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AROMATIZATION INHIBITION ALONE OR IN COMBINATION WITH GnRH AGONISTS FOR THE TREATMENT OF PREMENOPAUSAL BREAST CANCER PATIENTS

M. DOWSETT,[1]* R. C. STEIN[2] and R. C. COOMBES[3]

[1]Department of Biochemistry, Royal Marsden Hospital, Fulham Road, London SW3 6JJ, [2]Department of Oncology, Middlesex Hospital, London W1P 8BT and [3]Department of Medical Oncology, Charing Cross Hospital, London W6 8RF, England

Summary—Aromatase inhibition in postmenopausal women causes a marked fall in the plasma levels of oestrogens and is an effective treatment for breast cancer, however, trials with aminoglutethimide found that this aromatase inhibitor was ineffective in suppressing plasma oestrogen levels in premenopausal breast cancer patients. We found that the more potent inhibitor, 4-hydroxyandrostenedione (4-OHA), which can suppress oestrogen synthesis in rodents and non-human primates with intact ovarian function, was also unsuccessful as an oestrogen suppressant in premenopausal women at its maximum tolerated dose (500 mg/week i.m.). GnRH agonists are effective suppressants of ovarian oestrogen synthesis but oestrogen production from peripheral sites is unaffected. Our studies of a combination of the GnRH agonist goserelin and 4-OHA demonstrated that the combination caused greater oestrogen suppression than goserelin alone and led to objective clinical response in 4/6 breast cancer patients after their relapse from treatment with goserelin as a single agent. The combination of a GnRH agonist and an aromatase inhibitor should be subjected to clinical trials.

INTRODUCTION

Aromatase inhibitors are recognized as effective therapeutic agents in postmenopausal patients with oestrogen-dependent breast cancer. However, in premenopausal patients consistent suppression of plasma oestrogen levels was not achieved with the prototype aromatase inhibitor, aminoglutethimide (AG) [1–3]. Intermittent increases in plasma gonadotrophin levels were observed [1], and the resultant increase in the stimulation of ovarian steroidogenesis appears to have adequately compensated for any AG-induced blockade of aromatase. All the same it is possible that with a more potent inhibitor of aromatase, suppression of ovarian oestrogen synthesis may be achievable.

4-Hydroxyandrostenedione (4-OHA; CGP 32349) is a more potent inhibitor than AG and has the additional advantage of being a suicide inhibitor i.e. it permanently inactivates the enzyme by irreversibly binding to the active site [4]. In contrast to AG, 4-OHA was able to suppress oestradiol levels in mature rats [5]. LH levels rose in AG-treated rats in a similar way to that in premenopausal women, but 4-OHA-treated rats showed a decrease in LH levels which appears to be due to the minor androgenic nature of 4-OHA. Further studies have demonstrated that 4-OHA can suppress ovarian oestrogen formation in non-human primates [6]. We have therefore examined the possibility that 4-OHA could effectively suppress ovarian oestrogen synthesis in a small group of premenopausal patients with advanced breast cancer.

4-OHA IN PREMENOPAUSAL PATIENTS

Five premenopausal patients (median age 46 years, range 42–52) with progresssive locally advanced or metastatic breast cancer were treated with 4-OHA 500 mg/week i.m. [7]. Two patients had received previous endocrine treatment but this had ceased at least 3 weeks before starting 4-OHA. Serum oestradiol and gonadotrophin levels were measured by radioimmunoassay at weekly intervals just prior to each injection. Treatment was continued for between 4 and 19 weeks (median 8 weeks).

No consistent fall in serum levels of oestradiol or obvious compensatory rise in

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*To whom correspondence should be addressed.

gonadotrophins occurred in any of the five patients. The results are plotted separately for three patients who were unambiguously premenopausal (Fig. 1) and for two patients who showed an apparently postmenopausal endocrine profile (low oestradiol and raised gonadotrophin level) at the start of 4-OHA treatment (Fig. 2) despite documentation of their menstrual activity shortly prior to treatment.

In the former group of patients there was one instance in each of two of the patients of serum oestradiol levels which were <100 pmol/l (the laboratory upper 95% confidence limit for postmenopausal women) (Fig. 1). These occurred towards the end of each patient's treatment and could conceivably be due to a cumulative effect of 4-OHA. Gonadotrophin levels were not postmenopausal in these same two samples. In the other 20 samples available from these 3 patients there was no indication of suppressed ovarian oestrogen synthesis.

In the other two patients, despite 4-OHA treatment there were several points at which the originally low oestradiol values rose above 100 pmol/l (Fig. 2). In one of the two patients after 6 weeks treatment there was a steady rise

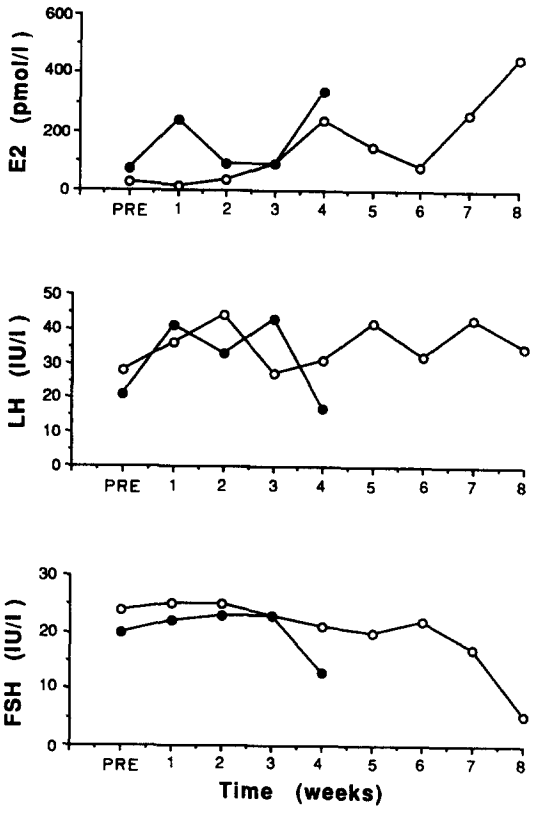

Fig. 2. Endocrine changes in 2 perimenopausal breast cancer patients treated with 500 mg 4-OHA i.m. weekly. The same symbol represents the same patient in each panel.

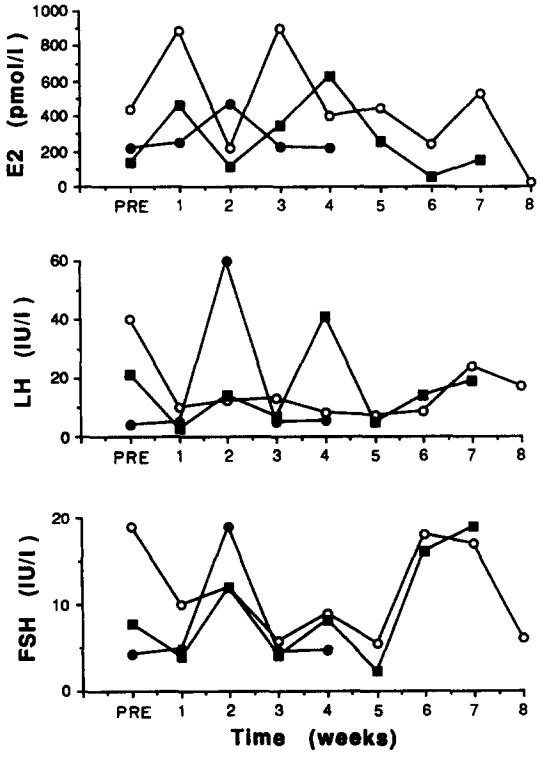

Fig. 1. Endocrine changes in 3 premenopausal breast cancer patients treated with 500 mg 4-OHA i.m. weekly. The same symbol represents the same patient in each panel.

in oestradiol levels accompanied by a reduction in gonadotrophin levels. Thus in these two patients who, could be categorized as being perimenopausal with reduced ovarian function initially, 4-OHA did not inhibit the oestrogen synthesis which accompanies intermittent follicular activity and is characteristic of perimenopausal women.

All five patients continued to menstruate during treatment. None of the five patients showed an objective response to treatment although one remained stable for 8 weeks of treatment. Four of the patients received the gonadotrophin releasing hormone (GnRH) agonist, goserelin (ICI: Zoladex), on discontinuation of 4-OHA and three of these had an objective response lasting 9, 12 and 25 months, respectively.

It seems fair to conclude that, at what we have previously found to be the maximum tolerated dose of 4-OHA, premenopausal aromatase activity is not inhibited sufficiently to suppress plasma oestradiol levels. Since no marked increase in gonadotrophin secretion occurred it appears that the unaffected levels of oestrogen were not due to an overt increase

in gonadal drive but probably to ineffective inhibition. It is probably significant that the suppression observed previously in both rats and non-human primates was after giving much more 4-OHA (50 mg/kg/day and *ca.* 30 mg/kg/day, respectively) [5, 6] than the tolerable dose in patients (*ca.* 10 mg/kg/week). The very high intra-ovarian concentration of androgen substrate for aromatization may also be an important factor in reducing the effectiveness of 4-OHA since substrate can protect aromatase from inactivation by 4-OHA.

A lack of objective response in only 5 patients does not exclude that 4-OHA could be clinically effective in a substantial proportion of patients. The 95% confidence intervals for response are 0–52% after making such an observation. It is notable that despite our own experience of the non-efficacy of the AG in premenopausal women [2] Wander *et al.* [3] reported a 28% response rate in the absence of significantly suppressed plasma oestradiol levels. However, if responses to 4-OHA were to occur it seems that they would be through a mechanism that is independent of circulating oestrogens. It has been suggested that inhibition of intra-tumoural aromatase activity is a possible explanation for the AG-induced responses noted above [8].

GnRH AGONISTS IN PREMENOPAUSAL PATIENTS

The more conventional medical treatment for premenopausal breast cancer is with a GnRH agonist which has been said to achieve a steroidal profile in plasma equivalent to ovariectomy as a result of the profound fall in gonadotrophin secretion achieved. To reflect this the treatment is sometimes referred to as a "medical ovariectomy" or "medical castration". A number of analogues have been shown to be clinically effective in premenopausal breast cancer patients.

Peripheral aromatization seems to be largely unaffected by GnRH agonist treatment although the suppression of ovarian androgen synthesis reduces the plasma levels of substrate for aromatase. Thus plasma oestrogens within the postmenopausal range are generally observed in patients during agonist therapy. Results from our recent study in patients with endometriosis who were treated with goserelin for 6 months indicate that in some patients a low but progressively increasing level of follicular activity may also occur which leads to a partial recovery of oestradiol synthesis [8]. It has been noted with a number of GnRH agonists that the initially suppressed FSH levels increase with time back to approximately pretreatment levels in the absence of a recovery of LH levels [8–12]. In our recent studies a concurrent but more variable increase in serum oestradiol levels was observed such that at the end of treatment (weeks 20 and 24) oestradiol levels were greater in 15 patients than at week 4 lower in 2, and the same in 1 (Fig. 3). It is clear from the use of aromatase inhibitors in postmenopausal women that these very low oestradiol levels in such patients can still be mitogenic to breast carcinomas. It is therefore possible that by the addition of an aromatase inhibitor to patients under treatment with a GnRH agonist, a more complete and potentially more effective treatment may be achieved. We have therefore conducted 2 studies in which 4-OHA has been administered to premenopausal patients with advanced breast cancer already on treatment with the GnRH agonist goserelin.

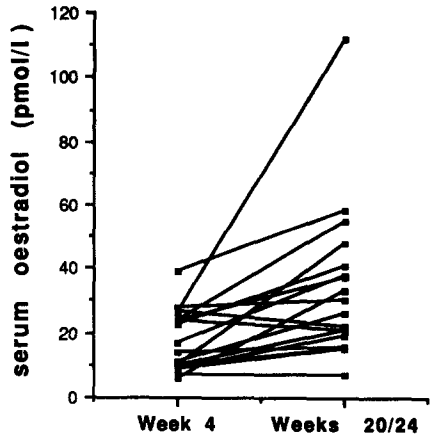

Fig. 3. Serum levels in 18 premenopausal patients with endometriosis treated with goserelin (3.6 mg s.c. monthly) after 4 weeks and after 20/24 weeks (mean).

GOSERELIN + 4-OHA IN PREMENOPAUSAL PATIENTS

Endocrine study

The aim of this study was to determine whether 4-OHA led to additional oestrogen suppression in goserelin-treated patients. Five patients who were already on treatment with once monthly s.c. injections of 3.6 mg goserelin and whose disease was either stable or in remission were given three injections of 500 mg 4-OHA i.m. at weekly intervals. Blood samples were taken on five occasions, the first two being during the week prior to starting 4-OHA and

the others at weekly intervals after starting 4-OHA. Serum levels of oestradiol are shown for each of these samples in Fig. 4. The mean ($\pm$SEM) serum oestradiol level prior to 4-OHA treatment was 23.6 ± 4.1 pmol/l. Seven days after the first injection of 4-OHA the serum levels of oestradiol had fallen in all 5 patients to a mean of 6.1 ± 1.9 pmol/l ($P < 0.001$, t-test), and continued suppression below the levels on Zoladex alone was found during this 3 week study.

LH and FSH in this combination study are shown for each patient in Fig. 5. Addition of 4-OHA had no effect on LH levels in any of the patients. In 3 patients there was a small increase in FSH levels but overall this was not statistically significant.

Clinical study

The encouraging endocrine data above provided support for a clinical study of this combination treatment. Six premenopausal patients who had experienced a partial response to goserelin but who had subsequently relapsed were treated with 250 mg 4-OHA i.m./2 weeks in addition to continued treatment with goserelin. The dose of 250 mg 4-OHA/2 weeks is known to produce oestrogen suppression which is closely similar to that with 500 mg weekly [13]. Four patients (67%) experienced objective responses which lasted for between 8 and 24 months. One patient had disease stabilization for a period of 5 months whilst the sixth patient continued to progress.

A further patient who had stable disease over a 2 month period of treatment with goserelin was similarly treated with additional 4-OHA. Her disease remained stable for a further 10 months.

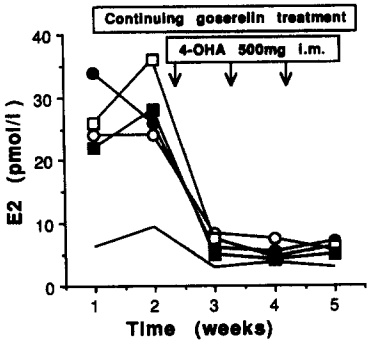

Fig. 4. Serum oestradiol levels in 5 premenopausal breast cancer patients during the addition of 500 mg 4-OHA i.m. weekly to ongoing treatment with goserelin (3.6 mg s.c. monthly).

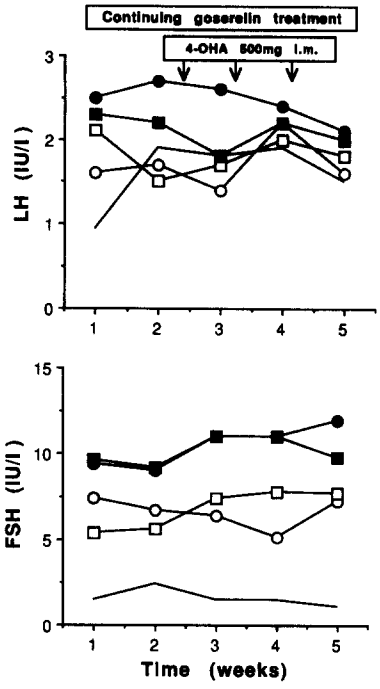

Fig. 5. Serum LH and FSH in 5 premenopausal breast cancer patients during the addition of 500 mg 4-OHA i.m. weekly to ongoing treatment with goserelin (3.6 mg s.c. monthly).

The results indicate that in breast cancer which has responded to treatment with a GnRH agonist the tumour is still sensitive to further oestrogen suppression at relapse. This suggests that the tumour has become adapted to grow in the presence of low, postmenopausal levels of oestrogen but retains a dependence on these low levels for progression. Addition of an aromatase inhibitor to the treatment of GnRH agonist-relapsed patients is clearly a worthwhile therapeutic stop. However, the important question to ask is whether first-line treatment with the combination of a GnRH agonist and an aromatase inhibitor may be more effective than sequential treatment as described above. This possibility should be evaluated in a clinical trial. Experience with experimental models indicates that the alternative combination of a GnRH agonist with tamoxifen is less attractive: in rodents carrying carcinogen-induced mammary tumours there is a better tumour regression to GnRH agonist or castration alone than to either of these plus tamoxifen.

CONCLUSION

The treatment of premenopausal breast cancer with 4-OHA alone cannot be justified by its

effects on circulating hormone levels. With the availability of effective well-tolerated and conveniently administered GnRH agonists there seems little reason to pursue aromatase inhibitors as suppressants of ovarian oestrogenesis for breast cancer treatment. It is possible, however, that potent, short-acting inhibitors could find a role in managing fertility (e.g. in ovulation induction schedules). The endocrine and clinical results of the combination of a GnRH agonist with an aromatase inhibitor are impressive and make it important to test this combination in a large comparative clinical trial.

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