Estrogen and Its Receptors in Cancer

George G Chen,[1] Qiang Zeng,[1] Gary MK Tse[2]

[1]Department of Surgery, The Chinese University of Hong Kong, Prince of Wales Hospital, Shatin, N.T., Hong Kong SAR, China
[2]Department of Anatomical and Cellular Pathology, The Chinese University of Hong Kong, Prince of Wales Hospital, Shatin, N.T., Hong Kong SAR, China

Published online 18 July 2008 in Wiley InterScience (www.interscience.wiley.com).
DOI 10.1002/med.20131

Abstract: The involvement of estrogen and its receptors in the development of cancer has been known for years. However, the exact mechanism responsible is far from clear. The estrogen-mediated carcinogenic process is complicated by recent findings, which reveal that estrogens have multiple functions in cells, which can be either adverse or beneficial, and that the effects of estrogen may be cell-type or organ dependent. The estrogenic effect may be also greatly influenced by the state of two estrogen receptors, ERα and ERβ. This review will discuss the role and function of estrogens and its receptors in cancers of three categories: (1) Breast cancer and gynecologic cancers, (2) Cancers of endocrine organs, (3) Lung cancer and cancers of digestive system. We will also review some novel treatments aiming to interfere with relevant pathways mediated by estrogens and its receptors. Med Res Rev, 28, No. 6, 954–974, 2008

Key words: estrogen; estrogen receptor; cancer

1. INTRODUCTION

Estrogen regulates the growth, differentiation, and physiology of the reproductive process. Estrogen also influences the pathological processes of hormone-dependent cancers, such as breast, endometrial, prostate and ovarian and thyroid cancers.[1,2] Estrogens are a group of steroid compounds which function as the primary female sex hormones. While estrogens are present in men and women, their levels are significantly higher in women of reproductive age. They are mainly produced by the adrenal cortex and ovary, and three estrogens occur naturally in the female.[1,3] In pre-menopausal women, 17β-estradiol (E2), produced by the ovary, is the estrogen produced in the largest quantity and is the most potent as it has the highest affinity for its receptors. In pre-menopausal

Contract grant sponsor: CUHK direct grant; *Contract grant number:* 2041163; *Contract grant sponsor:* the Research Grants Council of the Hong Kong Special Administrative Region; *Contract grant numbers:* CUHK4390/03M and CUHK4556/05M
Correspondence to: G.G. Chen, Department of Surgery, Prince of Wales Hospital, Shatin, New Territories, Hong Kong, China.
E-mail: gchen@cuhk.edu.hk

women, circulating estradiol levels fluctuate from 40 to 200–400 pg/mL during the menstrual cycle.[3] After menopause, estradiol levels drop to less than 20 pg/mL. The second endogenous estrogen is estrone, a less potent metabolite of estradiol. Estrone is produced from androstenedione, the immediate precursor of estrone, in adipose tissue. In post-menopausal women, the ovary ceases to produce estradiol while the adrenal gland continues to produce androstenedione. As the result, the level of estrone remains unchanged while the plasma level of estradiol falls significantly. The third endogenous estrogen is estriol (E3), also a metabolite of estradiol. Estriol is the principal estrogen produced by the placenta during pregnancy, and is found in smaller quantities than estradiol and estrone in non-pregnant women.[1,3]

The biological actions of estrogens are traditionally mediated by binding to one of two specific estrogen receptors (ERs), ERα or ERβ, which belong to the nuclear receptor (NR) superfamily, a family of ligand-regulated transcription factors. ERα and ERβ are encoded by separate genes, ESR1 and ESR2 respectively. Although binding of estrogens to ERs results in a variety of changes in cell functions, the mechanism of its action can be classified in two pathways: genomic and non-genomic.

A. Genomic Pathway

Estrogen influences the cellular events through two main pathways, genomic and non-genomic (Fig. 1).[1] In the genomic pathway, estrogen exerts its function *via* ERα and ERβ. In general, this classical pathway of estrogen involves estrogen-dependent formation of nuclear ER homo- or heterodimers, and the subsequent binding of this nuclear estrogen-ER complex binds to estrogen response element (ERE) sequences in the promoter region of estrogen-responsive genes, resulting in the recruitment of coregulatory proteins (co-activators or co-repressors) to the promoter, which leads to an increase or decrease in mRNA levels, the production of associated proteins and finally a physiological response. The genomic pathway typically occurs over the course of hours. There is evidence that ERα and ERβ can regulate transcription of some genes independent of ERE by interacting with other DNA-bound transcription factors, rather than binding directly to DNA,[1,2] which may explain that about one third of estrogen-induced genes lack functional ERE.[4] AP-1, SP-1, forkhead box (Fox), oct, nuclear factor kappaB (NF-*k*B) and GATA-3 are some of known non-ERE DNA-bound transcription factors that interact with ERs.[5–7] Interestingly, the level of E2 that determines the saturation of liganded/unliganded ERs may have opposite effects on the expression of TNFα *via* interacting with glucocorticoid receptor-interacting protein 1 (GRIP1), c-jun and NF-*k*B.[5]

B. Non-Genomic Pathway

In the non-genomic pathway, estrogen acts either through the ER located in or adjacent to the plasma membrane, or through other non-ER plasma membrane-associated estrogen-binding proteins (Fig. 1).[1–4,8] The non-genomic action of estrogen results in cellular responses such as increased levels of calcium or nitric oxide and the activation of multiple intracellular kinase cascades including mitogen-activated protein kinase (MAPK), phosphoinositide 3-kinase (PI3K), protein kinase A (PKA), protein kinase C (PKC). The ER may be targeted to the plasma membrane by adaptor proteins such as caveolin-1 or Shc (an SH2 containing proto-oncogene). AP-1 response elements, for instance, may be regulated indirectly through interactions between ERs and the AP-1 transcription factors c-fos and c-jun. On the other hand, AP-1-dependent transcription may also be directly and efficiently activated by binding of E2 to the cytoplasmic ERs that may form a complex with SRC-1, p300, ubiquitin ligase E6-AP, Mdm2, Carm, and pol II.[9,10] These transcription factors regulate genes involved in many cellular processes, including proliferation, differentiation, cell motility, and apoptosis. The non-genomic effects occur within a few minutes, which is too rapid to be mediated by biosynthesis of RNA or new proteins.

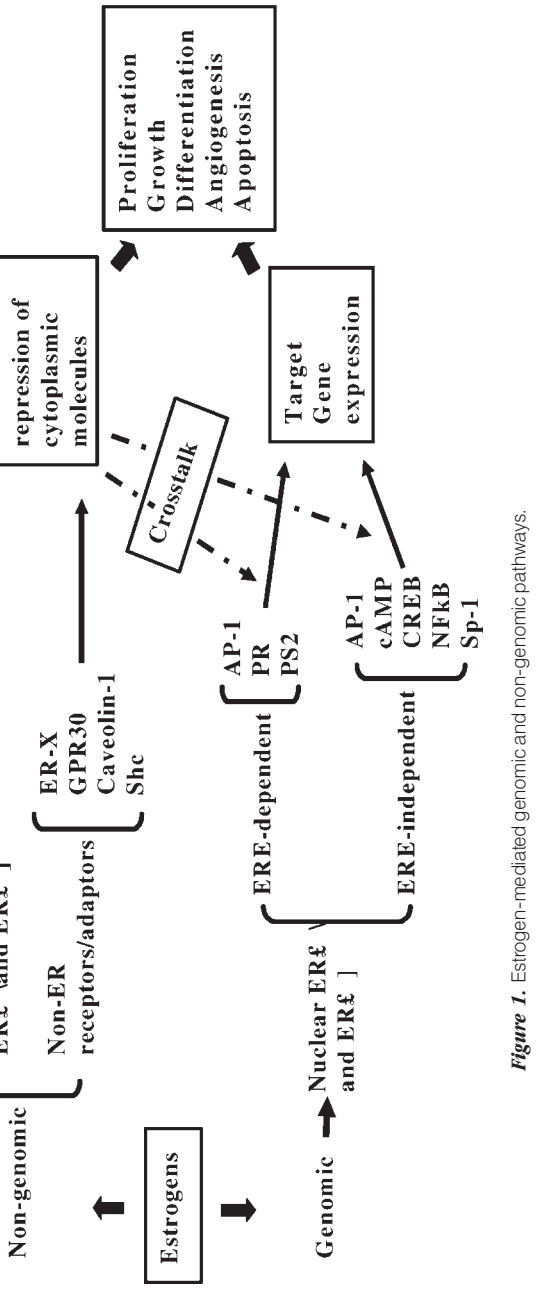

Figure 1. Estrogen-mediated genomic and non-genomic pathways.

C. Non-Genomic Pathway Crosstalk to Genomic Pathway

Though the genomic and non-genomic pathways function differently, studies have indicated that there is a crosstalk between them (Fig. 1).[4,8] The rapid non-genomic activation of cytoplasmic signaling pathways has been shown to regulate gene expression independent of ERE. For example, ERK activation in the non-genomic pathway enhances AP-1-mediated gene expression. The non-genomic signaling can activate MAPK to stimulate the phosphorylation and recruitment of coactivators such as SRC-1 and the phosphorylation of SRC-1 enhances nuclear ER transcriptional activity.

D. G Protein-Coupled Receptor-30

Although the transcriptional effects of estrogen in many cases can be explained by the binding of estrogen to its traditional receptors, ERα and ERβ, it is now widely accepted that some of its rapid effects cannot be attributed to ERα and ERβ. Recent studies have discovered some novel estrogen receptors.[1,11] Among them, G protein-coupled receptor-30 (GPR30), an orphan member of the seven transmembrane receptor, has been shown to involve in cancer development. Estrogen through GPR30 can stimulate MAPK and ERK1/2, and transactivation of the epidermal growth factor receptor (EGFR) independent of ERα and ERβ in breast cancer SKBR3 cells.[12,13] However, GPR30 is not involved in estrogen-mediated MAPK activation in breast cancer MCF-7 cells.[14] The findings suggest that the GPR30-mediated pathway of estrogen might be cell type-dependent. E2-GPR30 interaction has similar characteristics as the non-genomic signaling of E2-ERs and thus it can be regarded as an alternative non-genomic pathway.

E. Affinity of Estrogen Receptors and SERMs: Selective Estrogen Receptor Modulators

ERα and ERβ differ mostly in the N-terminal A/B and F domains, exhibiting a 15% and 18% identity respectively.[1,2] The ligand binding domain (E domain) is moderately conserved between both receptors and shows a 59% amino acid identity. Despite the identity between ERα and ERβ in the ligand binding domain, the two receptors exhibit differences in ligand binding specificity.[15] A number of compounds have been found to have different binding affinities to ERs. These compounds are capable of acting as ER agonists in some tissues but as antagonists in others. Therefore, they are selective estrogens and antiestrogens at different target tissues and are termed as selective ER modulators (SERM).[16,17] From the therapeutic point of view, the ideal SERM should be antiestrogenic in cancer tissues but proestrogenic in the vasculature and brain. Chemically, SERM can be classified in five groups: triphenylethylenes, benzotiophenes, tetrahydronaphtylens, indoles, and benzopyrans. Chemicals in the latter three groups are still under the development or clinical trials. Triphenylethylenes are developed for the treatment of the estrogen-dependent breast cancer. The representative of this category is tamoxifene (ICI 46474) that is clinically used as a first-line endocrine treatment. However, tamoxifene has proestrogenic effect on the endometrium and its treatment is associated with an increase in the incidence of endometrial cancer. For this reason, its use for the endocrine treatment may be limited though it is still recommended for the treatment of ER-positive breast cancer.[17] Benzotiophenes constitute a second chemical group. Raloxifene is the main molecule for this class. The major advantage of raloxifene over tamoxifene is that it does not have a stimulatory effect on the endometrium and thus will not increase risk for endometrial cancer.[16,17] Therefore, it is now used to prevent breast cancer in high-risk women.[17,18] Table I summarizes the values of the relative binding affinity of some SERM to ERα and ERβ.[19,20] The mechanism responsible for the action of SERM is unclear. However, it is now believed that the action cannot completely explained by ER models. The relative expression of co-activators and co-repressors within a cell affects the function of SERM.[18]

Table I. Values of the Relative Binding Affinity of Some SERM to ERα and ERβ

SERM	ERα	ERβ
E2	100	100
genistein	4	87
ICI 164,384	85	166
tamoxifen(4-OHT)	257	232
tamoxifen	4	3
toremifene	10	90
raloxifene	69	16
DPN	0	18
8ß-VE2	0	83
ERB-041	0	72
WAY-200070	0	180
WAY-202196	0	133
PPT	49	0
16-LE2	57	0

Relative binding affinity was calculated as ratio of concentrations of E2 to reduce the specific radioligand binding by 50%.
The value for E2 is arbitrarily set at 100.

F. Aromatase and its Inhibitors

The importance of ER in the development and progression of hormonally sensitive tumors has been well recognized. However, it has also been noted that some of these tumors can develop in an ER-free environment. The occurrence of mammary tumor in ER knockout mice is the best evidence to support this concept.[21] People were once puzzled by the fact the incidence of breast cancer is even higher in post-menopausal women when the ovaries have ceased to produce estrogens. It is now known that in post-menopausal women, estrogen levels in breast tissue are 10–50 times the level in blood and concentrations of E2 are higher in malignant tissues than in non-malignant tissues.[22] Therefore, estrogen can be *de novo* produced in some types of tumors including breast cancer, indicating a significant role of the peripheral aromatization of ovarian and adrenal androgens in cancer development.[23] Evidence also shows that estrogen metabolites may also be carcinogenic.[23] The above information leads to the application of aromatase inhibitors such as anastrozole, letrozole and exemestane to attenuate estrogen biosynthesis and thus to treat certain types of hormonally sensitive tumors. Recent large clinical trial have shown that aromatase inhibitors are superior to tamoxifen for prolonging disease-free survival in post-menopausal women with ER-positive breast cancer in adjuvant scenario.[24] In addition, aromatase inhibitors have been used in the treatment of metastatic breast cancer in post-menopausal women.[24] These agents might also have a potential role in the prevention setting.[25]

G. Estrogen Receptor Variants

The function of estrogens can be influenced by a balance between wild-type ERs and their functional exon-skipping variants, which may encode proteins interfering with the co-expressed wild-type forms in a dominant negative manner or by becoming constitutively and ligand-indpendently activated.[1,26–28] In breast cancer MCF7 cells, in addition to a predominant full-length 66 kDa form of ERα (ERα66), the ERα gene also produces a 46 kDa isoform (ERα46), lacking N-terminal domains.[29] ERα46 has been shown to suppress not only the activation function-1 (AF1) activity of ERα66 but also the transcription of target genes.[29] However, whether the presence of ERα46 has any association with anti-tumor treatment remains unknown. Recently, a report describes the function of

ERβ and its variants in breast cancer.[30] Two ERβ splice isoforms, one lacking ERβ1 exons 1, 2, and 5 (ERβδ125) and the other lacking exons 1, 2, 5, and 6 (ERβδ1256) are identified in MDA-MB231 breast cancer cells. Over-expression of wild-type ERβ, but not the two exon-deleted variants exerts strong antitumor effect on either ERα-positive or ERα-negative breast cancer cells. This antitumor effect is reflected by inhibition of E2-stimulated cell growth, enhancement of apoptosis, and increase in the antiproliferative effect of tamoxifen on tumor cells.[30] In addition to the above isoforms or variants of ERs, a number of other variants have been found in different types of tissues or cells, which has been well summarized elsewhere.[2,31]

2. ESTROGEN AND ESTROGEN RECEPTORS IN CANCERS

Estrogen and its receptors are involved in the development of many types of malignant tumors. These tumors are generally classified in four groups, (1) breast and gynecologic cancers (cervical endometrial, ovarian), (2) endocrine gland cancers (adrenocortical, ovarian, pancreatic, prostate and thyroid), (3) digestive cancers (colorectal, esophageal, liver and pancreatic), and (4) lung carcinoma. Most of these tumors can express both ERα and ERβ (Table II). ERα appears to promote the proliferation of breast, gynecologic cancers and endocrine gland cancers but to inhibit the proliferation of digestive cancers and lung cancer. In contrast to ERα, ERβ suppresses the proliferation of tumor cells in the former groups but increases it in the latter groups. The prognostic significance of ER change is inconsistent among these tumors. However, the decreased level of ERβ is usually associated with poor prognosis (Table II). Most of the above-mentioned tumors are able to *de novo* biosynthesize E2 through the action of aromatase (Table III). The locally produced E2 may then function in a paracrine manner to stimulate the proliferation and growth of cells or/and to render cells more resistant to apoptosis. However, it is noted that the function of E2 in at least two organs colorectum and esophagus to inhibit proliferation and growth of tumor cells, which is opposite to its function in the most other organs. The differential effects of estrogens on organs are further supported by the epidemiological study on risk of cancers in patients who receiving estrogen-replacement therapy (ERT) or hormone-replacement therapy (HRT)/oral contraceptives (OC) (Table IV). The risk

Table II. ERα and ERβ in Cancers, and Their Impacts on Cell Proliferation and Prognosis

Site of cancer	ERα	ERβ	increase in proliferation	poor prognosis	References
Adrenocort	+	+	ERβ↓,ERα↑	ND	101
Breast	+/-	+/-	ERβ↓	ERβ↓	2,22,61,62,68,69
				ERα↓	139
Cervix	+	ND	ERα↑	ND	56
Colorectum	↓,-,~,	↓	ERα↓, ERβ↓	ND	125-128,140
Endometrium	↓	↓	ERα↑	ERα↓	57,76,141,142
Esophagus	+	+	ND	ERα↑ERβ↓	131,132
Liver	↑	↑	ND	vER	133,134,143
Lung	+	+	ERα↓ERβ↓	ERα↑ERβ-	135,136,138,144
Ovary	+	+	ERβ↓	ERβ↓	63,145
Pancreas	+	+	ND	ND	102,113
Prostate	+	↓	ERα↑ERβ↓	ERβ-	97,104,107,146
Thyroid	↑,~	↓,~	ERα↑ERβ↓	ND	109-112,147

+, expression/positive; −, no expression/negative; ↑, increase (compared with non-tumor or normal tissues/cells); ↓, decrease (compared with non-tumor or normal tissues/cells); ~, no effect or no change; ND, not data available; vER, variant ERs.

Table III. Estrogen Synthesis in Cancers and its Influence on Cell Functions

	Estrogen synthesis	Estrogen function		References
		Proliferation	Apoptosis	
Adrenocort	+	↑	ND	101
Breast	+	↑	↓	22,23,47,48,77,148
Cervix	+	↑	↓	39,149,150
Colorectum	+	↓ or ↑	↑	116,117,125,128,151
Endometrium	+	↑	ND	40,59,60,152,153
Esophagus	ND	↓, ~	ND	129,130
Liver	+	ND	ND	154
Lung	+	ND	ND	155,156
Ovary	+	↑	ND	42,43,157
Pancreas	ND	↑	ND	102
Prostate	+	↑	ND	98,103,104
Thyroid	+	↑	↓	105,110,58

+, positive; −, negative; ↑, increase (compared with non-treated group); ↓, decrease (compared with non-treated group); ~, no change; ND, not data available.

for the development of breast, gynecologic cancers and endocrine gland cancers is generally increased in patients who have female hormone treatments. In contrast, the risk for the development of digestive cancers and lung cancer is generally decreased in patients who receive these treatments. Currently there is no an explanation for these observations of obviously opposite impacts of female hormones on cancers. Nevertheless, the findings strongly indicate that the function of female hormones may be organ-dependent.

Table IV. Relationship Between Female Hormone Use and Cancer

cancer	ERT	EP (HRT, OC)	References
Adrenocort	ND	ND	
Breast	↑	↑	32-35
Cervix	↑	↑,-	38,159,160
Colorectum	↓	↓	34,35,115,118-121
Endometrium	↑	↑	33-35,120,161
Esophagus	ND	↓	122
Liver	ND	↓	35
Lung	↓	↓	123,124
Ovary	↑	↑	34,120,121,162-164
Pancreas	-	-	165
Prostate	ND	ND	
Thyroid	↑	↑	87-90

-, no relationship; ↑, increase (compared with non-treated group); ↓, decrease (compared with non-treated group); ND, not data available.
ERT, estrogen-replacement therapy; EP, estrogen + progestin; HRT, hormone-replacement therapy; OC, oral contraceptives.

A. Breast and Gynecologic Cancers (Cervical, Endometrial, Ovarian)

The role of estrogens and ERs has been extensively investigated in breast cancer. Epidemiologically, the incidence of breast cancer is significantly higher in women who receive ERT or HRT, especially in post-menopausal women with long-term of hormone treatments.[32–35] There is increasing evidence to show that breast tumors express a high level of aromatase to biosynthesize estrogen locally.[22,23] The consequence of such a high level of aromatase is that the tumor is able to produce estrogens locally in an autocrine fashion, resulting in much higher estrogen levels in the tumor tissue than non-tumor tissues or circulation system.[22,23] The involvement of estrogen in the breast cancer development is further evident by study using aromatase inhibitors. The inhibition of aromatase by its chemical inhibitors or siRNA has been shown to suppress the proliferation of breast cancer cells in culture and reduce the growth of breast tumor in animal experiments as well as patients with breast cancer.[22,23,36,37] Similar to breast cancer, there are also solid supports of the involvement of estrogen in the development of gynecologic tumors including cervical, endometrial, ovarian cancers. Female hormone therapies are positively associated with these cancers (Table IV). Hormone therapy may also help to create a carcinogenic environment for certain gynecologic tumors. For example, HPV infection in cervical cancer may benefit from HRT.[38] The majority of these gynecologic tumors also has an autocrine mechanism to produce intratumoral estrogen *via* aromatase.[39–43] The expression of aromatase in the tumor can also affect the survival of patients. For example, there is an association between intratumural aromatase expression and poor survival in endometrial cancer.[44]

The carcinogenic effect of estrogen is executed at least in three ways (Fig. 2). First, estrogens promote the proliferation of cells *via* ER-mediated genomic or/and non-genomic pathways. A increasing number of estrogen-induced molecules have been identified and these molecules can function to promote cell proliferation, growth, to reduce sensitivity to apoptotic stimuli, to enhance invasiveness. For example, E2 can stimulate the LRP16 gene expression *via* ERα in breast cancer and the induced LRP16 can interfere with ERα-mediated transcription of E-cadherin, resulting in the reduction of E-cadherin and subsequently the invasive growth of breast cancer and endometrial cancer.[45,46] Similarly, E2 may up-regulate the level of Wnt11 *via* an ER-dependent mechanism in breast cancer.[47] The induced Wnt offers the resistance of tumor cells to apoptosis and thus increases the survival.[48] Second, estrogen promotes cell proliferation *via* cell membrane-related but ER-independent phosphorylation of target molecules. This pathway is first shows in ER-negative breast cancer cells (MDA-MB-435 and MAD-MD-231).[49] Estrogen can activate Akt by inducing rapid phosphorylation at Ser(473) of this protein and this activation can be blocked by the inhibitors of PI3K and Src kinase but not by the ER antagonist ICI 182780 (fulvestrant).[49] A similar finding is also demonstrated in endometrial cancer cells, and neither ERE activation nor ICI 182780 can affect E2-mediated Akt or Erk1/2 activation and cell proliferation.[50,51] In another study, effects of E2 and E2-BSA that cannot enter the cell on protein kinase C (PKC) is investigated in ER-negative HCC38 breast cancer cells.[52] It shows that both E2 and E2-BSA can rapidly increase PKC *via* phosphatidylinositol-dependent phospholipase C and G protein and that the action is not affected by ER-antagonist ICI 182780, ER-agonist diethylstilbestrol, and antibodies to ERα and ERβ, indicating that E2-mediated PKC activity is *via* membrane pathways which is involving neither ERα nor ERβ. Interestingly, PKC activity is found to be positively correlated with the severity of breast cancer and such a correlation is even greater in ER-negative tumors.[52] Third, direct genotoxic effects of estrogen metabolites on DNA damage, mutation and cell transformation. It has been well known that some of estrogen metabolites are carcinogens due to their toxicity to DNA and the toxic effect is independent of ER. The toxicity is mainly related to the feature of reactive oxygen species in these metabolites. The details of this aspect are well summarized by three review articles published recently.[22,23,53]

The relationship and the balance between ERα and ERβ may greatly influence the development of tumors and the treatment. The early study has suggested that as normal breast tissues become

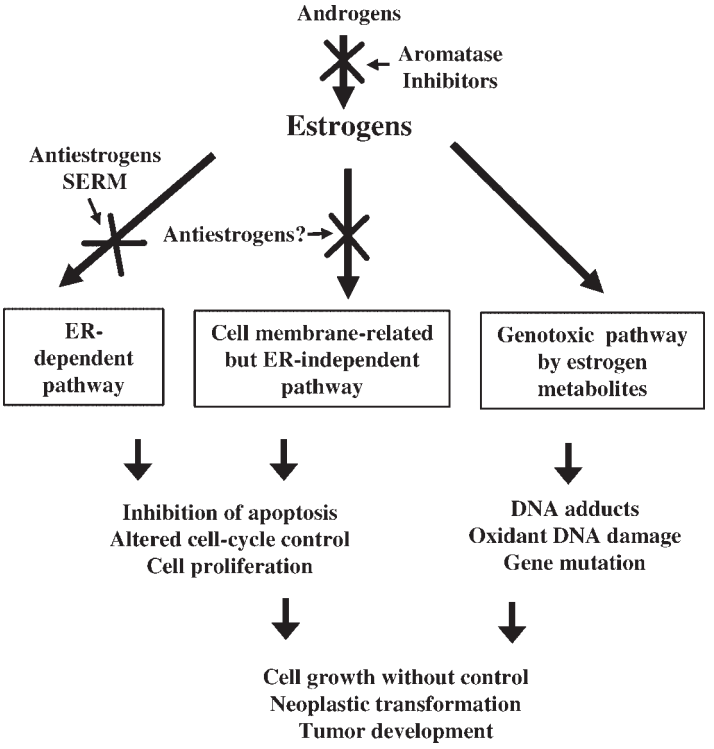

Figure 2. The carcinogenic pathways of estrogen.

tumorigenic, the amount of ERα RNA increases whereas the amount of ERβ decreases.[54] However, probably due to the reliability or accuracy of ERβ antibodies that were available in the market of the early 2000s, results of ERβ protein in tumors are inconsistent as to the prognostic value of ERβ, showing that ERβ is a good survival marker of breast cancer in some studies but a bad one in other reports.[2] Studies into the ERβ function in breast and gynecologic cancers are few until recently. A recent study of 512 breast cancer tissue samples reveals that 78% of the tumors are ERα positive and 50% are ERβ positive.[55] Functionally, the activation of ERα is associated with the proliferation and growth of tumor cells,[48,56–60] whereas the activation of ERβ promotes apoptosis, suppresses the malignant transformation and inhibits the growth of the tumor cells.[61–67] Over-expression of ERβ prevents establishment and growth of breast tumors in a subcutaneous xenograft mouse model.[62] Restoration of ERβ in ovarian cancer cells results in enhancement of apoptosis of tumor cells, and a strong inhibition of their proliferation and invasion.[65] E2-meditaed proliferation of endometrial cells is blocked by transfection with ERα antisense DNA[57,59] but not with ERβ antisense DNA.[59] The inhibitory effect of ERβ may associate with its ability to decrease the expression of c-myc, cyclin A, cyclin D1 and cyclin E and to increase the levels of p21(cip1) and p27(Kip1).[66,67] ERβ may also act as an ERβ antagonist to directly interact with ERα since ERβ has shown to negatively regulate the transactivation of ERα in breast cancer cells (MCF7).[68,69] The experiment on ER-knockout mice has also suggested a stimulatory role of ERα and an inhibitory effect of ERβ in the proliferation of different estrogen-target tissues.[27,70,71] Taken together, the functional experiments have demonstrated that ERα and ERβ have completely different roles in the breast and gynecologic cancers, in which ERα functions as a tumor promoter whereas ERβ as a tumor suppressor. This concept is in agreement with the clinical values of ERβ detected in tumor tissues. The presence of ERβ in breast, endometrial and ovarian tumors is associated with better prognosis or a longer disease-free survival,[55,69,72–75] and the level of ERβ is significantly decreased in higher grade endometrial

cancers.[76] Therefore, ERβ is not a surrogate for ERα in breast and gynecologic cancers. ERα and ERβ can interact with each other and they both have their own pathological and clinical relevance in tumor progression and prognosis. The activity or the level of ERs, especially ERα can be also regulated by hypermethylation/hypomethylation and phosohorylation. The hypomethylation or the phosohorylation of ERα may lead to resistance of breast cancer to antiestrogen treatments. The details of the aspect has been recently reviewed.[77,78]

The different functions of ERα and ERβ may complicate the carcinogenic aspects of estrogen *via* above-mentioned three pathways that can act in an additive or synergistic fashion to induce tumors and promote their growth. Such multiple channels and different layers of complexity in estrogen carcinogenesis have required anti-tumor treatments to be finely tuned and more specifically targeted. The treatment established earlier such as tamoxifen to target ER non-selectively appears to be not specific enough to block the carcinogenic effect of estrogen in a given organ (Fig. 2). Pure antiestrogens such as ICI 182780 that bind the receptor to impair ER dimerization are also limited in its efficacy and specificity. Recently, ER-selective agonists or antagonists such as DPN (an ERβ agonist and MPP (an ERα antagonist) have been shown to inhibit the cell proliferation or to enhance apoptosis.[79,80] The ERα-selective antagonist or ERβ-selective agonist may have potential to be more specific in target tumor cells. Aromatase inhibitors that completely erase estrogen seem to be the most effective therapeutic agents since it blocks all three proposed processes (Fig. 2). However, estrogen-deprivation therapy may associate with potential serious side-effects as estrogens in physiological concentrations are required for numerous functions of a cell. Therefore, the usefulness of this therapeutic strategy remains to be further evaluated.

B. Endocrine Gland Cancers (Adrenocortical, Pancreatic, Prostate, and Thyroid)

There is ample evidence to support that estrogens play a role in the development of endocrine gland cancers. Epidemically, the incidence of thyroid cancer is three to five times more frequent in woman than in men and the gender difference in the incidence is particularly obvious for women of reproductive age relative to men.[81–86] The use of oral contraceptives or female hormone therapy appears to result in a moderately increased risk of developing thyroid cancer or pancreatic cancer.[87–92] Although prostate cancer only occurs in men, the estrogenic factor is still evident. Prostate cancer develops in older men at a time when the level of serum testosterone is in decline, and the level of estrogen remains unchanged or increases with age.[93] The net result is a significant increase the ratio of testosterone to estrogen allowing the regulation of prostate gland to be shifted pre-dominately under the control of estrogen. African-American men have a twofold increased risk of prostate cancer as compared to their Caucasian counterparts and this increased incidence is correlated with a higher level of estrogens in the former than in the latter.[94] There is not relevant information about adrenocortical cancer probably due to this tumor is very rare. Animal studies agree with the epidemiological data and suggest that exogenous E2 may promote thyroid tumors. Ovary intact rats with the highest level of bioavailable E2 has the highest incidence of thyroid tumor development, and the ovariectomized rats with reduced concentrations of bioavailable E2 show a decrease in the incidence of thyroid tumors.[95] Importantly, when ovariectomized rats are supplemented with E2, they develop a higher incidence of thyroid tumors than ovariectomized rats that are not supplemented with E2.[95,96] The prostate of the adult male animals (rats, dogs, and monkeys) can develop carcinogenic features when the animal is treated with estrogen together with testosterone,[97–99] and aromatase knockout mice, which are estrogen deficient but have an increase in androgens, do not develop prostate cancer.[100] Similar to breast and gynecologic cancers, most endocrine gland tumors are now known to be able to biosynthesize estrogen in a paracrine manner since these malignant tissues can express a high level of aromatase.[98,101–105]

Estrogen may carry out its carcinogenic function in endocrine gland tumors *via* the three channels described in breast and gynecologic cancers. However, few studies have been performed to

investigate the genotoxic role of estrogens as well as its phosphorylation function in endocrine gland tumors, leaving these two possible pathways unproved. Estrogen-mediated genomic and non-genomic pathways in endocrine gland tumors have been well examined and the study has been extensively focused on the shift in balance between ERα and ERβ in tumor cells with a decreased level of ERβ. In prostate cancer, E2-mediated the activation of ERα is associated with aberrant proliferation, inflammation and the development of malignancy, whereas E2-mediated the activation of ERβ is associated with anti-proliferation, differentiation and apoptosis.[97,99,106–108] The importance of ERα rather than ERβ during the hormonal induction of prostate cancer is well demonstrated in the following animal study though it has not been proved in human. Pre-malignant prostatic intraepithelial neoplasia (PIN) lesions observed in the mouse model of prostate tumors induced by testosterone and E2 are characterized by significantly increased ERα expression within the lesion itself, and interestingly, PIN lesions can be induced in ERβ knockout but not ERα knockout mice.[99] There is also strong evidence to support the inhibitory function of ERβ in prostate tumors. Knockdown of ERβ by its siRNA in prostate cells increases the expression of genes highly relevant to tumor cell proliferation such as prostate-derived Ets factor, the catalytic subunit of the telomerase, and ERα. In contrast, the up-regulation of ERβ by histone deacetylase inhibitor valproic acid (VPA) or tectorigenin results in antiproliferative effects by down-regulation of these genes.[106] Therefore, the loss or decreased ERβ has been consistently associated with the progression of prostate tumors.[97,99] The decreased level of ERβ may attribute to the apoptosis of ERb cells which have been shown to have increased levels of Bax, poly(ADP-ribose) polymerase and caspase-3.[108]

In thyroid cancer, the functions of ERα and ERβ mimic that in prostate cancer. The expression of ERα appears to be increased in thyroid tumor tissues while ERβ is frequently undetected.[109] E2 can promote the proliferation of thyroid cells and this effect is positively related to ERα but negatively to ERβ, as ERα agonist PPT enhances cell proliferation whereas ERβ agonist DPN inhibits it.[110] The E2-mediated cell proliferation is associated with a rapid (within minutes) increase in the phosphorylation of Erk1/2 and subsequently a reduced Bcl-2 but a decreased Bax level. The knockdown of ERα significantly attenuated the E2-mediated Bcl-2 and pERK1/2 expression. In contrast, the knockdown of ERβ markedly enhanced them.[110] The E2-mediated regulation of Erk1/2 and Bcl-2 family molecules is believed to be *via* both genomic and non-genomic pathways since its effects can be achieved in minutes and also maintained for a few days.[110] The inhibitory role of ERα is re-inforced using ERα selective antagonist MPP, which can effectively block E2-mediated proliferation to a similar level by ICI182780.[110,111] The pro-proliferative function of ERα and the anti-proliferative function of ERβ have been further evident in another study using an adnoviral vector carrying ERα (Ad-ERα) or ERβ (Ad-ERβ). Ad-ERalpha infection stimulates thyroid cancer cell growth, in contrast, Ad-ERbeta infection suppresses their growth by inducing apoptosis.[112] Furthermore, estrogen and anti-estrogen suppresses AP1 activity in Ad-ERalpha-infected cells, whereas upon Ad-ERbeta infection estrogen further stimulates AP1 activity which in turn is suppressed by anti-estrogen, suggesting that each ER acts differently through a non-ERE-mediated pathway.[112]

In pancreatic cancer, E2 has been shown to stimulate the growth of tumor cells probably in a ER-related pathway.[102] The expression of ERα mRNA is increased whereas ERβ is significantly decreased in the advantage of the tumor,[113] suggesting a positive role of ERα but a negative role for ERβ in pancreatic cancer. In adrenocortical cancer, E2 enhances the proliferation of cancer cells.[101] Compared with the normal adrenal cortex and adrenocortical adenomas, carcinomas are characterized by significantly lower ERβ levels, ERα upregulation, and aromatase overexpression.[101]

Taken together, the above findings demonstrate the opposite function of ERα and ERβ in the tumors of endocrine system and most of these tumors have been shown to express a high level of pro-proliferative ERα but a low level of anti-proliferative ERβ. These findings raise the possibility of targeting ERα or stimulating ERβ as a possible therapy. In this aspect, the selective ERα antagonist Toremifene has been used to reduce the incidence of prostate cancer in men with high grade prostatic

intraepithelial neoplasia, showing that Toremifene decreases the incidence of prostate cancer by 1 year.[114] In the strategy to activate ERβ, ERβ selective agonist DPN has successfully reduced the proliferation of prostate cancer cells and thyroid cancer cells,[80,110] suggesting that ERβ specific agonists might be valid candidates for new pharmacological approaches against tumors of endocrine system.

C. Lung Cancer and Cancers of Digestive System (Colorectal, Esophageal, and Liver)

The role of estrogen in lung cancer and digestive cancers (colorectal, esophageal, and liver) appears to be different from the other two categories described in the previous sections. The protective role of estrogen in lung cancer and digestive cancers is strongly suggested by the epidemiological observations. The current HRT users shows approximately 30% reduced incidence of colorectal cancers [115] and similar observations have been reported by a number of other reports.[34,116–121] The mechanism of protection may associate with selective regulation of apoptotic genes in colon cancer cells by E2.[119] The E2-protective role is also noted in liver, esophageal, and lung cancers.[35,122–124]

In experiments, E2 induces colon cancer cell apoptosis in an ERα-related pathway.[125] It increases hTNF-alpha gene expression, which in turn activates caspase-8, -9, and caspase-3 and leads to the DNA fragmentation and apoptosis. In the other hand, E2 plus ERalpha down-regulates beta-catenin signalings to suppress proliferation and metastasis of colorectal cells.[125] The study suggests that efforts aiming at enhancing ERα level or activity may be an alternative therapy against colorectal cancer.[125] The finding is somewhat supported by the fact that ERα is either reduced or undetected in colorectal cancer cells.[126–128] However, the activation of ERβ has also shown to be associated with the inhibition of colon cancer.[127] The relationship between ERα and ERβ in colorectal cancer remains unknown and the exact function of ERα and ERβ in colorectal cancers also needs further experiments to confirm. However, it appears that the function of ERs in colorectal cancers differs from that in breast cancer, gynecological and endocrine cancers. Similar to colorectal cancer, E2 also inhibits the growth of esophageal cancer cells in an ERα-dependent pathway since the inhibitory effect is lost in ERα-negative esophageal cancer cells.[129,130] Though esophageal cancer cells also express ERβ,[131] the role of ERβ is unknown. However, positive expression of ERα in addition to negative expression of ERβ is an unfavorable independent prognostic indicator in squamous cell carcinoma of the esophagus.[132] There are very limited reports on the function of ERα and ERβ in hepatocellular carcinoma (HCC) and their role remain unclear, though HCC cells are known to express both.[133] There is a study indicating that the presence of variant liver ER transcripts in the tumor is the strongest negative predictor of survival in inoperable HCC and their presence is associated with spontaneous survival significantly worse than in patients with wild-type ERs.[134]

Non-small cell lung cancer cells (NSLCC) express both ERα and ERβ in the nucleus as well as extra-nuclear sites,[135] and the block of either of them by their siRNA can results in a significant reduction in the proliferation of cells.[136] The finding is supported by application of ER antagonist ICI 182780 leading to the inhibition of NSLCC proliferation.[137] Therefore, the functional role of ERα and ERβ seems to benefit the growth of the tumor since the inhibition or block of both leads to the arrest the tumor growth. The functional relationship between both ERs is complicated by the observations that ERα expression and the absence of ERβ expression are associated with a poorer prognosis for NSCLC patients, and that the absence of ERβ serves as a marker identifying patients at high risk even at an early clinical stage.[138] Obviously, further studies need to clarify this point as well as the signaling pathway of E2 in lung cancer.

3. FUTURE PROSPECTS

A better understanding of the molecular mechanisms by which estrogen stimulates cell growth can provide new insights into diagnosis, treatment and prevention in estrogen-associated tumors. Loss of

estrogen or its receptors contributes to the development or progression of various tumors. Both activation (*via* estrogen agonists) and inhibition (*via* estrogen antagonists) of ER action are therapeutic strategies currently used in the clinical setting. ER antagonists, SERM and aromatase inhibitors are effective for the treatment of breast cancer and endometrial cancer, and their usefulness and efficacy in the treatment of other hormone-dependent cancers such as prostate cancer and thyroid cancer awaits further study. At the same time, other novel strategies to selectively target or stimulate ERα or ERβ may appear to be more effective for certain tumors such as prostate and thyroid cancers. Since both the genomic and non-genomic responses of ERα and ERβ can be exquisitely coordinated and be co-functional in physiological and pathological conditions, it will be certainly a great advantage if a treatment can specifically block adverse effects of ERs but leave beneficial effects of ERs intact. Nevertheless, any single therapy or therapies in combination should be thought to optimally induce apoptosis or death of cancer cells without damaging healthy cells. Clearly, estrogen and its receptors have been implicated in the pathogenesis of several cancers but their definitive role has yet to be fully established, especially in lung cancer and cancer of digestive system. Understanding the role that estrogen and its receptors may play in the risk or severity of the tumor will no doubt increase our ever-expanding knowledge of the relationship among estrogen, ERs, and cancers.

4. ABBREVIATIONS

16-LE2	3,17-dihydroxy-19-nor-17-pregna-1,3,5(10)-triene-21,16-lactone
8β-VE2	8-vinylestra-1,3,5(10)-triene-3,17β-diol
Ad-ERα	adnoviral vector carrying ERα
Ad-ERβ	adnoviral vector carrying ERβ
AF1	activation function-1
AP-1	activator protein 1
DPN	diarylpropionitrile
E2	17β-estradiol
E3	estriol
EGFR	epidermal growth factor receptor
ERB-041	2-(3-fluoro-4-hydroxyphenyl)-7-vinyl-1,3-benzoxazol-5-ol
ERE	estrogen response element
ERs	estrogen receptors
ERT	estrogen-replacement therapy
ERα46	46 kDa isoform
ERα66	66 kDa form of ERα
ERβδ125	ERβ1 exons 1, 2, and 5
ERβδ1256	ERβ1 exons 1, 2, 5, and 6
Fox	forkhead box
GPR30	G protein-coupled receptor-30
GRIP1	glucocorticoid receptor-interacting protein 1
HCC	hepatocellular carcinoma
HPA	hypothalamic-pituitary-adrenal
HRT	hormone-replacement therapy
MAPK	mitogen-activated protein kinase
NF-*k*B	nuclear factor *kappa*B
NR	nuclear receptor
NSLCC	non-small cell lung cancer cells
OC	oral contraceptives

PI3K	phosphoinositide 3-kinase
PIN	prostatic intraepithelial neoplasia
PKA	protein kinase A
PKC	protein kinase C
PPT	propylpyrazole triol
SERM	selective ER modulators
VPA	valproic acid
WAY-200070	7-bromo-2-(4-hydroxyphenyl)-1,3-benzoxazol-5-ol
WAY-202196	3-(3-fluoro-4-hydroxy-phenyl)-7-hydroxy-naphthonitrile

ACKNOWLEDGMENTS

The authors thank Ms. Christina Lou for her skilled assistance with the illustrations and manuscript preparation. The work from the authors' laboratory is supported by CUHK direct grant; grant number: 2041163, and the Research Grants Council of the Hong Kong Special Administrative Region; grants number: CUHK4390/03M and CUHK4556/05M.

REFERENCES

1. Ascenzi P, Bocedi A, Marino M. Structure-function relationship of estrogen receptor alpha and beta: Impact on human health. Mol Aspects Med 2006;27:299–402.
2. Pearce ST, Jordan VC. The biological role of estrogen receptors alpha and beta in cancer. Crit Rev Oncol Hematol 2004;50:3–22.
3. Ruggiero RJ, Likis FE. Estrogen: Physiology, pharmacology, and formulations for replacement therapy. J Midwifery Women's Health 2002;47:130–138.
4. Pietras RJ, Márquez-Garbán DC. Membrane-associated estrogen receptor signaling pathways in human cancers. Clin Cancer Res 2007;13:4672–4676.
5. Cvoro A, Tzagarakis-Foster C, Tatomer D, Paruthiyil S, Fox MS, Leitman DC. Distinct roles of unliganded and liganded estrogen receptors in transcriptional repression. Mol Cell 2006;21:555–564.
6. Carroll JS, Brown M. Estrogen receptor target gene: An evolving concept. Mol Endocrinol 2006;20:1707–1714.
7. Eeckhoute J, Keeton EK, Lupien M, Krum SA, Carroll JS, Brown M. Positive cross-regulatory loop ties GATA-3 to estrogen receptor alpha expression in breast cancer. Cancer Res 2007;67:6477–6483.
8. Levin ER. Integration of the extranuclear and nuclear actions of estrogen. Mol Endocrinol 2005;19:1951–1959.
9. Björnström L, Sjöberg M. Estrogen receptor-dependent activation of AP-1 via non-genomic signalling. Nucl Recept 2004;2:3.
10. Cascio S, Bartella V, Garofalo C, Russo A, Giordano A, Surmacz E. Insulin-like growth factor 1 differentially regulates estrogen receptor-dependent transcription at estrogen response element and AP-1 sites in breast cancer cells. J Biol Chem 2007;282:3498–3506.
11. Prossnitz ER, Arterburn JB, Smith HO, Oprea TI, Sklar LA, Hathaway HJ. Estrogen signaling through the transmembrane G protein-coupled receptor G PR30. Annu Rev Physiol 2008;70:165–190.
12. Filardo EJ, Quinn JA, Sabo E. Association of the membrane estrogen receptor, GPR30, with breast tumor metastasis and transactivation of the epidermal growth factor receptor. Steroids (in press).
13. Thomas P, Pang Y, Filardo EJ, Dong J. Identity of an estrogen membrane receptor coupled to a G protein in human breast cancer cells. Endocrinology 2005;146:624–632.
14. Pedram A, Razandi M, Levin ER. Nature of functional estrogen receptors at the plasma membrane. Mol Endocrinol 2006;20:1996–2009.
15. Ruff M, Gangloff M, Wurtz JM, Moras D. Estrogen receptor transcription and transactivation: Structure-function relationship in DNA- and ligand-binding domains of estrogen receptors. Breast Cancer Res 2000;2:353–359.

16. Morello KC, Wurz GT, DeGregorio MW. Pharmacokinetics of selective estrogen receptor modulators. Clin Pharmacokinet 2003;42:361–372.
17. Baumann CK, Castiglione-Gertsch M. Estrogen receptor modulators and down regulators: Optimal use in postmenopausal women with breast cancer. Drugs 2007;67:2335–2353.
18. Jordan VC, O'Malley BW. Selective estrogen-receptor modulators and antihormonal resistance in breast cancer. J Clin Oncol 2007;25:5815–5824.
19. Kuiper GG, Lemmen JG, Carlsson B, Corton CJ, Safe SH, van der Saag PT, van der Burg B, Gustafsson J-A. Interaction of estrogenic chemicals and phytoestrogens with estrogen receptor beta. Endocrinology 1998;139:4252–4263.
20. Harris HA. Estrogen receptor-beta: Recent lessons from in vivo studies. Mol Endocrinol 2007;21:1–13.
21. Bocchinfuso WP, Korach KS. Mammary gland development and tumorigenesis in estrogen receptor knockout mice. J Mammary Gland Biol Neoplasia 1997;2:323–334.
22. Yager JD, Davidson NE. Estrogen carcinogenesis in breast cancer. N Engl J Med 2006;354:270–282.
23. Russo J, Russo IH. The role of estrogen in the initiation of breast cancer. J Steroid Biochem Mol Biol 2006;102:89–96.
24. Gligorov J, Pritchard K, Goss P. Adjuvant and extended adjuvant use of aromatase inhibitors: Reducing the risk of recurrence and distant metastasis. Breast 2007;16(Suppl 3):S1–S9.
25. Kendall A, Dowsett M. Novel concepts for the chemoprevention of breast cancer through aromatase inhibition. Endocr Relat Cancer 2006;13:827–837.
26. Flouriot G, Brand H, Denger S, Metivier R, Kos M, Reid G, Sonntag-Buck V, Gannon F. Identification of a new isoform of the human estrogen receptor-alpha (hER-alpha) that is encoded by distinct transcripts and that is able to repress hER-alpha activation function 1. EMBO J 2000;19:4688–4700.
27. Fuqua SA, Wolf DM. Molecular aspects of estrogen receptor variants in breast cancer. Breast Cancer Res Treat 1995;35:233–241.
28. Price RH Jr, Butler CA, Webb P, Uht R, Kushner P, Handa RJ. A splice variant of estrogen receptor beta missing exon 3 displays altered subnuclear localization and capacity for transcriptional activation. Endocrinology 2001;142:2039–2049.
29. Métivier R, Penot G, Carmouche RP, Hübner MR, Reid G, Denger S, Manu D, Brand H, Kos M, Benes V, Gannon F. Transcriptional complexes engaged by apo-estrogen receptor-alpha isoforms have divergent outcomes. EMBO J 2004;23:3653–3666.
30. Treeck O, Juhasz-Boess I, Lattrich C, Horn F, Goerse R, Ortmann O. Effects of exon-deleted estrogen receptor beta transcript variants on growth, apoptosis and gene expression of human breast cancer cell lines. Breast Cancer Res Treat (in press).
31. Heldring N, Pike A, Andersson S, Matthews J, Cheng G, Hartman J, Tujague M, Ström A, Treuter E, Warner M, Gustafsson JA. Estrogen receptors: How do they signal and what are their targets. Physiol Rev 2007;87:905–931.
32. Willey SC, Cocilovo C. Screening and follow-up of the patient at high risk for breast cancer. Obstet Gynecol 2007;110:1404–1416.
33. Mourits MJ, De Bock GH. Exogenous steroids for menopausal symptoms and breast/endometrial cancer risk. Int J Gynecol Cancer 2006;16(Suppl 2):494–496.
34. Anonymous. Hormone replacement therapy and cancer. Gynecol Endocrinol 2001;15:453–465.
35. Fernandez E, Gallus S, Bosetti C, Franceschi S, Negri E, La Vecchia C. Hormone replacement therapy and cancer risk: A systematic analysis from a network of case-control studies. Int J Cancer 2003;105:408–412.
36. Christov K, Grubbs CJ, Shilkaitis A, Juliana MM, Lubet RA. Short-term modulation of cell proliferation and apoptosis and preventive/therapeutic efficacy of various agents in a mammary cancer model. Clin Cancer Res 2007;13:5488–5496.
37. Brodie A, Sabnis G, Jelovac D. Aromatase and breast cancer. J Steroid Biochem Mol Biol 2006;102:97–102.
38. Kutza J, Smith E, Levy B, Jian J, Haugen T, Turek L. Use of hormone replacement therapy (Hrt) And detection of human papillomavirus (Hpv) DNA in postmenopausal women. Ann Epidemiol 2000;10:465–466.
39. Nair HB, Luthra R, Kirma N, Liu YG, Flowers L, Evans D, Tekmal RR. Induction of aromatase expression in cervical carcinomas: Effects of endogenous estrogen on cervical cancer cell proliferation. Cancer Res 2005;65:11164–11173.
40. Fowler JM, Ramirez N, Cohn DE, Kelbick N, Pavelka J, Ben-Shachar I, Morrison C. Correlation of cyclooxygenase-2 (COX-2) and aromatase expression in human endometrial cancer: Tissue microarray analysis. Am J Obstet Gynecol 2005;192:1262–1271.

41. Li YF, Hu W, Fu SQ, Li JD, Liu JH, Kavanagh JJ. Aromatase inhibitors in ovarian cancer: Is there a role? Int J Gynecol Cancer (in press).
42. Krasner C. Aromatase inhibitors in gynecologic cancers. J Steroid Biochem Mol Biol 2007;106:76–80.
43. Bulun SE, Chen D, Lu M, Zhao H, Cheng Y, Demura M, Yilmaz B, Martin R, Utsunomiya H, Thung S, Su E, Marsh E, Hakim A, Yin P, Ishikawa H, Amin S, Imir G, Gurates B, Attar E, Reierstad S, Innes J, Lin Z. Aromatase excess in cancers of breast, endometrium and ovary. J Steroid Biochem Mol Biol 2007;106:81–96.
44. Segawa T, Shozu M, Murakami K, Kasai T, Shinohara K, Nomura K, Ohno S, Inoue M. Aromatase expression in stromal cells of endometrioid endometrial cancer correlates with poor survival. Clin Cancer Res 2005;11:2188–2194.
45. Han WD, Si YL, Zhao YL, Li Q, Wu ZQ, Hao HJ, Song HJ. GC-rich promoter elements maximally confers estrogen-induced transactivation of LRP16 gene through ERalpha/Sp1 interaction in MCF-7 cells. J Steroid Biochem Mol Biol 2008;109:47–56.
46. Meng YG, Han WD, Zhao YL, Huang K, Si YL, Wu ZQ, Mu YM. Induction of the LRP16 gene by estrogen promotes the invasive growth of Ishikawa human endometrial cancer cells through the downregulation of E-cadherin. Cell Res 2007;17:869–880.
47. Vendrell JA, Ghayad S, Ben-Larbi S, Dumontet C, Mechti N, Cohen PA. A20/TNFAIP3, a new estrogen-regulated gene that confers tamoxifen resistance in breast cancer cells. Oncogene 2007;26:4656–4667.
48. Lin Z, Reierstad S, Huang CC, Bulun SE. Novel estrogen receptor-alpha binding sites and estradiol target genes identified by chromatin immunoprecipitation cloning in breast cancer. Cancer Res 2007;67:5017–5024.
49. Tsai EM, Wang SC, Lee JN, Hung MC. Akt activation by estrogen in estrogen receptor-negative breast cancer cells. Cancer Res 2001;61:8390–8392.
50. Treeck O, Diedrich K, Ortmann O. The activation of an extracellular signal-regulated kinase by oestradiol interferes with the effects of trastuzumab on HER2 signalling in endometrial adenocarcinoma cell lines. Eur J Cancer 2003;39:1302–1309.
51. Guo RX, Wei LH, Tu Z, Sun PM, Wang JL, Zhao D, Li XP, Tang JM. 17 beta-estradiol activates PI3K/Akt signaling pathway by estrogen receptor (ER)-dependent and ER-independent mechanisms in endometrial cancer cells. J Steroid Biochem Mol Biol 2006;99:9–18.
52. Boyan BD, Sylvia VL, Frambach T, Lohmann CH, Dietl J, Dean DD, Schwartz Z. Estrogen-dependent rapid activation of protein kinase C in estrogen receptor-positive MCF-7 breast cancer cells and estrogen receptor-negative HCC38 cells is membrane-mediated and inhibited by tamoxifen. Endocrinology 2003;144:1812–1824.
53. Mueck AO, Seeger H. Breast cancer: Are estrogen metabolites carcinogenic? Climacteric 2007;10(Suppl 2):62–65.
54. Leygue E, Dotzlaw H, Watson PH, Murphy LC. Altered estrogen receptor alpha and beta messenger RNA expression during human breast tumorigenesis. Cancer Res 1998;58:3197–3201.
55. Borgquist S, Holm C, Stendahl M, Anagnostaki L, Landberg G, Jirstrom K. Oestrogen receptors alpha and beta show different associations to clinicopathological parameters and their co-expression might predict a better response to endocrine treatment in breast cancer. J Clin Pathol 2008;61:197–203.
56. Au WW, Abdou-Salama S, Al-Hendy A. Inhibition of growth of cervical cancer cells using a dominant negative estrogen receptor gene. Gynecol Oncol 2007;104:276–280.
57. Taylor AH, al-Azzawi F, Pringle JH, Bell SC. Inhibition of endometrial carcinoma cell growth using antisense estrogen receptor oligodeoxyribonucleotides. Anticancer Res 2002;22:3993–4003.
58. Boggess JF, Zhou C, Bae-Jump VL, Gehrig PA, Whang YE. Estrogen-receptor-dependent regulation of telomerase activity in human endometrial cancer cell lines. Gynecol Oncol 2006;103:417–424.
59. Zhang Y, Liao Q, Chen C, Yu L, Zhao J. Function of estrogen receptor isoforms alpha and beta in endometrial carcinoma cells. Int J Gynecol Cancer 2006;16:1656–1660.
60. Zhang Y, Liao Q, Chen C, Yu L, Zhao J. Function of estrogen receptor isoforms alpha and beta in endometrial carcinoma cells. Int J Gynecol Cancer 2006;16:1656–1660.
61. Lin CY, Strom A, Li Kong S, Kietz S, Thomsen JS, Tee JB, Vega VB, Miller LD, Smeds J, Bergh J, Gustafsson JA, Liu ET. Inhibitory effects of estrogen receptor beta on specific hormone-responsive gene expression and association with disease outcome in primary breast cancer. Breast Cancer Res 2007;9:R25.
62. Behrens D, Gill JH, Fichtner I. Loss of tumourigenicity of stably ERbeta-transfected MCF-7 breast cancer cells. Mol Cell Endocrinol 2007;274:19–29.
63. Treeck O, Pfeiler G, Mitter D, Lattrich C, Piendl G, Ortmann O. Estrogen receptor {beta}1 exerts antitumoral effects on SK-OV-3 ovarian cancer cells. J Endocrinol 2007;193:421–433.

64. Treeck O, Pfeiler G, Horn F, Federhofer B, Houlihan H, Vollmer A, Ortmann O. Novel estrogen receptor beta transcript variants identified in human breast cancer cells affect cell growth and apoptosis of COS-1 cells. Mol Cell Endocrinol 2007;264:50–60.
65. Lazennec G. Estrogen receptor beta, a possible tumor suppressor involved in ovarian carcinogenesis. Cancer Lett 2006;231:151–157.
66. Paruthiyil S, Parmar H, Kerekatte V, Cunha GR, Firestone GL, Leitman DC. Estrogen receptor beta inhibits human breast cancer cell proliferation and tumor formation by causing a G2 cell cycle arrest. Cancer Res 2004;64:423–428.
67. Ström A, Hartman J, Foster JS, Kietz S, Wimalasena J, Gustafsson JA. Estrogen receptor beta inhibits 17beta-estradiol-stimulated proliferation of the breast cancer cell line T47D. Proc Natl Acad Sci USA 2004;101:1566–1571.
68. Zhao C, Matthews J, Tujague M, Wan J, Strom A, Toresson G, Lam EW, Cheng G, Gustafsson JA, Dahlman-Wright K. Estrogen receptor beta2 negatively regulates the transactivation of estrogen receptor alpha in human breast cancer cells. Cancer Res 2007;67:3955–3962.
69. Macaluso M, Montanari M, Noto PB, Gregorio V, Surmacz E, Giordano A. Nuclear and cytoplasmic interaction of pRb2/p130 and ER-beta in MCF-7 breast cancer cells. Ann Oncol 2006;17(Suppl 7):vii27–vii29.
70. Dupont S, Krust A, Gansmuller A, Dierich A, Chambon P, Mark M. Effect of single and compound knockouts of estrogen receptors a and b on mouse reproductive phenotypes. Development 2000;127:4277–4291.
71. Fuqua SA, Schiff R, Parra I, Moore JT, Mohsin SK, Osborne CK, Clark GM, Allred DC. Estrogen receptor beta protein in human breast cancer: Correlation with clinical tumor parameters. Cancer Res 2003;63:2434–2439.
72. Sugiura H, Toyama T, Hara Y, Zhang Z, Kobayashi S, Fujii Y, Iwase H, Yamashita H. Expression of estrogen receptor beta wild-type and its variant ERbetacx/beta2 is correlated with better prognosis in breast cancer. Jpn J Clin Oncol 2007;37:820–828.
73. Shupnik MA. Estrogen receptor-beta: Why may it influence clinical outcome in estrogen receptor-alpha positive breast cancer? Breast Cancer Res 2007;9:107.
74. Gruvberger-Saal SK, Bendahl PO, Saal LH, Laakso M, Hegardt C, Eden P, Peterson C, Malmstrom P, Isola J, Borg A, Ferno M. Estrogen receptor beta expression is associated with tamoxifen response in ERalpha-negative breast carcinoma. Clin Cancer Res 2007;13:1987–1994.
75. Chan KK, Wei N, Liu SS, Xiao-Yun L, Cheung AN, Ngan HY. Estrogen receptor subtypes in ovarian cancer: A clinical correlation. Obstet Gynecol 2008;111:144–151.
76. Chakravarty D, Srinivasan R, Ghosh S, Gopalan S, Rajwanshi A, Majumdar S. Estrogen receptor beta1 and the beta2/betacx isoforms in nonneoplastic endometrium and in endometrioid carcinoma. Int J Gynecol Cancer 2007;17:905–913.
77. Murphy LC, Weitsman GE, Skliris GP, Teh EM, Li L, Peng B, Davie JR, Ung K, Niu YL, Troup S, Tomes L, Watson PH. Potential role of estrogen receptor alpha (ERalpha) phosphorylated at Serine118 in human breast cancer in vivo. J Steroid Biochem Mol Biol 2006;102:139–146.
78. Stearns V, Zhou Q, Davidson NE. Epigenetic regulation as a new target for breast cancer therapy. Cancer Invest 2007;25:659–665.
79. Davis AM, Ellersieck MR, Grimm KM, Rosenfeld CS. The effects of the selective estrogen receptor modulators, methyl-piperidino-pyrazole (MPP), and raloxifene in normal and cancerous endometrial cell lines and in the murine uterus. Mol Reprod Dev 2006;73:1034–1044.
80. Pravettoni A, Mornati O, Martini PG, Marino M, Colciago A, Celotti F, Motta M, Negri-Cesi P. Estrogen receptor beta (ERbeta) and inhibition of prostate cancer cell proliferation: Studies on the possible mechanism of action in DU145 cells. Mol Cell Endocrinol 2007;263:46–54.
81. Correa P, Chen VW. Endocrine gland cancer. Cancer 1995;75(S1):338–352.
82. Libutti SK. Understanding the role of gender in the incidence of thyroid cancer. Cancer J 2005;11:104–105.
83. Smailyte G, Miseikyte-Kaubriene E, Kurtinaitis J. Increasing thyroid cancer incidence in Lithuania in 1978-2003. BMC Cancer 2006;6:284–289.
84. Scheiden R, Keipes M, Bock C, Dippel W, Kieffer N, Capesius C. Thyroid cancer in Luxembourg: A national population-based data report (1983-1999). BMC Cancer 2006;6:102–111.
85. Machens A, Hauptmann S, Dralle H. Disparities between male and female patients with thyroid cancers: Sex difference or gender divide? Clin Endocrinol 2006;65:500–505.
86. Ward LS, Assumpcao LV. The impact of gender in differentiated thyroid cancer. Clin Endocrinol 2007;66:752.
87. Ron E, Kleinerman RA, Boice JD Jr, Livolsi VA, Flannery JT, Fraumeni JF. A population-based case-control study of thyroid cancer. J Natl Cancer Inst 1987;79:1–112.

88. Preston-Martin S, Bernstein L, Pike MC, Maldonado AA, Henderson BE. Thyroid cancer among young women related to prior thyroid disease and pregnancy history. Br J Cancer 1987;55:191–195.
89. McTiernan AM, Weiss NS, Daling JR. Incidence of thyroid cancer in women in relation to previous exposure to radiation therapy and history of thyroid disease. J Natl Cancer Inst 1984;73:575–581.
90. Persson I, Yuen J, Bergkvist L, Schairer C. Cancer incidence and mortality in women receiving estrogen and estrogen-progestin replacement therapy—Long-term follow-up of a Swedish cohort. Int J Cancer 1996;67:327–332.
91. Fernandez E, La Vecchia C, D'Avanzo B, Negri E. Menstrual and reproductive factors and pancreatic cancer risk in women. Int J Cancer 1995;62:11–14.
92. Bueno de Mesquita HB, Maisonneuve P, Moerman CJ, Walker AM. Anthropometric and reproductive variables and exocrine carcinoma of the pancreas: A population-based case-control study in The Netherlands. Int J Cancer 1992;52:24–29.
93. Kaufman JM, Vermeulen A. The decline of androgen levels in elderly men and its clinical and therapeutic implications. Endocr Rev 2005;26:833–876.
94. Rohrmann S, Nelson WG, Rifai N, Brown TR, Dobs A, Kanarek N, Yager JD, Platz EA. Serum estrogen, but not testosterone, levels differ between black and white men in a nationally representative sample of Americans. Clin Endocrinol Metab 2007;92:2519–2525.
95. Thiruvengadam A, Govindarajulu P, Aruldhas MM. Modulatory effect of estradiol and testosterone on the development of N-nitrosodiisopropanolamine induced thyroid tumors in female rats. Endocrine Res 2003;29:43–51.
96. Mori M, Naito M, Watanabe H, Takeichi N, Dohi K, Ito A. Effects of sex difference, gonadectomy, and estrogen on N-methyl-N-nitrosourea induced rat thyroid tumors. Cancer Res 1990;50:7662–7667.
97. Carruba G. Estrogen and prostate cancer: An eclipsed truth in an androgen-dominated scenario. J Cell Biochem 2007;102:899–911.
98. Prins GS, Korach KS. The role of estrogens and estrogen receptors in normal prostate growth and disease. Steroids 2008;73:233–244.
99. Risbridger GP, Ellem SJ, McPherson SJ. Estrogen action on the prostate gland: a critical mix of endocrine and paracrine signaling. J Mol Endocrinol 2007;39:183–188.
100. McPherson SJ, Wang H, Jones ME, Pedersen J, Iismaa TP, Wreford N, Simpson ER, Risbridger GP. Elevated androgens and prolactin in aromatase-deficient mice cause enlargement, but not malignancy, of the prostate gland. Endocrinology 2001;142:2458–2467.
101. Barzon L, Masi G, Pacenti M, Trevisan M, Fallo F, Remo A, Martignoni G, Montanaro D, Pezzi V, Palù G. Expression of aromatase and estrogen receptors in human adrenocortical tumors. Virchows Arch 2008;452:181–191.
102. Konduri S, Schwarz RE. Estrogen receptor beta/alpha ratio predicts response of pancreatic cancer cells to estrogens and phytoestrogens. J Surg Res 2007;140:55–66.
103. Carruba G. Estrogen and prostate cancer: An eclipsed truth in an androgen-dominated scenario. J Cell Biochem 2007;102:899–911.
104. Ricke WA, McPherson SJ, Bianco JJ, Cunha GR, Wang Y, Risbridger GP. Prostatic hormonal carcinogenesis is mediated by in situ estrogen production and estrogen receptor alpha signaling. FASEB J 2008;22:1512–1520.
105. Dalla Valle L, Ramina A, Vianello S, Fassina A, Belvedere P, Colombo L. Potential for estrogen synthesis and action in human normal and neoplastic thyroid tissues. J Clin Endocrinol Metab 1998;83:3702–3709.
106. Stettner M, Kaulfuss S, Burfeind P, Schweyer S, Strauss A, Ringert RH, Thelen P. The relevance of estrogen receptor-beta expression to the antiproliferative effects observed with histone deacetylase inhibitors and phytoestrogens in prostate cancer treatment. Mol Cancer Ther 2007;6:2626–2633.
107. Walton TJ, Li G, Seth R, McArdle SE, Bishop MC, Rees RC. DNA demethylation and histone deacetylation inhibition co-operate to re-express estrogen receptor beta and induce apoptosis in prostate cancer cell-lines. Prostate 2008;68:210–222.
108. Cheng J, Lee EJ, Madison LD, Lazennec G. Expression of estrogen receptor beta in prostate carcinoma cells inhibits invasion and proliferation and triggers apoptosis. FEBS Lett 2004;566:169–172.
109. Kawabata W, Suzuki T, Moriya T, Fujimori K, Naganuma H, Inoue S, Kinouchi Y, Kameyama K, Takami H, Shimosegawa T, Sasano H. Estrogen receptors (alpha and beta) and 17beta-hydroxysteroid dehydrogenase type 1 and 2 in thyroid disorders: possible in situ estrogen synthesis and actions. Mod Pathol 2003;16:437–444.
110. Zeng Q, Chen GG, Vlantis AC, van Hasselt CA. Estrogen mediates the growth of human thyroid carcinoma cells via an estrogen receptor—ERK pathway. Cell Prolif 2007;40:921–935.
111. Zeng Q, Chen GG, Vlantis AC, Tse GM, van Hasselt CA. Different contributions of estrogen receptor risoforms to the development of thyroid papillary and anaplastic cancers. J Pathol 2008;214:425–433.

112. Cho MA, Lee MK, Nam KH, Chung WY, Park CS, Lee JH, Noh T, Yang WI, Rhee Y, Lim SK, Lee HC, Lee EJ. Expression and role of estrogen receptor alpha and beta in medullary thyroid carcinoma: Different roles in cancer growth and apoptosis. J Endocrinol 2007;195:255–263.
113. Satake M, Sawai H, Go VL, Satake K, Reber HA, Hines OJ, Eibl G. Estrogen receptors in pancreatic tumors. Pancreas 2006;33:119–127.
114. Price D, Stein B, Sieber P, Tutrone R, Bailen J, Goluboff E, Burzon D, Bostwick D, Steiner M. Toremifene for the prevention of prostate cancer in men with high grade prostatic intraepithelial neoplasia: Results of a double-blind, placebo controlled, phase IIB clinical trial. J Urol 2006;176:965–970.
115. al-Azzawi F, Wahab M. Estrogen and colon cancer: Current issues. Climacteric 2002;5:3–14.
116. Slattery ML, Ballard-Barbash R, Edwards S, Caan BJ, Potter JD. Body mass index and colon cancer: An evaluation of the modifying effects of estrogen (United States). Cancer Causes Control 2003;14:75–84.
117. al-Azzawi F, Wahab M. Estrogen and colon cancer: Current issues. Climacteric 2002;5:3–14.
118. Newcomb PA, Zheng Y, Chia VM, Morimoto LM, Doria-Rose VP, Templeton A, Thibodeau SN, Potter JD. Estrogen plus progestin use, microsatellite instability, and the risk of colorectal cancer in women. Cancer Res 2007;67:7534–7539.
119. Qiu Y, Langman MJ, Eggo MC. Targets of 17beta-oestradiol-induced apoptosis in colon cancer cells: A mechanism for the protective effects of hormone replacement therapy? J Endocrinol 2004;181:327–337.
120. Gambacciani M, Monteleone P, Sacco A, Genazzani AR. Hormone replacement therapy and endometrial, ovarian and colorectal cancer. Best Pract Res Clin Endocrinol Metab 2003;17:139–147.
121. Burkman RT. Reproductive hormones and cancer: Ovarian and colon cancer. Obstet Gynecol Clin North Am 2002;29:527–540.
122. Gallus S, Bosetti C, Franceschi S, Levi F, Simonato L, Negri E, La Vecchia C. Oesophageal cancer in women: Tobacco, alcohol, nutritional and hormonal factors. Br J Cancer 2001;85:341–345.
123. Schabath MB, Wu X, Vassilopoulou-Sellin R, Vaporciyan AA, Spitz MR. Hormone replacement therapy and lung cancer risk: A case-control analysis. Clin Cancer Res 2004;10:113–123.
124. Schwartz AG, Wenzlaff AS, Prysak GM, Murphy V, Cote ML, Brooks SC, Skafar DF, Lonardo F. Reproductive factors, hormone use, estrogen receptor expression and risk of non small-cell lung cancer in women. J Clin Oncol 2007;25:5785–5792.
125. Hsu HH, Cheng SF, Chen LM, Liu JY, Chu CH, Weng YJ, Li ZY, Lin CS, Lee SD, Kuo WW, Huang CY. Over-expressed estrogen receptor-alpha up-regulates hTNF-alpha gene expression and down-regulates beta-catenin signaling activity to induce the apoptosis and inhibit proliferation of LoVo colon cancer cells. Mol Cell Biochem 2006;289:101–109.
126. Campbell-Thompson M, Lynch IJ, Bhardwaj B. Expression of estrogen receptor (ER) subtypes and ERbeta isoforms in colon cancer. Cancer Res 2001;61:632–640.
127. Nakayama Y, Sakamoto H, Satoh K, Yamamoto T. Tamoxifen and gonadal steroids inhibit colon cancer growth in association with inhibition of thymidylate synthase, survivin and telomerase expression through estrogen receptor beta mediated system. Cancer Lett 2000;161:63–71.
128. Fiorelli G, Picariello L, Martineti V, Tonelli F, Brandi ML. Functional estrogen receptor beta in colon cancer cells. Biochem Biophys Res Commun 1999;261:521–527.
129. Utsumi Y, Nakamura T, Nagasue N, Kubota H, Harada T, Morikawa S. Effect of 17 beta-estradiol on the growth of an estrogen receptor-positive human esophageal carcinoma cell line. Cancer 1991;67:2284–2289.
130. Ueo H, Matsuoka H, Sugimachi K, Kuwano H, Mori M, Akiyoshi T. Inhibitory effects of estrogen on the growth of a human esophageal carcinoma cell line. Cancer Res 1990;50:7212–7215.
131. Liu L, Chirala M, Younes M. Expression of estrogen receptor-beta isoforms in Barrett's metaplasia, dysplasia and esophageal adenocarcinoma. Anticancer Res 2004;24:2919–2924.
132. Nozoe T, Oyama T, Takenoyama M, Hanagiri T, Sugio K, Yasumoto K. Significance of immunohistochemical expression of estrogen receptors alpha and beta in squamous cell carcinoma of the esophagus. Clin Cancer Res 2007;13:4046–4050.
133. Wang AG, Lee KY, Kim SY, Choi JY, Lee KH, Kim WH, Wang HJ, Kim JM, Park MG, Yeom YI, Kim NS, Yu DY, Lee DS. The expression of estrogen receptors in hepatocellular carcinoma in Korean patients. Yonsei Med J 2006;47:811–816.
134. Villa E, Moles A, Ferretti I, Buttafoco P, Grottola A, Del Buono M, De Santis M, Manenti F. Natural history of inoperable hepatocellular carcinoma: Estrogen receptors' status in the tumor is the strongest prognostic factor for survival. Hepatology 2000;32:233–238.
135. Pietras RJ, Marquez DC, Chen HW, Tsai E, Weinberg O, Fishbein M. Estrogen and growth factor receptor interactions in human breast and non-small cell lung cancer cells. Steroids 2005;70:372–381.

136. Kawai H, Ishii A, Washiya K, Konno T, Kon H, Yamaya C, Ono I, Ogawa J. Combined overexpression of EGFR and estrogen receptor alpha correlates with a poor outcome in lung cancer. Anticancer Res 2005;25:4693–4698.
137. Hershberger PA, Vasquez AC, Kanterewicz B, Land S, Siegfried JM, Nichols M. Regulation of endogenous gene expression in human non-small cell lung cancer cells by estrogen receptor ligands. Cancer Res 2005;65:1598–1605.
138. Kawai H, Ishii A, Washiya K, Konno T, Kon H, Yamaya C, Ono I, Minamiya Y, Ogawa J. Estrogen receptor alpha and beta are prognostic factors in non-small cell lung cancer. Clin Cancer Res 2005;11:5084–5089.
139. Bentzon N, During M, Rasmussen BB, Mouridsen H, Kroman N. Prognostic effect of estrogen receptor status across age in primary breast cancer. Int J Cancer 2008;122:1089–1094.
140. Arai N, Strom A, Rafter JJ, Gustafsson JA. Estrogen receptor beta mRNA in colon cancer cells: Growth effects of estrogen and genistein. Biochem Biophys Res Commun 2000;270:425–431.
141. Ali SH, O'Donnell AL, Mohamed S, Mousa S, Dandona P. Overexpression of estrogen receptor-alpha in the endometrial carcinoma cell line Ishikawa: Inhibition of growth and angiogenic factors. Gynecol Oncol 2004;95:637–645.
142. Shabani N, Mylonas I, Jeschke U, Thaqi A, Kuhn C, Puchner T, Friese K. Expression of estrogen receptors alpha and beta, and progesterone receptors A and B in human mucinous carcinoma of the endometrium. Anticancer Res 2007;27:2027–2033.
143. Waalkes MP, Liu J, Chen H, Xie Y, Achanzar WE, Zhou YS, Cheng ML, Diwan BA. Estrogen signaling in livers of male mice with hepatocellular carcinoma induced by exposure to arsenic in utero. J Natl Cancer Inst 2004;96:466–474.
144. Schwartz AG, Prysak GM, Murphy V, Lonardo F, Pass H, Schwartz J, Brooks S. Nuclear estrogen receptor beta in lung cancer: Expression and survival differences by sex. Clin Cancer Res 2005;11:7280–7287.
145. Chan KK, Wei N, Liu SS, Xiao-Yun L, Cheung AN, Ngan HY. Estrogen receptor subtypes in ovarian cancer: a clinical correlation. Obstet Gynecol 2008;111:144–151.
146. Yu S, Wong YC, Wang XH, Ling MT, Ng CF, Chen S, Chan FL. Orphan nuclear receptor estrogen-related receptor-beta suppresses in vitro and in vivo growth of prostate cancer cells via p21(WAF1/CIP1) induction and as a potential therapeutic target in prostate cancer. Oncogene (in press).
147. Arain SA, Shah MH, Meo SA, Jamal Q. Estrogen receptors in human thyroid gland. An immunohistochemical study. Saudi Med J 2003;24:174–178.
148. Stender JD, Frasor J, Komm B, Chang KC, Kraus WL, Katzenellenbogen BS. Estrogen-regulated gene networks in human breast cancer cells: Involvement of E2F1 in the regulation of cell proliferation. Mol Endocrinol 2007;21:2112–2123.
149. Wang Q, Li X, Wang L, Feng YH, Zeng R, Gorodeski G. Antiapoptotic effects of estrogen in normal and cancer human cervical epithelial cells. Endocrinology 2004;145:5568–5579.
150. Chen D, Carter TH, Auborn KJ. Apoptosis in cervical cancer cells: Implications for adjunct anti-estrogen therapy for cervical cancer. Anticancer Res 2004;24:2649–2656.
151. Fiorelli G, Picariello L, Martineti V, Tonelli F, Brandi ML. Estrogen synthesis in human colon cancer epithelial cells. J Steroid Biochem Mol Biol 1999;71:223–230.
152. Pathirage N, Di Nezza LA, Salmonsen LA, Jobling T, Simpson ER, Clyne CD. Expression of aromatase, estrogen receptors, and their coactivators in patients with endometrial cancer. Fertil Steril 2006;86:469–472.
153. Watanabe J, Kamata Y, Seo N, Okayasu I, Kuramoto H. Stimulatory effect of estrogen on the growth of endometrial cancer cells is regulated by cell-cycle regulators. J Steroid Biochem Mol Biol 2007;107:163–171.
154. Granata OM, Cocciadiferro L, Miceli V, Polito LM, Campisi I, Carruba G. Metabolic profiles of androgens in malignant human liver cell lines. Ann N Y Acad Sci 2006;1089:262–267.
155. Mah V, Seligson DB, Li A, Márquez DC, Wistuba II, Elshimali Y, Fishbein MC, Chia D, Pietras RJ, Goodglick L. Aromatase expression predicts survival in women with early-stage non small cell lung cancer. Cancer Res 2007;67:10484–10490.
156. Weinberg OK, Marquez-Garban DC, Fishbein MC, Goodglick L, Garban HJ, Dubinett SM, Pietras RJ. Aromatase inhibitors in human lung cancer therapy. Cancer Res 2005;65:11287–11291.
157. Ding JX, Feng YJ, Yao LQ, Yu M, Jin HY, Yin LH. The reinforcement of invasion in epithelial ovarian cancer cells by 17 beta-Estradiol is associated with up-regulation of Snail. Gynecol Oncol 2006;103:623–630.
158. Lee ML, Chen GG, Vlantis AC, Tse GMK, Leung BCH, van Hasselt CA. Induction of thyroid papillary carcinoma cell proliferation by estrogen is associated with an altered expression of Bcl-xL. Cancer J 2005;11:113–121.

159. Lacey JV Jr, Brinton LA, Barnes WA, Gravitt PE, Greenberg MD, Hadjimichael OC, McGowan L, Mortel R, Schwartz PE, Kurman RJ, Hildesheim A. Use of hormone replacement therapy and adenocarcinomas and squamous cell carcinomas of the uterine cervix. Gynecol Oncol 2000;77:149–154.
160. Yasmeen S, Romano PS, Pettinger M, Johnson SR, Hubbell FA, Lane DS, Hendrix SL. Incidence of cervical cytological abnormalities with aging in the women's health initiative: A randomized controlled trial. Obstet Gynecol 2006;108:410–419.
161. Rebbeck TR, Troxel AB, Wang Y, Walker AH, Panossian S, Gallagher S, Shatalova EG, Blanchard R, Bunin G, DeMichele A, Rubin SC, Baumgarten M, Berlin M, Schinnar R, Berlin JA, Strom BL. Estrogen sulfation genes, hormone replacement therapy, and endometrial cancer risk. J Natl Cancer Inst 2006;98:1311–1320.
162. Lacey JV Jr, Brinton LA, Leitzmann MF, Mouw T, Hollenbeck A, Schatzkin A, Hartge P. Menopausal hormone therapy and ovarian cancer risk in the National Institutes of Health-AARP Diet and Health Study Cohort. J Natl Cancer Inst 2006;98:1397–1405.
163. Beral V, Million Women Study Collaborators, Bull D, Green J, Reeves G. Ovarian cancer and hormone replacement therapy in the Million Women Study. Lancet 2007;369:1703–1710.
164. Greiser CM, Greiser EM, Dören M. Menopausal hormone therapy and risk of ovarian cancer: Systematic review and meta-analysis. Hum Reprod Update 2007;13:453–463.
165. Duell EJ, Holly EA. Reproductive and menstrual risk factors for pancreatic cancer: a population-based study of San Francisco Bay Area women. Am J Epidemiol 2005;161:741–747.

George G Chen *received his PhD from the University of Glasgow, UK, his Medical degrees from Shanghai 2nd Medical University and Fujian Medical University, China. He is currently a professor and surgical laboratory director in The Chinese University of Hong Kong, Hong Kong. His laboratory focuses primarily on how the apoptotic molecules contribute to the carcinogenesis, He has published approximately 100 full-papers.*

Qiang Zeng *received his PhD from The Chinese University of Hong Kong, Hong Kong and his MD from Xiang Ya School of Medicine, Central South University, China. His PhD project was in the area of estrogen and its receptors in human thyroid cancer. He is currently a post-doctoral fellow in Florida Atlantic University, USA.*

Gary M.K. Tse *(MBBS, FRCPC, FCAP) is an Associate Consultants with interest in breast, head and neck pathology and in autopsy. He works closely with breast radiologists, surgeons and scientists in the areas of breast, head and neck cancers.*