

Endocrine Effects of Tamoxifen Plus Exemestane in Postmenopausal Women with Breast Cancer

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ABSTRACT

Purpose: In some specific circumstances, combined hormonal therapies for breast cancer seem to be more effective than single maneuvers. In two laboratory mammary cancer models, the combination of the aromatase inactivator exemestane plus tamoxifen gives a higher response rate than is found with either agent alone.

To evaluate the endocrine effects of the combination of exemestane and tamoxifen, we studied 33 postmenopausal women disease-free following primary treatments for breast cancer who were taking tamoxifen for at least 3 months.

Design: After observation for symptoms on tamoxifen for 4 weeks, blood samples were taken and patients were begun additionally on exemestane 25 mg p.o. qd. Eight weeks later, blood samples were again taken, and exemestane was discontinued.

Results: A decrease in alkaline phosphatase was found with exemestane treatment ($P = 0.06$), whereas no change in osteocalcin level was observed. A decrease in high-density lipoprotein cholesterol level was found ($P = 0.0025$), whereas total cholesterol, low-density lipoprotein cholesterol and triglyceride levels showed no changes with exemestane treatment. Estradiol, estrone, and estrone sulfate levels decreased to immeasurable or very low levels with exemestane treatment (all $P < 0.001$). No significant changes in frequencies of common drug-associated side effects, such as vasomotor symptoms or weight change, were found.

Conclusions: Based on the absence of adverse endocrine effects with the addition of exemestane to tamoxifen therapy observed in this study, further clinical evaluation of the efficacy of this combination is warranted.

INTRODUCTION

Whereas hormonal therapies are useful for some patients with metastatic breast cancer and as adjuvant treatment in a majority of patients, these often well-tolerated approaches are

limited in their benefits. Patients with metastatic breast cancers progress eventually on hormonal treatment, despite presence of hormonal receptors in primary breast cancers indicating greater likelihood of response to hormonal treatments. Fifteen to 30% of patients given adjuvant hormonal therapies develop recurrent breast cancer from which they die (1). Despite the development of effective new hormonal agents such as aromatase inhibitors or inactivators, there is need for hormonal therapies which give greater response rates and longer times to treatment failure in metastatic disease and greater disease-free and overall survival in adjuvant settings.

Whereas single sequential hormonal maneuvers (oophorectomy, antiestrogen therapies, aromatase inhibitor, or inactivator treatment) have been the hormonal approaches found most effective, recently there are increasing data suggesting that some specific combined hormonal therapies are more effective, specifically oophorectomy (surgical, radiation, or chemotherapeutic with a luteinizing hormone–releasing hormone agonist) plus tamoxifen as adjuvant therapy in premenopausal women with stage I/II hormone receptor–positive disease (2–4). For combinations involving aromatase inhibitors however, to date, data regarding combination therapies have indicated no benefits from this approach.

Laboratory data have provided limited support for combination aromatase inhibitor–antiestrogen therapy. In a MCF-7 nude mouse model, Lu et al. found the combinations of anastrozole and tamoxifen and letrozole and tamoxifen essentially no better than anastrozole or letrozole alone (5). The estrogen agonist effect of tamoxifen was suggested to be responsible for these findings. The translation of findings from this work to the human situation is uncertain, because the doses used in women are greater; and whereas a “tickler” agonist effect of tamoxifen is observed clinically in some women with metastatic breast cancer (tumor flare), such patients often go on to have durable favorable responses. These findings have led to a sequential hormonal treatment adjuvant study of letrozole in women who have completed 5 years of tamoxifen therapy, which showed significant benefit from the follow-up letrozole treatment (6), and more recently, shorter-term tamoxifen therapy of 2 to 3 years followed by exemestane or continuing tamoxifen showed superiority for the sequential tamoxifen/exemestane approach (7). A large adjuvant study of anastrozole, tamoxifen, or the two agents combined has shown superiority of the anastrozole-alone treatment over both tamoxifen and the combination (8).

Exemestane is a steroidal aromatase inactivator (AI) for which the laboratory and developing clinical treatment results data seem different in potentially significant ways from data for anastrozole and letrozole. Exemestane does not bind to steroidal receptors. In a DMBA carcinogen rat mammary cancer model, exemestane and tamoxifen combined were found to give a significantly higher response rate in established tumors than the rates for tamoxifen or exemestane alone, and the combination was more effective as a preventive intervention than either agent alone (9). In a recent limited report using an MCF-7 nude mouse

Received 8/10/04; revised 10/29/04; accepted 11/19/04.

Grant support: Pharmacia, Inc.

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model, the same group which reported absence of additive effect or synergism with anastrozole or letrozole and tamoxifen, found that exemestane and tamoxifen were additive in their tumor growth suppressant effects (10).

In women with metastatic breast cancer, exemestane is associated with higher response rates than tamoxifen (11); gives responses in cases where aminoglutethimide, anastrozole, or letrozole have failed (12); and is superior to megestrol in median survival (13). Such a benefit was not found in a letrozole/megesterol study (14), whereas a similar benefit was suggested in a pooled analysis of two phase III studies of anastrozole (15). Exemestane gives responses in patients with visceral disease who have failed antiestrogens (16). The intratumoral aromatase inactivator effects of exemestane are suggested to be potentially more significant than the estrogen lowering effects (17, 18).

In a recently reported pilot study, Rivera et al. found that the addition of tamoxifen treatment to exemestane in women with metastatic breast cancer was associated with no significant change in the pharmacokinetics of exemestane and nonsignificant differences in three estrogen markers (19).

In summary, these data suggest that exemestane is a particularly active aromatase-targeting agent, and that the combination of exemestane and tamoxifen could be more active than exemestane or tamoxifen alone, and in contrast to the combination of anastrozole or letrozole and tamoxifen, a combination without pharmacokinetic or pharmacodynamic interactions. Evaluating this combination in the metastatic, but in particular in the adjuvant setting, requires data demonstrating an *absence* of significant toxicity and *absence* of interference with the bone-preserving and lipid estrogen agonist effects of tamoxifen (20, 21).

The current communication reports data on these issues; a companion report will present additional data on the pharmacokinetic interactions of these two agents.

MATERIALS AND METHODS

Under a protocol approved by the University of Wisconsin Committee for the Protection of Human Subjects (HSC 2000-549), we recruited 33 postmenopausal women diagnosed ≤ 4 years previously with stage I/II invasive breast cancer who were receiving tamoxifen 20 mg p.o. qd as single dose adjuvant therapy for a minimum of 3 months (and had been receiving tamoxifen up to 10 years). Postmenopausal status was defined as:

- 1 year since last menstrual period and no prior oophorectomy or hysterectomy, or
- prior bilateral oophorectomy, or
- previous hysterectomy, one or both ovaries intact, either ≥ 60 years of age or follicle-stimulating hormone level in the postmenopausal range.

Patients were considered premenopausal and ineligible if they did not meet these criteria for postmenopausal status. Participants had to have had a normal physical examination without evidence of signs or symptoms of metastatic cancer, a normal chest X-ray ≤ 14 days of entry on study, and an Eastern Cooperative Oncology Group performance status of 0, 1, or 2.

Patients using estrogenic or progesterone therapies other than tamoxifen were not eligible.

Patients had to have adequate organ and hematopoietic system function as indicated by the following laboratory values, obtained ≤ 14 days before entry on study: serum creatinine < 1.5 mg/dL; alkaline phosphatase $\leq 1.5 \times$ institutional upper limit of normal; absolute neutrophil count $\geq 1,500/\text{mL}^3$; total bilirubin $\leq 1.5 \times$ institutional upper limit of normal; and platelet count $\geq 100,000/\text{mL}^3$. All participants provided written informed consent.

Design. We conducted an open label study of sequential design. After entry on study, participants continued their tamoxifen treatment and provided symptoms data and answered the FACT B4 quality-of-life questionnaire (22). After 4 weeks, the symptoms checklist and FACT B4 instrument were again given, and blood samples were obtained before the daily tamoxifen dose was taken for evaluation of estradiol, estrone, and estrone sulfate, alkaline phosphatase, osteocalcin, total and high-density lipoprotein (HDL) cholesterol, and triglycerides. Exemestane 25 mg p.o. qd was begun, and over the next 8 weeks a symptoms diary was kept. After this time on one day, before the daily doses of exemestane and tamoxifen were taken, symptoms checklist and the FACT B4 instrument were again given, and blood samples for the same evaluations as previously were obtained. At baseline, 4 and 12 weeks, physical examinations including weight were done.

Laboratory Methods Assays for Estradiol (E2), Estrone (E1), and Estrone Sulfate (E1S). Samples were drawn into chilled blood collection tubes containing sodium heparin as anticoagulant and immediately placed in ice-water before centrifugation ($1,000\text{--}1,200 \times g$ for 10-15 minutes at 4°C) within 30 minutes of collection. The harvested plasma was transferred to screw-cap polypropylene storage tubes and stored frozen at approximately -20°C until analysis.

Samples were assayed for plasma concentrations of E1, E1S, and E2 using solid-phase extraction and high-performance liquid chromatography purification followed by highly sensitive RIA, according to a procedure described previously (23). The lower limits of quantitation of the assay were 1.8 pg/mL for E1, 6 pg/mL for E1S, and 0.7 pg/mL for E2. Estrogen concentrations below the lower limits of quantitation were recorded as the value of that limit.

Statistical Methods. Because they did not generally conform to Normal distributions, differences in laboratory values at weeks 4 and 12 were tested with the Wilcoxon signed-rank test (24). Means and medians are reported, where they could be calculated.

RESULTS

Thirty-three women were recruited to the study. Two women developed bothersome symptomatology after 4 weeks of combined therapy, increase in hot flashes (1) and rash (1), and elected to stop their exemestane. Participants had a mean age of 57.3 years (range, 74-43 years) and averaged 2.9 years from diagnosis (range, 0.9-8.8 years). Mean body mass index of participants was 27.2 (range, 19.6-37.2).

There were no significant weight changes in participants during the 8 weeks of added exemestane treatment. As shown in Table 1, participants had reductions in HDL cholesterol ($P = 0.0025$) and alkaline phosphatase ($P = 0.06$) but no

Table 1 Clinical chemistry results

Variable	Treatment	No. subjects assessed	Mean value	Sign rank for differences in values
Alkaline phosphatase (units/L)	Tamoxifen	30	73.4	0.08
	Tamoxifen + Exemestane		70.4	
Osteocalcin (ng/mL)	Tamoxifen	29	2.9	0.50
	Tamoxifen + Exemestane		2.7	
Total cholesterol (mg/dL)	Tamoxifen	30	195.8	0.34
	Tamoxifen + Exemestane		189.5	
HDL cholesterol (mg/dL)	Tamoxifen	30	64.2	0.0025
	Tamoxifen + Exemestane		59.8	
LDL cholesterol (mg/dL)	Tamoxifen	27	104.4	0.56
	Tamoxifen + Exemestane		106.8	
Triglycerides (mg/dL)	Tamoxifen	29	138.4	0.36
	Tamoxifen + Exemestane		132.7	
Estradiol (pg/mL)	Tamoxifen	29	5.8	<0.001
	Tamoxifen + Exemestane		A	
Estrone (pg/mL)	Tamoxifen	29	28.9	<0.001
	Tamoxifen + Exemestane		B	
Estrone sulfate (pg/mL)	Tamoxifen	28	364	<0.001
	Tamoxifen + Exemestane		25.1	

NOTE. A = all values below lower limit of quantification; B = more than half of values below lower limit of quantification.

statistically significant changes in osteocalcin, total, low-density lipoprotein cholesterol, or triglyceride levels. Estradiol, estrone, and estrone sulfate levels decreased >90%, consistent with magnitudes of effects expected with exemestane.

For 30 subjects completing 8 weeks of combined therapy, symptom checklist data showed no significant changes in frequencies of common drug-associated side effects such as vasomotor symptoms. Similarly, the FACT B quality-of-life questionnaire data showed no significant changes.

DISCUSSION

The current study is the same in design (nonrandomized, longitudinal) as a series of studies investigating AI or inactivator/selective estrogen receptor modulator/tamoxifen combinations (19, 25–27). Whereas there has been some loss of data for each of the individual variables investigated, these losses are small and are unlikely to have compromised the broad conclusions. This report complements that of Rivera et al., which addressed, in particular, pharmacokinetic changes and symptoms in patients on exemestane who had tamoxifen added to their treatment (19). In the current and referenced studies of AI/selective estrogen receptor modulator combinations, the sample sizes have been too small to allow definitive conclusions about symptomatic effects.

The principal findings reported here are encouraging with respect to the exemestane/tamoxifen combination. We found a suggestion that adding exemestane to tamoxifen led to further decreases in bone resorption markers. Tamoxifen alone decreases bone resorption and loss of bone mineral density, and limited data have suggested a similar effect of exemestane alone also (20, 28). In the current study however, only osteocalcin and all alkaline phosphatase were evaluated; assessment of other markers such as bone-specific alkaline phosphatase may have provided a clearer or more nuanced picture of the effects of this exemestane/tamoxifen combination in bone. However, we found a decrease in cardiovascular disease protective HDL cholesterol levels but no other evidence of impact of the exemestane/tamoxifen combination on the hepatic-mediated lipid-lowering effects of tamoxifen

alone and no evidence of an adverse symptom profile with this combination (21). Finally, we found no evidence that the presence of tamoxifen altered exemestane's profound estrogen-lowering capacity, which data support similar findings by Rivera et al. (19).

The laboratory data of Rivera et al. showing essentially superimposable plasma concentration time profiles for exemestane alone and exemestane plus tamoxifen are in contrast to the findings of lower letrozole levels and lower anastrozole levels in the presence of tamoxifen (19, 20, 29). The laboratory MCF-7 model data of Brodie et al. further suggest differences of these nonsteroidal and steroidal AI/selective estrogen receptor modulator combinations (10). It has been speculated that the explanation for the absence of additive benefit of anastrozole and tamoxifen together over either alone is a consequence of the agonist effect of tamoxifen in the extremely low-estrogen environment this AI creates (8). If there is a true additive effect of exemestane and tamoxifen as the two available animal data studies suggest, what might explain such differences (9, 10)? There may be differences in rodent metabolism of exemestane and the nonsteroidal AIs. Exemestane or its metabolites may have secondary effects (beyond the estrogen-lowering properties), which act to blunt the posited tickler estrogenic effect of tamoxifen. For example, Rivera et al. suggested an androgenic effect of exemestane as supported by other data (12, 30). Ongoing elegant studies of gene transcription effects of exemestane alone and in combination may provide further insight into the actions of exemestane alone or in the presence of tamoxifen.⁴

Defining an optimal and efficient approach to further investigate the exemestane/tamoxifen combination in human studies is challenging. A metastatic trial has been proposed by the North Central Cancer Treatment Group. A neoadjuvant study has particular challenges: in particular, in defining the best study end points. Clinical responses in bigger tumors can be difficult to recognize; tumors can become replaced by stroma of similar volume, and when serial tissue biopsies are used to monitor end

⁴ A. Elias, personal communication, April 1, 2004.

points, these can become more difficult to obtain with responses. Finally, the usefulness of a response seen in the neoadjuvant big-tumor setting, in predicting response in the micrometastatic setting is questionable (31). Adjuvant hormonal therapy studies are large expensive endeavors because of often expected small differences in treatment; the Tamoxifen and Exemestane Adjuvant Multicenter trial of 5 years of adjuvant exemestane versus tamoxifen seeks to accrue 4,400 women.

In summary, the current report suggests that the pharmacokinetic interaction of tamoxifen and exemestane does not adversely affect bone metabolism markers but lowers HDL cholesterol whereas not changing other hepatic lipid/lipoprotein markers. These findings support timely further investigations of this combination hormonal therapy.

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