2. Gonadal hormone synthesis, secretion, transport, metabolism, action
a. Steroid hormones
(1). Biosynthesis
Understand the physiologic and clinical importance of free (unbound) sex steroid hormone concentrations and the regulation of SHBG
Free or unbound sex steroid hormone is not bound to proteins and is the active form in cells/organs.

SHBG is increased by estrogen, tamoxifen, phenytoin, hyperthyroidism, and cirrhosis

SHBG is decreased by exogenous androgens, glucocorticoids, GH (acromegaly), hypothyroidism, obesity, and hyperinsulineamic states.

Know the genes and biosynthetic pathways contributing to androgen production

In Leydig cell, precursor cholesterol synthesized de novo or from plasma pool (receptor-mediated endocytosis of LDL). Cholesterol transferred by StAR protein to inner mitochondrial membrane (stimulated by LH, which also upregulates the biosynthetic enzymes). Side chain cleaved once by CYP11A1, then 17-hydroxylation and cleavage of 17,20 bond by CYP17 (17α-hydroxylase and 17,20-lyase), then side chain cleaved again by 17β-HSD, and the A ring is oxidized 3β-HSD. Testosterone is major product of testis (5-6mg/day in men), but DHT, androsterone, 5α-androstane-3α, 17β-diol, androstenedione, 17-hydroxyprogesterone, progesterone, and pregnenolone are also made in small amounts.
Testosterone is converted in peripheral tissues (primarily at androgen target tissues) to dihydrot testosterone by 5α-reductase (2 isoenzymes: type 1 encoded on chromosome 5; type 2 encoded on chromosome 2; type 1 expressed in skin and liver predominantly after puberty, also in sebaceous glands of scalp; type 2 in male urogenital tract early in embryogenesis, and in hair follicle of scalp. DHT is responsible for many aspects of male sexual development and virilization. DHT if further metabolized to 17-ketosteroids and excreted in the urine. Alternatively, testosterone and other androgens with Δ⁴, 3-keto configuration (like androstenedione) are converted to estrogens by aromatase in many extraglandular tissue but most significantly in adipose tissue.

Know the genes and biosynthetic pathways contributing to estrogen production
-In men, most of the estrogens are formed from peripheral aromatization of androgens. Only 15% comes from the testis (also by aromatization of androstenedione and testosterone).-In women: ovarian steroidogenesis requires involvement of both theca (androstenedione production) and granulosa (aromatization); peripheral aromatization of androgens in extraglandular tissues (mostly estrone, which is weaker, is produced)

(2). Specific steroids
(a). Testosterone (See also V.C.4.a.(3))
Know the organs that produce testosterone in men and women and the relative proportion secreted by each organ
-Men: Leydig cell of testis (98%); adrenal cortex in zona reticularis (1%); peripheral conversion androstenedione to testosterone (1%)
Women: theca cell of ovary (33%), peripheral conversion of androstenedione (produced by ovary and adrenal) to testosterone (67%)

Know the relative roles of secretion and peripheral metabolism in the production of testosterone in men and women
See above

Know the relative bioavailability of the plasma free, albumin-bound, and SHBG-bound fractions of plasma testosterone
Bioavailability: free = 100% available; albumin-bound = nearly all available; SHBG-bound = practically none available (1000-fold higher affinity for SHBG than albumin)

Know the factors regulating SHBG synthesis and its serum concentrations
See above

Know the metabolic fates of testosterone

Know the intracellular signaling pathway of androgens within target cells
Testosterone enters cells by passive diffusion. In cells that express 5α-reductase, converts to DHT.
Testosterone and DHT bind to same androgen receptor, but affinity is higher for DHT. Hormone-receptor complexes attach to hormone response elements in DNA to regulate transcription.
Testosterone-receptor complex doesn’t bind DNA as efficiently as DHT-receptor complex. This is why DHT formation so important in development. Androgen receptors highest concentration in organs of male reproduction, areas of the brain. Present in both Sertoli and Leydig cells. Small amounts in skeletal muscle, heart, vascular smooth muscle, and placenta.

Major actions of androgens: regulation of gonadotropin secretion, initiation and maintenance of spermatogenesis, formation of male phenotype during sexual differentiation, promotion of sexual maturation at puberty, and control of libido
Know the characteristics of anti-androgens and their potential uses
Block androgen synthesis: leuprolide acetate: GnRH agonist → decrease LH; medroxyprogesterone acetate inhibits androgen synthesis directly and also inhibits LH secretion; anti fungal agents of imidazole class (ketoconazole) block cytochrome P450 enzymes in steroid hormone biosynthesis; spironolactone also impairs androgen biosynthesis and also inhibits binding of androgen to androgen receptor

5α-reductase inhibition: selectively blocks androgen action in the tissues (prostate, hair follicles) – finasteride (Propecia)

Block action of androgens: androgen receptor antagonists (flutamide, cyproterone acetate)

Know the structure and function of the androgen receptor and the steroids to which they respond
Androgen receptor encoded on long arm of X chromosome. 917 amino acids. Active form is a homodimer. Hormone-binding and DNA-binding domains have high homology with progesterone, glucocorticoid, and mineralocorticoid receptors. N-terminus is quite different and is polymorphic

Know patterns of fetal concentrations of estrogens, progestins, androgens, and gonadotropins at varying developmental ages

(b). Estradiol (See also V.C.4.a.(3))
Understand the sex steroid pattern of the normal menstrual cycle
Fsh stimulates granulosa cells of a follicle. This causes increased production of estradiol. With conjunction with increased estradiol levels, FSH stimulates the granulosa cells to express more LH receptors. This leads to increase in progesterone and 17OH progesterone which may play a role in the
positive feedback on the estrogen primed pituitary that leads to the LH surge. FSH also stimulates several steroidogenic enzymes including CYP19, aromatase, and 3-B-hydroxysteroid dehydrogenase.

-LH principally stimulates androstenedione production and to a lesser degree testosterone production in the theca cell.

-After ovulation the corpus luteum forms and produces progesterone to maintain the endometrium after ovulation. CL is maintained by LH and then hCG once fertilization occurs.

<table>
<thead>
<tr>
<th>SEX STEROIDS*</th>
<th>Early Follicular</th>
<th>Preovulatory</th>
<th>Midluteal</th>
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<tr>
<td>Progesterone (mg)</td>
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</tr>
<tr>
<td>17-Hydroxyprogesterone (mg)</td>
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<td>4</td>
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<tr>
<td>Dehydroepiandrosterone (mg)</td>
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<tr>
<td>Androstenedione (mg)</td>
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<td>4.7</td>
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<td>Testosterone (Eg)</td>
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<td>126</td>
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<tr>
<td>Estrone (Eg)</td>
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<td>350</td>
<td>250</td>
</tr>
<tr>
<td>Estradiol (Eg)</td>
<td>36</td>
<td>380</td>
<td>250</td>
</tr>
</tbody>
</table>

From Baird DT, Fraser IS. Blood production and ovarian secretion rates of estradiol-17β and estrone in women throughout the menstrual cycle. J Clin Endocrinol Metab 38: 1009-1017. 1974. @ The Endocrine Society.

*Values are expressed in milligrams or micrograms per 24 hours.
Know the organs that secrete estradiol in males and females and the relative proportion secreted by each organ.

In males, 0.1% testosterone is converted to estradiol; 10-15% comes directly from the testes while the rest is from peripheral tissues such as adipose.

In females, androstenedione from the ovaries and adrenal glands can be converted peripherally to estrone and then estradiol, however majority of androstenedione formed by the theca cells is converted to estradiol by the granulosa cells.

Know the role of secretion relative to peripheral metabolism in production of estradiol in men and women.

See above.

**Know the relative bioavailability of the plasma free, albumin-bound, and SHBG-bound fraction of plasma estradiol**

The majority of estradiol is bound, although it has less binding affinity than testosterone for SHBG (38%), and more for albumin (60%) with 2% being unbound.

Know the metabolic fate of estradiol.

Deactivation includes conversion to less active estrogens such as estrone and estriol. Estriol is the major urinary metabolite. Estradiol is conjugated in the liver by sulfate and glucuronide formation and as such excreted via the kidneys. Some of the water soluble conjugates are excreted via the bile duct, and partly reabsorbed after hydrolysis from the intestinal tract.
Know the intracellular signaling pathway of estrogen action within target cells
Similar to testosterone

Know the characteristics of anti-estrogens and their potential uses
Block estrogen production:
- leuprolide acetate or GnRH agonist could be used for central precocious puberty which ultimately reduces ovarian estrogen production
- Aromatose inhibitors: testolactone; inhibits conversion of androgens to estrogens; in MAS this would lead to decreased ovarian estrogen production, decrease frequency of menses, and slow rate of bone maturation

Know the characteristics of estrogen analogues and their potential uses
- Estrogen blocker: tamoxifen; used in treatment of precocious puberty in MAS by blocking the effects of endogenous estrogen; also being used in early pubertal gynecomastia but its effectiveness has not been established
- diethylstilbestrol is a phosphorylated, nonsteroidal estrogen used as palliative treatment of advanced prostate cancer and metastatic Breast CA, and feminizing treatment of male transsexuals

(c). Progestins (See also V.C.4.a.(3))

Know the relationship of progesterone secretion to granulosa cell luteinization
After ovulation occurs the granulosa cells luteinize and become large cells. Through LH mediation and hCG mediation these cells produce progesterone.

Understand the mechanism of action of progesterone in target cells
Progesterone acts with PR and this ligand/receptor effects transcription similar to estrogen and testosterone/DHT.

Know the cells of origin of progesterone in testes and ovaries
In the preovulatory state theca cells are active in steroidogenesis while the granulose cell produce estradiol from androstenedione.

The granulose/large cells are more active in steroidogenesis and production of progesterone after ovulation occurs.

In testes, steroidogenesis occurs in the leydig cells.

Understand the synthesis of progesterone
Recognize the effects of progestins on fluid and electrolyte balance
Progestins +/- estradiol can increase aldosterone production leading to hypertension

Know the relative androgenicity of the synthetic progestins used in oral contraceptives
High: Norgestrel (0.3mg), levonorgestrel (0.15mg), norethindrone acetate (1.5-2.5mg)

Moderate: norethindrone (1mg); norethindrone acetate (1mg); ethynodiol (1mg), levonorgestrel triphasics

Low: desogestrel; drospirenone; norgestimate; norethindrone (0.4-0.5mg)

(d). Androstenedione
Know the secretion of androstenedione relative to testosterone by the interstitial cells of ovaries and testes
Ovaries secrete more androstenedione relative to testosterone while the testes secrete more testosterone relative to androstendione

Know the relative contribution of the peripheral metabolism of androstenedione to the synthesis of testosterone and estrone
Males, majority of androstenedione is converted to testosterone predominantly by the testes. Small proportion of androstenedione is converted to estrogen and testosterone peripherally. However the majority of estrogen comes from peripheral metabolism in males.

In females, majority of androstenedione is converted to estrone/estradiol. Small portion of androstendione goes into circulation and converted to androgens or estrogens.
(e). 17-hydroxyprogesterone
Know the cells of origin of 17-hydroxyprogesterone in testes and ovaries
Leydig cells in testes and Theca cells in the ovaries

b. Peptide hormones
(1). Müllerian inhibiting factor
Know the cell of origin of Müllerian inhibiting factor
-males: AMH is secreted by the fetal Sertoli cells with peak level occurring at the time of mullerian duct regression (9-12 weeks)
-female: small amounts from Granulosa
Know the control of Müllerian inhibiting factor and changes in concentrations throughout development
-AMH gene expression is activated by the SRY gene. AMH transcription is initiated by SOX9 with up-regulation being mediated by SF1, WT1, and GATA4
-concentrations of AMH rise from birth to relatively high levels in the first year in newborn males, decrease by age 10, and decrease further during puberty
Newborn females have low or non-detectable levels of AMH, which rise only slightly thereafter; serum AMH concentrations are virtually non-detectable in most girls just before puberty. (can be detected in adults)
Know the effects of Müllerian inhibiting factor

(2). Inhibin/activin
Know the cells of origin of inhibin/activin
-inhibin/activin are heterodimeric glycoprotein products primarily from the sertoli cells of the testes and the ovarian granulose cells
-a number of other tissues, including the adrenal, pituitary, and placenta, also synthesize these peptides.
Know the control of inhibin/activin secretion
- FSH induces synthesis and secretion of gonadal inhibin

Know the effects of inhibin/activin at the pituitary and gonadal levels
-pituitary effects:
- inhibins selectively suppress FSH secretion without simultaneous suppression of LH secretion; thus, they provide one of the mechanisms whereby the pituitary can release differential amounts of FSH and LH, even though there appears to be only a single gonadotropin-releasing factor
- activins have been shown to stimulate both basal and GnRH-induced FSH release from the anterior pituitary as well as increase FSH beta mRNA levels by enhancing transcription
-gonadal effects:
-activins and inhibins also have local actions within the ovary influencing granulosa cell growth and differentiation, the responsiveness of the ovary to gonadotropins, steroid hormone production, follicular development, and oocyte maturation

Know the cell of origin of follistatin
-pituitary protein follistatin which is a glycosylated single protein chain binds and inactivates activin

Know the control of follistatin secretion
A substantial portion of pituitary follistatin is likely to originate from gonadotropes and, therefore, is subject to regulation by factors that influence this cell type including GnRH, the autocrine action of activin B, inhibins, and gonadal steroids

↓ activin from αfollistatin ➔ ↓ FSH
Know the effects of follistatin at the pituitary level

(3). Testis determining gene

Know the locus of the SRY gene and its role in testis determination

- SRY is a member of a family of DNA-binding proteins bearing a high mobility group (HMG) box and maps to the short arm of the Y chromosome Yp11.3, very close to the pseudoautosomal region 1 (PAR1). The SRY gene product bends DNA and exposes target genes for transcription.

Par1 on Yp and PAR2 on Yq are the only regions of the Y chromosome that undergo meiotic recombination with homologous sequences of the X chromosome during male spermatogenesis.

- SRY gene produces a transcription factor that is involved in the differentiation of the bipotential gonad. SRY activates SOX9 and SF1, which stimulates proliferation of sertoli precursors. These precursors organize around germ cells into tubular cords and start secreting AMH.

Know that the SRY gene might be translocated to another chromosome and the results of such a change -proximity of SRY to PAR1 makes it susceptible to translocation to the X chromosome following aberrant recombination and provides an explanation for approx 88% of XX males and for a lower proportion of XY females

Know that SRY is a high mobility group gene and how it functions

See above