POSTTRANSLATIONAL MODIFICATIONS AND ITS ROLE IN HABITUAL MISCARRIAGE: PROGNOSIS, DIAGNOSIS AND NEW APPROACHES TO THERAPY

Ziganshin A.M.[1], Dikke G.B.[2], Musina A.M.[1], Bayanova R.R.[1], Frolov A.L.[3]

[1] Bashkir State Medical University (3 Lenin St., 450008 Ufa, Russian Federation)
[2] F.I. Inozemtsev Academy of Medical Education (22 Moskovsky Ave., St. Petersburg 190013, Russian Federation)
[3] Republican Clinical University of the Ministry of Health of the Republic of Belarus, Perinatal Center (Batyrskaya str., 31, Ufa 450106, Russian Federation)

Corresponding author:
Aidar M. Ziganshin,
e-mail: zigaidar@yandex.ru

RESUME

The prevalence of recurrent pregnancy loss (RPL) in women ranges from 1 to 5 %. Among the known causes of RPL, the role of post-translational protein modifications (PTMP) has been studied in recent years. These are protein structure transformations that complete the formation of their molecule or participate in the regulation of the functions of this molecule, catalyzed by specific enzymes.
The aim. *To assess the role of PTMP in the pathogenesis of RPL, as well as to determine potential biomarkers and therapeutic targets of RPL.*
Material. *A search of publications by keywords was conducted in the electronic databases PubMed/MEDLINE and Google Scholar, published before December 2024.*
Results. *PTMB plays an important role in the processes of trophoblast invasion, endometrial decidualization and embryo implantation, which makes them significant for understanding reproductive dysfunction. The use of mass spectrometry to study PTMB opens up new possibilities for the diagnosis and prognosis of RPL. Epigenetic therapy of RPL demonstrates efficacy and a lower probability of side effects compared to traditional methods. Despite significant prospects, research in this area is accompanied by difficulties associated with heterogeneity of terminology and ethical issues.*
Conclusion. *PTMB in the context of RPL can contribute to the improvement of diagnostic and therapeutic strategies in reproductive medicine. Further development of methodologies for studying PTMB is needed.*

Key words: *recurrent miscarriage; posttranslational protein modifications; mass spectrometry, epigenetic therapy*

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ПОСТТРАНСЛЯЦИОННЫЕ МОДИФИКАЦИИ БЕЛКОВ И ИХ РОЛЬ В ПРИВЫЧНОМ НЕВЫНАШИВАНИИ БЕРЕМЕННОСТИ: ПРОГНОЗЫ И ДИАГНОСТИКА

Зиганшин А.М.[1], Дикке Г.Б.[2], Мусина А.М.[1], Баянова Р.Р.[1], Фролов А.Л.[3]

[1] ФГБОУ ВО «Башкирский государственный медицинский университет» (450008, г. Уфа, ул. Ленина, д. 3, Россия)
[2] ЧОУ ДПО «Академия медицинского образования имени Ф.И. Иноземцева» (190013, г. Санкт-Петербург, ул. Московский пр-кт, д. 22 литер М, Россия)
[3] ГБУЗ МЗ РБ «Республиканский клинический перинатальный центр» (450106, г. Уфа, ул. Батырская, д. 31, Россия)

Автор, ответственный за переписку: **Зиганшин Айдар Миндиярович,** e-mail: zigaidar@yandex.ru

РЕЗЮМЕ

Распространенность привычного невынашивания беременности (ПНБ) у женщин по всему миру составляет от 1 до 5 %. Среди известных причин ПНБ в последние годы изучается роль посттрансляционных модификаций белков (ПТМБ) – превращения структуры белков, завершающие формирование их молекулы или участвующие в регуляции функций этой молекулы, и катализируемые специфическими ферментами.

Цель. *Оценить роль ПТМБ в патогенезе ПНБ, а также определить потенциальные биомаркеры и терапевтические мишени ПНБ.*

Материал. *Проведен поиск публикаций по ключевым словам в электронных базах данных PubMed/MEDLINE и Google Scholar, опубликованных до декабря 2024 г.*

Результаты. *ПТМБ играют важную роль в процессах инвазии трофобласта, децидуализации эндометрия и имплантации эмбриона, что делает их значимыми для понимания нарушений репродуктивной функции. Использование масс-спектрометрии для исследования ПТМБ открывает новые возможности для диагностики и прогноза ПНБ. Эпигенетическая терапия ПНБ демонстрирует эффективность и меньшую вероятность побочных эффектов по сравнению с традиционными методами. Несмотря на значимые перспективы, исследования в этой области сопровождаются трудностями, связанными с неоднородностью терминологии и этическими вопросами.*

Заключение. *ПТМБ в контексте ПНБ может способствовать улучшению диагностических и терапевтических стратегий в репродуктивной медицине. Необходимы дальнейшие разработки методологий для изучения ПТМБ.*

Ключевые слова: *привычное невынашивание беременности, посттрансляционные модификации белков; масс-спектрометрия, эпигенетическая терапия*

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INTRODUCTION

Recurrent pregnancy loss (RPL), or habitual miscarriage, is a woman's history of two or more clinical pregnancy losses before 22 weeks of gestation, according to Russian clinical guidelines [1]. The European Society of Human Reproduction and Embryology (ESHRE) defines RPL as two or more pregnancy losses before 24 weeks [2]. The prevalence of RPL ranges from 1 % to 5 %, but its true incidence is difficult to estimate due to heterogeneity in definitions and criteria [3]. The etiology of RPL remains unresolved and includes many modifiable and non-modifiable factors. Even after a thorough assessment of the etiology and risk factors for RPL, up to 75 % of cases remain unexplained [3].

Achieving satisfactory pregnancy outcomes through available treatment methods remains a complex and challenging process, particularly in cases of unexplained recurrent miscarriages. Furthermore, RPL causes significant suffering to many families and results in serious socio-economic losses [3].

In recent years, the importance of post-translational modifications (PTMs) of proteins in various physiological and pathological processes has become known [4]. PTMs of proteins are transformations of protein structure catalyzed by specific enzymes that complete the formation of a protein molecule or participate in the regulation of its functions. PTMs of proteins are the process of adding or removing chemical groups from amino acid residues in a polypeptide chain, which increases the functional diversity of proteins and is defined as modification of amino acid side chains after protein synthesis [5]. These modifications influence cell growth and differentiation, participate in maintaining the integrity of the cell cycle and apoptosis, and regulate numerous biological processes. Research into PTMs of proteins is actively conducted in the context of oncological, cardiovascular, and metabolic diseases [6].

Pregnancy is the result of the interplay of multiple regulatory systems, and an abnormality at any of these stages can lead to pregnancy termination. Recent genetic studies have confirmed that the frequency of aneuploidy in RPL and sporadic miscarriages is not statistically significantly different. This indicates that the underlying mechanisms of sporadic miscarriage and RPL may be analogous [7].

PTMs of proteins play an important role in regulating embryo implantation, embryonic development, placental formation, and the maternal and fetal immune response, which is crucial for understanding the pathological mechanisms of RPL [5]. Studying PTMs of proteins may shed light on the role of modified proteins in the RPL pathogenesis and facilitate the development of more effective diagnostic/prognostic tools and more targeted treatments. However, the number of studies examining the role of PTMs of proteins in RPL remains limited to date, and the data obtained are not systematized, which serves as the basis for this review.

THE AIM OF THE REVIEW

To assess the role of PTMs of proteins in the pathogenesis of RPL, as well as to determine potential biomarkers and therapeutic targets of RPL.

MATERIALS AND METHODS

Publication search strategy

We searched PubMed/MEDLINE and Google Scholar for publications. The search strategy included the following keywords and their combinations in Russian and English: привычное невынашивание беременности; постранслящионные модификации белков; гликозилирование; фосфорилирование; сумоилирование; убиквитинирование; метилирование; ацетилирование; пальмитоилирование; (miscarriage; pregnancy loss; posttranslational modifications; glycosylation; phosphorylation; sumoylation; ubiquitination; methylation; acetylation; palmitoylation). The search was performed among studies published up to December 2024.

All authors independently screened the titles and abstracts of the identified articles. When relevant studies were identified, the full text of the corresponding article was retrieved. Duplicates and incomplete versions of articles were excluded. Full-text versions of articles were assessed for compliance with the following inclusion criteria: the article was published in English or Russian between 2014 and 2024; it was published in a peer-reviewed scientific journal; the article is a literature review, experimental study, or clinical trial that contains the specified keywords; the article describes the role of the specific PTMs of proteins in reproductive function.

RESULTS AND DISCUSSION

PTMs of proteins are a non-template process and are responsible for the formation of multiple protein forms. Approximately 400 types of PTMs of proteins reactions are known, each affecting a limited range of proteins. It has now been established that protein synthesis does not end at the ribosomes; afterward, the next stage of protein transformation begins is processing, or PTMs of proteins. After a peptide (protein) chain has been released from the ribosome, it assumes its biologically active form, i.e., folds in a specific manner. However, this process is not always possible until the newly formed polypeptide chain undergoes processing, which is the final stage of protein biosynthesis. PTMs of proteins occur in the rough endoplasmic

reticulum, where ribosomes are located on its surface, as well as in the Golgi complex. The functional significance of PTMs of proteins reactions is that they enable the activity of a protein or entire groups of proteins to be controlled in response to the changing cellular requirements. The specificity of these interactions opens up the possibility of selectively influencing the processing and, consequently, the function of specific proteins [8].

Advances in mass spectrometry over the past two decades have significantly expanded the number of known PTMs of proteins in biology, and as instrumentation continues to improve, this number is likely to continue to grow. The following is a description of some PTMs in proteins that have already been studied in detail.

GLYCOSYLATION

Glycosylation is a common and complex process in PTMs of proteins. It is based on a mechanism through which certain amino acid residues in proteins are linked to carbohydrates, forming glycosidic bonds under the influence of enzymes called glycosyltransferases, which mediate various biological functions and modulate protein activity [9].

There are several types of glycosylation, including N-, O-, C-, S-, and P-glycosylation. Each type of glycosylation has its own unique characteristics and functions, and can influence the stability, longevity and functional activity of proteins. These processes are also important for cell adhesion and interactions between cells in the immune system.

O-Glucosamination (O-GlcNAcylation) is a common PTM of proteins that significantly influences the functional properties of proteins. This process is regulated by two important enzymes: O-GlcNAc transferase (OGT), which adds the glucosamine residue, and O-Glucosaminase (OGA), which removes it. O-Glucosamination plays a crucial role in maintaining genomic stability, epigenetic regulation, regulation of protein synthesis and degradation, metabolic pathways, signaling cascades, and apoptosis. Research suggests that OGT may be associated with various pathologies, including adverse changes in fetal nervous system development [10]. This emphasizes the significance of O-Glucosaminylation in understanding not only typical physiological processes, but also the pathological mechanisms underlying numerous diseases.

OGA expression is essential for embryonic and fetal viability. In the study conducted by de Lima Castro M. et al. (2023), an increase in perinatal mortality was observed among experimental mice with the OGA gene deleted. This phenomenon is likely associated with the dynamics of circulating glucose levels and a decrease in glycogen storage in the liver, indicating the sensitivity of glycosylation to the metabolic state of the body [11].

Hypoxia that occurs during placental development is one of the main causes of various pregnancy complications, vascular placental abnormalities, and miscarriage [12]. Increased O-Glucosamination caused by OGA loss has a significant impact on placental function. This can lead to weakened vascularization in the area between the mother's blood vessels and the placenta [13]. Ruane P.T. et al. suggested that O-Glucosamination promotes accelerated trophoblast differentiation [14]. Additionally, Liu J. et al. suggest that cystathionine γ-lyase (CSE) and nuclear receptor subfamily 4, group A, member 3 (NR4A3) are activated by high levels of O-Glucosamination; this activation suppresses trophoblast syncytialization and leads to H_2S production [15].

O-Glucosamination serves as a key regulator of transcriptional activity. Aquaporin-3 (AQP3) expression is enhanced by O-Glucosamination, which interacts with transcription factor protein 1 (SP1). A decrease in human placental trophoblast migration is observed with AQP3 inhibition. One of the targets of O-Glucosamination at Ser40 is a histone H_2A variant, which is essential for trophoblast stem cell differentiation [16]. Hypoxia-inducible factor-1 alpha (HIF-1α) activity, which is crucial for placental vascular development, increases with decreased O-Glucosamination levels [17].

Placental growth and glucose and amino acid transport are primarily controlled by two key protein kinases: AMP – activated protein kinase (AMPK) and the mammalian target of rapamycin (mTOR). An important enzyme in the hexosamine biosynthetic pathway (HBP) is fructose-6-phosphate amidotransferase, also known as glutamine: fructose-6-phosphate amidotransferase (GFAT). This enzyme plays a critical role in controlling trophoblast proliferation via the PI3K/Akt/mTOR signaling pathway, which is necessary for maintaining optimal nutrient balance in the placenta [18].

AMPK regulates the localization, expression, and selectivity of OGT. It has the ability to directly or indirectly suppress mTOR activity and phosphorylate GFAT protein, reducing its activity. In the human placenta, it shapes the mTOR signaling pathway and maintains OGT levels, thereby influencing trophoblast differentiation [19].

Poor embryo quality and endometrial dysfunction are considered the main factors leading to RPL. During oocyte maturation, OGA expression increases and O-Glucosamination levels decrease. O-Glucosamination improves cell proliferative, migratory, and invasive abilities, as well as cell adhesion, which facilitates successful embryo implantation by regulating endometrial receptivity [20].

There is evidence that O-Glucosamination may be associated with the development of chronic inflammatory processes in the placenta. Dysfunction in the regulation of the hexosamine signaling pathway (HSP) and O-Glucosamination may be the mechanism responsible for the embryotoxic effects caused by hyperglycemia [21]. Therefore, reduced glycosylation

of the prominin-1 protein may also negatively impact the ability of the blastocyst to implant in the uterus.

N-glycosylation. Within the context of N-glycosylation, the most active enzymes are N-acetylglucosamine transferases V (GnT-V) and III (GnTIII). Although the role of GnT-III during pregnancy in healthy women and RPL has not been fully elucidated, GnT-V is believed to influence trophoblast invasion by altering β1,6-GlcNAc levels on integrin α5β1. N-glycosylation, a key regulator of intercellular communication, plays an important role in the normal function of the immune system.

Yu M. et al. found that N-glycosylation influences endometrial receptivity [22]. Some studies have also shown that alpha-1,3-mannosyltransferase (ALG3) and components of the oligomeric Golgi complex 5 (COG5) may lead to congenital disorders of glycosylation [23, 24]. These glycosylation mechanisms may be closely linked to the pathogenesis of RPL.

Protein O-fucosylation. Protein O-fucosyltransferase 1 (poFUT1) is a key enzyme that catalyzes the O-fucosylation of proteins that affect embryo implantation. This enzyme regulates cyclin protein production and activates the MAPK and PI3K/Akt signaling pathways, which promotes trophoblast proliferation [25]. Epiregulin enhances poFUT1 expression by increasing the O-fucosylation of plasminogen activator and activating the PI3K/Akt signaling pathway. This, in turn, promotes the epithelial-mesenchymal transition (EMT) of the trophoblast and improves embryo implantation.

PoFUT1 also positively correlates with O-fucosylation of the Notch1 molecule within certain limits. A decrease in poFUT1 activity is accompanied by a decrease in Notch1 activity, which leads to a decrease in the transcriptional activity of prolactin and insulin-like growth factor binding protein-1 (IGFBP1), as well as to impaired decidualization of endometrial stromal cells [26]. Fucosyltransferase IV (FUT4) plays an important role in the biosynthesis of α1,3-fucosylated glycans carried by glycoproteins. The miR-200c/FUT4/α-1,3-fucosylation (LeY)/CD44/Wnt/β-catenin signaling cascade significantly contributes to uterine receptivity. MiR-200c inhibits α-1,3-fucosylation, while LeY activates CD44 by interacting with FUT4, leading to inhibition of the Wnt/β-catenin signaling pathway and decreased endometrial receptivity [27].

Thus, protein glycosylation significantly influences the functions of trophoblast, decidual stromal cells, and decidual immune cells, playing an important role in maintaining immunological tolerance during pregnancy. Therefore, studies of protein glycosylation processes are of significant interest in understanding the immunopathogenesis of RPL.

PHOSPHORYLATION

Phosphorylation is the most common modification of PTMs in proteins. It has been suggested that abnormal placentation or impaired trophoblast invasion may be the main causes of RPL in women [28]. Decreased endometrial plasticity is also associated with RPL. Patients with RPL exhibit a significant decrease in the expression levels of exocyst complex component 2 (SEC5) in decidual macrophages, which inhibits M2 polarization and STAT6 phosphorylation [29]. M2 cells, in addition to reducing the inflammatory response, play an important role in tissue repair and maintaining fetal immune stability throughout pregnancy.

Extravillous trophoblasts (EVT) are actively stimulated to proliferate and invade when decidual macrophages polarize into M2 *in vitro*. Formyl peptide receptor 2 (FPR2) can regulate trophoblast functions through the PI3K/AKT signaling pathway [30]. The ability of EVTs and primary cell lines to migrate and invade is significantly reduced by high levels of ezrin protein and its activated phosphorylated form. According to Gao L. et al., increased levels of trophoblast-produced lactic acid in the decidua of women with RPL can induce macrophage polarization in M1, which occurs via the HIF-1α/SRC/LDHA pathway. RPL is also associated with abnormal β3-integrin expression [31].

Cai X. et al. demonstrated that phosphorylated Nur77 protein controls endometrial receptivity via the β3-integrin/FAK pathway [32]. STAT3 hyperphosphorylation is observed in RPL, which inhibits the proliferation of regulatory T lymphocytes (Treg) and reduces the secretion of transforming growth factor (TGF)-1β and interleukin (IL)-10 [33]. High levels of SPARCL protein are responsible for decreased ERK phosphorylation and Fos and Jun expression, which suppresses EVTs migration and invasion [34]. By suppressing STAT3 phosphorylation, insufficient indoleamine 2,3-dioxygenase (IDO) activity can impair trophoblast proliferation and migration, ultimately leading to RPL.

The activity of various signaling pathways is regulated through modifications in phosphorylation. Due to the significant impact of protein phosphorylation on most vital processes, it is crucial to develop methods for targeting specific phosphorylated molecules. Continued developments in phosphorylation technology and research are opening up new opportunities for the diagnosis and treatment of RPL.

SUMOYLATION

Sumoylation involves the addition of small ubiquitin-like modifiers (SUMO) to specific targets in a covalent and reversible manner. Four different SUMO subtypes have been identified. Notably, they appear to exhibit spatial specificity: SUMO-1 is localized to the nuclear envelope of oocytes, while SUMO-2 and SUMO-3 are located intranuclearly. During meiosis, SUMO-1 is predominantly localized

to the spindle poles, while SUMO-2 and SUMO-3 are concentrated in the centrosome [35]. These observations suggest a specific role for SUMO in oocyte development.

Sumoylation is regulated by the sentinel-specific protease (SENP) family [35]. Septin2 can be modified by SUMO; septins are required for chromosome congression and meiotic progression [35].

Jones K.T. noted that if securin remained unchanged, meiotic progression via the anaphase-promoting complex (APC) would be hampered [36]. In transgenic mouse models, knockdown of the desumoylation enzymes SENP1 and SENP2 results in pregnancies with nonviable embryos, as well as abnormal placental trophoblast differentiation and cell proliferation [37]. Yu H.I. et al. found that SENP2-mediated extraembryonic and embryonic development requires SUMO2/3 [38]. A study conducted by Huang C.J. et al. showed that SENP7 deficiency results in progressive embryonic transformation, with these embryos exhibiting varying degrees of DNA damage and difficulty in progressing to the blastocyst stage [39]. Sumoylation is essential for oocyte maturation and is critical for controlling the activity of cytoskeletal proteins [40], but its specific molecular mechanism remains unclear. Studies have shown that the absence of UCB9 in chicken cells leads to the cumulative accumulation of chromosomal abnormalities [41]. Nacerddine K. et al. noted that embryos lacking UCB9 died due to chromosome segregation defects [42].

Polo-like kinase 1 (PLK1), a member of the polo-like and serine/threonine kinase family, plays an important role in the phosphorylation of serine-137 and threonine-210, which significantly affects the activity of spindle checkpoints. PLK1's function in organizing microtubules and spindle poles is linked to SUMO-1, and its localization and kinetochore function in PLK1 are modified and regulated by SUMO-2/3 [43].

Errors in egg meiosis and cell cycle regulation can predispose eggs to aneuploidy, potentially leading to miscarriage. Thus, SUMO studies provide a new theoretical basis for screening euploid embryos, thereby expanding our understanding of embryonic development.

UBIQUITINATION

Ubiquitination is a post-translational modification process that involves the covalent attachment of ubiquitin to target proteins through a series of enzymatic reactions. Ubiquitination regulates various cellular processes, such as DNA repair, the cell cycle, autophagy, and transcriptional regulation. The enzymes responsible for ubiquitination are divided into three classes: ubiquitin-activating enzymes (E1), ubiquitin-conjugating enzymes (E2), and ubiquitin ligases (E3). These enzymes are responsible for the activation, binding, and ligation of ubiquitin, ensuring the normal course of ubiquitination. E1 proteins activate ubiquitin using ATP, with UBA1 being a member of the E1 family and playing a key role in fertilization. E2 proteins determine the specific mode of ubiquitin chain attachment, while E3s link a target protein to a specific E2, allowing for the selection of the type of protein that is ubiquitinated. Deubiquitinating enzymes (DUBs) include several classes, such as ubiquitin-specific proteases (USPs), ovarian tumor proteases (OTUs), ubiquitin carboxyl-terminal hydrolases (UCHs), Josephin/MPN domain-associated metallopeptidases (JAMMs), and monocyte chemotactic protein-inducible proteins (MCPIPs) [44].

USP25 and USP36 are members of the deubiquitinating enzyme (DUB) family. A study conducted by Ding J. et al. showed that USP25 levels are reduced in placental villous tissue from patients with RPL. The miR-27a-R3p/USP25 axis may influence trophoblast migration and invasion by controlling downstream Wnt signaling [45]. This finding highlights the importance of USP25 in the process of trophoblast invasion and possible mechanisms through which it participates in the RPL pathogenesis.

Furthermore, it has been suggested that another deubiquitinating enzyme, USP2a, may deubiquitinate β-catenin, thus promoting trophoblast invasion through the PI3K/Akt/GSK3β/β-catenin pathway [46]. Collectively, these findings suggest a potential role for USP2a in the regulation of processes essential for normal trophoblast development.

USP36, through its regulation of DHX33–DEAH box RNA helicase, significantly contributes to ribosomal RNA synthesis and mRNA translation. Additionally, it controls nucleolar activity through deubiquitinization of proteins such as nucleophosmin/B23 and fibrillarin. USP36 deficiency at the morula stage induces apoptosis, which, in turn, can cause preimplantation death [47]. These data highlight the importance of DUBs in regulating key processes in reproductive biology and indicate their potential role in reproductive disorders.

E3 ubiquitin ligases are key components of the ubiquitin-protease system, playing an important role in the regulation of invasion and migration of human placental trophoblasts. Such E3 ligases include β-TrCP, Fbxw8, Cullin family proteins, and Cbl family proteins. These ligases are involved in the control of the trophoblast cell cycle, including apoptosis, proliferation, and differentiation. Trophoblast apoptosis is regulated by proteins such as Mcl-1 and MDM2, while proliferation and differentiation depend on Fbxw8 [48]. MDM2 is also required for maintaining cell cycle integrity, highlighting its importance in cellular regulation. Underregulation of SKP2 protein in decidual tissue has been associated with RPL.

Wu L. et al. suggest that MALAT1, an antisense transcript associated with lung adenocarcinoma metastasis, may recruit an E3 ligase to participate

in trophoblast invasion [49]. Furthermore, E3 ubiquitination of MIB2 plays an important role in the control of oocyte meiosis by activating DLL3, which regulates oocyte meiosis through the AKT pathway [50].

Oocyte maturation is closely linked to oocyte ubiquitination processes, which play an important role in the regulation of meiosis. Cyclin B1 is a protein essential for this process. Ubiquitination of Beclin 1, an important regulator of autophagy, promotes Vps34 activity, but the Wiskott–Aldrich syndrome protein (WASP) inhibits Beclin 1 ubiquitination, leading to Vps34 inactivation and inhibition of autophagy [51]. Furthermore, it was recently found that lnc-HZ08 promotes PI3K ubiquitination and degradation, which suppresses trophoblast growth through activation of the PI3K/pAkt/p21/CDK2 pathway, which in turn can lead to miscarriage [52].

Another study found that an OTU deubiquitinase, which has specificity for linear linkages, can activate the linear ubiquitin chain assembly machinery (LUBAC). This mechanism prevents autoubiquitination, which is associated with linear polyubiquitin, and is linked to cell death and type I interferon production [53]. Importantly, many physiological processes in the body, such as autophagy, cellular metabolism, and apoptosis, are mediated by protein ubiquitination.

However, despite significant progress in understanding the role of ubiquitination in various cellular processes, further research is needed to identify the impact of ubiquitination on RPL and potential therapeutic options.

METHYLATION

Protein methylation is an enzymatic process in which methyl groups are transferred to specific amino acid residues in proteins. Common targets for methylation include lysine, arginine, histidine, cysteine, and asparagine. This modification can influence protein function, their interactions with other molecules, and the regulation of various cellular processes.

Lysine residues can undergo different levels of methylation: monomethylation, dimethylation, and trimethylation. Conversely, arginine residues can be monomethylated, as well as symmetrically or asymmetrically dimethylated. These differences in the methylation process influence the functional properties of proteins and their roles in cellular processes.

Protein methylation can be classified into two main categories depending on which proteins are modified: histone and non-histone. Histone methylation is largely associated with the regulation of gene transcription, as it influences chromatin structure and the accessibility of DNA to transcription machinery. Non-histone methylation, in turn, affects all proteins and plays a crucial role in various cellular functions, including transcriptional signaling, the cell cycle, and metabolism.

Non-histone methylation, regulated by protein arginine methyltransferases (PRMTs) and protein lysine methyltransferases, is an important mechanism that influences cellular signaling pathways, protein stability, and mRNA translation, and is involved in many critical biological processes. Arginine methyltransferase 1, 3, and 6 have been identified within the female reproductive system, indicating their potential role in the regulation of reproductive functions [54].

Studies have shown that PRMT1 plays a crucial role in repairing damaged DNA, and mouse embryos that have lack of this protein do not survive. The absence of PRMT3 also leads to reduced embryo size and delayed growth and development [55]. These data indicate that arginine methyltransferases play a critical role in normal embryonic development and may be associated with RPL.

Research has shown a link between PRMT3, asymmetric dimethylarginine (ADMA), and nitric oxide (NO), where ADMA, by inhibiting nitric oxide synthase (NOS), is formed as a result of arginine methylation in PRMT proteins [56]. A recent study has shown that women with RPL have reduced NOS activity compared to control subjects. This may adversely affect pregnancy outcomes [56].

Furthermore, methylation of CXC, Rab, and CAAX proteins is completed by a reaction catalyzed by isoprenylcysteine carboxylmethyltransferase (LCMT). Studies have shown that LCMT-deficient embryos die during the middle stages of gestation. However, the precise mechanism through which LCMT affects embryonic development is still not fully understood [57].

Histone methylation plays a crucial role in the regulation of gene expression and maintenance of genomic stability, which in turn significantly affects the embryonic development prior to implantation. The methylation of histones such as H3K4, H3K27, H3K9, and H3K36 is associated with important processes that occur during the early stages of mammalian embryonic development [58]. Methylation of the H3K9 residue, in particular, is involved in maintaining genomic stability and in repressing the expression of cell type-specific genes. Abnormalities in H3K9me3 reprogramming may lead to impaired genomic activation in zygotes, highlighting its importance for proper embryonic development [59]. G9a, a key enzyme responsible for H3K9 methylation, plays an important role in processes associated with hypoxia, cancer, and early embryonic development. Studies have shown that G9aMT expression and methylated histone H3-K9 levels were significantly lower in fresh endometrial decidual tissue from women with RPL compared to average levels [60]. However, it remains unclear how exactly H3-K9 methylation levels and G9a activity relate to RPL. Understanding this interaction may be critical for developing new approaches to diagnosing and treating conditions associated with RPL. Further research in this area is required

to clarify the precise mechanisms of histone methylation in relation to reproductive biology and its influence on pregnancy outcomes.

DNA methylation has been suggested as a potential mechanism that contributes to RPL development [61]. However, histone H3K27 methylation also plays a significant role in DNA methylation-independent processes associated with imprinting. Specifically, the H3K27me3 modification modulates gene expression by suppressing its activity and influencing cellular differentiation, as well as contributing to the development and progression of various diseases.

During embryonic development, dynamic changes in H3K27me3 may indicate the choice of cellular differentiation pathways. This marker is also involved in the regulation of bivalent genes, maintaining the pluripotency of embryonic stem cells. H3K27me3 has been identified as an imprinting marker, XIST, which plays an important role in X-chromosome inactivation [62].

Interestingly, loss of H19/IGF2 imprinting has been observed in the decidua of patients with RPL. This discrepancy appears to be closely linked to a deficiency in the inhibitory histone marker H3K27me3 [63]. This methylation imbalance may indicate a potential predisposition to RPL.

Fatima N. et al. used a structural equation model to substantiate the correlation between methyltransferases and embryonic development in patients with RPL [64]. Such studies highlight the complexity of the molecular mechanisms associated with methylation and demonstrate that methylated protein modifications may have unpredictable hidden implications for RPL therapy. This opens up new perspectives for developing treatment approaches and understanding the causes of RPL.

ACETYLATION

Acetylation is the process of transferring acetyl groups to the lysine residues or N-terminus of a protein. This mechanism is particularly important in regulating histone activity, where histone acetyltransferases and deacetylases (HDACs) maintain cellular homeostasis. Histone acetylation is associated with the activation of gene expression, while deacetylation often leads to gene silencing.

Studies have shown that inhibiting HDAC enzyme activity with trichostatin A may limit trophoblast invasion. Moreover, trichostatin A also promotes decidualization of endometrial stromal cells, which is essential for maintaining normal reproductive function [65].

Histone acetylation is a crucial process that regulates cytokine expression. ATP citrate lyase (ACLY) is an important regulator of this process. A study conducted by Chen X. et al. showed a significant decrease in ACLY levels in the chorionic villi of placentas in patients with RPL compared to the control group. This decrease leads to inhibition of histone acetylation and causes an imbalance in the ratio of M2 and M1 macrophages, which may negatively affect immune responses in the placenta [66].

Additionally, Wang P. et al. noted that patients with RPL have decreased HDAC levels in chorionic villi and increased transcriptional activity of the transcription factor EB (TFEB). Increased TFEB activity leads to excessive activation of autophagy, which may suppress trophoblast growth [67]. This suggests that disturbances in acetylation mechanisms and their regulators may contribute to the development of RPL.

Class I HDACs (histone acetyl-independent deacetylases), including HDAC1, HDAC2, HDAC3, and HDAC8, play important roles in preimplantation embryonic development and other vital processes such as checkpoint activation and DNA repair. These enzymes are involved in maintaining DNA methylation homeostasis and regulating gene expression, which is critical for normal embryonic development. HDAC1 and HDAC2 promote genome-wide DNA methylation; embryos with impaired function of these HDACs are destined to die at the morula stage [68]. This underscores their importance in early embryonic development, when proper methylation is essential for normal cellular differentiation and genomic stability.

HDAC3 deficiency also has consequences: as shown in a study conducted by Bhaskara S. et al., its deficiency leads to prolongation of the S-phase of the cell cycle and DNA damage [69]. This suggests that HDAC3 has a critical role in cell cycle control and maintaining genomic integrity in cells.

Furthermore, a study conducted by Kim T.H. et al. noted decreased HDAC3 expression in the uterus of patients with endometriosis-induced infertility [70]. This opens up new perspectives for understanding the connection between changes in HDAC activity and reproductive disorders, which could lead to significant insights into the development of new therapeutic approaches.

Overall, these data highlight the critical importance of class I HDACs for embryonic development and reproductive function, as well as the need for further study of their role in RPL.

Non-histone acetylation is an important process in which acetyl groups are attached to lysine residues of proteins other than histones. This process is carried out by lysine acetyltransferases (KATs) and lysine deacetylases (KDACs), which play a key role in regulating protein functionality in cells [71]. KAT families can be classified into three main groups: GCN5, CBP/p300, and MYST, each with its own specific functions and mechanisms of action. These enzymes are involved in the acetylation of various proteins, including transcription factors, enzymes responsible for metabolism, and proteins

involved in cell signaling, thereby influencing a variety of cellular processes. Lysine deacetylases (KDACs), on the other hand, are divided into two main classes: Zn^{2+}-dependent and NAD^{+}-dependent sirtuin deacetylases. Zn^{2+}-dependent KDACs, such as HDAC1 and HDAC2, play a role in deacetylation, leading to the suppression of gene expression, while NAD^{+}-dependent sirtuin deacetylases (e.g., SIRT1) regulate various cellular processes, including metabolism and aging. Although KAT and KDAC have been studied, many aspects of their interactions with specific substrates and the mechanisms they use to regulate protein functionality remain uncertain [71]. Further research in this area is expected to better understand the role of non-histone acetylation in various physiological and pathophysiological processes, including RPL.

The discovery of α-tubulin acetylation and the transcription factor p53 has been an important step in our understanding of the role of non-histone acetylation in cellular processes. For example, post-ovulatory oocyte aging is associated with abnormal α-tubulin acetylation, which may impact female fertility [72]. Studies have shown that defects in α-tubulin acetylation mechanisms lead to impaired meiotic spindle assembly in oocytes and sperm, thereby reducing fertility and contributing to abnormalities in the morphology of female and male gametes. These findings suggest specific aspects that are relevant to germ cells [73].

KAT6A, a lysine acetyltransferase, plays an important role in regulating the expression of a number of developmental genes, including genes responsible for the formation of the heart and nervous system [74]. Abnormal protein acetylation has also been observed in infants with congenital heart defects [75].

Studies of mitochondrial deacetylases, such as Sirt3, have shown that they can modulate oxidative stress in blood vessels and exert a protective effect on endothelial function [76].

Therefore, numerous studies highlight that acetylation significantly influences embryonic development through transcription, translation, and protein interaction mechanisms. These processes may play a crucial role in RPL development, providing new theoretical foundations and approaches for further research in this area. Understanding the impact of non-histone acetylation on reproductive health creates new opportunities for the development of therapeutic approaches aimed at improving pregnancy outcomes and offspring health.

PALMITOYLATION

Palmitoylation, or S-palmitoylation, is an important PTM in proteins that results in the addition of palmitic acid to proteins. This process is mediated by enzymes such as palmitoyltransferases and depalmitoylases. Palmitoylation influences the subcellular localization, stability, and functional activity of proteins, which in turn may play a critical role in cellular signaling and homeostasis [77]. This modification is particularly important for regulating cellular activity, including cell differentiation and apoptosis. Understanding the mechanism of palmitoylation may aid in the development of new therapeutic approaches for the treatment of these pathologies.

Interestingly, some studies have found a direct correlation between RPL and the palmitoylation process [78-80]. This may open up new horizons for studying the mechanisms underlying RPL and facilitating the search for new solutions.

THE POTENTIAL OF PTMs IN PROTEINS IN THE PREDICTION OF RECURRENT PREGNANCY LOSS

Omics technologies, including genomics, epigenomics, transcriptomics, proteomics, and metabolomics, provide a holistic and comprehensive approach to studying biological systems. Proteomics, which includes the study of PTMs in proteins, the subject of this review, is the systematic and comprehensive study of the types, structures, and functions of proteins expressed within cells or tissues. Proteomic methods used to analyze PTMs in proteins can be divided into antibody-based and mass spectrometry (MS)-based analyses. Protein microarrays, immunohistochemistry, and Western blotting have demonstrated widespread popularity as effective tools for antibody-based analysis of PTMs in proteins. MS-based analysis is also a powerful method for studying PTMs in proteins. However, most proteomic studies investigating RPL have been performed using 2D-DIGE or quantitative methods such as iTRAQ in combination with MS-based approaches. Through the investigation of the relative associations between protein modifications and physiopathological changes, biomarkers for disease diagnosis can be identified. By examining and integrating data obtained through various omics approaches, it is possible to uncover and gain a more profound understanding of underlying molecular interactions and their related longitudinal effects [6, 15].

Although RPL pathogenesis is somewhat understood, specific diagnostic biomarkers and potential regulatory targets for RPL have not been identified. Therefore, researchers have carried out various omics studies using decidual tissue, villous tissue, and blood from patients with RPL. Several studies have so far provided insight into the potential use of PTMs in proteins markers for RPL prognosis.

OPG and Syndecan-1 transcript and protein expression, which were significantly lower in decidual samples from women with RPL than in women with normal pregnancies, are likely markers of RPL [6, 15].

PTMs IN PROTEINS AND NEW APPROACHES TO THE TREATMENT OF RECURRENT PREGNANCY LOSS

Epigenomics, the study of epigenetic modifications at the genome level, is closely related to PTMs in proteins. The most important and well-studied epigenetic modification is DNA methylation. Current epigenetic studies of RPL primarily focus on DNA methylation [54-57]. Epigenetic mechanisms are well-established in fetal and maternal pathophysiology. A new RPL risk gene, *CREB5*, was identified, and Yu M. et al. showed that its hypomethylation increased its expression and caused trophoblast cell dysfunction, leading to recurrent pregnancy loss [22]. Environmental factors that cause *CREB5* hypomethylation should be the focus of further research, as this may suggest approaches to causal interventions to prevent RPL [22]. Epigenetic therapy involves the application of medications or other methods to target these epigenetic processes. Methylation analyses have identified new molecular targets for epigenetic therapy. In therapeutic approaches, dCas9 may serve as a targetable platform for various effector proteins. However, given the current limitations, only a few epigenetic medications have been introduced into clinical practice so far. Further research into the field of epigenetic therapy is therefore required.

CONCLUSION

PTMs in proteins play a significant role in the pathophysiological mechanisms of RPL, affecting processes such as trophoblast invasion and implantation. The use of mass spectrometry has provided new opportunities for their investigation, allowing for the identification of post-translational modifications that can be used as biomarkers. Epigenetic therapies may offer more targeted and fewer adverse effects compared to conventional RPL treatments, emphasizing the need for continued research into the relationship between PTMs in proteins and reproductive health. However, research in this area faces challenges, including inconsistent terminology and ethical concerns, which hinder scientific collaboration and require the development of new methodologies to better understand this field.

Conflicts of interest

No potential conflict of interest relevant to this article reported.

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REFERENCES

1. Rasmark Roepke E, Matthiesen L, Rylance R, Christiansen OB. Is the incidence of recurrent pregnancy loss increasing? A retrospective register-based study in Sweden. *Acta Obstet Gynecol Scand*. 2017; 96(11): 1365-1372. doi: 10.1111/aogs.13210

2. Tetruashvili NK. Habitual miscarriage. *Obstetrics and Gynecology: News. Opinions. Training.* 2017; 4(18): 70-87. (In Russ.). [Тетруашвили Н.К. Привычный выкидыш. *Акушерство и гинекология: Новости. Мнения. Обучения*. 2017; 4(18): 70-87.]. doi: 10.24411/2303-9698-2017-00010

3. Dimitriadis E, Menkhorst E, Saito S, et al. Recurrent pregnancy loss. *Nat Rev Dis Primers.* 2020; 6(1): 98. doi: 10.1038/s41572-020-00228-z

4. Malyshkina AI, Nazarova AO, Batrak NV, et al. Sociomedical characteristics of patients with recurrent miscarriage. *Russian Bulletin of Obstetrician-Gynecologist*. 2014; 14(6): 4348. (In Russ.). [Малышкина А.И., Назарова А.О., Батрак Н.В., и др. Медико-социальная характеристика пациенток с привычным невынашиванием беременности. *Российский вестник акушера-гинеколога*. 2014; 14(6): 4348].

5. Pogorelova TN, Gun'ko VO, Nikashina AA, Alliluev IA, Botasheva TL. Post-translational modifications and differential expression of proteins in placental insufficiency. *Russian Journal of Human Reproduction*. 2016; 22(6): 115119. (In Russ.). [Погорелова Т.Н., Гунько В.О., Никашина А.А., и др. Посттрансляционная модификация и дифференциальная экспрессия белков при плацентарной недостаточности. *Проблемы репродукции*. 2016; 22(6): 115119]. doi: 10.17116/repro2016226115-119

6. Pieroni L, Iavarone F, Olianas A, et al. Enrichments of post-translational modifications in proteomic studies. *J Sep Sci*. 2020; 43(1): 313-336. doi: 10.1002/jssc.201900804

7. Pan S, Chen R. Pathological implication of protein post-translational modifications in cancer. *Mol Aspects Med.* 2022; 86: 101097. doi: 10.1016/j.mam.2022.101097

8. Tur-Torres MH, Garrido-Gimenez C, Alijotas-Reig J. Genetics of recurrent miscarriage and fetal loss. *Best Pract Res Clin Obstet Gynaecol*. 2017; 42: 11-25. doi: 10.1016/j.bpobgyn.2017.03.007

9. Eichler J. Protein glycosylation. *Curr Biol.* 2019; 29(7): R229-R231. doi: 10.1016/j.cub.2019.01.003

10. de Lima Castro M, Dos Passos RR Jr, Justina VD, et al. Physiological and pathological evidence of O-GlcNAcylation regulation during pregnancy related process. *Placenta.* 2023; 141: 43-50. doi: 10.1016/j.placenta.2023.04.018

11. Keembiyehetty C, Love DC, Harwood KR, et al. Conditional knock-out reveals a requirement for O-linked N-Acetylglucosaminase (O-GlcNAcase) in metabolic homeostasis. *J Biol Chem*. 2015; 290(11): 7097-113. doi: 10.1074/jbc.M114.617779

12. Pestrikova TYu, Iurasova EA, Tkachenko VA. Placental insufficiency as the underlying condition of the complications and outcomes of the gestation period. *Russian Bulletin of Obstetrician-Gynecologist*. 2020; 20(1): 515. (In Russ.). [Пестрикова Т.Ю., Юрасова Е.А., Ткаченко В.А. Плацентарная недостаточность как базовая патология осложнений и исходов гестационного периода. *Российский вестник акушера-гинеколога*. 2020; 20(1): 515]. doi: 10.17116/rosakush2020200115

13. Yang YR, Jang HJ, Lee YH, et al. O-GlcNAc cycling enzymes control vascular development of the placenta by modulating the levels of HIF-1α. *Placenta*. 2015; 36(10): 1063-8. doi: 10.1016/j.placenta.2015.08.001

14. Ruane PT, Tan CMJ, Adlam DJ, et al. Protein O-GlcNAcylation promotes trophoblast differentiation at implantation. *Cells*. 2020; 9(10): 2246. doi: 10.3390/cells9102246

15. Liu J, Shao X, Qin W, et al. Quantitative chemoproteomics reveals O-GlcNAcylation of cystathionine γ-lyase (CSE) represses trophoblast syncytialization. *Cell Chem Biol*. 2021; 28(6): 788-801.e5. doi: 10.1016/j.chembiol.2021.01.024

16. Lima VV, Dela Justina V, Dos Passos RR Jr, et al. O-GlcNAc modification during pregnancy: focus on placental environment. *Front Physiol*. 2018; 9: 1263. doi: 10.3389/fphys.2018.01263

17. Lai Y, Fu Z, Gao Y, Ma N, Li L. Hypoxia-inducible factors (HIFs) in early pregnancy: implications for miscarriage†. *Biol Reprod*. 2024; 111(5): 987-999. doi: 10.1093/biolre/ioae139

18. Shi L, Kang K, Wang Z, et al. Glucose Regulates Glucose Transport and Metabolism via mTOR Signaling Pathway in Bovine Placental Trophoblast Cells. *Animals (Basel)*. 2023; 14(1): 40. doi: 10.3390/ani14010040

19. Watkins AJ, Lucas ES, Marfy-Smith S, et al. Maternal nutrition modifies trophoblast giant cell phenotype and fetal growth in mice. *Reproduction*. 2015; 149(6): 563-75. doi: 10.1530/REP-14-0667

20. Han X, Li X, Liu H, et al. OGlcNAc modification influences endometrial receptivity by promoting endome trial cell proliferation, migration and invasion. *Oncol Rep*. 2019; 42(5): 2065-2074. doi: 10.3892/or.2019.7317

21. Wang AJ, Wang A, Hascall V. Detoxification of Hyperglycemia-induced Glucose Toxicity by the Hexosamine Biosynthetic Pathway. *Front Biosci (Landmark Ed)*. 2024; 29(2): 71. doi: 10.31083/j.fbl2902071

22. Yu M, Qin H, Wang H, et al. N-glycosylation of uterine endometrium determines its receptivity. *J Cell Physiol*. 2020; 235(2): 1076-1089. doi: 10.1002/jcp.29022

23. Mortimer NT, Fischer ML, Waring AL, et al. Extracellular matrix protein N-glycosylation mediates immune self-tolerance in Drosophila melanogaster. *Proc Natl Acad Sci USA*. 2021; 118(39): e2017460118. doi: 10.1073/pnas.2017460118

24. Ferrer A, Starosta RT, Ranatunga W, et al. Fetal glycosylation defect due to ALG3 and COG5 variants detected via amniocentesis: complex glycosylation defect with embryonic lethal phenotype. *Mol Genet Metab*. 2020; 131(4): 424-429. doi: 10.1016/j.ymgme.2020.11.003

25. LiuC, Liang X, Wang J, et al. Protein O-fucosyltransferase 1 promotes trophoblast cell proliferation through activation of MAPK and PI3K/Akt signaling pathways. *Biomed Pharmacother*. 2017; 88: 95-101. doi: 10.1016/j.biopha.2017.01.026

26. Yang Y, Zhang D, Qin H, Liu S, Yan Q. poFUT1 promotes endometrial decidualization by enhancing the O-fucosylation of Notch1. *EBioMedicine*. 2019; 44: 563-573. doi: 10.1016/j.ebiom.2019.05.027

27. Zheng Q, Zhang D, Yang Yu, et al. MicroRNA-200c impairs uterine receptivity formation by targeting FUT4 and α1,3-fucosylation. *Cell Death Differ*. 2017; 24(12): 2161-2172. doi: 10.1038/cdd.2017.136

28. Dosiou C, Giudice LC. Natural killer cells in pregnancy and recurrent pregnancy loss: endocrine and immunologic perspectives. *Endocr Rev*. 2005; 26(1): 44-62. doi: 10.1210/er.2003-0021

29. Yang L, Zhang X, Gu Y, et al. SEC5 is involved in M2 polarization of macrophages via the STAT6 pathway, and its dysfunction in decidual macrophages is associated with recurrent spontaneous abortion. *Front Cell Dev Biol*. 2022; 10: 891748. doi: 10.3389/fcell.2022.891748

30. Li A, Li S, Zhang C, et al. FPR2 serves a role in recurrent spontaneous abortion by regulating trophoblast function via the PI3K/AKT signaling pathway. *Mol Med Rep*. 2021; 24(6): 838. doi: 10.3892/mmr.2021.12478

31. Gao L, Xu QH, Ma LN, et al. Trophoblast-derived Lactic Acid Orchestrates Decidual Macrophage Differentiation via SRC/LDHA Signaling in Early Pregnancy. *Int J Biol Sci*. 2022; 18(2): 599-616. doi: 10.7150/ijbs.6781690/cells12050711

32. Cai X, Jiang Y, Cao Z, et al. Mst1-mediated phosphorylation of Nur77 improves the endometrial receptivity in human and mice. *EBioMedicine*. 2023; 88: 104433. doi: 10.1016/j.ebiom.2022.104433

33. Liu B, Wu H, Huang Q, et al. Phosphorylated STAT3 inhibited the proliferation and suppression of decidual Treg cells in unexplained recurrent spontaneous abortion. *Int Immunopharmacol*. 2020; 82: 106337. doi: 10.1016/j.intimp.2020.106337

34. Liu X, Zhao J, Luan X, et al. SPARCL1 impedes trophoblast migration and invasion by down-regulating ERK phosphorylation and AP-1 production and altering EMT-related molecule expression. *Placenta*. 2020; 89: 33-41. doi: 10.1016/j.placenta.2019.10.007

35. Chang HM, Yeh ETH. SUMO: From Bench to Bedside. *Physiol Rev.* 2020; 100(4): 1599-1619. doi: 10.1152/physrev.00025.2019

36. Jones KT. Anaphase-promoting complex control in female mouse meiosis. *Results Probl Cell Differ.* 2011; 53: 343-63. doi: 10.1007/978-3-642-19065-0_15

37. Yamaguchi T, Sharma P, Athanasiou M, et al. Mutation of SENP1/SuPr-2 reveals an essential role for desumoylation in mouse development. *Mol Cell Biol*. 2005; 25(12): 5171-82. doi: 10.1128/MCB.25.12.5171-5182.2005

38. Yu HI, Hsu T, Maruyama EO, et al. The requirement of SUMO2/3 for SENP2 mediated extraembryonic and embryonic development. *Dev Dyn.* 2020; 249(2): 237-244. doi: 10.1002/dvdy.125

39. Huang CJ, WuD, Jiao XF, et al. Maternal SENP7 programs meiosis architecture and embryo survival in mouse. *Biochim Biophys Acta Mol Cell Res.* 2017; 1864(7): 1195-1206. doi: 10.1016/j.bbamcr.2017.03.005

40. Snider NT, Omary MB. Post-translational modifications of intermediate filament proteins: mechanisms and functions. *Nat Rev Mol Cell Biol.* 2014; 15(3): 163-77. doi: 10.1038/nrm3753

41. Hayashi T, Seki M, Maeda D, et al. Ubc9 is essential for viability of higher eukaryotic cells. *Exp Cell Res.* 2002; 280(2): 212-21. doi: 10.1006/excr.2002.5634

42. Nacerddine K, Lehembre F, Bhaumik M, et al. The SUMO pathway is essential for nuclear integrity and chromosome segregation in mice. *Dev Cell.* 2005; 9(6): 769-79. doi: 10.1016/j.devcel.2005.10.007

43. Feitosa WB, Hwang K, Morris PL. Temporal and SUMO-specific SUMOylation contribute to the dynamics of Polo-like kinase 1 (PLK1) and spindle integrity during mouse oocyte meiosis. *Developmental biology*. 2018. 434(2): 278-291. doi: 10.1016/j.ydbio.2017.12.011

44. Wang J, Zhou Q, Ding J, et al. The conceivable functions of protein ubiquitination and deubiquitination in reproduction. *Front Physiol.* 2022; 13: 886261. doi: 10.3389/fphys.2022.886261

45. Ding J, Cheng Y, Zhang Y, et al. The miR-27a-3p/USP25 axis participates in the pathogenesis of recurrent miscarriage by inhibiting trophoblast migration and invasion. *J Cell Physiol*. 2019; 234(11): 19951-19963. doi: 10.1002/jcp.28593

46. Wang J, Ding J, Zhang S, et al. Decreased USP2a expression inhibits trophoblast invasion and associates with recurrent miscarriage. *Front Immunol*. 2021; 12: 717370. doi: 10.3389/fimmu.2021.717370

47. Fraile JM, Campos-Iglesias D, Rodríguez F, et al. Loss of the deubiquitinase USP36 destabilizes the RNA helicase DHX33 and causes preimplantation lethality in mice. *J Biol Chem*. 2018; 293(6): 2183-2194. doi: 10.1074/jbc.M117.788430

48. Feng J, Yin H, Baturuhu Dai Y, et al. Research progress of E3 ubiquitin ligase regulating biological be havior of human placental trophoblast cells. *Front Endocrinol (Lausanne)*. 2023; 14: 1124041. doi: 10.3389/fendo.2023.1124041

49. Wu L, Liu Q, Fan C, et al. MALAT1 recruited the E3 ubiquitin ligase FBXW7 to induce CRY2 ubiquitin-mediated degradation and participated in trophoblast migration and invasion. *J Cell Physiol*. 2021; 236(3): 2169-2177. doi: 10.1002/jcp.30003

50. Chen LJ, Zhang NN, Zhou CX, et al. Gm364 coordinates MIB2/DLL3/Notch2 to regulate female fertility through AKT activation. *Cell Death Differ*. 2022; 29(2): 366-380. doi: 10.1038/s41418-021-00861-5

51. Xia P, Wang S, Du Y, et al. WASH inhibits autophagy through suppression of Beclin 1 ubiquitination. *EMBO J.* 2013; 32(20): 2685-96. doi: 10.1038/emboj.2013.189

52. Xie J, Liang T, Zhao J, et al. Lnc-HZ08 regulates BPDE-induced trophoblast cell dysfunctions by promoting PI3K ubiquitin degradation and is associated with miscarriage. *Cell Biol Toxicol*. 2022; 38(2): 291-310. doi: 10.1007/s10565-021-09606-z

53. Heger K, Wickliffe KE, Ndoja A, et al. OTULIN limits cell death and inflammation by deubiquitinating LUBAC. *Nature.* 2018; 559(7712): 120-124. doi: 10.1038/s41586-018-0256-2

54. WeiH, Mundade R, Lange KC, Lu T. Protein arginine methylation of non-histone proteins and its role in diseases. *Cell Cycle.* 2014; 13(1): 32-41. doi: 10.4161/cc.27353

55. Swiercz R, Cheng D, Kim D, Bedford MT. Ribosomal protein rpS2 is hypomethylated in PRMT3-deficient mice. *J Biol Chem.* 2007; 282(23): 16917-23. doi: 10.1074/jbc.M609778200

56. Hao F, Tang LC, Sun JX, et al. Decreased nitric oxide content mediated by asymmetrical dimethylarginine and protein l-arginine methyltransferase 3 in macrophages induces trophoblast apoptosis: a potential cause of recurrent miscarriage. *Human Reproduction*. 2021; 36(12): 3049-3061. doi: 10.1093/humrep/deab225

57. Bergo MO, Leung GK, Ambroziak P, et al. Isoprenylcysteine carboxyl methyltransferase deficiency in mice. *J Biol Chem*. 2001; 276(8): 5841-5. doi: 10.1074/jbc.C000831200

58. Xu R, Li C, Liu X, Gao S. Insights into epigenetic patterns in mammalian early embryos. *Protein Cell*. 2021; 12(1): 7-28. doi: 10.1007/s13238-020-00757-z

59. Matoba S, Liu Y, Lu F, et al. Embryonic development following somatic cell nuclear transfer impeded by persisting histone methylation. *Cell.* 2014; 159(4): 884-95. doi: 10.1016/j.cell.2014.09.055

60. Fatima N, Ahmed SH, Salhan S, Rehman SM, Kaur J, Owais M, Chauhan SS. Study of methyl transferase (G9aMT) and methylated histone (H3-K9) expressions in unexplained recurrent spontaneous abortion (URSA) and normal early pregnancy. *Mol Hum Reprod.* 2011; 17(11): 693-701. doi: 10.1093/molehr/gar038

61. Zhou Q, Xiong Y, Qu B, et al. DNA Methylation and Recurrent Pregnancy Loss: A Mysterious Com-

pass? *Front Immunol*. 2021; 12: 738962. doi: 10.3389/fimmu.2021.738962

62. Inoue A, Jiang L, Lu F, Suzuki T, Zhang Y. Maternal H3K27me3 controls DNA methylation-independent imprinting. *Nature*. 2017; 547(7664): 419-424. doi: 10.1038/nature23262

63. Wen X, Zhang Q, Zhou L, et al. Intrachromosomal Looping and Histone K27 Methylation Coordinately Regulates the lncRNA H19-Fetal Mitogen IGF2 Imprinting Cluster in the Decidual Microenvironment of Early Pregnancy. *Cells*. 2022; 11(19): 3130. doi: 10.3390/cells11193130

64. Fatima N, Ahmed SH, Chauhan SS, et al. Structural equation modelling analysis determining causal role among methyltransferases, methylation, and apoptosis during human pregnancy and abortion. *Scientific Reports*. 2020; 10(1): 12408. doi: 10.1038/s41598-020-68270-1

65. Sakai N, Maruyama T, Sakurai R, et al. Involvement of histone acetylation in ovarian steroid-induced decidualization of human endometrial stromal cells. *J Biol Chem*. 2003; 278(19): 16675-82. doi: 10.1074/jbc.M211715200

66. Chen X, Song QL, Li ZH, et al. Deletion of ACLY Disrupts Histone Acetylation and IL-10 Secretion in Trophoblasts, Which Inhibits M2 Polarization of Macrophages: A Possible Role in Recurrent Spontaneous Abortion. *Oxid Med Cell Longev*. 2022; 2022: 5216786. doi: 10.1155/2022/5216786

67. Wang P, Zhao C, Zhou H, et al. Dysregulation of Histone Deacetylases Inhibits Trophoblast Growth during Early Placental Development Partially through TFEB-Dependent Autophagy-Lysosomal Pathway. *Int J Mol Sci*. 2023; 24(15): 11899. doi: 10.3390/ijms241511899

68. Zhao P, Wang H, Wang H, et al. Essential roles of HDAC1 and 2 in lineage development and genome-wide DNA methylation during mouse preimplantation development. *Epigenetics*. 2020; 15(4): 369-385. doi: 10.1080/15592294.2019.1669375

69. Bhaskara S, Chyla BJ, Amann JM, et al. Deletion of histone deacetylase 3 reveals critical roles in S phase progression and DNA damage control. *Mol Cell*. 2008; 30(1): 61-72. doi: 10.1016/j.molcel.2008.02.030

70. Kim TH, Yoo JY, Choi KC, et al. Loss of HDAC3 results in nonreceptive endometrium and female infertility. *Sci Transl Med*. 2019; 11(474): eaaf7533. doi: 10.1126/scitranslmed. aaf7533

71. Narita T, Weinert BT, Choudhary C. Functions and mechanisms of non-histone protein acetylation. *Nat Rev Mol Cell Biol*. 2019; 20(3): 156-174. doi: 10.1038/s41580-018-0081-3

72. Lee AR, Thanh Ha L, Kishigami S, Hosoi Y. Abnormal lysine acetylation with postovulatory oocyte aging. *Reprod Med Biol*. 2013; 13(2): 81-86. doi: 10.1007/s12522-013-0172-y

73. Kalebic N, Sorrentino S, Perlas E, et al. αTAT1 is the major α-tubulin acetyltransferase in mice. *Nat Commun*. 2013; 4: 1962. doi: 10.1038/ncomms2962

74. Wiesel-Motiuk N, Assaraf YG. The key roles of the lysine acetyltransferases KAT6A and KAT6B in physiology and pathology. *Drug Resist Updat*. 2020; 53: 100729. doi: 10.1016/j.drup.2020.100729

75. Fukushima A, Zhang L, Huqi A, et al. Acetylation contributes to hypertrophy-caused maturational delay of cardiac energy metabolism. *JCI Insight*. 2018; 3(10): e99239. doi: 10.1172/jci.insight.99239

76. Dikalova AE, Pandey A, Xiao L, et al. Mitochondrial Deacetylase Sirt3 Reduces Vascular Dysfunction and Hypertension While Sirt3 Depletion in Essential Hypertension Is Linked to Vascular Inflammation and Oxidative Stress. *Circ Res*. 2020; 126(4): 439-452. doi: 10.1161/CIRCRESAHA.119.315767

77. Zhou B, Hao Q, Liang Y, Kong E. Protein palmitoylation in cancer: molecular functions and therapeutic potential. *Mol Oncol*. 2023; 17(1): 3-26. doi: 10.1002/1878-0261.13308

78. Wu L, LiJ, Xu HL, et al. IL-7/IL-7R signaling pathway might play a role in recurrent pregnancy losses by increasing inflammatory Th17 cells and decreasing Treg cells. *Am J Reprod Immunol*. 2016; 76(6): 454-464. doi: 10.1111/aji.12588

79. Zhang M, Zhou L, Xu Y, et al. A STAT3 palmitoylation cycle promotes TH17 differentiation and colitis. *Nature*. 2020; 586(7829): 434-439. doi: 10.1038/s41586-020-2799-2

80. Ding J, Yin T, Yan N, et al. FasL on decidual macrophages mediates trophoblast apoptosis: A potential cause of recurrent miscarriage. *Int J Mol Med*. 2019; 43(6): 2376-2386. doi: 10.3892/ijmm.2019.4146

Information about the authors

Aydar M. Ziganshin – Dr. Sc. (Med.), Professor of the Department of Obstetrics and Gynecology with a course at the Institute of Additional Professional Education of the Bashkir State Medical University of the Ministry of Health of the Russian Federation; e-mail: Zigaidar@yandex.ru, https://orcid.org/0000-0001-5474-1080

Galina B. Dikke –Dr. Sc. (Med.), Professor, Department of Obstetrics and Gynecology with a Course of Reproductive Medicine, Inozemtsev Academy of Medical Education, e-mail: galadikke@yandex.ru, https://orcid.org/0000-0001-9524-8962

Aliya M. Musina – 5th year student of the Faculty of Medicine of the Bashkir State Medical University of the Ministry of Health of the Russian Federation, e-mail: aliya_musina_2020@mail.ru, https://orcid.org/0009-0008-6183-9930

Regina R. Bayanova – 5th year student of the Faculty of Medicine of the Bashkir State Medical University of the Ministry of Health of the Russian Federation, e-mail: Reginaradik44@mail.ru, https://orcid.org/0009-0002-2348-3474

Alexey L. Frolov – Cand. Sc. (Med.), Honored Doctor of the Republic of Bashkortostan, distinguished physician of the Russian Federation and the Republic of Bashkortostan, Head of the Operating Unit. No. 2 of the State Budgetary Healthcare Institution of the Republican Clinical Perinatal Center of the Ministry of Health of the Republic of Bashkortostan; e-mail: frolrpc@yandex.ru, https://orcid.org/0009-0004-0678-1503