Human steroid biosynthesis for the oncologist

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Abstract

In 2005, results from the Arimidex, Tamoxifen Alone or in Combination (ATAC) trial ushered in a new era of endocrine therapy for hormone-responsive malignancies. This study demonstrated that, compared to tamoxifen (a selective estrogen receptor modulator [SERM]), anastrozole (aromatase inhibitor [AI]) prolonged time to recurrence and disease-free survival for postmenopausal women with breast cancer. The advantage was even greater for those with estrogen receptor-positive (ER^+) tumors, and anastrozole was better tolerated than tamoxifen. Since then, AIs have become first-line adjuvant therapy for ER+ breast cancer in postmenopausal women.

In late 2010, a trial comparing abiraterone acetate (a 17-hydroxylase/17,20-lyase [CYP17A1] inhibitor) plus prednisone versus prednisone alone in men with castration resistant-prostate cancer (CRPC) previously treated with docetaxol chemotherapy was terminated early due to the survival benefit in the abiraterone acetate arm. This result not only validated a new therapy for CRPC but, with the antecedent phase I-II abiraterone studies, shattered our understanding of the molecular mechanisms underpinning CRPC development and progression.

AIs and CYP17A1 inhibitors will be widely used by oncologists, yet fellowship programs provide little training in steroid biosynthesis, compared with training in the biology of standard chemotherapies. Consequently, these drugs might be used without an appreciation of their caveats and pitfalls. The purpose of this review is to acquaint practicing oncologists with the fundamental principles and pathways of steroid biosynthesis, to improve their understanding of how and why these drugs work, and to alert these physicians to potential problems related to the drugs' mechanisms of action.

Introduction

The hormonal dependence of breast and prostate cancers has been exploited for decades as a therapeutic strategy. Dramatic and lifesaving tumor regression or stabilization was first achieved with surgical oophorectomy in breast cancer [1] and orchiectomy in prostate cancer [2], but recurrences were common despite elimination of all gonad-derived androgens or estrogens, respectively. Recognizing that the adrenal glands provides a source of androgen precursors, surgical adrenalectomy was added to ovariectomy for breast cancer [3], and later chemical adrenalectomy with aminoglutethimide proved equivalent [4]. Efforts to provide more complete and less invasive hormone ablation strategies in prostate cancer led to the use of estrogens such as diethylstilbestrol (DES) and progestins [5], later replaced by

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long-acting gonadotropin-releasing hormone (GnRH) agonists such as leuprolide acetate [6]. Androgen receptor (AR) antagonists such as bicalutamide and nilutimide provided the next contribution to therapy in prostate cancer [7], whereas in breast cancer, the anti-estrogen tamoxifen, which is actually a selective estrogen receptor modulator or SERM, became the reigning standard of care for many years [8] until dethroned by the aromatase inhibitors (AIs) in the past decade [9]. Abiraterone acetate, a 17-hydroxylase/17,20-lyase (CYP17A1) and thus androgen biosynthesis inhibitor, has been recently approved for the treatment of castration resistant-prostate cancer (CRPC) [10]. Consequently, the evolution of treatment has generally progressed from surgery to receptor antagonists to hormone synthesis inhibitors. Since these therapies should theoretically all achieve the same purpose, why are the enzyme inhibitors gaining favor over time?

To understand how these drugs work and why they might provide more complete hormone ablation than other therapies, a basic understanding of steroid biosynthesis is essential. Unfortunately, oncology training programs rarely provide more than a rudimentary introduction to steroidogenesis in their curricula. In this article, we will develop a logical and clinically relevant framework to understand steroid biosynthesis in normal and pathologic states. These basic principles will then illustrate why these drugs are more effective in some patient populations than in others, when these drugs should be combined with other agents, and how resistance develops. A comprehensive review of human steroid biosynthesis has recently appeared [11], and the reader is directed to this reference for any details not covered herein.

The big picture

All steroid hormones derive from cholesterol, and in normal human physiology, only a few tissues convert 27-carbon cholesterol to 21-carbon pregnenolone, the first committed intermediate in steroidogenesis. The adrenal cortex, Leydig cells of the testis, theca and granulosa cells of the ovary, and trophoblasts of the placenta are the only cells in the body capable of synthesizing enough pregnenolone to contribute to circulating concentrations of steroids [12]. Subsequent conversions within the steroid-producing cells yield either active hormones or hormone precursors, and the exact products depend on which downstream enzymes are expressed in those cells (Figure 1A,B). These steroids enter the circulation to act on target cells, but extensive metabolism occurs outside the steroidogenic tissues. Consequently, circulating concentrations of steroids only partially reflect their biological activity, and different cells each experience a unique hormonal milieu depending on the balance of enzymes and other factors present in each cell [13]. For example, circulating testosterone (T) is actually three potential hormones. T binding to AR elicits androgenic effects in some tissues such as spermatic tubules, stimulating sperm production. In the prostate, 5α-reductase irreversibly converts T to dihydrotestosterone (DHT), which unlike T uniquely mediates formation of the prostate and the male external genitalia in fetal life, as well as development of prostatic hyperplasia [14]. Conversely, aromatase activity in breast tissue metabolizes T to the potent estrogen estradiol (E2), which if excessive causes gynecomastia (Figure 1C).

The first step is the hardest

The conversion of cholesterol to pregnenolone is generally the slowest, most complex, and acutely regulated step in steroid production [15]. The side chain cleavage enzyme (P450scc or CYP11A1) uniquely catalyzes this transformation, which really involves three chemical steps, at a maximum rate of roughly 7 turnovers per minute, pathetically slow for an enzyme. In addition, *access* of cholesterol to CYP11A1 is restricted topologically within the specialized mitochondria of steroidogenic cells. The mobile pool of cholesterol capable of

entering steroid biosynthesis resides on the outer mitochondrial membrane, whereas CYP11A1 and its cofactor protein ferredoxin reductase (FdxR) are attached to the inner mitochondrial membrane (Figure 2). In the mitochondrial matrix, electrons from reduced nicotinamide adenine dinucleotide phosphate (NADPH) channel from FdxR to a soluble iron-sulfur protein ferredoxin (Fdx) and then to CYP11A1. This reaction uses two electrons and one molecule of oxygen in each of three cycles to cleave cholesterol to pregnenolone. Every pulse of luteinizing hormone (LH) yields a corresponding pulse of T via the induction of intracellular cyclic adenosine monophosphate (cAMP), which "opens the gate" for cholesterol entry to the steroidogenic pathway. The familiar cosyntropin stimulation test also elicits a sharp rise in cortisol in 30 min, due to cAMP elevation and cholesterol mobilization. How does this rapid mobilization of cholesterol occur?

The protein that drives cholesterol from the outer to inner mitochondrial membranes is the steroidogenic acute regulatory (StAR) protein [16] (Figure 2), whose mechanism of action is incompletely understood. The x-ray crystal structure of the StAR homolog MLN64 contains a binding pocket for one molecule of cholesterol [17], suggesting that StAR physically carries cholesterol across the intermembranous space; however, other evidence suggests that StAR acts while residing on the outer membrane [18]. StAR phosphorylation is required for the activation and termination of its action [19], reflective of its transient nature. Some steroidogenic tissues such as the placenta do not express StAR yet still produce abundant steroids, possibly using MLN64 instead. In model cells, StAR co-expression increases pregnenolone production 7-fold above that observed using the CYP11A1 catalytic system alone [20].

Conceivably, additional tissues, including cancer cells, might convert cholesterol to pregnenolone, and evidence supporting this mechanism of T synthesis in CRPC progression has been found in some [21],[22] but not all studies [23]. Nevertheless, for a cell to convert cholesterol to pregnenolone, the minimum requirements include expression of CYP11A1, FdxR, and Fdx; sufficient cholesterol, oxygen, and NADPH content in a permissive mitochondrial environment; and ideally StAR and its phophorylation machinery. Of course, the quantities of pregnenolone synthesis required for local *de novo* T production sufficient to stimulate CRPC growth are many orders of magnitude lower than that required to raise circulating steroid concentrations, challenging the limits of detection by modern assay methods.

Steroidogenesis is directional

An important concept in steroid biosynthesis is that most steps are irreversible and that classes of endogenous steroid hormones are defined both by their biologic activities and their chemical structures. These structures, in turn, are determined by the activities of those enzymes catalyzing their biosynthesis, and the nomenclature is mercifully helpful for the novice in most cases. Most of these enzymes are cytochromes P450, and their naming is thus CYP followed by number, letter, and number (CYP17A1, CYP3A4, etc) [24]. In addition, the 5α-reductase reactions are irreversible (Figure 1B).

The terminal steps of steroidogenesis and the peripheral conversions are mediated by the many hydroxysteroid dehydrogenases (HSDs) (Figure 1A,B), and these reactions are reversible, although each enzyme has a strong directional preference in intact cells [25],[26]. Thus, pairs of enzymes drive steroid flux in opposite directions, one from hydroxysteroid to ketosteroid and another back from ketosteroid to hydroxysteroid. An example is 17βHSD3, the critical enzyme for converting androstenedione (AD) to T in Leydig cells. Boys with 17βHSD3 deficiency are born with ambiguous genitalia from T and DHT deficiency *in utero* [27]. Conversely, 17βHSD2 oxidizes T almost completely to AD in peripheral tissues,

attenuating the androgenic activity of T in cells containing this enzyme [28]. One caveat regarding the reversibility of these transformations is that the two 3βHSD-isomerase enzymes [29], by virtue of their second ($\Delta^5 \rightarrow \Delta^4$-isomerase) activity, also mediate irreversible reactions (Figure 1C). The many human HSD enzymes are each encoded by separate genes with characteristic tissue-specific expression profiles, catalytic activities, and substrate preferences [30]. The function of only a few enzymes are known with certainty, particularly for those expressed in peripheral tissues, and many of these enzymes are poor catalysts for HSD reactions. Consequently, a cancer cell might contain several 17βHSD isoforms competing for steroid substrates, and the final biologic potency of an entering steroid will be determined by the net result of these activities.

Another mechanism of terminating steroid action is conjugation, similar to the "Phase II" conjugation reactions of drug metabolism, including glucuronidation and sulfation. For steroids, free hydroxyl groups are sulfated or glucuronidation by sulfotransferases (SULT) and uridine diphosphate glucuronyltransferase (UGT) enzymes, respectively, which also have a number, letter, and number nomenclature (SULT2A1, UGT2B17) [31]. The UGT enzymes include UGT1A1, a major bilirubin conjugating enzyme; low UGT1A1 activity causes Gilbert's disease and predisposes to irinotecan toxicity [32]. Glucuronylation is generally an irreversible process, which tags the steroid for excretion in the urine by organic anion transporters, whereas sulfation increases protein binding and prolongs circulating half-life. At least some steroid sulfates are desulfated in specific tissues, such as estrone sulfate conversion to estrone in breast cancer cells [33].

You can go your own way

The classical pathways have been constructed based on the dominant enzyme activities and hormone concentrations observed in normal adrenal and gonadal tissues, reinforced by changes in flux observed in enzyme deficiency states. Nevertheless, minor pathways also exist, but we hide these "under the rug" since their contributions are insignificant during normal physiology. For example, 21-deoxycortisol (progesterone hydroxylated in the 17- and 11-positions) is made in the normal adrenal but is not part of a mainstream pathway and generally not measured by clinical laboratories. Elevated 21-deoxycortisol, however, is a more specific marker steroid for 21-hydroxylase deficiency than the customary 17-hydroxyprogesterone [34], and 21-deoxycortisol is used as a second-tier newborn screening test in some states.

In addition, more than one pathway might converge at the same product. While pathway A might be the major route to steroid X in tissues whose primary job is to export X as a circulating hormone, other tissues might use pathway B or C. How is DHEA converted to T and DHT, as must occur to explain hirsutism in women? Well, textbooks usually say DHEA → AD → T → DHT (enzyme sequence 3βHSD, 17βHSD, SRD5A), with these reactions occurring mainly in liver and skin. There is no reason why DHEA should not be converted to androst-5-ene-3β,17β-diol (A5diol) via 17βHSD first and then by 3βHSD to T, and this sequence is probably dominant in the normal testis [35] (Figure 1B). Furthermore, we have recently shown that CRPC specimens and cell lines predominantly use the sequence DHEA → AD → 5α-androstane-3,17-dione (5αdione) → DHT (enzyme sequence 3βHSD, SRD5A1, 17βHSD), entirely bypassing T [36] (Figure 3). Even more bizarre, a complex alternate or "backdoor" pathway to DHT in the tammar wallaby pouch young testis involves 5α-reduction of 17-hydroxyprogesterone prior to the 17,20-lyase reaction of CYP17A1 [37,38]. In this pathway, the testis exports the 5α-reduced androgens androsterone and 5α-androstane-3α,17β-diol, that latter of which is oxidized to DHT in the target tissues to mediate male prostate and genital differentiation in that species [39]. The lesson is that

alternate routes might bypass blocks in classical pathways, as long as enzymes capable of catalyzing these reactions are expressed in the tissue and cells of interest.

Feedback mechanisms or lack thereof

Under normal circumstances, tropic hormones regulate steroid production, and the product steroid then suppresses its own production in a classical negative feedback loop. In the adrenal, adrenocorticotropin (ACTH) stimulates production of cortisol and dehydroepiandrosterone sulfate (DHEAS), yet only cortisol provides negative feedback on ACTH (Figure 4A). Similarly, luteinizing hormone (LH) stimulates androgen production by the Leydig cells of the testis (mainly T, Figure 4B) and theca cells of the ovary (mainly AD), and these androgens suppress LH production. In the ovary, follicle-stimulating hormone (FSH) acts on the granulosa cells surrounding the developing oocyte to induce aromatase production, thus allowing conversion of AD and T to E2 (Figure 4C), and FSH also induces small amounts of E2 production in the testes. E2 and even progesterone also suppress LH production, whereas FSH negative feedback is mainly due to inhibin B, which is a protein hormone derived from the Sertoli cells of the testes and granulosa cells of the ovary.

Based on the paradigms described, damage to the pituitary (tumor, trauma, radiation) will lower LH, FSH, and ACTH production and therefore reduce production of T, E2, cortisol, DHEAS, and all the precursor steroids and metabolites as well. Surgical removal of the adrenal glands raises ACTH, which can be lowered with exogenous cortisol replacement. Gonadectomy will cause LH and FSH to rise, and androgen or estrogen replacement lowers LH but not FSH, since inhibin B remains absent. Similarly, natural menopause occurs when all follicles and thus granulosa cells are depleted, and estradiol production falls while LH and more specifically FSH rise. Long-acting GnRH agonists act by desensitizing the pituitary gonadotropes and suppressing LH (and FSH) and therefore T and E2 synthesis. Supraphysiologic doses of exogenous androgens and estrogens or progestins also lower LH, T, and E2 production—but not FSH—but directly exert hormone actions on the body. For example, high-dose DES, a potent estrogen, is used to treat prostate cancer by increasing negative feedback on the gonadal axis, which suppresses LH and thus T production. The estrogenic action of DES, however, causes gynecomastia in a high proportion of patients and predisposes to thrombotic events, limiting its utility. Similarly, dexamethasone or prednisone suppress ACTH and thus both cortisol and DHEAS synthesis. Steroid receptor antagonists relieve the negative feedback of these hormones and cause a rise in the tropic hormones from the pituitary and subsequently the endogenous steroids themselves.

T and DHT in prostate cancer

Based on the preceding discussion, several strategies to block T and DHT synthesis and/or action can be envisioned, and most of these have already found application in clinical practice (Figure 5). Surgical orchiectomy, DES, leuprolide acetate, and bicalutamide are all successful examples of different strategies to block testicular T production or action, and dutasteride inhibits conversion of T to DHT by 5α-reductase enzymes. The problem with AR antagonists is that most are partial agonists or incomplete antagonists, and up-regulation of LH and thus T might overcome the blockade, prompting the development of more potent compounds such as MDV3100 [40]. Gonadal suppression strategies fail to address the abundant DHEAS and DHEA derived from human adrenal gland, which are only 3 or 2 steps upstream of T (Figure 3), with redundant pathways mediating these transformations [13]. Adrenal androgen elimination in CRPC probably explains the additional therapeutic benefit of ketoconazole [41], a CYP17A1 inhibitor, and dexamethasone or prednisone, which suppress ACTH and thus adrenal DHEA(S) production [42].

Similarly, abiraterone is a potent and specific CYP17A1 inhibitor, explaining its recent success in treating CRPC [10,43,44]. Despite its ideal molecular target and potency, abiraterone is not used as monotherapy. Like AR antagonists, inhibition of T synthesis will remove negative feedback and increase LH production in an effort to restore homeostasis. The mechanism of abiraterone action involves irreversible binding to the heme iron in the active site of CYP17A1, so inhibition continues even after the drug has been completely cleared from the circulation, until new enzyme is synthesized. Consequently, abiraterone is used with GnRH agonist, to maintain LH suppression and prevent the synthesis of new CYP17A1 protein in the testis. When studied in noncastrate men with prostate cancer, abiraterone alone lowers T only transiently to castrate levels [45], and therefore abiraterone is unlikely to be effective as monotherapy in prostate cancer.

In the pivotal abiraterone trial, all participants received prednisone 5 mg twice daily as well, which was generally started during docetaxol treatment. Since abiraterone also ablates adrenal CYP17A1 activity, prednisone might seem unnecessary for efficacy, but CYP17A1 inhibition also blocks cortisol synthesis and induces a syndrome of 17-hydroxylase deficiency [46,47] (17OHD, Figure 5). Biglieri described the clinical and biochemical consequences of 17OHD, a rare form of congenital adrenal hyperplasia, nearly a half-century ago [48]. Loss of negative feedback from cortisol deficiency causes ACTH to rise, activating StAR, driving cholesterol into steroidogenesis, and flooding the available downstream enzymes with pregnenolone (Figure 6). The block at CYP17A1 leaves only pathways involving 3βHSD2, 21-hydroxylase (CYP21A2), and 11β-hydroxylase (CYP11B1) and limits steroidogenesis to corticosterone and its precursor 11-deoxycorticosterone (DOC). Steroid flux in the adrenal of a patient with 17OHD resembles that of the rodent adrenal, which also lacks CYP17A1 and makes corticosterone as the principal glucocorticoid. In 17OHD, circulating corticosterone concentrations rise over 100 times normal to >10,000 ng/dL (~1 μM) in a new steady-state [47], which prevents glucocorticoid insufficiency in most patients [46]. In order to make this much corticosterone, however, DOC production also rises markedly, and at these concentrations (>10 nM), DOC is a potent mineralocorticoid receptor (MR) agonist, inducing volume expansion with kaluresis and suppressing plasma renin activity (Figure 6). Consequently, the clinical presentation of complete 17OHD is sexual infantilism regardless of chromosomal sex, hypertension, and hypokalemia [48].

In the phase I-II trials of abiraterone, prednisone was omitted [43,44], and hypertension and hypokalemia were commonly observed. DOC and corticosterone rose markedly, as is seen in genetic 17OHD [49]. These consequences were treated effectively with addition of either MR antagonist to block the action of DOC on MR (eplerenone in these studies) or corticosteroid to prevent the ACTH rise and thus lower DOC production. Thus, prednisone co-therapy with abiraterone acetate serves two purposes: to limit synthesis of new adrenal CYP17A1, which might overcome of the blockade in DHEAS synthesis in between abiraterone acetate doses, and to prevent accumulation of DOC with its propensity to cause hypertension and hypokalemia. This triple-regimen is the evolution of the "total androgen blockade" concept proposed by Labrie [50] upon considering all potential androgen sources and peripheral steroid metabolic pathways. It is possible that smaller prednisone doses and other glucocorticoids might accomplish the same purposes, but other regimens have not been studied.

In considering other targets, only three enzymes besides CYP17A1 are required for T synthesis (Figure 3). CYP11A1 and 3βHSD inhibitors would block synthesis of all steroid hormones and cause adrenal insufficiency as well. A 17βHSD inhibitor, acting on the final step, might seem ideal, but many enzymes catalyze this reaction besides testicular 17βHSD3, so the exact target(s) are unknown. Although AKR1C3 (17βHSD5) is highly

expressed in many CRPC specimens [51], inhibition of this enzyme might select for clones that express alternative 17βHSD isoenzymes. In contrast, CYP17A1 is the ONLY enzyme that converts pregnenolone to DHEA, so this target leaves no alternative pathway to T open. The relative contributions of the *de novo* steroidogenesis from cholesterol all the way to T and DHT versus terminal metabolism of adrenal-derived DHEA(S) within the CRPC tumor to androgens is debated and likely to vary amongst specific tumors [52]. Regardless of origin, complete blockade is of all CYP17A1 activity will ablate all sources of T and DHT production, but the complete endocrinologic consequences of these manipulations must be considered to optimize patient outcomes.

Estrogens in breast cancer

Similarly, steroidogenesis in the ovary of reproductive-age women derives from feedback-regulated axes as in the male, but steroid and germ cell production in the female is cyclical, adding to complexities beyond the scope of this article. Since E2 is the major hormone providing negative feedback, inhibition of aromatase will increase LH and FSH production, which increases aromatase expression in an effort to overcome the blockade (Figure 4C). When AIs are administered to premenopausal women, E2 decreases only transiently, and subsequently a new steady-state is reached. In contrast, the postmenopausal ovary makes little or no estrogen, and E2 deriveds from androgen precursors (DHEAS, AD) of ovarian and adrenal origin [53]. The aromatase enzyme in postmenopausal women is expressed mainly in adipose tissues, including those of the breast [54], but also in breast cancers themselves [55]. Since LH and FSH, which are already elevated in postmenopausal women, do not regulate aromatase expression in these tissues, the potent, irreversible AIs used clinically are very effective in suppressing E2 synthesis in postmenopausal women and treating ER+ breast cancers in these women [56].

In contrast, the use of AIs in premenopausal women with ER+ breast cancer would be ineffective, unless combined with therapy to prevent overcoming the blockade. Such treatments might include surgical or chemical oophorectomy and suppression of LH and FSH with high-dose progestins or long-acting GnRH agonists. Preliminary reports have demonstrated the feasibility of "total estrogen blockade" in premenopausal women with ER+ breast cancers using leuprorelin and anastrazole [57]. Whether such combination therapies will prove effective in this population remains unknown.

Because AI therapy in women who have ceased menses for over 12 months—and thus are clinically menopausal—effectively suppresses E2 and eliminates most negative feedback on the gonadotropes, LH and FSH might rise even higher. In some women, this gonadotropin surge, which mimics the mid-cycle surge, induces ovulation and/or menses in some women [58]. Often only one cycle is experienced, but this phenomenon, which is rather distressing to patient and doctor alike, also brings into question whether the menopausal status has been correctly assigned. Will such patients still benefit from AI therapy? In general, patients with a single episode of menses on an AI can be considered postmenopausal, although serum estradiol measurements before and during AI therapy are required in this setting [59]. As long as serum estradiol remains low, AI therapy remains an option.

Similar to the tumor progression typically observed in CRPC, tumor progression eventually occurs in most cases of metastatic breast cancer treated with AIs. The mechanisms responsible for the escape from control are being studied vigorously in several groups. Serum concentrations of E2 remain markedly reduced at progression in nearly all cases [60],[61], although intratumoral concentrations of E2 might be sufficient to activate ER yet elude detection in the most sensitive assays. Furthermore, the steroids accumulating above

the AI blockade are androgens, and AR is expressed in many breast cancers [62]. Could these tumors be converting from estrogen-dependent to androgen-dependent growth mechanisms? In addition, androgen metabolites such as androstanediols also accumulate, and some of these compounds activate ER, thwarting the intention of the treatment [63]. In either case, strategies similar to those employed in CRPC might be effective salvage therapies, by blocking all androgen and estrogen production, and some trials testing this hypothesis are underway.

Summary and conclusions

Endocrine therapy has been a mainstay for the treatment of breast and prostate cancer for decades, and enzyme inhibitors have taken center stage as first-line therapies in recent years. Treatment strategies to eliminate production of key steroids must consider the optimal targets, alternate pathways that might bypass the blockade, and the endocrine consequences of deranged steroid flux. In many instances, combination therapies with other agents are necessary to prevent side-effects of upstream steroids and/or physiologic adaptations, which might overcome the blockades. Every new drug brings its unique challenges, which derive from the basic physiology of how the biochemical machinery in different tissues orchestrates the synthesis of steroid hormones. Armed with an understanding of these fundamental concepts and principles of steroid biosynthesis, the practicing oncologist will be prepared to avoid these pitfalls and optimally manage the prostate and breast cancer patients on these treatments.
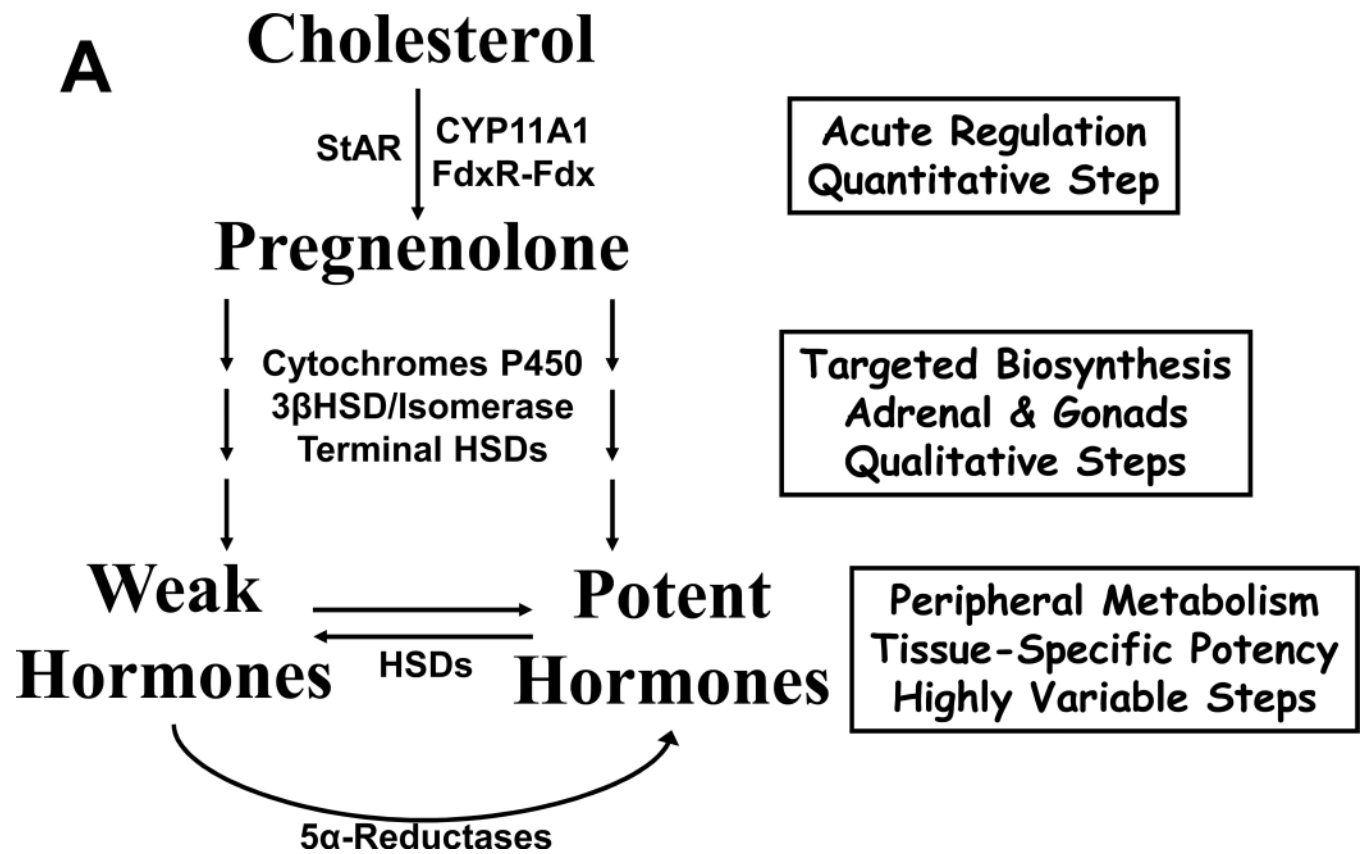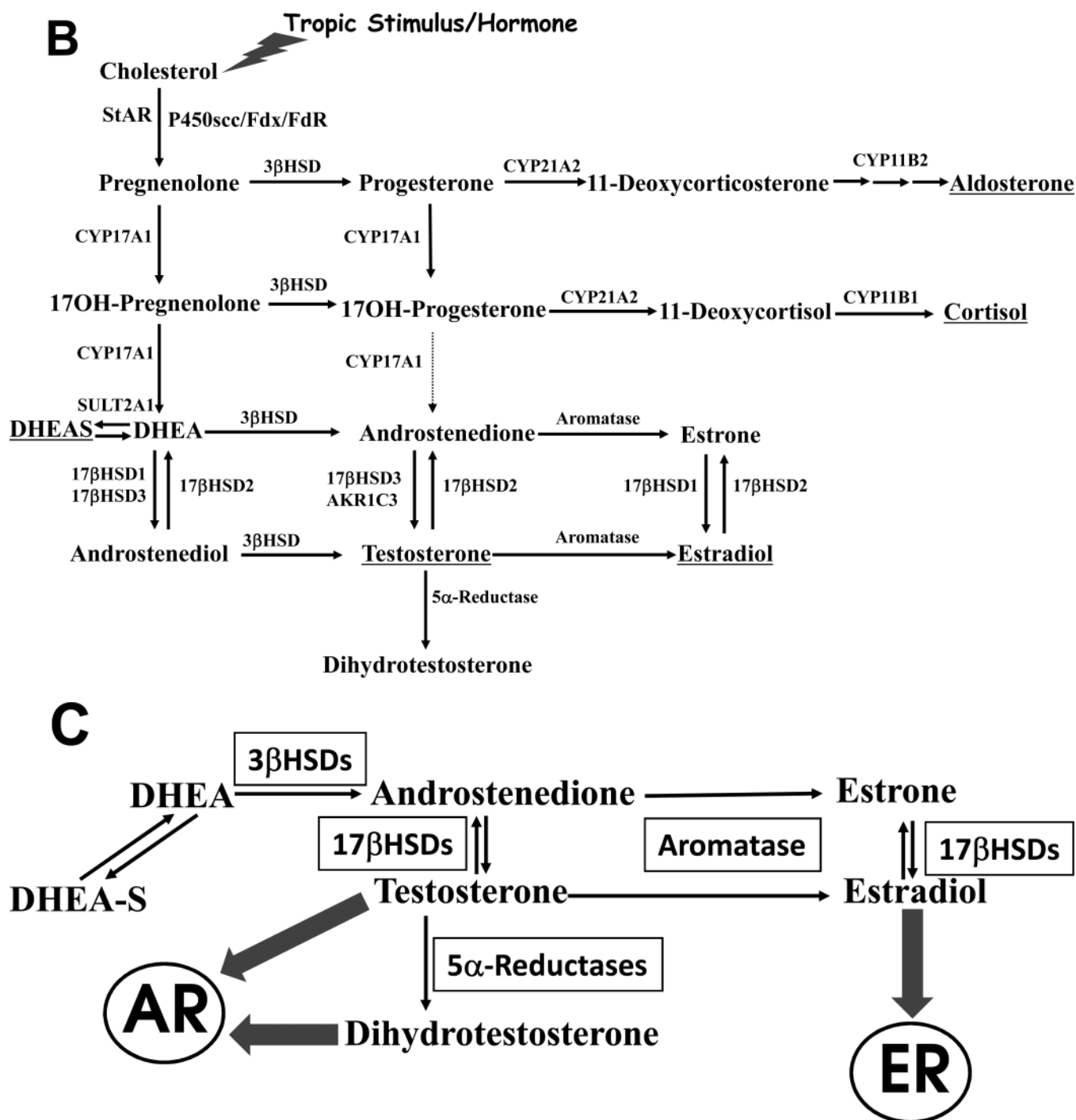
Acknowledgments

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A   Cholesterol

StAR | CYP11A1
       FdxR-Fdx

Pregnenolone

Cytochromes P450
3βHSD/Isomerase
Terminal HSDs

Weak
Hormones   → ← HSDs   Potent
                                 Hormones

5α-Reductases

Acute Regulation
Quantitative Step

Targeted Biosynthesis
Adrenal & Gonads
Qualitative Steps

Peripheral Metabolism
Tissue-Specific Potency
Highly Variable Steps

FIGURE 1.
A, A conceptual model for steroid biosynthesis. Classical steroid production begins with a tropic stimulus, which drives the conversion of cholesterol to pregnenolone and maintenance of the downstream enzymes in the steroidogenic cell. Peripheral tissues and target cells further metabolize these steroids. B, Major steroidogenic pathways in human beings. Enzymes are indicated above and aside arrows, and main product steroids of the adrenals, testis, and ovary are underlined. Thin line from 17-hydroxy-(17OH)-progesterone to androstenedione indicates minor pathway. C, Canonical terminal steps of T synthesis from

DHEA(S) and fates of T in different tissues. T acts directly on AR or indirectly after metabolism to DHT, and aromatase changes the biological activity of T via metabolism to E2.
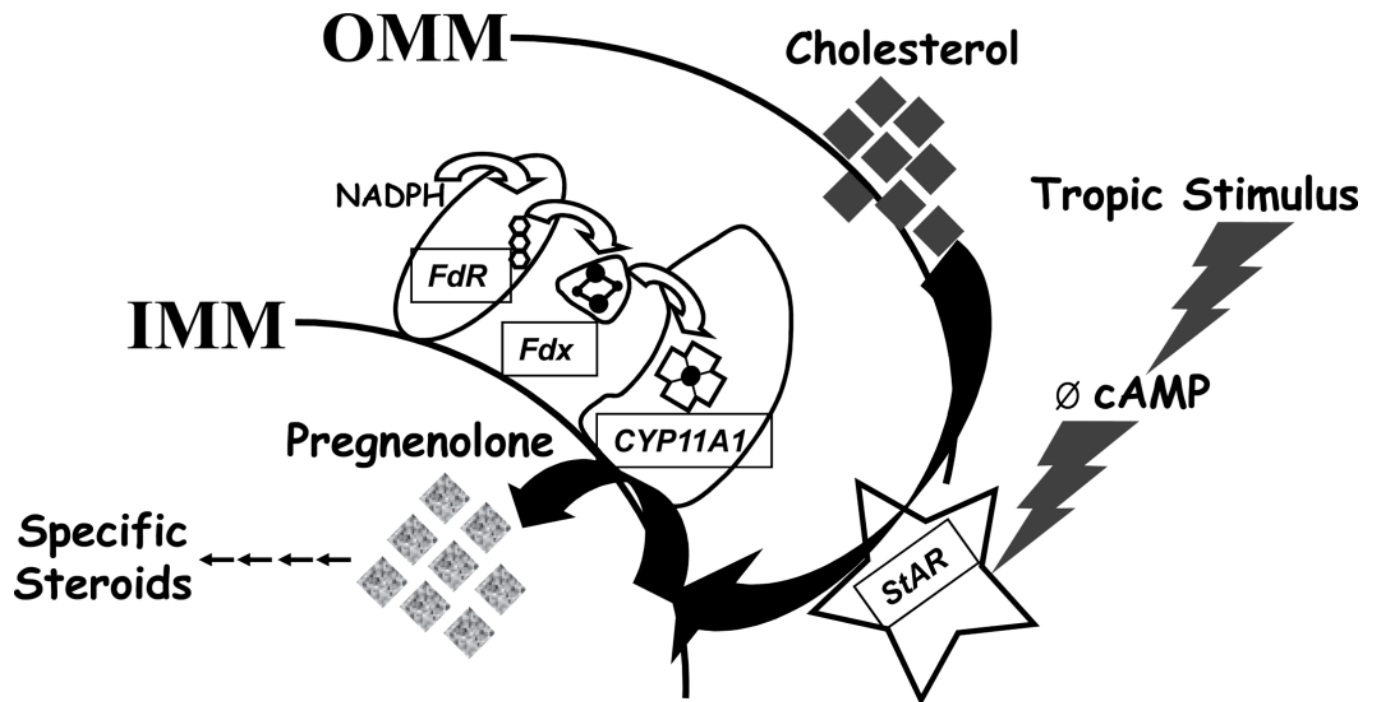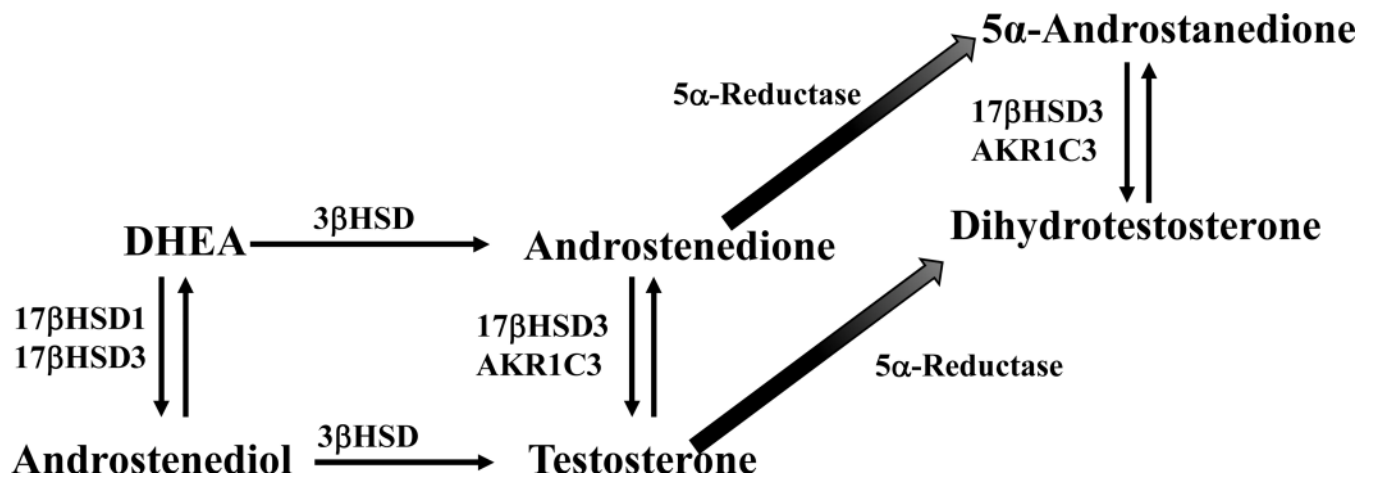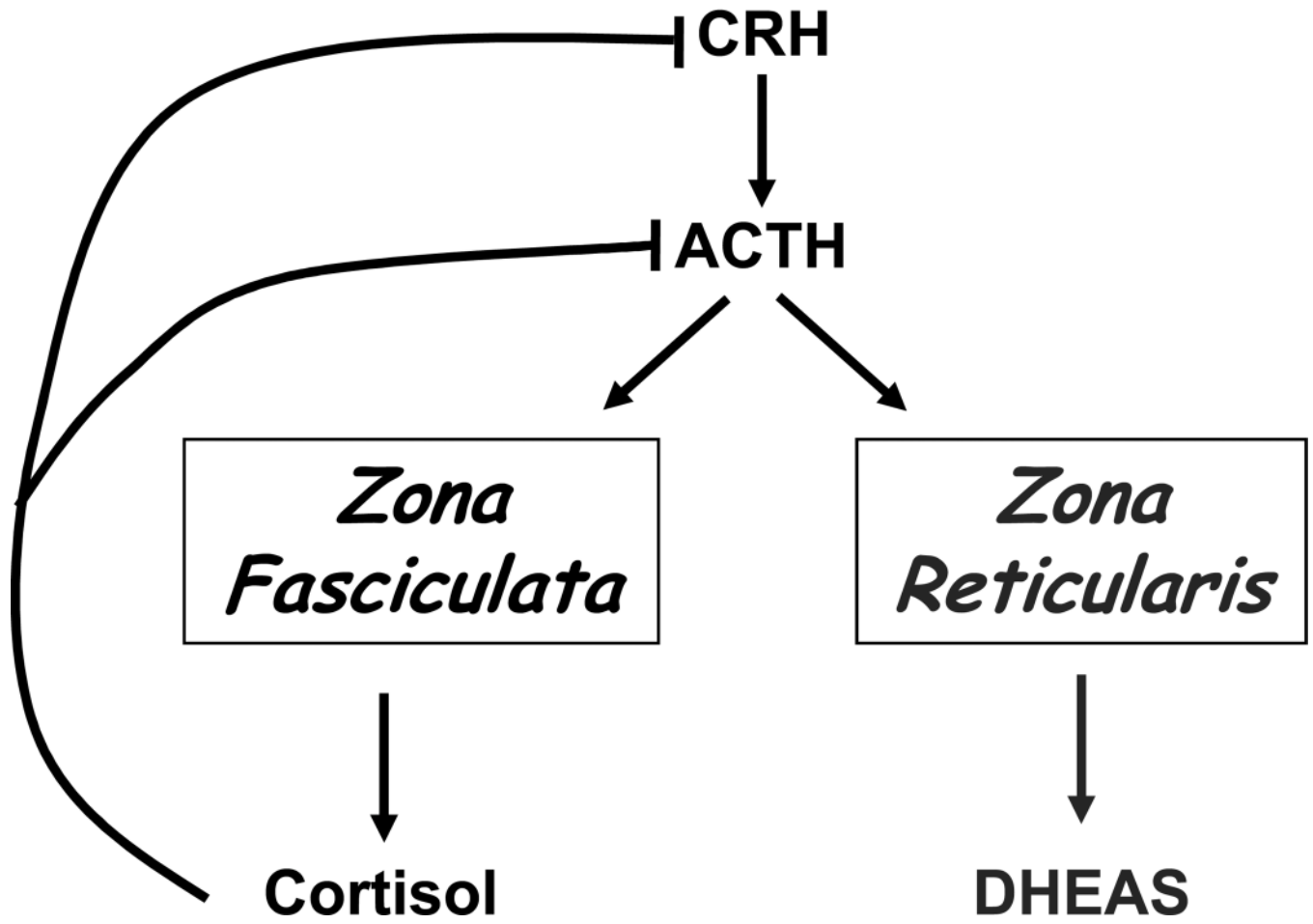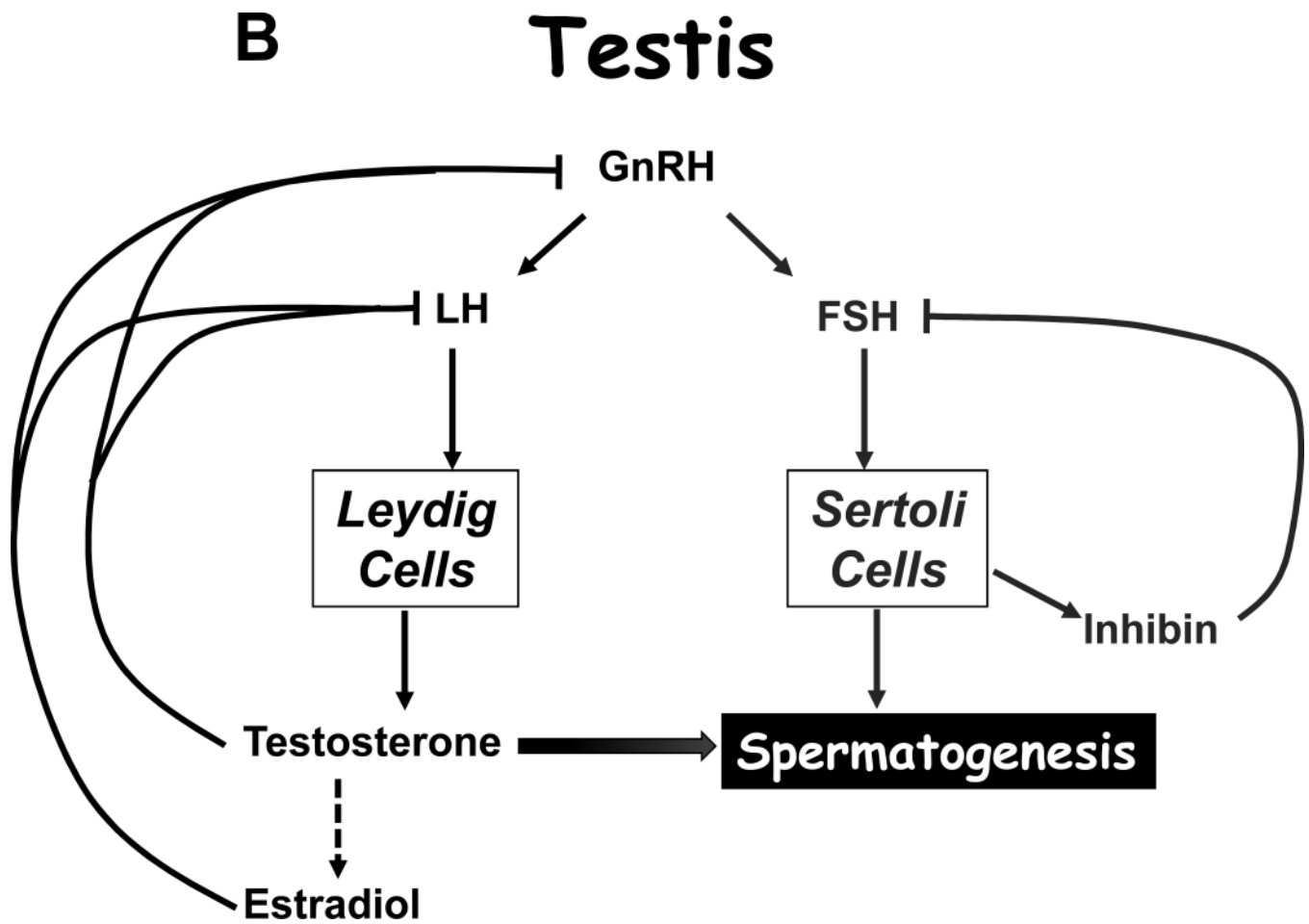
FIGURE 2.
Initiation of steroid biosynthesis in the mitochondria via StAR and CYP11A1. Stimulated by increased cAMP, StAR induces the movement (large solid arrow) of cholesterol in the outer mitochondrial membrane (OMM) to the inner mitochondrial membrane (IMM), where FdR and CYP11A1 reside. Electrons from NADPH flow (open arrows) to the flavin (three hexagons) in FdR, to the iron-sulfur cluster (dots and lines) in the soluble mitochondrial matrix protein Fdx to the heme (Maltese cross and dot) of CYP11A1. These electrons plus molecular oxygen enable CYP11A1 to convert cholesterol to pregnenolone (small solid arrow) in three cycles, and pregnenolone is then liberated into pathways, which lead to specific steroids (cortisol, T, etc.).

FIGURE 3.
Termninal steps of androgen biosynthesis from DHEA. In this pseudo-three-dimensional diagram, reactions to the right (3βHSD) and to the back (5α-reductase) are irreversible, whereas vertical reactions are reversible and catalyzed by several 17βHSD enzymes. Flux from DHEA to DHT might follow several pathways, and an individual steroid molecule might cycle vertically many times before finally moving through the 3βHSD and/or 5α-reductase reactions.

A   Adrenal

CRH

ACTH

Zona
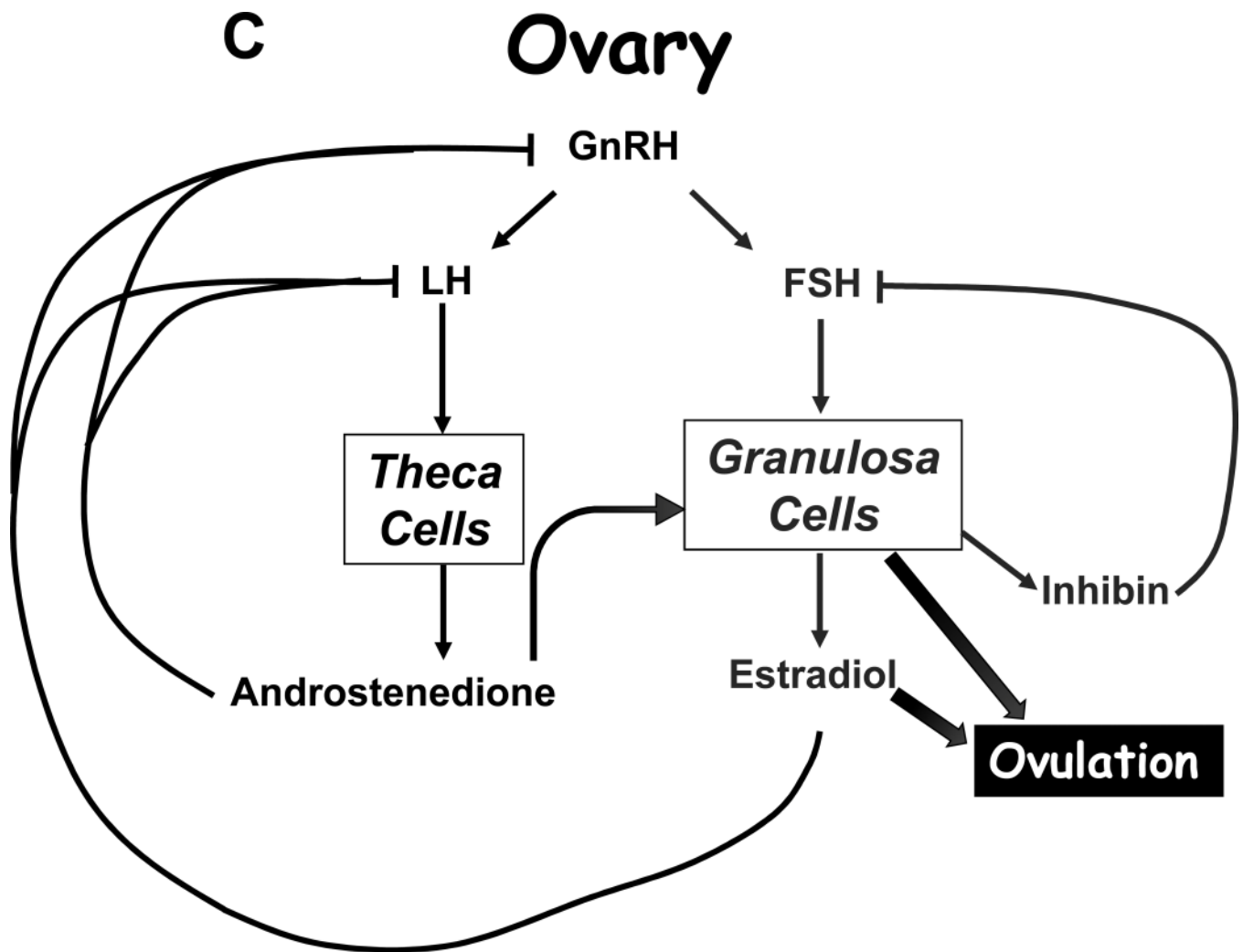Fasciculata

Zona
Reticularis

Cortisol

DHEAS

FIGURE 4.
Hypothalamic-pituitary-target gland axes for the A, adrenal; B, testis; and C, ovary. Block
arrows indicate actions of hormones, and flat end to lines indicate negative feedback.
Curved block arrow in panel C indicates that androstenedione from the theca cell diffuses to
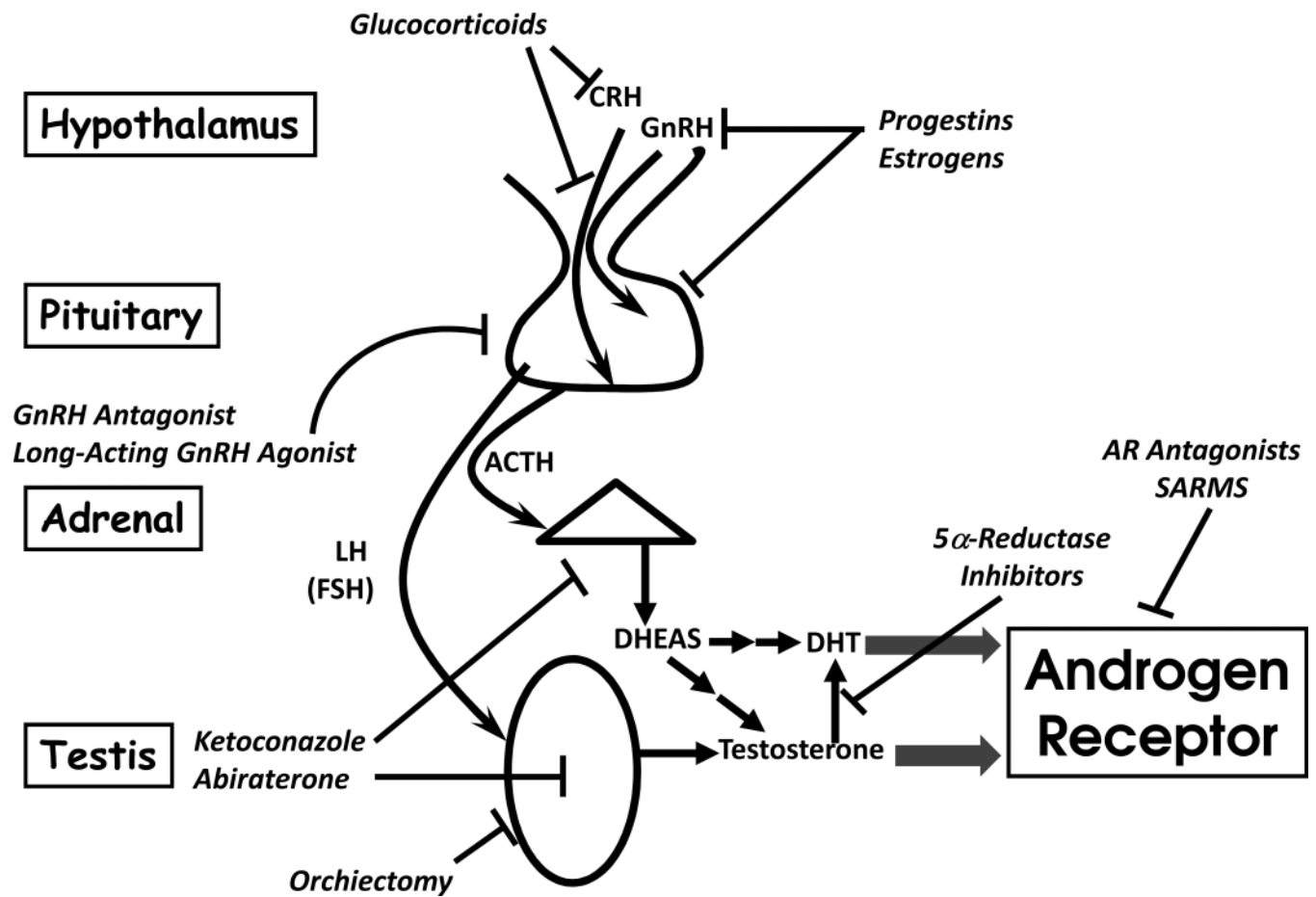the granulosa cell, where aromatization occurs.

FIGURE 5.
Endocrine therapy strategies for prostate cancer, which all inhibit androgen production or antagonize the androgen receptor. Flat end to lines indicate site of action on process or tissues, including hypothalamus and pituitary (top), adrenal (triangle in middle), and testis (oval at bottom). Block arrows indicate receptor activation.
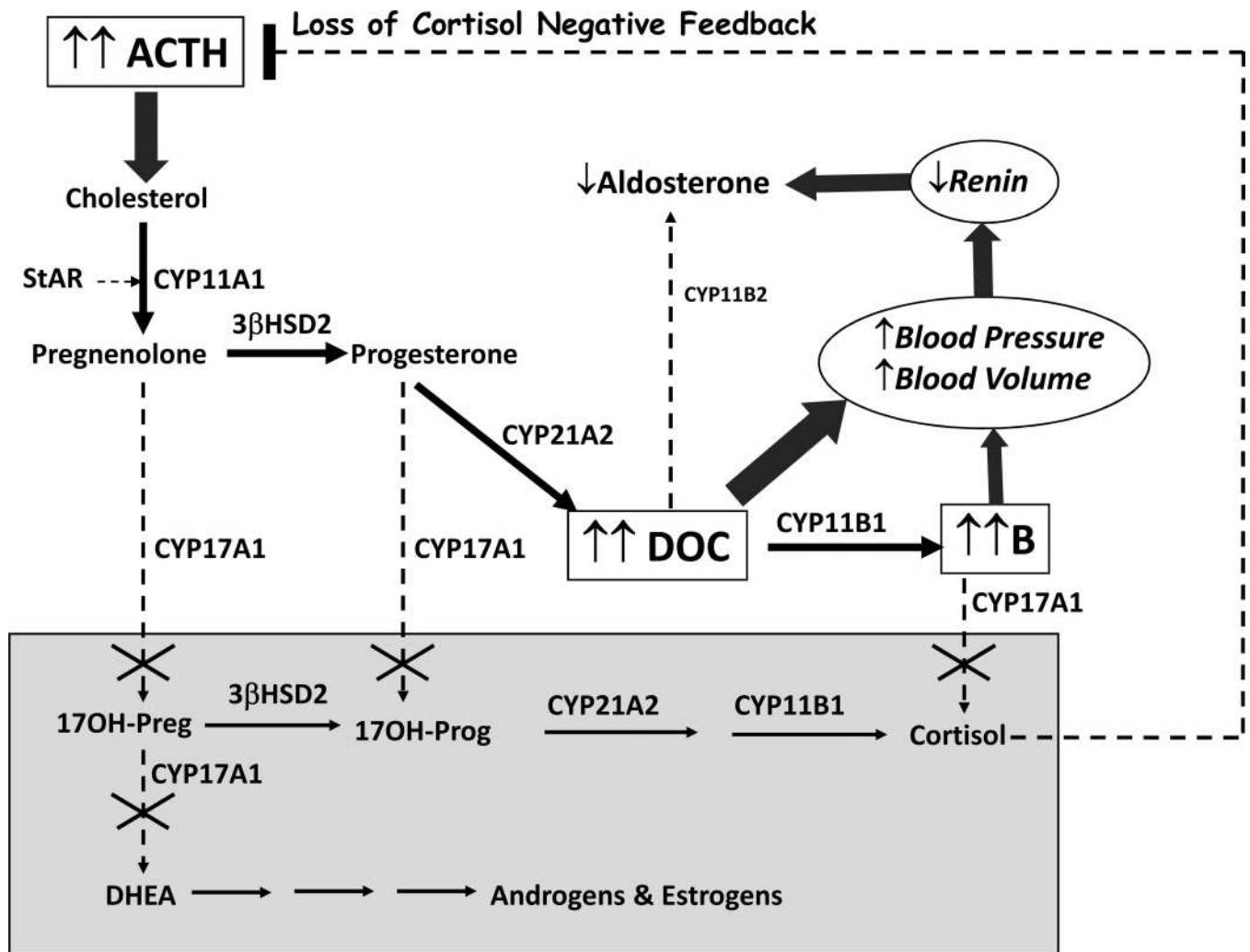
FIGURE 6.
Physiology of genetic 17-hydroxylase deficiency, equivalent to abiraterone acetate monotherapy. Loss of CYP17A1 activities (dashed arrows with X) eliminates production of all steroids in grey box, including androgens, estrogens, and cortisol. Loss of cortisol negative feedback increases ACTH and flux of cholesterol to pregnenolone (block arrow). The only available pathways (thick arrows) go to DOC and corticosterone (B), which expand blood volume, increase blood pressure, and suppress both plasma renin activity and aldosterone production (block arrows).