tamoxifen on plasma insulin-like growth factor I in patients with breast cancer. *Cancer Res* 1989, **49**, 1882–1884.
37. Committee on Diet, Nutrition and Cancer. Assembly of Life Sciences, National Research Council. *Diet, Nutrition, and Cancer*. Washington, DC: National Academy Press, 1982, 91.
38. Colletti RB, Copeland KC, Devlin JT, Roberts JD, McAuliffe TL. Effect of obesity on plasma insulin-like growth factor-I in cancer patients. *Int J Obes* 1991, 15, 523–527.
39. Emerman JT, Leahy M, Gout PW, Bruchovsky N. Elevated growth hormone levels in sera from breast cancer patients. *Horm Metab Res* 1985, 17, 421–424.
40. Love RR and Rose DP. Elevated bioactive prolactin in women at risk for familial breast cancer. *Eur J Cancer Clin Oncol* 1985, 21, 1553–1554.
41. Vennin Ph, Peyrat J-Ph, Bonneterre J, Louchez MM, Harris AG, Demaille A. Effect of the long-acting somatostatin analogue SMS 201-995 (Sandostatin) in advanced breast cancer. *Anticancer Res* 1989, **9**, 153–156.
42. Pollak MN, Costantino J, Polychronakos C, *et al.* Effect of tamoxifen on serum insulin-like growth factor I levels of stage I breast cancer patients. *J Natl Cancer Inst* 1990, 82, 1693–1697.

Acknowledgements—The authors thank Pr Huart and his colleagues from the Centre Regional de Transfusion Sanguine (Lille-France) and Dr Bukowski from the Barbieux Medical Center (Roubaix, France) for blood collection of control patients, and John Hall for reading the English. Supported by grants from the Fédération Nationale des Centres de Lutte contre le Cancer (Ligue Nationale Contre le Cancer funds, Paris, France), and the Association pour la Recherche sur le Cancer, Villejuif, France.

Eur J Cancer, Vol. 29A, No. 4, pp. 497–500, 1993.
Printed in Great Britain

0964–1947/93 $6.00 + 0.00
Pergamon Press Ltd

Oestrogenic Effects of Adjuvant Tamoxifen in Postmenopausal Breast Cancer

Tommy Fornander, Lars E. Rutqvist, Nils Wilking, Kjell Carlström and Bo von Schoultz

Oestrogenic influence of the non-steroidal anti-oestrogen tamoxifen may have consequences for the morbidity pattern among women on long-term adjuvant treatment. Subclinical oestrogenic effects of adjuvant tamoxifen on the tissue level was studied among 16 postmenopausal women in three different organ systems: the pituitary, the liver and bone. After 3 months of adjuvant tamoxifen prolactin levels decreased 66% ($P<0.001$) in comparison with pretreatment levels. There was an 80% increase in basal growth hormone after 3 months of treatment in comparison with pretreatment levels, which did not reach statistical significance ($P = 0.07$). Sex hormone binding globulin levels increased 39% ($P<0.01$) and IGF-1 (somatomedin C) levels decreased 20% ($P<0.05$). The levels of bone GLA protein (BGP; osteocalcin), a marker of bone osteoblastic activity, decreased 28% ($P<0.01$). In 13 of the patients bone mineral density (BMD) was measured before treatment and after 1 year. No significant change in BMD was observed. The results thus suggest a clear oestrogenic effect of tamoxifen on the pituitary, liver and bone in postmenopausal women.
Eur J Cancer, Vol. 29A, No. 4, pp. 497–500, 1993.

INTRODUCTION

TAMOXIFEN is widely used as an adjunct to surgery in the treatment of primary breast cancer. There are few acute side-effects and the substance is generally well tolerated [1–4]. The effect in human breast cancer is thought to be predominantly anti-oestrogenic through a blockade of oestrogen receptors (ER) in the cancer cells [5, 6]. Other mechanisms have been suggested to explain effects observed in tumours with a low ER content [7]. Although the anti-oestrogenic action of tamoxifen is well established in breast cancer, effects in other human tissues are not fully known. Tamoxifen is one of several triphenylethylene substances all of which are partial oestrogenic agonists/antagonists. The oestrogenic and anti-oestrogen effects of tamoxifen have been shown to be both species- and organ-specific. In the uterus of immature and ovariectomized mice, tamoxifen acts as a pure oestrogen agonist during short-term treatment, but in the immature rat uterus the substance is a partial oestrogenic agonist/antagonist [8–11]. In the chick, tamoxifen is a pure antagonist of oestrogen-stimulated growth of the oviduct [12]. This task seems to be even more complicated considering also the duration of treatment, e.g. long-term tamoxifen therapy to mice has been reported to produce anti-oestrogenic actions [13]. It is obviously not possible to extrapolate results from one system to another and firm conclusions about tamoxifen effects on different tissues in man cannot be drawn from animal results. Although data on tamoxifen effects on several biochemical markers have been published, the information is incomplete and

Correspondence to T. Fornander.
T. Fornander, L.E. Rutqvist and N. Wilking are at the Radiumhemmet, Karolinska Hospital, S-10401 Stockholm; K. Carlström is at the Department of Obstetrics and Gynecology, Huddinge University Hospital, Huddinge; and B. von Schoultz is at the Department of Obstetrics and Gynecology, Karolinska Hospital, Stockholm, Sweden.
Revised 11 Aug. 1992; accepted 1 Oct. 1992.

in some cases conflicting [14–20]. Here we report further biochemical evidence for subclinical oestrogenicity at the tissue level in certain organ systems known to be influenced by oestrogens, i.e. the pituitary, the liver and bone. The pituitary response to oestrogenic stimulation was assessed by analysis of basal levels of prolactin (PRL) and growth hormone (hGH). Serum levels of sex hormone-binding globulin (SHBG) and insulin-like growth factor 1 (IGF-1; somatomedin C), were used as markers for liver metabolic influence. Changes in serum levels of bone GLA protein (BGP; osteocalcin), were used as a measure of oestrogenicity in bone. The mentioned variables are all well established markers for oestrogenic influence in the respective organs.

PATIENTS AND METHODS

The patients included were 16 post-menopausal women with a mean age of 63 years (range 51–75 years) who all were about to begin adjuvant tamoxifen treatment 40 mg daily as an adjunct to surgery for early breast cancer. A total of 6 patients were in stage I, the rest in stage II. All women were more than 6 months post menopausal and free from known metabolic diseases, or diseases of the liver or skeleton. 1 patient had a constant about 40% increase in creatinine. With this exception, routine analyses of haemoglobin, platelets, white blood cell count, albumin, alanine aspartate aminotransferase, alkaline phosphatase and creatinine were within the normal range. The study was approved by the Karolinska Institute Ethics Committee. Informed consent was obtained from all participants.

Basal blood samples were collected 1–10 days before the start of adjuvant tamoxifen, i.e. about 6 weeks after surgery for breast cancer. Samples were also collected 12 weeks from the start of tamoxifen therapy. At both occasions, the blood samples were drawn intravenously (i.v.) between 8.00 and 8.30 a.m after an overnight fast. Serum was separated after centrifugation and stored at −70°C until assayed. Each single analysis was determined for all women together in a single assay.

Hormone analyses

Serum concentrations of hGH were determined by a two-site immunoradiometric assay (Pharmacia hGH IRMA, Pharmacia Diagnostics AB, Uppsala, Sweden). PRL was measured by radioimmunoassay (RIA) using a commercial kit (Pharmacia Prolactin RIA 100, Pharmacia Diagnostics AB, Sweden). Bound and free ^{125}I-PRL were separated by precipitation with polyethylene glycol. Serum SHBG was determined by a RIA using a commercial kit from MILAB (Malmö, Sweden). Serum levels of IGF-1 were determined radioimmunologically after precipitation of binding proteins with ethanol–acetic acid, using a commercial kit from Immunonuclear Corp. (Stillwater, MN, U.S.A.). Serum concentrations of BGP were determined by RIA using a polyclonal rabbit anti-bovine osteocalcin antibody (OSTK-PR, Compagnie Oris, Industrie S. A., France). The sensitivity of this assay is 0.35 ng/ml. The intra- and interassay variations for hGH were 7 and 12%, respectively, 9 and 11% for PRL, 4 and 8% for SHBG, 10 and 12% for IGF-1, and 7 and 10% for BGP.

Bone mineral measurements

13 of the patients in this study also participated in a concomitant prospective study of bone mineral changes during tamoxifen treatment. Single photon absorptiometry (SPA) at two levels of the distal forearm was performed before treatment and after 1 year. The method was described previously [21]. These patients were also interviewed about factors known to be of importance for the rate of bone mineral loss [21]. For this subset of patients the mean age was 62 years (range 52–74), the mean length was 165 cm (range 157–174), the mean weight was 68 kg (range 58–105) and the mean age at time of menopause was 50 years (range 45–55). A total of 4 patients had a history of oestrogen replacement therapy for 2–20 years and 5 patients were smokers (≥ 5 cigarettes/day). No patient was known for alcohol abuse.

Table 1. Serum levels (mean and range) of hormones and hormone-sensitive proteins before and after 3 months of tamoxifen 40 mg daily

	Before treatment	After 3 months	*P* value
hGH (mU/l)	3.4 (0.5–26)	6.1 (0.5–25)	0.066
PRL (μg/l)	9.9 (3.5–22)	3.4 (2.0–5.9)	<0.001
SHBG (nM)	42 (14–89)	58 (24–141)	0.001
IGF-1 (nmol/l)	16 (8.4–23)	13 (6.7–24)	0.028
BGP (ng/ml)	8.4 (1.5–13)	6.1 (1.8–12)	0.002

Statistical methods

Wilcoxon signed rank test for paired observations was used to determine the level of significance of differences between zero and treatment values.

RESULTS

Mean values and range for PRL, hGH, SHBG, IGF-1 and BGP before and after 3 months treatment with tamoxifen 40 mg daily are given in Table 1 and percental changes are illustrated by Fig. 1. The basal serum concentration of PRL decreased during treatment (all 16 women; $P<0.001$). The mean increase in basal hGH was 80%, but due to individual variation this change did not reach statistical significance. In 15 out of 16 women increased levels of SHBG were recorded. The mean increase was 39% compared to pretreatment values ($P<0.01$). Mean values for IGF-1 showed a 20% decline (reduced values in

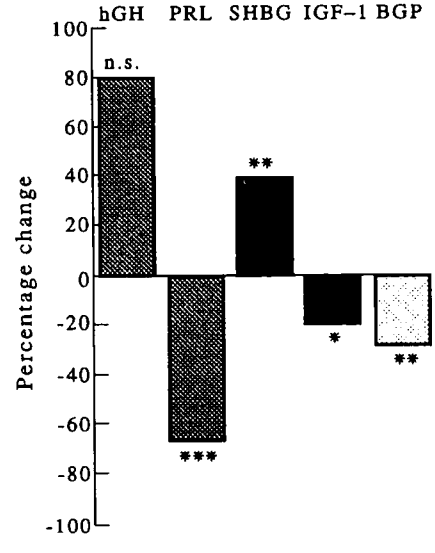

Fig. 1. Oestrogenic influence of tamoxifen treatment on different tissue variables expressed as per cent change in pretreatment values. Significant difference between pretreatment and treatment mean values are denoted by *($P<0.05$), **($P<0.01$), *($P<0.001$). ▩ = pituitary level, ■ = liver proteins, □ = bone marker.**

13 of 16 women; $P<0.05$). The mean BGP level decreased with 28% (reduced values in 13 of 16 women; $P<0.01$).

No statistically significant change in bone mineral density (BMD) was seen either at the most distal level of the forearm representing mainly trabecular bone (±0%; $P=0.7$) or at the slightly more proximal level representing mainly cortical bone (−1%; $P=0.2$) (data not presented). The changes in BMD were too small to be possible to be correlated to the changes in BGP.

DISCUSSION

Recently a 6-fold increase in the incidence of endometrial cancer was observed in the Stockholm adjuvant tamoxifen trial [22]. This observation has not been confirmed in other trials using a dose of tamoxifen lower than 40 mg daily, as was the case in the Stockholm trial [2, 4, 23, 24]. Thus, the possibility of a dose–effect relationship or a threshold effect cannot be ruled out. Observations of oestrogenic effects in the female genital tract, however, have been reported also with lower doses of tamoxifen [25–29]. These and other observations indicate that this anti-oestrogen may have clearly oestrogenic effects in human organ systems. Our current understanding of clinical tamoxifen effects in different organs is far from complete. In the present study we found further evidence of oestrogenic effects of tamoxifen on the tissue level in three different organ systems, the pituitary, liver and bone.

Oestrogen treatment is well known to enhance the hGH secretion in both men and women [30]. The possible influence of tamoxifen treatment on hGH has not been extensively studied. Unchanged hGH concentration with tamoxifen added to previously unopposed oestrogen therapy has been reported [14, 31]. We found a tendency for increases in the levels of hGH also with tamoxifen. A weak oestrogen is well known to cause a reduction in PRL levels while, on the other hand, high doses of a potent oestrogen have been shown to increase the PRL levels [15]. In this study we observed a 66% fall in basal PRL levels. With 2 mg 17β-oestradiol daily only a 30% reduction was reported [15, 32]. With tamoxifen 40 mg daily 36 and 71% decreases in PRL have been observed after 1 and 3 months, respectively. On the other hand Paterson *et al.* [14] found no effect on PRL with 40 mg tamoxifen daily during a treatment period of 1–8 weeks. Thus, in this study, tamoxifen appears to act as a weak oestrogen on pituitary function with regard to hGH and PRL levels, in addition to the previously reported about 30–50% decrease in baseline gonadotropin levels [15, 33].

The serum concentrations of a great many liver-derived proteins are known to increase after oral oestrogen administration and have been used as markers for the hepatocellular impact of oral oestrogens [34, 35]. SHBG has previously been reported to increase significantly with long-term treatment [17]. In this study treatment with tamoxifen gave a 39% increase in SHBG. This should be compared with an increase of 25% following 2 mg oestradiol-17β daily or 75% following 50 μg oral ethinyl oestradiol daily [36]. IGF-1, another polypeptide produced by the liver, was observed to decrease by 20%. This is similar to an about 35% decrease reported with 10 μg ethinyl-oestradiol treatment [31]. A significant decrease in IGF-1 with tamoxifen in lower dose (20 mg daily) has also been observed by others [20]. Other previously reported effects of tamoxifen on liver-derived proteins are, e.g. changes in pregnancy zone protein, transcortin and lipoproteins [15, 37–40]. The observed effects of tamoxifen in this study thus implies that tamoxifen acts as a clear oestrogen on several aspects of liver metabolism.

Osteocalcin is a bone matrix γ-carboxyglutamate protein (BGP), synthesised by proliferating osteoblasts. A small fraction is released to the circulation and it has been proposed as a useful marker for bone mineral metabolism [41]. Oestrogen replacement therapy, known to reduce the post-menopausal bone loss, has been found to reduce the circulating BGP level by about 50% [42]. Reports on tamoxifen treatment and BGP levels are scanty. Fentiman *et al.* who studied BGP and bone mineral content in premenopausal patients found no significant changes in osteocalcin or bone mineral content [18]. In this study, however, we found a significant 28% decrease in BGP levels with tamoxifen. There are previous but scanty animal and clinical indications of tamoxifen acting as an oestrogenic agonist also on bone tissue [43, 44]. In a recent study it was found that women treated with tamoxifen 40 mg daily for 5 years had increased BMD compared to controls and those treated for 2 years, but the differences were not statistically significant [21]. 13 of the patients in the present study concomitantly participated in a prospective study of BMD. Preliminary data showed no statistical significant change in BMD in cortical bone or in trabecular bone. The complete results from the prospective bone mineral measurements of these and additional tamoxifen patients will be presented in a coming paper including a comparison with endocrinologically untreated patients. Our observation of decreased BGP levels is, however, clear biochemical evidence of an oestrogenic effect of tamoxifen in bone. Further studies are thus indicated to establish if tamoxifen in fact prevents post-menopausal bone loss.

This study was made on a small number of patients and the results should therefore be judged cautiously considering the great variability of the hormone patterns in humans. These data, however, add to previous reports that tamoxifen exerts clear oestrogenic influence in humans on three different tissue levels, the pituitary, the liver and bone. Both endogenous oestrogen levels and oestrogen replacement therapy have been related to health and illness in postmenopausal women. The consequences of long-term tamoxifen treatment should therefore be evaluated through careful monitoring of the randomised trials. Other anti-oestrogens with more pure anti-oestrogenic effects are currently being developed. These agents are potentially attractive due to expectations of a faster and better antitumour effect. However, the changed balance between agonistic/antagonistic oestrogen effects may also have implications with regard to side-effects which may be different and more unfavourable than with tamoxifen.

1. Ludwig Breast Cancer Study Group. Randomised trial of chemo-endocrine therapy, endocrine therapy and mastectomy alone in postmenopausal patients with operable breast cancer and axillary node metastasis. *Lancet* 1984, i, 1256–1260.
2. CRC Adjuvant Breast Trial Working Party. Cyclophosphamide and tamoxifen as adjuvant therapies in the management of breast cancer. Preliminary Analysis by the CRC Adjuvant Breast Trial Working Party. *Br J Cancer* 1988, 57, 604–607.
3. Baum M, Brinkley DM, Dosset JA, *et al.* Controlled trial of tamoxifen as single adjuvant agent in the management of early breast cancer. Analysis at Eight Years by "Nolvadex" Adjuvant Trial Organisation. *Br J Cancer* 1988, 57, 608–611.
4. Ribeiro G, Swindell R. The Christie hospital adjuvant tamoxifen trial: status at 10 years. *Br J Cancer* 1988, 57, 601–603.
5. Jordan VC. Antitumor activity of antiestrogen ICI 46,474 (tamoxifen) in dimethylbenzantracene (DMBA)-induced rat mammary carcinoma model. *J Steroid Biochem* 1974, **4**, 354.
6. Lippman M, Bolan G, Huff K. Interactions of antiestrogen with human breast cancer in long-term tissue culture. *Cancer Treat Rep* 1976, **60**, 1421–1429.
7. Patterson MB, Furr B, Wakeling A, Battersby MA. The biology

and physiology of "Nolvadex" (tamoxifen) in the treatment of breast cancer. *Breast Cancer Res Treat* 1982, **2**, 363–374.
8. Terenius L. Two modes of interaction between oestrogen and antioestrogen. *Acta Endocrinol* 1970, **64**, 47–58.
9. Terenius L. Structure–activity relationships of antioestrogens with regard to interaction with 17β-oestradiol in the mouse uterus and vagina. *Acta Endocrinol* 1971, **66**, 431–447.
10. Martin L, Middleton E. Prolonged oestrogenic and mitogenic activity of tamoxifen in the ovariectomized mouse. *J Endocrinol* 1978, **78**, 125–129.
11. Furr B, Jordan VC. The pharmacology and clinical uses of tamoxifen. *Pharmacol Ther* 1984, **25**, 127–205.
12. Sutherland RL, Mester J, Baulieu EE. Tamoxifen is a potent "pure" antioestrogen in the chick oviduct. *Nature* 1977, **267**, 434–435.
13. Jordan VC, Lababidi MK. Anti-oestrogenic and anti-tumor properties of prolonged tamoxifen therapy in C3H/OUJ mice. *Eur J Cancer* 1990, **26**, 718–721.
14. Paterson AG, Turkes A, Groom GV, Webster DJ. The effect of tamoxifen on plasma growth hormone and prolactine in postmenopausal women with advanced breast cancer. *Eur J Cancer Clin Oncol* 1983, **19**, 919–922.
15. Helgason S, Wilking N, Carlström K, Damber MG, von Schoultz B. A comparative study of the estrogenic effects of tamoxifen and 17β-estradiol in postmenopausal women. *J Endocrinol Metab* 1982, **54**, 404–408.
16. Sakai F, Cheix F, Clavel N, *et al.* Increases in steroid binding globulins induced by tamoxifen in patients with carcinoma of the breast. *J Endocrinol* 1978, **76**, 219–226.
17. Jordan VC,Fritz NF, Tormey DC. Long-term adjuvant therapy with tamoxifen: effects on sex hormone binding globulin and antithrombin III. *Cancer Res* 1987, **47**, 4517–4519.
18. Fentiman IS, Caleffi A, Rodin B, Fogelman I. Bone mineral content of women receiving tamoxifen for mastalgia. *Br J Cancer* 1989, **60**, 262–264.
19. Szamel I, Hindy I, Vincze B, Kerpel-Fronius S, Eckhardt S. Influence of antiestrogen drugs on the sex hormone and sex hormone-binding globulin levels in breast cancer patients. *Ann N Y Acad Sci* 1988, **538**, 265–279.
20. Pollak M, Constantino J, Polychronakos C, *et al.* Effect of tamoxifen on serum insulinlike growth factor 1 levels in stage I breast cancer patients. *J Natl Cancer Inst* 1990, **82**, 1693–1697.
21. Fornander T, Rutqvist LE, Sjöberg HE, Blomqvist L, Mattsson A, Glas U. Long-term adjuvant tamoxifen in early breast cancer: effect on bone mineral density in postmenopausal women. *J Clin Oncol* 1990, **8**, 1019–1024.
22. Fornander T, Rutqvist LE. Adjuvant tamoxifen and second cancers. *Lancet* 1989, **i**, 616.
23. Stewart HJ, Knight GM. Tamoxifen and the uterus and endometrium. *Lancet* 1989, **i**, 375–376.
24. Fisher B, Costantino J, Redmond C, *et al.* A randomized clinical trial evaluating tamoxifen in the treatment of patients with node-negative breast cancer who have estrogen-receptor-positive tumors. *N Eng J Med* 1989, **320**, 479–484.
25. Ferrazzi E, Cartei G, Mattarazo R, Fiorentino M. Oestrogen-like effect of tamoxifen on vaginal epithelium. *Br Med J* 1977, **I**, 1351–1352.
26. Boccardo F, Bruzzi P, Rubagotti A, Rosso R. Estrogen-like action of tamoxifen on vaginal epithelium in breast cancer patients. *Oncology* 1981, **38**, 281–285.
27. Budel VM, Paridaens RJ, Spetschinsky A, Heuson JC, Gompel C. Effects of aminoglutethemide plus hydrocortisone on the genital tract of postmenopausal women with advanced breast cancer. A clinical and cytologic survey. *Anticancer Res* 1986, **6**, 709–712.
28. Nuovo MA, Nuovo GJ, McCaffrey RM, Levine RU, Barron B, Winkler B. Endometrial polyps in postmenopausal patients receiving tamoxifen. *Int J Gynecol Pathol* 1989, **8**, 125–131.
29. Neven P, DeMuylder X, VanBelle Y, Vanderick G, DeMuylder E. Tamoxifen and the uterus and endometrium. *Lancet* 1989, **i**, 375.
30. Dawson-Huges B, Stern D, Goldman J, Reichlin S. Regulation of growth hormone and somatomedin-C secretion in postmenopausal women: effect of physiological estrogen replacement. *J Clin Endocrinol Metab* 1986, **63**, 424–432.
31. Fröhlander N, von Schoultz B. Growth hormone and somatomedin C during postmenopausal replacement therapy with oestrogen alone and in combination with antioestrogen. *Maturitas* 1988, **9**, 297–302.
32. Helgason S, Carlström K, Damber M-G, von Schoultz B. Gonadotrophins and prolactin during estrogen replacement therapy. *Acta Obstet Gynecol Scand* 1981, **60**, 515–516.
33. Coombes RC, Powles TJ, Rees LH, *et al.* Tamoxifen, aminoglutethimide and danazol: effect of therapy on hormones in postmenopausal patients with breast cancer. *Br J Cancer* 1982, **46**, 30–34.
34. Geola FL, Frumar AM, Tataryn IV, *et al.* Biological effects of various doses of conjugated equine estrogens in postmenopausal women. *J Clin Endocrinol Metab* 1980, **51**, 620–625.
35. Helgason S. Estrogen replacement therapy after the menopause. Estrogenicity and metabolic effects. *Acta Obstet Gynecol Scand* 1982, **107** (suppl.), 7–29.
36. Helgason S, Damber J-E, Damber M-G, von Schoultz B, Selstam G, Södergård R. A comparative longitudinal study on sex hormone binding globulin capacity during estrogen replacement therapy. *Acta Obstet Gynecol Scand* 1982, **61**, 97–100.
37. Fex G, Adielsson G, Mattson W. Oestrogen-like effects of tamoxifen on the concentration of proteins in plasma. *Acta Endocrinol* 1981, **97**, 109–113.
38. Rössner S, Wallgren A. Serum lipoproteins and proteins after breast cancer surgery and effects of tamoxifen. *Atherosclerosis* 1984, **52**, 339–346.
39. Bruning PF, Bonfrer JMG, Hart AAM, *et al.* Tamoxifen, serum lipoproteins and cardiovascular risk. *Br J Cancer* 1988, **58**, 497–499.
40. Bertelli G, Pronzato D, Amoroso D, *et al.* Adjuvant tamoxifen in primary breast cancer: influence on plasma lipids and antitrombin III levels. *Breast Cancer Res Treat* 1988, **12**, 307–310.
41. Brown JP, Delmas PD, Malaval L, Edouard C, Chapuy MC, Menunier PJ. Serum bone GLA-protein: a specific marker for bone formation in postmenopausal osteoporosis. *Lancet* 1984, **i**, 1091–1093.
42. Johansen JS, Riis BJ, Delmas PD, Christiansen C. Plasma BGP: an indicator of spontaneous bone loss and of the effect of oestrogen treatment in postmenopausal women. *Eur J Clin Invest* 1988, **18**, 191–195.
43. Jordan VC, Phelps E, Lindgren JU. Effects of anti-estrogen on bone in castrated and intact female rats. *Breast Cancer Res Treat* 1987, **10**, 31–35.
44. Turken S, Siris E, Seldin D, Flaster E, Hyman G, Lindsay R. Effects of Tamoxifen on spinal bone density in 10 postmenopausal women with breast cancer. *J Natl Cancer Inst* 1989, **81**, 1086–1088.

Acknowledgements—The study was supported by the King Gustaf V Jubilee Fund, the Swedish Cancer Society and the Swedish Medical Research Council (5982).