Pulsatile low dose
luteinizing hormone-releasing hormone treatment for
induction of follicular maturation
and ovulation in women with amenorrhoea

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Abstract. Chronic intermittent low dose luteinizing hormone-releasing hormone (LRH) therapy was given to 7 normoprolactinaemic women with long-standing secondary amenorrhoea, low-normal gonadotrophin levels and low endogenous oestrogen production (WHO group I). Four of them were involuntarily infertile. A portable, computerized infusion pump (Zyklotat, Ferring) delivered 20 μg LRH every 90 min through a chronic indwelling catheter inserted subcutaneously in the fat tissue of the lower abdominal wall. The LRH administration was continued until menstruation or pregnancy. Nine treatment courses over 21 to 69 days were given. Twelve ovulatory cycles were induced by the pulsatile LRH treatment. The corpus luteum function was normal in 7 cycles. Temporary interruptions of the pump function in 2 patients and haematomas at the catheter site in another 2 patients may have caused the luteal phase insufficiency. One of the 4 infertile women became pregnant. She developed ovarian hyperstimulation during her first treatment cycle. The ovarian enlargement regressed despite continued LRH therapy and a second ovulatory cycle was induced.

Thus, pulsatile administration of low doses of LRH can induce follicular maturation, ovulation and normal corpus luteum function in hypo- or normogonadotrophic women with secondary amenorrhoea with low endogenous oestrogen production. The LRH therapy can be given with the same pulse frequency throughout the induced cycle. Should pregnancy not occur the pulsatile LRH treatment can be continued without interruption to induce a second ovulatory cycle.

Chronic intermittent high dose LRH treatment can induce follicular maturation and ovulation in women with amenorrhoea (see Nillius 1979 for review). By prolonged pulsatile treatment with 500 μg LRH every 8 h it is possible to produce normal ovulatory cycles in women devoid of ovarian activity before the treatment (Nillius et al. 1975). Recent studies on the neuroendocrinological control of the rhesus monkey menstrual cycle by Knobil et al. (1980) have stimulated interest in chronic intermittent low dose LRH treatment of women with amenorrhoea. The low doses of LRH (1–20 μg) have to be administered at shorter intervals (60–120 min) to produce optimal gonadotrophin secretion. Pulsatile low dose LRH treatment has recently proved to be effective in inducing normal ovulatory menstrual cycles in hypo-gonadotrophic women with primary and secondary amenorrhoea (Leyendecker 1979; Crowley & McArthur 1980; Keogh et al. 1981; Shoemaker et al. 1981). Small portable, computerized infusion pumps have made this mode of LRH administration practically possible for clinical use.

Here we report on use of the computerized infusion pump Zyklotat (Ferring) for chronic intermittent low dose LRH treatment of 7 normoprolactinaemic women with secondary amenorrhoea, low-normal serum gonadotrophin levels and low endogenous oestrogen secretion (WHO group I). Follicular maturation and ovulation were induced in all the women by the pulsatile low dose LRH treatment.
Materials and Methods

Patients

Seven women, aged 20–32 years (median 26 years) with secondary amenorrhoea of 2.5–11 years duration (median 4 years) volunteered for the LRH treatment. Four of them (patient 2, 3, 4 and 5) were involuntarily infertile. Six of them had developed amenorrhoea in relation to weight loss but had regained normal weight when the treatment was instituted.

Hormonal evaluation

All had normal prolactin (Prl) levels in serum and normal sellar X-ray. The thyroid and adrenal function was normal. None responded with withdrawal bleeding after progesterone injection (50 mg im). Clomiphene citrate was ineffective in inducing ovulation. The pre-treatment basal gonadotrophin levels and their responses to iv LRH ( Hoechst AG, 100 µg) are shown in Table 1. All the women had follicle-stimulating hormone (FSH) levels in serum within the normal follicular phase range, while 3 of them (patients 1, 3 and 7) had luteinizing hormone (LH) levels below that range. All but one of the volunteers (patient 5) responded with gonadotrophin release to LRH. Patient 2 had exaggerated FSH and LH responses to LRH at her first pre-treatment test. Before her second treatment course the basal LH level was below the normal follicular phase range while the basal FSH level was normal. The gonadotrophin response pattern to LRH was then prepubertal with greater FSH than LH increase. A prepubertal response pattern to LRH was seen in 2 other women (patients 1 and 6). Exaggerated FSH response to LRH was seen in 2 of the women (patients 1 and 6) and 2 other had exaggerated LH responses (patients 4 and 5).

LRH therapy

The chronic intermittent LRH treatment was administered by means of a portable computerized infusion pump (Zyklotat, Ferring GmbH, Kiel, FRG). The pump was connected to a chronic indwelling catheter inserted sc into the fat tissue of the lower abdominal wall. The same catheter was used throughout the treatment course(s) except in one woman (patient 5). The pump delivered 20 µg LRH (Ferring) in 50 µl solution during 1 min every 90 min. The LRH solution contained 62.5 IU heparin per ml. The women were treated as outpatients. The treatment did not interfere with their daily life activities.

The therapy was monitored by clinical examinations, basal body temperature (BBT) recordings and frequent venous blood samples for determinations of FSH, LH, oestriadiol and progesterone.

The LRH administration was continued with the same pulse frequency until menstruation or pregnancy. In 3 of the 4 infertile women (patients 2, 3 and 5) the treatment was continued without interruption to induce a second ovulatory cycle when they did not conceive during the first ovulatory cycle.

Table 1.

Pre-treatment serum gonadotrophin levels and gonadotrophin response to iv LRH (100 µg) in 7 women with secondary amenorrhoea.

<table>
<thead>
<tr>
<th></th>
<th>Serum FSH (µg/l)</th>
<th>Serum LH (µg/l)</th>
</tr>
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<tbody>
<tr>
<td></td>
<td>Pre-treatment</td>
<td>Response to</td>
</tr>
<tr>
<td></td>
<td></td>
<td>LRH</td>
</tr>
<tr>
<td>Normal mean</td>
<td>1.1</td>
<td>0.3</td>
</tr>
<tr>
<td>and range*</td>
<td>(0.4–3.1)</td>
<td>(0–1.1)</td>
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<tr>
<td>Patients</td>
<td></td>
<td></td>
</tr>
<tr>
<td>1</td>
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<td>1.2</td>
</tr>
<tr>
<td>2</td>
<td>1.8</td>
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<td>3</td>
<td>0.4</td>
<td>0</td>
</tr>
<tr>
<td>4</td>
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<tr>
<td>5</td>
<td>1.8</td>
<td>1.0</td>
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<tr>
<td>6</td>
<td>0.9</td>
<td>2.0</td>
</tr>
<tr>
<td>7</td>
<td>1.2</td>
<td>0.8</td>
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</table>

* Early follicular phase, n = 10 (from Bergh et al. 1978).

** Second treatment course.
Table 2.
A summary of the results of the pulsatile low dose LRH therapy in 7 women with secondary amenorrhoea.

<table>
<thead>
<tr>
<th>Patient</th>
<th>Treatment course length (days)</th>
<th>Menstrual cycle length (days)</th>
<th>Pre-treatment oestradiol (pmol/l)</th>
<th>Oestradiol maximum (pmol/l)</th>
<th>Progesterone maximum (nmol/l)</th>
<th>Luteal phase length (days)</th>
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<td>23</td>
<td>35</td>
<td>1020</td>
<td>30</td>
<td>20</td>
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<tr>
<td>2</td>
<td>49*</td>
<td>21</td>
<td>40</td>
<td>10200</td>
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<tr>
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<tr>
<td>3</td>
<td>69*</td>
<td>37</td>
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<td>71</td>
<td>1260</td>
<td>70</td>
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</table>

* Two consecutive induced menstrual cycles.

**Criteria of ovulation and normal corpus luteum function**

An increasing oestradiol level in serum to above 500 pmol/l followed by an increase in the serum progesterone concentration to at least 6 nmol/l were taken as criteria for follicular maturation and ovulation. The corpus luteum function was judged to be normal if the maximal progesterone concentration in serum exceeded 32 nmol/l and the luteal phase had a duration of at least 10 days.

**Hormone assay methods**

FSH and LH in serum were measured by a radioimmunosorbert technique with indirectly coupled antibodies (Wide et al. 1973). The results were expressed in μg/l using highly purified FSH and LH preparations as reference standards (Roos 1968; Roos et al. 1975). Normal values of gonadotrophins in serum during the early follicular phase of the menstrual cycle are given in Table 1. Immunoreactive oestradiol in serum was measured by using a radioimmunoassay, in principle according to Hotchkiss et al. (1971), using [2,4,6,7-3H]oestradiol-17ß and an antiserum (Miles-Yeda Ltd.) to oestradiol-6-(O-carboxymethyl)-oxide-BSA conjugate. The reference ranges for oestradiol in serum were: early follicular and late luteal phase: 60–200 pmol/l; midcycle peak: 500–1300 pmol/l. Progesterone was assayed by a similar radioimmunomethodological method.

**Results**

A summary of the results of the LRH treatment is given in Table 2. Twelve menstrual cycles were induced during nine treatment courses in the 7 volunteers. The pulsatile low dose LRH therapy induced follicular growth and maturation, as evidenced by increased oestradiol levels in serum, in all the women. All the cycles were ovulatory. An ovulatory cycle with normal corpus luteum function induced by the pulsatile low dose LRH therapy is illustrated in Fig. 1.

Normal corpus luteum function occurred in 7 of the 12 induced menstrual cycles. A maximal progesterone level of below 32 nmol/l was seen in 5 cycles, indicating corpus luteum insufficiency. In 2 of them, the infusion pump temporarily stopped functioning during the luteal phase and then the progesterone levels decreased. This is illustrated in Fig. 2 which shows a cycle with a normal pre-ovulatory oestradiol peak and a midcycle-like LH surge followed by an increase of the progesterone concentration up to a plateau of about 30 nmol/l. The pulsatile LRH administration was then interrupted when the pump stopped to function during...
Serum levels of FSH, LH, oestradiol and progesterone during a normal ovulatory cycle induced by sc pulsatile low-dose LRH treatment of a 29-year-old woman with secondary amenorrhoea (patient 6). FSH and LH response to iv LRH before and after treatment are also shown.

Fig. 1.

Serum levels of FSH, LH, oestradiol and progesterone during an ovulatory cycle induced by sc pulsatile low-dose LRH treatment of a 20-year-old woman with secondary amenorrhoea (patient 1). The luteal phase is inadequate probably due to temporary interruption of pump function. FSH and LH response to iv LRH before and after treatment are also shown.

Fig. 2.
Serum levels of FSH, LH, oestradiol and progesterone during an ovulatory cycle induced by sc pulsatile low-dose LRH treatment of a 26-year-old woman with secondary amenorrhoea (patient 4). The corpus luteum function is insufficient.

36 h. The progesterone level rapidly decreased to 6 nmol/l. Reactivation of the corpus luteum function occurred when the LRH therapy was reinitiated. The progesterone level increased to 16 nmol/l during the menstrual-like bleeding. Luteal phase insufficiency with a maximal progesterone concentration of 14 nmol/l also occurred during the second treatment course in patient 4 (Fig. 3). A local sc bleeding at the catheter site may have interfered with the absorption of LRH in this patient.

One pregnancy occurred in the 4 infertile women. Patient 2 who became pregnant was treated twice. Two menstrual cycles were induced during the first treatment course (Fig. 4). Ovarian hyperstimulation occurred during the first cycle. The oestradiol level in serum rapidly increased to a maximum of 10 200 pmol/l on treatment day 9 and ultrasonography showed multiple ovarian cysts with a diameter of up to 5 cm. The ovarian enlargement regressed despite continued LRH therapy and a new ovulatory cycle was induced. The pump was out of function during 3 days and then the gonadotrophin and oestradiol levels rapidly decreased. When the LRH administration was reinitiated both gonadotrophin and oestradiol levels increased to normal mid-cycle concentrations. Ovulation then occurred. The corpus luteum function was insufficient with a maximal progesterone level of 25 nmol/l. The pump stopped again after 10 days of treatment, oestradiol and progesterone level rapidly decreased and a uterine bleeding ensued. The LRH therapy was re instituted 3 weeks later. The patient conceived during this ovulatory cycle (Fig. 5). Follow-up with ultrasonography has shown a normally developing singleton pregnancy.

Two consecutive ovulatory cycles with normal corpus luteum function were induced by uninterrupted pulsatile LRH therapy in patient 3 (Fig. 6). She had no FSH or LH responses to iv LRH at the pre-treatment test. Before she developed amenorrhoea she had tried to become pregnant during 3 years. She had previously been given two treatment courses with human gonadotrophins without conceiving. One of the other infertile women (patient 5) had previously had a child after 8 treatment courses with human gonadotrophins. During the two menstrual cycles induced by the LRH therapy she had inadequate luteal phase, perhaps because
Serum levels of FSH, LH, oestradiol and progesterone during 2 consecutive menstrual cycle induced by sc pulsatile low-dose LRH treatment of a 24-year-old woman with secondary amenorrhoea (patient 2). Ovarian hyperstimulation occurred during the first induced cycle. The infusion pump did not function during 3 days of the follicular phase and stopped again after another 10 days of treatment during the second menstrual cycle. This resulted in disturbed follicular maturation and insufficient corpus luteum function. FSH and LH response to iv LRH before and after treatment are also shown.

Discussion

This study shows that chronic intermittent administration of low doses of LRH can induce ovulatory menstrual cycles in weight loss-related amenorrhoeic women with low endogenous oestrogen production. A portable computerized infusion pump proved to be practical and useful for the long-term pulsatile treatment. The same infusion pump and LRH dose was successfully used by Leyendecker et al. 1980a,b) to induce ovulation in 7 women with 'hypothalamic amenorrhoea'. They gave LRH iv and interrupted the pulsatile treatment soon after ovulation. Corpus luteum function was then supported by repeated injections of human chorionic gonadotrophin (hCG). Intermittent sc administration of a lower dose (25 ng/kg) was used by Crowley & McArthur (1980) to induce a normal menstrual cycle in a patient with hypogonadotrophic hypogonadism. A sc pulsatile treatment regimen was also used by Keogh et al. (1981) to induce ovulation followed by pregnancy in one patient with 'hypothalamic anovulatory infertility'.

Women with weight loss-related amenorrhoea have previously proved to be ideal candidates for chronic intermittent LRH therapy (Nillius et al. 1975; Nillius & Wide 1979). They have a prepubertal gonadotrophin response pattern to LRH with relatively greater FSH than LH increases. The
Fig. 5.
Serum levels of FSH, LH, hCG, oestradiol and progesterone during a normal conceptual cycle induced by the second sc pulsatile low-dose LRH treatment course of the 24-year-old woman with secondary amenorrhoea from Fig. 4. FSH and LH response to iv LRH before and after treatment are also shown.

Fig. 6.
Serum levels of FSH, LH, oestradiol and progesterone during 2 consecutive normal ovulatory cycles induced by uninterrupted pulsatile low-dose LRH treatment of a 25-year-old woman with secondary amenorrhoea (patient 3). FSH and LH response to iv LRH before and after treatment are also shown.
great initial FSH release is favourable for optimal induction of follicular maturation and thus also for adequate corpus luteum function. However, absent pre-treatment gonadotrophin responses to LRH do not exclude successful induction of follicular maturation and ovulation by pulsatile high and low dose LRH therapy (Nillius & Wide 1979 and Fig. 6).

During follicular maturation the FSH and LH response pattern change, presumably due to steroid modulation of the pituitary responsiveness to LRH. The LH release increases and the FSH release decreases when the maturing follicle(s) secrete increasing amounts of oestrogen (e.g. Figs. 1 and 2). This change of the FSH/LH ratio during prolonged LRH treatment should theoretically prevent hyperstimulation of the ovaries. However, overstimulation with ovarian cyst formation and excessively high oestrogen secretion did occur during the pulsatile low dose LRH treatment in one of our patients (Fig. 4).

Inadequate luteal phases often occurred during ovulation induction by pulsatile high dose LRH (Nillius & Wide 1975). In the present study, insufficient corpus luteum function was observed during 5 of the 12 induced cycles. In one of these treatment courses the infusion pump stopped to function for 3 days during the late follicular phase (Fig. 4). It is interesting to note that the follicle could be reactivated and made to ovulate after 3 days without pulsatile gonadotrophin stimulation. However, the interrupted gonadotrophin stimulation probably led to some damage of the follicle as ovulation was followed by an inadequate luteal phase.

Repeated ovulatory menstrual cycles can be induced by pulsatile low dose LRH therapy (Fig. 6). This has also been shown by Schoemaker et al. (1981), who successfully induced 3 consecutive ovulatory cycles in one patients with 'postpill amenorrhoea'. It is interesting to note the return of FSH responsiveness to LRH when the progesterone and oestrogen levels decreased during the late luteal phase (Figs. 4 and 6). During the prolonged treatment the same sc catheter was used without problems for periods of up to 69 days. However, in one patient bleeding at the catheter site necessitated reinsertion of a new sc catheter. Heparin in the LRH solution may have caused the bleedings and is therefore no longer used. The pulsatile low dose LRH therapy was well accepted by all the women.

Acknowledgments

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References


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