PERGAMON

European Journal of Cancer 39 (2003) 572–579

European Journal of Cancer

www.ejconline.com

Review

Anti-oestrogenic chemoprevention of breast cancer—the need to progress

Trevor J. Powles*

Royal Marsden Hospital, Downs Road, Sutton, Surrey SM2 5PT, UK

Received 12 November 2002; accepted 15 November 2002

Abstract

Clinical trials of the selective oestrogen receptor modulator (SERM), tamoxifen, have shown an early reduction in risk of breast cancer in healthy women of approximately 40%, but with associated risks and benefits to normal tissues. An overall clinical benefit and the identification of the women at risk of breast cancer who may gain benefit from tamoxifen has not been clearly established. The identification of those women at risk who are most likely to gain benefit, and the development of other SERMs and aromatase inhibitors which might be more active and have a more beneficial spectrum of activity on normal tissues in healthy women is essential, if the aim of preventing breast cancer in healthy women is to be achieved.

Keywords: Tamoxifen; Breast cancer; Risk factors; Chemoprevention; Side-effects

1. Introduction

The concept that oestrogen is involved in the development of most breast cancers [1–3] has encouraged the use of antioestrogens, such as tamoxifen in healthy women to prevent the disease. Clinical trials using tamoxifen started over 15 years ago [4], and have now accrued over 25 000 healthy women, but results at this time have produced what appears to be conflicting and in many ways unconvincing results. Although, overall, there appears to be a reduction in the early incidence of breast cancer [5], this effect is not large, especially for poor prognosis invasive cancers, and there is no evidence that this reduction will be maintained in the long term. Furthermore, there is no clear cut evidence indicating which women gain a risk reduction with tamoxifen and whether this is of clinical benefit compared with treating the cancers as they arise. Toxicity by tamoxifen, in these otherwise healthy women including an increase in the risk of endometrial cancer, is not insignificant.

This paper aims to put into perspective the results of the trials so far, evaluate the risk factors which have been identified which may predispose to oestrogenic promotion of breast cancer, and review the future strategy for anti-oestrogenic prevention of breast cancer in healthy women.

2. Breast cancer prevention trials

A pilot trial was started at the Royal Marsden Hospital in 1986, designed to evaluate the feasibility and safety of tamoxifen given to healthy women to prevent breast cancer. The early results showed that tamoxifen appeared to have a 'selective' anti-oestrogenic effect on the breast with potentially beneficial oestrogenic effects on normal tissues [4] later defined as selective oestrogen receptor modulation (SERM). This encouraged the start in 1992 of multicentre trials in the USA, UK, Italy and other parts of the world. The Royal Marsden trial continued as a single centre trial.

The early results of all of these well conducted, large, double-blind randomised, placebo-controlled trials have now been reported and are summarised in Table 1 and in the text below.

* Tel.: +44-20-8661-3361; fax: +44-20-8661-7313.
E-mail address: trevor.powles@rmh.nthames.nhs.uk (T.J. Powles).

0959-8049/03/$ - see front matter
doi:10.1016/S0959-8049(02)00771-2

Table 1
Summary of the randomised tamoxifen chemoprevention of breast cancer trials

Trial name [Ref.]	No. of women	Age limits (Median)	Breast cancer risk criteria for entry	Previous LCIS or ADH (%)	% postmeno-pausal	Use of HRT	Median follow-up (months)	Total no Breast Cancers (Relative Risk and Confidence Intervals) Placebo Tam	Comments
NSABP1-1 [7]	13,388	> 34 (52)	Gail estimated risk of 1.66/1000/year or previous LCIS (25)	17	61	No	69	244 124 (RR 0.51, CI 0.39–0.66)	Most benefit for LCIS and ADH Reduction in fractures
Italian National Trial [9,10]	5408	35–70 (51)	Hysterectomy (74% had also had an ovariectomy) with no special risk factors for breast cancer	0	NA	Yes	94	45 34 (RR0.76 CI 0.47–1.60)	Only women taking HRT had a significant benefit
Royal Marsden Trial [11]	2494	30–70 (47)	Estimated age-adjusted 4-fold risk based on family history	0	34	Yes	120	75 62 (RR0.83 CI NA)	66% premenopausal with high risk of carrying breast cancer gene
The International breast cancer intervention trial [12]	7140	35–70 (50.9)	Estimated age adjusted 2- to 3-fold risk based on family history and other risk factors	5	49.9	Yes	50	101 68 (RR0.67 CI 0.49–0.91)	Lesser effect than P1 Especially or Invasive cancers

LCIS, lobular carcinoma *in situ*; HRT, hormone replacement therapy; ADH, atypical ductal hyperplasia; NA, not available; NSABP, National Surgical Adjuvant Breast Project.

2.1. The National Surgical Adjuvant Breast Project (NSABP P-1)

The NSABP P-1 trial randomised 13 388 healthy women with an estimated risk of breast cancer of at least 1.66/100 women/5 years determined by the Gail model [6]. This model includes age, number of previous benign breast biopsies, age at menarche and first live birth, and number of first degree relatives with breast cancer. In 1998, the results of this trial were published [7] and the observed risk reduction of breast cancer by 49% ($P = 0.0001$) was considered so striking that the trial was prematurely closed and unblinded before a full evaluation of the overall clinical benefits and risks, including mortality, could be made. This decision meant that no further randomised data from a longer follow-up could be obtained from this trial and the crucial question of whether it was better to reduce risk rather than treat the disease as it arose remained uncertain.

This was particularly so because most of the 85 less cancers which had not occurred in the women on tamoxifen compared with placebo were likely to be small, oestrogen receptor (ER)-positive and lymph node-negative cancers and highly curable if allowed to occur. Furthermore, the toxicity caused by tamoxifen in the 6681 healthy women who had received the drug, on average, for nearly 4 years, was more than had been anticipated, with increases in endometrial cancer, polyps and cysts, thromboembolism, cataracts and vasomotor symptoms.

A mathematical model was developed in an attempt to balance the observed risk reduction against the toxicity [8]. Those who were likely to gain most benefit were the younger women who had an increased risk of breast cancer, particularly those with benign pathology such as lobular carcinoma *in situ* (LCIS) or atypical ductal hyperplasia (ADH) or postmenopausal women who had had a hysterectomy. These results did not establish that giving tamoxifen to healthy women to prevent breast cancer was better than treating the cancers as they arose.

It is clearly encouraging that this trial was able to show a very significant and substantial overall risk reduction for breast cancer but, unfortunately, many critical questions remained unanswered by this trial.

2.2. The Italian National Trial (INT)

Women accrued to this trial had no special risk factors for breast cancer but, because of concerns about the effects of tamoxifen on the uterus, all women needed to have had a hysterectomy (74% of whom had also had an oophorectomy). The results of this trial reported in 1998 [9], with an update in 2002 [10], showed no significant overall risk reduction for breast cancer, although there was a risk reduction for those women

who received hormone replacement therapy (HRT). The women in this trial were generally at a low risk of breast cancer, probably because most had had a previous oophorectomy. This may have compromised any subsequent tamoxifen risk reduction unless the women were also receiving HRT.

2.3. The Royal Marsden Trial (RMHT)

In 1986, when this trial started, because of the possible unknown toxicity of tamoxifen in healthy women, only women with a strong family history of breast cancer were considered eligible. The participants were therefore younger and had an age-corrected risk for breast cancer of approximately 4-fold which was higher than participants in all the other trials. Furthermore, women with high risk benign breast pathology such as LCIS were excluded. The results, reported in 1998 [11], showed no risk reduction for breast cancer by tamoxifen.

2.4. The International Breast Intervention Study l (IBIS-I)

The results of this trial, which randomised 7140 women with an age-corrected risk for breast cancer of approximately 2- to 3-fold, were similar to the NSABP P-1 trial [12]. There was a smaller, but still significant, risk reduction for all breast cancers of approximately 32%, but for invasive cancers this was not significant (64 tamoxifen, 85 placebo, Odds Ratio (OR) 0.75, 95% Confidence Interval (CI) of 0.54–1.04). The 21 less invasive breast cancers which did not occur in women on tamoxifen were all ER-positive, and mostly axillary lymph node-negative cancers and of less than 2 cm. If they had been allowed to occur they would have been mostly curable.

The toxicity for this trial was similar to that reported for the NSABP P-1 trial, again raising the questions of whether there was likely to be any overall clinical benefit and whether prevention, by the fairly indiscriminant use of tamoxifen, given for 5 years to large numbers of healthy, relatively low-risk women is better than the treatment of the few extra cancers as they arise. This is particularly important in this trial because there was a significant ($P=0.01$) increase in overall mortality for women on tamoxifen.

2.5. The multiple outcomes of raloxifene evaluation (MORE)

Independent of the tamoxifen trials, in 1994, a trial was started in women with osteoporosis [13] evaluating the prevention of fracture potential of another selective oestrogen receptor modulator (SERM), raloxifene. Women in this trial had annual mammography to monitor the effect of this drug on breast cancer incidence.

Annual mammography in this osteoporosis trial showed that after 30 months of follow-up, there was a 76% reduction in the incidence of breast cancer [14,15] associated with a significant reduction in the incidence of osteoporotic fractures in the spine [16], in serum cholesterol [17], and in cardiac events in women at increased risk of heart disease [18]. Like tamoxifen, there was an increase in thromboembolism and vasomotor symptoms. However, there was no evidence of an increase in the risk of endometrial cancer, polyps, cysts or endometrial thickening. Raloxifene, unlike tamoxifen, had no oestrogenic effects on the uterus, indicating that its oestrogenic activity is relatively impeded. A similar impeded oestrogenic activity on breast cells could account for the relatively high risk reduction of breast cancer reported in this trial, compared with the tamoxifen trials.

2.6. Overview (meta-analysis)

An overview of the four tamoxifen breast cancer prevention trials has recently been undertaken evaluating the main outcomes of breast and endometrial cancer incidence, vascular events and mortality [5]. The combined data showed a 38% reduction in breast cancer incidence (95% CI 28–46%, $P=0.001$). There was an increase in endometrial cancer events (Relative Risk (RR) 2.4, CI 1.5–4.0) and thromboembolic events (RR 1.9, CI 1.4–2.6), but not in cardiovascular events. Overall, there was no significant effect on mortality (RR 0.91, CI 0.70–1.18). The data, together with the data from the very large breast cancer adjuvant treatment trials [19], indicate that it is very unlikely that mortality is increased by tamoxifen.

The range in the risk reduction results of the four individual tamoxifen trials on breast cancer incidence are only just statistically compatible, indicating that factors other than statistical variation are likely to account for the differences in results. The risk characteristics of the women in these trials could be a major factor. For example, in the P-1 trial, younger women who had ADH and LCIS, which are usually ER-positive, were included. This could have accounted for a higher chemoprevention effect in these women. In contrast, in the Marsden trial, women with high-risk benign pathology were excluded, whereas a high proportion of younger women who were likely to be carrying high-risk breast cancer predisposing genes, which may predispose to ER-negative cancers, were included. It is possible that these women were less likely to have a risk reduction by tamoxifen. The risk criteria for women in the IBIS trial were somewhere between that for the P-1 trial and the Marsden trial.

Although, at first sight, a 38% risk reduction of breast cancer using tamoxifen seems substantial, the absolute reduction in the numbers of women who develop breast

cancer in the prevention setting is small in comparison with the very large numbers of healthy women who need prolonged, potentially toxic treatment to achieve this. To make prevention of breast cancer by endocrine intervention a feasible proposition, the efficacy of the treatments must be maximised. This can be done in two ways. Firstly, by using agents which are more effective and less toxic than tamoxifen and secondly by defining risk factors which will identify those women at significant risk who are most likely to gain benefit from the use of these agents. By eliminating those women unlikely to gain benefit from future trials, the efficacy and risk benefit ratio will improve. What risk factors need to be evaluated and what agents should be tested?

3. Risk factors for breast cancer

Many factors have been implicated in breast cancer risk including increasing age [20] early menarche and late menopause [6,21–23] family history [6,21–24] early age at first live birth [6,21,23], benign breast biopsy [6,21,23] including ADH [25] and LCIS [7] cytological ductal atypia [25–30] increased breast mammographic density [31–33], high bone density [34,35], high circulating oestrogen levels [3,36], high plasma insulin-like growth factor levels [37,38], high body mass [33,39,40] and use of oral contraception and hormone replacement therapy [41]. Most of these risk factors are likely to have an underlying oestrogenic basis and some have an established inherited component (Table 2).

Risk factors may be caused by genetic mutations which increase the risk of breast cancer by increasing oestrogenic activity in the body. This can be caused by a high production of oestrogen or by increased cellular sensitivity to normal levels of oestrogen [42]. Other genetic mutations such as *BRCA 1* may cause breast cancer by non-oestrogenic mechanisms such as defects in the control of cell replication, or DNA repair. However, the incidence of the second copy mutation and other required gene mutations may be increased by oestrogen promotion of cell replication, thereby increasing the penetrance of the carrier state.

Table 2
Risk factors for breast cancer

Risk factors	Likely to be endocrine based	Likely to be inherited	[Refs]
Increasing age	+ or −		[20]
Early menarche	+		[6,21–23]
Late menopause	+		[6,21–23]
Family history	+ or −	+ + +	[6,21–24]
Early first pregnancy	+		[6,21,23]
Benign breast biopsy		?	[6,21,23]
Cellular atypia	+	?	[25–30]
Lobular carcinoma in situ	+	?	[7]
High breast mammographic density	+	+ +	[31–33]
High bone mineral density	+	+ +	[34,35]
High serum oestrogens	+	?	[3,36]
High plasma IGF		?	[37,38]
High body mass		?	[33,39,40]
Use of oral contraceptives	+		[41]
Use of HRT	+		[41]

IGF, insulin-like growth factor.

As an example of an inherited oestrogenic risk factor, high mammographic breast density is of special interest, because it does appear to be predominantly genetically determined [43–45] and related to higher circulating levels of oestrogen [43]. Furthermore, it is increased by the use of HRT [46] and reduced by the use of tamoxifen [43,44], indicating that it is a phenotypic marker of a genetic oestrogenic predisposition to breast cancer risk which could be used to identify and monitor an anti-oestrogenic intervention.

Many of the known risk factors are interrelated, probably because of an underlying increased oestrogenic risk. For example, the link between the predictive value of bone density and a family history of breast cancer depends on high levels of plasma oestrogen [47] and it is these women who have the risk reduction of breast cancer with raloxifene [48].

Combinations of risk factors may be synergistic and produce models which better predict breast cancer. For example, the Gail model [6] is based on the number of first degree relatives with breast cancer, age at menarche, age at first live birth, and the number of benign breast biopsies. Most of these risk factors are related to oestrogen. In contrast, the Claus model [49], developed in a younger high-risk population, is based on a complete family history of breast and other cancers. This model identifies women who are likely to have high penetrance breast cancer predisposing genes, which may not be primarily oestrogen based. Models for risk which include risk factors which are not oestrogen based may identify women at high risk of developing cancers which are resistant to tamoxifen. These women, although at high risk, would not gain benefit from tamoxifen prevention of breast cancer.

An algorithm needs to be developed which is based on endocrine risk factors such as mammographic breast density, bone mineral density, plasma oestrogen levels, menstrual, obstetric and lactation history, benign ER-positive high-risk histology such as ADH or LCIS, use of oral contraceptives and HRT and any other factors which may have an oestrogenic basis. The identification of the genes which may predispose to these risks would really make the prospect of endocrine prevention compelling. What genes could be involved?

It is likely that most breast cancers are initiated by single or multiple genetic defects [50]. Some of these genetic events will need little promotion in order to develop into clinical cancers. Others will depend on promotion to become clinical cancers. Oestrogen is

probably the most important promoter for breast cancer and any inherited genetic polymorphisms which cause an increase in tissue oestrogen levels or in the sensitivity of the cells to oestrogen, may be important inherited risk factors for breast cancer. These inherited risks may be reflected in a family history for breast cancer which will not be as well defined as for the high-risk genes such as BRCA, but may be identified by inherited phenotypic features such as high mammographic density, high bone mineral density, high-risk benign pathology such as LCIS, ADH and atypia, high levels or more prolonged pre-menopausal levels (early menarche or late menopause) of circulating oestrogens and other less well defined features such as body mass.

The likely gene targets for enhanced oestrogenic activity and 'super' promotion are the genes involved in oestrogen synthesis [51], in oestrogen metabolism and in oestrogen binding and those genes that determine the sensitivity of ERs (ERα and ERβ) to oestrogen [42]. The components of the ER most likely to increase the oestrogen sensitivity of ER would be changes in the activity of co-regulators, particularly overexpression of a co-activator such as AIB1 [52] or a mutation of a co-repressors such as NCoR or SMRT. Mutations of the ERs, particularly ERβ which modulates the oestrogen activation of ERα could cause 'super' promotion, as could mutations in critical parts of ERα which bind the co-repressors [53–55]. Other factors such as HER2 [52], epidermal growth factor (EGF) and insulin-like growth factor 1 (IGF1) [56], and other hormone receptors such as the progesterone receptors interact with ER and inherited polymorphisms which affect the activity of any of these could increase the oestrogenic promotion of breast cancer.

Inherited mutations in the ER mechanism which increases the sensitivity to oestrogen would undoubtedly have an effect on the balance of oestrogenic and anti-oestrogenic effects of tamoxifen so that tamoxifen could lose its prevention effect and even become a promoter of breast cancer.

Having identified the risk factors which make it more likely that women will gain benefit from an endocrine intervention what are likely to be the most effective and least toxic endocrine agents to be tested.

4. Agents which may be more effective and less toxic than tamoxifen

It therefore seems that the preventive effect of tamoxifen depends on its anti-oestrogenic anti-proliferative effect being more than its oestrogenic proliferative effect and this may vary according to the genetic risk factors involved in the development of the cancer. Other SERMS may have less oestrogenic activity because of differences in structure. For example, tamoxifen is oestrogenic on the uterus whereas raloxifene, which has a side chain which masks a critical part of the ER, preventing the recruitment of the coactivator SRC1, is anti-oestrogenic on the uterus [57]. This impeded oestrogenic activity of raloxifene might make it more active than tamoxifen for prevention of breast cancer and account for the encouraging results from the MORE trial. Raloxifene and tamoxifen are now being directly compared for risk reduction and toxicity in the NSABP P-2 trial which will accrue 22 000 women with a similar Gail risk to those women in the P-1 trial. Other SERMs such as lasofoxifene are being evaluated for breast cancer risk reduction.

For the most part, the spectrum of activity of these SERMs on normal tissues and on cancer cells will depend on their chemical structures. It should therefore be possible to achieve multiple benefits on normal tissues and an enhanced anticancer effect compared with tamoxifen (Table 3).

Another approach is to look at agents which have only anti-oestrogenic activity such as the aromatase inhibitors or the so called 'pure' anti-oestrogens. The lack of any oestrogenic activity should make these

Table 3
Properties of an ideal serm

Organ	Objective	Surrogate marker
Breast	Reduce breast cancer	Breast density
Bone	Reduce osteoporotic fracture	Bone mineral density
		Bone markers
Lipids	Reduce atheroma/stroke/mycardial infarction	Lipid profile
Brain	Reduce Alzheimers	Cognitive tests
	Improve cognitive function	Cognitive tests
	Reduce vasomotor symptoms	Gonadotrophin levels
	Maintain libido	None
Uterus/ovaries	No polyps, cysts,	Uterine ultrasound
	No endometrial hyperplasia or cancer	Endometrial sampling
Pelvic floor	No Incontinence/uterine prolapse	None
Clotting	No increase in thromboembolism	Clotting factors
Eye	No increase in cataract or retinopathy	Ophthalmic examination
Other cancers	Not genotoxic	Structural clearance

agents more active for breast cancer risk reduction than tamoxifen. The third generation non-steroidal aromatase inhibitors, such as anastrozole (Arimidex) and letrozole are very powerful inhibitors of oestrogen synthesis [58] and have no innate oestrogenic activity. These agents have been shown to be more active than tamoxifen in postmenopausal women for the treatment of advanced breast cancer [59–62] and anastrozole has been shown to be more active than tamoxifen at reducing the risk of relapse and the risk of developing new breast cancers in patients with operable breast cancer [63].

One problem with using these agents for prevention is that they are only effective in postmenopausal women. Another worrying issue is the toxicity, which has not been well established at this time, because of the small numbers of patients who have had a significant duration of treatment. The levels of oestrogen achieved by these agents are very low [58] and not previously achieved *in vivo*. The long-term effects of these very low levels of oestrogen on bone, the pelvic floor, the cardiovascular system, lipids or the brain are not known. The early report from the Arimidex, Tamoxifen Alone or in Combination (ATAC) trial indicate an increased fracture rate after only 30 months of treatment with anastrazole [63]. Some tissue specificity with aromatase inhibitors may be possible, because of the variation in the tissue-specific promoters of aromatase in different tissues allowing some normal tissue sparing of aromatase inhibitor activity [64]. Alternatively, it is possible that the third generation steroidal aromatase inhibitors such as exemestane may have less anti-oestrogenic impact than the non-steroidal aromatase inhibitors on normal tissues [59]. Clearly, the many potential advantages of using a SERM (Table 3) will not occur with aromatase inhibitors.

If raloxifene is better than tamoxifen in the P-2 trial a back-to-back comparison of raloxifene with anastrazole to directly compare efficacy and toxicity will be essential. Generally it would seem more likely that the development of a SERM with better risk reduction of breast cancer than tamoxifen, together with other additional benefits and with no toxicity on the uterus, is likely to be a more promising agent for breast cancer prevention than the use of the powerful aromatase inhibitors which are likely to have detrimental anti-oestrogenic side-effects. The complexity of using an 'add on' prevention of these anti oestrogenic complications by use of such agents as the bisphosphonates to prevent fractures is unattractive.

5. Conclusions

At this time, the NSABP P-1, the IBIS and the MORE trial have clearly shown that it is possible to achieve a risk reduction for breast cancer by using anti-oestrogenic agents. Whether this will provide long-term, overall clinical benefit, and in which women at risk, remains uncertain.

In order to maximise the chance of achieving clinical benefit by endocrine prevention, the strategy for the next generation of trials should be to develop an algorithm of risk factors which target women who have a high risk of developing endocrine-dependent cancers. These high risk, but likely to be endocrine-responsive, women should be recruited to the trials to test the next generation of agents which may be more active at preventing breast cancer, be less toxic, and also have other benefits.

To do this, the statistical design of these trials will be critical. It is essential that multiple outcomes can be evaluated so that real long-term clinical benefit can be identified. The design of the huge P-1 trial, with such statistical power that only the early risk reduction of breast cancer could be detected before the trial was stopped, will not do this. It is likely that for prevention trials multiple benefits such as prevention of breast cancer, osteoporotic fractures and heart disease will be required, and this will need to be carefully balanced against as little toxicity as possible, in order to achieve an overall clinical benefit. Surrogate markers of some of these outcomes will be required (Table 3).

For these reasons, the trial design for prevention trials needs to be reviewed so that the early risk reduction of breast cancer is not the single outcome. The trials should be allowed to mature so that an agreed spectrum of multiple outcomes (or agreed surrogate markers of these outcomes) can be evaluated and the possibility of an overall predetermined spectrum of events in many tissues should be agreed to produce an event rate which is the 'gold standard' for prevention trials. Only in this way will we be able to answer the fundamental question as to whether prevention of breast cancer is better than cure.

With the new agent for reducing the oestrogenic drive to breast cancer cells, and by selection of the women at risk who are most likely to gain benefit, it is likely that we could prevent the development of most breast cancers.

References

1. Lacassagne A. Hormonal pathogenesis of adenocarcinoma of the breast. *Am J Cancer* 1936, **27**, 217–225.
2. Clemons M, Goss P. Estrogen and the risk of breast cancer. *N Engl J Med* 2001, **344**, 276–285.
3. Cauley J, Lucas F, Kuller L. Elevated serum estradiol and testosterone concentrations are associated with a high risk for breast cancer: study of Osteoporotic Fractures Research Group. *Ann Intern Med* 1999, **130**, 270–277.
4. Powles T, Hardy J, Ashley S, *et al*. A pilot trial to evaluate the acute toxicity and feasibility of tamoxifen for prevention of breast cancer. *Br J Cancer* 1989, **60**, 126–131.
5. Cuzick J, Powles T, Veronesi U, *et al*. Overview of the main outcomes in the breast cancer prevention trials. *Lancet* (In press).

6. Gail M, Brintom L, Byar D, Corle D, Green S, Schairer D. Projecting individualised probabilities of developing breast cancer for white females who are examined annually. *J Natl Cancer Inst* 1989, **81**, 1879–1886.
7. Fisher B, Costantino J, Wickerham D, *et al*. Tamoxifen for prevention of breast cancer: report of the National Surgical Adjuvant Breast and Bowel Project P-1 Study. *JNCI* 1998, **90**, 1371–1388.
8. Gail M, Costantino J, Bryant J, *et al*. Weighing the risks and benefits of tamoxifen treatment for preventing breast cancer. *JNCI* 1999, **91**, 1829–1846.
9. Veronesi U, Maisonneuve P, Costa A, *et al*. Prevention of breast cancer with tamoxifen: preliminary findings from the Italian randomised trial among hysterectomised women. *Lancet* 1998, **352**, 93–97.
10. Veronesi U, Maisonneuve P, Sacchini V, Rotmensz N, Boyle P. Tamoxifen for breast cancer among hysterectomised women. *Lancet* 2002, **359**, 1122–1124.
11. Powles T, Eeles R, Ashley S, *et al*. Interim analysis of the incidence of breast cancer in the Royal Marsden Hospital tamoxifen randomised chemoprevention trial. *Lancet* 1998, **352**, 98–101.
12. IBIS investigators. First results from the International Breast Cancer Intervention Study (IBIS-1): a randomised prevention trial. *Lancet* 2002, **360**, 817-824.
13. Delmas P, Bjarnason N, Mitlak B, *et al*. Effects of raloxifene on bone mineral density, serum cholesterol concentrations, and uterine endometrium in postmenopausal women. *N Engl J Med* 1997, **337**, 1641–1647.
14. Cummings S, Eckert S, Krueger K, *et al*. The effect of raloxifene on risk of breast cancer in postmenopausal women. Results from the MORE randomized trial. *JAMA* 1999, **281**, 2189–2197.
15. Cauley J, Norton L, Lippman M, *et al*. Continued breast cancer risk reduction in postmenopausal women treated with raloxifene: 4 year results from the MORE trial: multiple outcomes of raloxifene evaluation. *Breast Cancer Research and Treatment* 2001, **65**, 125–134.
16. Ettinger B, Black D, Mitlak B, *et al*. Reduction of vertebral fracture risk in postmenopausal women with osteoporosis treated with raloxifene: results from a 3-year randomized clinical trial. Multiple outcomes of raloxifene evaluation (MORE) investigators. *JAMA* 1999, **282**, 637–645.
17. Walsh B, Kuller L, Wild R, *et al*. Effects of raloxifene on serum lipids and coagulation factors in healthy postmenopausal women. *JAMA* 1998, **279**, 1445–1451.
18. Barrett-Connor E, Grady D, Sashegyi A. Raloxifene and cardiovascular events in osteoporotic postmenopausal women: four-year results from the MORE (multiple outcomes of raloxifene evaluation) randomized trial. *JAMA* 2002, **287**, 847–857.
19. Early Breast Cancer Triallists' Collaborative Group E. Tamoxifen for early breast cancer: an overview of the randomised trials. *Lancet* 1998, **351**, 1451–1467.
20. Ries L, Kosary C, Hankey B, Miller B, Clegg L, Edwards B. *SEER Cancer Statistics Review, 1973–1996*. Bethesda, MD, National Cancer Institute, 1999.
21. Rockhill B, Weinberg C, Newman B. Population attributable fraction estimation for established breast cancer risk factors: considering the issues of high prevalence and unmodifiability. *Am J Epidemiol* 1998, **147**, 826–833.
22. Madigan M, Ziegler R, Benichou J, Byrne C, Hoover R. Proportion of breast cancer cases in the United States explaiend by well-established risk factors. *J Natl Cancer Inst* 1995, **87**, 1681–1685.
23. Bruzzi P, Green S, Byar D, Brinton L, Schairer C. Estimating the population attributable risk for multiple risk factors using case-control data. *Am J Epidemiol* 1985, **122**, 904–914.
24. Slattery M, Kerber R. A comprehensive evaluation of family history and breast cancer risk: the Utah Population Database. *JAMA* 1993, **270**, 1563–1568.
25. Dupont W, Page D. Risk factors for breast cancer in women with proliferative breast disease. *N Engl J Med* 1985, **312**, 146–151.
26. Wrensch M, Petrakis N, Miike R. Breast Cancer risk in women with abnormal cytology in nipple aspirates of breast fluid. *J Natl Cancer Inst* 2001, **93**, 1791–1798.
27. Wrensch M, Petrakis N, King E. Breast cancer risk associated with abnormal cytology in nipple aspirates of breast fluid and prior history of breast biopsy. *Am J Epidemiol* 1993, **137**, 829–833.
28. Fabian C, Kimler B, Zalles C. Short-term breast cancer prediction by random periareolar fine-needle aspiration cytology and the Gail risk model. *J Natl Cancer Inst* 2000, **92**, 1217–1227.
29. Dooley W, Ljung B, Veronesi U. Ductal lavage for detection of cellular atypia in women at high risk for breast cancer. *J Natl Cancer Inst* 2001, **93**, 1624–1632.
30. Fabian C, Kimler B. Breast cancer chemoprevention: Current challenges and a look toward the future. *Clin Breast Cancer* 2002, **3**, 113–124.
31. Colditz G. Relationship between estrogen levels, use of hormone replacement therapy, and breast cancer. *J Natl Cancer Inst* 1998, **90**, 814–823.
32. Monninkhof E, van der Schouw Y, Peeters P. Early age at menopause and breast cancer: are leaner women more protected? A prospective anlaysis of the Dutch DOM cohort. *Breast Cancer Res Treat* 1999, **55**, 285–291.
33. Hall H, Coates R, Uhler R. Stage of breast cancer in relation to body mass index and bra cup size. *Int J Cancer* 1999, **82**, 23–27.
34. Ganry O, Peng J, Dubreuil A. Is there a reduced risk of breast cancer among women with hip fractures? *Eur J Epidemiol* 1999, **15**, 313–315.
35. Cauley J, Lucas F, Kuller L, *et al*. Bone mineral density and the risk of breast cancer in older women. *J Am Med Assoc* 1996, **276**, 1404–1408.
36. Hankinson S, Willett W, Manson J. Plasma sex steroid hormone levels and risk of breast cancer in postmenopausal women. *J Natl Cancer Inst* 1998, **90**, 1292–1299.
37. Yu H, Roha T. Role of the insulin-like growth factor family in cancer development and progression. *J Natl Cancer Inst* 2000, **92**, 1472–1489.
38. Hankinson S, Willett W, Colditz G. Circulating concentrations of insulin-like growth factor-I and risk of breast cancer. *Lancet* 1998, **351**, 1393–1396.
39. La Vecchia C, Negri E, Franceschi S. Body mass index and postmenopausl breast cancer: an age-specific analysis. *Br J Cancer* 1997, **75**, 441–444.
40. Cauley J, Gutai J, Kuller L. The epidemiology of serum sex hormones in postmenopausal women. *Am J Epidemiol* 1989, **129**, 1120–1131.
41. Colditz G, Hankinson S, Hunter D. The use of estrogens and progestins and the risk of breast cancer in postmenopausal women. *N Engl J Med* 1995, **332**, 1589–1593.
42. Powles T. Anti-oestrogenic prevention of breast cancer—the make or break point. *Nature Reviews Cancer* 2002, **2**, 787–794.
43. Ursin G, Pike M, Spicer D, Porrath S, Reitherman R. Can mammographic densities predict effects of tamoxifen on the breast? *J Natl Cancer Inst* 1996, **88**, 128–129.
44. Atkinson C, Warren R, Bingham S, Day N. Mammographic patterns as a predictive biomarker of breast cancer risk: effect of tamoxifen. *Cancer Epidemiol Biomarkers Prev* 1999, **8**, 863–866.
45. Boyd N, Dite G, Stone J, *et al*. Heritability of mammographic density, a risk factor for breast cancer. *New Eng J Med* 2002, **347**, 886–894.
46. Ursin G, Astrahan M, Salane M. The detection of changes in mammographic densities. *Cancer Epidemiol Biomarkers Prev* 1998, **7**, 43–47.
47. Andrykowski M, Curran S, Carpenter J. Rheumatoid symptoms following breast cancer treatment: a controlled comparison. *J Pain Symptom Manag* 1999, **18**, 85–94.

48. Lippman M, Krueger K, Eckert S, *et al*. Indicators of lifetime estrogen exposure: effect on breast cancer incidence and interaction with raloxifene therapy in the multiple outcomes of raloxifene evaluation study participants. *J Clin Oncol* 2001, **19**, 3111–3116.
49. Claus E, Risch N, Thompson W. Autosomal dominant inheritance of early-onset breast cancer: implications for risk prediction. *Cancer* 1994, **73**, 643–651.
50. Pharoah P, Antoniou A, Bobrow M, Zimmern R, Easton D, Ponder B. Polygenic susceptibility to breast cancer and implications for prevention. *Nat Gene* 2002, **31**, 33–36.
51. Haiman C, Hankinson S, Spiegelman D. The relationship between a polymorphism in CYP17 with plasma hormone levels and breast cancer. *Cancer Res* 1999, **59**, 1015–1020.
52. Osborne C, Vardou V-J, Hilsenbeck S, *et al*. The estrogen receptor coactivator AIB1 (SRC3) in combination with HER-2 is a prognostic and predictive marker in patients with breast cancer. Proceedings American Society of Clinical Oncology, Orlando. *J Clin Oncol* 2002, **21**, 33 a.
53. Wolf D, Jordan V. The estrogen receptor from a tamoxifen stimulated MCF-7 tumor variant contains a point mutation in the ligand binging domain. *Breast Cancer Res Treat* 1994, **31**, 129–138.
54. Fuqua S. A hypersensitive estrogen receptor-alpha mutation in premalignant breast lesions. *Cancer Res* 2000, **60**, 4026–4029.
55. Zhang Q, Borg A, Wolf D, Oesterreich S, Fuqua S. An estrogen receptor mutant with strong hormone-independent activity from a metastatic breast cancer. *Cancer Res* 1997, **57**, 1244–1249.
56. Martin M. A role for AKT in mediating the estrogenic functions of epidermal growth factor and insulin-like growth factor I. *Endocrinology* 2000, **141**, 4503–4511.
57. Shang Y, Brown M. Molecular determinants for the tissue specificity of SERMs. *Science* 2002, **295**, 2465–2468.
58. Geisler J, Haynes B, Anker G, Dowsett M, Lonning P. Influence of letrozole and anastrozole on total body aromatization and plasma estrogen levels in postmenopausal breast cancer patients evaluated in a randomized, cross-over study. *J Clin Oncol* 2002, **20**, 751–757.
59. Goss P, Strasser K. Aromatase inhibitors in the treatment and prevention of breast cancer. *J Clin Oncol* 2001, **19**, 881–894.
60. Bonneterre J, Thurlimann B, Robertson J. Anastrozole versus tamoxifen as first-line therapy for advanced breast cancer in 668 postmenopausal women: results of the tamoxifen or arimidex randomized group efficacy and tolerability study. *J Clin Oncol* 2000, **18**, 3748–3757.
61. Nabholtz J, Buzdar A, Pollak M. Anastrozole is superior to tamoxifen as first-line therapy for advanced breast cancer in postmenopausal women: results of a North American multicenter randomized trial. *J Clin Oncol* 2000, **18**, 3758–3767.
62. Mouridsen H, Gershanovich M, Sun Y. Superior efficacy of letrozole versus tamoxifen as first-line therapy for postmenopausal women with advanced breast cancer: results of a phase III study of the International Letrozole Breast Cancer Group. *J Clin Oncol* 2001, **19**, 2596–2606.
63. ATAC Trialists' Group. Anastrozole alone or in combination with tamoxifen versus tamoxifen alone for adjuvant treatment of postmenopausal women with early breast cancer: first results of the ATAC randomised trial. *Lancet* 2002, **359**, 2131-2139.
64. Simpson E, Dowsett M. Aromatase and its inhibitors: significance for breast cancer therapy. *Recent Prog Horm Res* 2002, **57**, 317–338.