Pharmaceutical Sciences & Analytical Research Journal
ISSN: 2640-6659

Academic Strive
Researcher's perception is our reality

Review Article *Volume 6 Issue 3*

Uterine Endometrial Receptivity and Biochemical Markers

Ibrahim MA*

College of Pharmacy, University of Mosul, Iraq

***Corresponding author:** Manal Abdulmunem Ibrahim, College of Pharmacy, University of Mosul, Mosul, Iraq, Tel: +9647728430744; Email: alfarhamanal@uomosul.edu.iq

Received Date: August 12, 2024; **Published Date:** August 26, 2024

Abstract

Background: The failure of embryo implantation means the inability to achieve pregnancy and infertility. Uterine receptivity is an important topic in gynecology and depends on the presence of different biomolecules in the body and the uterus. Aim: This review is to show the modern insights on biomolecules that may related to uterine receptivity.

Methods: The website Pubmed is searched for the phrase "biochemical markers of uterine receptivity". Forty-eight articles published between 2019 and 2023 were selected.

Main Findings: These biomolecules that increase uterine receptivity include: Genes (HOXA-10, LIF, CTNNA-2, and others genes). Proteins in uterine fluid (annexins, integrins, mucin, and profilin), CD-44, osteopontin, Legumina, IGFB-7, CK-7 proteins in endometrial tissues. The miRNA in extracellular vesicles of the uterine fluid. Cell surface markers on exosomes of uterine fluid, such as, CD-63, and Hormones and progesterone. The infertile women may show; lower levels of 17HSDB-2, AZGP-1, TPPP-3, and S-10013, higher levels of cystatin B, CBG, and fetuin. Sixteen miRNAs are dysregulated in women with implantation failure. Dysregulated immune profile. Increase CD-9 surface markers. 6-Lower estrogen receptor (ER beta) and progesterone receptors. Low progesterone and a higher level of mid-luteal estradiol. 8- Increase total oxidant status. Defect in ribosomal subunits, mitochondrial dysfunction, abnormal metabolic process, low vitamin D, and increased vaginal microbe. A higher proportion of uterine senescent cells. Alter immune response of the uterus due to HSV, HPV, and chronic endometritis. Berberine, metformin, lovastatin, and lifestyle modification may enhance uterine receptivity of PCOS. Platelet-rich plasma treatment may increase uterine receptivity. Hyperactivated immune profile treated by prednisolone and high dose of progesterone. Hypoactivated immune profile treated by HCG and endometrial scratching.

Conclusion: Different biomolecules may affect uterine receptivity. Identification of these markers is an efficient way to increase the possibility of implantation success.

Keywords: Window of Implantation; Polycystic Ovary Syndrome; Micro RNA; Uterine Natural Killer Cells

Abbreviations

WOI: Window of Implantation; IUI: Intrauterine Insemination; IVF: *In Vitro* Fertilization; ICSI: Intra-Cytoplasm Sperm Injection; DIGE: Two-Dimensional-Differential in Gel-Electrophoresis, LC-MS/MS: Nanobore-Liquid Chromatography-Tandem Mass-Spectrometry; iTRAQ: Isobaric-Tag for Relative and Absolute-Quantitation; LIF: Leukemia Inhibitory Factors; CTNNA-2: Catenin Alpha 2; MLL-1: Mixed Lineage Leukemia 1 Protein; EZH-2: Enhancer of Zest Homolog 2; DNA: Deoxyribonucleic Acid; RNA: Ribonucleic Acid; HSD-17B2: 17b-Hydroxysteroid Dehydrogenase; CK-7: Cytokeratin 7; AZGP-1: Zinc-α-2-Glycoprotien; TPPP-3: Tubulin Polymerization Endorsing Protein Family Member 3; CBG: Cortisol Binding Globulin; PCR: Polymerase Chain Reaction; TLRs: Toll-Like Receptors; uNK: Uterine Natural Killer Cells; ERA: Expression Microarray Analysis; HPV: Human Papilloma Virus; HSV: Human Simplex Virus Infection; bFGF2: Fibroblast Growth Factor Basic; PCOS: Polycystic Ovary Syndrome; α v B-3: Alpha-v-Betal-3 Receptors; LPAR-3: Lysophosphatidic Acid Receptor 3; IGFBP-7: Insulin-Like Growth Factor Binding 7; RIF: Recurrent Implantation Failure.

Introduction

The normal endometrium becomes receptive to the embryo through the time of the window of implantation (WOI), which always happens 7 days after the peak of LH and lasts for 2 to 6 days. Steroid hormones lead to the maturation of endometrium and affect gene expression for initiating WOI time [1]. After menstruation, the endometrium is in the proliferative phase, and then becomes early secretory, mild secretory, and late secretory phase before the second menstruation [1].

The implantation failure may be attributed to the type of the embryo, the endometrium, and the interaction between the two. Embryonic and maternal biomarkers may include chromosomal abnormalities, maternal immune dysfunction, endometrial vascularization, and blood flow. The vascularization flow index is significantly higher in fertile women than in infertile women [2].

The endometrial embryo implantation entails three steps: apposition, adhesion, and invasion. During apposition chemokines and other molecules in uterine fluid are important for nexus between the fetus and the endometrium. The adhesion step requires adhesion molecules, such as, integrins, mucin, and osteopontin. The invasion step is regulated by steroid hormones, cytokines, transporting factors, and cell cycle molecules [3]. Therefore, different molecules may be proposed as important biochemicals to accomplish uterine receptivity, failure of implantation means failure to achieve pregnancy and hence infertility. Even advanced methods to treat infertility, such as, intrauterine insemination (IUI), in vitro fertilization (IVF), and intra-cytoplasm sperm injection (ICSI) may be ended in implantation failure. Recurrent implantation failure means the failure to achieve pregnancy due to implantation failure for three times of transferring embryos in IVF procedures [4].

In this review, we searched the website of Pubmed about the statement " Biochemical markers of uterine receptivity" We selected articles published between 2019 and 2023 that are either original research, clinical research, scientific reports, or meta-analysis that deal with human beings. Review articles and animal research are neglected. 48 articles fit these criteria. This search is to show modern insights into molecules that may affect uterine receptivity. The information obtained can be written under the following topics.

Specimen Collection and Methods of Determination

The specimens may be 1- Endometrial biopsy, 2-Uterine fluid, 3- Extracellular vesicles, and 4-Endometrial cervix.

Endometrial Biopsy: this specimen is used for sequencing and analyzing DNA, RNA, or proteome analysis. The proteome analysis for endometrial tissue biopsy may be done by two-dimensional-differential in gel-electrophoresis (DIGE), nanobore-liquid chromatography-tandem mass-spectrometry (LC-MS/MS), and isobaric-tag for relative- and absolute-quantitation (iTRAQ). An improved technique iTRAQ LC-MS/MS was employed to screen specific proteins related to endometrial receptivity which detected 263 differentially expressed proteins in women with implantation failure [5].

The tests used for sequencing the important genes affected are ERA test [6], ER Map [7], WIN-test [8], be READY [1], and TAC-seq. The (Rs ERT test) is an RNA seq-based endometrial receptivity test used for analyzing the RNA of endometrial tissue to predict the WOI period and improve pregnancy rate. They found that the pregnancy rate increased by 20% when transferring blastocysts during exact WOI time [9]. The rsERT, comprising 175-biomarker-genes, showed an average accuracy of 98.4% [9].

Uterine Fluid: They utilize uterine fluid as it contains extracellular vesicles, RNA, DNA, regulating protein, ions, lipids, and other biofactors. The uterine fluid can be obtained by non-invasive methods to determine the receptivity of the endometrium as they found 800 genes within the uterine fluid may be involved in implantation biology. They utilize non-invasive RNA-sequence-based tests by analyzing transcriptomic profiles [10]. The uterine fluid collection may be a less invasive routine practice when compared to a biopsy [11].

Extracellular Vesicles from Uterine Fluid: Uterine receptivity may link to extracellular vesicles. these vesicles

are secreted from endometrial cells lining the glands and contain cargo for example lipids, proteins, and nucleic acids. These extracellular vesicles known as exosomes (30-150nm), or microvesicles (100-350 nm), these structures are important for cell-to-cell communication [12]. The isolation of endometrial secretome (proteins expressed by endometrium and secreted to extracellular space) is done by uterine lavage then isolation of extracellular vesicles by ultracentrifugation or sucrose cushion [13]. This is a simple, non-invasive method and can be carried out during transvaginal ultrasonography during WOI [14]. The transcription of uterine fluid extracellular vesicles is correlated with the endometrium tissue transcription and includes genes known to regulate cell adhesion and implantation [10].

Cervix Biopsy: The Human cervical epithelium during the period of pre-implantation exhibited an elevated level of LIF which may be considered as a biomolecule that detect uterine receptivity without invasive endometrial damage [15].

Biomolecules that are Related to Uterine Receptivity:

Genes: Significant differences in transcripted genes between fertile-females and those with intermittent implantation failure are found. 122 differentially expressed genes are downregulated, and 66 are upregulated in females with intermittent implantation failure [16]. Moreover, the genes that interact with many other genes and are most closely associated with the disease called hub genes may play an important role in recurrent implantation failure. Three to ten hub genes strongly correlated with signaling pathways and immune response in recurrent implantation failure [16,17]. The immune pathways were remarkably decreased while lipid catabolism pathways were remarkably increased in those patients [17].

However, endometrial receptivity can be assessed by 72 genes, four of them are housekeeping genes (genes that are expressed in all cells in normal and pathological conditions for cellular basic function) using uterine fluid-derived extracellular vesicles transcriptome [18] or by using uterine biopsy [1]. The examples of important genes are as follows:

- The HOXA-10 gene, one of 39 genes of the homeobox (HOX) gene family responsible for embryonic development, is crucial for embryo implantation and decidualization and encodes the protein Homeobox protein Hox-A10 which is important for protein binding and DNA binding [19].
- The LIF gene (leukemia inhibitory factors), encodes for the pleiotropic cytokines important for hematopoietic differentiation, leukemia cells terminal differentiation, induction of neural cell differentiation, and the immune tolerance at the maternal-fetal interface [15].
- CTNNA-2 (catenin alpha 2) gene is important for uterine receptivity [20]. The CTNNA-2 is a protein-coding gene that regulates cell-cell adhesion between cadherin adhesion receptors and the cytoskeleton [21].
- The ZEB-1 gene encodes a zinc finger transcription factor that binds the HOXA-10 gene controlling its expression and modulating endometrial receptivity through epithelial mesenchymal transition promotion. ZEB-1 gene is highly expressed at mRNA and protein levels in human endometrium during the mid-secretory phase of the menstrual cycle. Also, it promotes mesenchymal transition in carcinogenesis [22].
- Moreover, higher expressions of ABCG-2 and ALDH-1A1 genes are detected in receptive women's uterus. ABCG-2 gene encodes for proteins of transport, heme transport, and protein binding, while the ALDH-1A1 gene encodes for metabolism, retinol metabolic process, and oxidoreductase activity [23].
- The shift from the pre-receptive to the receptive phase of the endometrium showed an altered manifestation of specific genes, such as, ICAM-1, NFKB-1A, UCAM-1, LIF, VEGF, TLR-5+ suggesting their enrollment in the endometrial receptivity [24].

Proteins: The uterine fluid proteins can be used to estimate uterine receptivity. A study revealed that over 3000 proteins can be found in uterine fluid, 367 of these proteins undergo significant alterations when endometrium is transformed from early secretory endometrium to mid-secretory endometrium during WOI. While women with repeated implantation failure showed an altered mid-secretory endometrial profile in the uterine fluid after proteomic analysis using mass spectrophotometry [25].

Segura-Benitez M, et al. [26] study revealed that extracellular vesicle proteins are important for uterine receptivity and the best method of isolation of these vesicles is by ultracentrifugation, 218 proteins were present in the extracellular vesicles, and 82 were selected as novel biomarkers for endometrial receptivity [26]. These proteins may include annexins, collagen VI, integrins, mucins, and profilin 1. Annexins are upregulated during the receptive endometrial phase and act as adherent molecules between embryo and endometrial, while integrins mediate cell adhesion to collagens and laminin. The profilin 1 is manifested by endometrial epithelial cells and necessary for embryo attachment, while other metalloproteinase proteins affect tight junctions in trophoblastic cells. Moreover, integrin α.v.β-3, VEGF, TNF-α, and LIF in uterine fluid were higher in fertile women. Integrin α.v.β-3 has the best prediction value for endometrial receptivity among other biomarkers [2].

Integrins are cell adhesion molecules important in cell-cell adhesion and cell-extracellular matrix adhesion. With the start of gestation, integrin manifestation is important for trophoblast adherance and embryo penetration of the decidua [27]. Several integrins types exist that act alongside

other cell adhesion molecules, such as, selectins, and cadherins. The integrin molecules are folded into U-shaped embedded in the cell membrane not simply hooks but also signals cells to do certain actions, such as, attachment, differentiation, or death [28].

The proteins in the endometrial tissues are deemed the main direct effective biomolecule and the last effector of transcriptional gene translation [5], such as, mucin 1, and cyclooxygenase-2. Mucin 1 glycoprotein is required for adhesion, while mucin 16 overexpression on the endometrial cell surface may hinder implantation [29]. The cyclo-oxygenase-2 is necessary for decidualization because it is important for arachidonic acid conversion to prostaglandin E-2 which is important for implantation [30].

Also, osteopontin and CD-44 play a significant role in uterine receptivity. The CD-44 is a transmembrane glycoprotein implicated in the migration and adhesion of endothelial cells. The osteopontin is a phosphoglycoprotein that acts as a bridge between the endometrium surface and trophoblast through interaction between α.v.β-3 integrin and CD-44. CD-44 and osteopontin are increased in the secretory phase in the endometrial tissue of fertile women during the window of implantation to form a complex that is vital in fetus recognition [31].

Moreover, the legumina protein, the cysteine endopeptidase that hydrolyzes asparaginyl bonds, may regulate trophoblast invasion and endometrial remodeling. The glycoprotein IGFBP-7 may regulate the IGF-1 metabolism interacts with IL-6 expressed in the endometrium, and plays an important role in decidualization. Hepatocyte growth factor, is important for cell proliferation because it binds with hepatocyte growth factor receptors. CK-7(cytokeratin 7) is upregulated to three folds in receptive endometrium [32].

Other proteins that act as regulators for gene transcription, such as, MLL-1 (mixed lineage leukemia 1 protein) which is the master regulator of transcription of HOX genes, this protein function as methyltransferase and DNA transcription factor. Additionally, the EZH-2 protein (Enhancer of Zest homolog 2) a histone-lysine N-methyltransferase enzyme encoded by the EZH-2 gene acts as a negative regulator of gene expression and negative regulator of DNA binding transcription factors [33]. A study found that ratio of MLL-1:EZH-2 was low in the uterine secretion of non-receptive women, those women exhibited low MLL-1 and high EZH-2, because EZH-2 inhibits HOXA-10 expression and decreased decidualization, while MLL-1 has importance in downstream effect on HOXA-10 gene [19].

There are 263 differentially manifested proteins in the endometrial tissue of patients with repeated implantation failure [5]. The 17b-hydroxysteroid dehydrogenase (HSD-17B2), zinc-α-2-glycoprotien (AZGP-1), tubulin polymerization endorsing protein family member 3 (TPPP-3), and S-100A13 are significantly lower in the non-receptive uterus, therefore may be considered as fundamental biochemical factor for endometrial receptivity and findings of repeated implantation failure [5,34]. HSD-17B2 is an enzyme that catalyses the generation and inactivation of estrogen and androgen, the synthesis of active progesterone, and the oxidation of estradiol to estrone. This enzyme is allocated in the endoplasmic reticulum and is potentially manifested in glandular and luminal cells. The AZGP-1 protein is a secretory adipokine regulated by the thyroid, androgen, and glucocorticoid hormones. It is important for lipolysis, glucose transport, and decreasing inflammatory factors. The TPPP-3 is allocated in the nucleus and presented in endometrial ciliated cells to alleviate the reliability of the microtubule system which is the main component of the mitotic spindle that controls cell division and aggregation. And, S-100A13 is a small calcium-binding protein important for calcium homeostasis and cell proliferation [5,34].

The endometrial tissue biopsy specimens showed 82 differentially manifested proteins in women with recurrent implantation failure, 55 proteins are upregulated and 27 are downregulated [35]. Cystatin B the intracellular inhibitor of thiol proteinase cathepsin B reported an increase in miscarriage cases [32]. Also, higher levels of CBG (cortisol binding globulin) and the Fetuin-A protein are presented in women with repeated implantation failure [35]. CBG is important for cortisol delivery, inflammation, and metabolism, its increase may relate to low progesterone levels. Fetuin-A is the major carrier of free fatty acids important for free fatty acid-induced insulin resistance [36] and decreases embryo implantation [35]. Another protein that decreases uterine receptivity is podocalyxine expressed by the human endometrial epithelium. This protein decreases implantation by rendering the epithelium nonadhesive. Moreover, this protein may suppress gene expression of cell adhesion (LIF), stimulate anti-implantation genes, such as, (LEFTY-2), and increase expression proteins of adherence, and tight junction, such as, E. cadherin and claudin. However, the luminal epithelium must decrease this factor to switch to the receptive phase during the window of implantation [37].

miRNA: The mature mi-RNA or micro-RNA are small noncoding regulatory RNA (19-25 nucleotides) that regulate hundreds of mRNA through complementation with the non-translated regions of their target transcripts that result in inhibition or translation promotion or negatively affect gene expression by degradation of m RNA. The miRNA can be determined by real-time PCR [38]. These molecules participate in angiogenesis by modulating the expression of proteins that promote vessel growth [39]. The micro RNA present in extracellular vesicles of uterine fluid modulates implantation by affecting target genes of the epithelial cells

[40]. Therefore, miRNAs act as epigenetic regulators of endometrial receptivity and embryo implantation through post-translational modification. The implantation failure may be related to the dysregulation of miRNAs [40].

A study on the endometrial fluid used the endometrial receptivity array technique that depends on tissue gene expression of micro RNA by using real-time PCR during the window of implantation found that 61 mi RNA are dysregulated in women with recurrent implantation failure when compared to healthy women, 34 are upregulated and 27 are downregulated [40]. While, the study of Tiantian Li, et al. [12] found that after ultracentrifugation and separation of extracellular vesicles from uterine fluid, 12 endometrial extravascular small noncoding RNA identified are related to endometrial receptivity and are associated with the biological function of the immune response, extracellular matrix remodeling, and cell junction. Moreover, this study identified a small noncoding RNA that termed hsa-miR-362-3p as a highly expressed mi RNA in non-pregnant women with implantation failure.

Moreover, the miR-183 family shows an estrogen-dependent upregulation in endometrial cells and has a positive effect on the migration and proliferation of these cells. The miR-183-5P mediates the regulation of the CTNNA-2 gene in the endometrial cell and enhances the effect of estrogen on endometrial receptivity [20]. However, no significant differences in mi-RNA expression are found between the natural cycle and hormone replacement therapy cycle [41].

Immune Cells: A study revealed that 75% of recurrent pregnancy loss is usually due to a dysregulated immunological profile that when treated increases life birth to 55% in those women [42]. Innate immune cells, such as, natural killer cells, macrophages, and dendritic cells are abundant at the implantation site. These cells express toll-like receptors (TLRs) that when stimulated result in the expression of anti-inflammatory cytokines. Also, gene expressions of TLR signaling molecules, such as, TRIB2 and TLR9 showed a difference between females with repeated implantation failure and fertile women [24]. Also, the immune cells infiltration is reduced in recurrent implantation failure, such as, CD56 natural killer cells, dendritic, Th-1, Th-2, regulatory T cells, and macrophages [17].

The uterine natural killer cells (uNK) in fertile women are important for trophoblast invasion [43]. The CD-56+ uNK cells associate with the transcriptional biomolecules of endometrial receptivity considered by gene expression microarray analysis (ERA test) (44). While CD-16+ natural killer cells resulted in embryo rejection in infertile women [16], and a very high level of uterine natural killer cells is a predictor of miscarriage [43].

The T-helper cells cytokines (Th-2 cytokines) favor the implantation process, while the T-helper cytokines (Th-1 cytokines) are harmful for implantation [43]. Also, the B-cell activation plays an important role in repeated implantation failure, therefore CD-20 receptors of B-cell are increased in abortion [32].

The dysregulated immune profile can be categorized into hyper-activated, hypo-activated, and a mixed immune profile. The hyper-activated profile resulted in pregnancy loss by direct rejection of the embryo. The mixed profile is characterized by immune over-activation excess of Th1 cytokines, with immature uNK cells [42]. Also, the hyper-activated or hypo-activated immune profile can be classified according to the maturation of uterine natural killer cells, IL-15 (a marker of uNK cell activation), and IL-18 (a marker of angiogenesis) [43].

Surface markers: CD-9 is a cell surface glycoprotein containing palmitoylation site that allows CD-9 to interact with lipids and proteins, such as, integrins producing negative regulation of cell proliferation [45]. CD-63 is a protein associated with the membrane of intracellular vesicles or cell surface expression. It can be expressed in stromal cells of the endometrium and function in protein binding and positive regulation of integrin-mediated signaling pathways [46]. The exosomes (30-100 nm) in the uterus are secreted from the endometrium epithelium contain miRNA and exhibit cell surface markers CD-63 and CD-9. The exosomes in fertile and infertile women showed differences in CD-9 and CD-36 exhibition. The exosome production should be increased in the mid-secretory phase for implantation, the CD-63 expression is higher in fertile women and reaches the highest level in the period of implantation window, but the CD-9 expression is increased in non-receptive women and can be used as a biomarker of infertility [47].

Receptors: It is known that there are two types of estrogenic receptors in the endometrium tissues, these are ER alpha and ER beta. The ER alpha receptors are expressed during the follicular phase of the menstrual cycle, while ER beta receptors are the dominant estrogen receptors subtype and are expressed within the vascular endothelium during the window of implantation, these receptors are important for angiogenesis and vascular remodeling. The study of Al-Lamee H, et al. [48] found that infertile women have a lower level of estrogen receptor type (ER beta) and progesterone receptors. Conditions that lead to decreased estrogen levels, such as, GnRH agonists may result in a significant reduction of endometrial progesterone receptors and ER.β. that lead to infertility [48].

Steroid Hormones: The normal hormonal levels during implantations are crucial, as the endometrial receptivity window occurs in the mid-secretory phase after sufficient

time from progesterone exposure. The progesterone inhibits the synthesis of cholesterol in the epithelial compartment resulting in inhibition of epithelial cell proliferation during the mid-secretory phase. Moreover, abnormal progesterone signaling leads to infertility and other gynecological disease [49].

The binding of progesterone with the progesterone receptors resulted in the activation of the expression of PGR-regulated genes, such as, homeobox gene (HOXA-10), bone morphogenesis protein-2(BMP-2), MMP-2, SERPINE-1, MNMT, and WNT-5A, EMP-1, IER-3. Moreover, numerous epithelial cell surface markers are upregulated and presented upon PGR binding, such as, CLDN-4, CLDN-8, and KLF-4 [49]. Also, after 72 hours from progesterone release, podocalyxin protein an adhesion transmembrane sialomucine and negative regulator of uterine receptivity is downregulated [50]. Normally, the podocalyxin protein tends to be higher in low progesterone levels because it increases epithelial polarity during non-receptive phases of the endometrium. This protein should be decreased during the window of implantation period when the endometrium becomes receptive [50]. The administration of progesterone and human chorionic gonadotrophin HCG after ovarian stimulation resulted in increased VEGF level and miR-17-5P which stimulate angiogenic pathway, endometrial vascular activity, and endometrial receptivity [39].

The progesterone hormone increases the expression of 17b-hydroxysteroid dehydrogenase enzyme (HSD-17B2), but low progesterone levels cause overexpression of cortisol binding globin (CBG), which dysregulates endometrial immune condition [34,35]. However, the infertile women showed lower levels of HSD-17B2 and higher estradiol levels than fertile women [34]. Also, the level of the mid-luteal estradiol is reversely linked to markers of endometrial receptivity maturation [44].

Cytokines, Microbiota, and Infections: Women with idiopathic infertility showed lower levels of TGFb-1 (transforming growth factor), bFGF-2 (fibroblast growth factor basic), and a high level of DEFa-1 (alpha-defensin). The expression of these markers is correlated with the incidence of endometrial peptostreptococcus, human papilloma virus (HPV), history of repeated human simplex virus infection (HSV), and abortion [51].

TGFb-1 is an extracellular multifunctional polypeptide cytokine produced by white blood cells that controls some cellular functions, such as, cell growth, proliferation, differentiation, and T-cell regulation [52]. The bFGF2 (fibroblast growth factor basic) interacts with the transmembrane receptors, such as, integrin and influences cell proliferation and tissue vascularization. The FGF2 expression increases in the glandular epithelium of the secretory phase of the endometrium of fertile women acting as an integrin ligand important for adhesion, development, differentiation, and angiogenesis [53]. The DEFa-1 (α-defensin) is produced by neutrophils and epithelial cells upon infection. it is an inducible bacteriolytic protein that acts on gram-positive and gram-negative bacteria [54].

Furthermore, it is found that Enterococcus faecalis especially with superoxide-producing E. faecalis may result in opportunistic chronic endometritis and lead to infertility because of its effect on the expression of cytokines that promote apoptosis and damage uterine receptivity [55]. Therefore, uterine cavity infection may alter cytokine pathways crucial for blastocyst growth and implantation [51]. Women with recurrent HSV, HPV, miscarriage, and chronic endometritis should undergo an assessment of their immune biomarkers [51,55].

Oxidative Stress: The total antioxidant status and enzyme prolidase enzyme activity were higher in patients with unexplained infertility. Parolidase enzyme is important for protein metabolism, matrix remodeling, inflammation, angiogenesis, and cell proliferation. The enzyme parolidase is increased in oxidative stress and can be used as an oxidative stress marker in various diseases [56].

Other Factors: Women with recurrent implantation failure showed down-regulation of the ribosomal proteins, mitochondrial upset and abnormal metabolic routes, such as, hormones and lipids [5]. The normal metabolism and mitochondrial upsets are important for fetal implantation and gestation. Therefore, lysophosphatidic acid receptor 3, and glucose transporter 1 were linked to endometrial receptivity [5]. In general, women with recurrent implantation failure had lower vitamin D levels, border lower progesterone levels, and more vaginal microbe compared with control [57]).

Effect of Age and Gynecological Disease on the Endometrial Receptivity

Aging: The endometrial aging is separated from individual age. The stemness is inversely associated with senescence in human endometrial stromal cells and sixteen human genes expression was shown to change with the aging of the endometrium [58]. The endometrial stromal cells in non-receptive women have a higher proportion of senescent cells (cells that stop multiplying do not die, but release chemicals that trigger inflammation, increase expression m RNA of CDKN-1A genes, and expression of senescence secretions). The CDKN-1A gene functions for DNA damage, regulation of cell death, cellular senescence, cytokine-mediated pathway, and negative regulation of vascular proliferation [59].

Therefore, the implantation failure may be due to senescence promotion induced by stress, oxidative stress, or DNA damage [23].

On the other hand, autophagy -the breakdown of old cells in the body and reuse so that cells operate more effectively- is highly present in normal human proliferative, secretory, and decidual tissue manifested by autophagy-related markers, such as, LC3 and P 62 indicating that autophagy may be essential for embryo implantation [60].

Polycystic Ovary Syndrome (PCOS) and Uterine Receptivity: Although of the ovulatory cycle, PCOS women may have infertility. This may be related to abnormal expression of certain receptors, such as, alpha-v-betal-3 receptors (α v B-3), and lysophosphatidic acid receptor 3 (LPAR-3). The first receptor is a type of integrin and acts as a receptor for phagocytosis on macrophages and dendritic cells. The second receptor is a protein-coupled receptor that binds the lipid signaling molecule lysophosphatidic acid which evokes calcium mobilization. The abnormality of these receptors in PCOS may be revised by berberine or metformin administration [61]. The lipid metabolism-related genes can modulate embryo implantation by affecting adhesion molecules, adipokines, and other lipids. The abnormality of lipid metabolism in women with PCOS, and hence the implantation failure can be treated by lovastatin administration or quercetin by their effect on blood lipids [62]. Moreover, the lifestyle modification in PCOS may modulate endometrial proteomes, such as, an increase of legumain, insulin-like growth factor receptors, keratin, type II cytoskeletal 7 and cystatin B, and a decrease of CD20 beta lymphocyte antigen [32]. Also, downregulated genes in receptive endometrium showed more downregulation in obese PCOS upon weight loss [63].

In polycystic ovarian women, the levels of osteopontin and CD44 receptors are increased in circulation and local secretions, but decreased in endometrial tissues which leads to implantation failure, because of hindering endometrium-trophoblast interaction by saturating osteopontin and CD44 receptors on the surface of blastocysts [31]. However, a study showed that women with recurrent implantation failure, if PCOS or not undergo displaced or transition in the window of implantation time [1]. Therefore, in women with shifted implantation time, the pregnancy rate can be increased after using a personalized window of implantation determination [1].

Endometriosis and Uterine Receptivity: The impaired endometrial receptivity in women with endometriosis may related to gene polymorphism of muc-1 and cox-2 [30], or due to their proteomic difference from fertile women [64]. Women with minimal to mild endometriosis showed upregulation of six proteins associated with endometrial receptivity. The higher upregulated protein after metformin treatment is insulin-like growth factor binding 7(IGFBP-7). Therefore, metformin may enhance endometrial receptivity in endometriosis by improving the expression of endometrial receptivity marker IGFBP-7 [64].

Anti-Phospholipid Syndrome: Women with anti-phospholipid positivity exhibit recurrent implantation failure due to their inhibition of LIF and HOXA-10 expression, or due to abnormal uterine pinpode development during the window of the implantation period [65].

Treatment of Recurrent Implantation Failure

The treatment of immune hyperactivated women is by prednisolone 20 mg/day, vitamin E twice a day, and high doses of progesterone because progesterone decreases the expression of pro-inflammatory cytokine. The treatment by prednisolone continues until 8 weeks of pregnancy because it decreases Th1 cytokines and IL-15 mRNA overexpression. Also, heparin can be effective by modulating the complements effect, and intravenous intralipid can be used with high-dose progesterone to decrease immune activation and increase the Th-2 effect [42].

The treatment of hypo-activated women is done by endometrial scratching and administration of HCG injection 1500 IU on days 4,6, and 8 after oocyte retrieval for women undergoing IVF to trigger the maturation of uNK cells [61], also sexual intercourse is recommended [42,43].

The procedure of platelet-rich plasma treatment may increase uterine receptivity because of the increased expression of microRNA (miR-211-3p) which increases the chances of pregnancy. However, the increase of IGF-1 levels after platelet-rich plasma treatment is related to poor pregnancy outcomes [38].

Metformin is recommended to increase uterine receptivity for women with minimal to mild endometriosis [66], and PCOS women [61]. Also, the low uterine receptivity of PCOS women can be treated by berberine, Lovastatin, and quercetin [61,62].

Conclusion

No single molecule or receptor can affect uterine receptivity, but there are large numbers of mediators. Certain gynecological diseases may affect uterine receptivity, such as, endometriosis and PCOS or it may be to unknown causes. The age of women is not a reflection of endometrial receptivity. Different biomolecules may affect uterine receptivity, such as, certain genes, their transcriptional molecules, proteins, receptors, hormones, uterine fluid microvesicles contents,

and miRNAs. Moreover, infection, immunity, and microbiota play important roles in this respect. Characterization and identification of biomarkers for the receptive endometrium is an effective method for increasing the probability of successful embryo implantation.

References

1. Meltsov A, Saare M, Teder H, Paluoja P, Arffman RK, et al. (2023) Targeted gene expression profiling for accurate endometrial receptivity testing. Sci Rep 13(1): 13959.
2. Wang L, Lv S, Mao W, Pei M, Yang X (2020) Assessment of endometrial receptivity during implantation window in women with unexplained infertility. Gynecol Endocrinol 36(10): 917-921.
3. Hernandez-Vargas P, Munoz M, Dominguez F (2020) Identifying biomarkers for predicting successful embryo implantation: applying single to multi-OMICs to improve reproductive outcomes. Hum Reprod Update 26(2): 264-301.
4. Coughlan C, Ledger W, Wang Q, Liu F, Demirol A, et al. (2014) Recurrent implantation failure: definition and management. Reprod Biomed Online 28(1): 14-38.
5. Yang J, Wang L, Ma J, Diao L, Chen J, et al. (2023) Endometrial proteomic profile of patients with repeated implantation failure. Front Endocrinol Lausanne 14: 1144393.
6. Diaz-Gimeno P, Horcajadas JA, Martinez-Conejero JA, Esteban FJ, Alama P, et al. (2011) A genomic diagnostic tool for human endometrial receptivity based on the transcriptomic signature. Fertil Steril 95(1): 50-60.
7. Enciso M, Carrascosa JP, Sarasa J, Martinez-Ortiz PA, Munne S, et al. (2018) Development of a new comprehensive and reliable endometrial receptivity map (ER Map/ER Grade) based on RT-qPCR gene expression analysis. Hum Reprod 33(2): 220-228.
8. Haouzi D, Assou S, Mahmoud K, Tondeur S, Reme T, et al. (2009) Gene expression profile of human endometrial receptivity: comparison between natural and stimulated cycles for the same patients. Hum Reprod 24(6): 1436-1445.
9. He A, Zou Y, Wan C, Zhao J, Zhang Q, et al. (2021) The role of transcriptomic biomarkers of endometrial receptivity in personalized embryo transfer for patients with repeated implantation failure. J Transl Med 19(1): 176.
10. He A, Wu H, Zou Y, Wan C, Zhao J, et al. (2023) Can biomarkers identified from the uterine fluid transcriptome be used to establish a noninvasive endometrial receptivity prediction tool? A proof-of-concept study. Reprod Biol Endocrinol 21(1): 20.
11. Giacomini E, Scotti GM, Vanni VS, Lazarevic D, Makieva S, et al. (2021) Global transcriptomic changes occur in uterine fluid-derived extracellular vesicles during the endometrial window for embryo implantation. Hum Reprod 36(8): 2249-2274.
12. Li T, Greenblatt EM, Shin ME, Brown TJ, Chan C (2021) Cargo small non-coding RNAs of extracellular vesicles isolated from uterine fluid associate with endometrial receptivity and implantation success. Fertil Steril 115(5): 1327-1336.
13. Beal JR, Ma Q, Bagchi IC, Bagchi MK (2022) Role of Endometrial Extracellular Vesicles in Mediating Cell-to-Cell Communication in the Uterus: A Review. Cells 12(22): 2584.
14. Saraee F, Shekari F, Moini A, Sadeghi M, Ghaznavi P, et al. (2022) Human uterine fluid lavage-derived extracellular vesicle isolation: a comparative study for minimally invasive endometrial receptivity assessment. Reprod Biomed Online 45(3): 457-472.
15. Fukui Y, Hirota Y, Aikawa S, Ishizawa C, Iida R, et al. (2022) Uterine Receptivity is Reflected by LIF Expression in the Cervix. Reprod Sci 29(5): 1457-1462.
16. Lai ZZ, Zhang J, Zhou WJ, Shi JW, Yang HL, et al. (2023) Identification of potential biomarkers and immune infiltration characteristics in recurrent implantation failure using bioinformatics analysis. Front Immunol 14: 992765.
17. Li F, Gao W, Li Y, Wang Y, Liu L, et al. (2023) Potential Biomarkers and Endometrial Immune Microenvironment in Recurrent Implantation Failure. Biomolecules 13(3): 406.
18. Meltsov A, Giacomini E, Vigano P, Zarovni N, Salumets A, et al. (2023) Letter to the Editor - Pilot proof for RNA biomarker-based minimally invasive endometrial receptivity testing using uterine fluid extracellular vesicles. Eur J Obstet Gynecol Reprod Biol 287: 237-238.
19. Zou K, Du Q, Chen X, Tang P, Liang H (2023) MLL1:EZH2 Ratio in Uterine Secretions and Endometrial Receptivity in Patients with Endometriosis. Lab Med 54(1): 90-97.
20. Akbar R, Ullah K, Rahman TU, Cheng Y, Pang HY, et al. (2020) miR-183-5p regulates uterine receptivity and enhances embryo implantation. J Mol Endocrinol 64(1): 43-52.

21. GeneCards Suite (2024) Aliases for CTNNA2 Gene.

22. Ran J, Yang HH, Huang HP, Huang HL, Xu Z, et al. (2020) ZEB1 modulates endometrial receptivity through epithelial-mesenchymal transition in endometrial epithelial cells in vitro. Biochem Biophys Res Commun 525(3): 699-705.

23. Tomari H, Kawamura T, Asanoma K, Egashira K, Kawamura K, et al. (2020) Contribution of senescence in human endometrial stromal cells during proliferative phase to embryo receptivity. Biol Reprod 103(1): 104-113.

24. Basatvat S, Russell JM, Saare M, Thurston LM, Salumets A, et al. (2021) Potential innate immunity-related markers of endometrial receptivity and recurrent implantation failure (RIF). Reprod Biol 21(4): 100569.

25. Kasvandik S, Saarma M, Kaart T, Rooda I, Ehrenberg A, et al. (2020) Uterine Fluid Proteins for Minimally Invasive Assessment of Endometrial Receptivity. J Clin Endocrinol Metab 105(1): dgz019.

26. Segura-Benitez M, Carbajo-Garcia MC, Corachan A, Faus A, Pellicer A, et al. (2022) Proteomic analysis of extracellular vesicles secreted by primary human epithelial endometrial cells reveals key proteins related to embryo implantation. Reprod Biol Endocrinol 20(1): 3.

27. Merviel P, Challier JC, Carbillon L, Foidart JM, Uzan S (2001) The role of integrins in human embryo implantation. Fetal Diagn Ther 16(6): 364-371.

28. (2024) Integrin. In Wikipedia.

29. Liu L, Wang Y, Chen X, Tian Y, Li TC, et al. (2020) Evidence from three cohort studies on the expression of MUC16 around the time of implantation suggests it is an inhibitor of implantation. Journal of Assisted Reproduction and Genetics 37(5): 1105-1115.

30. Budihastuti UR, Dasuki D, Sadewa AH, Utoro T (2023) Endometrial receptivity defects MUC-1 and COX-2 polymorphisms in endometriosis. J Med Life 16(10): 1503-1507.

31. Paravati R, De Mello N, Onyido EK, Francis LW, Younas K, et al. (2020) Differential regulation of osteopontin and CD44 correlates with infertility status in PCOS patients. J Mol Med (Berl) 98(12): 1713-1725.

32. Abdulkhalikova D, Sustarsic A, Bokal EV, Jancar N, Jensterle M, et al. (2022) The Lifestyle Modifications and Endometrial Proteome Changes of Women With Polycystic Ovary Syndrome and Obesity. Front Endocrinol (Lausanne) 13: 888460.

33. (2024) EZH2. In Wikipedia.

34. Yang J, Wang L, Ma J, Diao L, Chen J, et al. (2023) Endometrial proteomic profile of patients with repeated implantation failure. Front Endocrinol (Lausanne 14: 1144393.

35. Wang C, Feng Y, Zhou WJ, Cheng ZJ, Jiang MY, et al. (2021) Screening and identification of endometrial proteins as novel potential biomarkers for repeated implantation failure. PeerJ 9: e11009.

36. (2023) Alpha-2-HS-glycoprotein. In Wikipedia.

37. Heng S, Samarajeewa N, Wang Y, Paule SG, Breen J, et al. (2021) Podocalyxin promotes an impermeable epithelium and inhibits pro-implantation factors to negatively regulate endometrial receptivity. Sci Rep 11(1): 24016.

38. Babian S, Salehpour S, Nazari L, Ghorbanmehr N (2022) The expression level of mir-21-3p in platelet-rich plasma: A potential effective factor and predictive biomarker in recurrent implantation failure. Mol Reprod Dev 89(10): 498-505.

39. Salmasi S, Sharifi M, Rashidi B (2021) Evaluating the effect of ovarian stimulation and exogenous progesterone on CD31-positive cell density, VEGF protein, and miR-17-5p expression of endometrium immediately before implantation. Biomed Pharmacother 133: 110922.

40. von Grothusen C, Frisendahl C, Modhukur V, Lalitkumar PG, Peters M, et al. (2022) Uterine fluid microRNAs are dysregulated in women with recurrent implantation failure. Hum Reprod 37(4): 734-746.

41. Grasso A, Navarro R, Balaguer N, Moreno I, Alama P, et al. (2020) Endometrial Liquid Biopsy Provides a miRNA Roadmap of the Secretory Phase of the Human Endometrium. J Clin Endocrinol Metab 105(3): dgz146.

42. Cheloufi M, Kazhalawi A, Pinton A, Rahmati M, Chevrier L, et al. (2021) The Endometrial Immune Profiling May Positively Affect the Management of Recurrent Pregnancy Loss. Front Immunol 12: 656701.

43. Coutanceau B, Santos ED, Blanchard NS, Louboutin AS, Boitrelle F, et al. (2023) Should the Treatment of Patients with Repeated Embryo Implantation Failure Be Adapted as a Function of the Endometrial Cytokine Profile? A Single-Center Experience. Biomedicines 11(3): 817.

44. Saxtorph MH, Persson G, Hallager T, Petersen KB, Eriksen JO, et al. (2020) Are different markers of endometrial

receptivity telling us different things about endometrial function. Am J Reprod Immunol 84(6): e13323.

45. (2024) CD9. In Wikipedia.

46. (2024) CD63. In Wikipedia.

47. Uyar Y, Ozgul M, Gokap S, Ok G, Tan A, et al. (2020) The correlation between unexplained infertility and exosomes. Ginekol Pol 91(5): 240-246.

48. Al-Lamee H, Ellison A, Drury J, Hill CJ, Drakeley AJ, et al. (2022) Altered endometrial oestrogen-responsiveness and recurrent reproductive failure. Reprod Fertil 3(1): 30-38.

49. Chi RA, Wang T, Adams N, Wu SP, Young SL, et al. (2020) Human Endometrial Transcriptome and Progesterone Receptor Cistrome Reveal Important Pathways and Epithelial Regulators. J Clin Endocrinol Metab 105(4): e1419-e1439.

50. Paule SG, Heng S, Samarajeewa N, Li Y, Mansilla M, et al. (2021) Podocalyxin is a key negative regulator of human endometrial epithelial receptivity for embryo implantation. Hum Reprod 36(5): 1353-1366.

51. Tapilskaya NI, Savicheva AM, Shalepo KV, Budilovskaya OV, Gzgzyan AM, et al. (2023) Local Immune Biomarker Expression Depending on the Uterine Microbiota in Patients with Idiopathic Infertility. Int J Mol Sci 24(8): 7572.

52. (2024) TGF beta 1. In Wikipedia.

53. (2024) Fibroblast growth factor 2. In Wikipedia.

54. (2024) Defensin. In Wikipedia.

55. Zhang Z, Li T, Xu L, Wang Q, Li H, et al. (2021) Extracellular superoxide produced by Enterococcus faecalis reduces endometrial receptivity via inflammatory injury. Am J Reprod Immunol 86(4): e13453.

56. Isbilen E, Kulaksizoglu S, Kirmizioglu M, Komurcu OK, Tabur S (2022) Role of prolidase activity and oxidative stress biomarkers in unexplained infertility. Int J Gynaecol Obstet 156(3): 430-435.

57. Saxtorph MH, Hallager T, Persson G, Petersen KB, Eriksen JO, et al. (2020) Assessing endometrial receptivity after recurrent implantation failure: a prospective controlled cohort study. Reprod Biomed Online 41(6): 998-1006.

58. Chen P, Yang M, Wang Y, Guo Y, Liu Y, et al. (2022) Aging endometrium in young women: molecular classification of endometrial aging-based markers in women younger than 35 years with recurrent implantation failure. J Assist Reprod Genet 39(9): 2143-2151.

59. (2024) CDKN1A. In Wikipedia.

60. Su Y, Zhang JJ, He JL, Liu XQ, Chen XM, et al. (2020) Endometrial autophagy is essential for embryo implantation during early pregnancy. J Mol Med (Berl 98(4): 555-567.

61. Wang Z, Nie K, Su H, Tang Y, Wang H, et al. (2021) Berberine improves ovulation and endometrial receptivity in polycystic ovary syndrome. Phytomedicine 91: 153654.

62. Liu Y, Yao Y, Sun H, Zhao J, Li H, et al. (2022) Lipid metabolism-related genes as biomarkers and therapeutic targets reveal endometrial receptivity and immune microenvironment in women with reproductive dysfunction. J Assist Reprod Genet 39(9): 2179-2190.

63. Bergant G, Abdulkhalikova D, Sustarsic A, Peterlin B, Bokal EV, et al. Expression of Markers of Endometrial Receptivity in Obese Infertile PCOS Women before and after the Weight Loss Program-A Preliminary Study. Cells 12(1): 164.

64. Huang X, Xiao L, Long Y, Pei T, Luo B, et al. (2022) Comparative Proteomic Analysis Reveals Metformin Improves the Expression of Biomarkers of Endometrial Receptivity in Infertile Women with Minimal/Mild Endometriosis. Reprod Sci 29(9): 2593-2606.

65. Tan X, Ding J, Pu D, Wu J (2021) Anti-phospholipid antibody may reduce endometrial receptivity during the window of embryo implantation. J Gynecol Obstet Hum Reprod 50(6): 101912.

66. Huang X, Xiao L, Long Y, Pei T, Luo B, et al. (2022) Correction to: Comparative Proteomic Analysis Reveals Metformin Improves the Expression of Biomarkers of Endometrial Receptivity in Infertile Women with Minimal/Mild Endometriosis. Erratum for Reprod Sci 29(12): 3532.