

# Commentary

## Endogenous hormones and the aetiology of breast cancer

Timothy J Key and Pia K Verkasalo

University of Oxford, Oxford, UK

Received: 16 July 1999  
 Accepted: 22 July 1999  
 Published: 23 August 1999  
 © Current Science Ltd

After several decades of epidemiological and laboratory research, how well do we understand the role of endogenous hormones in the aetiology of breast cancer? Early studies showed that risk varies with several hormonal events: risk is increased by early menarche and late menopause, and decreased by giving birth at a young age and by high parity. The protective effect of pregnancy is probably due to hormonally induced differentiation of breast epithelial cells, causing a reduction in the number of susceptible cells. The effects of age at menarche and menopause indicate that the duration of exposure to cyclic ovarian function is an important determinant of breast cancer risk, but it has proved difficult to establish whether oestradiol and/or progesterone is responsible, or to show whether variations in hormone levels between women have any important effect on risk.

Most of the early epidemiological studies of endogenous hormones and breast cancer sought to test the simple hypothesis that high oestrogen levels would increase risk. The first studies used a case-control design; hormone levels in women with breast cancer were compared with those in women without breast cancer. This design is relatively fast and cheap, but interpretation is limited by the possibility that any differences observed could be caused by the tumour or by the treatment, rather than being markers of a long-standing hormonal environment that has led to the development of the disease. To avoid this problem it is necessary to conduct prospective studies in which samples are collected from many thousands of healthy women who are followed for the occurrence of breast cancer; hormone levels are then measured in the stored samples from women who develop breast cancer and compared with measurements from women who remain free of breast cancer. These prospective studies

### Important note about how to cite this article

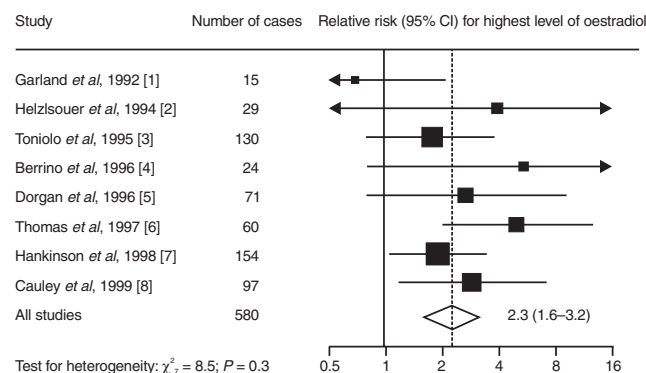
This article is also available online in the *Breast Cancer Research* website. To avoid confusion, please ensure that only the online version of the article is cited in any reference, as follows:

Key TJ, Verkasalo PK: Endogenous hormones and the aetiology of breast cancer [commentary]. <http://breast-cancer-research.com/vol1no1/23aug99/editorial/2>

are slow and expensive to conduct, but during the past few years several have matured and important results have been published on endogenous hormones and breast cancer risk in postmenopausal women.

Figure 1 shows the current summary of these prospective data for oestradiol in postmenopausal women [1–8]. For each study, we have plotted the most adjusted estimate of relative risk of breast cancer for women with high levels of oestradiol in that study compared with women with low oestradiol, together with a weighted average of the results

Figure 1



Prospective studies of oestradiol and breast cancer risk in postmenopausal women. For each study, the relative risk plotted is for women with the highest oestradiol concentration (tertile or quartile) compared with women with the lowest concentration. The 'all studies' estimate was calculated by weighting the individual relative risks by the inverse of their estimated variances, on a logarithmic scale. The relative risk for the study by Garland *et al* 1992 [1] was calculated by us from published data.

from all the studies. Seven out of eight studies found increased relative risks in association with high levels of oestradiol, and the pooled relative risk is 2.3 (95% confidence interval 1.6–3.2). Thus, the data strongly support the hypothesis that high levels of oestradiol in postmenopausal women increase breast cancer risk. The studies that have measured bioavailable oestradiol and/or oestrone as well as total oestradiol have resulted in broadly similar risk estimates for these different measurements (not shown). This would be expected because bioavailable oestradiol is very strongly correlated with total oestradiol in postmenopausal women. Likewise, serum oestrone is the main precursor of oestradiol in postmenopausal women, and thus the two hormones are strongly correlated with each other.

For premenopausal women, data on oestradiol and breast cancer risk have been published from only four prospective studies [2,9–11] with a total of 179 cases of breast cancer. The numbers for progesterone are even smaller; data have been published from three studies [2,9,11] with a total of only 99 cases. None of these studies found statistically significant associations between oestradiol or progesterone and breast cancer risk. However, the design and interpretation of these studies is complicated by the large physiological variations in serum concentrations of both oestradiol and progesterone during the menstrual cycle. In a study of the repeatability of serum hormone levels, Muti *et al* [12] took two samples 1 year apart from 60 premenopausal women, attempting to collect the two samples under identical conditions on the same day of the luteal phase of the menstrual cycle; correlations between the two samples were high for androgens and peptide hormones, but very low for oestradiol. This suggests that more than one sample is needed to estimate a woman's oestradiol level in the luteal phase of the menstrual cycle reliably. Furthermore, oestradiol levels during the mid-cycle peak [11], or cumulative exposure to oestradiol over the entire cycle, may be of importance in relation to breast cancer risk. Future studies should ideally collect several samples from each woman and from each of at least two menstrual cycles.

In addition to the observational epidemiological studies of endogenous hormones and breast cancer risk, recently published trials of the preventive effects of selective oestrogen receptor modulators have provided important further evidence for the role of oestradiol in the aetiology of breast cancer. The first trials [13] have suggested that both tamoxifen and raloxifene may reduce breast cancer rates. Much more information is needed to substantiate these findings and to study the effects of these drugs on breast cancer mortality, but these early results do imply that blocking the action of oestrogens can reduce the incidence of breast cancer.

What about other hormones such as androgens, insulin-like growth factor-I and prolactin? Unlike oestradiol,

serum concentrations of these hormones do not change dramatically at menopause, and the strong protective effect of early menopause is likely to be due to changes in oestradiol (and possibly progesterone) rather than changes in androgens or peptide hormones. The recent prospective studies have reported that, like oestradiol, serum concentrations of testosterone, androstenedione and dehydroepiandrosterone are positively associated with breast cancer risk in postmenopausal women [4–8,14–16]. These sex hormones are all products of the same metabolic pathway and their serum concentrations are positively correlated with each other. Multivariate analyses of the existing studies have not produced a consistent conclusion as to which steroid hormone is most closely related to risk. More data should help, but the results will require cautious interpretation because the measurements for different hormones are not equally informative. For example, in postmenopausal women the reproducibility of measurements has been found to be higher for testosterone than for oestradiol [17]; this could result in testosterone being apparently more strongly associated with risk than oestradiol simply because the measurement for testosterone is more informative.

As well as the steroid hormones, recent prospective studies [18,19] have suggested that the peptide hormones insulin-like growth factor-I (in premenopausal women) and prolactin (in postmenopausal women) may also be related to breast cancer risk. The metabolism of these peptides has links to the metabolism of sex hormones, but the serum concentrations of these hormones are not strongly correlated with oestrogens and their effects on breast cancer could well be independent.

The most likely mechanism by which oestradiol and perhaps other hormones affect breast cancer risk is by controlling the mitotic rate of the breast epithelial cells. High mitotic rates can increase cancer risk by increasing the chance of mutations occurring and of being replicated before they are repaired, and can also increase the growth of early tumours [20]. Studies of the mitotic rate in the breast epithelial cells of premenopausal women [21] have shown that some mitoses occur throughout the menstrual cycle, but that there is a peak in activity during the mid-to-late luteal phase when serum concentrations of both oestradiol and progesterone are high. Experiments using human breast tissue grafted into mice [22,23] show that oestradiol is a mitogen. No mitogenic effect was seen in human breast tissue in mice after administration of progesterone [23], but this does not exclude the possibility of a mitogenic effect under physiological conditions in women [24]. These data imply that oestradiol alone may increase breast cancer risk through the stimulation of mitosis, but it remains possible that, in premenopausal women, progesterone may augment this effect of oestradiol.

In addition to stimulating mitosis, it has been suggested that oestradiol could also increase breast cancer risk because some of its metabolites such as the catechol oestrogen 4-hydroxyoestradiol might cause direct DNA damage through the formation of free radicals [25,26]. Much of the experimental evidence for this hypothesis is derived from studies on kidney cancer in hamsters, which may well behave differently from human breast cancer. Early reports that women with genetically determined reduced inactivation of catechol oestrogens are at increased risk of breast cancer have not been confirmed [27]. More data are needed before this hypothesis can be evaluated.

Mutations in high-risk genes such as *BRCA1* increase the risk for several hormone-related cancers. The mechanisms for these effects may include interactions with oestrogen as well as direct effects on cell proliferation and apoptosis; for example, wild-type *BRCA1* may suppress oestrogen-dependent transcription [28]. Inherited mutations in high-risk genes are involved in only a small proportion of breast cancers, but a substantial component of breast cancer risk may be determined by the sum of multiple smaller genetic effects. Recent twin studies on breast cancer [29,30], which account for all modes of genetic transmission, indicate that hereditary genetic factors may contribute to as much one-third of the variation in breast cancer incidence within a population.

*CYP17* codes for an enzyme that catalyzes two steps in the synthesis of oestradiol, and work on *CYP17* is among the first of a new generation of studies, looking at whether common genetic polymorphisms may affect breast cancer risk by affecting hormone metabolism. Two studies [31,32] have looked at serum oestradiol concentrations in relation to a single base pair polymorphism in the promoter region of *CYP17*, which might affect gene transcription; both studies reported slightly higher oestradiol concentrations in women with the putatively more active polymorphism. However, studies of this polymorphism in *CYP17* have not demonstrated a significant elevation in breast cancer risk [32–36]. Several other mechanisms are also possible: polymorphisms in other genes encoding enzymes in the steroid synthesis and metabolism pathway (*CYP11A1*, *3 $\beta$ -HSD*, *17 $\beta$ -HSD*, *CYP19*) may affect steroid levels; polymorphisms in the genes encoding peptides (prolactin, insulin-like growth factor-I) may influence the serum concentration or the intrinsic activity of the hormone; polymorphisms in hormone receptor genes [oestrogen receptor (ER)- $\alpha$ , ER- $\beta$ ] may increase the biological response to a given hormone level; and polymorphisms in genes encoding hormone-binding proteins [sex hormone binding globulin (SHBG), insulin-like growth factor binding protein 3] could affect risk by altering hormone bioavailability. Testing these hypotheses in large epidemiological studies is now technically straightforward and significant polymorphisms are likely to be identified in the near future.

What about environmental determinants of endogenous hormone levels, such as diet and exercise? Despite burgeoning interest in the effects of diet on both hormones and breast cancer, the only established link is with obesity. In postmenopausal women, obesity causes a substantial increase in bioavailable oestradiol due to increased production from androstenedione and oestrone and a decrease in SHBG [37], and obese postmenopausal women have about a two-fold increased risk for breast cancer. More data from prospective studies are needed to show whether the effect of obesity on breast cancer risk can be explained by its effects on oestradiol, but it is already clear that breast cancer rates could be reduced by reducing the prevalence of obesity in postmenopausal women [38]. The possible protective effect of exercise is less firmly established, but moderate exercise in young women might perhaps reduce breast cancer risk by reducing exposure to both oestradiol and progesterone [39].

Hypotheses by which nutrition could affect oestradiol metabolism abound. For example, fat might increase synthesis, fibre might increase excretion, phyto-oestrogens might block the stimulation of receptors, and indoles might accelerate catabolism. None of these hypotheses has yet been strongly supported, and perhaps the most promising current hypothesis is that alcohol may increase breast cancer risk by increasing endogenous oestradiol levels [40,41]. Establishing the effects of diet on hormone metabolism is important, because studies of migrants show that increases in breast cancer risk can be observed as soon as 10 years after migrating from East Asia to the USA [42]. Japanese women living in rural Japan have much lower serum oestrogen levels than white Americans, but Japanese-Americans now have serum oestrogen levels as high as white Americans [43]. The increases in oestrogen levels and breast cancer risk may both be determined by the ‘westernization’ of diet.

The evidence that oestradiol is an important determinant of breast cancer risk is now strong. Research during the next few years may be expected to confirm the importance of oestradiol, to clarify the roles of other hormones, and to establish the environmental and genetic determinants of endogenous hormone levels.

## References

1. Garland CF, Friedlander NJ, Barrett-Connor E, Khaw K-T: **Sex hormones and postmenopausal breast cancer: a prospective study in an adult community.** *Am J Epidemiol* 1992, **135**:1220–1230.
2. Helzlsouer KJ, Alberg AJ, Bush TL, Longcope C, Gordon GB, Comstock GW: **A prospective study of endogenous hormones and breast cancer.** *Cancer Detect Prev* 1994, **18**:79–85.
3. Toniolo PG, Levitz M, Zeleniuch-Jacquotte A, et al: **A prospective study of endogenous estrogens and breast cancer in postmenopausal women.** *J Natl Cancer Inst* 1995, **87**:190–197.
4. Berrino F, Muti P, Micheli A, et al: **Serum sex hormone levels after menopause and subsequent breast cancer.** *J Natl Cancer Inst* 1996, **88**:291–296.

5. Dorgan JF, Longcope C, Stephenson HE Jr, *et al*: **Relation of prediagnostic serum estrogen and androgen levels to breast cancer risk.** *Cancer Epidemiol, Biomarkers & Prev* 1996, **5**:533–539.
6. Thomas HV, Key TJ, Allen DS, *et al*: **A prospective study of endogenous serum hormone concentrations and breast cancer risk in postmenopausal women on the island of Guernsey.** *Br J Cancer* 1997, **76**:401–405.
7. Hankinson SE, Willett WC, Manson JE, *et al*: **Plasma sex steroid hormone levels and risk of breast cancer in postmenopausal women.** *J Natl Cancer Inst* 1998, **90**:1292–1299.
8. Cauley JA, Lucas FL, Kuller LH, *et al*: **Elevated serum estradiol and testosterone concentrations are associated with a high risk for breast cancer.** *Ann Intern Med* 1999, **130**:270–277.
9. Wysowski DK, Comstock GW, Helsing KJ, Lau HL: **Sex hormone levels in serum in relation to the development of breast cancer.** *Am J Epidemiol* 1987, **125**:791–799.
10. Rosenberg CR, Pasternack BS, Shore RE, Koenig KL, Toniolo PG: **Premenopausal estradiol levels and the risk of breast cancer: a new method of controlling for day of the menstrual cycle.** *Am J Epidemiol* 1994, **140**:518–525.
11. Thomas HV, Key TJ, Allen DS, *et al*: **A prospective study of endogenous serum hormone concentrations and breast cancer risk in premenopausal women on the island of Guernsey.** *Br J Cancer* 1997, **75**:1075–1079.
12. Muti P, Trevisan M, Micheli A, *et al*: **Reliability of serum hormones in premenopausal and postmenopausal women over a one-year period.** *Cancer Epidemiol Biomarkers Prev* 1996, **5**:917–922.
13. Cummings SR, Eckert S, Krueger KA, *et al*: **The effect of raloxifene on risk of breast cancer in postmenopausal women.** *J Am Med Ass* 1999, **281**:2189–2197.
14. Dorgan JF, Stanczyk FZ, Longcope C, *et al*: **Relationship of serum dehydroepiandrosterone [DHEA], DHEA sulfate, and 5-androstene-3 $\beta$ ,17 $\beta$ -diol to risk of breast cancer in postmenopausal women.** *Cancer Epidemiol Biomarkers Prev* 1997, **6**:177–181.
15. Gordon GB, Bush TL, Helzlsouer KJ, Miller SR, Comstock GW: **Relationship of serum levels of dehydroepiandrosterone and dehydroepiandrosterone sulfate to the risk of developing postmenopausal breast cancer.** *Cancer Res* 1990, **50**:3859–3862.
16. Zeleniuch-Jacquotte A, Bruning PF, Bonfrer JMG, *et al*: **Relation of serum levels of testosterone and dehydroepiandrosterone sulfate to risk of breast cancer in postmenopausal women.** *Am J Epidemiol* 1997, **145**:1030–1038.
17. Hankinson SE, Manson JE, Spiegelman D, *et al*: **Reproducibility of serum hormone levels in postmenopausal women over a 2–3-year period.** *Cancer Epidemiol Biomarkers Prev* 1995, **4**:649–654.
18. Hankinson SE, Willett WC, Colditz GA, *et al*: **Circulating concentrations of insulin-like growth factor-I and risk of breast cancer.** *Lancet* 1998, **351**:1393–1396.
19. Hankinson SE, Willett WC, Michaud DS: **Plasma prolactin levels and subsequent risk of breast cancer in postmenopausal women.** *J Natl Cancer Inst* 1999, **91**:629–634.
20. Preston-Martin S, Pike MC, Ross RK, Jones PA, Henderson BE: **Increased cell division as a cause of human cancer.** *Cancer Res* 1990, **50**:7415–7421.
21. Anderson TJ, Ferguson DJP, Raab GM: **Cell turnover in the 'resting' human breast: influence of parity, contraceptive pill, age and laterality.** *Br J Cancer* 1982, **46**:376–382.
22. McManus MJ, Welsch CW: **The effect of estrogen, progesterone, thyroxine, and human placental lactogen on DNA synthesis of human breast ductal epithelium maintained in athymic nude mice.** *Cancer* 1984, **54**:1920–1927.
23. Laidlaw IJ, Clarke RB, Howell A, *et al*: **The proliferation of normal human breast tissue implanted into athymic nude mice is stimulated by estrogen but not progesterone.** *Endocrinology* 1995, **136**:164–171.
24. Pike MC, Ursin G, Spicer DV: **Experiments on proliferation of normal human breast tissue in nude mice do not show that progesterone does not stimulate breast cells [Letter].** *Endocrinology* 1996, **137**:1505–1506.
25. Yager JD, Liehr JG: **Molecular mechanisms of estrogen carcinogenesis.** *Ann Rev Pharmacol Toxicol* 1996, **36**:203–232.
26. Cavalieri EL, Stack DE, Devanesan PD, *et al*: **Molecular origin of cancer: catechol estrogen-3, 4-quinones as endogenous tumor initiators.** *Proc Natl Acad Sci USA* 1997, **94**:10937–10942.
27. Millikan RC, Pittman GS, Tse C-KJ, *et al*: **Catechol-O-methyltransferase and breast cancer risk.** *Carcinogenesis* 1998, **19**:1943–1947.
28. Fan S, Wang J-A, Yuan R, *et al*: **BRCA1 inhibition of estrogen receptor signaling in transfected cells.** *Science* 1999, **284**:1354–1356.
29. Ahlbom A, Lichtenstein P, Malmström H, *et al*: **Cancer in twins: genetic and nongenetic familial risk factors.** *J Natl Cancer Inst* 1997, **89**:287–293.
30. Verkasalo PK, Kaprio J, Koskenvuo M, Pukkala E: **Genetic predisposition, environment and cancer incidence: a nationwide twin study in Finland, 1976–95.** *Int J Cancer* 1999, in press.
31. Feigelson HS, Shames LS, Pike MC, Coetzee GA, Stanczyk FZ, Henderson BE: **Cytochrome P450c17 $\alpha$  gene (CYP17) polymorphism is associated with serum estrogen and progesterone concentrations.** *Cancer Res* 1998, **58**:585–587.
32. Haiman CA, Hankinson SE, Spiegelman D, *et al*: **The relationship between a polymorphism in CYP17 with plasma hormone levels and breast cancer.** *Cancer Res* 1999, **59**:1015–1020.
33. Feigelson HS, Coetzee GA, Kolonel LN, Ross RK, Henderson BE: **A polymorphism in the CYP17 gene increases the risk of breast cancer.** *Cancer Res* 1997, **57**:1063–1065.
34. Dunning AM, Healey CS, Pharoah PDP: **No association between a polymorphism in the steroid metabolism gene CYP17 and risk of breast cancer.** *Br J Cancer* 1998, **77**:2045–2047.
35. Helzlsouer KJ, Huang H-Y, Strickland PT: **Association between CYP17 polymorphisms and the development of breast cancer.** *Cancer Epidemiol, Biomarkers Prev* 1998, **7**:945–949.
36. Weston A, Pan C-f, Bleiweiss IJ, *et al*: **CYP17 genotype and breast cancer risk.** *Cancer Epidemiol Biomarkers Prev* 1998, **7**:941–944.
37. Siiteri PK, MacDonald PC: **Role of extraglandular estrogen in human endocrinology.** In: *Handbook of physiology*. Edited by Geiger SR, Astwood EB, Greep RO. Washington DC: American Physiological Society, 1973:615–629.
38. La Vecchia C, Negri E, Franceschi S, *et al*: **Body mass index and postmenopausal breast cancer: an age-specific analysis.** *Br J Cancer* 1997, **75**:441–444.
39. Bernstein L, Henderson BE, Hanisch R, Sullivan-Halley J, Ross RK: **Physical exercise and reduced risk of breast cancer in young women.** *J Natl Cancer Inst* 1994, **86**:1403–1408.
40. Smith-Warner SA, Spiegelman D, Yaun S-S, *et al*: **Alcohol and breast cancer in women. A pooled analysis of cohort studies.** *JAMA* 1998, **279**:535–540.
41. Muti P, Trevisan M, Micheli A, *et al*: **Alcohol consumption and total estradiol in premenopausal women.** *Cancer Epidemiol Biomarkers Prev* 1998, **7**:189–193.
42. Ziegler RG, Hoover RN, Pike MC, *et al*: **Migration patterns and breast cancer risk in Asian-American women.** *J Natl Cancer Inst* 1993, **85**:1819–1827.
43. Probst-Hensch NM, Pike MC, McKean-Cowdin R, Stanczyk FZ, Kolonel LN, Henderson BE: **Ethnic differences in postmenopausal plasma estrogen levels high estrone levels in Japanese-American women despite low weight.** *Br J Cancer* 1999, in press.

**Authors' address:** Imperial Cancer Research Fund, Cancer Epidemiology Unit, University of Oxford, Oxford, UK

**Correspondence:** Timothy J Key, Imperial Cancer Research Fund, Cancer Epidemiology Unit, University of Oxford, Gibson Building, Radcliffe Infirmary, Oxford OX2 6HE, UK. Tel: +44 (0)1865 311933; fax: +44 (0)1865 310545; e-mail: key@icrf.icnet.uk