Aromatase: Contributions to Physiology and Disease in Women and Men

Aromatase (estrogen synthetase; EC 1.14.14.1) catalyzes the demethylation of androgens’ carbon 19, producing phenolic 18-carbon estrogens. Aromatase is most widely known for its roles in reproduction and reproductive system diseases, and as a target for inhibitor therapy in estrogen-sensitive diseases including cancer, endometriosis, and leiomyoma (141, 143). However, all tissues contain estrogen receptor-expressing cells, the majority of genes have a complete or partial estrogen response element that regulates their expression (61), and there are plentiful nonreceptor effects of estrogens (79); therefore, the effect of aromatase through the provision of estrogen is almost universal in terms of health and disease. This review will provide a brief but comprehensive overview of the enzyme, its role in steroidogenesis, the problems that arise with its functional mutations and mishaps, the roles in human physiology of aromatase and its product estrogens, its current clinical roles, and the effects of aromatase inhibitors. While much of the story is that of the consequences of the formation of its product estrogens, we also will address alternative enzymatic roles of aromatase as a demethylase or nonenzymatic actions of this versatile molecule. Although this short review is meant to be thorough, it is by no means exhaustive; rather, it is meant to reflect the cutting-edge, exciting properties and possibilities of this ancient enzyme and its products.

What is Aromatase, Where is it Expressed, How is This Regulated, and What is the Outcome?

P450 aromatase clears androgens by their irreversible conversion to estrogens. Aromatase is the sole member of family 19 of the P450 superfamily of enzymes, termed CYP19. Aromatase is a microsomal protein that is found throughout the vertebrate phylum (141), but the human gene is unique compared with the rest of its superfamily. Specifically, the human gene includes one untranslated exon (I) that regulates tissue-specific promoters in the 5′-protein region, thereby responding to a wealth of tissue-specific regulation of aromatase expression (140). For example, aromatase expression in the ovary is regulated by cAMP but in the breast is regulated by prostaglandins (156). Expressions in the brain, adipose tissue, and placenta have been shown to be regulated by compounds addressing promoters that are coded at varying distances in the transcript from the enzyme (140). This means that conclusions regarding the effect of conditions and regulator availability must actually be tested before they can be accepted actions in biological systems. This also includes tissue-specific rates of aromatase expression [placenta (143), fat, liver, muscle, brain, breast (102), and prostate (145)]. Finally, although the mechanism is not settled, there are sex differences; in our studies, males have two- to threefold the activity of females in every tissue tested (99).

In mammalian systems, Cyp19 uses C19 androgens as their substrate and enzymatically removes C19 to form a phenolic A-ring in the steroid. Depending on the androgen’s attached prosthetic groups, the product of androstenedione, testosterone, 16-OH-androstenedione, or fetal estradiol, respectively, are estrone (E1), estradiol (E2), estriol (E3), or estetrol (E4) (47, 141, 143). In the late 1900s, there appeared reports of studies indicating that CYP19 could demethylate cocaine, and de-ethylate 7-ethoxycoumadin and 2 and 6 hydroxylate estradiol. Much of this work was...
substantiated by blockade of the effects with a monoclonal antibody to aromatase, and there was a dose-related inhibition with testosterone (105, 106, 133, 151). Unfortunately, this important work has not continued.

**Aromatase/Estrogens Have a Wide Range of Developmental and Adult Sources**

**Pregnancy.** Aromatase is expressed in the earliest tested syncyiotrophoblast layer of the human placenta (122); however, the majority of the estrogen formed is estriol, a weak ER ligand produced from 16α-OH androstenedione made by the fetal adrenal cortex (42). As a result, pregnancy is a “progesterone-dominated state.” Since progesterone is a smooth muscle relaxant that maintains the quiescence of the uterine muscle and promotes the maintenance of a dehydrated, tight cervix (65), during this period myometrial contractility is subdued and labor is held in abeyance. At the end of pregnancy, there is a fall in progesterone or a fall in progesterone receptor (PR) action (7) followed by the onset of labor (59, 88). We first ascribed the onset of labor to the fall in the “progesterone block” (34) that allows disinhibition of estrogen receptor (ER) expression and increased myometrial activity (124). The circulating estrogens also induce oxytocin receptors (87), potassium channels (113), and gap junctions (124) to sensitize the uterine muscle to electrical activity. Thus it is estrogen that causes the onset of labor, when the progesterone block is removed. In fact, in some countries, it was common practice to administer high-dose estradiol to induce labor (112).

**Brain.** The developing brain expresses aromatase and produces estrogen. The proof of aromatase action by the human fetal hypothalamus was the first organ-based example of peripheral, i.e., nongonadal, nonplacental conversion of androgen to estrogen (100). This finding supported the “aromatization hypothesis” of the sexual differentiation of the regulation of GnRH/gonadotrophin regulation and sexual behavior (9, 95, 145).

**Gonad.** The gonad is the chief source of aromatase in adults of both sexes. In *men*, the source of androgens is the Leydig cells that are outside of the seminiferous tubules cells. The spermatocys cells express aromatase and convert testicular androgens to estrogens (40). Since both compartments of the testis (Leydig cells and spermatocys cells) are self-renewing throughout life, men continue to secrete both androgens and estrogens throughout life, albeit at decreasing rates with age [andropause (148)]. Although local and peripherally formed estrogens are the main steroid regulators of pituitary gonadotropins (98). The role of estrogens in spermatogenesis and male fertility remains unresolved.

**Women** express aromatase in the cells that invest the developing oocyte (the granulosa cells of the developing follicle) and their postovulatory successors, the corpus luteum cells. The source of the androgen is the stromal-theca cells of the ovary. The roles of estrogen in regulating and maintaining the reproductive tract are widely known and will not be elaborated here (67).

**Other.** Aromatase expression has been shown in every organ tested. During the reproductive era, the circulating estrogen from the gonads is the main source of estrogen action. During the prepubertal and post-reproductive eras, locally formed phenolic estrogen receptor binders are the main source, and the outcomes of their action follow patterns from their original source.

**Regulatory and Homeostatic Functions of Aromatase/Estrogen**

**Clearance of androgen by aromatase results in substitution of 100- to 1,000-fold more bioactive estrogen molecules.** The conversion of androgen to estrogen by aromatase has a dual effect: 1) removal of the androgen molecule plus 2) the production of an estrogen molecule. While the former may have only a slight effect in the concentration of androgen, estrogens are mole for mole 100-1,000 times more active than androgens. Thus, although the amount of aromatase and the percentage of androgen converted to estrogen may be quantitatively small, often being below 1% in any tissue, the effect in terms of hormonal action may be great. Two examples follow. 1) Removal (detoxification) of androgens is necessary for the growth of the granulosa cells since androgens induce follicular atresia (41), and 2) estrogen induces FSH receptors that drive granulosa cell growth and aromatase expression in what will be the dominant follicle(s). Thus the two functions of aromatase are combined to bring about critical regulation of follicle number and development (123, 161). We believe that this “detoxification of androgens by aromatase” is the means by which monotonous vs. polytocious reproduction are regulated (56).

**Steroid hormone binding protein (SHBG) induction in the liver.** SHBG regulates the amount of unbound or “free/bioavailable” androgen and estrogen in the blood. In addition to high-affinity binding to specific binding proteins, such as SHBG, steroids and other molecules are electrostatically associated with albumin in the blood. This does not interfere with their biological function. Both hormones bind to SHBG, and their clearance of by the gut and kidneys reflects the amount of free or unbound hormone. SHBG expression is differentially regulated: androgens downregulate SHBG, whereas estrogens upregulate SHBG expression. Therefore, elevated androgen is the cause of
increased clearance of both estrogens and androgens, whereas elevated estrogens have the opposite effect. While the depletion may not be significant under most circumstances, it has been shown that, in cases of aromatase deficiency, there may be androgenization or even virilization, as is seen in aromatase-deficient pregnant women (134). In addition, since bound steroids cannot enter cells, they are biologically inactive. In this optic, the differential regulation of SHBG binding is of biological importance and conveys the major biological discrepancies in action of the two classes of sex steroids (see above and Ref. 118).

As an important exception, it is important to keep in mind that rodents do not express SHBG; their sex steroids are all “free.” They have very rapid clearance of sex steroids, and their sex steroids circulate at low levels, yet they are in homeostasis. This must be taken into account when evaluating effects of/on sex steroids in rodents (60, 71).

**Regulation of gonadotropins.** In both women and men, estradiol is the key steroidal hormone regulating feedback control of the gonadotrophins. Regardless of the level of testosterone, antagonist selective estrogen receptor modulators (SERMs) induce a rise of gonadotrophins (98).

**The sites of aromatase expression and action.** The placenta and gonads are the primary sources of aromatase. Before puberty and following menopause, estrogen arises mainly from extra-gonadal sites; “peripheral conversion” (56, 143). Precursor androgens arise from the ovaries, testes, and adrenal cortex after menopause/andropause (55). Adipose tissue is sufficiently abundant that even the low levels of aromatase in fat make it the chief source of estrogen in the postmenopausal woman; therefore, postmenopausal estrogen levels often correlate with body fat or BMI. This also is the case in aging men (81, 143).

**Circulating estrogens vs. locally produced estrogens cellular effects.** The chief organs expressing aromatase are the placenta and the gonads. These organs produce large amounts of estrogens that have massive local effects and contribute to the circulating estrogen at rates that generally overpower effects of locally produced estrogens in other organs (56, 122, 123). However, the placenta and ovary/testis are transient sources of aromatase; pregnancies end, and gonadal failure is inevitable. When this happens, there is increased impact of locally formed aromatase/estrogen. We have discussed elsewhere the straightforward effects of locally formed estrogen in the brain (99). However, effects of local aromatase in changing and heterogeneous cells masses are not always a simple matter. For example, while the normal female breast harbors aromatase expressed by the breast tissue and immunocytes, local and circulating estrogens affect the rate of apoptosis in the breast and endometrium (142), but the proportional cellular contributions change as the normal breast undergoes carcinogenesis (150). In this case, the locally formed estrogen appears to regulate the cellular immune response and may play role in the lack of response to malignant transformation (142) (see **FIGURE 3** below).

**Molecular effects.** The human vascular endothelium expresses a complete aromatase/estrogen/estrogen receptor system (39) that can regulate nitric acid synthetase (27), cell adhesion molecules (30), and the ability to sialylate neural cell adhesion molecule (NCAM), to obviate its ability to tether monocytes. We have proposed that the latter is one of the means by which estrogen is athero-preventative (35).

There are other examples of the effects of locally formed estrogens (139, 163). Not all are well understood; for example, the demonstration of the presence of aromatase in synaptosomes and axon terminals (96). While this aromatase could simply be converting androgens to estrogens, one must consider whether aromatase could have other chemical actions, for example, as a demethylase, deformylase, C-C, or acyl-carbon bond cleaver (2, 106, 115, 155). Some of these actions of aromatase have been woven into the drug-discovery process (57, 152). Nonclassical forms/expression of aromatase have been suggested (147, 162).

**Outcomes of Aromatase Action**

**Classical Estrogen Actions**

Aromatase expression and the action of its product estrogens have most commonly been related to reproduction. Quantitatively, in humans, the most important sources of aromatase are the placenta and the gonads. The placenta is the successor to the corpus luteum when pregnancy follows ovulation. This has been the prototype for biochemical studies on aromatase (121, 140). While aromatase expression is present in both the activated ovarian theca and the granulosa cells, the classical formulation is that, under regulation by the gonadotrophins, the theca cells produce androgens that are aromatized by the granulosa cells of the developing ovarian follicle and their successor corpus luteum cells (87, 123). The ovarian estrogens also drive the endometrial cycle (143). In the testis, aromatase expression is mainly associated with endocrine effects on spermatogenesis (145). However, estrogen secretion by the testes is of systemic importance. For example, genetic males deficient in androgen receptor expression secrete sufficient estrogen to develop a normal female extragenital phenotype and gonadotropin levels (97, 123).
**Homeostatic Estrogen Actions**

Less well appreciated are the widespread and pervasive effects of estrogen in maintaining immune homeostasis, metabolic homeostasis, bone homeostasis, etc. However, these actions of estrogens are fundamental to good health (54, 153). Reduction or absence of aromatase/estrogen has abundantly demonstrated the 24/7 need for estrogen throughout the body (80, 93) (see below where we discuss the pathological outcomes of low aromatase/estrogen). The ubiquitous effects of estrogens on all systems is not surprising since estrogen receptors are widespread and the majority of genes have estrogen response elements (15).

The picture of estrogen’s homeostatic actions is complicated by their variety, independent regulation, and overlapping domains; clashing outcomes of estrogen actions on interacting systems occur and may be confusing. For example, although estrogen protects against arteriosclerotic vascular disease (ASVD; see below), estrogen also induces thrombogenic proteins in the liver. While each of these functions is salutary, when a woman who is long past menopause has subclinical ASVD takes estrogen, as in the case of menopausal hormone treatment (MHT), the elevation of thrombogenic proteins plus deteriorating vascular endothelium may cause increased intravascular clotting in individuals with subclinical or clinical vascular disease. Treating women from 50 to 70 yr old (age at menopause, ~50 yr) with estrogen and failing to note that increased harm from venous thromboembolism was limited to the older, longer postmenopausal women was the basis of the initial misinterpretation by the WHI that concluded that estrogen given to all postmenopausal women is harmful (149). This erroneous conclusion has since been corrected (63a, 80).

While most of the information on this topic comes from observations on women, estrogen also plays key roles in homeostasis in men (32).

**Cellular Kinetics**

Estrogen is a survival agent that antagonizes apoptosis of breast cells, endometrial cells, etc. (76). This has been raised as a possible cause of overgrowth of endometrial and breast cells that may be related to dysplasia and cancer. The Women’s Health Initiative (WHI), however, has shown that conjugated equine estrogen alone reduces the incidence of invasive breast cancer (29, 117).

**Metabolic Effects of Aromatase/Estrogen**

Estradiol is a powerful regulator of metabolism. In addition to direct effects on liver proteins and lipids (44), estradiol regulates fuel metabolism. The chief means is through regulation of insulin sensitivity. Estradiol regulates insulin receptor expression and ligand estrogen receptors in the pancreas, clears circulating androgens, and affects gluconeogenesis (3). These effects are the bases of the protection against diabetes progress so well shown by the WHI (17).

Paradoxically, extremely high levels of circulating estradiol have been associated with worsening insulin insensitivity. This has recently been ascribed to estrogen binding to insulin and insulin receptors (117).

**Immune Regulation by Aromatase/Estrogen**

By regulating pro- and anti-apoptotic forces in the immune response, estrogens determine cellular homeostatic mechanisms (13, 92). Aromatase/estrogen effects on the immune system are the most pervasive, and arguably most important, regulatory functions in the body. They include actions in all of the systems in this section, from bone to cardiovascular to metabolism. Because of the limits of space for this review, and to avoid repetition, we have chosen to expand on these actions below in the section on diseases.

**Vascular Homeostasis by Aromatase/Estrogen**

The occurrence of cardiovascular disease and related deaths in estrogen-deficient women with premature menopause has been well documented (70, 110, 135, 144), as have the preventative effects of estrogen in naturally menopausal women who are treated within the first few years of its onset (31).

**Confusion over the WHI’s results.** Starting in 2002, the cardioprotective effects of menopausal estrogen treatment were obscured by the initial interpretation of the WHI (120) in which the concurrent administration to 50- to 79-yr-old women of equine estrogens (CEE; Premarin) plus the androgen- and corticoid receptor-binding progestin medroxyprogesterone resulted in increased intravascular clotting and failed to show cardio-protection. Despite serious design problems in the WHI, the hazard of all menopausal hormone treatment was widely, although not uniformly, accepted and the administration of MHT decreased by ~40% (58, 80). For a decade or more, doctors and patients have turned against the use of estrogen in the postmenopausal woman. While the expected mortality of this outcome is still being disputed (91a, 127a), the profusion of non-estrogen remedies for symptoms, bone and genital health, and continuing rise in metabolic syndrome-related conditions is seriously concerning (43, 58, 80). Importantly, subsequent subanalyses and prospective observational studies by the WHI on women who had received conjugated estrogens alone (ET) have confirmed the protection by estrogen (80, 83).
Additional cardio-studies have shown imaging evidence of athero-protection by estrogen, with or without progestins (51, 63).

The mechanisms of aromatase’s/estrogen’s cardio-protection are under study (12, 131). We will describe our relevant studies, but space limitations do not allow the description of the many interesting reports by others (89).

**Estrogen prevents monocyte capture by endothelial cells.** Arteriosclerosis is driven by inflammation. To develop plaque, it is necessary to create a collection of subendothelial monocytes/macrophages that will become foam cells (78). The adhesion of circulating monocytes to endothelial cells in response to vascular signals of inflammation (cytokines, growth factors) is the initial step in the formation of arterial plaque (77). The capture of monocytes requires the deployment of neural cell adhesion molecule (NCAM) by the mutually attracted monocytes and endothelial cells. This allows the formation of NCAM:NCAM binding that tethers the monocytes to the vascular endothelium (50, 78). Estrogen receptors (ER) and aromatase are expressed in the endothelium of human coronary and other vessels (39), and ER is expressed by monocytes (91). We recently showed the presence of estrogen-sensitive neural cell adhesion molecule (NCAM) sialylases in vascular endothelium (107). Since polysialylated NCAM (PSA-NCAM) does not adhere to apposed NCAM molecules, we tested and showed the expression of estradiol-induced sialylation of NCAM in human vascular endothelium (107) (**FIGURE 1**). We then grew in vitro monolayers of human arterial endothelial cells (HAEC) and showed that estradiol (and its precursor steroids testosterone and dehydroepiandros-}

**FIGURE 1. Cultured human umbilical vessel endothelial cell**
A cultured human umbilical vessel endothelial cell (HUVEC) stained for immunoreactive polysialylated neural cell adhesion molecule (PSA-NCAM; arrow). Note that the cell is polarized, showing the tuft of PSA-NCAM strands only at one area of the cell. The sialylation of NCAM blocks the formation of NCAM: NCAM tethers that arrest cells flowing through the vessels (107).

...terone) inhibits the adhesion of monocytes to the endothelial cells (35) (**FIGURE 2**). The activity of the estrogen precursor steroids implies that local aromatase may have a role in their estrogen-like action. These studies support an estrogen-driven means of interfering with monocyte capture and the formation of plaque (39). Further studies are underway.

**Brain Development and Homeostasis by Aromatase/Estrogen**

Aromatase is expressed by the brain in the human fetus, human adult, as well as many other species (10, 99). We showed that this is the basis of sexual differentiation in the hypothalamus (82). Estrogen is neuroprotective (86, 104, 125). The hippocampus expresses aromatase, and estradiol induces the development of dendritic spines by hippocampal dendrites. The spine length positively regulates long-term potentiation of neurons and could influence such functions as memory (159). Our imaging studies showing that estradiol affects brain activity have been confirmed and extended by others (129).

**Brain Function**

We and others have shown imaging evidence exists of estrogen action on brain function; thus far, this is largely descriptive and not mechanistic (130).

The hippocampus acts like a computer chip, receiving sensory system messages and distributing incoming information to many brain areas for action (73). Although there is clear evidence of estrogen’s lengthening hippocampal dendritic spines, which should increase pre-processing of incoming information and improve memory (155), there is no evidence that estrogen has other than an immediate effect on memory dysfunction following loss of ovarian function. The effect of estrogen on human memory remains unresolved (62, 132). However, the KEEPS long-term trial of menopausal hormone therapy in recently menopausal women confirmed the mood-sparing effect of estrogen plus progesterone (53).

**Bone Homeostasis by Aromatase/Estrogen**

Menopause or other loss of gonadal steroids results in osteoporosis. Administration of estrogen prevents this loss in women and men. Since aromatase is expressed by both osteoblasts and osteoclasts, it must be that local formation of estrogen is not sufficient to maintain bone homeostasis (154).

It would be remiss not to remark on the importance of aromatase and bone homeostasis in men. Studies of aromatase-deficient men have shown unfused epiphyses and osteopenia, and a remarkably similar phenotype to elderly men with a CYP19 gene polymorphism that alters aromatase
activity, which all highlight the importance of aromatase in bone homeostasis in men (52). Additionally, age-related decline in estrogen can predict bone mass in men similar to that of postmenopausal women (70) via the process of bone resorption (45). These issues grow in importance as the administration of aromatase inhibitors to men becomes more common.

**Genital Homeostasis by Aromatase/Estrogen**

Aromatase is expressed by many of the genital organs, and they are highly dependent on estrogen for their growth and maintenance. However, genital atrophy is rapidly evident following loss of ovarian estrogen secretion, again indicating that peripheral aromatization is not sufficient to maintain normal bone mass genital health or function (69, 158).

**Other Aromatase/Estrogen Effects**

There are other homeostatic actions of aromatase/estrogen throughout the body, including regulation of glucose homeostasis (85); however, space does not allow a complete treatment.

**Aromatase and Disease**

As with other enzymes, abnormal aromatase expression due to inappropriate gene regulation, or defects or mutations in the gene or aromatase protein itself can have clinical effects. In such cases, there could be clinical value in regulating aromatase action, either as a primary goal or as a way of blocking unwanted effects of aromatase products.

**Aromatase Abnormalities**

Complete absence of the CYP19A1 gene is an autosomal recessive disorder (98). In females, this can result in ambiguous genitalia at birth and/or hypergonadotrophic hypogonadism at puberty with no development of secondary sex characteristics. The presence of androgens from the adrenal gland can cause progressive virilization. The first report of this disorder was in an 18-yr-old with primary amenorrhea and pseudohermaphroditism who was eventually found to have had two missense mutations in the heme-binding region of the aromatase gene (33, 140). In males, lacking aromatase has a smaller clinical effect, with normal genitalia and pubertal maturation; however, they tend to be extremely tall due to delayed epiphyseal closures, develop osteoporosis, and have decreased libido (25). The most significant early warning sign of an affected fetus is maternal virilization due to faulty placental clearance of androgens, but the disease is often not found until after birth or puberty (36). Low aromatase in adipose tissue also results in insulin resistance and an abnormal lipid profile (160).

**Disorders Secondary to Aromatase Dysregulation**

Lack of gonadotrophins or ovarian failure results in decreased aromatase expression. This can have secondary effects on organs that rely on estrogen, such as the bones, brain, liver, skin, and genital tissues (165). For example, delayed increase in gonadal aromatase and estrogen formation leads to delayed puberty, late epiphyseal closure, and tall stature (22). These cases are treated as forms of premature gonadal failure, usually by administration of sex-appropriate estrogen or androgen, or, in cases where fertility is an issue, by gonadotropin or GnRH administration. On the other hand, precocious increase in ovarian aromatase/estrogen results in precocious puberty, including early epiphyseal closure and short stature (46). In many cases, the abnormalities during puberty are caused by inappropriate mutation or function of the hypothalamic mechanism and effects on GnRH/gonadotropin secretion. Uncommonly, tumors of the brain or pituitary may cause dysregulation of gonadotrophin secretion or hormone-secreting tumors of the gonad result in precocious puberty, but the cause is not determined in most instances. These “functional” dysregulations are treated with aromatase inhibitors or GnRH downregulation (46). Other examples of this dynamic can be seen especially related to reproduction and the balance of estrogen:androgen action and require similar treatment (49, 98, 145, 160).
Abnormalities in Aromatase/Estrogen Contribute to the Contemporary Causes of Illness and Death

During the past century, the average age at death doubled, and the most common causes of death changed from acute illnesses (hemorrhage, infection, etc.) to chronic diseases of aging. This age-related shift in causes of illness and death to represent arteriosclerotic vascular disease, diabetes, and degenerative brain disease and cancer is related to metabolic syndrome, risky lifestyle, and diet and inflammatory tone. As the population ages and becomes increasingly aromatase/estrogen deficient, the diversity and importance of aromatase/estrogen’s effects is increasingly appreciated (43, 111, 119).

Absence/Overexpression of Estrogen Can Result in Immunopathology

The immune system depends on aromatase/estrogen for homeostasis. The body is patrolled by immunocytes that are first-responders to injury. This includes the microglia in the nervous system. It has previously been shown that monocytes can penetrate areas of axonal damage in the brain and convert into microglia, which may help the brain regenerate and aid in plasticity (11).

We have used the increased understanding of the mechanism(s) by which aromatase/estrogen regulates to growth and death of immunocytes to characterize homeostasis (maintenance of normal interactions between tissues and first-responder immunocytes; FIGURE 3). Note that aromatase/estrogen regulates the Fas-FasL, the CD40L-CD40, and the TGFβR-TGFβ systems, as well as the myriad of cytokines and growth factors involved in immune homeostasis.

Aromatase/Estrogen and the Onset of Cancer

Postmenopausal estrogen treatment has been shown to protect against the diagnosis of invasive breast duct cancer (see above). Estrogen-related cancers have long been studied, and there are no reports of direct effects

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**FIGURE 3.** Estrogen regulates the cellular immune response

*Figure* shows that estrogen regulates the cellular immune response to constantly occurring micro-injuries in the brain by regulating immunocyte proliferation and death (apoptosis). In the absence of the modulating influence of estrogen, the cellular immune response may attack neurons, fibers of passage, and other normal components in the area to drive a cycle of auto-degeneration apparent as pathology. *Figure* is modified from Ref. 137 and used with permission.
of the primary estrogens (estrone, estradiol, estriol) causing cancer in humans. Study of clear cell vaginal cancers in children whose mothers received diethylstilbestrol did not reveal genomic clues to the origin of the cancers (18). Similarly, the in vivo progression of breast ducts and endometrial epithelial cells through malignant transformation has not revealed a direct effect of estradiol, except for excessive proliferation that might be associated with increased opportunities to undergo genetic mutation. Proposals that metabolites of estradiol could form adducts that result in genetic abnormalities have not been confirmed in vivo, although the search continues for the presence of this mechanism in humans (26).

The same is true for endometrial cancer, in which case the focus has shifted from aromatase/estrogen to the role of metabolic diseases (153a). The mechanism of tamoxifen (a mixed agonist-antagonist selective estrogen receptor modulator)-induced endometrial cancer remains obscure (64). To be sure, there are many reports of in vitro malignant transformation in the presence of estrogen, and animal models have shown similar outcomes, and the acid test production of malignant transformation of normally proliferating cells to cancers has yet to be reported.

**Estrogen Prevention of the Diagnosis of Cancer**

Although the anti-cancer role of estrogen has been seen in the case of breast cancers (29), the reported decrease in colon cancer diagnosis and deaths remains uncertain (23, 37, 74). Since most reports in this area deal with diagnosis rather than prevalence, they are difficult to interpret; that is, the reports may be dealing with effects on tumor progression rather than tumor initiation.

**Recent Developments**

Translational science has allowed for vast and rapid advances in the fields of medicine and human biology and physiology. In this section, we will discuss current translational uses and recent developments in the world of aromatase.

**Aromatase Inhibitors**

Numerous aromatase inhibitors (AI) have been developed. The first aromatase inhibitors were nonspecific anti-cytochrome p450 agents. The most tested and used clinically is amnioglutethimide, which was originally developed as an anticonvulsant. It was found to have monoamine oxidase activity and is soporific. This was a severe drawback for the use of amnioglutethimide, although its anti-p450 actions were employed for acute treatment as a “medical adrenalectomy” in a select subgroup of breast cancer patients (127).

As the AI field progressed, two types of specific AI were developed: type 1 were steroid analogs of androstenedione that irreversibly bind to the same binding site as normal aromatase substrates and “inactivate” or stop the enzyme from functioning. Type 2 AIs were nonsteroidal molecules that bind reversibly to the heme group of the aromatase enzyme. The type 2 AIs were developed clinically, and three “generations” are in clinical use today. Third-generation AIs are approximately three times as potent as their predecessors. The third-generation, nonsteroidal AIs in widespread use today are letrozole and anastrozole (143).

The AI adverse effects are the same as other blockers of gonadal function. However, the menopausal symptoms are more severe, the psychological aberrations are more often reported, and bone mineral loss is worse (66). In cases of long-term AI treatment, there are reports of accelerated cardiovascular disease compared with the lack of such progress among women receiving SERMs (5, 128). When feasible in cases of treatment of nonmalignant diseases, AI treatment may be accompanied by administration of low-dose estrogen to diminish symptoms, etc., so-called “add-back” progestin or estrogen therapy (1).

**Use of AIs against breast cancer.** One of the major uses of AIs is in risk reduction for recurrence and adjunctive treatment of estrogen receptor-positive breast cancer (8). Ductal breast cancer is currently the most common cancer of women in western Europe/North America, and the use of aromatase inhibitors challenges the current “gold standard” use of the SERM tamoxifen and the lesser used raloxifene (16, 146). The theory is that, unlike SERMs, the inhibition or inactivation of aromatase suppresses serum estrogen levels and has no partial agonist activity (143). Studies show that tamoxifen and raloxifene are approximately one-third less effective than aromatase inhibitors in preventing new breast cancers or as adjunctive treatments for active breast cancers (5). However, the length of life of the SERM-treated women is about the same as that of AI-treated women (5). At this point, the regimens remain unsettled (146), and concerns have been raised regarding cardiovascular complications of AIs (8, 164) and mood disturbances (116). Since decreased estrogen has been implicated in cardiovascular disease and depression, the marked effect of AIs compared with partial agonist SERMs is in keeping with present knowledge (see above).

**Use of AIs in men.** Prostate cancer expresses aromatase and estrogen receptors (66). However, although there has been some study into specialized treatment with AIs in prostate and other cancers (19), treatment with AIs has been disappointing. It is thought that alternative pathways
develop to obviate the effects of aromatase inhibition. As well, there is important loss of bone mineral mass in these men (20) and even some data on use of AIs on prepubertal boys of short stature to delay epiphyseal closure (84).

There have been several additional uses of AIs in men. For example, improved hormonal profiles have been seen in men who struggle with infertility and low testosterone (108, 114). Additionally, there have been some positive case reports on AI use in cognition (28) and even in male breast cancer (6).

**Infertility treatment.** In addition to satisfactory experience in inducing ovulation with the SERM clomiphene, the use of AIs appears to furnish better chances of ovulation induction without increasing the risk of ovarian hyper-stimulation than does the administration of gonadotropins (48).

**Endometriosis treatment.** The finding of aromatase overexpression in endometriosis led to successful treatment with AI (21). However, this treatment is still in the formative stages (109).

**Leiomyomas and AIs.** Uterine leiomyomas express estrogen receptors and respond to gonadotrophin suppression by high-dose GnRH. Therefore, clinical trials using AIs have begun. While it is too early to predict the long-term outcomes, the results are promising for tumor shrinkage and improved surgical results (75).

**Conclusions**

Begun as a chemist’s explanation of the synthesis of estrogen (126, 136), studies on aromatase/estrogen have grown in importance as the role and mechanisms of action of estrogens have come front and center (10, 14, 24, 82, 90, 99, 104). And, while xenoestrogens, especially the plant and incidental nonsteroidal SERMs, have increasingly claimed attention, aromatase and its products and blocking agents have kept their important place in biology and medicine (38, 157). The role of gonadotrophins in driving the gonadal aromatase, the role of peripheral aromatization, the effects of testosterone clearance, and substitution by 100- to 1,000-fold more potent estradiol clarifies why small amounts of aromatase are critical to bodily function.

Moreover, the presence of tissue-specific co-transactivational factors in individual organs means that only biological testing can determine the qualitative effects of estrogens or SERMs.

Estrogen is the preferred treatment for prevention of menopausal symptoms and complications, and protection against aging-related diseases and metabolic syndrome/inflammation-related diseases, including Type 2 diabetes and brain dystrophies (58, 80, 138).

The medical use of aromatase inhibitors and the complications of anti-aromatase molecules will be pertinent for decades to come (4, 5, 8, 68, 103, 104, 128). However, a word of caution: Because these drugs infringe the myriad of known, essential estrogen actions, pharmacovigilence of the effects of AI will be of particular importance in the use of aromatase blockers and targeted aromatase stimulators—when they become available. ■

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**References**


10. Balthazart GF, Ball J. New insights into the regulation and revised manuscript; J.K.B. and F.N. approved final version of manuscript.


