women with primary breast cancer [4,5]. In a small phase II study of 19 postmenopausal patients with advanced breast cancer (ABC) failing on tamoxifen therapy, 37% of patients achieved a partial response and a further 32% achieved disease stabilisation. The median duration of response was 26 months [6]. No negative effects were observed on the liver, brain or genital tract. These clinical data confirmed the lack of cross-resistance between ICI 182780 and tamoxifen and suggested a prolonged duration of tumour control. To date, there are no clinical data available for the effects of ICI 182780 on bone and only a small amount of data (from non-comparative studies) for effects upon lipid profiles and the endometrium.

However, ICI 182780 is being studied versus the non-steroidal aromatase inhibitor anastrozole and tamoxifen in two separate phase III studies as part of a large clinical programme involving postmenopausal women with ABC who have either failed on tamoxifen therapy or not received prior tamoxifen therapy.

Another steroidal anti-oestrogen believed to be devoid of any oestrogen-agonist activity is the Roussel compound RU 58668. It is structurally similar to ICI 182780 and possesses all the properties of a pure anti-oestrogen in pre-clinical studies [7]. At present there are no clinical data available for this anti-oestrogen, but the pre-clinical data for RU 58668 suggest that it could be used for the treatment of ER-positive patients who have failed prior tamoxifen therapy and in breast cancer prevention [7]. The non-steroidal anti-oestrogen EM-800, which was developed as an oral 'pure' anti-oestrogen and behaves as a 'pure' anti-oestrogen in relation to the breast, uterus, vagina and hypothalamo-pituitary-gonodal axis [8], has recently been shown to inhibit bone loss and to reduce serum cholesterol levels [8], and may in fact turn out to be SERM rather than a SERD. There are no published clinical data for this compound.

Thus, ICI 182780, clearly offers an effective alternative in the clinic for patients whose disease has become resistant to tamoxifen [6], and might be more effective than tamoxifen as a first-line endocrine therapy. The results of the phase III studies and the clinical effects of ICI 182780 on bone density and the serum lipid profile are awaited.

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Determination of tamoxifen and its metabolites in the endometrial tissue of long-term treated women

G. Giorda [a,*], L. Franceschi [b], D. Crivellari [a], M.D. Magri [a], A. Veronesi [a], C. Scarabelli [a], M. Furlanut [b]

[a] *Gynecological Oncology Department, Centro di Riferimento Oncologico, Via Pedemontana Occidentale, I-33081 Aviano, Italy*
[b] *Department of Pharmacology, DPMSC, University of Udine, Italy*

Abstract

Concentrations of tamoxifen and its metabolites were analysed in the endometrium of 23 post-menopausal asymptomatic breast cancer patients who were on chronic tamoxifen therapy. Small endometrial samples were collected during diagnostic hysteroscopy. Analysis of both serum and tissue for these compounds was performed by mass spectrometry. Tamoxifen and its metabolites were

* Corresponding author. Fax: +39-434-659439.
E-mail address: ggiorda@ets.it (G. Giorda).

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far more concentrated in the endometrium than in serum; tamoxifen was also significantly more concentrated in endometrium with hyperplastic changes than in atrophic endometrium. Endometrial polyps of an additional 9 women showed a trend to a lesser concentration of compounds. Increased concentration of tamoxifen compounds could possibly be explained by the avidity of these compounds for oestrogen receptors (ER).

Keywords: Endometrium; Tamoxifen; Tissue distribution

It is still not determined whether tamoxifen and its metabolites are concentrated in human endometrial tissue. Only one study [1] has reported a determination of endometrial tamoxifen and metabolite concentration in 1 patient after 28 days withdrawal of the drug. The need for large samples (usually recovered from surgical specimens) for chromatographic methods has probably limited the determination of tamoxifen and its metabolites in human endometrial tissue. We have, therefore, chosen a sensitive method such as mass spectrometry (MS) to identify both tamoxifen and three of its metabolites, namely N-desmethyltamoxifen (metabolite X), N-Didesmethyl-tamoxifen (metabolite Z) and 4-hydroxytamoxifen (metabolite B).

23 postmenopausal breast cancer patients who had been taking tamoxifen for at least 2 months and had their last tablet within 24 h, underwent outpatient diagnostic hysteroscopy and small forceps endometrial biopsy (mean: 2 mg of endometrial tissue). Endometrial biopsies were duplicated, one for metabolite analysis and one for pathological examination. The endometrial sample and blood serum from the same patients were immediately frozen in liquid nitrogen for subsequent analysis. Identification of tamoxifen and metabolites extracted were performed with LC-MS [1]. A convenient calibration curve and some accepted assumptions have been made to extrapolate the small endometrial samples (mean: 2 mg) to 1 g of tissue and to consider 1 g of tissue equivalent to 1 ml of serum. Quantitative determinations of tamoxifen and its metabolites were then expressed as ng per either ml of serum or g of endometrial tissue. These results were then correlated with hysteroscopic features and endometrial pathology. Nine additional postmenopausal women with macroscopically non-malignant endometrial polyps were further examined and a mean 100 mg of each polyp was analysed.

Table 1
Serum and endometrial concentrations and ratio of endometrial to serum concentrations (□) of tamoxifen and metabolites in 23 postmenopausal asymptomatic women

	Serum (ng/ml)	Endometrium (ng/g)	Δ[a]
Tamoxifen	102±44	1887±762	22
Metabolite B	7±5	1952±1283	400
Metabolite X	318±158	4240±1642	17
Metabolite Z	28±16	2047±1809	100

[a] Ratio of endometrial to serum concentrations.

Serum concentration of tamoxifen and metabolites showed that all women were on chronic treatment. The concentrations and tissue/serum ratio (Δ) are reported in Table 1.

No correlation was found between tissue concentrations and hysteroscopic features (endometrial vascular congestion, cystic atrophy and mucosal adhesions). A significant correlation ($P < 0.05$) was instead found between tamoxifen concentration in 6 hyperplastic (2286 ng/g) versus 17 not hyperplastic endometria (1766 ng/g). In the polyps, tamoxifen and its metabolites were less concentrated than in the endometrium, but not to a significant extent.

In agreement with studies in rats, these preliminary data show that both tamoxifen and its metabolites are far more concentrated in the endometrium than in serum with a magnitude similar to other human non-malignant tissues [2]. This active concentration could possibly be explained by the avidity of these compounds for oestrogen receptors, but this has to be further confirmed by analysis of receptor distribution in the endometrium, especially in hyperplastic ones that should harbour a greater number of oestrogen receptors. The difference encountered in endometrial polyps could be biased by the presence of abundant stroma in the polyp samples compared with the superficial endometrial samples.

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