Neuroendocrinology Letters ISSN 0172–780X


REVIEW ARTICLE

# Endocrine modulation and the fragile balance of homeostasis – An overview

**Günter Vollmer, Susanne Starcke, Jannette Wober & Oliver Zierau**

Molecular Cell Physiology & Endocrinology, Faculty for Mathematics and Science, Institute for Zoology, Germany.

*Correspondence to:* Molecular Cell Physiology and Endocrinology
Institute of Zoology
Technical University Dresden, GERMANY.
Mommsenstr. 13
01062 Dresden
PHONE: +49 351/463-31922; FAX: +49 351/463-31923
E-MAIL: guenter.vollmer@mailbox.tu-dresden.de

*Submitted:* *May 2, 2002*
*Accepted:* *May 5, 2002*

*Key words:* **phytoestrogens; estrogen receptor-α and β; development; menopause; tissue specificity; endocrine disruption**

*Neuroendocrinology Letters 2002; **23**(suppl 2):37–42 pii: NEL230802R04 Copyright © Neuroendocrinology Letters 2002*

**Abstract**

Endocrine modulation by natural and synthetic chemicals and the eventually resulting beneficial or adverse effects for human and animal health are controversially debated not only among scientists but particularly in the public. Most information is available on so-called environmental estrogens, however the amount of information on substances interfering with other hormonal axes steadily increases, particularly on those exhibiting (anti)androgenic activities. The aim of this paper is to summarize existing data and to give an overview on the potential pathways leading to interferences of environmental hormones with homeostasis and eventually resulting health effects. Experimental evidence suggests the hypothesis that fetal and neonatal organisms may be at risk if exposed to environmental estrogens. In contrary, it appears as if phytoestrogens, particularly those with selective estrogen receptor modulator-(SERM-)like activities have the potential to be useful in medical application, both as dietary means and as pharmaceuticals. Lacking valid information about the detailed analysis of the molecular mode of action for environmental estrogens, the possibility for an ultimate classification of environmental estrogens in "dangerous endocrine disruptors" and phytoestrogens in "useful pharmaceuticals" cannot be supported conclusively. Nevertheless both activities are likely.

## Exposure to environmental hormones

Most information is available on so-called environmental estrogens, however, the amount of information on substances interfering with other hormonal axes steadily increases, particularly on those exhibiting (anti)androgenic activities. Xenobiotic substances capable to interfere with estrogen function add up to >230 individual compounds [1, 2]. They comprise naturally occurring compounds e.g. endogenous estrogens, phyto-and mycoestrogens, as well as man-made chemicals e.g. oral contraceptives or industrial products with hormone-like activities (for review see [3]). However, it has to be kept in mind that some of these synthetic xenobiotics accused to cause effects in the male reproductive tract or affect its function e.g. sperm quality and quantity, occur in the environment in concentrations orders of magnitude lower than those estrogens which are used for oral contraception and hormonal replacement therapies [4] or contained in meat of the daily diet [5]. Further, the exposure to hormonally active xenobiotics can be neglected to the amount of phytoestrogens ingested with the diet or through herbal potions use in so-called "life

style medicine" [4]. The latter are at least able to induce hormonal changes in females and may exhibit toxicity in males. Despite numerous effects described in many different experimental systems there is no conclusive evidence about the capability of environmental hormones to induce impacts on human health [6]. Potential risks and benefits of exposure or use of environmental estrogens, particularly of phytoestrogens, which occur in high concentrations, will be discussed below.

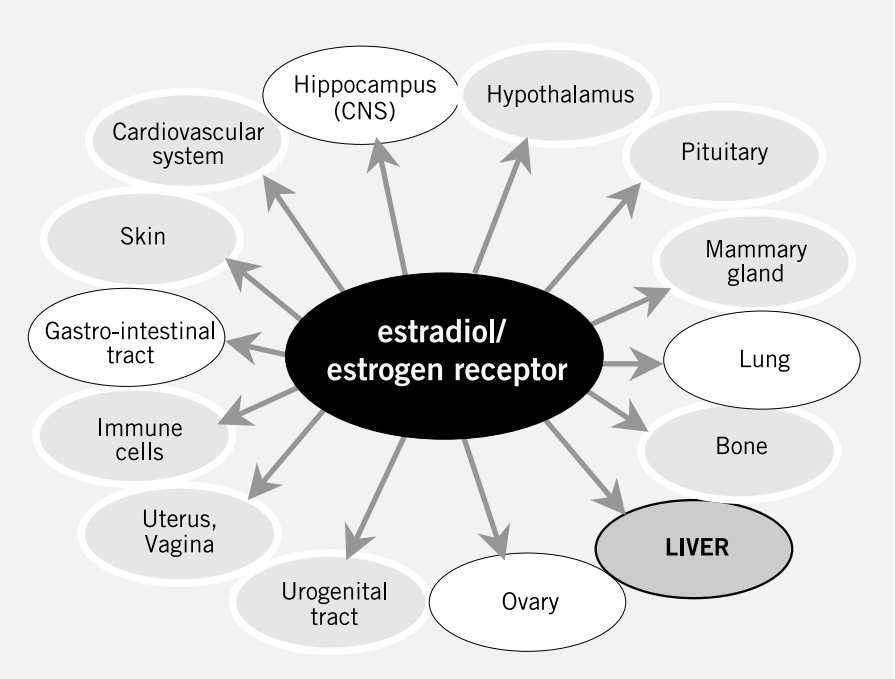
### Molecular mechanisms triggered by hormones from the environment

The molecular mechanisms triggered by hormonal substances from the environment can be subdivided into two categories: nuclear receptor mediated responses and direct effects. In the case of environmental estrogens the first class of effects is triggered by the binding to estrogen receptors-α (ERα) and -β (ERβ) thereby activating genomic responses e.g. alteration of gene expression. Over the last few years the picture of receptor mediated estrogen action became rather complex. First of all there exist two receptor subtypes and several splice variants thereof. Several ligands have been identified binding with different affinities to the two receptor subspecies α and β. Most prominent in this regard is the phytoestrogen genistein with its clear-cut preference for the ERβ [7]. Upon ligand binding they can form homo- and heterodimers with differing transcriptional activity [8], [9]. To make things even more complicated the steric conformation of the ligand binding domain is dependent on the ligand bound [10], [11, 12]. This is important because the ligand binding domain harbors the activation function-2 (AF-2) which in turn is necessary for the interaction with so-called co-activators or co-repressors [13], [14]. In other words, the bound ligand determines whether the hormone receptor complex is accessible for molecules capable of enhancing or suppressing transcriptional activity [15]. Upon pure combinatorial considerations a multiplicity of potential action modes of a given estrogenic compound arises.

However, there are further important issues needed to be taken into consideration. ERs have been found in virtually almost all organs and cell types which have been looked at, however some of these organs exhibit a dramatic difference in the relative amount of ER subtypes-α and –β ([16]; Fig. 1). Finally, ER complexes not only function in the classical view of ligand activated transcription factors, but are also capable to bind to other transcription factors like AP-1, SP1 or NFκB and modulate those transcriptional activities [15]. In summary, the picture arises that environmental estrogens exhibit cell and organ specific effects and functions commonly referred to as selective estrogen receptor modulator (SERM) activity. This SERM activity is clinically relevant as already proven for the synthetic SERM Raloxifen, used in the treatment of osteoporosis, and represents the theoretical basis for any considerations of potential beneficial effects of SERM phytoestrogens.

Molecular modes of action of environmental hormones comprise interactions with key steroid metabolic enzymes like sulfotransferase and sulfatase, 3β-hydroxysteroid dehydrogenase, 17β-hydroxysteroid dehydrogenase and aromatase [17, 18], [19], [20]. Further, they are suspected to bind to steroid binding proteins such as sex hormone binding globulin [21] and α-fetoprotein, particularly of rodents [22], thereby altering the ratio of free (available) hormone and protein bound hormone, particularly relevant for estrogens.

Further, natural and synthetic hormones trigger rapid responses thereby circumventing receptor mediated mechanisms of gene expression. Signal transduction pathways involved are those leading to an increase of intracellular calcium levels [23], [24] and to activation of mitogen activated protein kinases (MAP-kinases) [25]. These mechanisms lead to functional consequences such as alterations in oxidative, inflammatory and angio-



genic pathways, on energy metabolism and on inhibition of tyrosine kinases.

Finally, cross-talk mechanisms between steroid receptors and growth factor mediated signal transduction cascades have been described. By this mechanism growth factors like IGF-I, EGF and others, as well as increased intracellular cAMP levels are capable of activating ERs in a ligand independent manner (for review see [26]).

## Potential risks and benefits of endocrine modulation

Taken together all the knowledge on molecular mechanisms triggered by environmental hormones a very complex, multifaceted picture arises. As a consequence two immediate questions come up. Which are the risks following exposure to environmental hormones, which are benefits expectable from pharmacological use of environmental hormones, particularly of phytoestrogens? We are far away from being able to answer these questions ultimately, but some issues appear to be obvious following review of the available literature. It appears as if the answer to the above raised questions clearly correlates with the respective life stage at the time point of exposure. To date evidence accumulates that exposure of pre- or perinatal organisms induce severe health risks, whereas there is reason to believe that the ageing population particularly postmenopausal women may benefit from "exposure" (treatment) with phytoestrogens.

## Neonatal and perinatal exposure to (environmental) estrogens

Evidence for interference of endocrine disrupting chemicals with a developing organism is abundant and has been thoroughly reviewed recently [27]. Environmental exposure has usually to be regarded as being chronically and in low doses; a situation which is hard to mimic under laboratory conditions. Therefore, examples for risks by environmental hormones will be discussed which are clearly supported by experimental laboratory data.

*Low dose effects and fetal exposure*: A topic which is most controversially discussed in the literature is the topic of low dose effects of environmental estrogens and fetal exposure. There was one study reporting increase in prostate weight of male mice following low dose exposure of fetuses to bisphenol A [21]. This piece of work stimulated an important discussion about the probability of low dose effects and non-linear dose response curves. Although the issue of increased prostate weight following bisphenol A exposure has been contradicted by two consecutive studies using larger groups of animals [28], [29], the discussion on low dose effects on the developing embryo still persists [30], as well as the discussion on non-linear, inverted dose-response curves. Because in a similar experimental setting as for the studies on impacts of exposure of environmental estrogens on male fetuses, the same group tested for influences in females finding an advanced onset of estrus cyclicity which has to be regarded as earlier onset of puberty [31].

*Neonatal exposure and imprinting*: The neonatal period is critical in the development of the future hormone-receptor connections. The first encounter between the developing receptor and the target hormone provokes the hormonal imprinting needed for the maturation of the signal transduction system. Changes to this imprinting can be evoked by absence or excess of molecules similar to the target hormone, the latter being already recognized in 1983 [32]. Consequently, and as a result of the DES disaster [33], these effects have been thoroughly and extensively studied for this compound (for review see [34]). Mechanistically it has been proposed that there is a phenomenon of "pathway" imprinting [27] of signal transduction cascades as well as a genomic imprinting through DNA-methylation [35]. In addition, histone acetylation representing a generally accepted mechanism for imprinting [36], [37] has not been extensively studied as a potential mechanism of hormonal imprinting. Now that the experimental tools are available to investigate the imprinting phenomenon on a molecular level, the phenomenon of hormonal imprinting as a target for hormonal deregulation has attracted considerable attention again.

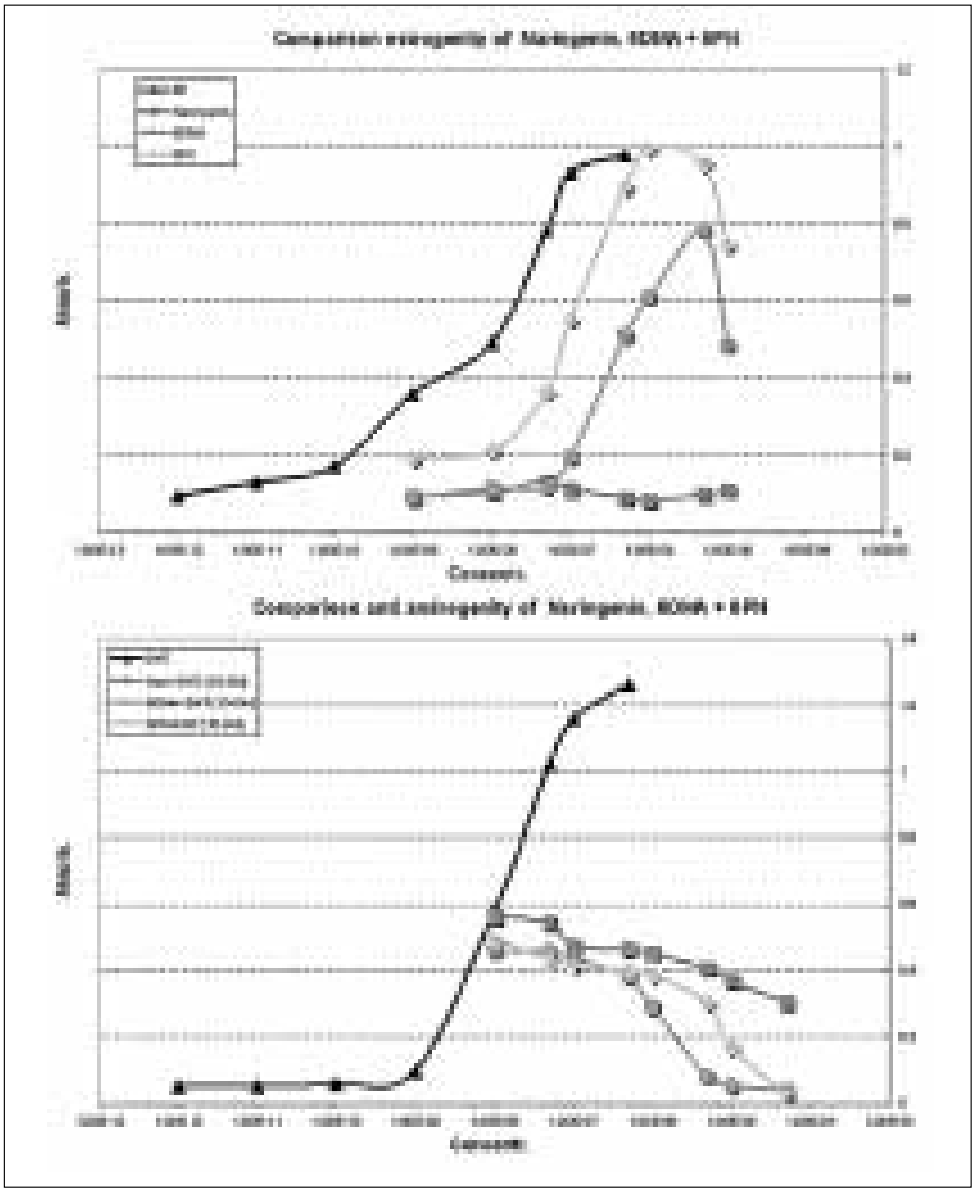
Proposing imprinting as a major regulator in the determination of functional hormonal circuits the question arises what is at stake if imprinting occurs at inappropriate time or with inappropriate doses of hormone? Recent studies exemplify the consequences of inappropriate imprinting.

In two strains of male rats neonatal imprinting with estradiol-benzoate leads to consequences in weight and sizes of the male reproductive organs. For the prostate a temporally biphasic response with an apparent non-linear dose response could be shown. At postnatal day 35 low doses induced increase in prostate weight, whereas high doses induced a significant reduction in prostate weight. While the low dose effect was completely abolished at postnatal day 90 the high dose effect persisted [38]. Similar high dose effects were detectable in other male sexual organs e.g. testis, epididymis, seminal vesicle and coagulating gland. In addition testosterone metabolism was compromised by the down-regulation of various testosterone converting enzymes at high doses of estradiol-benzoate [39].

Neonatal imprinting with the antiestrogen Tamoxifen led to considerable alterations in the sexual behavior of adult animals. In animals of both sexes the typical activities and responses (lordosis in females and mounting and intromission in males) had been vanished by almost 100%. Interestingly there was almost no alteration following treatment of animals with the antigestagen Mifepristone using the identical protocol [40].

Whereas in the studies mentioned above, derivatives of natural hormones or synthetic antihormones were used there is one study performed with the phytoestrogen genistein. Neonatal exposure of mice to this particular phytoestrogen induced endometrial adenocarcinoma in adult animals [41].

Taken together, the studies on endocrine modulation in pre- and neonatal animals demonstrate that inappropriate exposure to estrogenic hormones can lead to permanent and detrimental effects in both male and female organisms. Particularly the stage of hormonal imprint-



ing appears to be a very sensitive target for endocrine disruption of hormone function.

### Hormonal modulation and ageing

Screening literature with regard to potential targets for the beneficial use of the accumulated knowledge on hormone action immediately the ageing population becomes apparent. Epidemiological and experimental studies suggest that phytoestrogens are potent candidate molecules for hormone replacement strategies. Dietary habits or respectively treatment with phytotherapeutica, containing estrogen-like activities, apparently led to a decrease of the incidence rate for carcinoma of breast, prostate and colon [42], [43]. Further, they are believed to protect against loss of bone mass and bone density [44] thereby slowing down the process of osteoporosis [45]. Finally preliminary data suggest that they may also be useful for the maintenance of the integrity of vessel walls and in the prevention of atherosclerosis [46]. In both organ systems regulation or deregulation is mediated by almost identical set of genes, which at least in part are regulated by estrogens. Intimate involvement of the estradiol/estrogen receptor system in osteoporosis and atherosclerosis is also suggested by results from studies with knock-out mice. Animals with targeted disruption of ERα or ERβ exhibit a compromised regulation of blood pressure, reactivity of the capillaries, regulation of weight and blood lipids [47], [48].

Another potential target for the use of phytoestrogens is the brain. There are preliminary data, both positive and negative, on possible effects on the progression of degenerative brain diseases [49], [50]. It is also likely that general behavior in common [51], [52] can be modulated as suggested by studies on anxiety [53], visual spatial memory [54] and sexual behavior [40].

In summary, the picture arises that environmental estrogens eventually exhibit cell and organ specific effects and function as a SERM. However, at present and based on our current knowledge it is much too early to conclusively judge the pharmaceutical potential of phy-

toestrogens. In the view of the available literature it appears likely that phytoestrogens or diets containing phytoestrogens and plant extracts will be increasingly be used to prevent age correlated complaints or diseases as well as osteoporosis or cardiovascular diseases originated by the drop of the body's own hormone production . For a conclusive evaluation of phytoestrogens a lot more data about their molecular mode of action are needed, particularly on the organ selectivity of function of some phytoestrogens. Further, it appears necessary to screen for new phytoestrogens, especially for those exhibiting the characteristic of SERM. This screening for new lead substances is essential because some of the known phytoestrogens e.g. coumestrol are pure agonists with some adverse side effects [55, 56], others like genistein seem to be troublesome if the neonatal organisms is exposed [41]. A new promising class of phytoestrogens may be represented by prenylated naringenins (Fig. 2) which at least in *in vitro* systems show both estrogenic and antiandrogenic potentials and therefore may have the potential of representing phytoestrogens with SERM-like quality.

In conclusion, phytoestrogens represent a heterogenous class of substances with a strong potential to be used in pharmacological research particularly in the search for novel lead substances for hormone replacement therapy. Although these substances may have the potential to benefit the ageing female population care has to be taken since the same class of substances may act as endocrine disrupting chemicals in exposed fetal or neonatal organisms.

REFERENCES


1 Fang H, Tong W, Shi LM, et al. Structure-activity relationships for a large diverse set of natural, synthetic, and environmental estrogens. Chem Res Toxicol 2001; **14**:280–94.
2 Hong H, Tong W, Fang H, et al. Prediction of estrogen receptor binding for 58,000 chemicals using an integrated system of a tree-based model with structural alerts. Environ Health Perspect 2002; **110**:29–36.
3 Murkies AL, Wilcox G, Davis SR. Clinical review 92: Phytoestrogens. J Clin Endocrinol Metab 1998; **83**:297–303.
4 Nilsson R. Endocrine modulators in the food chain and environment. Toxicol Pathol 2000; **28**:420–31.
5 Ibarreta D, Daxenberger A, Meyer HH. Possible health impact of phytoestrogens and xenoestrogens in food. Apmis 2001; **109**:161–84.
6 Degen GH, Bolt HM. Endocrine disruptors: update on xenoestrogens. Int Arch Occup Environ Health 2000; **73**:433–41.
7 Kuiper GG, Carlsson B, Grandien K, et al. Comparison of the ligand binding specificity and transcript tissue distribution of estrogen receptors alpha and beta. Endocrinology 1997; **138**:863–70.
8 Cowley SM, Hoare S, Mosselman S, Parker MG. Estrogen receptors alpha and beta form heterodimers on DNA. J Biol Chem 1997; **272**:19858–62.
9 Pettersson K, Grandien K, Kuiper GG, Gustafsson JA. Mouse estrogen receptor beta forms estrogen response element-binding heterodimers with estrogen receptor alpha. Mol Endocrinol 1997; **11**:1486–96.
10 Brzozowski AM, Pike AC, Dauter Z, et al. Molecular basis of agonism and antagonism in the oestrogen receptor. Nature 1997; **389**:753–8.
11 Pike AC, Brzozowski AM, Walton J, et al. Structural aspects of agonism and antagonism in the oestrogen receptor. Biochem Soc Trans 2000; **28**:396–400.
12 Pike AC, Brzozowski AM, Hubbard RE, et al. Structure of the ligand-binding domain of oestrogen receptor beta in the presence of a partial agonist and a full antagonist. Embo J 1999; **18**:4608–18.
13 Wong CW, Komm B, Cheskis BJ. Structure-function evaluation of ER alpha and beta interplay with SRC family coactivators. ER selective ligands. Biochemistry 2001; **40**:6756–65.
14 Bramlett KS, Wu Y, Burris TP. Ligands specify coactivator nuclear receptor (NR) box affinity for estrogen receptor subtypes. Mol Endocrinol 2001; **15**:909–22.
15 Nilsson S, Makela S, Treuter E, et al. Mechanisms of estrogen action. Physiol Rev 2001; **81**:1535–65.
16 Gustafsson JA. Estrogen receptor beta – a new dimension in estrogen mechanism of action. J Endocrinol 1999; **163**:379–83.
17 Kirk CJ, Harris RM, Wood DM, Waring RH, Hughes PJ. Do dietary phytoestrogens influence susceptibility to hormone-dependent cancer by disrupting the metabolism of endogenous oestrogens? Biochem Soc Trans 2001; **29**:209–16.
18 Krazeisen A, Breitling R, Moller G, Adamski J. Phytoestrogens inhibit human 17beta-hydroxysteroid dehydrogenase type 5. Mol Cell Endocrinol 2001; **171**:151–62.
19 Le Bail JC, Champavier Y, Chulia AJ, Habrioux G. Effects of phytoestrogens on aromatase, 3beta and 17beta-hydroxysteroid dehydrogenase activities and human breast cancer cells. Life Sci 2000; **66**:1281–91.
20 Makela S, Poutanen M, Lehtimaki J, Kostian ML, Santti R, Vihko R. Estrogen-specific 17 beta-hydroxysteroid oxidoreductase type 1 (E.C. 1.1.1.62) as a possible target for the action of phytoestrogens. Proc Soc Exp Biol Med 1995; **208**:51–9.
21 Nagel SC, vom Saal FS, Welshons WV. The effective free fraction of estradiol and xenoestrogens in human serum measured by whole cell uptake assays: physiology of delivery modifies estrogenic activity. Proc Soc Exp Biol Med 1998; **217**:300–9.
22 Milligan SR, Khan O, Nash M. Competitive binding of xenobiotic oestrogens to rat alpha-fetoprotein and to sex steroid binding proteins in human and rainbow trout (Oncorhynchus mykiss) plasma. Gen Comp Endocrinol 1998; **112**:89–95.
23 Dopp E, Vollmer G, Hahnel C, Grevesmuhl Y, Schiffmann D. Modulation of the intracellular calcium level in mammalian cells caused by 17beta-estradiol, different phytoestrogens and the anti-estrogen ICI 182780. J Steroid Biochem Mol Biol 1999; **68**:57–64.
24 Chambliss KL, Shaul PW. Rapid activation of endothelial NO synthase by estrogen: evidence for a steroid receptor fast-action complex (SRFC) in caveolae. Steroids 2002; **67**:413–9.
25 Migliaccio A, Di Domenico M, Castoria G, et al. Tyrosine kinase/p21ras/MAP-kinase pathway activation by estradiol- receptor complex in MCF-7 cells. Embo J 1996; **15**:1292–300. abs.html.
26 Cenni B, Picard D. Ligand-independent Activation of Steroid Receptors: New Roles for Old Players. Trends Endocrinol Metab 1999; **10**:41–46. _00000121 _00000121.
27 McLachlan JA, Newbold RR, Burow ME, Li SF. From malformations to molecular mechanisms in the male: three decades of research on endocrine disrupters. Apmis 2001; **109**:263–72.
28 Ashby J, Tinwell H, Haseman J. Lack of effects for low dose levels of bisphenol A and diethylstilbestrol on the prostate gland of CF1 mice exposed in utero. Regul Toxicol Pharmacol 1999; **30**:156–66.
29 Cagen SZ, Waechter JM, Jr., Dimond SS, et al. Normal reproductive organ development in CF-1 mice following prenatal exposure to bisphenol A. Toxicol Sci 1999; **50**:36–44.
30 Melnick R, Lucier G, Wolfe M, et al. Summary of the National Toxicology Program's Report of the Endocrine Disruptors Low-Dose Peer Review. Environ Health Perspect 2002; **110**:427–31.
31 Howdeshell KL, Hotchkiss AK, Thayer KA, Vandenbergh JG, vom Saal FS. Exposure to bisphenol A advances puberty. Nature. 1999; **401**:763–4.
32 Bern HA, Mills KT, Jones LA. Critical period for neonatal estrogen exposure in occurrence of mammary gland abnormalities in adult mice. Proc Soc Exp Biol Med. 1983; **172**:239–42.
33 Herbst AL. Exogenous hormones in pregnancy. Clin Obstet Gynecol 1973; **16**:37–50.
34 McLachlan JA. Environmental signaling: what embryos and evolution teach us about endocrine disrupting chemicals. Endocr Rev. 2001; **22**:319–41.
35 Li S, Ma L, Chiang T, et al. Promoter CpG methylation of Hox-a10 and Hox-a11 in mouse uterus not altered upon neonatal diethyl-

stilbestrol exposure. Mol Carcinog 2001; **32**:213–9.
36 Pennisi E. Behind the scenes of gene expression. Science 2001; **293**:1064–7.
37 Jenuwein T, Allis CD. Translating the histone code. Science 2001; **293**:1074–80.
38 Putz O, Schwartz CB, Kim S, LeBlanc GA, Cooper RL, Prins GS. Neonatal low- and high-dose exposure to estradiol benzoate in the male rat: I. Effects on the prostate gland. Biol Reprod 2001; **65**:1496–505.
39 Putz O, Schwartz CB, LeBlanc GA, Cooper RL, Prins GS. Neonatal low- and high-dose exposure to estradiol benzoate in the male rat: II. Effects on male puberty and the reproductive tract. Biol Reprod 2001; **65**:1506–17.
40 Csaba G, Karabelyos C. The effect of a single neonatal treatment (hormonal imprinting) with the antihormones, tamoxifen and mifepristone on the sexual behavior of adult rats. Pharmacol Res 2001; **43**:531–4.
41 Newbold RR, Banks EP, Bullock B, Jefferson WN. Uterine adenocarcinoma in mice treated neonatally with genistein. Cancer Res 2001; **61**:4325–8.
42 Adlercreutz CH, Goldin BR, Gorbach SL, et al. Soybean phytoestrogen intake and cancer risk. J Nutr 1995; **125**:757S–770S.
43 Ingram D, Sanders K, Kolybaba M, Lopez D. Case-control study of phyto-oestrogens and breast cancer. Lancet 1997; **350**:990–4.
44 Mei J, Yeung SS, Kung AW. High dietary phytoestrogen intake is associated with higher bone mineral density in postmenopausal but not premenopausal women. J Clin Endocrinol Metab 2001; **86**:5217–21.
45 Sherman S. Preventing and treating osteoporosis: strategies at the millennium. Ann N Y Acad Sci 2001; **949**:188–97.
46 Teede HJ, Dalais FS, Kotsopoulos D, Liang YL, Davis S, McGrath BP. Dietary soy has both beneficial and potentially adverse cardiovascular effects: a placebo-controlled study in men and postmenopausal women. J Clin Endocrinol Metab 2001; **86**:3053–60.
47 Nilsson BO, Ekblad E, Heine T, Gustafsson JA. Increased magnitude of relaxation to oestrogen in aorta from oestrogen receptor beta knock-out mice. J Endocrinol 2000; **166**:R5–9.
48 Korach KS. Estrogen receptor knock-out mice: molecular and endocrine phenotypes. J Soc Gynecol Investig 2000; **7**:S16–7.
49 Lambert JC, Harris JM, Mann D, et al. Are the estrogen receptors involved in Alzheimer's disease? Neurosci Lett 2001; **306**:193–7.
50 Wang CN, Chi CW, Lin YL, Chen CF, Shiao YJ. The neuroprotective effects of phytoestrogens on amyloid beta protein- induced toxicity are mediated by abrogating the activation of caspase cascade in rat cortical neurons. J Biol Chem 2001; **276**:5287–95.
51 Halbreich U, Kahn LS. Role of estrogen in the aetiology and treatment of mood disorders. CNS Drugs 2001; **15**:797–817.
52 Lephart ED, West TW, Weber KS, et al. Neurobehavioral effects of dietary soy phytoestrogens. Neurotoxicol Teratol 2002; **24**:5–16.
53 Lund TD, Lephart ED. Dietary soy phytoestrogens produce anxiolytic effects in the elevated plus-maze. Brain Res 2001; **913**:180–4.
54 Lund TD, West TW, Tian LY, et al. Visual spatial memory is enhanced in female rats (but inhibited in males) by dietary soy phytoestrogens. BMC Neurosci 2001; **2**:20.
55 Whitten PL, Patisaul HB, Young LJ. Neurobehavioral actions of coumestrol and related isoflavonoids in rodents. Neurotoxicol Teratol 2002; **24**:47–54.
56 Whitten PL, Lewis C, Russell E, Naftolin F. Potential adverse effects of phytoestrogens. J Nutr 1995; **125**:771S–776S.