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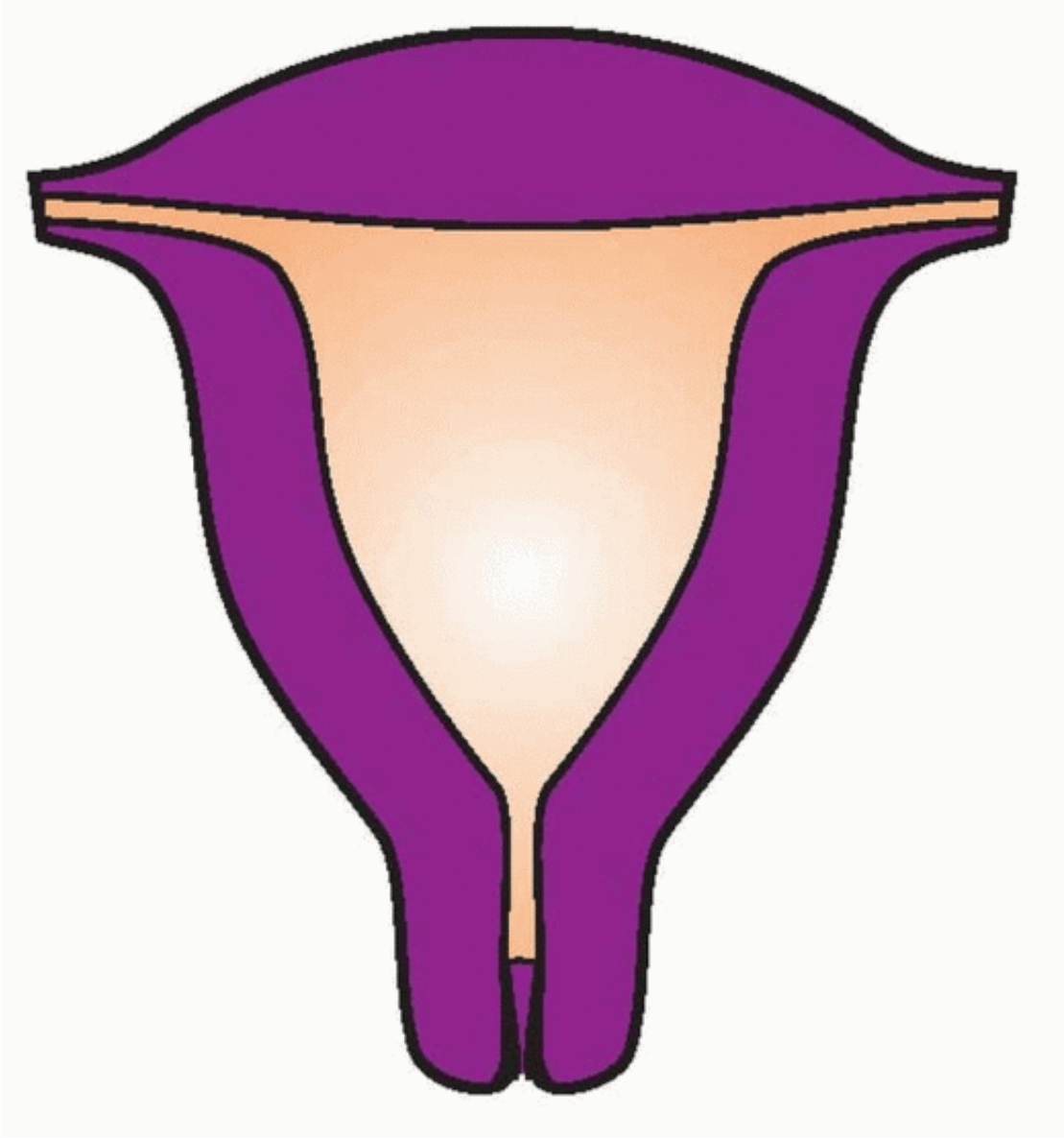
The Uterus



Anatomic knowledge of the uterus was slow to accumulate.[1,2] Papyrus writings from 2500 B.C. indicate that the ancient Egyptians made a distinction between the vagina and uterus. Because the dead had to be embalmed,

dissection was precluded, but prolapse was recognized because it was important to return the uterus into its proper place prior to mummification. Next to the Egyptian papyri in antiquity were Hindu writings in which descriptions of the uterus, tubes, and vagina indicate knowledge gained from dissections. This was probably the earliest description of the fallopian tubes.

There is little information in Greek writings about female anatomy; however, Herophilus (fourth century B.C.), the great anatomist in Alexandria and the originator of scholarly dissection, recorded the different positions of the uterus. Soranus of Ephesus (98-138 A.D.) accurately described the uterus (probably the first to do so), obviously from multiple dissections of cadavers. He recognized that the uterus is not essential for life, acknowledged the presence of leiomyomas, and treated prolapse with pessaries.

Herophilus and Soranus were uncertain about the function of the fallopian tubes, but Galen, Rufus, and Aetisu correctly guessed their function. Galen promoted the practice of bleeding for the treatment of almost every disorder. In his argument that nature prevented disease by discharging excess blood, Galen maintained that women were healthier because their superfluous blood was eliminated by menstruation.[3] The writings of Galen (130-200 A.D.) represented the knowledge of medicine for over 1,000 years until the end of the medieval Dark Ages. Galen's description of the uterus and tubes indicates that he had only seen the horned uteri of animals.

In the 16th century, Berengarius, Vesalius, Eustachius, and Fallopius made significant contributions to the anatomic study of the female genitalia. Berengarius (Giacomo Berengario da Carpi) was the first anatomist to work with an artist. His anatomic text, published in 1514, depicted dissected subjects as if they were still alive.

Gabriele Fallopio (or Fallopius) published his work, *Observationes Anatomicae*, in Venice in 1561, 1 year before his death from pleurisy at age 40. He provided the first descriptions of the clitoris and the hymen and the first exact descriptions of the ovaries and the tubes. He named the vagina and the placenta and called the tubes the uteri tuba (the trumpet of the uterus), but soon they were known universally as the fallopian tubes. It was his professor and mentor at the University of Padua, however, Andreas Vesalius, who was the first to accurately reveal the presence of the endometrial cavity.

Development of the Müllerian System

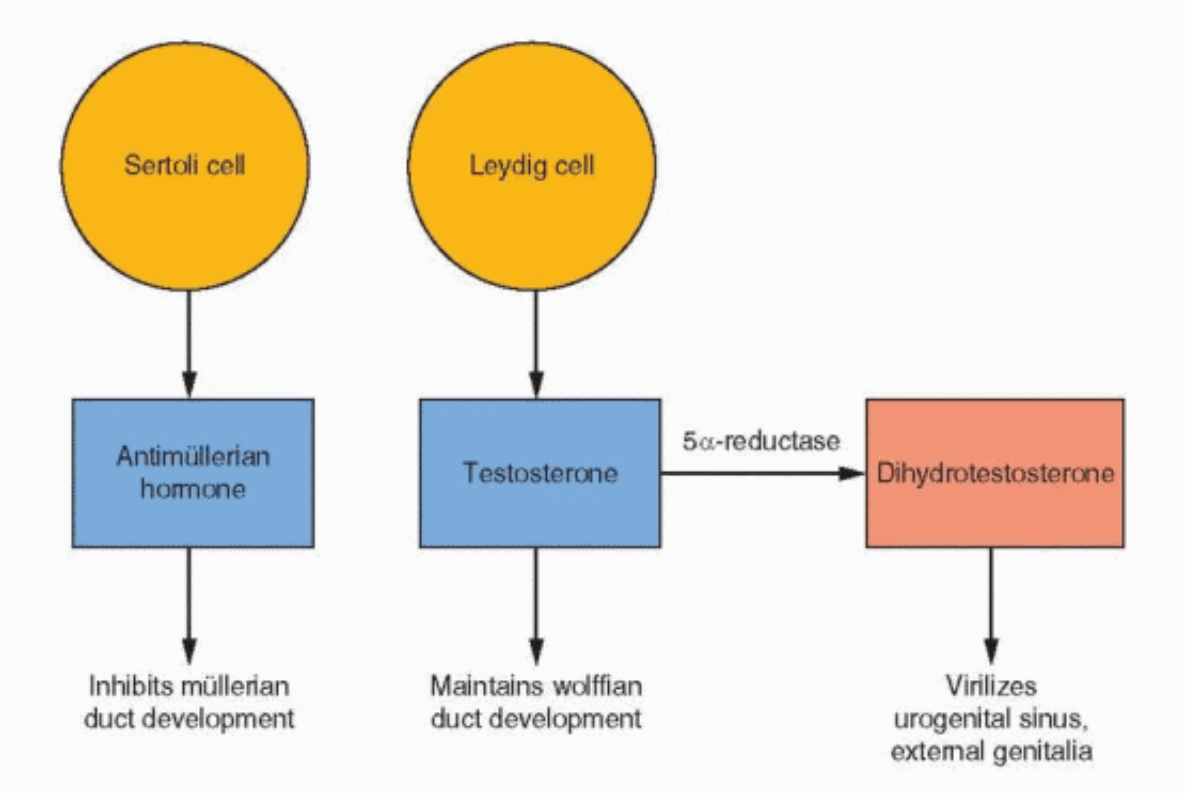
The wolffian (mesonephric) and müllerian (paramesonephric) ducts are discrete primordia that temporarily coexist in all embryos during the ambisexual period of development (up to 8 weeks). Thereafter, one type of duct system persists normally and gives rise to special ducts and glands, whereas the other disappears during the third fetal month, except for nonfunctional vestiges.

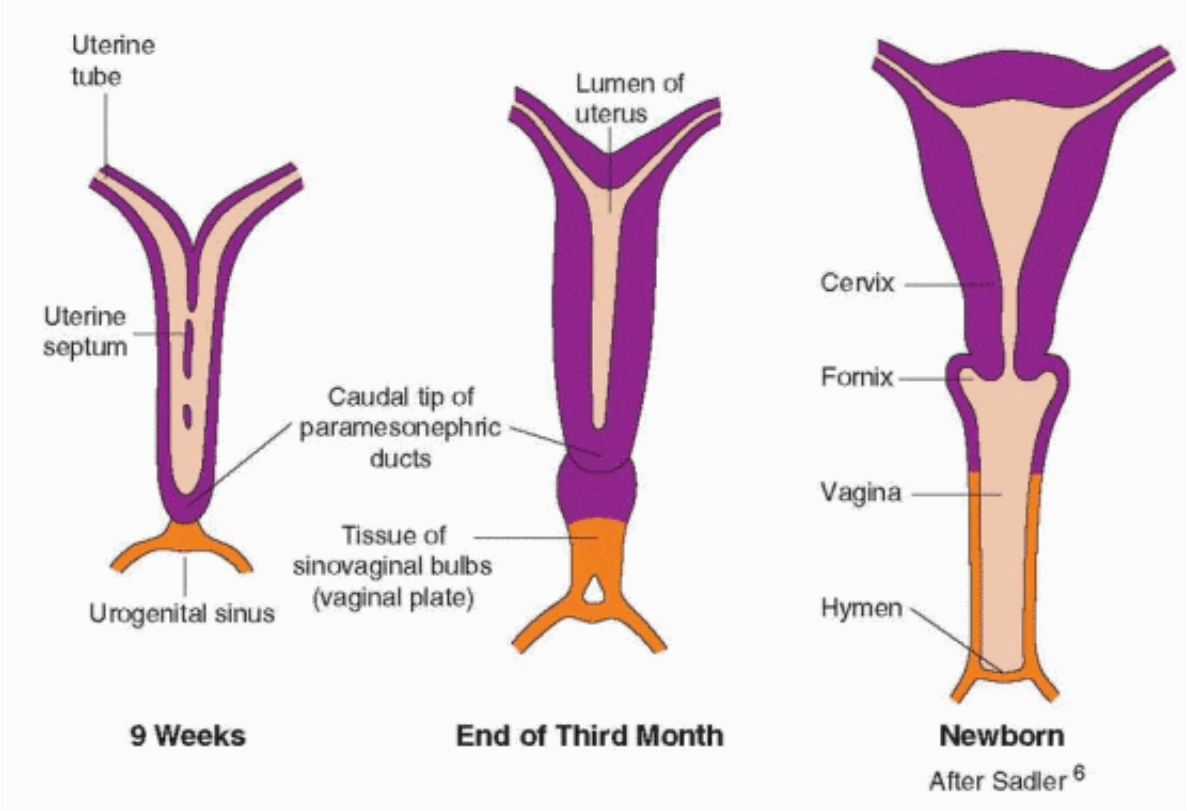
Hormonal control of mammalian somatic sex differentiation was established by the classic experiments of Alfred Jost.[4] In Jost's landmark studies, the active role of male-determining factors, as opposed to the constitutive nature of female differentiation, was defined as the directing feature of sex differentiation. This principle applies not only to the internal ducts but to the gonad, external genitalia, and even the brain. The critical factors in determining which of the duct structures stabilize or regress are the secretions from the testes: AMH (antimüllerian hormone, also known as müllerian-inhibiting substance or müllerian-inhibiting factor) and testosterone.

AMH is a member of the transforming growth factor-β family of glycoprotein differentiation factors that include inhibin and activin. The gene for AMH has been mapped to chromosome 19. AMH is synthesized by Sertoli cells soon after testicular differentiation and is responsible for the ipsilateral regression of the müllerian ducts by 8 weeks. Despite its presence in serum up to puberty, lack of regression of the uterus and tubes is the only consistent expression of AMH gene mutations. In the absence of AMH, the fetus will develop fallopian tubes, uterus, and

upper vagina from the paramesonephric ducts (the müllerian ducts). This development requires the prior appearance of the mesonephric ducts, and for this reason,

abnormalities in development of the tubes, uterus, and upper vagina are associated with abnormalities in the renal system.





The internal genitalia possess the intrinsic tendency to feminize. In the absence of a Y chromosome and a functional testis, the lack of AMH allows retention of the müllerian system and development of fallopian tubes, uterus, and upper vagina. In the absence of testosterone, the wolffian system regresses. In the presence of a normal ovary or the absence of any gonad, müllerian duct development takes place. This process is discussed in greater detail in Chapter 9.

The paramesonephric ducts come into contact in the midline to form a Y-shaped structure, the primordium for the uterus, tubes, and the upper one-third of the vagina.[5] The fallopian tubes, uterus, and the upper portion of the vagina are created by the fusion of the müllerian ducts by the tenth week of gestation. Canalization to create the uterine cavity, the cervical canal, and the vagina is complete by the 22nd week of gestation. Under the epithelium lies mesenchymal tissue that will be the origin of the uterine stroma and smooth muscle cells. By the 20th week of pregnancy, the uterine mucosa is fully differentiated into the endometrium.

The endometrium, derived from the mucosal lining of the fused müllerian ducts, is essential for reproduction and may be one of the most complex tissues in the human body. It is always changing, responding to the cyclic patterns of estrogen and progesterone of the ovarian menstrual cycle and to a complex interplay among its own autocrine and paracrine factors.

The Histologic Changes in Endometrium During an Ovulatory Cycle

The sequence of endometrial changes associated with an ovulatory cycle has been carefully studied by Noyes in the human and Bartlemez and Markee in the subhuman primate.[7,8,9,10] and [11]

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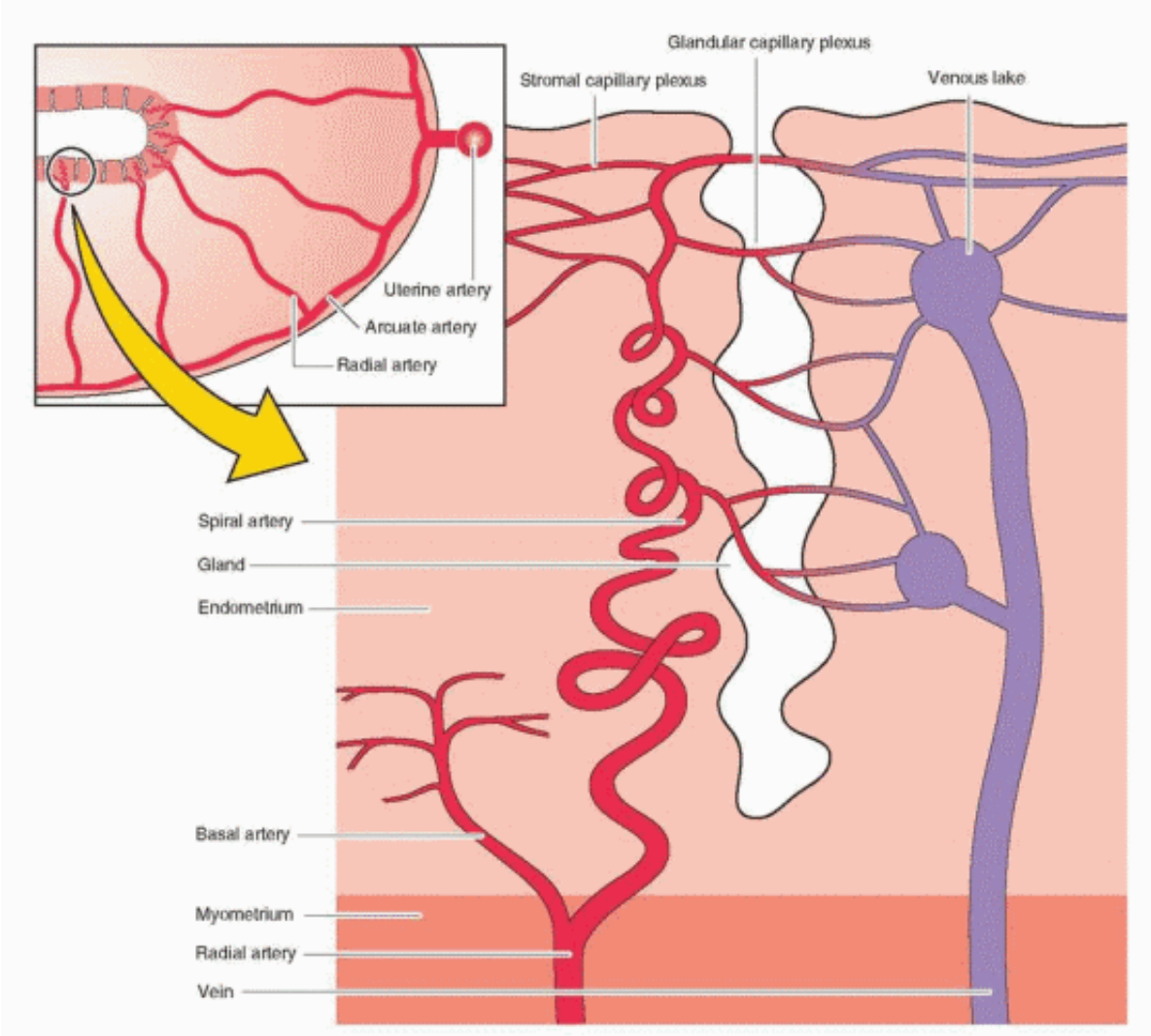
From these data a description of menstrual physiology has been developed based on specific anatomic and functional changes within glandular, vascular, and stromal components of the endometrium.[12,13] and [14] These changes will be discussed in five phases: (1) the menstrual endometrium, (2) the proliferative phase, (3) the

secretory phase, (4) preparation for implantation, and finally, (5) the phase of endometrial breakdown. Although these distinctions are not entirely arbitrary, it must be recalled that the entire process is an integrated evolutionary cycle of endometrial growth and regression, which is repeated some 400 times during the adult life of the human female.

The endometrium can be divided morphologically into an upper two-thirds “functionalis” layer and a lower one-third “basalis” layer. The purpose of the functionalis layer is to prepare for the implantation of the blastocyst; therefore, it is the site of proliferation, secretion, and degeneration. The purpose of the basalis layer is to provide the regenerative endometrium following menstrual loss of the functionalis.[15]



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The Uterine Vasculature

The two uterine arteries that supply the uterus are branches of the internal iliac arteries. At the lower part of the uterus, the uterine artery separates into the vaginal artery and an ascending branch that divides into the arcuate arteries. The arcuate arteries run parallel to the uterine cavity and anastomose with each other, forming a vascular ring around the cavity. Small centrifugal branches (the radial arteries) leave the arcuate vessels, perpendicular to the endometrial cavity, to supply the myometrium. When these arteries enter the endometrium, small branches (the basal arteries) extend laterally to supply the basalis layer. These basal arteries do not

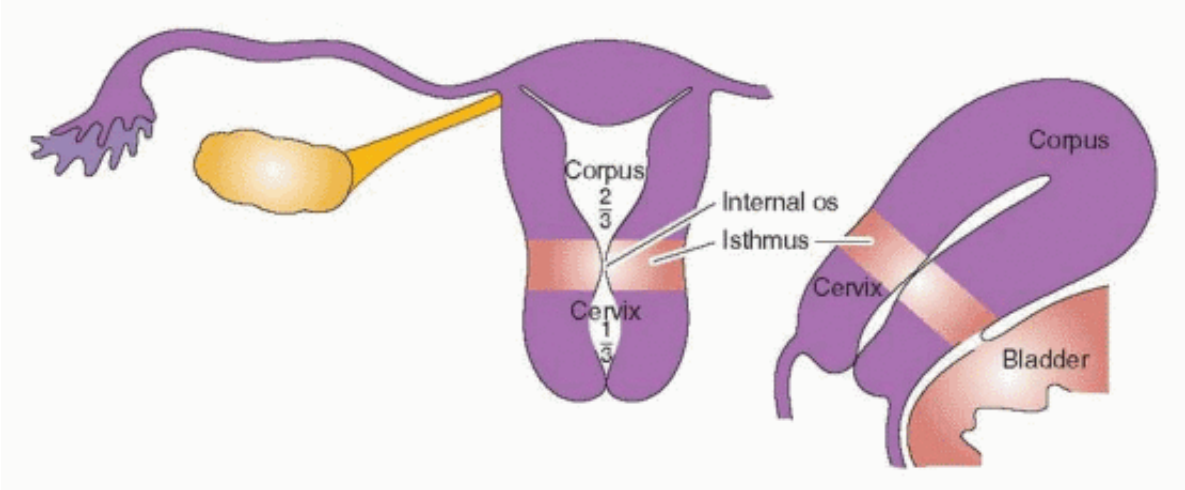
demonstrate a response to hormonal changes. The radial arteries continue in the direction of the endometrial surface, now assuming a corkscrew appearance (and now called the spiral arteries), to supply the functionalis layer of the endometrium. It is the spiral artery (an end artery) segment that is very sensitive to hormonal changes. One reason that the functionalis layer is more vulnerable to vascular ischemia is that there are no anastomoses among the spiral arteries. The endometrial glands and the stromal tissue are supplied by capillaries that emerge from the spiral arteries at all levels of the endometrium. The capillaries drain into a venous plexus and eventually into the myometrial arcuate veins and into the uterine veins. This unique vascular architecture is important in allowing a repeated sequence of endometrial growth and desquamation.

The Menstrual Endometrium

The menstrual endometrium is a relatively thin but dense tissue. It is composed of the stable, nonfunctioning basalis component and a variable, but small, amount of residual stratum spongiosum. At menstruation, this latter tissue displays a variety of functional states including disarray and breakage of glands, fragmentation of vessels and stroma with persisting evidence of necrosis, white cell infiltration, and red cell interstitial diapedesis. Even as the remnants of menstrual shedding dominate the overall appearance of this tissue, evidence of repair in all tissue components can be detected. Endometrial regeneration originates in epithelial and stromal stem cells.[16] Endometrial epithelial stem cells are located in the base of the endometrial glands and stromal stem cells around blood vessels in the basalis layer.

The menstrual endometrium is a transitional state bridging the more dramatic proliferative and exfoliative phases of the cycle. Its density implies that the shortness of height is not entirely due to desquamation. Collapse of the supporting matrix also contributes significantly to the shallowness. Reticular stains in Rhesus endometrium confirm this

“deflated” state. Nevertheless, as much as two-thirds of the functioning endometrium is lost during menstruation. The more rapid the tissue loss, the shorter the duration of flow. Delayed or incomplete shedding is associated with heavier flow and greater blood loss.



DNA synthesis is occurring in those areas of the basalis that have been completely denuded by day 2-3 of the menstrual cycle (the endometrium in the isthmic area, the narrow area between the cervix and the corpus, and the endometrium in the cornual recesses at the ostia of the tubes remain intact). The new surface epithelium emanates from the flanks of stumps of glands in the basalis layer left standing after menstrual desquamation.[17] Rapid re-epithelialization follows the proliferation of the cells in the basalis layer and the surface epithelium in

the isthmic and tubal ostial endometrium. This epithelial repair is supported by underlying fibroblasts. The stromal fibroblast layer forms a compact mass over which the resurfacing epithelium can “migrate.” In addition, it is likely that the stromal layer contributes important autocrine and paracrine factors for growth and migration. Because hormone levels are at their nadir during this repair phase, the response may be due to injury rather than hormone mediated. However, the basalis layer is rich in its content of estrogen receptors. This “repair” is fast; by day 4 of the cycle, more than two-thirds of the cavity is covered with new epithelium.[17] By day 5-6, the entire cavity is re-epithelialized, and stromal growth begins.

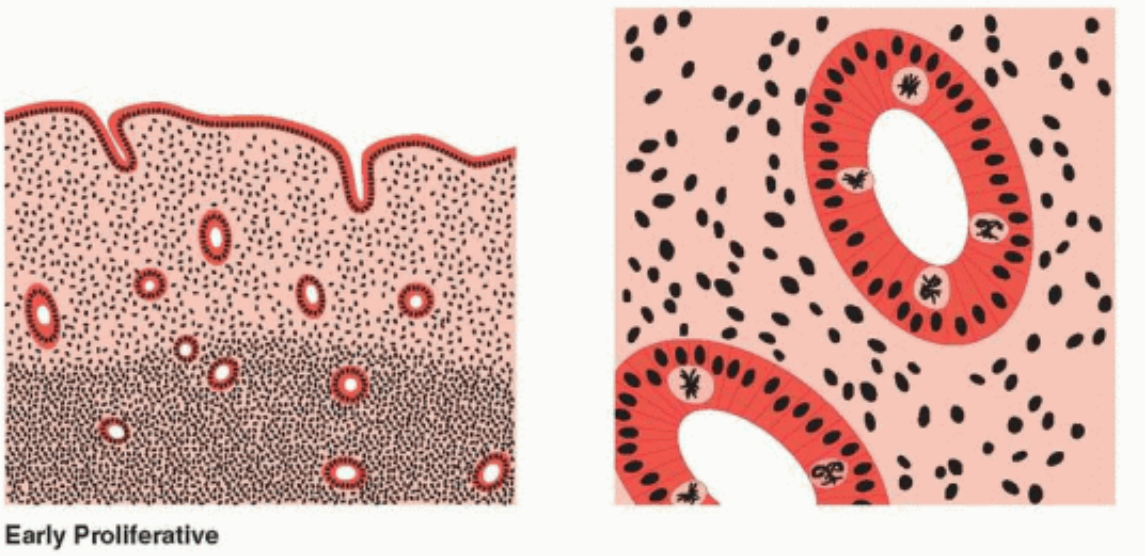
The Proliferative Phase

The proliferative phase is associated with ovarian follicle growth and increased estrogen secretion. Undoubtedly as a result of this steroidal action, reconstruction and growth of the endometrium are achieved. The glands are most notable in this response. At first they are narrow and tubular, lined by low columnar epithelium cells. Mitoses become prominent and pseudostratification is observed. As a result, the glandular epithelium extends peripherally and links one gland segment with its immediate neighbor. A continuous epithelial lining facing the endometrial cavity is formed. The stromal component evolves from its dense cellular menstrual condition through a brief period of edema to a final loose syncytial-like status. Coursing through the stroma, spiral vessels extend (unbranched and uncoiled in the early proliferative phase) to a point immediately below the epithelial binding membrane. Here they form a loose capillary network. All of the tissue components (glands, stromal cells, and endothelial cells) demonstrate proliferation, which peaks on days 8-10 of the cycle, reflecting rising estradiol levels in the circulation and maximal estrogen receptor concentration in the endometrium.[18] This proliferation is marked by increased mitotic activity and increased nuclear DNA and cytoplasmic RNA synthesis, which is most intense in the functionalis layer in the upper two-thirds of the uterus, the usual site of blastocyst implantation.

During proliferation, the endometrium grows from approximately 0.5 mm to 3.5-5.0 mm in height of a singular layer. Restoration of tissue constituents has been achieved by estrogeninduced new growth as well as incorporation of ions, water, and amino acids. The stromal ground substance has re-expanded from its menstrual collapse. Although true tissue growth has occurred, a major element in achievement of endometrial height is “reinflation” of the stroma.

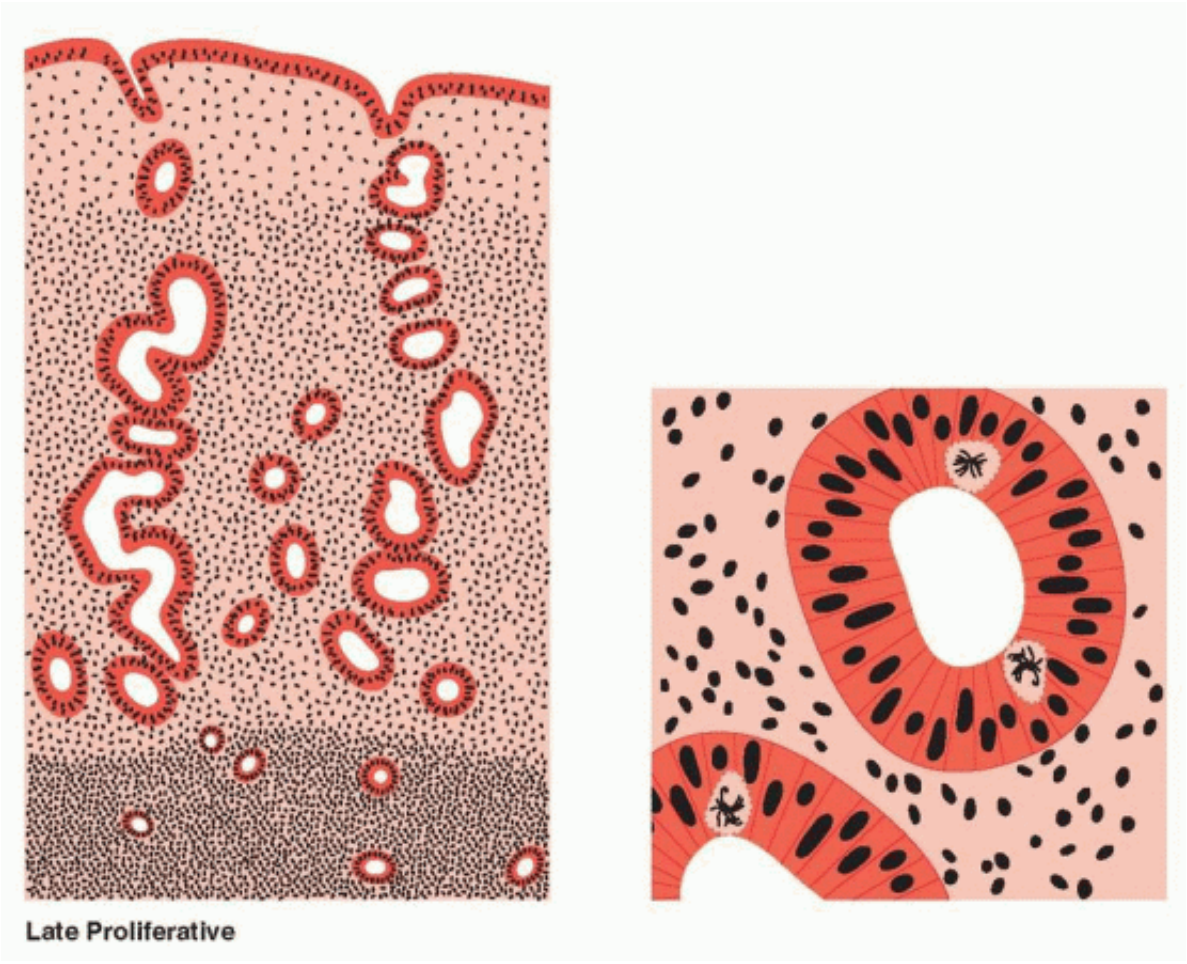
An important feature of this estrogen-dominant phase of endometrial growth is the increase in ciliated and microvillous cells. Ciliogenesis begins on days 7-8 of the cycle.[17] This response to estrogen is exaggerated in hyperplastic endometrium that is the result of hyperestrogenism. The concentration of these ciliated cells around gland openings and the ciliary beat pattern influence the mobilization and distribution of endometrial secretions during

the secretory phase. Cell surface microvilli, also a response to estradiol, are cytoplasmic extensions and serve to increase the active surface of cells.

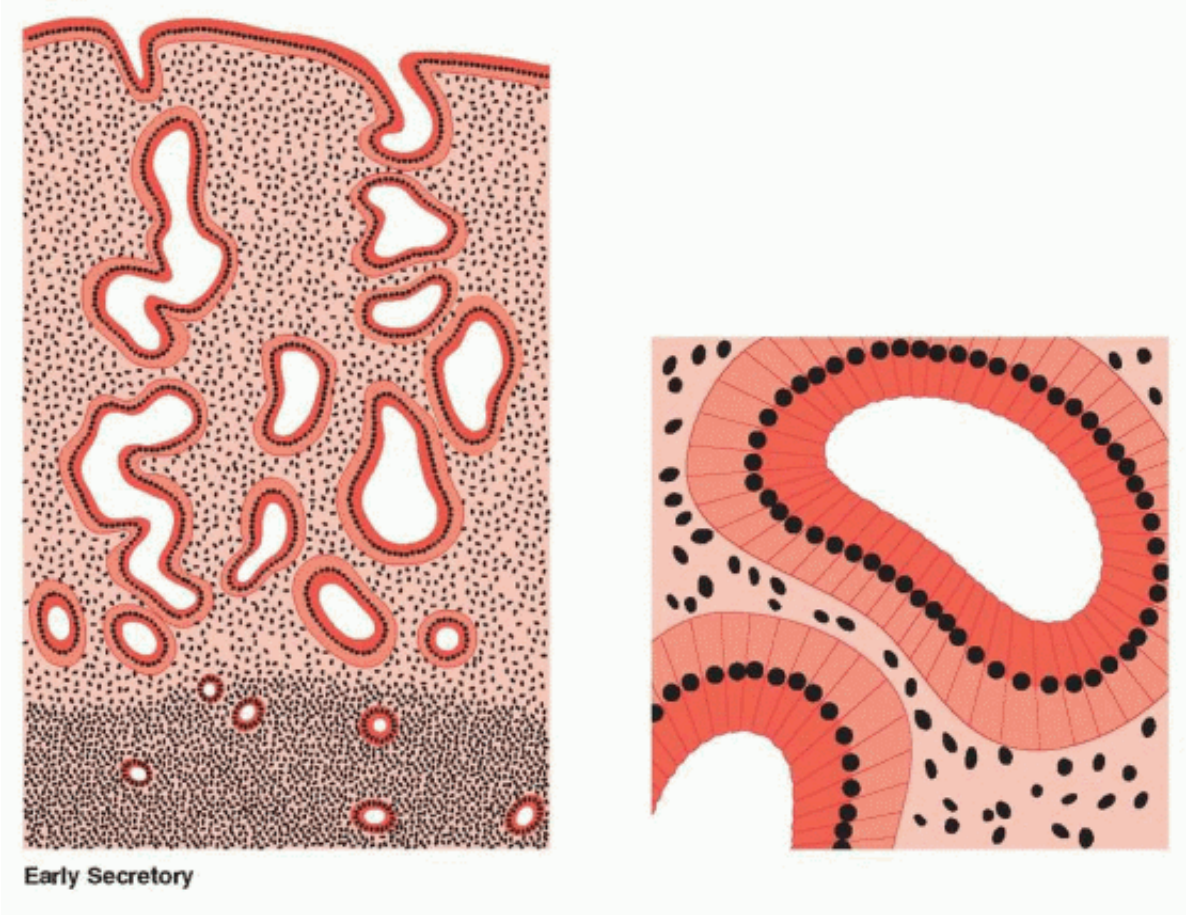


Early Proliferative

At all times, a large number of cells derived from bone marrow are present in the endometrium. These include lymphocytes and macrophages, diffusely distributed in the stroma.



Late Proliferative



The Secretory Phase

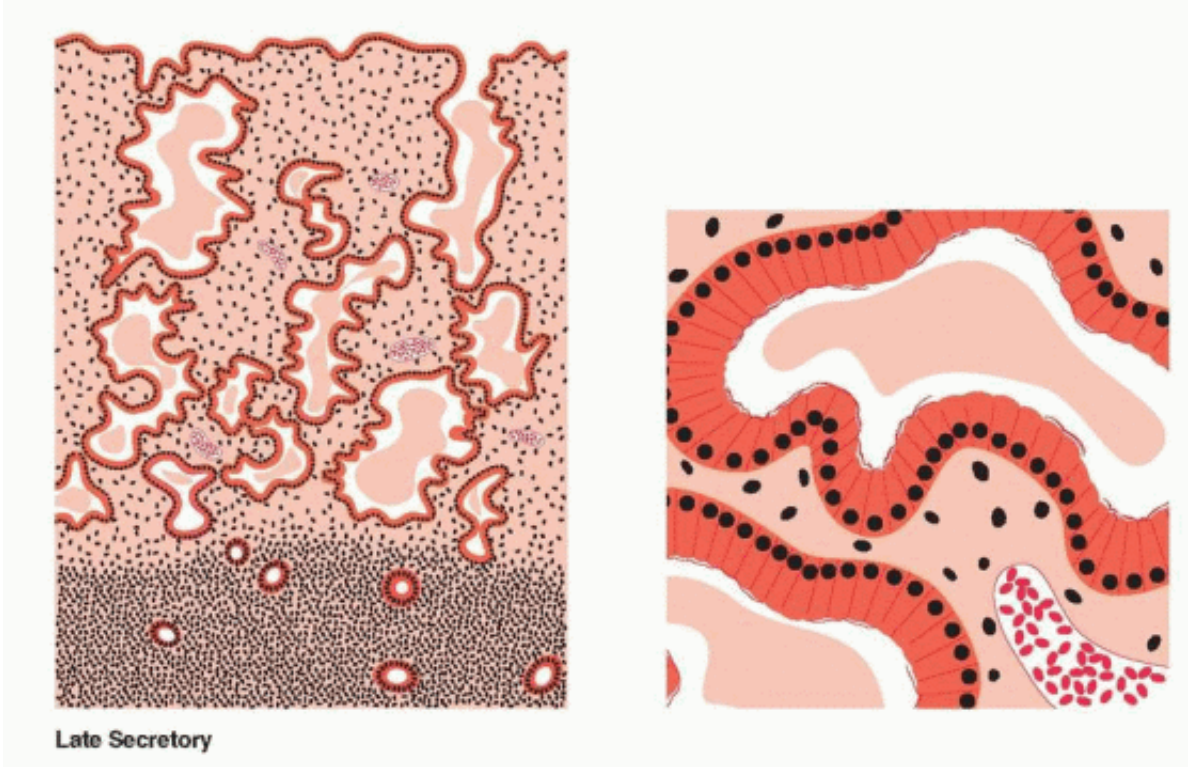
After ovulation, the endometrium now demonstrates a combined reaction to estrogen and progesterone activity. Most impressive is that total endometrial height is fixed at roughly its preovulatory extent (5-6 mm) despite continued availability of estrogen. Epithelial proliferation ceases 3 days after ovulation.[19] This restraint or inhibition is believed to be induced by progesterone. This limitation of growth is associated with a decline in mitosis and DNA synthesis, significantly due to progesterone interference with estrogen receptor expression and progesterone stimulation of 17β-hydroxysteroid dehydrogenase and sulfotransferase, which convert estradiol to estrone sulfate (which is rapidly excreted from the cell).[20,21] In addition, estrogen stimulates many oncogenes that probably mediate estrogeninduced growth. Progesterone antagonizes this action by suppressing the estrogen-mediated transcription of oncogene mRNA.[22]

Individual components of the tissue continue to display growth, but confinement in a fixed structure leads to progressive tortuosity of glands and intensified coiling of the spiral vessels. The secretory events within the glandular cells, with progression of vacuoles from intracellular to intraluminal appearance, are well known and take place over a 7-day postovulatory interval. At the conclusion of these events, the glands appear exhausted, the tortuous lumina variably distended, and individual cell surfaces fragmented in a sawtooth appearance. Stroma is increasingly edematous, and spiral vessels are prominent and densely coiled.

The first histologic sign that ovulation has occurred is the appearance of subnuclear intracytoplasmic glycogen vacuoles in the glandular epithelium on cycle days 17-18. Giant

mitochondria and the “nucleolar channel system” appear in the gland cells. The nucleolar channel system has a

unique appearance due to progesterone, an infolding of the nuclear membranes. Individual components of the tissue continue to display growth, but confinement in a fixed structure leads to progressive tortuosity of glands and intensified coiling of the spiral vessels. These structural alterations are soon followed by active secretion of glycoproteins and peptides into the endometrial cavity. Transudation of plasma also contributes to the endometrial secretions. Important immunoglobulins are obtained from the circulation and delivered to the endometrial cavity by binding proteins produced by the epithelial cells. The peak secretory level is reached 7 days after the midcycle gonadotropin surge, coinciding with the time of blastocyst implantation.



The Implantation Phase

Significant changes occur within the endometrium from the 7th to the 13th day postovulation (days 21-27 of the cycle). At the onset of this period, the distended tortuous secretory glands have been most prominent with little intervening stroma. By 13 days postovulation, the endometrium has differentiated into three distinct zones. Something less than one-fourth of the tissue is the unchanged basalis fed by its straight vessels and surrounded by indifferent spindle-shaped stroma. The midportion of the endometrium (approximately 50% of the total) is the lace-like *stratum spongiosum*, composed of loose edematous stroma with tightly coiled but ubiquitous spiral vessels and exhausted dilated glandular ribbons. Overlying the spongiosum is the superficial layer of the endometrium (about 25% of the height) called the *stratum compactum*. Here the prominent histologic feature is the stromal cell, which has become large and polyhedral. In its cytoplasmic expansion one cell abuts the other, forming a compact, structurally sturdy layer. The necks of the glands traversing this segment are compressed and less prominent. The subepithelial capillaries and spiral vessels are engorged.

At the time of implantation, on days 21-22 of the cycle, the predominant morphologic feature is edema of the endometrial stroma. This change may be secondary to the estrogenand progesterone-mediated increase in prostaglandin and vascular endothelial growth factor (VEGF) production by the endometrium that cause an increase in capillary permeability. Receptors for the sex steroids are present in the muscular walls of the

endometrial blood vessels, and the enzyme system for prostaglandin synthesis is present in both the muscular walls and the endothelium of the endometrial arterioles. Mitoses are first seen in endothelial cells on cycle day 22. Vascular proliferation leads to the coiling of the spiral vessels, a response to the sex steroids, the prostaglandins, and the autocrine and paracrine factors produced in response to estrogen and progesterone.

During the secretory phase, so-called K (Körnchenzellen) cells appear, reaching a peak concentration in the first trimester of pregnancy. These are granulocytes that have an immunoprotective role in implantation and placentation. They are located perivascularly and are believed to be derived from the blood. By day 26-27, the endometrial stroma is infiltrated by extravasated polymorphonuclear leukocytes. The majority of the leukocytes are killer cells and macrophages, believed to be involved in the process of endometrial breakdown and menstruation. The appearance and function of these cells are regulated by the complex array of peptides and cytokines in the endometrium in response to hormonal signaling.

The gene expression pattern in the endometrium throughout the menstrual cycle is being established, with a focus on the implantation window.[23,24] and [25] As expected, microarray analyses reveal a changing pattern of gene expression that correlates with each hormonal and morphological stage in the endometrial menstrual cycle.[26] Ultimately this will yield a comprehensive picture, with the gene signature of each event in the estrogen and progesterone regulation of the endometrium. The regulating growth factors, cytokines, and peptide hormones that are essential for implantation will be identified.

The stromal cells of the endometrium respond to hormonal signals, synthesize prostaglandins, and, when transformed into decidual cells, produce an impressive array of substances, some of which are prolactin, relaxin, renin, insulin-like growth factors (IGFs), and insulinlike growth factor binding proteins (IGFBPs). The endometrial stromal cells, the progenitors of decidual cells, were originally believed to be derived from the bone marrow (from cells invading the endometrium), but they are now considered to emanate from the primitive uterine mesenchymal stem cells.[27]

The decidualization process begins in the luteal phase under the influence of progesterone and mediated by autocrine and paracrine factors. On cycle days 22-23, predecidual cells can be identified, initially surrounding blood vessels, characterized by cytonuclear enlargement, increased mitotic activity, and the formation of a basement membrane. The decidua, derived from stromal cells, becomes an important structural and biochemical tissue of pregnancy. Decidual cells control the invasive nature of the trophoblast, and the products of the decidua play important autocrine and paracrine roles in fetal and maternal tissues.

Lockwood and his colleagues assign a key role to decidual cells in both the process of endometrial bleeding (menstruation) and the process of endometrial hemostasis (implantation and placentation).[28,29] and [30] Implantation requires endometrial hemostasis and the maternal uterus requires resistance to invasion. Inhibition of endometrial hemorrhage can be attributed, to a significant degree, to appropriate changes in critical factors as a consequence of decidualization; e.g., lower plasminogen activator levels, reduced expression of the enzymes that degrade the stromal extracellular matrix (such as the metalloproteinases), and increased levels of plasminogen activator inhibitor-1. Withdrawal of estrogen and progesterone support, however, leads to changes in the opposite directions, consistent with endometrial breakdown.

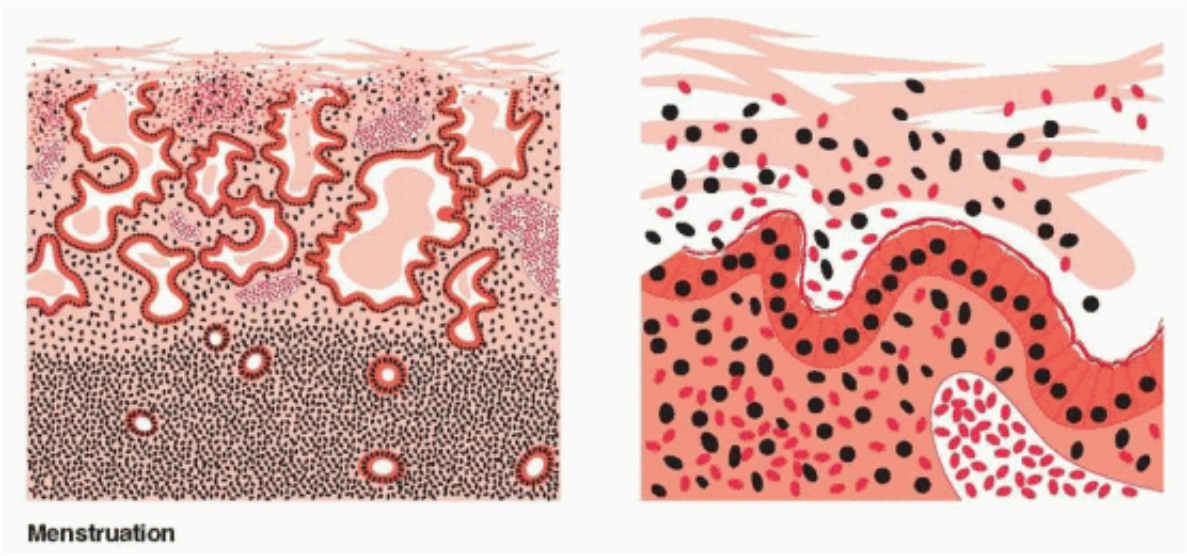
The Phase of Endometrial Breakdown

Predecidual transformation has formed the “compacta” layer in the upper part of the functionalis layer by day 25 (3 days before menstruation). In the absence of fertilization, implantation, and the consequent lack of sustaining quantities of human chorionic gonadotropin from the trophoblast, the otherwise fixed lifespan of the corpus

luteum is completed, and estrogen and progesterone levels wane.

The withdrawal of estrogen and progesterone initiates important endometrial events: vasomotor reactions, the process of apoptosis, tissue loss, and, finally, menstruation. The most prominent immediate effect of this hormone withdrawal is a modest shrinking of the tissue height and remarkable spiral arteriole vasomotor responses. The classic concept of the vascular sequence was constructed from direct observations of Rhesus endometrium transplanted to the anterior chamber of the eye.[7,8] With shrinkage of height, blood flow within the spiral vessels diminished, venous drainage was decreased, and vasodilation ensued. Thereafter, the spiral arterioles underwent rhythmic vasoconstriction and relaxation. Each successive spasm was more prolonged and profound, leading eventually to endometrial blanching. Thus these reactions were proposed to lead to menstruation because of endometrial ischemia and stasis caused by vasoconstriction of the spiral arterioles. A new model of menstruation, as discussed in Chapter 15, emphasizes enzymatic autodigestion of the functional layer of the endometrium and its capillary plexus.

In the first half of the secretory phase, acid phosphatase and potent lytic enzymes are confined to lysosomes. Their release is inhibited by progesterone stabilization of the lysosomal membranes. With the waning of estrogen and progesterone levels, the lysosomal membranes are not maintained, and the enzymes are released into the cytoplasm of epithelial, stromal, and endothelial cells and eventually into the intercellular space. These active enzymes will digest their cellular constraints, leading to the release of prostaglandins, extravasation of red blood cells, tissue necrosis, and vascular thrombosis. This process is one of *apoptosis,* (programmed cell death, characterized by a specific morphologic pattern that involves cell shrinkage and chromatin condensation culminating in cell fragmentation) mediated by cytokines.[31] An important step in this breakdown is the dissolution of cell-tocell adhesion by key proteins. Binding of endometrial epithelial cells utilizes transmembrane proteins, *cadherins,* that link intercellularly with each other and intracellularly with catenins that are bound to actin filaments.[32]



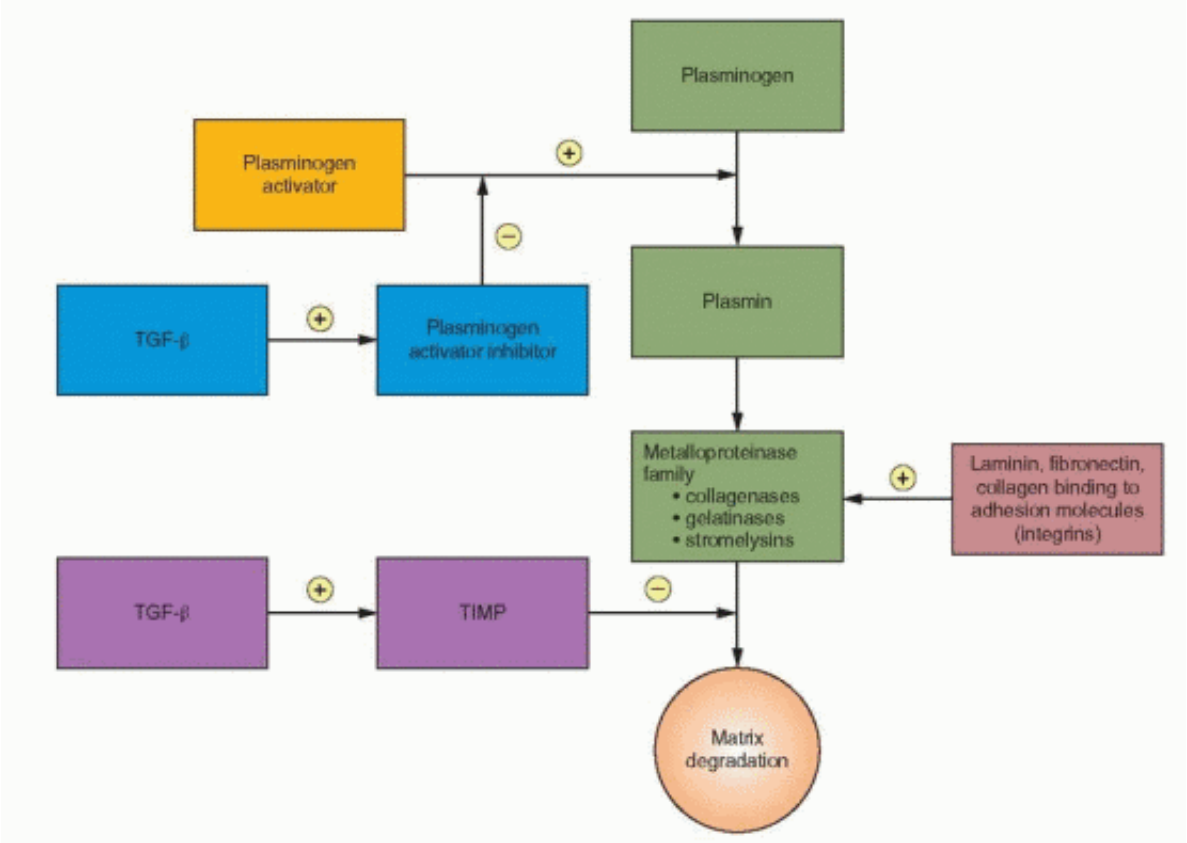
Endometrial tissue breakdown also involves a family of enzymes, matrix metalloproteinases, that degrade components (including collagens, gelatins, fibronectin, and laminin) of the extracellular matrix and basement membrane.[33,34] The metalloproteinases include collagenases that degrade interstitial and basement membrane collagens; gelatinases that further degrade collagens; and stromelysins that degrade fibronectin, laminin, and glycoproteins. The expression of metalloproteinases in human endometrial stromal cells follows a pattern

correlated with the menstrual cycle, indicating a sex steroid response as part of the growth and remodeling of the endometrium with a marked increase in late secretory and early menstrual endometrium.[35] Progesterone withdrawal from endometrial cells increases VEGF production and induces matrix metalloproteinase secretion, probably from both endometrial stromal cells and leukocytes, which is followed by the irreversible breakdown of cellular membranes and the dissolution of extracellular matrix.[36,37] and [38] Appropriately, this enzyme expression increases in the decidualized endometrium of the late secretory phase, during the time of declining progesterone levels. With the continuing progesterone secretion of early pregnancy, the decidua is maintained and metalloproteinase expression is suppressed, in a mechanism mediated by transforming growth factor-beta (TGF-ß).[39] In a nonpregnant cycle, metalloproteinase expression is suppressed after menses, presumably by increasing estrogen levels.

Metalloproteinase activity is restrained by specific tissue inhibitors designated as TIMP.[40] The balance of metalloproteinase and TIMP activity is an important event in successful implantation. Thus, progesterone withdrawal can lead to endometrial breakdown through a mechanism that is independent of vascular events (specifically ischemia), a mechanism that involves cytokines.[31] During bleeding, both normal and abnormal, there is evidence indicating that specific genes are activated in the endometrium; one such gene has the structural features of the TGF-ß family.[41]

There is considerable evidence to support a major role for a cytokine, tumor necrosis factor-a (TNF-a), in menstruation.[31] TNF-a is a transmembrane protein whose receptor belongs to the nerve growth factor/TNF family for inducing apoptotic signals. The key change is an increase in secretion because TNF-a secretion by endometrial cells reaches a peak at menstruation, but there is no cycle change in receptor content. TNF-a inhibits endometrial proliferation and induces apoptosis; this cytokine causes a loss of adhesion proteins (the cadherin-catenin-actin complex) and induces cell-to-cell dissolution. In addition to endometrial cells, TNF-a also causes damage to vascular endothelium.

Progesterone withdrawal is also associated with an increase in vascular endothelial growth factor receptor concentrations in the stromal cells of the layers of endometrium destined to be sloughed.[42] Although the vascular endothelial growth factor system is usually involved with angiogenesis, in this case these factors are involved in the preparation for menstrual bleeding, perhaps influencing the expression of matrix metalloproteinases (MMPs). Endometrial genes without classic steroid response elements can respond to the sex steroids by utilizing a family of proteins (the Sp family) that mediate steroid activity at the level of transcription (acting in a fashion similar to the steroid receptors). These proteins, induced by progesterone in stromal (decidual) and epithelial cells, can activate tissue factor, plasminogen activator inhibitor-1, IGF binding protein-1, uteroglobin, and uteroferrin. Tissue factor is involved in the clotting mechanism to sustain hemostasis. Uteroglobin is a small protein expressed in endometrial epithelial cells.[43] The physiologic function of uteroglobin is uncertain. Uteroglobin, with high affinity, binds progestins and may play a role in immunosuppression. Uteroglobin gene expression is stimulated by estrogen, and this response is enhanced by progesterone. Human endometrium can secrete ß-endorphin, yet another candidate for involvement in endometrial immunologic events, and its release is inhibited by both estrogens and glucocorticoids.[44]



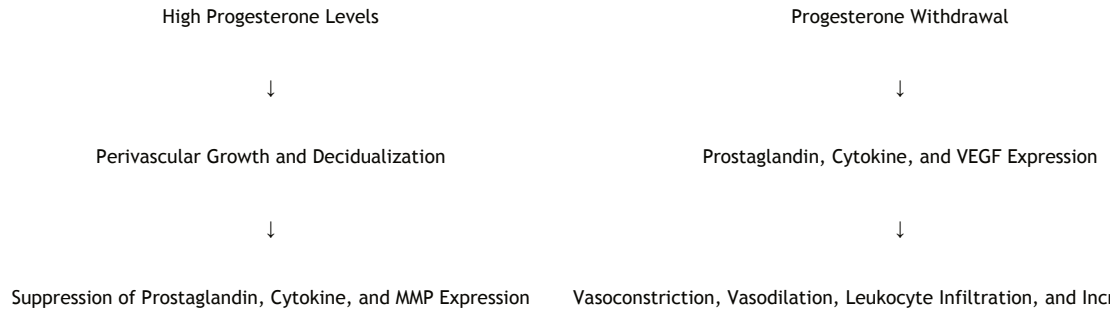
Eventually, considerable leakage occurs as a result of diapedesis, and finally, interstitial hemorrhage occurs due to breaks in superficial arterioles and capillaries. White cells migrate through capillary walls, at first remaining adjacent to vessels but then extending

throughout the stroma. The leukocytes add important regulatory cytokines, chemokines, and enzymes that are involved in the degradation of the extracellular matrix. During arteriolar vasomotor changes, red blood cells escape into the interstitial space. Thrombinplatelet plugs also appear in superficial vessels. The prostaglandin content (PGF_{2a} and PGE_2) in the secretory endometrium reaches its highest levels at the time of menstruation. The vasoconstriction and myometrial contractions associated with the menstrual events are believed to be significantly mediated by prostaglandins from perivascular cells and the potent vasoconstrictor endothelin-1, derived from stromal decidual cells.

As ischemia and weakening progress, the continuous binding membrane is fragmented, and intercellular blood is extruded into the endometrial cavity. New thrombin-platelet plugs form intravascularly upstream at the shedding surface, limiting blood loss. Increased blood loss is a consequence of reduced platelet numbers and inadequate hemostatic plug formation. Menstrual bleeding is influenced by activation of clotting and fibrinolysis. Fibrinolysis is principally the consequence of the potent enzyme plasmin, formed from its inactive precursor plasminogen. Endometrial stromal cell tissue factor (TF) and plasminogen activators and inhibitors are involved in achieving a balance in this process. TF stimulates coagulation, initially binding to factor VII. TF and plasminogen activator inhibitor-1 (PAI-1) expression accompanies decidualization, and the levels of these factors may govern

the amount of bleeding.[30,45] PAI-1, in particular, exerts an important restraining action on fibrinolysis and proteolytic activity.[46] Blood loss is also controlled by constriction of the spiral arteries, mediated by the perivascular cells, myofibroblasts that surround the spiral arteries.[47] These cells respond to progesterone

withdrawal by expressing prostaglandins, cytokines, and MMPs, causing not only cycling vasoconstriction and vasodilation but also modulating leukocyte entry (an important additional source of metalloproteinases) into the endometrium. Disordered growth and function of the perivascular cells are likely contributing factors in menstrual bleeding problems.



With progressive enzymatic degradation of the endometrium, the subsurface capillary and venous vascular system is disrupted, causing hemorrhage and escape of blood into the endometrial cavity. Additional ischemic breakdown ensues with necrosis of cells and defects in vessels adding to the menstrual effluvium. Degeneration extends to the deepest extent of the functional layer where rupture of the basal arterioles contributes to bleeding. A natural cleavage point exists between basalis and spongiosum, and, once breached, the loose, vascular, edematous stroma of the spongiosum desquamates and collapses. The process is initiated in the fundus and inexorably extends throughout the uterus. In the end, the typical deflated, shallow, dense, menstrual endometrium results. Within 13 hours, the endometrial height shrinks from 4 to 1.25 mm.[13] Menstrual flow stops as a result of the combined effects of prolonged vasoconstriction of the radial arteries and the spiral arteries in the basalis, tissue collapse, vascular stasis, and estrogeninduced “healing.” In contrast to postpartum bleeding, myometrial contractions are not important for control of menstrual bleeding. Thrombin generation in the basal endometrium in response to extravasation of blood is essential for hemostasis. Thrombin promotes the generation of fibrin, the activation of platelets and clotting cofactors, and angiogenesis.

The basalis endometrium remains during menses, and repair takes place from this layer. This endometrium is protected from the lytic enzymes in the menstrual fluid by a mucinous layer of carbohydrate products that are discharged from the glandular and stromal cells.[48]

Normal Menses

Approximately 50% of the menstrual detritus is expelled in the first 24 hours of menstrual flow. The menstrual fluid is composed of the autolysed functionalis, inflammatory exudate, red blood cells, and proteolytic enzymes (at least one of which, plasmin, lyses fibrin clots as they form). The high fibrinolytic activity advances emptying of the uterus by liquefaction of tissue and fibrin. If the rate of flow is great, clotting can and does occur.

Most women (90%) have menstrual cycles with an interval of 24 to 35 days (Chapter 6).[49,50] Menarche is followed by approximately 5-7 years of increasing regularity as cycles shorten to reach the usual reproductive-age pattern. In the 40s, cycles begin to lengthen again. The usual duration of flow is 4-6 days, but many women flow as little as 2 days and as much as 8 days. The normal volume of menstrual blood loss is 30 mL; greater than 80 mL is abnormal. Normal and abnormal characteristics and definitions of menstrual flow are discussed in detail in Chapter 15.

A Teleologic Theory of Endometrial-Menstrual Events

Menstruation is a very recent phenomenon in the evolutionary time line. It occurs in very few species, even among viviparous animals. An unabashedly teleologic view of menstrual events was offered many years ago by Rock et al.[51] The basic premise of this thesis is that every endometrial cycle has, as its only goal, nourishing support of an early embryo. Failure to accomplish this objective is followed by orderly elimination of unutilized tissue and prompt renewal to achieve a more successful cycle.

The ovum must be fertilized within 12-24 hours of ovulation. Over the next 4 days, it remains unattached within the tubal lumen utilizing tubal fluids and residual cumulus cells to sustain nutrition and energy for early cellular cleavage. After this stay, the solid ball of cells (morula), which is the embryo, leaves the tube and enters the uterine cavity. Here the embryo undergoes another 2-3 days of unattached but active existence. Fortunately, by this time endometrial gland secretions have filled the cavity and they bathe the embryo in nutrients. This is the first of many neatly synchronized events that mark the conceptus-endometrial relationship. By 6 days after ovulation, the embryo (now a blastocyst) is ready to attach and implant. At this time, it finds an endometrial lining of sufficient depth, vascularity, and nutritional richness to sustain the important events of early placentation to follow. Just below the epithelial lining, a rich capillary plexus has been formed and is available for creation of the trophoblast-maternal blood interface. Later, the surrounding zona compactum, occupying more and more of the endometrium, will provide a sturdy splint to retain endometrial architecture despite the invasive inroads of the burgeoning trophoblast.

Failure of the appearance of human chorionic gonadotropin, despite otherwise appropriate tissue reactions, leads to the vasomotor changes associated with estrogen-progesterone withdrawal and menstrual desquamation. However, not all the tissue is lost, and, in any event, a residual basalis is always available, making resumption of growth with estrogen a relatively rapid process. Indeed, even as menses persists, early regeneration can be seen. As soon as follicle maturation occurs (in as short a time as 10 days), the endometrium is ready once again to perform its reproductive function.

The Uterus Is an Endocrine Organ

The uterus is dynamic. It not only responds and changes in a sensitive fashion to classic hormonal signals (the endocrine events of the menstrual cycle) but it is also composed of complex tissues, with important autocrine and paracrine functions that serve not only the uterus but also the contiguous tissues of the fetoplacental unit during pregnancy. The most dynamic component of the uterus is the endometrium.

Endometrial Products

The endometrium secretes many substances, the functions of which (and their interrelationships) represent a major investigative challenge.[52] In addition to producing a nourishing, supportive environment for the early embryo, the endometrium plays an important role in suppressing the immune response within the pregnant uterus. The mechanisms controlling the immune response in decidual cells are not understood, but hormonal influence is undoubtedly important.

The presence of the cytokine family, involved in inflammation and immune responses, is not surprising in a tissue that undergoes cyclic degeneration. The interleukins stimulate the production of prostaglandins as well as other cytokines.[53] Colony-stimulating factor-1 is a cytokine that influences cellular proliferation and the presence of macrophages. Interferon-γ is produced by activated T lymphocytes and inhibits endometrial epithelial

proliferation. Leukemia-inhibiting factor (LIF) is expressed in response to a variety of other cytokines

and growth factors. Like the interleukins, LIF is most abundant during the progesteronedominated secretory phase and early decidua and may have a role in embryo implantation.[54, 55] Tumor necrosis factor-a (TNF-a) gene expression is present in endometrium, and its activity is increased during the proliferative phase, decreased early in the secretory phase, and increased again in the midsecretory phase.[56] TNF-a exerts multiple influences on cellular growth.

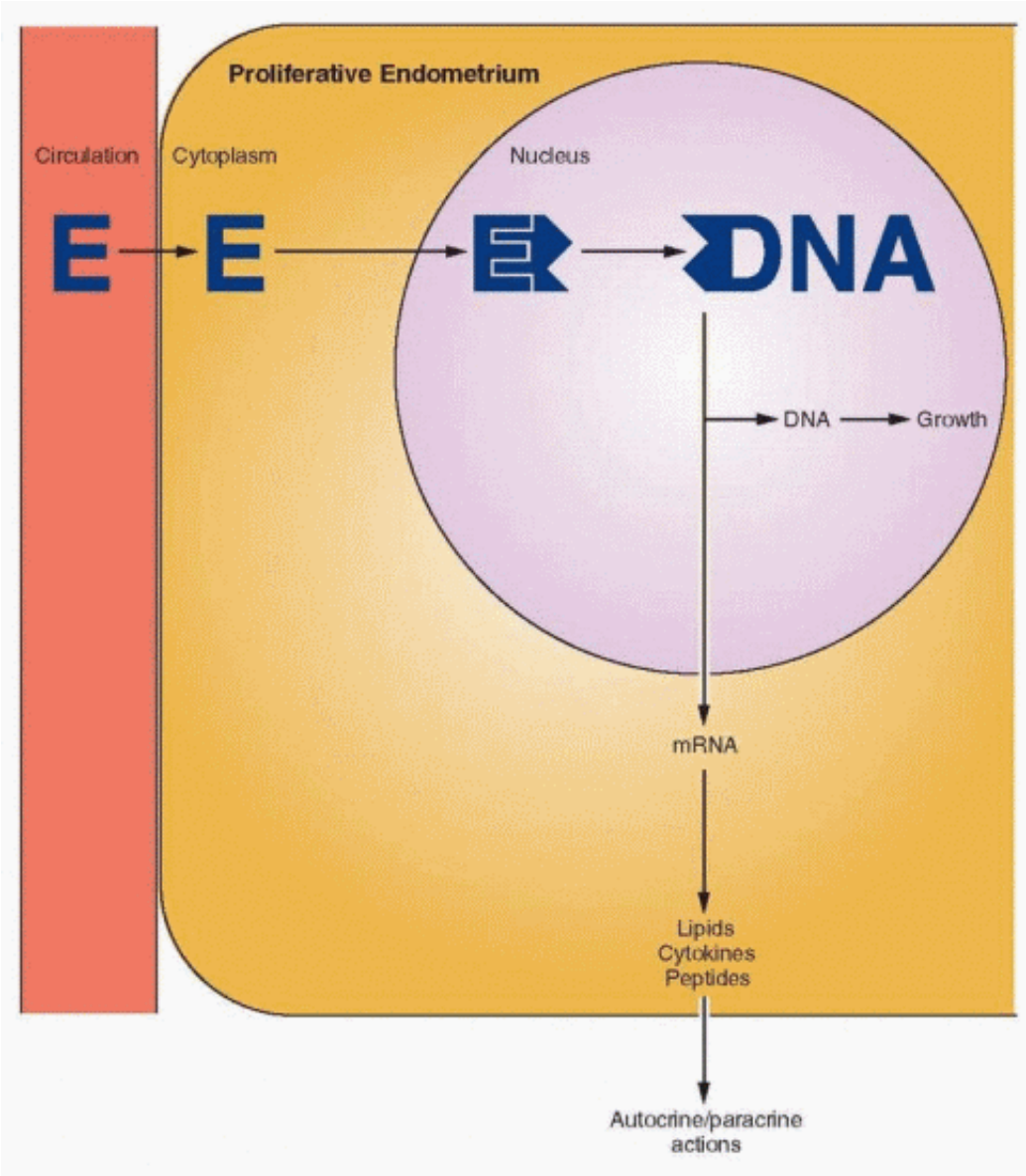
A Partial List of Endometrial Regulating Molecules

Lipids	*Cytokines*	*Peptides*
Prostaglandins	Interleukin-1α	Prolactin
Thromboxanes	Interleukin-1ß	Relaxin
Leukotrienes	Interleukin-6	Prorenin and Renin
	Interferon-γ	Endorphin
	Colony-stimulating factor-1	Endothelin-1
	Tumor necrosis factor-α	Corticotropin-releasing hormone
	Leukemia-inhibiting factor	Fibronectin
		Uteroglobin
		Lipocortin-1
		Parathyroid hormone-like protein
		Integrins
		Epidermal growth factor family

		EGF
		Heparin-binding EGF
		TGF-α
	Insulin-like growth factor family	
		IGF-I
		IGF-II
		IGFBPs 1-6
	Transforming growth factor-ß family	
		Activins
		Inhibins
		Follistatin
	Platelet-derived growth factor	
	Fibroblast growth factor	
	Vascular endothelial growth factor	
	Gonadotropin-releasing hormone (GnRH)	

Growth factors are peptides that bind to specific cell membrane receptors and initiate intracellular signaling pathways. Because the growth factors are potent mitogens, it is also not surprising that the follicular phase of the cycle, associated with proliferative activity of the endometrium, is marked by dramatic alterations in growth

factors. Estrogen stimulates gene expression for epidermal growth factor (EGF) (and its receptor) and insulin-like growth factor (IGF) production. In turn, EGF elicits estrogen-like actions by interacting with the estrogen receptor mechanism.[57] EGF, a potent mitogen, is present in endometrial stromal and epithelial cells during the follicular phase of the cycle and in the stromal cells during the luteal phase.[58] Transforming growth factor-a (TGF-a) and EGF work through the same receptor and are important mediators of estrogen-induced growth of the endometrium. TGF-a levels peak at midcycle, in contrast to EGF levels, which are relatively stable and noncyclic.[59,60] and [61] Platelet-derived growth factor is a potent mitogen localized to stromal cells.



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The insulin-like growth factors promote cellular mitosis and differentiation. They are expressed in a pattern controlled by estrogen and progesterone. IGF-I is predominant in proliferative and early secretory endometrium, and IGF-II appears in the mid to late secretory phase and persists in early pregnancy decidua.[62] Endometrial IGF-I expression is correlated with the circulating estrogen levels during the menstrual cycle.[63] This suggests that IGF-I synthesis is regulated by estrogen and mediates estrogen-induced growth of the endometrium, and IGF-II is

involved in differentiation in response to progesterone. Evidence in the monkey indicates that IGF-I is the primary regulator of myometrial growth in response to estrogen as well as to estrogen plus progesterone.[64]

As elsewhere in the body, the myometrial IGF activity is modulated by the IGF binding proteins, which respond to the sex steroids in a differential manner; IGFBP-2 parallels IGF-I response, whereas IGFBP-3 is decreased in muscle but increased in vascular endothelium by estrogen.[65] IGFBP-4 and IGFBP-5 respond to estrogen but are unaffected by the addition of progesterone. IGFBP-1, as discussed later, is a major product of decidualized endometrium.

Gonadotropin-releasing hormone (GnRH) is present in endometrium and in increased amounts in secretory endometrium and decidua.[66] In human decidual cells, GnRH increases the expression of matrix metalloproteinases, suggesting a role for GnRH in the regulation of the enzymes involved in implantation.[67] Like all of these molecules, GnRH is involved in signaling pathways associated with cell proliferation and breakdown, interacting with adhesion factors such as integrins, enzymes, and angiogenic substances.[68]

Human myometrial smooth muscle and endometrial stromal cells express mRNA for parathyroid hormone-like protein, the function of which is unknown.[69] Transforming growth factor-ß (TGF-ß) stimulates the production of the parathyroid hormone-like protein. TGF-ß production is greatest in the secretory phase and may inhibit cellular proliferation by increasing IGFBP-3 synthesis.

Prostaglandins are produced by both epithelial and stromal cells, and the prostaglandin content in the endometrium reaches a peak level in late secretory endometrium. The predominant prostaglandin produced by endometrium is prostaglandin F_{2a}, a potent stimulus for myometrial contractions.[70] Endometrial prostaglandin production decreases dramatically after implantation, suggesting the presence of an active mechanism for suppression.[71] The production of prostaglandins requires estrogen support, but the increased production by secretory endometrium indicates progesterone enhancement, and acute withdrawal of progesterone promotes a further increase.[70] Endometrial stromal cells produce prostacyclin and thromboxane in response to estrogen, a response that can be blocked by progestins.[72] The myometrium principally produces prostacyclin, utilizing precursors derived from the endometrium. However, receptors for all members of the prostaglandin family are present on human myometrial cells, and contraction of the myometrium is a major consequence of prostaglandin F_{2a}.[73]

Thromboxane is synthesized by uterine tissues. Gene expression for the thromboxane synthase and for the thromboxane receptor can be identified in endometrial glands, stromal cells, myometrial smooth muscle, and uterine blood vessels.[74] Thromboxane A_2 is a potent vasoconstrictor and stimulator of smooth muscle cells. Because of its rapid metabolism, it is limited to autocrine and paracrine activity.

Women with excessive menstrual bleeding have alterations in the normal rates of prostaglandin production. For this reason, effective reductions in menstrual blood loss can be achieved with treatment utilizing one of the nonsteroidal anti-inflammatory agents that inhibit prostaglandin synthesis. These agents are also effective treatment for prostaglandinmediated dysmenorrhea.

Fibronectin and laminin are extracellular matrix substances that are secreted by stromal cells of the endometrium in response to progesterone.[75] These proteins are important

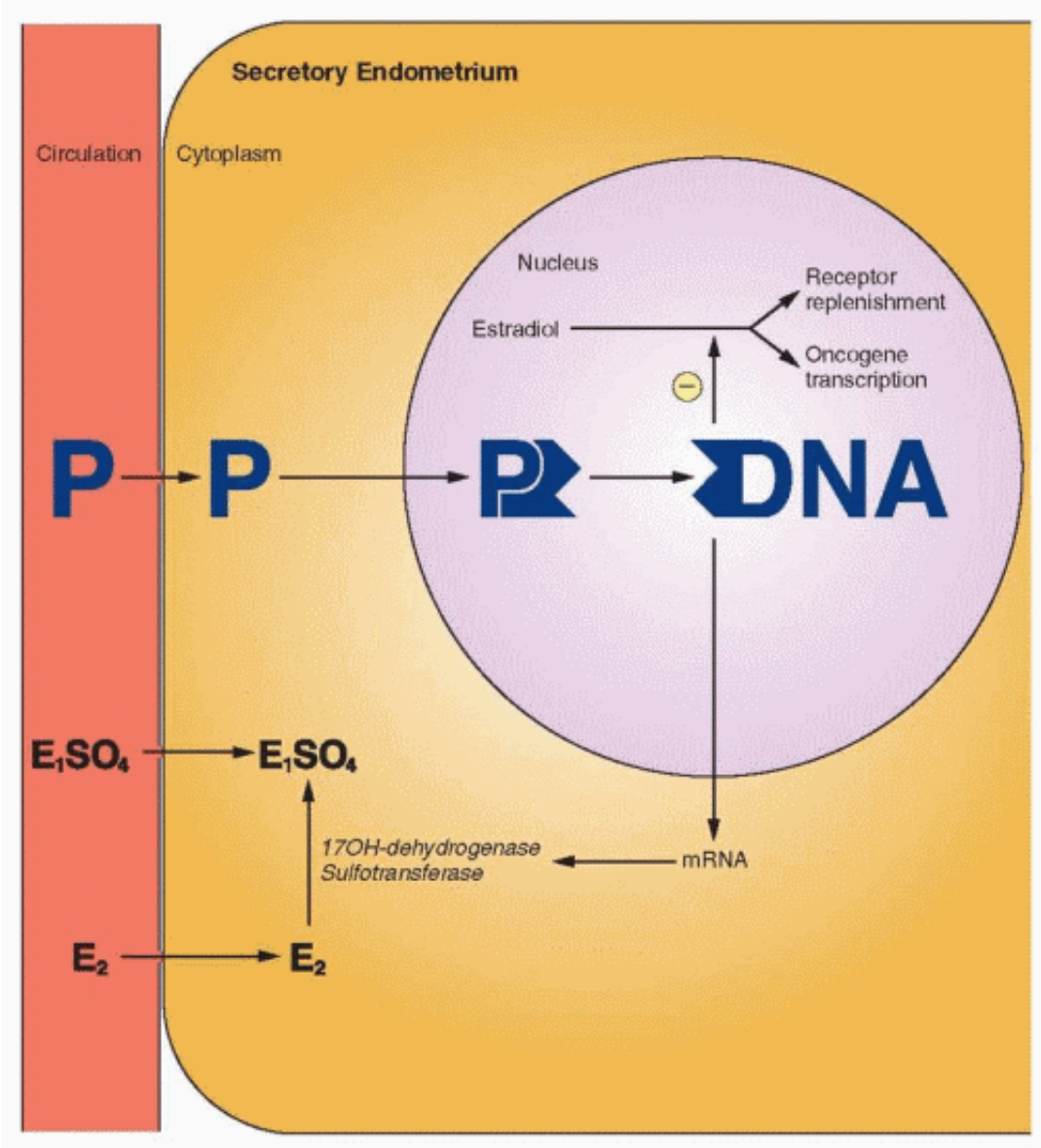
adhesion molecules during implantation. Integrins are a family of glycoproteins that function as receptors for proteins such as collagen, fibronectin, and laminin. The integrins are highly expressed in endometrium and are

important for cell-to-cell and cell-to-matrix interactions.[76] The expression of integrins is regulated by cytokines and growth factors, not estrogen and progesterone.[77]

Endothelins are potent vasoconstrictors produced in the vascular endothelial cells. The vasoconstrictor activity of endothelin-1, present in the endometrium, is balanced by the fact that it promotes the synthesis of the vasodilators nitric oxide and prostacyclin. Endothelin-1 is synthesized in endometrial stromal cells and the glandular epithelium, stimulated by both TGF-ß and interleukin-1a.[78] Endothelin-1 is at least one agent responsible for the vasoconstriction that shuts off menstrual bleeding. It is also a potent stimulator of myometrial contractions and can contribute to dysmenorrhea. Finally, endothelin-1 is a mitogen and can promote the healing re-epithelialization of the endometrium. Human decidual cells also synthesize and secrete endothelin-1, from where it may be transported into the amniotic fluid.[79]

Angiogenesis, the formation of new blood vessels, is an essential process in tissue growth and development. Angiogenesis is necessary for tumor growth, and, in normal tissues, it is usually kept in check by regulating factors. The female reproductive tissues (specifically ovarian follicles, the trophoblast, and the endometrium), however, must experience periodic

and rapid growth and regression. In these tissues, angiogenesis is part of normal events. The endometrium is a major source for angiogenic factors during the menstrual cycle and during pregnancy.[80] Vascular endothelial growth factors (VEGFs), a collection of specific mitogens for endothelial cells, are abundantly expressed in human endometrium, reaching a peak that correlates with the maximal angiogenesis reached during the secretory phase.[81,82] The VEGF family contains six growth factors and utilizes three different receptors. During the proliferative phase, estrogen stimulates VEGF synthesis. VEGF expression is also stimulated by hypoxia, specifically the hypoxia associated with endometrial breakdown, and the new blood vessel growth as well as the re-epithelialization of the endometrium in the new proliferative phase are dependent on these growth factors in response to estrogen.[83,84]



Angiogenesis is also influenced by many other growth factors and other substances such as fibronectin and prostaglandins. The fibroblast growth factor family, in particular, is highly mitogenic for endothelial cells and endometrial stromal cells. Angiopoietins sustain the endometrium by preventing apoptosis and stabilizing blood vessels. The endometrium also produces inhibitory proteins, and the final growth of blood vessels reflects the balance between the inhibitory and stimulatory factors.

In all types of endometrial and myometrial cells, estrogen receptor expression reaches a maximum in the late proligerative phase.[85,86] The concentration is greatest in the glandular epithelium. During the early secretory phase, estrogen receptor expression declines, followed by an increase in the mid and late secretory phases. These changes reflect the cyclic changes in estradiol (which increases estrogen receptor expression) and progesterone (which decreases estrogen receptor expression). Although estrogen receptor-beta is present in human endometrium, it is less prominent than estrogen receptor-alpha and exhibits less change during the cycle, except when it becomes the predominant estrogen receptor in the endometrial vasculature in the late secretory period.[87] Estrogen stimulation of proliferation is largely, if not totally, mediated by estrogen receptor-alpha.

Progesterone receptor expression in endometrial glandular epithelium reaches a maximum in the late proliferative

and early secretory phases (reflecting induction of progesterone receptor by estrogen) and then declines to nearly undetectable levels by the midpoint of the secretory phase. Stromal cells in the endometrium show only minor fluctuations in progesterone receptors during the menstrual cycle. Decidualizing stromal cells exhibit strong progesterone receptor expression, although progesterone receptors are absent from decidual epithelial cells. Smooth muscle cells of the uterus demonstrate strong progesterone receptor expression throughout the menstrual cycle. Many of the events in uterine growth and function are regulated by the interplay between estrogen and progesterone. In general, progesterone antagonizes estrogen stimulation of proliferation and metabolism. This antagonism can be explained by the effects of progestins on the estrogen receptor (a decrease in levels), on the enzymes that lead to excretion of estrogen from cells, and by progesterone suppression of estrogen-mediated transcription of oncogenes.

Androgen receptor is present in endometrium at all stages of the menstrual cycle, in postmenopausal endometrium, and in the decidua of pregnancy.[88] Surprisingly, the androgen receptor concentration is constant throughout the cycle. Androgens suppress the proliferative effects of estrogen on the endometrium, and experimental evidence suggests that the suppressive effects on the endometrium induced by antiprogestational agents are mediated by the androgen receptor.[89]

The complexity of the endometrium can be appreciated by viewing the results of complementary DNA microarray studies. In one effort directed just to the endometrial breakdown associated with menstruation, 571 transcripts were identified that were involved in 131 biochemical pathways, including thyroid hormone synthesis and metabolism![90] Gene expression studies are just beginning to profile the patterns associated with specific hormones and pharmacologic agents.[91]

The Decidua

The decidua is the specialized endometrium of pregnancy. The biochemical dialogue between the fetoplacental unit and the mother must pass back and forth through the decidua. The classic view of the decidua conformed to its designation as a thin line in anatomic diagrams, a minor, inactive structural component. We now know that the decidua is a vigorous, active tissue.

Decidual cells are derived from the stromal cells of the endometrium, under the stimulation of progesterone. This transformation is regulated by members of the transforming growth factor beta family, including activin A.[92,93] In addition, ghrelin acting via the growth hormone receptor is involved in this process.[94]

Decidual cells appear during the secretory phase and continue to proliferate during early pregnancy, eventually lining the entire uterus including the implantation site. The decidual cell is characterized by the accumulation of glycogen and lipid droplets and the new expression of a host of substances, including prolactin, relaxin, renin, insulin-like growth factors (IGFs), and insulin-like growth factor binding proteins (IGFBPs). There is no evidence that these proteins are secreted into the circulation; therefore, they serve as autocrine and paracrine agents.[95,96]

Riddick was the first to detect prolactin in the decidualizing endometrium of the late luteal phase.[97] The amino acid sequence and the chemical and biologic properties of decidual prolactin are identical to those of pituitary prolactin. Decidual prolactin synthesis and release are controlled by the placenta, fetal membranes, and decidual factors. Dopamine, bromocriptine, and thyrotropin-releasing hormone (TRH), in contrast to their action in the pituitary, have no effect on decidual synthesis and release of prolactin. A protein named decidual prolactin-releasing factor has been purified from the placenta, and an inhibiting protein, which blocks the stimulatory activity of the releasing factor, has been purified from decidua.[96] IGF-1, relaxin, and insulin all stimulate decidual

prolactin synthesis and release, each through its own receptor. The same decidual cells produce both prolactin and relaxin. Prolactin exerts an overall inhibitory effect on the process of decidualization, perhaps an autocrine mechanism to limit the extent of decidualization.[98]

Lipocortin-1 is a calcium- and phospholipid-binding protein, present in the placenta and decidua, that inhibits phospholipase A_2 and responds to glucocorticoids. Lipocortin-1 inhibits decidual prolactin release but in a mechanism independent of phospholipase action and independent of glucocorticoids. The prostaglandin system is not involved in decidual prolactin production, and corticosteroids do not affect decidual prolactin release.[99]

There is good reason to believe that amniotic fluid prolactin is derived from the decidua. In vitro experiments indicate that the passage of prolactin across the fetal membranes is in the direction of the amniotic cavity. The amniotic fluid concentration correlates with the decidual content, not with maternal circulating levels. Amniotic fluid prolactin reaches peak levels in the first half of gestation (about 4,000 ng/mL) when maternal plasma levels are approximately 50 ng/mL and fetal levels about 10 ng/mL. Maternal circulating prolactin reaches maximal levels near term. Finally, amniotic fluid prolactin is unaffected by bromocriptine treatment (which reduces both fetal and maternal circulating levels to baseline levels).

It is believed that decidual prolactin regulates amniotic fluid volume and electrolyte concentrations. Prolactin regulates water and ion transport in lower animals, and prolactin binds to amniotic membranes. Disorders in human pregnancy associated with abnormal amniotic fluid volumes may be explained by this mechanism, especially idiopathic polyhydramnios, which is associated with a decrease in the number of prolactin receptors in the

membranes. Prolactin may be involved in the regulation of surfactant synthesis in the fetus, and prolactin may inhibit uterine muscle contractility. Prolactin suppresses the immune response and helps to prevent immunologic rejection of the conceptus. Prolactin can also function as an autocrine and paracrine growth factor in the uterus.[100]

Fibroblast growth factor, derived from decidua, stimulates blood vessel growth in early pregnancy. Another factor, endothelial-cell-stimulating angiogenesis factor (a nonprotein mitogen), is also derived from decidua and contributes to the vascularization of the decidua during the first trimester of pregnancy.[101] The expression of corticotropin-releasing hormone (CRH) has been demonstrated in human decidua, and many actions for decidual CRH are possible: activation of prostaglandins, stimulation of myometrial contractions, and a contribution to both maternal and fetal stress responses during pregnancy and labor.[102]

Prorenin (the inactive precursor of renin) is produced in decidua in response to IGF-1, insulin, endothelin, and relaxin.[103,104] and [105] A uterine role for renin has not been determined.

The insulin-like growth factor-binding proteins, IGFBP-1, -2, -3, and -4, are produced by endometrial stromal cells.[106] Large amounts of IGFBP-1 are present in amniotic fluid. The IGFBPs appear to be regulated by insulin, the IGFs, and relaxin.[107] Relaxin is related structurally to insulin and the IGFs, and it stimulates IGFBP-1 production in endometrial stromal cells.[108] IGFBP-1 is considered to be a marker for decidualization. Because it binds growth-promoting IGFs, the appearance of IGFBP-1 contributes to differentiation rather than proliferation of the endometrial stromal cells.

IGFBP-1 begins to appear in midsecretory phase endometrium and reaches a level of major production in decidua by late in the first trimester of pregnancy. IGFBP-1, when first identified, was known as placental protein 12 and then as pregnancy-associated a-globulin. By the second trimester of pregnancy, high levels of IGFBP-1 are present in the amniotic fluid and the circulation, and then fall significantly during the third trimester. The decidual

production of IGFBP-1 is correlated with the morphologic and histologic changes induced by progesterone and regulated by progesterone, relaxin, insulin, IGF-I, and IGF-II. In fact, IGFBP-1 is a mediator of progesterone-induced decidualization of endometrial stromal cells.[109] Binding of the insulin-like growth factors to the IGFBPs would limit further mitogenic activity in the endometrium in the secretory phase and during pregnancy. In addition, decidual IGFBP-1 may contribute to the limitation of trophoblast invasion.

The continuous stimulation of IGFBP-1 production by human endometrium can be maintained in women as long as they retain an intrauterine device that releases a progestin into the endometrial cavity.[110] In endometrial samples from these women, areas of endometrial atrophy correlate with intense staining for IGFBP-1. This makes a strong argument for the importance of insulin-like growth factors for endometrial growth and the potential for prevention of endometrial growth by providing IGFBP-1.

The glycoprotein a subunit, common to follicle-stimulating hormone (FSH), luteinizing hormone (LH), thyroid-stimulating hormone (TSH), and hCG, is secreted into the circulation by the pituitary and placenta. A specific role for the a subunit has not been apparent; however, gonadotropin receptors are present in the endometrium, and a subunit acts synergistically with progesterone to induce decidualization of endometrial cells in vitro.[111] In addition, the a subunit stimulates decidual prolactin secretion.[112]

The chorion laeve, villous trophoblast, and decidua are all sites of TGF-ß production.[113] TGF-ß can signal its own production; thus, TGF-ß can be a messenger from fetal tissues to decidua. TGF-ß is also believed to play a role in limiting trophoblastic invasion.[114] This may be accomplished by stimulating the production of plasminogen activator inhibitor and the factor that causes tissue inhibition of metalloproteinases.[115]

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SUMMARY: The Uterus Is an Endocrine Organ

One cannot dispute the fact that the uterus is an endocrine organ, but the vast array of active substances with their complicated names can be bewildering and overwhelming. It is helpful to keep in mind a fundamental and relatively simple description: the endometrium is necessary for reproduction, and the synchronous, complex cycle of events is dependent on the endocrine guidance of estradiol and progesterone, modulated and mediated by the plethora of locally produced biochemical agents. Each and every signaling substance utilizes one of the pathways discussed in* Chapter 2 *and makes a contribution to the dynamic sequence of morphological and biochemical events repeatedly dedicated to nourishing and supporting an early embryo.

Anatomical Abnormalities of the Uterus

Congenital abnormalities of the müllerian ducts are relatively common, occurring in 7% to 10% of all women, and contributing to the problems of infertility, recurrent pregnancy loss, and poor pregnancy outcomes that occur in approximately 25% of women with uterine anomalies.[116,117,118,119,120] and [121] Major anomalies are about 3 times more common in women with recurrent miscarriages.[122] The problems encountered in pregnancy include preterm labor, breech presentations, and complications that lead to interventions and greater perinatal mortality. Cervical cerclage is often indicated for prevention of preterm labor due to these anomalies. In addition, these abnormalities can produce the symptoms of dysmenorrhea and dyspareunia and even amenorrhea. Endometriosis in young women, especially adolescents, should raise clinical suspicion of genital tract malformations. Because the embryologic origin of the ovaries is separate and distinct from that of the müllerian structures, patients with

müllerian anomalies have normal ovaries and ovarian function. Conception and implantation are not prevented. Surgical correction is recommended for pain, endometriosis due to obstruction, and poor obstetrical outcomes.

Incidence of Müllerian Defects

Fertile and infertile women	3-4%[123]
Women with recurrent miscarriages	5-10%[120]
Women with late miscarriages and preterm deliveries	>25%[120]

Anomalies can result from the failure of the müllerian ducts to fuse in the midline, to connect with the urogenital sinus, or to create the appropriate lumen in the upper vagina and uterus by resorption of the central vaginal cells and the septum between the fused müllerian ducts. Because fusion begins in the midline and extends caudally and cephalad, abnormal results can exist at either end. Formation of the uterine cavity begins at the lower pole and extends cephalad with dissolution of midline tissue; hence, incomplete resorption of tissue commonly yields persistence of the midline uterine wall intruding into the cavity. The molecular pathophysiology of these abnormalities has been insufficiently studied; however, the association with other somatic anomalies and occasional reports of familial transmission suggest genetic linkages.

Vaginal outflow tract obstruction can be minimal with a transverse septum or complete due to agenesis. A septum is the result of a defect in the connection of the fused müllerian

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ducts to the urogenital sinus or a failure of canalization of the vagina. The location of the septum varies, although it is usually in the upper or middle third of the vagina. Vaginal agenesis is the result of a complete failure in canalization; these patients present with amenorrhea or pain due to accumulated menstrual effluvium. Surgical correction is frequently necessary to relieve the relative constriction (and obstruction) of the vaginal canal. An absent vagina is usually accompanied by an absent uterus and tubes, the

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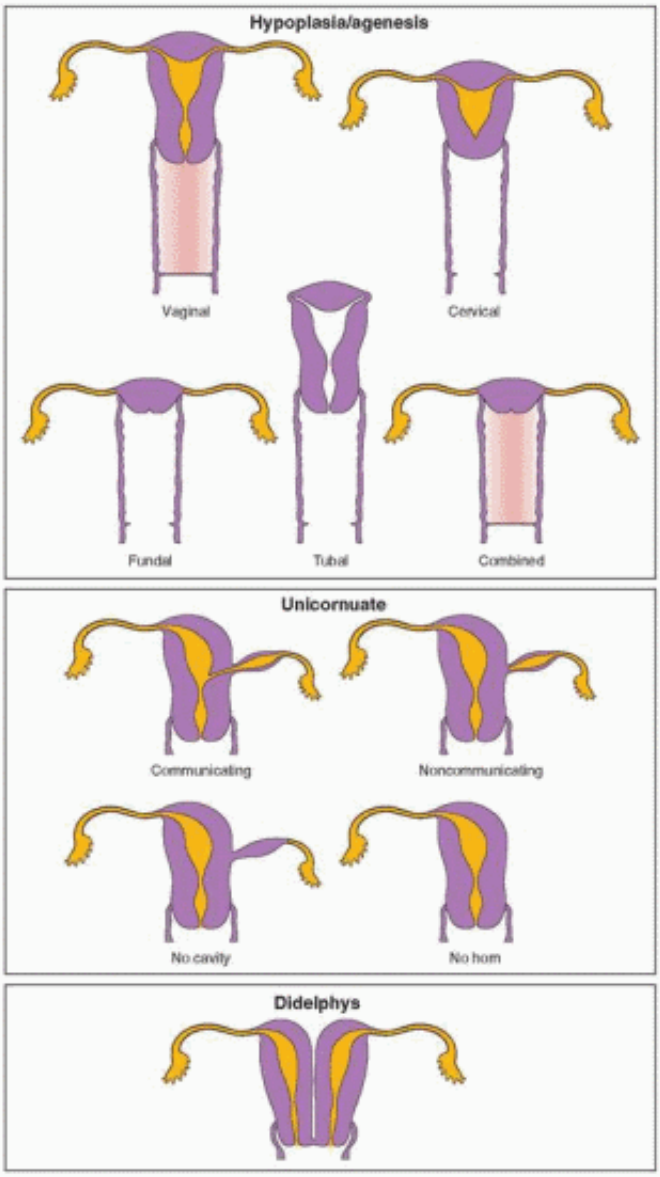
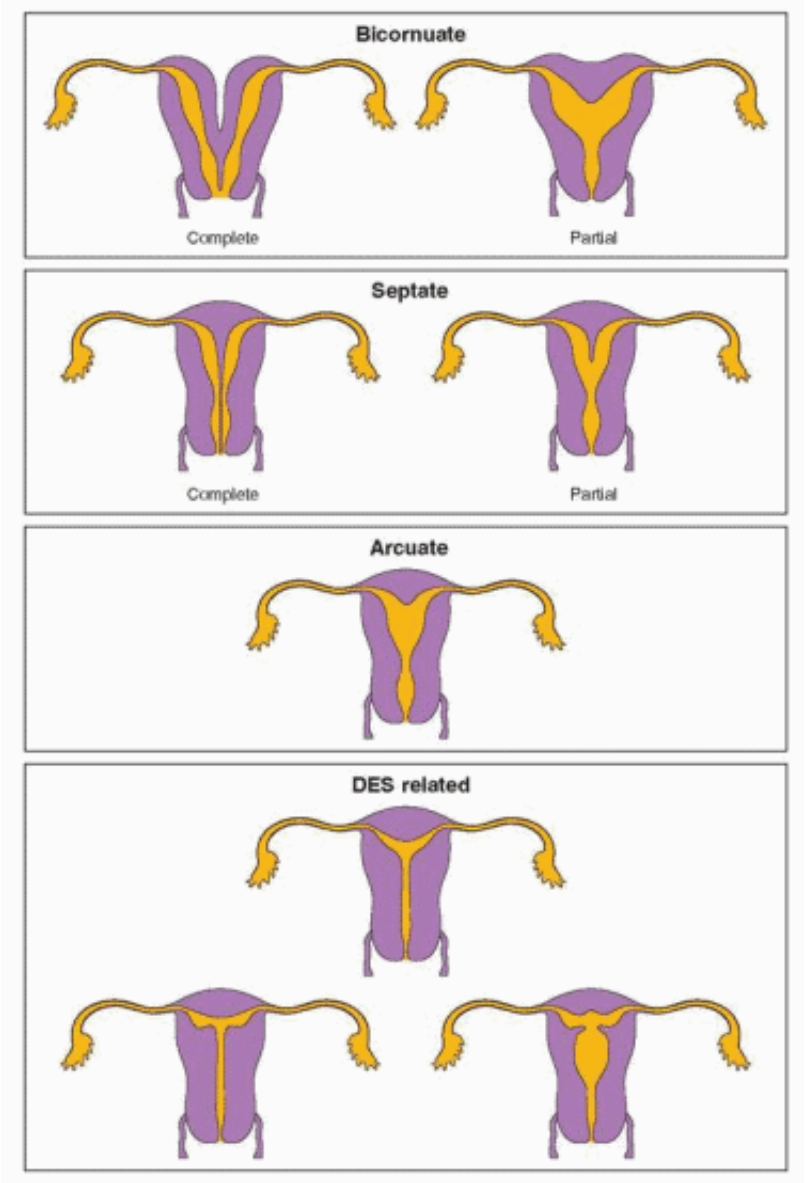
classic müllerian agenesis of the Mayer-Rokitansky-Kuster-Hauser syndrome (discussed in Chapter 11).


Hypoplasia/agenesis
Vaginal
Cervical
Fundal
Tubal
Combined
Unicornuate
Communicating
Noncommunicating
No cavity
No horn
Didelphys



Distribution of Specific Anomalies[123]

Septate uterus	35%
Bicornuate uterus	26%
Arcuate uterus	18%
Unicornuate uterus	10%

Uterus didelphys	8%

Uterine anomalies can be organized into the following categories.[124] Each of these can be associated with obstructions that present during adolescence with amenorrhea and cyclic pain.[125]

Uterus Didelphus (Double Uterus)

Lack of fusion of the two müllerian ducts results in duplication of corpus and cervix. These patients usually have no difficulties with menstruation and coitus, except when there is also a midline longitudinal vaginal septum. Occasionally, one side is obstructed and symptomatic. In addition, a double uterus is occasionally associated with an obstructed hemivagina (often with ipsilateral renal agenesis); early diagnosis and excision of the obstructing vaginal septum will preserve fertility. Pregnancy is associated with an increased risk of miscarriage, malpresentations, and premature labor, although many patients will have no reproductive difficulties.[123,126,127] Unification of the two endometrial cavities by metroplasty is indicated in patients with repeated poor obstetrical outcomes.

Unicornuate Uterus

An abnormality that is unilateral obviously is due to a failure of development in one müllerian duct (probably a failure of one duct to migrate to the proper location). The altered uterine configuration is associated with an increase in endometriosis and in obstetrical complications (early spontaneous miscarriage, ectopic pregnancy, abnormal presentations, intrauterine growth retardation, and premature labor).[126,128,129,130] and [131] There may be a rudimentary horn present, and implantation in this horn is followed by a very high rate of pregnancy wastage or tubal pregnancies. A rudimentary horn can also be a cause of chronic pain, and surgical excision is worthwhile. However, most rudimentary horns are asymptomatic because they are noncommunicating, and the endometrium is not functional. Because of the potential for problems, prophylactic removal of the rudimentary horn is recommended when it is encountered during a surgical procedure. Approximately 40% of patients with a unicornuate uterus will have a urinary tract anomaly (usually of the kidney).[132] Surgical reconstructive procedures do not improve obstetrical outcomes; however, cervical cerclage may be beneficial when indicated.

The Bicornuate Uterus

Partial lack of fusion of the two müllerian ducts produces a single cervix with a varying degree of separation in the two uterine horns. This anomaly is relatively common, and

pregnancy outcome has usually been reported to be near normal. Some, however, find a high rate of early miscarriage, preterm labor, and breech presentations.[119,126] With a history of repeated poor pregnancy outcome, surgical metroplasty is worth consideration.

The Septate Uterus and the Arcuate Uterus

Partial lack of resorption of the midline septum between the two müllerian ducts results in fibromuscular defects that range from a slight midline septum (the arcuate, heart-shaped cavity) to a significant midline division of the endometrial cavity. A total failure in resorption can leave a longitudinal vaginal septum (a double vagina). This defect is not a cause of infertility, but once pregnant, the greater the septum the greater the risk of recurrent

spontaneous miscarriage, especially in the second trimester. The complete septate uterus is associated with a high risk of spontaneous miscarriage, preterm labor, intrauterine growth retardation, and breech presentation.[119,133,134] Even a small septum is associated with these poor obstetrical outcomes.[135] Outcomes are excellent with treatment by hysteroscopy.[123,134,136,137,138,139] and [140] Post treatment miscarriage rates are approximately 10% in contrast to the 90% pretreatment rates with a complete septum. A longitudinal vaginal septum usually does not have to be excised (unless dyspareunia is a problem). In some reports, the arcuate uterus has no adverse impact on reproductive outcome.[126] Prophylactic surgery is considered appropriate for a septate uterus in older women and in women being treated with in vitro fertilization. A surgical procedure is not indicated for the arcuate uterus.

Very Rare Anomalies

Isolated agenesis of the cervix or the endometrium is incredibly rare. Absence of the cervix can lead to so much pain and obstruction that hysterectomy is the best solution. Attempts to preserve fertility by creating a fistulous communication between uterus and vagina have achieved some success, but repeat surgery due to reappearance of obstruction is common.[141,142] In asymptomatic patients, consideration should be given to preservation of structures for the possibility of pregnancy that can be achieved by means of one of the techniques of assisted reproduction (Chapter 32).

The Diethylstilbestrol-Associated Anomaly

Mothers who were treated in 1938 to 1975 with high doses of estrogen early in their pregnancies had children who developed a variety of anomalies, ranging from the hypoplastic T-shaped uterus to irregular cavities with adhesions.[143] Women with uterine abnormalities usually also have cervical defects. In these individuals, the chance of term pregnancy is decreased because of higher risks of ectopic pregnancy, spontaneous miscarriage, and premature labor.[144] An incompetent cervix is common. Poor outcome is correlated with an abnormal uterus on hysterosalpingography. No treatment is available beyond cervical cerclage.

Accurate Diagnosis of Anomalies

In the past, full diagnosis required surgical intervention, first laparotomy and then, more recently, laparoscopy. Today, vaginal ultrasonography, especially three-dimensional ultrasound,

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sonohysterography, and magnetic resonance imaging are highly accurate, and surgical intervention is usually not necessary.[145,146] and [147] Hysterosalpingography alone can yield inaccurate results due to a failure to perfuse both uterine horns on either side of a midline division, and cannot reliably distinguish bicornuate and septate uteri. Decisions should not be based on hysterosalpingography alone. Congenital anomalies of the müllerian ducts are frequently accompanied by abnormalities in the urinary tract, such as a horseshoe or pelvic kidney. Renal agenesis can be present on the same side as a müllerian defect.

Pedro Acién at the San Juan University Hospital in Alicante, Spain, is an acknowledged expert on the many and varied malformations of the female genital tract. He advocates a more complete classification, that includes müllerian anomalies with anomalies of the urogenital ridge, the mesonephric structures, and the cloaca.[148] The embryologic origins of the various anomalies and an understanding of unusual cases can be obtained through Acién's publications.[5,148]

Leiomyomas (Uterine Fibroids)

Uterine leiomyomas are benign neoplasms that arise from uterine smooth muscle and cause abnormal uterine bleeding and symptoms secondary to a large pelvic mass. It is hypothesized that leiomyomas originate from somatic mutations in myometrial cells, resulting in progressive loss of growth regulation.[149,150] The tumor grows as genetically abnormal clones of cells derived from a single progenitor cell (in which the original mutation took place). Studies indicate that leiomyomas are monoclonal.[151] Different rates of growth can reflect the different chromosomal abnormalities present in individual tumors. Multiple myomas within the same uterus are not clonally related; each myoma arises independently.

The presence of multiple myomas (which have a higher recurrence rate than single myomas) argues in favor of a genetic predisposition for myoma formation. There is about a 2.5-fold increased risk of developing myomas in first-degree relatives of women with these tumors.[152] Hereditary leiomyomatosis and renal cell carcinoma is an autosomal dominant syndrome with both cutaneous and uterine leiomyomas. The risk of renal cell carcinoma and that of leiomyosarcoma are increased in this syndrome.[153,154] The gene involved is *fumarate hydratase*, coding for an enzyme involved in the Kreb's cycle. A family history of cutaneous leiomyomata should trigger screening for this gene mutation. Renal cell cancer occurs in 10-16% of women with this syndrome. Studies of DNA polymorphisms will undoubtedly yield patterns identifying women at high risk for uterine leiomyomata, and perhaps risk for recurrence following ablation treatments and for malignant progression to leiomyosarcoma. Thus far, chromosomal abnormalities have been described in about 40% of myomas.[155] Another approach is to identify the microRNA pattern associated with leiomyoma size, growth rates, and ethnic prevalence.[156]

It is not certain whether leiomyosarcomas arise independently or from leiomyomas. However, the incidence of leiomyosarcomas in patients with leiomyomas is very low (less than 1%).[157] Gene profiling has not discovered shared abnormalities or a common molecular pathway comparing myomas with leiomyosarcomas.[158]

If surgical specimens are serially sectioned, about 77% of women who come to hysterectomy will have myomas, many of which are occult.[159] By the age of menopause, ultrasound can identify myomas in about 80% of black American women and 70% of white American women.[160] In the United States, about 40% of abdominal hysterectomies and 17% of vaginal hysterectomies are performed for leiomyomas.[161] The peak incidence for myomas requiring surgery occurs around age 45, approximately 8 cases per 1,000 women

each year.[162] In the United States, approximately 10-15% of women require hysterectomy for myomas. For unknown reasons, uterine leiomyomas are 2-3 times more prevalent in black women compared with white, Hispanic, and Asian women and account for 75% of hysterectomies among black women.[160,163,164]

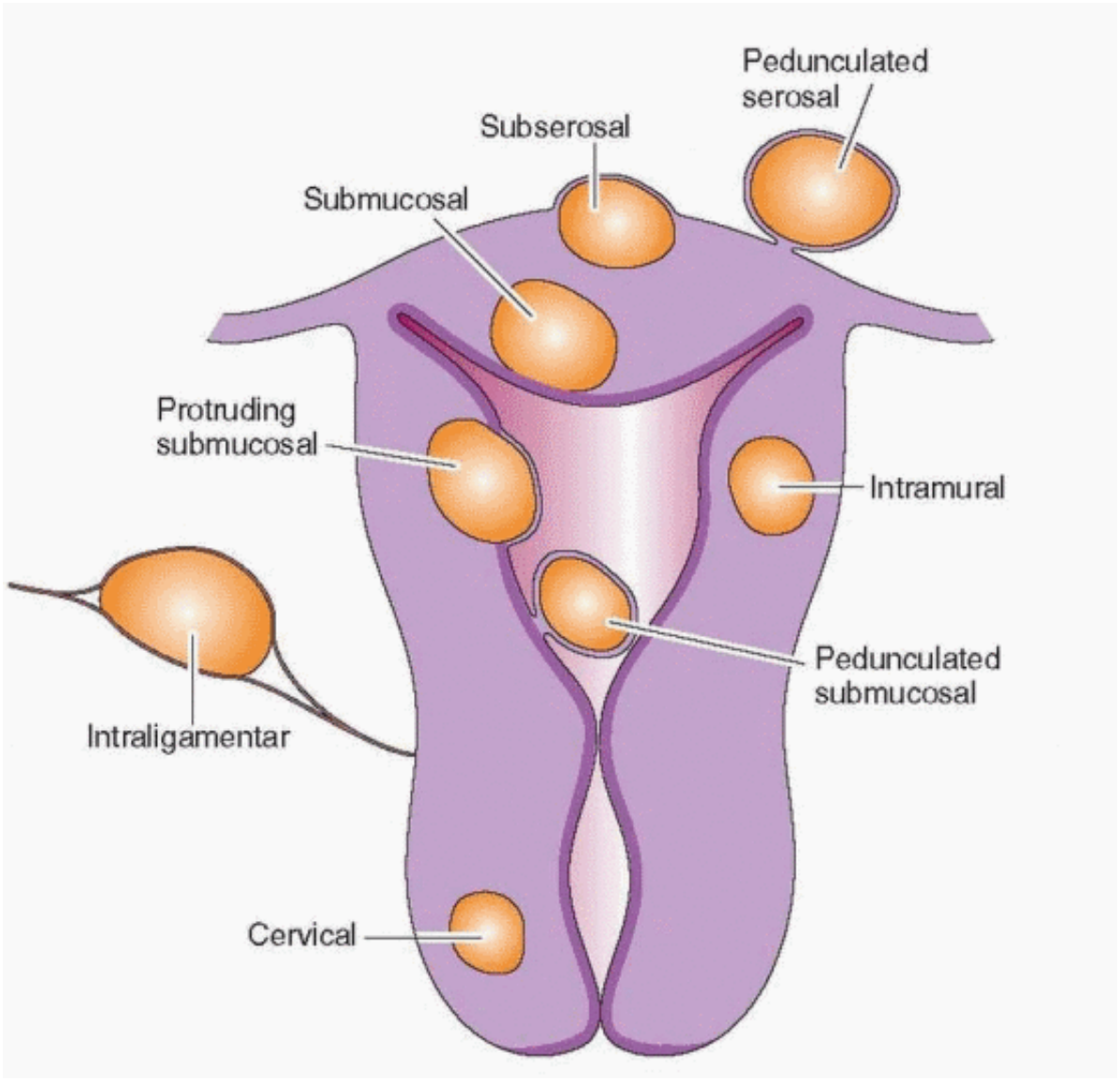
Myomas are present (diagnosed by ultrasonography) in about 30% of women, and in about 1-2% of pregnancies.[165,166] The risk of myoma is decreased with increasing parity and with increasing age at last term birth.[166,167] Women with at least two full-term pregnancies have half the risk for myomas. Smoking decreases the risk (presumably by decreasing estrogen levels), and obesity increases the risk (presumably by increasing estrogen levels). Although a lower risk for myomas is associated with factors that decrease estrogen levels, including leanness, smoking, and exercise, the use of oral contraceptives is not associated with an increased risk of uterine myomas, although the Nurses' Health Study reported a slightly increased risk when oral contraceptives were first used in early teenage years.[167,168] and [169]

The hormone sensitivity of leiomyomas is further indicated by the following clinical observations. Leiomyomas

develop during the reproductive (hormonally active) years and regress after menopause. Occasionally, leiomyomas grow during pregnancy, and the hypogonadal state induced by treatment with gonadotropin-releasing hormone (GnRH) agonists often causes shrinkage of myomas.

The environment within the leiomyoma is hyperestrogenic. The estradiol concentration is increased, and leiomyomas contain more estrogen and progesterone receptors.[170,171,172] and [173] Aromatase gene and enzyme expression are present in significant levels in leiomyomas.[174] Indeed, leiomyoma tissue is hypersensitive to estrogen and appears to have lost a regulatory influence that limits estrogen response.[175] Endometrial hyperplasia can be observed at the margins of submucosal myomas.[176] In the myometrium and in leiomyomas, peak mitotic activity occurs during the luteal phase, and mitotic activity is increased by the administration of high doses of progestational agents.[177,178] These facts indicate that progesterone stimulates mitotic activity in leiomyomas, although animal studies indicate both stimulation and inhibition of myometrial growth. Similarly, clinicians have reported both regression

and growth with progestational treatment. Nevertheless, most of the evidence supports a growth-promoting role for progestins. The association with estrogen can be explained by the estrogen enhancement of progesterone receptor expression.[179,180] Treatment with mifepristone, the progesterone antagonist, or with asoprisnil, a selective progesterone receptor agonist/antagonist, is associated with a reduction in leiomyoma size.[181,182]



At least one pathway for the growth-promoting effect of progestins is the induction of *BCL2* gene expression increasing the production of the Bcl-2 protein that inhibits apoptosis and promotes cell replication.[183] Bcl-2 protein expression is increased in leiomyoma cells and markedly increases in response to progesterone.[184] In contrast, normal myometrial cells do not respond to estradiol or progesterone with Bcl-2 protein expression, and there is no cyclic change throughout the menstrual cycle.

As in the normal uterus, the effects of estrogen and progestins on leiomyomas are mediated by growth factors.[185] EGF is overexpressed in myomas, EGF receptors are present in leiomyomas, and GnRH agonist treatment (and hypogonadism) decreases EGF concentration in myomas (but not in normal myometrium).[186,187] IGF-I and IGF-II and their receptors are abundant in myometrium and actively overexpressed in leiomyomas.[188,189] Leiomyomas express more IGF-II and less IGFBP-3 than myometrium, a situation that would enhance growth factor availability and activity in the tumor.[190] Leiomyoma cells express more parathyroid hormone-related protein (another growth factor) than normal myometrium.[191] Like the endometrium and myometrium, leiomyomas secrete prolactin, and prolactin functions in the uterus as a growth factor.[100] Even hematopoiesis is possible in a leiomyoma.[192]

One of the consequences of altered growth factor expression in myomas is an abnormal vasculature, characterized by a dilated venous plexus.[193] This morphologic feature may be the result of specific vascular regulators of angiogenesis, such as fibroblast growth factor and vascular endothelial growth factor. These changes probably contribute to the heavy menstrual bleeding associated with submucosal myomas.

Uterine growth and signaling molecules are highly expressed in leiomyomas.[194] As with all tumors, these pathways in leiomyomas may one day be targeted by gene therapy. For example, specific adenoviruses can deliver altered genes to myoma cells that can interfere with the gene expression required for tumor growth and cellular functions.

Reproductive Function and Leiomyomas

Leiomyomas are an infrequent cause of infertility, either by mechanical obstruction or distortion (and interference with implantation).[195,196] When a mechanical obstruction of fallopian tubes, cervical canal, or endometrial cavity is present and no other cause of infertility or recurrent miscarriage can be identified, myomectomy is usually followed by a prompt achievement of pregnancy in a high percentage of patients (usually within the first year).[196,197] Small submucosal myomas are best treated by hysteroscopic resection. Preoperative visualization is important, and mapping of myomas by sonohysterography or magnetic resonance imaging (MRI) is superior to standard ultrasonography (which is relatively inaccurate).[198] It is difficult to distinguish between submucosal myomas and endometrial polyps with ultrasonography.[199] Very large myomas (greater than 4-5 cm) and myomas that do not have greater than 50% protrusion into the cavity are not good candidates for hysteroscopic removal.

The 5-year recurrence rate after abdominal myomectomy for a single myoma is about 10%, and 25% with multiple myomas, with subsequent hysterectomy necessary in one-third of patients with recurrence.[200] In a series with long-term follow-up, the recurrence rate over 10 years after single myomectomy reached 27%.[201] Women who gave birth after

myomectomy had a recurrence rate (over 10 years) of 16%, compared to a rate of 28% in those who did not give birth. In an Italian study of recurrence, the rate at 5 years reached 55% in those who did give birth after surgery and 42% in those with no childbirth.[202] These differences may reflect the diligence and sensitivity of the ultrasonographic assessments.

An increased incidence of spontaneous miscarriage because of myomas has not been definitively documented in the literature. Myomectomy for infertility or recurrent miscarriage requires a deliberate and careful decision after all factors have been considered. Intracavitary myomas, however, usually require surgery. Submucosal myomas are associated with general cavitary alterations in the expression of proteins involved with implantation, not just an effect confined to the endometrium over the myoma.[203] Intramural myomas that do not affect the endometrial cavity do not affect implantation or increase the risk of miscarriage.[204, 205] Because of the rapid regrowth of myomas following cessation of GnRH agonist therapy, medical therapy for infertility is not recommended.

Most myomas do not grow during pregnancy.[206] When they do, most of the growth is in the first trimester, and most myomas regress in size after the pregnancy. The size of a myoma will not predict its course; large myomas will not necessarily grow more than small ones. Most pregnancies, in the presence of myomas, will, therefore, be uncomplicated (although a higher incidence of cesarean section has been observed).[165,207] Nevertheless, the risks of malpresentations, preterm delivery, and spontaneous miscarriage are increased.[208] So-called red degeneration of myomas is occasionally observed during late pregnancy, a condition due to central hemorrhagic infarction of the myoma. Pain is the hallmark of this condition, occasionally associated with rebound tenderness, mild fever,

leukocytosis, nausea, and vomiting. Usually pain is the only symptom and resolution follows rest and analgesic treatment.[209] Surgery should be a last resort. The larger the myoma, the greater the risk of premature labor.[210]

Medical Therapy of Leiomyomas

The goals of medical therapy for leiomyomas are to *temporarily* reduce symptoms and to reduce myoma size, and the therapy of choice is treatment with a GnRH agonist.[211] Any treatment that lowers endogenous estrogen levels should be effective, and therefore, the use of aromatase inhibitors is another option.[212] Prolonged medical regimens are expensive and complicated. With few exceptions, surgical treatment is preferred for symptomatic uterine leiomyomas. Medical therapy is provided preoperatively to improve anemia and reduce surgical complexity and recovery times.[213]

The short half-life of GnRH is due to rapid cleavage of the bonds between amino acids 5-6, 6-7, and 9-10. By altering amino acids at these positions, analogues of GnRH can be synthesized with different properties. Substitution of amino acids at the 6 position or replacement of the C-terminal glycine-amide (inhibiting degradation) produces agonists. An initial agonistic action (the so-called flare effect) is associated with an increase in the circulating levels of follicle-stimulating hormone (FSH) and luteinizing hormone (LH). This response is greatest in the early follicular phase when GnRH and estradiol have combined to create a large reserve pool of gonadotropins. After 1-3 weeks, desensitization and down-regulation of the pituitary produce a hypogonadotropic, hypogonadal state. The initial response is due to desensitization, the uncoupling of the receptor from its effector system, whereas the sustained response is due to a loss of receptors by down-regulation and internalization. Furthermore, postreceptor mechanisms lead to secretion of biologically inactive gonadotropins, which, however, can still be detected by immunoassay.

The GnRH analogues cannot escape destruction if administered orally. Higher doses administered subcutaneously can achieve nearly equal effects as those observed with intravenous

treatment; however, the smaller blood peaks are slower to develop and take longer to return to baseline. Other forms of administration include nasal spray, sustained-release implants, and intramuscular injections of biodegradable microspheres.

Treatment with GnRH Agonists

Summarizing the experience with GnRH agonist treatment of leiomyomas, the mean uterine size decreases 30-64% after 3-6 months of treatment.[211] Maximal response is usually achieved by 3 months. The reduction in size correlates with the estradiol level and with body weight. Menorrhagia, anemia, pelvic pressure, and urinary frequency all respond favorably to GnRH agonist treatment.[214,215] A decrease in operative blood loss can be achieved when the pretreatment uterus is as large as a 16-week pregnancy or larger. However, some studies find no benefit in terms of surgical blood loss or length of hospital stay, and surgical dissection may be more difficult because of softening of the myoma.

Why is there a variation in response? When one considers the many factors involved in myoma growth (estrogen, progesterone, growth factors, and receptors), it makes sense that not every myoma is the same. After cessation of GnRH agonist therapy, menses return in 4-10 weeks, and myoma and uterine size return to pretreatment levels in 3-4 months. The rapid regrowth is consistent with the fact that reduction in size is not due to a cytotoxic effect.

Preoperative GnRH agonist therapy offers several advantages for hysteroscopic removal of submucosal tumors. In addition to a decrease in myoma size, endometrial atrophy will improve visualization, and decreased vascularity

will reduce blood loss.

Leiomyomatosis Peritonealis Disseminata is a condition in which multiple small nodules of benign smooth muscle are found throughout the abdominal cavity and occasionally in the pulmonary cavity. This condition appears to be sensitive to estrogen because it has been aggravated by postmenopausal estrogen treatment, and regression has been achieved with GnRH agonist treatment.[216]

Adenomyosis is the ectopic presence of endometrial glands within the myometrium. This diagnosis can be made by magnetic resonance imaging, and successful treatment with a GnRH agonist has been reported.[217,218]

Side Effects with GnRH Agonists

Hot flushes are experienced by more than 75% of patients, usually in 3-4 weeks after beginning treatment. Approximately 5-15% of patients will complain of headache, mood changes, vaginal dryness, joint and muscle stiffness, and depression. About 30% of patients will continue to have irregular (although light) vaginal bleeding. It is useful to measure the circulating estradiol level. If the level is greater than 30 pg/mL, suppression is inadequate. A small number (10%) of patients will experience a localized allergic reaction at the site of injection of depot forms of GnRH analogues. More serious reaction is rare, but immediate and delayed anaphylaxis can occur, requiring intense support and management.[219]

Bone loss occurs with GnRH therapy, but not in everyone, and it is reversible (although it is not certain if it is totally reversible in all patients). A significant vaginal hemorrhage 5-10 weeks after beginning treatment is encountered in about 2% of treated women, due to degeneration and necrosis of submucosal myomas.[220] A disadvantage of agonist treatment is a delay in diagnosis of a leiomyosarcoma. Keep in mind that almost all leiomyosarcomas

present as the largest or only uterine mass. Close monitoring is necessary and surgery has been the usual recommendation when either enlargement or no shrinkage of myomas occurs during GnRH agonist treatment.[221] The use of Doppler ultrasonography or magnetic resonance imaging offers greater accuracy of evaluation. However, the incidence of leiomyosarcoma, even in patients with “rapidly growing leiomyomas,” is very low (less than 0.5%) and almost unheard of in premenopausal women.[157] In premenopausal women, a conservative approach is warranted.

Escape of suppression can result in an unexpected pregnancy. No adverse effects of fetal exposure to GnRH agonists have been reported, even when exposure has persisted throughout the early weeks of pregnancy.[222]

GnRH Agonists and Steroid Add-Back

Treatment with a GnRH agonist with steroid add-back has been explored to permit longterm therapy without bone loss.[211] Two strategies have been employed: simultaneous agonist and steroid add-back treatment or a sequential regimen in which the agonist is used alone for 3 months, followed by the combination of the agonist and steroid add-back. This long-term treatment is attractive for women who are perimenopausal, perhaps avoiding surgery. In addition, long-term treatment would be useful for women with coagulopathies, and in women with medical problems who need to postpone surgery.

Simultaneous treatment with agonist and medroxyprogesterone acetate (20 mg daily) or norethindrone (10 mg daily) effectively reduced hot flushing but was less effective (consistent with a major supportive role for progestins in myomas) in reducing uterine volume.[211,223] A sequential program, adding a traditional postmenopausal hormone

regimen (0.625 mg conjugated estrogens on days 1-25 and 10 mg medroxyprogesterone acetate on days 16-25) effectively reduced uterine volume and maintained the reduced volume for 2 years (and avoided any loss in bone density)[211] A daily 2.5 mg dose of tibolone also prevents bone loss and inhibits vasomotor symptoms without reducing the therapeutic efficacy of GnRH agonist treatment.[224] The addition of raloxifene to GnRH agonist treatment appeared to produce a greater reduction in leiomyoma size,[225] but the effect was not sufficiently different to be of clinical significance. Raloxifene treatment by itself, even in a large dose, failed to reduce leiomyoma size in premenopausal women, although in postmenopausal women, raloxifene produced a 30% to 40% reduction in size after 1 year.[226,227] Treatment with raloxifene, alendronate, or tibolone prevents the bone loss associated with agonist therapy, but only tibolone also prevents hot flushing.[224,228,229] and [230]

We recommend 1 month of GnRH agonist treatment followed by agonist treatment combined with a daily, continuous add-back of estrogen and progestin using one of the available postmenopausal daily regimens. In view of the sensitivity of leiomyoma tissue to progestational agents, it makes sense to keep the dose of progestin relatively low. Preoperative GnRH agonist treatment is not indicated in every patient with uterine leiomyomas. The best candidates for treatment are women with bleeding and anemia to allow time for a response to iron supplementation and when the surgeon's clinical judgment suggests that a reduction in size may influence the choice of technique (e.g., laparoscopic or vaginal hysterectomy instead of laparotomy).

Treatment with a GnRH Antagonist

GnRH antagonist treatment can suppress pituitary-gonadal function without the initial stimulatory (flare) response observed with GnRH agonists. Results with depot Cetrorelix

preoperative treatment of uterine fibroids are similar to those with GnRH agonist treatment; however, the response is faster (a maximal reduction in size within 14 days), probably because there is no initial flare response.[231,232]

Treatment with Mifepristone

Mifepristone, the progestin antagonist, effectively reduces the size of uterine leiomyomas and produces amenorrhea in most patients. The initial study was relatively short-term (12 weeks), and fibroid shrinkage was observed with doses of 25 and 50 mg daily.[181] A lower dosage is effective without the high rate of hot flushing observed at higher doses. In a 6-month study, a dose of 5 mg mifepristone daily was associated with a 48% reduction in uterine volume, a decrease in pressure and pain, an increase in hemoglobin levels, and a nonsignificant increase in hot flushing.[233] A similar reduction in uterine volume was observed in a 3-month study with the 5 mg dose, also with improvements in pain and bleeding.[234] However, long-term mifepristone treatment can result in endometrial hyperplasia, a consequence of the antiprogestin action of the drug. This endometrial effect makes mifepristone an unacceptable choice for on-going treatment of leiomyomas until large clinical trials are performed to establish its safety. Short-term treatment prior to surgery is appropriate. Asoprisnil, a progesterone receptor modulator, has also successfully reduced uterine volume and bleeding.[182] It is necessary to be cautious regarding the use of progesterone receptor modulators, as with progesterone antagonists, until endometrial safety is established.

Treatment with the Levonorgestrel-releasing Intrauterine System

When uterine enlargement because of leiomyomas is no greater than the size of a 12-week pregnancy, the

insertion of a levonorgestrel-releasing intrauterine system is followed by a decrease in uterine size in many but not all patients and a dramatic reduction in menstrual blood loss, with 40% of patients achieving amenorrhea.[235,236] and [237] The contraceptive efficacy is not diminished, but expulsion rates are higher. This method of treatment is not recommended when distortion of the uterine cavity is evident on examination with ultrasonography. The beneficial effect of locally applied levonorgestrel is unexplained, contrasting with the studies that indicate growth promotion of myomas by progestins.

Treatment with Uterine Artery Embolization

Uterine artery embolization effectively reduces bleeding, pain, and fibroid size.[238,239,240] and [241] In a procedure under local anesthesia that takes about one hour, a catheter is advanced from the femoral artery to the uterine arteries to allow direct injection of polyvinyl particles or gelatin microspheres that occlude the blood flow. Myomas undergo necrosis in response to the transient ischemia, but normal tissue generates fibrinolysis and survives. The procedure is not recommended for large fibroids. After 5 years, recurrence of symptoms is about 10% to 25%. Most patients experience pain, nausea, and low-grade fever with a very high white blood count for 1 to 2 days following the procedure. In addition, serious complications occur, including complication-related hysterectomy, amenorrhea, premature menopause, septicemia from uterine infection, bowel obstruction, and pulmonary embolus. Several deaths have been reported, giving a rate comparable to that with hysterectomy. A significant number of patients with larger myomas acquire intra-abdominal adhesions after the procedure.[242] The general recommendation is that embolization should not be performed

in women who desire to retain their fertility. However, a substantial number of completed pregnancies have been reported after the procedure[243,244]; nevertheless, the fertility rates and the complication rates after pregnancy is achieved are not known with certainty. A randomized comparison with myomectomy indicated a higher rate of infertility and miscarriages after embolization.[245]

Treatment with Ultrasound

A magnetic resonance mapping system for heat can be used to visualize myomas and direct high-energy ultrasound to destroy myomas.[246,247] The temperature achieved produces instant necrosis within a limited volume of tissue, and, therefore, the method requires multiple treatments over several hours. Thermal injury to skin and normal tissues are potential side effects. Overall safety and long-term efficacy remain to be established; the early pregnancy experience in 51 women documented a 41% live birth rate and a 28% miscarriage rate.[248]

Transient uterine ischemia can be produced by placing vaginal clamps in the vaginal, fornices, guided by ultrasonography, to compress the uterine arteries against the cervix for about 6 hours. Short-term studies have demonstrated efficacy comparable to embolization, but long-term follow-up data are not yet available.[249]

All references are available online at: http://www.clinicalgynendoandinfertility.com

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