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9

# Environmental oestrogens, cosmetics and breast cancer

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The established role of oestrogen in the development and progression of breast cancer raises questions concerning a potential contribution from the many chemicals in the environment which can enter the human breast and which have oestrogenic activity. A range of organochlorine pesticides and polychlorinated biphenyls possess oestrogen-mimicking properties and have been measured in human breast adipose tissue and in human milk. These enter the breast from varied environmental contamination of food, water and air, and due to their lipophilic properties can accumulate in breast fat. However, it is emerging that the breast is also exposed to a range of oestrogenic chemicals applied as cosmetics to the underarm and breast area. These cosmetics are left on the skin in the appropriate area, allowing a more direct dermal absorption route for breast exposure to oestrogenic chemicals and allowing absorbed chemicals to escape systemic metabolism. This review considers evidence in support of a functional role for the combined interactions of cosmetic chemicals with environmental oestrogens, pharmacological oestrogens, phyto-oestrogens and physiological oestrogens in the rising incidence of breast cancer.

**Key words:** oestrogen; environmental oestrogen; cosmetics; breast cancer; xeno-oestrogen; DDT; PCB; parabens; aluminium; cyclosiloxane; triclosan; uv screen; phyto-oestrogen; breast cyst; endocrine therapy.

## THE CASE FOR A LINK BETWEEN ENVIRONMENTAL OESTROGENS AND BREAST CANCER

The breast is an endocrine-sensitive organ. Development of the breast from puberty through cycles of pregnancy, lactation and involution is controlled by an intricate web of hormonal controls.[1] The functional milk-producing cells are found lining the system of ducts and alveoli, but it is the balance of hormonal controls involving cell–cell communication between these epithelial cells and surrounding

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stromal cells, together with interactions through the extracellular matrix, which provides the overall balance for growth and differentiation in this organ.[1] In an environment of exposure to a variety of endocrine-disrupting chemicals, it should perhaps, therefore, not be such a mystery that disorders of the breast have become so widespread and that cancer in this organ has become the major cancer of women in the Western world.[2]

Although breast cancer may not be a new disease, the incidence of female breast cancer has risen worldwide in unprecedented terms in recent decades.[2,3] In England and Wales alone, the incidence of female breast cancer has increased from 74.4 per 100 000 population (European age-standardized rate, EASR) (21 446 new cases recorded) in 1979–113.8 per 100 000 population (EASR) (35 903 new cases recorded) in 2000.[4] Although being female is a major risk factor, breast cancer does also occur in men at around 1/100th of the frequency,[5] and the incidence of male breast cancer is also increasing in England and Wales.[6]

Epidemiological studies show that 90% of breast cancers are environmental in origin,[2] but the main underlying environmental causes remain to be identified. Although loss of function of the BRCA1/BRCA2 genes,[7] diet, smoking, alcohol and radiation[2,3] have been identified as risk factors, the main influence in the development of breast cancer remains lifetime exposure to oestrogen.[2,3] Increased exposure to endogenous oestrogens can result from early onset of menarche, late onset of menopause, nulliparity, late age of first pregnancy, practices of breastfeeding, and to exogenous oestrogens from personal choices to use the contraceptive pill or hormone replacement therapy.[2,3] In addition to epidemiological evidence linking breast cancer incidence to oestrogen exposure, the ability of oestrogen to drive the growth of breast tumours in vivo is well documented in clinical studies,[8] and the mechanisms of oestrogen action on the growth of breast cancer cells in animal models and in vitro is extensively described in experimental studies.[8,9] This involvement of oestrogen in the progression of breast cancer is the basis for the successful use of endocrine therapy as a treatment for breast cancer.[10]

In view of the unquestionable involvement of oestrogen in the development, progression and treatment of breast cancer, questions have to be asked concerning potential interactions of the many chemicals in the environment which can be stored in breast tissues and which can interfere with the physiological actions of oestrogen. Numerous reports have investigated whether environmental oestrogens entering the human body through food, water and air may be linked to the development of breast cancer.[11–15] I have suggested that an alternative route for exposure may be through the long-term, regular application to the underarm and breast area of a variety of cosmetic ingredients with oestrogenic activity.[6,16,17] These cosmetic chemicals are applied frequently and left on the skin of the breast area, allowing for not only continuous direct dermal exposure but also subsequent absorption and accumulation in underlying tissues.[17] The extent to which chemicals absorbed by this route escape metabolism remains unknown, but they would certainly escape the systemic metabolism to which orally derived chemicals would be subject.[17] The proven link between endocrine-disrupting chemicals and reproductive abnormalities in aquatic wildlife is attributed to the near-continuous exposure of these organisms to the chemicals in the water in which they live.[18] Continuous exposure to cosmetic chemicals provides for the human breast an analogous situation to the continuous exposure of aquatic life to chemicals in water.[17]

## BREAST EXPOSURE TO ENVIRONMENTAL OESTROGENS

In addition to the physiological steroidal oestrogens, many compounds have now been found to have oestrogenic activity such that the human breast can be exposed to environmental oestrogens from a variety of sources (Table 1). This can include pharmacological oestrogens, plant oestrogens (phyto-oestrogens) and man-made oestrogen-mimicking chemicals (xeno-oestrogens). Of these, some are ingested by human choice (e.g. in contraceptives or hormone replacement therapy), but by far the greater number may be taken inadvertently from the environment e.g. in the diet or from application of cosmetics to human skin.

A common feature of one mechanism of action of all these compounds is their saturable and reversible binding to cellular oestrogen receptors (ER$\alpha$ and/or ER$\beta$), and the kinetics of this binding determines their oestrogenic potency (see earlier chapters in this issue). Many of these compounds, when present in sufficient quantity, give responses equal to that of 17$\beta$-oestradiol, and they may then be described as having full agonist activity. Compounds unable to give responses equal to that of 17$\beta$-oestradiol are said to have partial agonist activity. Antagonist activity could be expected to occur when two oestrogens of different potencies compete for binding to receptor.

In a physiological context, it is important to note that the female is normally exposed to physiological oestrogens at the highest levels only between puberty and menopause, and that these levels cycle during the normal monthly menstrual cycle. However, exposure to environmental oestrogens is not regulated in this way. Exposure can occur prior to puberty or after menopause, and even between puberty and menopause the levels will not cycle in a physiological way (Figure 1). Thus whilst route of exposure, quantity, chemical combinations and rate of clearance (metabolism/excretion) will all

**Table 1.** Exposure of the human breast to environmental oestrogens.

| Compounds | Source |
|---|---|
| Physiological oestrogens | Physiological: levels higher between puberty and menopause; levels cycle during monthly menstrual cycle |
| Pharmaceutical oestrogens | Contraceptive pill, hormone replacement therapy |
| Phyto-oestrogens (e.g. genistein, daidzein) | Diet: edible plant material (e.g. soy beans) |
| Organochlorines (e.g. DDT, dieldrin, lindane) | Diet: pesticides and herbicides in environment accumulated in animal fat |
| Polychlorinated biphenyls (PCBs) | Diet: by-products of electrical industry in environment accumulated in animal fat |
| Polychlorinated dioxins | By-products of incineration: can be inhaled or taken in the diet in animal fat |
| Bisphenol A | Epoxy resins and polycarbonate plastics |
| Phthalates | Plasticizers |
| Alkyl phenols (e.g. nonylphenol) | Detergents |
| Metallo-oestrogens | Cadmium in cigarette smoke, metal ions in water or diet, aluminium in cosmetics |
| Parabens | Preservatives in cosmetics, food and pharmaceuticals |
| Chemical constituents of cosmetics | See Table 2 |

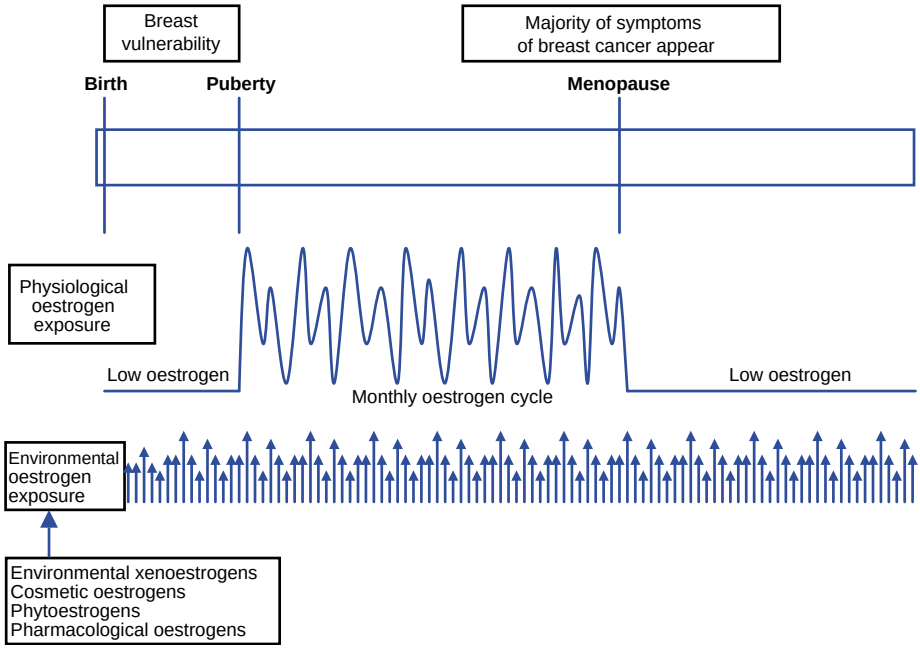


**Figure 1.** The potential importance of timing in the exposure of the human breast to environmental oestrogens. Levels of physiological oestrogens are highest only between puberty and menopause, and levels cycle during the monthly menstrual cycle. Exposure to environmental oestrogens can occur at any stage of life and is not regulated in a physiological way.

contribute to any resulting effects, timing could also be of relevance. There is evidence that the breast may be especially susceptible to carcinogenesis before puberty,[19] and studies of Japanese survivors of the atomic bomb in 1945 have shown that the highest risk of radiation-induced breast cancer was in girls who were prepubertal at the time of exposure.[20] On the other hand, since the majority of the symptoms of breast cancers arise in postmenopausal women,[2–4] exposure to chemicals after the menopause could also be significant. However, since the origin of breast cancer can occur many years before the symptoms become visible,[19] it is also possible that premenopausal exposure to chemicals could result in slow development of cancer detectable only in postmenopausal years. In the final analysis, since breast cancer is a multistage disease taking many years to develop, exposure to environmental oestrogens throughout all stages of life may play varied and additive roles.

## Pharmacological oestrogens

The link between breast cancer and use of the oral contraceptive pill has been extensively studied,[21] and publication of the million women study has documented an increase in breast cancer following the use of hormone replacement therapy.[22] This demonstrates that development of breast cancer can be influenced at all stages of life after puberty by voluntary exposure to exogenous oestrogens.

## Phyto-oestrogens

Phyto-oestrogens are compounds found naturally in plants and can be ingested by humans consuming certain foods (e.g. soy).[23] Consumption of phyto-oestrogens varies according to cultural cooking practices, being substantially higher in Japanese diets than in Western diets, and this has led to the assumption that soy diets of the East are the reason for lower incidence of breast cancer in those parts of the world.[23] This has in turn resulted in a widely held view that phyto-oestrogens are anti-oestrogenic to the breast. However, phyto-oestrogens do possess oestrogen agonist properties, and some have high potency—of the order of only tenfold lower than 17β-oestradiol itself.[23,24] It is now accepted that inhibitory actions of phyto-oestrogens at concentrations $> 10^{-5}$ M on cells in vitro—originally interpreted as anti-oestrogenicity—are not oestrogen-receptor-mediated, and, therefore, evidence for anti-oestrogenic actions of these compounds remains lacking in in vitro models.[24] This may explain the conflicting epidemiological reports and dietary trial results investigating the expected link between increased phyto-oestrogen consumption and reduced breast cancer.[23] Reports on the use of phyto-oestrogens to prevent breast cancer have been mixed,[23,25–27] and a recent prospective study of UK women showed that consumption of isoflavones increased breast cancer risk.[28] Animal models have also given conflicting results, showing that genistein is capable of reducing development of chemically induced mammary tumours in rats,[29–31] especially in younger rats,[32] whilst stimulating growth of subcutaneous MCF7-derived tumours in mice.[33,34] This could now be interpreted in the context of the diverse range of characterized mechanisms of action of phyto-oestrogens[23] without involvement of any anti-oestrogenic actions, such that the preventative actions of phyto-oestrogens on breast cancer could result from non-oestrogenic mechanisms such as antioxidant properties,[23] whilst the oestrogen agonist properties of phyto-oestrogens[23,24] would support growth of oestrogen-responsive breast cancers.

## Xeno-oestrogens

Xeno-oestrogens form a diverse group of man-made chemicals which have been released into the environment from agricultural spraying (herbicides, pesticides), as by-products of industrial processes and waste disposal (polychlorinated biphenyls (PCBs), dioxins), or as discharges from treatment systems (alkyl phenols). They are also found in household products in daily use, such as plastics (bisphenol A, phthalates) or flame retardants (polybrominated organics) and cosmetic products (e.g. parabens, cyclosiloxanes). They may be present in diet, and being lipophilic can pass up the food chain dissolved in fat and accumulate in humans at the top of the food chain.

Although exposure to individual xeno-oestrogens is only at low levels, many of these compounds are not readily metabolised, and due to their lipophilic properties they can accumulate over time in fatty tissues of the body. The breast has a high fat content, and the epithelial cells—which are the main target for cancer—are embedded in a fatty stroma. This provides an opportunity for concentration of lipophilic oestrogenic pollutants at the site of the target cells for breast cancer. These compounds accumulate with age, being lost at times of weight loss or during breast feeding, when they are passed on to the baby in the milk fat.[35] Epidemiological studies have shown that breast feeding can protect against breast cancer,[2,3] which may be explained by the low endogenous oestrogen exposure at this time, but a further contribution could result

from a concomitant reduction in breast burden of oestrogenic pollutants.[35] Since a majority of breast cancers become symptomatic after the menopause[2–4] when endogenous oestrogen levels are low, it is also possible that lifetime accumulation of xeno-oestrogens could play a significant role. The breast is known to be sensitive to exogenous oestrogens at this stage, because hormone replacement therapy can increase breast cancer development,[22] and this is also the stage of life when there would be the greatest body burden of xeno-oestrogen accumulation in the breast.

Use of animal models has demonstrated that xeno-oestrogens are capable of increasing breast cancer incidence in rodents. Organochlorines have been shown to increase chemically induced mammary tumours in rats.[36,37] Atrazine can cause mammary cancer in adult rats,[38] and nonylphenol (a metabolite of alkylphenols) can increase breast cancer incidence in mice.[39] The issue, then, is whether such compounds can have any effect in humans at the concentrations in which they are found in the human breast. Many xeno-oestrogens have now been detected in human body tissues, and in particular in breast adipose tissue[40–48] and human milk.[49–54] Results have been reported in a variety of different units, which makes inter-study comparisons difficult, but most measurements fall within the general range of 1–1000 ng/g fat. Following on from this approach, numerous studies have attempted to address the question of whether measured levels of individual xeno-oestrogens are higher in people with breast cancer than in those without, but the issue remains unresolved. Measurements using breast adipose tissue have suggested higher levels of organochlorine residues and PCBs in breast cancer patients than controls.[41,45,48] Studies using serum have produced varied results, some reporting an association,[55,56] but many larger studies finding no association.[57–60] However, all these studies have many inherent problems.

Firstly, breast cancer may start many years before symptoms appear,[19] and, therefore, the measurement of chemical load in those with breast cancer may not reflect exposure at the time of carcinogenesis.

Secondly, most of the large studies with statistical power have used serum for measurements, and this may not accurately reflect levels in the breast cells or breast fat in every individual.

Thirdly, complications may result from individual variations in susceptibility to chemicals such that absolute level of one specific chemical may not be the sole determinant.

Fourthly, humans are not exposed to single chemicals in isolation but to complex mixtures, and it may be the overall mixture of chemicals, which is important rather than specific single compounds. Similar oestrogenic load may be generated through exposure to different chemical mixtures. Some studies have attempted to address this issue,[48] but much more work on mixtures is needed.

Finally, it can be questioned whether it is even reasonable to expect breast cancer to be caused simply by the presence of a higher level of any specific chemical. Whilst proof of entry and retention of such chemicals in the breast must be a prerequisite, carcinogenesis may involve rather more stochastic mechanisms. It has long been accepted that cancer may arise not from one targeted cell but from within a generated field of susceptible cells.[61] Recent reports of genomic instability in the human breast[62,63] together with an oestrogenic environment generated from the presence of a mixture of xeno-oestrogens would provide a more general field favourable for both carcinogenesis and more long-term cancer growth and progression. In this context, it can be considered as significant that a wide range of xeno-oestrogens has been

detected in the human breast, but it may be naive to expect any overall measurable differences in specific chemicals between cancerous and non-cancerous tissues to either prove or refute a role for xeno-oestrogens in breast cancer development.

## OESTROGENIC COMPONENTS OF COSMETICS AND BREAST CANCER

In contrast to the diffuse and varied nature of environmental exposure to xeno-oestrogens through food, water and air, a mechanism of more direct breast exposure to xeno-oestrogens occurs through the application of oestrogenic chemicals of bodycare cosmetics to the underarm and breast area.[6,16,17] The strongest supporting evidence for a role of bodycare cosmetics in the rising incidence of breast cancer comes from published clinical observations dating back decades and showing a disproportionately large number of breast cancers in the upper outer quadrant (UOQ) of the breast, just the local area to which these chemicals are applied.[6,16] Early studies suggested that 31% of cancers occurred in the UOQ of the breast,[64] which may be in line with the accepted explanation that there is more target epithelial tissue in that region.[65] However, the proportion of breast cancers in the UOQ appears to rise with year of publication up to a level of 60.7% in 1994,[66] and within Great Britain is rising annually in a linear mode.[4]

Independent evidence for a role of underarm cosmetics in breast cancer comes from the recent report of increased levels of genomic instability found in the outer as compared with the inner quadrants of the breast.[62] Such a limited region of genetic changes is indicative of a local, non-systemic causative factor which would be consistent with an exogenous environmental factor such as local dermal absorption of cosmetic chemicals. It has long been accepted that a field of genetically altered cells would be a likely place for cancer to arise, and if it is regular application of chemicals over a prolonged period which is causing this field of genetically altered cells identified in outer breast quadrants,[62] then this would be a likely place for breast cancer to arise,[63] especially within an oestrogenic environment generated from the presence of a range of environmental oestrogens.

Two recent epidemiological studies have attempted to address the question directly, but the results are conflicting. In a population-based case-controlled study, Mirick and colleagues found no difference in the use of antiperspirant/deodorant products between a population of breast cancer patients and non-affected controls.[67] By contrast, McGrath found that within a population of breast cancer patients, those who used more antiperspirant/deodorant products were diagnosed with breast cancer at an earlier age.[68] This latter study suggests a dose–response relationship to the chemical exposure and critical sensitivity at a younger age, which is consistent with oestrogen action in breast cancer.[69]

An extensive array of cosmetics is applied on and around the human breast on a daily basis, including underarm antiperspirant/deodorant products, body lotions, body sprays, moisturizing creams, breast-firming creams, breast-enhancing creams, tanning creams and suncare products. These cosmetics are not rinsed off, as are shampoos or soaps, but the entire application is left on the skin each time, allowing for accumulation in the underarm and upper breast area. Such continuous overload of chemicals can result in absorption through the dermis, with the net result of chemical deposition in underlying local tissues.[6,16,17] If such cosmetic chemicals play a role in breast cancer development, then the challenge is to identify specific chemical culprits and their mode of action.

Cancer arises from genetic changes in somatic cells of the breast—mainly epithelial cells of the breast ducts—which result in loss of growth control in affected cells. If chemical components of underarm cosmetics are involved in these processes, they must be capable of causing alterations to DNA through genotoxic and/or non-genotoxic mechanisms, and of interfering with normal growth regulatory pathways, especially those of oestrogen.[6,16,17] Several components of cosmetics have genotoxic properties, including the aluminium salts,[70] cyclosiloxanes,[71] and triclosan.[72] However, in view of the role of oestrogens in breast cancer, interference in oestrogen action would be a likely component, and many components of cosmetics have now been shown to possess oestrogen-disrupting properties. Xeno-oestrogens include parabens, the antiperspirant aluminium salts, cyclosiloxanes, triclosan, ultraviolet (uv) screens, and phthalates (Table 2). Phyto-oestrogens are also added to cosmetics in the form of anthraquinones present in Aloe Vera, and in breast-enhancing creams in the form of 8-prenylnaringenin ('Push-up') and miroestrol/deoxymiroestrol (Pueraria creams) (Table 2). These oestrogenic components are discussed in more detail in the next sections.

**Table 2.** Chemical constituents of cosmetics applied to the underarm and breast area and which possess endocrine disrupting properties.

| Chemical constituent | Function in cosmetic | Endocrine disrupter action | Evidence indicating entry to human breast |
|---|---|---|---|
| Parabens | Preservative | Oestrogen agonist activity[75–86] | Measured in human breast[74] |
| Aluminium salts | Antiperspirant | Interfere with binding of oestrogen to oestrogen receptor and oestrogen-regulated gene expression[70] | Measured in blood following application to human underarm[115,117] |
| Cyclosiloxanes | Conditioning, spreading | Oestrogen agonist activity[127–129] | Penetration of human skin[130] |
| Triclosan | Deodorant/preservative | Endocrine disruption in wildlife[134] | Measured in human milk[137] |
| Suncreeens | Absorb ultraviolet light | Oestrogen agonist activity[138,144,145] | Penetration of human skin[140,141] and measured in human milk[143] |
| Phthalates | Plasticizer | Oestrogen agonist activity[147–149] | Measured in human milk[150] |
| Nonylphenol | Metabolite of alkylphenol ethoxylate, nonionic surfactant | Steroid metabolism[39] | |
| Anthraquinones (Aloe Vera) | Wound healing | Oestrogen agonist activity[152] | |
| 8-Prenylnaringenin ('push-up') | Enhance and increase size of breast | Oestrogen agonist activity[24] | Required function achieved in consumers from dermal application |
| Miroestrol/deoxymiroestrol ('Pueraria') | Enhance and increase size of breast | Oestrogen agonist activity[24] | Required function achieved in consumers from dermal application |

## Parabens

The alkyl esters of *p*-hydroxybenzoic acid (parabens) are added in concentrations of up to 0.8% as preservatives to thousands of cosmetic products,[73] and have been detected in human breast tumour tissue at an average concentration of 20 ng/g tissue.[74] Methylparaben, ethylparaben, *n*-propylparaben, *n*-butylparaben and isobutylparaben are the most widely used esters, and these have all now been shown to possess oestrogenic activity in assay systems in vitro and in vivo.[75–86] Oestrogenic activity of parabens increases with increasing length of the linear alkyl chain from methylparaben to *n*-butylparaben,[75,83] and with branching in the alkyl chain from *n*-propylparaben to isopropylparaben[82] and from *n*-butylparaben to isobutylparaben.[84] In addition to extending the linear alkyl chain length, the oestrogenic activity of methylparaben can also be increased by extending the methyl group with a structure containing an aromatic ring in benzylparaben.[85] The relative potencies of parabens within the same assay systems are summarized in Table 3. All parabens tested, together with their main metabolite *p*-hydroxybenzoic acid, possessed oestrogenic activity (Table 3). Molecular modelling has indicated that paraben molecules—either singly or in pairs—can bind into the ligand binding pocket of the crystal structure of the ligand binding domain (LBD) of the oestrogen receptor $\alpha$ (ER$\alpha$)[87] in place of the 17$\beta$-oestradiol, and with their phenolic hydroxyl groups positioned similarly to those of meso-hexoestrol and 17$\beta$-oestradiol.[83] This has given rise to the concept that parabens may be able to act as 'half-oestrogens', binding into the LBD in pairs rather than singly.[83]

Animal studies have shown that parabens are quickly absorbed from the gastrointestinal tract and from the blood, hydrolysed to *p*-hydroxybenzoic acid, conjugated, and the conjugate excreted in urine.[88–95] Parabens can also be absorbed

**Table 3.** Comparison of the relative oestrogen agonist activities of each of 6 parabens and their common metabolite in assay systems in MCF7 human breast cancer cells.

| Compound | Binding to ER | Reporter gene | Reporter gene | Cell proliferation | Reference |
|---|---|---|---|---|---|
| | Inhibition of 3H-E binding | 24 h ERE-CAT induction | 7 day ERE-CAT induction | Growth after 12–14 days | |
| | Molar excess to achieve 50% inhibition | Molar concentrations to achieve 50% of response with $10^{-8}$ M oestradiol | Molar concentrations to achieve 50% of response with $10^{-8}$ M oestradiol | Molar concentrations to achieve 50% of response with $10^{-8}$ M oestradiol | |
| Oestradiol | 3× | | $1\times10^{-11}$ M | $2\times10^{-11}$ M | 24 |
| *p*-Hydroxybenzoic acid | 5 000 000× | $>5\times10^{-4}$ M | $>10^{-3}$ M | | 105 |
| Methylparaben | 3 000 000× | $6\times10^{-4}$ M | $>5\times10^{-4}$ M | $1\times10^{-4}$ M | 83 |
| Ethylparaben | 500 000× | $>10^{-4}$ M | $>10^{-4}$ M | $2\times10^{-5}$ M | 83 |
| *n*-Propylparaben | 300 000× | $>10^{-4}$ M | $>10^{-4}$ M | $2\times10^{-6}$ M | 83 |
| *n*-Butylparaben | 100 000× | $>10^{-4}$ M | $>10^{-4}$ M | $9\times10^{-7}$ M | 83 |
| Isobutylparaben | 40 000× | $2\times10^{-6}$ M | $2\times10^{-7}$ M | $7\times10^{-7}$ M | 84 |
| Benzylparaben | 50 000× | $3\times10^{-6}$ M | $3\times10^{-6}$ M | $1\times10^{-6}$ M | 85 |

rapidly through skin.[96–100] Human skin has been shown to possess isoforms of carboxylesterase,[101] and it has been suggested that this enzyme activity would result in hydrolysis of dermally applied paraben esters to *p*-hydroxybenzoic acid in the skin.[102,103] However, it remains unknown whether the level of carboxylesterase is sufficient to hydrolyse all dermally applied parabens and, in particular, whether high consumer use of cosmetics or individual variations in carboxylesterase levels could result in incomplete hydrolysis.[17] Furthermore, recent studies indicate that *p*-hydroxybenzoic acid itself possesses its own intrinsic oestrogenic activity both in vitro in MCF7 human breast cancer cells[104,105] and in vivo in immature rodent uterotrophic assays.[86,106]

On the basis that parabens can be dermally absorbed, that they possess oestrogenic activity, and that they are present in the human breast, it has been suggested that they could contribute to aberrant oestrogen signalling in the human breast and could adversely influence the incidence of breast cancer.[17] Their endocrine disrupting properties have generated review[107] and reassessment[108] of the safety of their widespread uses.

## Aluminium salts

Aluminium salts are used as the active antiperspirant agent in cosmetics.[109] Their mode of action is thought to involve blocking of the sweat ducts which prevents the escape of sweat onto the skin surface, probably through the formation of a physical plug composed of precipitated salts and dead cells at the top of the sweat duct.[109] So effective is this action that antipersirants are now widely used in cosmetics. However, aluminium is known to be toxic,[110–112] and aluminium salts are permitted at high levels: aluminium chloride, aluminium chlorhydrate and aluminium zirconium chlorhydrate glycine complexes at up to 15, 20 and 25% by weight, respectively.[109] Dermal absorption of topically applied antiperspirant aluminium salts has been demonstrated through intact mouse skin[113,114] and the skin of the human underarm.[115–118] Aluminium in the form of aluminium chloride or aluminium chlorhydrate has been shown capable of interfering with the function of oestrogen receptors of human breast cancer cells both in terms of ligand binding and oestrogen-regulated gene expression.[70]

Other metals have already been shown capable of interfering with oestrogen action, defining a new class of endocrine disrupters termed metallo-oestrogens. Replacement of zinc in the zinc finger of ER with either nickel or copper could inhibit the binding of the DNA-binding domain of the ER to the ERE in the DNA.[119] Cadmium,[120] arsenite,[121] selenite,[122] copper, cobalt, nickel, lead, mercury, tin, chromium and vanadate[123] can all interfere with oestradiol binding to the ligand binding domain of the ER and influence oestrogen-regulated gene expression. Furthermore, oestrogenic affects of cadmium have also been demonstrated in vivo in the rodent uterus and mammary gland.[124] Aluminium can now be added to this list of metallo-oestrogens,[70] but the full range of its genotoxic potential[110–112] in breast cells remains to be determined.

## Cyclosiloxanes

Cyclosiloxanes and linear siloxanes have been incorporated into thousands of cosmetics as conditioning and spreading agents.[125,126] Octamethylcyclotetrasiloxane (D4) is a low-molecular-weight cyclic siloxane with high lipid solubility, and is permitted in personal care products at 40–60% by weight.[126] Research over the past four decades demonstrates the toxicity of D4[71] and its ability to interfere with the female

reproductive system.[127] D4 has been shown to possesses intrinsic oestrogenic activity in in vitro models and in in vivo uterotrophic assays,[128,129] and due to its high lipid solubility it can accumulate in fatty tissues after exposure.[126] Since D4 has been shown to be absorbed through human skin, with an average 1.09% of the administered dose being absorbed,[130] the effects of frequent cosmetic application and continuous exposure in leave-on cosmetics needs to be reassessed, especially in the light of studies demonstrating retention in body fat for at least 1 year.[131]

## Triclosan

Triclosan is an effective antiseptic[132] which is used widely in cosmetics as a preservative to prevent microbial growth in products during storage, and as a deodorant to prevent microbial growth on the skin surface which would interact with sweat to generate a smell. The action of triclosan is thought to reside in its ability to inhibit the enoyl-reductase enzymes of type II fatty acid synthases in susceptible bacteria.[133] However, it may have cytotoxic effects on human breast cancer cells,[133] and has been shown to possess endocrine-disrupting properties in aquatic species which indicate weak estrogenic and/or weak androgenic activity.[134] So ubiquitous is its use in consumer products that it has been detected in the aquatic environment[135] and in human tissues, both blood plasma[136] and milk.[137] Its presence in human milk must imply its passage through human breast epithelial cells.

## UV screens

Chemicals that absorb UVA (315–400 nm) or UVB (280–315 nm) radiation are used with increasing frequency in cosmetics due to the growing public concern over skin damage by UV light, and they can be added in concentrations of up to 10% for skin protection, despite uncertainty over the benefit for protection against melanoma.[138] Due to their widespread use and lipophilic properties, they can be expected to bioaccumulate in the environment, and measurements in fish have shown these UV screens to be present at a level comparable with that of PCBs and DDT.[139] In humans, they are known to be absorbed through skin,[140,141] have been detected in urine[141,142] and measured in human milk.[143] Benzophenone-3 (Bp-3), homosalate (HMS), 4-methyl-benzylidene camphor (4-MBC), octyl-methoxycinnamate (OMC) and octyl-dimethyl-PABA (OD-PABA) have all been demonstrated to possess oestrogenic activity in human breast cancer cells in culture and in the uterotrophic assay in vivo in rats.[138] 2,4-Dihydroxybenzophenone and 2,2′,4,4′-tetrahydroxybenzophenone have been reported to give a uterotrophic response in rats.[144] Dermal application of 4-MBC increased uterine weight at concentrations of 5 and 7.5% in olive oil.[138] 4-MBC and OMC have also been shown to have oestrogenic activity in vivo in fish.[145]

## Phthalates

Phthalates are used as plasticizers and as such are present ubiquitously in the environment. However, they are also present in many cosmetic products.[146] They have been shown to possess oestrogenic properties,[147–149] and their endocrine-disrupting properties are detailed elsewhere in this volume. Phthalates have been measured in human milk,[150] implying their presence in human breast, but the ubiquitous exposure to

phthalates through their many uses in plastics does not allow any implication concerning the origin of the phthalates measured in human milk.

### Musk fragrances

Nitromusks and polycyclic musk fragrances are found in a wide variety of cosmetics, perfumes and laundry detergents, and several possess oestrogenic activity.[151]

### Phyto-oestrogens

Phyto-oestrogens are used increasingly in cosmetics designed for application around the human breast. There is an increasing trend towards addition of Aloe Vera into personal care products, and the constituent anthraquinones are known to possess oestrogenic properties.[152] The phyto-oestrogens 8-prenylnaringenin and miroestrol/deoxymiroestrol are added to cosmetics for the purpose of breast enlargement, and are both known to possess high oestrogenic potency in human breast cancer cells.[24] Their popularity and sales are testimony to the fact that their stated function works, and thus that from dermal application they enter the body in sufficient quantity to influence physiological events in the human breast.

## COSMETIC CHEMICALS AND BENIGN BREAST DISEASE

Cancer represents only about 5% of clinical abnormalities of the breast, with benign conditions including fibroadenomas and breast cysts.[65] The reason for such a high incidence of benign conditions is unknown, but they are of concern because their presence can be an indicator of future development of cancer.[153] Interestingly, the UOQ is not only the most common site of the tumour in cancer but also of the abnormalities in benign breast conditions, including fibroadenoma, breast cysts,[154] and phyllodes tumour,[155] which suggests that if underarm cosmetics play a role in this phenomenon in cancer then they may also do so in benign conditions.

Since antiperspirants act by blocking sweat ducts,[109] and breast cysts result from blocked breast ducts,[65] it is possible that breast cysts could also arise from antiperspirant use. The action of antiperspirants is thought to arise through the formation of a physical plug in the sweat duct which is composed of precipitated cosmetic salts and dead cells.[109] The breast, being a modified sweat gland,[156] is also composed of a network of ducts but these drain out to the nipple for the purpose of milk secretion. Excessive application of antiperspirant cosmetics beyond the underarm to adjacent breast areas could result in absorption of antiperspirant chemicals (aluminium salts) and deposition in underlying breast tissues. This could facilitate subsequent blockage of breast ducts at varied points in the breast if sufficient chemicals are absorbed or accumulated over long periods of usage. It is, therefore, possible that the development of breast cysts could be an indicator of careless or excessive use of antiperspirant, which the user should heed. Similarly, the development of other benign tumours and their disproportionate incidence in the UOQ could also be an indicator of abnormal cell behaviour resulting from a level of cosmetic chemicals not well tolerated by the user, and oestrogenic chemicals may play a role here. Variations in product formulations and practices of usage, together with individual differences in absorption, metabolism and clearance of constituent chemicals, would be expected to give rise to

a wide range of susceptibilities and end phenotypes. If such a proposed link does exist, then the user, by reducing or eliminating cosmetic use, might have an opportunity to reduce the risk of subsequent cancer development.[153]

## COSMETICS AND INHERITED SUSCEPTIBILITY TO BREAST CANCER

Development of breast cancer through the inheritance of genetic susceptibility is assumed to have a different origin from the sporadic forms of breast cancer. The breast cancer susceptibility genes BRCA1 and BRCA2 act as tumour suppressor genes in that loss of their function is associated with increased risk of breast cancer, and their function is now known to be related to maintenance of DNA repair processes.[7] However, the identity of the source of the DNA damage subject to the compromised repair processes remains unknown. Although it is possible that cancers may result in these people from an inability to repair random replication errors, it is also possible that these people are simply more susceptible to genotoxic pollutant chemicals than the remainder of the population who have intact DNA repair systems. Direct and continuous dermal exposure to genotoxic and oestrogenic cosmetic chemicals provides a source of such chemicals local to the breast and should be investigated.[16]

## ENVIRONMENTAL OESTROGENS AND ENDOCRINE THERAPY

The ability to reduce breast tumour growth through manipulation of oestrogen action has played a central role in the endocrine therapy of breast cancer,[10] but little consideration has been given to the potential interaction of the presence of oestrogenic chemicals in the human breast on the effectiveness of this therapy in individual patients. Anti-oestrogens are used to compete with oestrogen for binding to the ER, and when attached to the ER can either partially or completely antagonize oestrogen action. Tamoxifen is the most widely used anti-oestrogen, and about one half of patients with ER-positive ($ER^+$) breast cancer can benefit from treatment.[10] Due to the partial agonist properties of tamoxifen, a new pure antagonist, fulvestrant (Faslodex, ICI 182 780), has been produced to supplement tamoxifen as first- or second-line endocrine therapy.[10] An alternative approach to reducing oestrogen action has been to use aromatase inhibitors which act to block oestrogen synthesis within the body.[10] Oestrogen is synthesized in the ovary and adrenal glands under control from the hypothalamus and pituitary through conversion of androgen to oestrogen by the enzyme aromatase. The key in this strategy has been to find drugs which can bind to aromatase either competitively or non-competitively to inhibit the enzyme activity. Inhibitors have been developed as either steroidal or non-steroidal, and come as first-, second- and third-generation drugs, each generation representing increased specificity and greater potency. The non-steroidal third-generation drugs anastrazole and letrozole fit well into the active site of the enzyme, and their high potency and good specificity have resulted in good clinical responses.[10]

The presence of oestrogenic chemicals in the human breast could potentially influence the effectiveness of these therapeutic approaches in the short or the long term. Development of resistance to endocrine therapy is a major clinical problem for which mechanisms remain unidentified. Whilst it is clear that breast cancer cells have a remarkable ability to circumvent any form of imposed growth inhibition in their

progression to a state of endocrine resistance,[8,9] it could also be that, in some cases, effectiveness of endocrine therapy is compromised not only by the emergence of oestrogen-resistant cells but also by the presence of environmental oestrogens in the breast. Theoretically it would seem self-defeating to reduce endogenous oestrogen synthesis with aromatase inhibitors and at the same time allow the breast to be exposed to a range of environmental oestrogen-mimicking chemicals. The action of these drugs is dependent on the ability to block endogenous oestrogen synthesis, and they would be ineffective against any parallel overload of environmental oestrogenic chemicals. Since environmental oestrogens tend to have a reduced binding affinity for ER relative to the physiological 17β-oestradiol,[76] anti-oestrogens would theoretically be capable of countering the action of environmental oestrogens as well as endogenous oestrogens. However, overload of oestrogenic chemicals could still result in the need for higher doses of tamoxifen or fulvestrant to give a clinical response. Perhaps, women undergoing endocrine therapy should be advised not to apply oestrogenic cosmetic chemicals around the breast, and certainly not to do so in conjunction with shaving, which would allow for particularly easy access for the chemicals. Since lipophilic oestrogenic chemicals stored and accumulated in human breast fat can be mobilized during weight loss, perhaps weight loss during endocrine therapy should also be viewed with concern.

## CONCLUSIONS AND APPEAL FOR FURTHER RESEARCH

There is no doubt that the human breast is exposed to a wide range of oestrogenic chemicals from environmental sources. At the present time, there is no proven link between the presence of such chemicals and the development of breast cancer, but this is because the necessary work has not been done, not because research has found no effect. Oestrogen is an established factor in breast cancer development, and the presence of such a range of oestrogenic insults within the human breast must be fully investigated. Although xeno-oestrogens have been identified within a wide range of environmental sources, it remains possible that other origins remain still to be identified. Furthermore, with so many and varied sources of oestrogenic chemicals, there is a need to try to summate the many environmental insults on oestrogen action. Most studies to date have considered single chemicals in isolation, which alone may or may not reach levels in the human breast which are equivalent to those needed for measurable effects in vitro. Research has demonstrated that the breast is subject to a multitude of oestrogenic chemicals at any one time, and these may interact with one another and with endogenous oestrogens. Taken together, the total oestrogenic insult may be more significant than when considering each chemical alone in isolation. Within any experimental assessment of mixtures, it will be important to ascertain the relative contribution of phyto-oestrogens (eaten in large amounts but cleared quickly) compared with that of xeno-oestrogens (entering the body in small amounts but accumulated). It will also be important to consider the relative effect of continuous direct dermal exposure to cosmetic oestrogens. Research will need to take into account issues of timing with respect to latency and periods of breast vulnerability, as well as individual differences in susceptibility. Only through much more extensive research can an overall understanding be gained of the full extent of the burden of aberrant oestrogen signalling caused by environmental oestrogens in the human breast.

Although interference in oestrogen action could be sufficient for reproductive and developmental abnormalities to arise, a central question still remains as to whether oestrogenic activity alone is adequate for breast cancer development. Cancer is known to involve genotoxic and non-genotoxic alterations to the genome of affected cells leading to alterations in growth control genes. Whilst an oestrogenic environment is known to influence breast cancer incidence, promotion, growth and progression, it remains uncertain as to its role in the genetic changes associated with initiation of cancer. It is possible that increased proliferation associated with an oestrogenic environment could simply enhance the effects of random replication errors. Long-term exposure to steroidal oestrogens is also known to be capable of generating genomic instability.[157] However, within the context of chemical mixtures, it is also possible that cancer could arise from exposure to both genotoxic and oestrogenic chemicals. Little is known of the genotoxic potential of xeno-oestrogens. Xeno-oestrogens have been shown to influence oestrogen-regulated reporter genes and a few single endogenous genes, but little is known of their effects on global patterns of gene expression or their ability to damage DNA. Cosmetic chemicals such as aluminium salts,[70] cyclosiloxanes[71] and triclosan[72] are already known to have DNA-damaging properties as well as oestrogenic activity. Our knowledge of the mechanisms of environmental oestrogen actions remains focused on cell proliferation. However, oestrogen action on breast cancer cell growth is known to influence more than proliferation, through actions on apoptosis, differentiation, cell adhesion and cell motility.[1,8,9] Oestrogen action in breast cancer can also be influenced through the interaction of two oestrogen receptors, ER$\alpha$ and ER$\beta$.[158] Some phyto-oestrogens have been shown to bind more strongly to ER$\beta$ than to ER$\alpha$,[23] and there may also be differences in xeno-oestrogen action through the two receptors.[159] Most research has focused on ER-mediated actions, but other mechanisms of action of endocrine-disrupting chemicals can involve alterations to the synthesis or metabolism of physiological hormones (see Chapters 2 and 4 of this issue). In this context, it is interesting that nonylphenol has recently been shown to alter oestrogen metabolism and to increase mammary tumour rates in mice.[39] It is evident that much more information is needed on the full profile of actions of environmental oestrogens and their interactions with other pollutant chemicals in the human breast.

The potential link between breast cancer and the application of oestrogenic chemicals in cosmetics provides an evidence-based hypothesis capable of being tested further. Although individual chemicals will have been tested by current safety guidelines, emergence of new data showing endocrine-disrupting activity of so many components warrants new investigation into their individual and combined effects. Furthermore, testing would normally involve relatively short-term high-dose studies of single compounds, and the long-term use of product formulations at lower levels over an entire lifetime warrants retrospective investigation. In a culture tending to increasing use of these products,[68] the lack of any advice concerning safe quantity or frequency of application should be of concern. Use of such chemicals by young children prior to puberty and by babies should be a further area for enquiry. Tendency for consumer use to involve underarm shaving prior to cosmetic application should also be considered, since shaving can create nicks and abrasions in the skin which allow particularly easy access for chemicals. If use of underarm and bodycare cosmetics is a factor in the development of breast cancer, then options for prevention could become a reality, either through individual decisions to cease usage or through alterations to product formulations. Measures to prevent breast cancer remain long-awaited.

**Practice Points**

- why is the incidence of breast cancer increasing?
  - research and clinical practice tends to focus on early detection and treatment, but there is a need to prevent breast cancer
- could oestrogenic chemicals play a role in the rising incidence of breast cancer?
  - many environmental chemicals with oestrogenic activity have been measured in the human breast
- could chemical constituents of underarm cosmetics be a cause of breast cancer?
  - many cosmetic chemicals possess genotoxic and oestrogenic properties, and these chemicals are applied to the breast area, allowing continuous direct dermal exposure; this provides a testable hypothesis, which if correct would open avenues to breast cancer prevention
- could antiperspirants cause breast cysts?
- should women undergoing endocrine therapy be advised to reduce their exposure to environmental oestrogens?

**Research agenda**

- controlled studies are investigating phyto-oestrogen action in breast cancer; systematic studies need to be carried out also for xeno-oestrogens
- research needs to investigate whether cosmetic chemicals applied to the underarm and breast area are a cause of breast cancer
- most research to date has centred around study of single chemicals in isolation. However, the human breast is exposed to a multitude of environmental oestrogens. Research now needs to investigate the effects of mixtures and to summate total oestrogenic burdens in the breast
- research is needed into mechanisms of susceptibility
- research needs to consider timing of xeno-oestrogen exposure and the effects of xeno-oestrogens during periods of breast vulnerability and latency

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