The Pathogenesis of Adenomyosis

2

Fang Xiao

Adenomyosis refers to an estrogen-dependent gynecological benign disease in which the endometrial glands and stroma invade the myometrium, resulting in localized or diffuse adenomyosis. It often combines with uterine fibroids, endometrial polyps, endometriosis, etc. The incidence of adenomyosis ranges from 1 to 70% [1] due to the variation of diagnostic criteria. The main clinical manifestations are progressive dysmenorrhea, chronic pelvic pain, menorrhagia, and infertility. With the improvement of magnetic resonance imaging (MRI), transvaginal ultrasound imaging, and other examination techniques, the detection rate of adenomyosis is also increasing [2, 3]. Although it has been nearly 100 years since the name of the disease is mentioned, the true pathogenesis of adenomyosis is still a mystery. This chapter will discuss and focus on the latest knowledge in the pathogenesis of adenomyosis.

2.1 Endometrial Invasion (Into the Myometrium) Theory

The theory of endometrial invasion into the myometrium is also called the basal endometrium invagination theory. Due to the lack of submucosa, the endometrium and the myometrial layers are in direct contact, and the barrier is relatively weak. The junction between the endometrium and the myometrium is called the endometrial-myometrial junctional zone (JZ), and some scholars call it the endometrial myometrial interface (EMI) [4]. Studies have shown that the thickness of JZ in patients with adenomyosis increased significantly. At present, most scholars believe that the thickness of JZ has to exceed 12 mm to diagnose adenomyosis [1, 5].

F. Xiao (✉)
Department of Gynecology and Obstetrics, Third Xiangya Hospital of Central South University, Changsha, Hunan, China

M. Xue et al. (eds.), *Adenomyosis*, https://doi.org/10.1007/978-981-33-4095-4_2

2.1.1 Endometrial Basal Layer Injury

Research studies found that women with a history of uterine or uterine cavity surgery, such as cesarean section, multiple births, multiple abortions, hysteroscopic myomectomy, diagnostic curettage, intrauterine device placement, etc., had almost 1.5 times the incidence of adenomyosis than those without such operations [5, 6]. Therefore these surgical procedures are high-risk factors for the formation of adenomyosis. The surgical operation of the uterus and uterine cavity can cause endometrium and EMI damage. The tissue injury and repair (TIAR) mechanism is activated, and the functional layer of the endometrium is recessed inward through the EMI; it gradually invades the myometrium and leads to the formation of adenomyosis [7]. In pregnancy, it is believed that the proliferation of uterine myocytes, the expansion of the uterine wall, and the involution of the postpartum uterus may damage the endometrium-myometrium JZ, resulting in the invasion of the endometrium into the myometrium.

On the other hand, obstetric complications, such as placental implantation, placental adhesions, uncoordinated uterine contractions, prolonged labor, cesarean section, and gynecological uterine surgery, such as laparoscopic myomectomy, hysteroscopic myomectomy, and endometrial ablation, can directly damage the EMI. Any uterine cavity surgery, like dilatation and curettage (D&C), polypectomy, and surgical abortion, may directly cause gaps or channels between the endometrium and the myometrium, resulting in the invasion of endometrium directly into the myometrium. Besides, adenomyosis is often accompanied by reproductive tract malformations, such as uterine malformation and vaginal septum. These two conditions lead to obstruction of menstrual flow and increase intrauterine pressure, and then it may cause the endometrium to invade into the muscle layer. Finally, congenital developmental abnormalities of the uterus may also have loose connections or gaps between the endometrium and myometrium, which can cause the endometrium to invade the myometrium and cause adenomyosis directly.

2.1.2 Enhancement of Endometrial Invasion and Adhesion Ability Theory

Matrix metalloproteinases (MMP) are a class of enzymes that degrade the extracellular matrix. It is suggested that the overexpression of endometrial metalloproteinase can enhance the endometrial tissue's invasiveness. Li et al. 2006 [8] found that there were high expressions of MMP 2, 3, and 9 in eutopic endometrial and ectopic endometrial tissues in patients with adenomyosis. Their expression levels were significantly higher than those in patients without adenomyosis, and MMP inhibitory enzymes −1 and 2– in endometrial tissue of adenomyosis were significantly lower than the endometrium in those without adenomyosis. Under the influence of various environmental or hormonal factors, the endometrial tissue itself may degrade the extracellular matrix components, including the basement membrane; thereby it can

destroy the natural barrier that prevents the endometrium from invading the myometrium, resulting in the formation of adenomyosis.

Cell adhesion molecules are membrane glycoprotein molecules that mediate the interaction between cells or between cells and substrates. They are involved in cell signaling and play an important role in the growth and differentiation of cells, cell migration, tumor metastasis, and wound healing. The expression of cell adhesion molecule-1 in the endometrium and the ectopic endometrium of the uterus with adenomyosis is significantly higher than that without adenomyosis. In addition to cell adhesion molecules, integrin β3 also plays a vital role in the process of cell invasion and adhesion. It not only connects cells and extracellular matrix but also regulates the occurrence of this process through signal transmission and reception. Xiao et al. 2013 [9] also found that the expression level of integrin β3 in the uterine endometrium is lower than that of the ectopic endometrium and uterine endometrium of the uterus without adenomyosis. The integrin expression level of the endometrium in a uterus with adenomyosis is reduced, which can destroy endometrial stability and decrease the basal adhesion of the basal layer, making it easier for the endometrium to invade the myometrium and eventually form the adenomyosis.

2.1.3 Abnormal JZ Between Endometrium and Myometrium

The endometrium-myometrium JZ was first proposed by Hricak H and others in 1983 when they initially used MRI to examine the female reproductive system [10]. Endometrium-myometrium JZ is a new uterine JZ which is different from the JZ in other tissues in the human body, and it lacks an identifiable protective layer – the submucosa. That is, there is no "intermediate buffer zone" between the endometrium and the myometrium, so they are in direct contact. JZ is a special functional unit, which has the functions of regulating trophoblast invasion, providing nutrition, facilitating sperm transport, menstrual hemostasis, and local immunity and is of great significance for maintaining the physiological function of the uterus.

In recent years, with the in-depth study of the pathogenesis of adenomyosis, it has been confirmed that the interface between the endometrium and myometrium is unique, regardless of whether it is structural, functional, histopathological, and by imaging. It plays a key role in the pathogenesis of adenomyosis. The uterine JZ is an estrogen-progestin-dependent transformation zone and shows periodic changes [11]. The embryonic origin of the subendometrial muscle layer is the same as the endometrium, and the anatomical structure is directly connected to the endometrium, so it is speculated that its biological function may be related to the endometrium. In adenomyosis, probably due to the overproduction of estrogen, it may promote oxytocin-mediated uterine activity, causing endometrial hyperplasia and peristaltic enhancement, thereby increasing mechanical stress and tension, chronic long-term abnormalities, and excessive uterine creeping movement may cause micro damage to JZ and activate self-renewing tissue damage repair mechanisms [7, 12, 13].

The activation of the JZ TIAR mechanism promotes the local production of interleukin-1 (IL-1) and induces the activation of COX-2, leading to the production of prostaglandin E2. The steroidogenic acute regulatory protein and cytochrome P450 aromatase are subsequently activated; androgen is formed and aromatized into estrogen, resulting in a high estrogen status in the endometrium. Estrogen acts through estrogen receptors (ER-β) to promote proliferation and healing. During the normal healing process in the normal endometrium, the increase in estrogen secretion gradually stops. However, in the uterus with adenomyosis, the high levels of estrogen stimulate oxytocin-mediated peristalsis through the ER-α receptor, inhibiting the healing process. A positive feedback mechanism is then generated; through which the chronic peristalsis in JZ promotes repeated cycles of injury and repair, resulting in the continuous rupture or damage of muscle fibers in the muscle wall. As each cycle gets worsen, the basal layer of the endometrium enters the myometrium, which eventually leads to the formation of adenomyosis [12, 13]. Also, a large number of studies have shown that abnormal amplitude, frequency, and direction of JZ contraction, especially active peristalsis or sluggish peristalsis, may be the cause of adenomyosis, endometriosis, and infertility [14]. Liu et al. 2013 [15] speculated that the up-regulation of NO (nitric oxide) in smooth muscle cells in the endometrium-myometrium JZ of patients with adenomyosis might be related to the abnormal contractile function of this site. Normal women's uterine contraction during menstrual period is from the fundus of the uterus to the cervix. This contraction helps hemostasis during the menstrual period and helps the exfoliated endometrial tissues exit the uterine cavity. Among patients with adenomyosis, this contraction movement during menstrual period is significantly reduced or not active at all. This abnormal movement makes menstrual blood more easily refluxed into the abdominal cavity, causing endometriosis, which can also explain the high association of adenomyosis and pelvic endometriosis.

2.1.4 The Proliferation of Ectopic Endometrial Cells and Unbalanced Apoptosis: [16]

Uterine adenomyosis is considered to be a proliferative disease. Therefore, the endometrial tissue proliferation ability and the inhibition of apoptosis have attracted much attention. Telomerase is a unique ribonucleoprotein. Its activity continuously adds telomere deoxyribonucleic acid (DNA)-repeating sequence at the end of chromosomes to prevent the loss of telomeres and make cells proliferate and immortalized. It is suggested that the ectopic endometrial cells of patients with adenomyosis under the activation of telomerase can enhance the proliferation ability of ectopic endometrial cells invading the uterine myometrium, promoting the proliferation of surrounding cells and participating in the formation of adenomyosis [16]. In adenomyosis, the expression level of telomerase is significantly higher than other tissues, and the expression level of telomerase in the lesions of severe diffuse adenomyosis is significantly higher than that of a local or mild adenomyosis.

Ki67 is a cell proliferation marker that is expressed only in the nucleus of circulating proliferating cells, not at resting cells. The expression of Ki67 in the ectopic endometrial glandular epithelium of adenomyosis was found to be significantly higher than the control group. It was also higher than that of the normal uterine endometrium in the secretory phase [17]. Besides, Xue et al. 2003 showed that markers related to the proliferation of adenomyosis, such as proliferating cell nuclear antigen, cyclin-1, and cyclin-dependent kinase transfer growth factor and epidermal growth factor, were up-regulated. It suggested that after the endometrial cells invaded the myometrium, their proliferative capacity was enhanced and was important in the development of adenomyosis.

B-cell lymphoma-2 gene (Bcl-2) is called the apoptotic suppressor gene, and survivin protein is an apoptosis-inhibiting protein. Survivin gene is the strongest apoptosis-inhibiting gene currently found. It can exert anti-apoptosis by directly inhibiting the apoptosis pathway. High expression of survivin can promote cell proliferation, participate in the regulation of cell mitosis, and resist various apoptosis-inducing factors, such as interleukin (interleukin, IL) -3, tumor necrosis factor (TNF) -α, chemotherapy, and radiation. The ectopic endometrial tissue of patients with adenomyosis showed a high expression of Bcl-2 and survivin [18], and the two showed a positive correlation. At the same time, Bax has an antagonistic role in the formation and progression of adenomyosis. Its level is down-regulated in the ectopic endometrial tissue in adenomyosis; thus, this leads to enhanced apoptosis inhibition, reduced apoptosis, and increased cell proliferation, participating in the pathogenesis of adenomyosis. Clinical applications of mifepristone and gonadotropin-releasing hormone analog (GnRHa) treatment can reduce levels of Bcl-2 and survivin expression.

Osteopontin (OPN) is a matricellular protein implicated in the pathogenesis of a variety of diseases. It also has an apoptosis-inhibiting effect. If OPN is lacking, the number of cell apoptosis increases significantly. The expression of OPN in the ectopic endometrium of patients with adenomyosis [19] is significantly higher than that of the control group, resulting in the enhanced anti-apoptosis ability of ectopic endometrial cells; thereby they are more likely to survive, and grow. Therefore abnormal apoptosis may be an important factor in the development of adenomyosis.

2.2 Epithelial to Mesenchymal Cells Transformation

Migration and invasion factors are considered to be the key factors for the progression of adenomyosis. Epithelial-mesenchymal transition (EMT) is when epithelial cells lose their cell polarity and become mesenchymal cells to improve their ability to migrate, invade, and resist apoptosis [20, 21]. There are many EMT inducer involved in the process of EMT, and they are E-cadherin, CK7 (an epithelial cell marker), and transforming growth factor-β [22–24]. Chen et al. 2010 [25] found that changes in EMT markers were related to serum E2 levels. Estrogen induced EMT of ER-positive endometrial cells, causing them to migrate and invade. Therefore

induced EMT of endometrial epithelial cells contributes to the development of adenomyosis.

From the embryonic development, the mesoderm mesenchyme of the urogenital system transforms into the epithelium to form the endometrium in a specific microenvironment. Mesenchymal epithelialization is not only the direct basis of endometriosis stem cell theory but can also be the direct basis of adenomyosis via EMT. The process of EMT involves a series of important protein molecules and signal transduction pathways, as discussed above. It is currently a hot research topic for endometriosis and adenomyosis. Since the role of the EMT process in the pathogenesis of adenomyosis is still unclear, further research is important.

2.3 Differentiation of Embryonic Multipotent Müllerian Duct Remnant

The Müllerian duct is the original embryonic structure that develops into the female uterus, fallopian tube, and upper vagina during fetal life [26]. The Müllerian duct is composed of surface epithelium and urogenital crest mesenchyme and can differentiate into endometrial glands and stroma [27]. It is speculated that the differentiation of the embryonic pluripotent Müllerian duct remnant in the uterine wall of adult females may lead to the establishment of ectopic endometrial tissue, forming the adenomyosis.

Adenomyosis has the typical characteristics of smooth muscle hyperplasia and fibrosis. In contrast, deep infiltrating endometriotic nodules are also considered to be a possible result of Müllerian duct differentiation, such as proposed by Dunnez et al. 1996 [28]. Adenomyoma and vaginal and rectal septum endometriosis are considered to be extrauterine adenomyosis lesions. They are similar to adenomyosis lesions in pathology and clinical characteristics, and it seems to support this pathogenesis theory.

It is reported in the literature that some patients with congenital genital tract malformation, mayer-rokitansky-küster-hauser (MRKH) syndrome, with their primordial uterus without a functional endometrium, can develop adenomyosis [29]. The finding in these patients did not support the theory of endometrial damage and invasion to explain the occurrence of adenomyosis. Therefore, some scholars proposed that adenomyosis could be developed from the remnant of the Müllerian ducts in the muscular layer.

2.4 Endometrial Stem Cell Theory

Gargett, 2007 [30], had found that there were very few epithelial and mesenchymal stem cells in the endometrium of adult women. Under normal circumstances, endometrial stem cells are at rest. Endometrial stem cells are located in the basal layer of the endometrium. After abnormal shedding, they can enter the pelvis through the fallopian tubes to form endometriosis. If endometrial stem cells migrate abnormally and invade the myometrium, then they form the adenomyosis. Chan et al. 2004 [31]

had identified active epithelial and mesenchymal stem cell populations from the basal layer of the endometrium and stroma from hysterectomy specimens. Kato, 2012 [32], confirmed that these stem cells were located among the basal cells of the endometrium and were responsible for the periodic repair of the endometrium after menstruation. In a normal endometrium, the presence of endometrial basal cells is a key factor for regeneration and renewal. However, it can also grow indefinitely and can even extend beyond the endometrium. Some scholars have found that under the stimulation of tissue damage, endometrial stem cells were activated, and proliferation, regeneration, and differentiation occurred, which could promote the invasion as ectopic endometrium. This phenomenon can also be explained by the "stem cell niche hypothesis"; that is to say, the stem cells are located in the niche, and under normal circumstances, the niche can prevent stem cells from proliferating, differentiating, and apoptotic, leaving them in a quiescent state. When the niche is destroyed, stem cells will proliferate, regenerate, and differentiate to varying degrees [33]. The activation of stem cells does not only promote tissue repair, and excessive activation can even lead to the occurrence of proliferative diseases, such as tumors. Therefore, the abnormal activation of endometrial stem cells may be one of the pathogenesis of adenomyosis.

In 2004, Taylor [34] found a donor-derived endometrial gland and mesenchymal cells in the endometrium of four female recipients who received HLA-unmatched bone marrow transplantation, which suggested that bone marrow stem cells could differentiate into endometrial cells. In mice and in vitro experiments [35–37], it also confirmed that bone marrow stem cells could indeed differentiate into mature endometrial cells. From these studies, it can be speculated that bone marrow stem cells may play a role in the development of adenomyosis.

Using electron microscopy, Ibrahim et al. 2015 [38] described another population of stem cells. These stem cells were located in the epithelial glands of the basal endometrium. Because their cytoplasm was electronically glowing, they were called pale cells. In the JZ of patients with adenomyosis, these stem cells were significantly less connected to the surrounding epithelial cells (loss of desmosome junctions). They presented more feet-like structures than in disease-free women. Therefore, it is suggested that in adenomyosis, pale cells might be displaced through the basement membrane, gain movement characteristics, and migrate to the interstitium and then transfer into the myometrium, where they formed new adenomyosis. Pale cell migration requires multiple signaling pathways and biological factors to co-regulate; the cell-to-cell tight connection junction is disintegrated, the expression of adhesion molecules is down-regulated; the basement membrane microrupture and the up-regulation of MMP may be involved in the migration process [39].

2.5 Abnormal Uterine Nerve

Under normal circumstances, the uterine nerves consist of sympathetic and parasympathetic nerves from S2 to S4. These nerve fibers pass through the uterosacral ligament to the paracervical tissue before entering the uterus and form the

Frankenhauser plexus at the posterolateral part of the cervix; the other part of innervation is introduced through the other ligaments of the uterus and the peritubal nerve fibers. Most of the uterine nerves are distributed at the interface of the endometrium and myometrium, that is, the nerve distribution in the inner 1/3 of the uterus is greater than the nerve distribution in the outer 2/3, and the nerve can reach the endometrial glandular epithelium and spiral arterioles. Due to the cyclical shrinkage of the endometrial function layer, the nerve fibers on this layer would have periodic denervation and nerve regeneration.

When various factors, such as pregnancy, miscarriage, childbirth, surgery, and infection, cause damage to the uterine nerves, the damaged nerves can release various neurotransmitters and neural mediators and can attract macrophages and release prostaglandins to the injured area, resulting in the accumulation of inflammatory factors and growth factors. Patients with adenomyosis who show clinical pain symptoms have a large number of abnormal nerve fiber hyperplasias at the interface of the endometrium and myometrium [40]. The abnormal nerve fibers are directly related to the patient's pain and thus involved in the mechanism of pain. These nerve injury and regeneration activities at the JZ may account for the abnormal contraction of the muscular layer and the increase of peristalsis in the JZ in adenomyosis, thus causing other symptoms of adenomyosis.

After treatment with hormonal drugs, such as oral contraceptives, progesterone or levonorgestrel intrauterine system (LNG-IUS), and GnRHa for adenomyosis, in addition to the decrease in nerve fibers in the uterine endometrium and myometrium, the expression of nerve growth factor (NGF) and its receptors also decreased. Therefore the symptoms of dysmenorrhea improved [41]. On the other hand, estrogen can increase the expression level of NGF in adenomyosis, thus increasing the severity of adenomyosis. Locally released NGF can stimulate the release of inflammatory mediators, such as serotonin, histamine, and tumor necrosis factor, by stimulating mast cell degranulation, thereby inducing inflammation. It has been shown that increased expression levels of NGF and its receptors in the ectopic endometrium of adenomyosis can increase the sensitivity of uterine nerves, aggravate the inflammatory response, and cause abnormal growth of local nerve fiber proliferation in adenomyosis. NGF can, in turn, stimulate the expression and release of focal neuropeptides to excite the neuron ends, to maintain the survival and development of sensory neurons, and participate in pain regulation, thus increasing pain or hyperalgesia in adenomyosis. Therefore, abnormal growth of uterine nerve fibers not only participates in the pathogenesis of adenomyosis but also participates in the mechanism of dysmenorrhea in adenomyosis.

2.6 The Theory of Increased Angiogenesis

Adenomyosis is often associated with endometrial proliferation, where the growth of endometrial cells requires blood vessels to provide nutrition and eliminate metabolic waste. When hysteroscopy is performed on patients with adenomyosis and about 50% of patients have abnormal vascularization in the endometrium [42].

Vascular endothelial growth factor (VEGF) is an important factor that promotes angiogenesis, and its expression level in tissues can reflect tissue angiogenesis. Mu et al. 2014 [43] had shown that in patients with adenomyosis, the number and activity of VEGF in the intima and serum were significantly higher than in normal people, and the level of VEGF in the serum of patients after surgical treatment was reduced compared with before treatment. Compared with a control group, the microvessel density (MVD) in the ectopic endometrium of patients with adenomyosis was significantly increased. Therefore, it can be speculated that the formation of new blood vessels affects the occurrence of adenomyosis, and it may also be the cause of menorrhagia. Liu et al. 2016 [44] found that platelets accumulated in patients with adenomyosis activated the TGF-β1/Smad3 signaling pathway, inducing EMT, fibroblast to myofibroblast transdifferentiation, smooth muscle cell proliferation, and fibrosis and then participated in the development of adenomyosis. Antiplatelet therapy is a potential treatment for adenomyosis, because angiogenesis is a necessary condition for the endometrium to invade the myometrium and continue to grow. The formation of blood vessels not only supplies the nutrients required for the growth of the ectopic endometrium but also may be an important way for the transfer of endometrial cells from the normal intima to the myometrium. In terms of molecular biology, adenomyosis VEGF with its angiogenic activity at EMI is significantly enhanced but does not change with the proliferation and secretion phase; VEGF not only accelerates the uterine endometrial gland invasion but also promotes ectopic endothelial cell proliferation. All the above findings support the theory of increased angiogenesis as the pathogenesis of adenomyosis.

2.7 High Estrogen and High Prolactin Theory

Patients with endometriosis have abnormal hypothalamic-pituitary-ovarian axis function and often have corpus luteum dysfunction. Both adenomyosis and endometriosis are estrogen-dependent diseases. Anti-estrogen drugs are used to treat endometriosis as well as adenomyosis. Obviously, androgen is the root cause of adenomyosis.

Aromatase, also called estrogen synthetase or estrogen synthase, is an enzyme responsible for a key step in the biosynthesis of estrogens. Patients with adenomyosis have high expression of aromatase in the endometrium, but in the endometrium of women without adenomyosis, its expression is difficult to be detected. In addition to aromatase, the activity of 17β-hydroxysteroid dehydrogenase type 2 that decomposes estrogen decreases, which ultimately leads to a high-estrogen environment in adenomyosis. After the estrogen combines with the corticotropin-releasing hormone and urocortin in the adenomyosis, these hormones stimulate cyclooxygenase and its isoenzyme C0X-2, to make an increase in PGE2. Both COX-2 and PGE2 are powerful inducers of aromatase activity and exert a positive feedback mechanism to aggravate dysmenorrhea. It is suggested that estrogen can indirectly promote uterine contraction and coordinate with oxytocin to regulate uterine contractility. Therefore the locally increased estrogen in adenomyosis can regulate uterine

contractility, resulting in abnormal contractions and increased peristalsis of the subendometrial myometrium. That is why the use of aromatase inhibitors to treat adenomyosis can reduce the uterine volume of adenomyosis and dysmenorrhea. It has the same effect as gestrinone. This finding supports the increase of estrogen levels in patients with adenomyosis; especially the local high estrogen environment is the main cause of adenomyosis [45].

In addition to estrogen, hyperprolactinemia can also directly induce adenomyosis. Prolactin can promote the binding of estrogen to its receptors and increase the effective biological activity of estrogen.

2.8 Immune Factors

Recent studies have found that the onset of adenomyosis may be related to abnormalities in humoral immunity, cellular immunity, and cytokines [45, 46]. In terms of humoral immunity, patients with adenomyosis may have abnormalities in immunoglobulin and complement. Abnormal cellular immunity of adenomyosis mainly includes natural killer cell reduction, macrophage increase, and helper T cell 1/helper T cell 2 (Th1/Th2) cell imbalance. The reduction of the killing capacity of natural killer cells is beneficial to the implantation of endometrial cells in the myometrium. After activation, macrophages can secrete a variety of inflammatory factors, such as interleukin (IL) -1β and TNF-α, and increase the oxidation reaction, resulting in increased oxygen consumption, resulting in oxidative stress, which in turn has a toxic effect on the embryo, affecting embryo implantation, and reduce the pregnancy rate. After treatment with GnRHa, the number of macrophage infiltration in the endometrium and uterine myometrium of patients with adenomyosis was significantly reduced and accompanied by a significant reduction of monocyte chemoattractant protein, (MCP -1), thereby increasing the clinical pregnancy rate of patients with adenomyosis, which may explain the immunological mechanism of adenomyosis with infertility [47].

2.9 Genes and Genetic Factors

Genetic factors may be involved in the occurrence of adenomyosis, but this process requires the joint action of external factors. At present, no single gene has been found that directly leads to the formation of adenomyosis. Studies have shown that related gene changes may be involved in the occurrence and development of adenomyosis, which is mainly achieved through apoptosis regulation, proliferation regulation, and mutation and inactivation of tumor suppressor genes [48, 49]. In adenomyosis lesions, the apoptosis control gene Bcl-2 shows continuous high expression without periodic changes, and the local ectopic endometrial tissue and the influence of the internal environment can up-regulate its expression. The

existence of Bcl-2 protein prevents ectopic endometrial cells of the adenomyosis from expressing and triggering cell surface receptors related to apoptosis, which reduces apoptosis and increases cell proliferation. It, in turn, promotes the occurrence and development of adenomyosis. At the same time, the high expression of survivin protein in the ectopic endometrial gland of adenomyosis significantly enhances the anti-apoptosis ability of ectopic endometrial cells. It also slows down apoptosis, thereby prolonging the survival time of endometrial cells invading the myometrium. Therefore, the high level of survivin protein may also be one of the causes of adenomyosis. In the uterine endometrium and ectopic endometrium of adenomyosis patients, the expression intensity of Fas/FasL is significantly weakened, and the imbalance causes ectopic endometrial cells to survive and escape the attacks from the body's immune system, which is an important mechanism for the pathogenesis of adenomyosis [50]. Adenomyosis patients have overexpression of many other genes closely related to cell proliferation, which promotes the growth of the intima, which leads to the occurrence and development of adenomyosis. These related genes mainly include proliferating cell nuclear antigen, proto-oncogene c-myc, proto-oncogene TrkB, and pituitary tumor transformation genes. The expression of these genes can promote cell proliferation, regulate the cell cycle, inhibit cell secretion, regulate apoptosis, promote cell migration, ectopic attachment, angiogenesis, and invasion, and ultimately promote the occurrence of adenomyosis [51]. Also, the inactivation of tumor suppressor genes and the occurrence of adenomyosis are closely related to the occurrence and development of adenomyosis [49].

In addition to epigenetic involvement in the pathogenesis of adenomyosis, a variety of oncogenes or tumor suppressor genes, metabolic genes, and cell signal regulation genes (or factors) have been found in recent years that may be related to the onset of adenomyosis. Because these oncogenes or metabolic genes can proliferate and invade, adenomyosis with these genes also has the characteristics of invasion and metastasis; therefore, most of the current researches use the discovered oncogenes to study adenomyosis.

In endometriosis, galectin-33 can up-regulate the expression of NGF. High expression of galectin-33 was also present in adenomyosis, and the induced high expression of NGF can promote ectopic endometrial proliferation and is related to the patient's pain. Because endometriosis has the biological behavior characteristics of malignant tumors, it is believed that some malignant biological behavior characteristics of endometriosis also exist in adenomyosis. For example, the onset of endometriosis involves mitogen-activated protein kinase (MAPK), Akt/protein kinase B (PKB), NF-κB, and phosphatidylinositol 3-kinase (Phosphatidylinositol3-kinase, PI3K), and other cell signaling pathways [52]. The pathogenesis of adenomyosis also involves these cell signaling pathways. However, patients with endometriosis have a family aggregation phenomenon, and adenomyosis is not yet clear whether this phenomenon exists. Therefore, the genetic research involving adenomyosis still needs much work.

2.10 Inflammation or Infection Factors

Most researchers have recognized the role of inflammation in the pathogenesis of endometriosis. Not surprisingly, many studies of ectopic endometrium and endometriosis showed a significant increase in IL-6 and other cytokines [53, 54]. Similarly, it is believed that inflammation or infection factors would also exist in adenomyosis. Therefore in patients with adenomyosis, whether it is serum or peritoneal fluid or endometrial tissue, abnormally elevated levels of IL-6 had been detected [55]. This finding showed that inflammation could be a causative factor of adenomyosis. The latest research also found that a variety of pathogenic microorganisms was detected in the vagina and cervical secretions of patients with adenomyosis, which was significantly higher than that of normal women [56]. This clinical finding, therefore, has a practical significance for the prevention and treatment of clinical adenomyosis.

2.11 Others

The incidence of adenomyosis has increased significantly in recent years. In addition to changes in people's lifestyles, environmental factors cannot be ignored. Stem cell receptors can be found in the endometrium of adenomyosis, especially at the junction of the endometrium and myometrium, namely JZ. Therefore, it is speculated that stem cells may also play a role in the pathogenesis of adenomyosis. Also, the bone marrow stem cell theory, biochemical theory, lymphatic dissemination, oxidative stress, and free radicals are considered to be involved in the pathogenesis of endometriosis. However, whether these phenomena or theories can be applied to the pathogenesis of adenomyosis still need much research to confirm.

In short, the pathogenesis of adenomyosis remains unclear. Although the above studies found that adenomyosis may be related to various factors, these factors do not act alone but a combination of various factors. Under the influence of various factors, such as uterine trauma, inflammation, or the patient's immune deficiency or possible inheritance, and migration and invasion capabilities of the endometrial cells, the endometrium then invades the myometrium, resulting in adenomyosis. It is believed that the pathogenesis of adenomyosis will be clarified and elucidated after more intensive researches in the future.

References

1. Graziano A, et al. Diagnostic findings in adenomyosis: a pictorial review on the major concerns. Eur Rev Med Pharmacol Sci. 2015;19(7):1146–54.
2. Tellum T, et al. Development of a clinical prediction model for diagnosing adenomyosis. Fertil Steril. 2018;110(5):957–964.e3.
3. Karamanidis D, et al. OC01: Transvaginal ultrasonography compared with magnetic resonance imaging for the diagnosis of adenomyosis. Ultrasound Obstet Gynecol. 2018;52(4):555.

4. Scoutt LM, et al. Junctional zone of the uterus: correlation of MR imaging and histologic examination of hysterectomy specimens. Radiology. 1991;179(2):403–7.
5. Agostinho L, et al. MRI for adenomyosis: a pictorial review. Insights Imaging. 2017;8(6):549–56.
6. Riggs JC, et al. Cesarean section as a risk factor for the development of adenomyosis uteri. J Reprod Med. 2014;59(1–2):20–4.
7. Leyendecker G, Wildt L. A new concept of endometriosis and adenomyosis: tissue injury and repair (TIAR). Horm Mol Biol Clin Invest. 2011;5(2):125–42.
8. Li T, Li Y-G, Pu D-M. Matrix metalloproteinase-2 and-9 expression correlated with angiogenesis in human adenomyosis. Gynecol Obstet Investig. 2006;62(4):229–35.
9. Xiao Y, et al. Expression of integrin β3 and osteopontin in the eutopic endometrium of adenomyosis during the implantation window. Eur J Obstet Gynecol Reprod Biol. 2013;170(2):419–22.
10. Hricak H, et al. Magnetic resonance imaging of the female pelvis: initial experience. Am J Roentgenol. 1983;141(6):1119–28.
11. Yifu S. There are some questions about uterine adenopathy. China Fam Plan Obstet Gynecol. 2018;10(1):3–6.
12. Leyendecker G, et al. Adenomyosis and endometriosis. Re-visiting their association and further insights into the mechanisms of auto-traumatisation. An MRI study. Arch Gynecol Obstet. 2015;291(4):917–32.
13. Shaked S, et al. Uterine peristalsis-induced stresses within the uterine wall may sprout adenomyosis. Biomech Model Mechanobiol. 2015;14(3):437–44.
14. Uduwela A, et al. Endometrial-myometrial interface: relationship to adenomyosis and changes in pregnancy. Obstet Gynecol Surv. 2000;55(6):390–400.
15. Liu Jingjing DH. Wangsha the expression and significance of nitric oxide in the smooth muscle cells in the endometrial-myoline junction region of uterine adenopathic patients. Chinese J Obstet Gynecol. 2013;48(007):504–7.
16. Kyo S, et al. Telomerase activity in gynecological tumors. Clin Cancer Res. 1996;2(12):2023–8.
17. Xue Q, Zhou Y, Liu Y. Expression of ki67 and CA_ (125) in Adenomyosis. J Pract Obstet Gynecol. 2003;5
18. Deng Y, Chunlan H, Xiaoling L. Expression of Survivin in Adenomyosis and its relationship with Bcl-2 and Bax protein. J Med Res. 2006;6
19. Xiao Y, et al. Expression of integrin beta3 and osteopontin in endometrium of patients with adenomyosis. Zhonghua Fu Chan Ke Za Zhi. 2009;44(5):354–8.
20. Polyak K, Weinberg RA. Transitions between epithelial and mesenchymal states: acquisition of malignant and stem cell traits. Nat Rev Cancer. 2009;9(4):265–73.
21. Acloque H, et al. Epithelial-mesenchymal transitions: the importance of changing cell state in development and disease. J Clin Invest. 2009;119(6):1438–49.
22. An M, et al. Interaction of macrophages and endometrial cells induces epithelial–mesenchymal transition-like processes in adenomyosis. Biol Reprod. 2017;96(1):46–57.
23. Khan KN, et al. Involvement of hepatocyte growth factor-induced epithelial-mesenchymal transition in human adenomyosis. Biol Reprod. 2015;92(2):35. 1-11
24. Oh SJ, et al. β-Catenin activation contributes to the pathogenesis of adenomyosis through epithelial–mesenchymal transition. J Pathol. 2013;231(2):210–22.
25. Chen YJ, et al. Oestrogen-induced epithelial–mesenchymal transition of endometrial epithelial cells contributes to the development of adenomyosis. J Pathol. 2010;222(3):261–70.
26. Sobel V, Zhu Y-S, Imperato-McGinley J. Fetal hormones and sexual differentiation. Obstet Gynecol Clin. 2004;31(4):837–56.
27. Spencer TE, et al. Comparative developmental biology of the mammalian uterus. Curr Top Dev Biol. 2005;68:85–122.
28. Donnez J, et al. Stereometric evaluation of peritoneal endometriosis and endometriotic nodules of the rectovaginal septum. Hum Reprod. 1996;11(1):224–8.

29. Enatsu A, et al. Adenomyosis in a patient with the Rokitansky-Kuster-Hauser syndrome. Fertil Steril. 2000;73(4):862–3.
30. Gargett CE. Uterine stem cells: what is the evidence? Hum Reprod Update. 2007;13(1):87–101.
31. Chan RW, Schwab KE, Gargett CE. Clonogenicity of human endometrial epithelial and stromal cells. Biol Reprod. 2004;70(6):1738–50.
32. Kato K. Stem cells in human normal endometrium and endometrial cancer cells: characterization of side population cells. Kaohsiung J Med Sci. 2012;28(2):63–71.
33. Long Wenjie GL, Fudi L, Xin S, Hui L. Stem cells and their relationship with adenomyosis*. Prog Mod Biomed. 2017;17(2):393–5.
34. Taylor HS. Endometrial cells derived from donor stem cells in bone marrow transplant recipients. JAMA. 2004;292(1):81–5.
35. Zhang W-B, et al. A study in vitro on differentiation of bone marrow mesenchymal stem cells into endometrial epithelial cells in mice. Eur J Obstet Gynecol Reprod Biol. 2012;160(2):185–90.
36. Morelli SS, Rameshwar P, Goldsmith LT. Experimental evidence for bone marrow as a source of nonhematopoietic endometrial stromal and epithelial compartment cells in a murine model. Biol Reprod. 2013;89(1):1–7.
37. Du H, Naqvi H, Taylor HS. Ischemia/reperfusion injury promotes and granulocyte-colony stimulating factor inhibits migration of bone marrow-derived stem cells to endometrium. Stem Cells Dev. 2012;21(18):3324–31.
38. Ibrahim MG, et al. Ultramicro-trauma in the endometrial-myometrial junctional zone and pale cell migration in adenomyosis. Fertil Steril. 2015;104(6):1475–1483.e3.
39. Zhang Yumin ZS. The study of pale cells and adenomyosis. Chin J Obstet Gynecol. 2018;1:39.
40. Zhang X, et al. Endometrial nerve fibers in women with endometriosis, adenomyosis, and uterine fibroids. Fertil Steril. 2009;92(5):1799–801.
41. Choi YS, et al. Effects of LNG-IUS on nerve growth factor and its receptors expression in patients with adenomyosis. Growth Factors. 2010;28(6):452–60.
42. Ota H, Tanaka T. Stromal vascularization in the endometrium during adenomyosis. Microsc Res Tech. 2003;60(4):445–9.
43. Mu Y, et al. Serum levels of vascular endothelial growth factor and cancer antigen 125 are related to the prognosis of adenomyosis patients after interventional therapy. Int J Clin Exp Med. 2015;8(6):9549.
44. Liu X, et al. Corroborating evidence for platelet-induced epithelial-mesenchymal transition and fibroblast-to-myofibroblast transdifferentiation in the development of adenomyosis. Hum Reprod. 2016;31(4):734–49.
45. Garcia L, Isaacson K. Adenomyosis: review of the literature. J Minim Invasive Gynecol. 2011;18(4):428–37.
46. Ota H, et al. Is adenomyosis an immune disease? Hum Reprod Update. 1998;4(4):360–7.
47. Garavaglia E, et al. Adenomyosis and its impact on women fertility. Iran J Reprod Med. 2015;13(6):327.
48. Vannuccini S, et al. Pathogenesis of adenomyosis: an update on molecular mechanisms. Reprod Biomed Online. 2017;35(5):592–601.
49. Liu Y, et al. Down-regulation of tumor suppressor PDCD4 expression in endometrium of adenomyosis patients. Current Res Transl Med. 2016;64(3):123–8.
50. Ye F, Zhang J, Qin J-q. Expression and significance of COX-2 and VEGF in adenomyosis. J Harbin Med Univ. 2006;3
51. Huang Y, et al. Expression of tyrosine kinase receptor B in eutopic endometrium of women with adenomyosis. Arch Gynecol Obstet. 2011;283(4):775–80.

52. Cakmak H, et al. Immune-endocrine interactions in endometriosis. Front Biosci (Elite Ed). 2009;1(2):429–43.
53. Keenan JA, et al. Interferon-gamma (IFN-γ) and Interleukin-6 (IL-6) in peritoneal fluid and macrophage-conditioned Media of Women with Endometriosis. Am J Reprod Immunol. 1994;32(3):180–3.
54. Bergqvist A, et al. Interleukin 1β, interleukin-6, and tumor necrosis factor-α in endometriotic tissue and in endometrium. Fertil Steril. 2001;75(3):489–95.
55. Yang JH, et al. Increased interleukin-6 messenger RNA expression in macrophage-cocultured endometrial stromal cells in adenomyosis. Am J Reprod Immunol. 2006;55(3):181–7.
56. Chen C, et al. The microbiota continuum along the female reproductive tract and its relation to uterine-related diseases. Nat Commun. 2017;8(1):1–11.