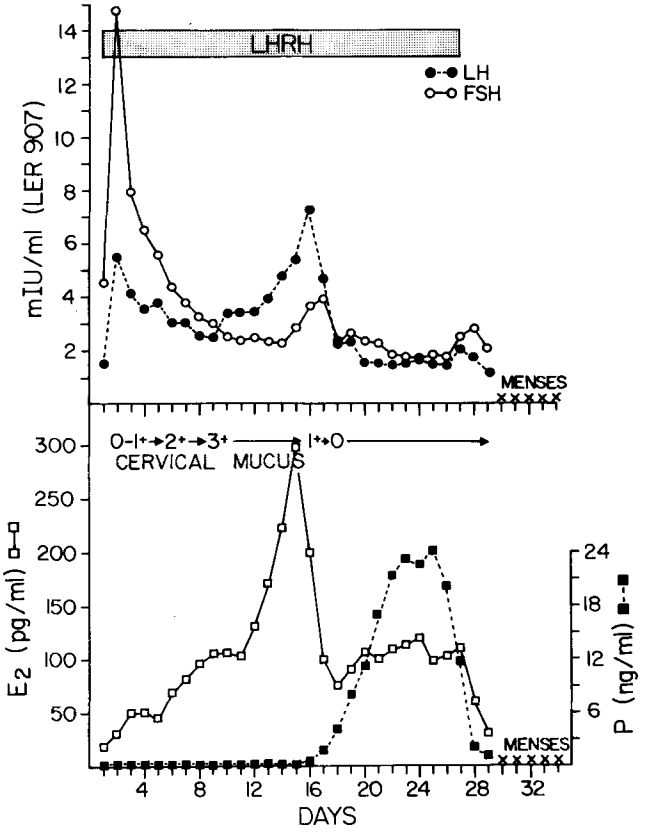

Fig. 2 Serum FSH, LH, estradiol and progesterone correlated with cervical mucus characteristics in a 24-year-old woman with Kallman's syndrome during 27-days of pulsatile LHRH therapy. Ultrasound examination on day 13 revealed a single follicle in the right ovary. Reproduced from Crowley & McArthur (6), with permission.

other presentations, it will not be disussed in further depth here. Suffice it to say that in 15 patients with a variety of hypothalamic disorders varying from congenital LHRH deficiency to hypothalamic amenorrhea in association with stress and post-recovery from weight loss, our experience concurs with those to be presented today and indicates that pulsatile LHRH administration represents a generally successful form of therapy which may well replace exogenous gonadotropins in the very near future.

ANTIFERTILITY EFFECTS OF LHRH ANALOGUES

Utilizing LHRH agonists as a method of continuously occupying the LHRH receptor on the pituitary membrane, it has been possible to effect a selective chemical castration therapy in a wide variety of circumstances. The first two

indications in which this method of treatment was published were the use of LHRH agonists to inhibit ovulation as a new method of contraception (17) and its use in precocious puberty (7). By means of single daily injections of an LHRH analogue, D-Trp6-Pro9-Net-LHRH, we have demonstrated dramatic clinical responses of children with centrally mediated precocious puberty. In 52 such patients treated to date, there has been a uniform suppression of all endogenous gonadotropin pulsations which were previously present as a marker of hypothalamic maturity prior to treatment in all of these children. In addition, a total ablation of the responsiveness of the pituitary to exogenous LHRH testing has been documented. A suppression of gonadal steroidogenesis to pre-pubertal levels has been observed accompanied by equally dramatic clinical responses including uniform cessation of menses, a striking arrest of regression of breast growth in females, and a decrease to 50 % of pretherapy levels of the testicular volume in the males. Thus it is clearly possible to induce suppression of the pituitary-gonadal axis in a safe and effective way using daily LHRHa administration. In addition, all of the patients withdrawn from LHRHa therapy following 2 months, 10 months and 3 years of continuous LHRHa administration have demonstrated a prompt return of spontaneous activity of the hypothalamic-pituitary-gonadal axis and reinitiation of the pubertal process (15). More recently we have investigated the impact of this selective removal of gonadarche from the pubertal process upon both growth and adrenal maturation since this therapeutic probe now permits us to pursue such a novel line of investigation (15). Similar approaches and successes have also been reported with LHRHa induced suppression of the pituitary in endometriosis (16); polycystic ovarian disease (5); metastatic prostate (2) and breast cancer (13); and uterine fibroids (10).

Thus it appears that LHRH and its analogues represent a major therapeutic advance in several areas of reproductive medicine. Theses agents will have an impact upon Pediatrics, Urology, Medical Oncology and Gynecology. These agents will permit a selective chemical castration to occur in several conditions in which such a removal of sex steroids is beneficial.

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