Danazol administration after gonadotrophin-releasing hormone analogue reduces rebound of uterine myomas

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We report the results of administration of danazol after suspension of gonadotrophin-releasing hormone analogue (GnRHa) therapy for uterine myomas. A total of 21 women with uterine myomas was treated with 100 mg danazol for 6 months after GnRHa therapy. Uterine volume and endocrine status were monitored monthly by ultrasound and assay of plasma gonadotrophins, oestradiol and progesterone. The results show a rebound of uterine volume about 30% less than in controls at the end of danazol therapy. Menstrual cyclicity returned after 65 ± 3 days in 16 subjects and five patients remained amenorrhoeic. Hormone assays confirmed renewed ovarian function in the women whose menstrual periods returned. Bone mineral content was substantially reduced during GnRHa treatment but improved significantly during danazol therapy even in the women who remained amenorrhoeic. These results show the utility of danazol in prolonging the therapeutic effects of GnRHa. The mechanism by which danazol inhibits rebound of uterine volume may be due to its antiprogesterone effects on uterine myomas.

Key words: danazol/GnRHa/myomas/oestradiol/progesterone

Introduction

Uterine fibroids are the most common benign tumours in women during their reproductive years and one of the most frequent causes of major surgery in women of reproductive age (Buttram and Reiter, 1981; Berkeley et al., 1983). Most symptoms experienced by women with uterine myomas are due either to large pelvic masses or uterine bleeding. Until now treatment of symptomatic patients has been primarily surgical, although there have been attempts at medical treatment with progesterone and gestrinone (Mixon and Hammond, 1961; Sadan et al., 1987; Countinho and Goncalves, 1989). Clinicians have long recognized that fibroids may depend on oestrogen for their growth. These tumours are rarely found before puberty, they increase in size during pregnancy and after oestrogen administration and regress after menopause and ovariectomy (Buttram and Reiter, 1981). Histological evidence of endometrial hyperplasia near submucous fibroids suggests that local oestrogen levels may be high. Further experimental evidence of oestrogen dependence is provided by studies showing a greater cytoplasmic oestrogen receptor frequency in leiomyomatous tissue than in adjacent normal myometrium (Sadan et al., 1987). A state of reduced oestrogen secretion led to a reduction in growth of fibroids and even their regression. The finding that menopause medically induced by gonadotrophin releasing hormone analogues (GnRHa) led to a reduction in fibroid volume opened the way for research into non-surgical means of treating fibroids (Filion et al., 1983; Friedman et al., 1987; Andreyko et al., 1988). Unfortunately, myomas tend to return to their original size when therapy is suspended (Letterie et al., 1989). Other problems with long term treatment with GnRHa have been menopausal symptoms and well documented bone loss (Comite and Jensen, 1988).

In order to overcome this problem, several groups have added progestins during and after GnRHa therapy. The fibroids began to grow again under these circumstances, despite the fact that progesterone was thought to inhibit its own receptor and thus reduce fibroid size (Maheux et al., 1986; Friedman et al., 1988). It has been demonstrated that uterine myomas contain a higher density of oestradiol and progesterone receptors than myometrium (Buchi and Keller, 1983).

Wilson et al. (1980) found that concentrations of cytoplasmic oestradiol receptors in fibroids were significantly greater than in normal myometrium and endometrium, and that the same was true for progesterone receptors but the difference was not statistically significant. Other authors confirmed this pattern showing a relation between the oestrogen receptor content in fibroid, myometrium and endometrium and the phase of the cycle (Soules and McCarty, 1982).

In this study we report the results of the use of danazol after suspension of analogue therapy. Danazol is a synthetic steroid used for many years in hormonal treatment of endometriosis (Dmowski and Cohen, 1975). The original clinical application of this steroid was based on its antigonadotrophic properties, demonstrated in animals and patients in whom the induction of amenorrhoea, inhibition of gonadal steroids and block of pituitary gonadotrophins was expected to be beneficial (Fanchimont and Cramilion, 1977; Dmowski et al., 1983). Danazol is in fact an isoxazole derivative of 17 α-ethinyl testosterone (ethisterone), another synthetic steroid. It exerts multiple effects at various sites of the female reproductive system, as direct and indirect consequences of its binding affinity for intracellular steroid receptors, its interaction with two circulating steroid binding
proteins, and its capacity to inhibit certain enzymes involved in gonadal and adrenal steroidogenesis (Barbieri et al., 1979).

Material and methods

Patients
Twenty-one women, aged 31–45 years, were enrolled in the study. Of these, 17 had symptomatic uterine leiomyomata found to be increasing in size. Four women were asymptomatic and were found to have leiomyomata on routine pelvic examination. The main symptoms included pelvic pressure and/or pain, menorrhagia, urinary frequency and dysmenorrhea. None of the women had used hormonal medication in the 3 months prior to enrolment or had even taken GnRHAs. None had any serious concomitant medical condition such as hormonal dysfunction. Before beginning therapy all women underwent gynaecological examination and cervical smear. Blood pressure was measured and routine blood tests, including lipid profile and clotting factors, were performed and found to be within normal limits; only red cell concentration and haemoglobin were below normal in some cases.

Treatment protocol
Informed consent was obtained from each patient before starting therapy. At the initial visit and every 3 months, patients underwent pelvic ultrasound (Siemens ultrasound with 5 MHz transabdominal transducer and 7.5 MHz trans-vaginal transducer; Sonoline SL2, Milan, Italy) to determine uterine myoma volume. These instrumental evaluations were performed in three dimensions either vaginally and/or abdominally. Blood samples were taken to assay oestradiol, progesterone, luteinizing hormone (LH), follicle stimulating hormone (FSH) and bone mineral content (BMC) was also determined every 2 months. The patients received an injection of long-acting GnRHAs, either goserelin (Zoladex, Zeneca, Milan, Italy) (12 patients) or triptorelin (Decapetyl 3.75, Ipsen, Milan, Italy) (nine patients), every 28 days for 6 months according to a randomized protocol. The difference between the two GnRHAs was insignificant for our study. Some years ago we showed a similar ovarian suppression after 12–15 days from injection in patients treated with goserelin or triptorelin (De Leo et al., 1992). GnRH therapy was initiated in mid to late luteal phase. Patients were seen at 2 month intervals during the treatment period for 1 year. After GnRH therapy all patients were treated with 100 mg danazol for a further 6 months (group B).

The control group was a previously conducted series consisting of 12 age-matched (34–41 years) women with uterine leiomyomata treated for 6 months with GnRH (triptorelin) and examined 6 months after the end of therapy (group A). This control group was composed of women treated previously, between November 1993 and June 1994 before the beginning the actual protocol. The women had menorrhagia, metrorrhagia, pelvic pain and anaemia. Age distribution in the two groups was comparable. In group A, mean age was 38.26 years and in group B 37.64 years.

Hormone assays
Venous blood samples were taken for assay of LH, FSH, oestradiol, progesterone and osteocalcin (OC) before the start of therapy and at 8-weekly intervals during the 1 year of treatment.

The blood was centrifuged and the serum stored at –20°C until analysis. Plasma FSH, LH, oestradiol, progesterone and OC levels were measured by double antibody radioimmunoassay with commercial kits. Samples were assayed in duplicate, at two different dilutions. All samples from a given subject were assayed together. Quality control pools at low, normal and high LH, FSH, oestradiol, progester-

Bone mineral density
Bone mineral density (BMD) was measured by dual X-ray absorptiometry (Ostescan Nim, Verona, Italy) in the distal radius of the non-dominant forearm before therapy and every 3 months.

Statistical analysis
Plasma hormone levels (as mean ± SD) were expressed in mIU/ml (LH; FSH), pg/ml (oestradiol; progesterone) and ng/ml (OC) (Table I). Comparisons between treatments were performed at each time point (weeks 12 and 24 after start of GnRH treatment and weeks 12 and 24 of follow-up) by analysis of variance (ANOVA). The comparison within treatment groups was made by Student’s t-test for paired and unpaired data.

Results
Oestradiol plasma levels in both treatment groups showed suppression throughout the 6 months of GnRH therapy (goserelin and triptorelin). Mean oestradiol concentrations showed a significant reduction (P < 0.01) during treatment with oestradiol plasma levels <20 pg/ml in both groups in weeks 12 and 24 of treatment (Table I). The patients were amenorrhoeic after the second injection. A significant reduction (P < 0.001) in uterine myoma volume was observed after 12 weeks of GnRH therapy. Maximum reduction was achieved after 6 months of treatment (Figure 1). Uterine dimensions throughout the study were determined by serial pelvic ultrasound examination. We did not observe significant differences in uterine volume reduction between groups treated with the two GnRHAs. All patients experienced partial or complete relief of symptoms while using GnRHAs. At the end of GnRH therapy we observed a gradual increase in uterine volume over a 6-month period in groups A and B. This increase was much greater in the group without danazol treatment (group A). At 6 months follow-up the rebound in uterine volume in group B was 31.2% less than in group A (P < 0.001).

Menstrual cyclicity returned after 65 ± 3 days from last GnRH injection in all women, except for five patients in group B who remained amenorrhoeic. Hormone assays confirmed that ovarian function was restored in the women whose menstrual periods returned. Basal oestradiol concentrations observed in follicular phase in the group treated with danazol (group B) were lower than in controls (group A) (Table I). The other hormones reported in Table I were similar in the two groups.

BMD declined during GnRHa treatment and was significantly higher 6 months after the end of therapy in all patients, including the five women remaining amenorrhoeic (Figure 2). In the patients treated with danazol, the increment in BMD was more rapid than in the group without danazol therapy. Osteocalcin levels (Figure 3) showed a significant increase in both groups during the 6 months of GnRHa therapy. After the end of therapy, osteocalcin levels returned to the normal range.
Table I. Hormone concentrations observed before and every 3 months during gonadotrophin-releasing hormone analogue (GnRHa) therapy in group A and B (follicular phase) and after discontinuing GnRHa in group A (control) and group B (treated with 100 mg/day of danazol). The blood samples were taken in follicular phase.

<table>
<thead>
<tr>
<th>Time (months)</th>
<th>Basal</th>
<th>GnRHa therapy</th>
<th>After GnRHa therapy</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>A</td>
<td>B</td>
<td>A</td>
</tr>
<tr>
<td>LH (mIU/ml)</td>
<td>8.4 ± 2</td>
<td>9.3 ± 2.5</td>
<td>2.1 ± 0.3</td>
</tr>
<tr>
<td>FSH (mIU/ml)</td>
<td>7.7 ± 1.1</td>
<td>6.5 ± 1.6</td>
<td>2.5 ± 0.3</td>
</tr>
<tr>
<td>Oestradiol (pg/ml)</td>
<td>71 ± 24</td>
<td>60 ± 12</td>
<td>10 ± 6</td>
</tr>
<tr>
<td>Progesterone (pg/ml)</td>
<td>320 ± 88</td>
<td>360 ± 150</td>
<td>120 ± 25</td>
</tr>
<tr>
<td>Prolactin (ng/ml)</td>
<td>9.5 ± 3.1</td>
<td>11.2 ± 2</td>
<td>7.0 ± 1.5</td>
</tr>
</tbody>
</table>

LH = luteinizing hormone.
FSH = follicle stimulating hormone.
A = analogue.
B = analogue + danazol.

Figure 1. Variation of uterine myoma volume (ml) during therapy with gonadotrophin-releasing hormone analogue (GnRHa).

Haemoglobin concentrations and haematocrit increased significantly during treatment with the two analogues. Haemoglobin increased from 11.1 ± 0.6 mg/ml before therapy to 12.7 ± 0.4 mg/ml after 6 months. Haematocrit (%) increased from 35.3 ± 1.4 to 39.6 ± 0.5 after 6 months. Serum cholesterol increased slightly from 185 ± 6 to 197 ± 18 mg/dl in the danazol group but there was no change in serum triglycerides.

During GnRHa therapy, vasomotor symptoms were reported by 75% of patients, vaginal dryness by 30% and transient headaches by 48%. These symptoms disappeared with the return of menstrual cyclicity when oestriadiol levels returned to the normal range for fertile women. The five amenorrhoeic women on danazol also reported the disappearance of menopausal symptoms.

Discussion

The present study shows the efficacy of GnRHa in the treatment of symptomatic and asymptomatic myomas and demonstrates that rebound of myoma volume after discontinuing therapy can be reduced by administering low doses of danazol.

The prevalence of myomas among patients of reproductive age and their tendency to regress after menopause suggest that this disorder is hormone dependent (Vollenhoven et al., 1990). Clinical and laboratory data demonstrate that an oestrogenic milieu is required for leiomyoma growth and suggest that inducing a state of reduced oestrogen secretion may be useful in the medical management of uterine myomas. The finding that induction of menopause by GnRHa led to a reduction in fibroid size opened the way to the use of these compounds in the treatment of myomas. The basic understanding was that oestrogen induced fibroid proliferation and, if suppressed, this would stop. The difficulty is that when GnRHa therapy is stopped, an increase in fibroid volume is observed (Letterie et al., 1989). In the present study, an increase in uterine volume was observed in association with increases in serum concentrations of oestradiol and progesterone as menstrual cyclicity resumed.

Recent data suggest that progestins, as well as oestrogens, stimulate fibroid growth (Tamaya et al., 1985; Rein et al., 1995). Both oestrogen and progesterone receptors have been detected in myometrium and myomas (Buchi and Keller, 1980). Myometrial and myoma tissue cultures supplemented with progesterone grew significantly. The observation that oestradiol plus medroxyprogesterone acetate shows that progesterone inhibits the ability of GnRHa to shrink uterine myomas (Carr et al., 1993). Myoma mitotic activity is higher in early luteal phase than in follicular phase (Buchi and Keller, 1983). Murphy et al. (1993) showed that an antiprogesterone, RU486, induced a reduction in myoma size and suggested the use of antiprogesterone in the treatment of myomas. These data support an important role for progesterone in the development and growth of uterine myomas.

The major problems with long term GnRHa treatment are menopausal symptoms and bone loss. Another difficulty is the fact that uterine volume usually returns to pretreatment values within 3–6 months of discontinuing GnRHa therapy.

Danazol binds to the progesterone receptor with an affinity that is 3% that of progesterone (Franchimont and Cramillon, 1977). Moreover, danazol is able to displace progesterone from human and myometrial cytosol suggesting a direct antiprogesterational activity in the human (Chamnes et al., 1980). For these reasons, we decided to use danazol after discontinuing GnRHa therapy in the treatment of uterine myomas.

The danazol dose given to the patients was well tolerated.
No patients reported side effects and all completed the protocol. We observed a slight but not significant increase in serum cholesterol while other haematological parameters were not modified.

The results of the present study suggest that it is possible to reduce the rebound of uterine volume by taking danazol at low doses after discontinuing GnRH therapy. The smaller rebound than controls allow a better evaluation of these patients. At the same time we observed a more rapid return of BMD to normal values.

These results show the use of danazol at low doses in prolonging the therapeutic effects of GnRHs. The mechanism of danazol in inhibiting rebound of myoma volume probably works through an antiprogestosterone effect on uterine myomas, even if a weak anti-oestrogenic effect cannot be excluded.

References


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