

NEW INSIGHTS INTO THE PATHOGENESIS OF OSTEOARTHRITIS

I. Momcheva¹, V. Gonchev², M. Valkov¹¹Department of Rheumatology, UMHAT – Burgas²University „Prof. Dr. As. Zlatarov“ – Burgas

Abstract. *The multifactorial pathogenesis of osteoarthritis (OA) and the diversity of its phenotypic manifestations have been subjects of discussion in recent years. This report summarizes the latest data from clinical studies regarding the pathogenesis (PG) of OA. Aspects of PG are examined: the consequences of trauma, the involvement of heat shock proteins, mitochondrial dysfunction, disturbances in the JAK/STAT signaling pathway, low-grade focal inflammation, the role of different macrophage populations, telomere shortening in the aging process, and more. Special attention is given to the involvement of genetic and epigenetic factors in the multifactorial pathogenesis of osteoarthritis. These factors, through mechanisms of gene expression modulation – such as DNA methylation and demethylation, histone modifications, acetylation and deacetylation, and activation of microRNAs – contribute to either silencing or enhanced expression of genes associated with OA. In this way, fine post-transcriptional tuning is maintained, which increases resistance to chronic diseases, including OA. Knowledge of epigenetic control could provide a more in-depth, holistic understanding of the etiology and pathogenesis of OA and offer future directions for its management.*

Key words: osteoarthritis, epigenetics, methylation, DNA, RNA, histones

INTRODUCTION

Osteoarthritis (OA) is the most common chronic joint disease and is becoming an increasingly serious public health issue worldwide. The disease's multifactorial pathogenesis and risk factors can be summarized on four levels.

The first level includes various risk factors and their initiating role in triggering pathogenic mechanisms. The risk factors associated with OA can be classified as intrinsic and individual. Intrinsic risk factors for OA are not influenced by external conditions and include genetics [1], aging [2], and sex [3]. Individual risk factors for OA encompass external conditions determined by lifestyle differences that may lead to the development or progression of the disease. These factors vary and are influenced by different living conditions and habits – diet, physical activity, occupational conditions, comorbidities and their treatment [4], obesity [5], joint loading [6], trauma [7], and metabolic syndrome [8]. The interaction of these factors contributes to the activation of multiple pathophysiological mechanisms leading to the initiation and progression of OA.

The second level involves the effects of potential pathophysiological mechanisms. Among these, low-grade inflammation plays a central role in the pathogenesis of OA. The activation of the inflammatory cascade through the secretion of pro-inflammatory

cytokines [IL-1, IL-6, TNF- α , etc.] and the activation of cellular and humoral factors of the innate immune response contribute to disease progression [9]. During aging and/or in the presence of risk factors such as trauma and excessive mechanical loading, the presence of cellular debris from structurally compromised cells in the joints accelerates the degeneration of joint tissues and the breakdown of the extracellular matrix [10, 11].

The third level includes changes in the intra-articular phenotype at the molecular level. Under the influence of the pathophysiological mechanisms mentioned at the second level, chondrocytes begin to produce catabolic factors such as matrix metalloproteinases [MMPs], ADAMTSs, and A-disintegrins, which are considered responsible for the degradation of the extracellular matrix [12, 13]. As the effects of increased catabolic activity accumulate, a gradual loss of aggrecans occurs, followed by the breakdown of type II collagen and the destruction of the collagen network [14]. Simultaneously, mesenchymal stem cells in the periosteum and/or synovium become activated and undergo chondrogenesis and ossification, explaining the formation of osteophytes [15].

The fourth level of pathogenesis involves histological changes in joint tissues over time. In the early stages of OA, mild cartilage damage leads to dysfunction and is accompanied by hypertrophy of

superficial layer chondrocytes [16]. As the disease progresses, the structural integrity and functional competence of the articular cartilage are progressively impaired. The subchondral bone undergoes remodeling, its trabecular structure changes, and new bone formation occurs [17]. Progressive damage to the menisci [18], degeneration of the ligamentous apparatus [19], and muscle atrophy further contribute to joint destabilization [20]. Low-grade focal inflammation of the synovial membrane and infrapatellar fat pad also plays a role in cartilage degeneration [21]. This joint-specific low-grade inflammation is responsible for the clinical manifestations of OA, accelerating pain in affected joints, swelling, and motor deficits [22]. In advanced disease, the full spectrum of histological changes and clinical manifestations of OA is observed.

Although classified into different levels, these pathogenic processes are interdependent and functionally connected. As a result, they give rise to corresponding clinical symptoms such as pain, swelling, stiffness, limited joint mobility, and muscle weakness. Early symptoms of OA may be localized, sporadic in frequency and occurrence, but their manifestation progresses as the disease advances. The next section of the review discusses four key topics in OA pathogenesis that contribute to a contemporary understanding of the disease and hold potential for the identification of biomarkers for early-stage OA.

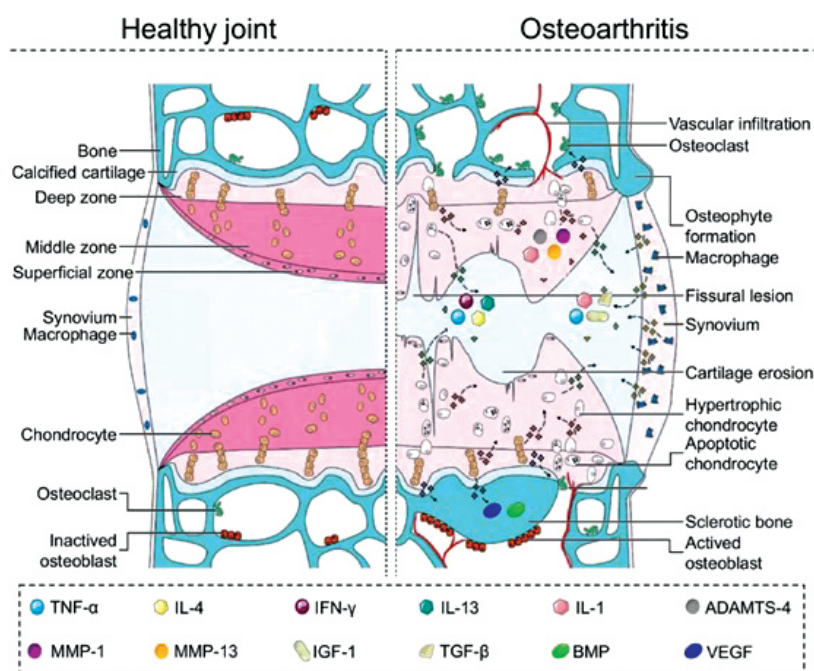
TRAUMA AND OSTEOARTHRITIS

Both in trauma and OA, the initial structural damage occurs in the superficial layers of the cartilage, leading to a loss of proteoglycans in the extracellular matrix and damage to the collagen fiber network [23]. Subsequently, chondrocytes degenerate and decrease in number, which in the early stages appears as minor localized damage in the superficial layers of the articular cartilage. Over time, these lesions may spread to the middle and deep layers of the cartilage [24]. As a result, under load, the cartilage in an osteoarthritic joint lacks the capacity of healthy joint cartilage. The low cellular composition of articular cartilage and its avascular nature further limit its ability to recover after injury [25]. When cartilage damage is inadequately, untimely, or ineffectively treated, the result is osteoarthritis.

HEAT SHOCK PROTEINS AND OSTEOARTHRITIS

Heat shock proteins (HSPs) are molecules produced under environmental stress conditions. They are involved in maintaining the structural integrity and function of cells by supporting cellular proteostasis. HSP families are classified based on their molecular weight, including mainly large HSPs (HSP90, HSP70, HSP60, HSP40) and small HSPs. They function as molecular chaperones, meaning they help large molecules fold and unfold [26]. In addition to their chaperone functions, they play a crucial role in cellular signal transduction and apoptosis regulation. Thus, HSP dysfunction is associated with numerous diseases, including cancer, neurodegenerative, inflammatory, autoimmune, cardiovascular, and other conditions. Extracellular HSPs act as stress warning signals, preparing other cells, particularly immune cells, to limit damage propagation.

A study aimed to identify age-related changes in the levels of HSPs, chemokines, and collagen degradation markers in the blood of patients with osteoarthritis [27]. The study revealed that the involvement of HSPs in OA pathogenesis appears to be a complex and multi-level process. Under the influence of external risk factors, HSP70 acts as an adjuvant



Signaling pathways and cartilage structural changes in OA, 2022, Elsevier

to the immune response, involving the chemokine CXCL17. It contributes to CXCL17-dependent ac-

tivation of dendritic cells and macrophages. This mechanism initiates localized low-grade inflammation. Chondrocytes and osteoblasts subjected to cytokine attack are unable to respond to HSP70 and HSP27 overexpression. As osteoarthritis progresses, the local tissue response advances, where hypertrophic differentiation processes of chondrocytes and subchondral bone cells play a crucial role.

Circulating HSP70 levels were investigated in patients with knee osteoarthritis, showing significantly higher values compared to healthy volunteers [28]. Data analysis from this study revealed that both circulating and synovial HSP70 levels are proportionally associated with the radiographic severity of knee OA. As OA progresses, HSP70 is detected in the chondrocytes of the deeper layers of the knee cartilage, suggesting that a possible mechanism is its altered localization within hyaline cartilage. In this aspect, HSP70 holds potential as a prognostic biomarker for OA monitoring as well as a clinical diagnostic tool.

JAK/STAT SIGNALING PATHWAY AND OA

The Janus kinase (JAK) and signal transducer and activator of transcription (STAT) signaling pathway was discovered more than a quarter of a century ago. As a pivotal mechanism in numerous vital cellular processes, the JAK/STAT pathway serves as a rapid signaling module from the membrane to the nucleus, inducing the expression of various pro-inflammatory and neoplastic mediators [29, 30]. Increasing evidence suggests that dysregulation of the JAK/STAT pathway is pathogenetically linked to various malignancies, autoimmune disorders, and degenerative diseases. The consequences of JAK/STAT pathway activation in OA result in an inflammatory response, inducing the production of pro-inflammatory cytokines such as TNF- α , IL-1 β , and IL-6, which contribute to OA pathogenesis [31].

There is some overlap in the levels of pro-inflammatory cytokines such as TNF- α , IL-1 β , and IL-6 in OA patients [32]. IL-1 β induces the production of IL-6, which activates catabolic processes and inhibits anabolic ones in articular cartilage [33]. Higher levels of IL-6 in serum and synovial fluid are associated with faster OA progression. IL-1 β also induces chondrocyte apoptosis [34]. This occurs because IL-1 β , through IL-6, activates MMPs and ADAMTSs – key mediators of catabolism that disrupt the metabolic balance of chondrocytes and activate cartilage degradation [35]. Numerous studies confirm that IL-6 induces JAK/STAT pathway activation in vari-

ous organs and systems. OA progression is closely linked to the activity of the IL-6/JAK-STAT axis [36]. IL-6 and its soluble receptor [sIL-6R] stimulate cartilage degeneration by promoting the production of ADAMTS-4, ADAMTS-5/11, MMP-1, MMP-3, and MMP-13. These matrix metalloproteinases are also regulated by STAT proteins [37]. Interrupting the IL-6/sIL-6R interaction may reduce MMP expression, limit extracellular matrix degradation, and slow OA progression. Accordingly, decreased serum levels of IL-6 and IL-17 could prevent concomitant low-grade synovial inflammation [38]. TNF- α also has the potential to activate matrix metalloproteinases [MMPs], which accelerate cartilage degeneration [39]. Similar regulatory effects may involve other pro-inflammatory cytokines, such as TNF- α , IL-4, IL-10, IL-13, IL-17, and IL-23. Notably, IL-4 has an anti-inflammatory effect on chondrocytes [40] and is thought to play a role in IL-4 signal regulation through the JAK/STAT pathway. IL-4, via the JAK/STAT pathway, has a chondroprotective role by inhibiting MMP-13 expression [41, 42]. The interaction of IL-10 with its two receptors, IL-10R1 and IL-10R2, can also activate the JAK/STAT pathway [43]. IL-23 may induce synovial inflammation and subchondral bone changes via STAT3 [44].

Apoptotic signaling pathways are also implicated in OA development. Since the JAK/STAT pathway plays a key role in apoptosis regulation, it is closely associated with inflammation and OA progression. Increased phosphorylation of JAK3 and STAT6 has been observed with IL-4 treatment combined with moderate mechanical loading [45].

Macrophages are the most common immune cells in synovial joints and are the main innate immune effector cells that initiate the inflammatory response in OA pathogenesis [46]. STAT1 and STAT3 influence macrophage-mediated inflammatory responses [47]. IL-13 and IL-4 activate M2 macrophages via the JAK2/STAT3 pathway, while IL-4 phosphorylates STAT3 and STAT6, stimulating DNA binding with STAT3. IL-13 induces TYK2 phosphorylation, subsequently activating STAT1 and STAT6 and increasing the DNA-binding activity of STAT1 [48]. IL-6 can activate M1 macrophages via the JAK2/STAT3 pathway [49]. Other studies suggest that an increased M1/M2 macrophage ratio correlates with OA severity. While activated M1 macrophages are associated with high production of pro-inflammatory cytokines such as IL-1, IL-6, IL-12, IL-23, and TNF- α [50], M2 macrophages contribute to lowering these cytokines, activating anti-inflammatory and immunosuppressive responses, and participating in pathogen clearance,

wound healing, tissue remodeling, and immune regulation. Enhancing M2 macrophage activity may counteract pathological OA changes [51].

The JAK/STAT signaling pathway is also involved in cartilage remodeling. Increased STAT3 levels have been associated with joint and muscle dysfunction in OA patients [52]. This suggests that JAK/STAT signaling may influence joint homeostasis, as well as subchondral bone remodeling and muscle loss. A study has demonstrated that activation of the JAK2/STAT3 pathway is linked to reduced type II collagen production in chondrocytes [53]. Activation of the JAK/STAT3 signaling pathway in chondrocytes can be induced by IL-1 β , leading to increased expression of MMPs [54].

The SOCS (Suppressor of Cytokine Signaling) protein family consists of negative feedback inhibitors of cytokine-induced signaling. They exert their function through the JAK/STAT signaling pathway as well. The SOCS family includes seven members: SOCS1, SOCS2, SOCS3, SOCS4, SOCS5, SOCS6, and SOCS7. SOCS1-3 proteins regulate cytokine signaling through negative feedback via the JAK/STAT pathway [55]. SOCS1 and SOCS3 can inhibit the activity of JAK1, JAK2, and TYK2, but not JAK3. SOCS4-7 regulate growth factor receptor signaling [56]. Activated STAT proteins can stimulate the transcription of SOCS genes and promote interactions between SOCS and phosphorylated JAKs. This interaction may lead to JAK/STAT pathway blockade [57].

Leptin has been shown to stimulate the expression of IL-6, MMP-1, MMP-3, and MMP-13 while reducing SOCS3 expression. Suppression of SOCS3 has the potential to induce inflammatory responses and enhance leptin-mediated MMP-3 expression in OA chondrocytes [58]. These findings suggest that SOCS proteins could serve as potential therapeutic targets for OA.

Dysregulation of the JAK/STAT pathway can lead to various pathological changes relevant to many clinical aspects of OA, including chondrocyte survival and apoptosis, regeneration attempts, and extracellular matrix degradation. Evidence suggests that modern antirheumatic drugs modulate JAK/STAT pathway activation [59]. It is crucial to determine the extent to which JAK inhibitors alter MMP expression in chondrocytes. Studies have shown that JAK inhibitors can maintain joint tissue homeostasis by modulating the effects of IL-6. Blocking JAK2 or JAK3 reduces the expression and enzymatic activity of MMP-13, ADAMTS-4, and ADAMTS-5, preventing type II collagen degradation. It has been

proposed that JAK inhibitors may support chondrogenic differentiation and promote cartilage regeneration by stimulating mesenchymal stem cell activity [60]. Both JAK1 and JAK2 inhibitors block TNF- α -induced STAT3 phosphorylation and STAT3-DNA binding [61]. These findings provide a rationale for developing OA therapies through modulation of the JAK/STAT signaling pathway activity.

EPIGENETICS IN THE PATHOGENESIS OF OA

Although OA is associated with a possible genetic predisposition, over time, the notion that the genome is not the primary cause of the disease has become more widely accepted. Numerous studies have failed to identify genes with high significance levels that are strongly associated with an increased prevalence of the disease. Current evidence supports the theory of polygenic inheritance of susceptibility. However, studies indicate that more than 80% of disease-associated genetic variants are located in non-coding regions of the genome [62]. This finding supports the hypothesis that epigenetics, as a key element in regulating gene expression, is involved in both the onset and progression of OA. The currently established paradigm views the disease as a process involving interactions between a broad range of genetic and environmental factors. Unlike genomic modifications, epigenetic modifications are more flexible and reversible. Epigenetic factors can induce transcriptional changes that may be critical in the pathogenesis of OA. Based on these findings, it is suggested that changes in gene expression, rather than alterations in the genetic code sequence, play a more significant role in the onset and progression of the disease [63].

Currently, the term „epigenetics“ refers to heritable and dynamic changes that do not involve alterations in the primary DNA sequence but rather adaptations in chromatin conformation to regulate gene expression [64]. The molecular basis of epigenetic mechanisms includes DNA methylation, histone modifications, and gene expression regulation through non-coding RNAs [65].

DNA methylation is one of the mechanisms that regulates human chondrogenesis. It has been established that changes in DNA methylation patterns correlate with OA manifestations [66]. Helmtrud I. and colleagues were the first to study specific methylation loci. They quantitatively analyzed the methylation of the proximal promoter regions of aggrecanase 1 and matrix metalloproteinase 2 (gelatinase A), MMP3 (stromelysin 1), MMP9 (gelatinase B), and MMP13

(collagenase 3) [67]. They demonstrated that the degree of demethylation at at least one CpG site in each enzyme was statistically higher in OA patients than in the control group. In a 2014 study by Fernandez-Tajes J. and colleagues, the authors identified a phenotypic subgroup of OA patients characterized by an increased local inflammatory response, which may be regulated by epigenetics [68]. Another study suggested that global DNA methylation could serve as a powerful tool for the molecular characterization of OA and as a potential diagnostic marker [69]. Subsequent studies on cartilage tissue have identified additional DNA methylation sites in OA, and these genes can be classified into four main categories:

- The first category relates to extracellular matrix homeostasis and includes COL2A1, COL9A1, COL10A1, ACAN, MMP2, MMP3, MMP9, MMP13, and ADAMTS-4 [70].
- The second category comprises molecules associated with inflammation, such as IL1B, IL8, IL32, TGF- β 2, IL1RN, and WNT11 [71].
- The third category includes SOX4, SOX9, RUNX2, and SOD2, which are involved in cartilage maintenance [72].
- The fourth category consists of growth factors such as BMP7, SOST, and GDF5 [73].

A study has established a link between DNA methyltransferase (DNMT) polymorphisms and primary knee osteoarthritis [74]. In response to IL-1, chondrocytes in the transitional zone increase the expression and activity of DNMT1. In 2018, Ellis H. and colleagues investigated DNA methylation at 850,000 sites across the genome using the MethylationEPIC BeadChip in samples from the UK Longitudinal Study [75]. They identified 548 significant DNA methylation loci among 2,907,234 genetic variations and 93,268 DNA methylation sites, indicating 548 significant DNA methylation loci. These findings suggest that DNA methylation may influence gene and non-coding RNA expression, as well as gene transcription and translation. It is known that DNA methylation varies significantly in tissues depending on disease stages [76]. This presents a challenge in determining from which structures biological samples should be collected for clinical methylation studies. Despite these challenges, discoveries in this field add new knowledge to the current understanding of the role of epigenetics in OA. Genome-wide studies further confirm and clarify the interaction between genetics and epigenetics in OA. Fernandez-Tajes and colleagues examined genome-wide DNA methylation in human joint chondrocytes to identify specific DNA methylation profiles in OA. They analyzed a

cohort of 25 OA patients and 20 healthy controls. In this patient subgroup, they identified 1,357 differentially methylated sites. Additionally, 450 genes were differentially expressed in patients with an elevated inflammatory response, which may be epigenetically regulated [77].

Another epigenetic mechanism involves post-translational histone modifications (PTMs). Initially, histones were thought to function as a static scaffold for DNA packaging. However, it was later discovered that they are dynamic proteins that undergo various post-translational modifications. There are five histone families [H1-H5], divided into two groups. Some lysine acetylations and methylations serve as activating signals for gene transcription. Histone H3 is modified by a large number of PTMs, including acetylation, methylation, and phosphorylation [78]. Histone methylation is crucial for proper genome programming during development, and disruptions in this process can lead to pathological conditions [79]. Broadly speaking, histones can be modified by histone deacetylases (HDACs) or histone acetyltransferases (HATs), leading to more or less condensed chromatin structures. Increased activation and expression of histone deacetylases may play a role in the pathogenesis of knee osteoarthritis [OA]. The upregulation of HDAC modulates the activity of miR-146a, which triggers the inflammatory response in knee OA [80]. UTX is a histone demethylase. In 2022, Lian W.-S. and colleagues investigated how UTX alters the phenotype of articular cartilage by stimulating anabolic processes or activating the phenotypic dedifferentiation of the extracellular matrix [81]. Evidence that histone post-translational modifications play a significant role in OA has increased over the past few years; however, the role of histones in the early stages of OA remains largely unknown. This is partly due to the difficulty of obtaining samples from the structurally complex articular cartilage, which has low cellular availability, and partly due to the invasiveness of the sampling procedure. Recently, a protocol for histone extraction from human articular cartilage was published, aiming to enhance the understanding of histone involvement in OA pathogenesis [82]. Similar to histone acetylation, changes in histone methylation are often associated with altered gene expression and modifications in signaling pathways in chondrocytes. In addition to histone methylation, the process of demethylation also plays a role in OA pathogenesis [83]. Increased levels of demethylases have been identified in cartilage from the tibial plateau in knee OA.

Sirtuins (SIRT) are NAD⁺ (nicotinamide adenine dinucleotide)-dependent enzymes. They function as deacetylases, removing acetyl groups from cellular proteins and thus altering their function. Deacetylation affects proteins in the cytoplasm, regulating cellular metabolism, as well as proteins in the nucleus, where it controls gene expression. It has been proven that SIRT and SIRT-dependent epigenetic control play a vital role in DNA repair, inflammation, and age-related diseases. There are seven types of sirtuins in humans, localized in different parts of the cell. SIRT1 is essential for maintaining hyaline cartilage homeostasis and is the most extensively studied sirtuin in joint pathology. Numerous *in vitro* and *in vivo* studies have demonstrated that SIRT1 is involved in OA pathogenesis, playing a role in apoptosis, cartilage matrix repair, and bone homeostasis. Increased expression of SIRT1 enhances the expression of genes responsible for extracellular matrix (ECM) production. SIRT6 has been shown to regulate chondrocyte proliferation and differentiation. Chondrocyte hypertrophy and proteoglycan production are reduced in SIRT6-deficient mice. SIRT3 is primarily located in the mitochondria, where its deacetylase activity regulates mitochondrial function, regeneration, and kinetics. Given these findings, SIRT3 is considered to play a crucial role in mitochondrial adaptations and aging delay [84].

MicroRNAs (miRNAs) are small single-stranded non-coding RNAs (ncRNAs) ranging from 21 to 25 nucleotides in length, functioning as post-translational suppressors [85]. They are involved in regulating biological processes such as cell proliferation, apoptosis, differentiation, and organogenesis. Numerous studies have demonstrated that miRNAs play a crucial role in the development and proper functioning of both innate and adaptive immunity [86]. Through a protein complex, they can induce the cleavage of messenger RNAs (mRNAs), which serve as their targets, thereby suppressing the expression of corresponding genes. A subclass of miRNAs, known as circulating miRNAs (cmiRNAs), has garnered attention due to their potential role in mediating cell-to-cell and tissue-to-tissue cross-signaling. These intracellular mediators are produced in various tissues in response to physiological and pathological stimuli and are secreted into the bloodstream, where they can travel to their target recipient tissues to modulate gene expression [87]. Furthermore, miRNAs can influence the translation of enzymes involved in histone modification and DNA methylation [88]. MiRNAs participate in numerous biological processes, including the regulation of innate and adaptive

immunity, genome stability maintenance, resistance to stress factors, cell growth, differentiation, proliferation, apoptosis, and inflammation. They are also implicated in tumorigenesis, cell migration, and invasion during the progression of various neoplasms. MiRNAs regulate the maturation and differentiation of immune cells, with certain miRNA groups involved in B- and T-cell regulation [89]. Overexpression of miR-150 has been shown to block B-cell development, while miR-199a-5p is associated with the inhibition of monocyte/macrophage differentiation [90]. Specific miRNAs are linked to the proper functioning of dendritic cells, their antigen-presenting ability, and cytokine production. The expression of Toll-like receptors (TLRs) on macrophages and dendritic cells is regulated by miRNAs [91]. The target gene of miRNA-155 in these cells is a suppressor of cytokine signaling. Consequently, a deficiency in miR-155 could lead to dysfunction in dendritic cells [92]. MiRNAs also influence the function of macrophages/monocytes [93], which are the primary cellular players in the low-grade inflammation observed in osteoarthritis (OA). Advances in molecular biology have revealed the multifaceted role of miRNAs as a „fine post-transcriptional tuning“ mechanism of the immune system. It has become evident that they play a critical role in the maturation, differentiation, and regulation of both innate and adaptive immunity [94].

The study of miRNAs is the subject of numerous clinical investigations in osteoarthritis. MiR-140 is specifically expressed in cartilage [95] and plays a significant role in chondrogenesis. In osteoarthritic cartilage, the expression of miR-140 is significantly reduced compared to healthy cartilage, leading to the expression of genes with negative effects in OA [96]. IL-1 β significantly decreases the expression of miR-140, while simultaneously increasing the expression of the catabolic MMP13 and ADAMTS5 [97]. MiR-146a is also expressed in cartilage, especially in the chondrocytes located in its superficial layer. These findings suggest that miRNAs may act as mediators in chondrocyte metabolism at the molecular level [98]. Several miRNAs with opposite roles have been identified in the pathophysiological processes of OA. While Kang and colleagues found that miR-204 inhibits prostaglandin synthesis and induces chondrocyte aging under stress conditions, Huang and colleagues argue that miR-204, together with its homolog miR-211, has a beneficial effect on cartilage homeostasis and suppresses the progression of osteoarthritis [99]. A deficiency of miR-204/-211 in mesenchymal progenitor cells increases the expression of catabolic markers, inducing OA in

mouse models [100]. MiRNAs can interact with histones and enzymes, creating complex epigenetic regulatory mechanisms. Yin and colleagues investigated the expression of miR-140-5p and miR-140-3p in synovial fluid in knee osteoarthritis, finding that both miRNAs were significantly reduced compared to levels obtained from synovial fluid from healthy knee joints. The conclusion is that the expression of miR-140-5p and miR-140-3p negatively correlates with the severity of OA [101]. Furthermore, Si and collaborators, in 2017, found that intra-articular injection of miR-140 regulates ECM homeostasis in an OA rat model, reducing disease progression. Conversely, miR-210 was found at elevated levels in synovial fluid from patients with early-stage OA and may serve as a diagnostic marker for the early detection of the disease [102]. In combination with other therapeutic strategies, miRNAs have potential as both biomarkers and therapeutic targets in OA. This strategy could aid in joint regeneration or slow disease progression in its early stages.

The JAK/STAT signaling pathway is linked to the expression of miRNA-224 in osteogenic differentiation. On the other hand, miRNA-224-5p can activate cartilage degeneration and worsen OA by activating the JAK2/STAT signaling pathway [103]. It has been shown that miRNA-149-5p is an early biomarker for OA, and JAK inhibitors exhibit therapeutic effects in OA by inhibiting its expression [104]. The miR-149/JAK1/IL-6/TNF- α axis plays an important role in maintaining joint tissue homeostasis. Specifically, miR-224-5p activates the JAK2/STAT pathway, leading to cartilage degeneration and accelerating symptom onset in OA patients [105]. Tofacitinib affects the expression of miR-149 and inhibits the JAK/IL-6/TNF- α axis, reducing the manifestations of OA in mice. MiR-223 may enhance inflammatory processes in the osteoarthritic joint by activating the JAK2/STAT1 signaling pathway in chondrocytes, increasing MMP-3 expression, and accelerating ECM degradation [106]. Hypoxic effects may enhance repair processes in cartilage in OA by stimulating chondrocyte proliferation while simultaneously suppressing apoptosis through miR-18/JAK/STAT signaling [107].

Mature miRNAs bind to complementary iRNAs – target gene sequences – and block gene expression as well as suppress translation [108]. In 2020, Lian L. P. and colleagues found that in chondrocytes of patients with arthroplasty, miR-483, miR-22, miR-377, miR-103, miR-16, miR-223, miR-2b, miR-23b, and miR-509 were elevated, while miR-29a, miR-140, miR-25, miR-337, miR-210, miR-26a, and miR-373 were reduced [109]. The genetic signatures of these

miRNAs differentiate chondrocytes in osteoarthritis from healthy chondrocytes. MiR-140 is associated with OA and can be detected in peripheral circulation or synovial fluid in advanced disease [110]. In the articular cartilage of OA patients, miR-146a is overexpressed. In these cases, the extracellular matrix is actively remodeled by concomitant low-grade inflammation, which leads to changes in the biomechanical properties of chondrocytes and accelerates the progression of OA [111]. These findings support the idea that miR-146a plays a significant role in the inflammatory component of OA [112]. Other miRNAs related to extracellular matrix degradation include miR-137, miR-449, miR-221, miR-30a, miR-19b-3p, miR-634, miR-29, miR-107, miR-497-5p, miR-26a, and miR-101. Some miRNAs, such as miR-378 in synovial fluid, can alter their expression levels as the disease progresses. Based on these conclusions, Bartholomew B. in 2014 suggested that intra-articular injection of certain miRNAs has the potential to reverse disease progression, which could be revolutionary in the treatment of osteoarthritis [113]. Wang and colleagues investigated the involvement of both lncRNAs and miRNAs in the onset and development of knee osteoarthritis. They found that 186 lncRNAs and 684 miRNAs were expressed in the degenerated chondrocytes of knee osteoarthritis [114]. The role of miR-146a-5p was analyzed in another study [115], where the authors discovered a connection with a protein involved in chondrocyte apoptosis and autophagy processes: miR-146a-5p stimulates apoptosis and suppresses autophagy in chondrocytes. It was found that miR-146a-5p is highly expressed in tissue from knees with osteoarthritis. Intra-articular injection of an antagonist of miR-146a-5p positively affected the progression of knee osteoarthritis. Recent studies suggest that miRNAs also play a crucial role in mitophagy, a process of autophagy that degrades damaged or dysfunctional mitochondria. Inhibiting miR-126-5p suppresses mitophagy and mitochondrial degradation, ensuring cartilage resistance to damage. This phenomenon is explained by the fact that mitochondria are powerful regulators of chondral homeostasis, and it has been established that they degenerate during the progression of knee osteoarthritis [116]. Overexpression of miR-103a-3p inhibits chondrocyte apoptosis and inflammation in OA [117]. Another study by Endisha and colleagues found that the expression of miR-34a-5p in both cartilage and synovium increases in patients with knee osteoarthritis. The synovium and cartilage of mice treated with intra-articular injections of miR-34a-5p showed signs of degeneration (disruption of

cartilage integrity, appearance of fissures, synovial proliferation, etc.) similar to those seen in OA [118]. Intra-articular injection of miR-34a-5p has a pro-inflammatory effect in knee osteoarthritis. A study by Tavallaei and colleagues observed elevated levels of miR-27b-3p in the synovial fluid of patients with late-stage knee osteoarthritis. The levels of this miRNA correlated with the severity of OA. The effects of injecting miR-27b-3p into the knees of mouse models were described as a phenotype with signs of synovitis, synovial fibrosis, increased inflammatory marker levels, and a pro-fibrotic response in fibroblast-like synoviocytes [119].

Another manuscript by Jin and colleagues evaluates the effect of miRNAs contained in exosomes from human mesenchymal stem cells (MSCs) in the treatment of gonarthrosis. Exosomes derived from MSCs of bone marrow prevent cartilage degeneration and subchondral bone remodeling in a rat model of gonarthrosis. They help maintain the chondrocyte phenotype by inducing type II collagen synthesis and inhibiting IL-1 β -induced aging and apoptosis. The exosomal lncRNA MEG-3 also slows aging and apoptosis of chondrocytes induced by IL-1 β [120]. Unlike microRNAs, which mainly rely on complementary RNA sequence pairing to suppress target genes, lncRNAs act in a significantly more complex manner [121]. lncRNAs have various mechanisms for regulating gene expression, including epigenetic regulation, transcriptional regulation, and post-transcriptional regulation [122]. Recent research has revealed the mechanism of axial regulation of the highly expressed lncRNA in osteoarthritic joints [123]. Of importance is the interaction between lncRNAs and miRNAs [124]. Through spatial conformation, the secondary structure of lncRNA may exert an adsorptive effect on miRNA, changing the contact concentration of miRNA and regulating the production of inflammatory genes or transcription factors in osteoarthritic joints [125].

Currently, the epigenetics of OA mainly focuses on chondrocytes, but several publications also address the epigenetic effects on the subchondral bone [126]. Through zonal analysis of the tibia, Zhang Y proposes a new hypothesis that subchondral epigenetic changes play a more significant role than those in cartilage in the progression of osteoarthritis.

Due to the differential expression of epigenetic changes in various joint tissues – chondrocytes, subchondral bone, synovium, ligaments, and other joint tissues – conflicting experimental results may occur. The heterogeneity of cells and tissues in the OA joint can compromise the accuracy of current

knowledge about gene function and epigenetic regulation. Future approaches to epigenetics in osteoarthritis would be more rational if refined single-cell studies were conducted. Single-cell studies would provide more precise subtyping of diseased tissues. In the future, these techniques could be used in OA research and provide a new perspective on understanding its pathogenesis.

CONCLUSION

In the pathological process of osteoarthritis, numerous epigenetic variables can influence the expression of genes associated with the disease [127]. An integrative study of all epigenetic mechanisms is crucial for better perspectives in the search for new biomarkers and treatments. Some of these epigenetic mechanisms have been studied for decades in other diseases or as part of the aging process. It has also been confirmed that these mechanisms regulate the expression of metalloproteases and the integrity of the cartilage matrix. We now know much more about the mechanisms and regulation of chromatin in OA, and the specific role played by chromatin modifiers, as well as the impact of histone modifications on the chondrocyte phenotype. Aberrant gene expression during aging is regulated by various epigenetic mechanisms and also plays an important role in the pathogenesis of osteoarthritic disease. Histone deacetylases have long been linked to aging, especially the Sirtuin family. Since aging is one of the risk factors for OA, research on the involvement of sirtuins in the etiology of the disease is ongoing. As a form of non-coding genetic information, epigenetics plays various regulatory roles at the molecular level in the initiation and progression of the disease [128]. Epigenetic regulations lead to interconnected and dynamic changes in the progression of the disease. OA-related signaling pathways, transcription factors, inflammatory factors, ECM proteins, and other variables are differentially sensitive to epigenetic control. However, further studies are still needed to fully understand the mechanisms of epigenetic regulation of the disease and to incorporate them into clinical practice. All of these studies, as well as the combination of genetic and epigenetic variables, contribute to a new understanding of the pathophysiology of cartilage diseases [129].

Библиография / References:

1. Ratneswaran A, Kapoor M. Osteoarthritis year in review: genetics, genomics, epigenetics. *Osteoarthritis and Cartilage* 2021;29:151–160. (10.1016/j.joca.2020.11.003)

2. Rahmati M, Nalesso G, Mobasheri A, Mozafari M. Aging and osteoarthritis: central role of the extracellular matrix. *Ageing Research Reviews* 2017;40,20–30. (10.1016/j.arr.2017.07.004)
3. Contartese D, Tschon M, De Mattei M, Fini M. Sex specific determinants in osteoarthritis: A systematic review of pre-clinical studies. *International Journal of Molecular Sciences* 2020;21. (10.3390/ijms21103696)
4. Thomas S, Browne H, Mobasheri A, Rayman MP. What is the evidence for a role for diet and nutrition in osteoarthritis? *Rheumatology (Oxford)* 2018;57,iv61–iv74. (10.1093/rheumatology/key011)
5. Stürmer T, Günther KP, Brenner H. Obesity, overweight and patterns of osteoarthritis: the Ulm osteoarthritis Study. *Journal of Clinical Epidemiology* 2000;53,307–313. (10.1016/s0895-4356(99)00162-6)
6. Ter Heegde F, Luiz AP, Santana-Varela S, et al. Osteoarthritis-related nociceptive behaviour following mechanical joint loading correlates with cartilage damage. *Osteoarthritis and Cartilage* 2020;28,383–395. (10.1016/j.joca.2019.12.004)
7. Thomas AC, Hubbard-Turner T, Wikstrom EA, Palmieri-Smith RM. Epidemiology of posttraumatic osteoarthritis. *Journal of Athletic Training* 2017;52,491–496. (10.4085/1062-6050-51.5.08)
8. Kuusalo L, Felson DT, Wang N, et al. Metabolic osteoarthritis – relation of diabetes and cardiovascular disease with knee osteoarthritis. *Osteoarthritis and Cartilage* 2021;29,230–234. (10.1016/j.joca.2020.09.010)
9. Scanzello CR, Goldring SR. The role of synovitis in osteoarthritis pathogenesis. *Bone* 2012;51,249–257. (10.1016/j.bone.2012.02.012).
10. Woodell-May JE, Sommerfeld SD. Role of inflammation and the immune system in the progression of osteoarthritis. *Journal of Orthopaedic Research* 2020;38,253–257. (10.1002/jor.24457)
11. Xie J, Wang Y, Lu L, et al. Cellular senescence in knee osteoarthritis: molecular mechanisms and therapeutic implications. *Ageing Research Reviews* 2021;70,101413. (10.1016/j.arr.2021.101413)
12. Sun K, Jing X, Guo J, et al. Mitophagy in degenerative joint diseases. *Autophagy* 2021;17,2082–2092. (10.1080/15548627.2020.1822097)
13. Mehana EE, Khafaga AF, El-Blehi SS. The role of matrix metalloproteinases in osteoarthritis pathogenesis: an updated review. *Life Sciences* 2019;234,116786. (10.1016/j.lfs.2019.116786)
14. Yang CY, Chanalaris A, Troeberg L. LADAMTS and ADAM metalloproteinases in osteoarthritis – looking beyond the “usual suspects”. *Osteoarthritis and Cartilage* 2017;25,1000–1009. (10.1016/j.joca.2017.02.791)
15. Goldring MB, Goldring SR. Articular cartilage and subchondral bone in the pathogenesis of osteoarthritis. *Annals of the New York Academy of Sciences* 2010;1192,230–237. (10.1111/j.1749-6632.2009.05240.x)
16. Van der Kraan PM, Van den Berg WB. Osteophytes: relevance and biology. *Osteoarthritis and Cartilage* 2007;15,237–244. (10.1016/j.joca.2006.11.006)
17. Madry H, Kon E, Condello V, et al. Early osteoarthritis of the knee. *Knee Surgery, Sports Traumatology, Arthroscopy* 2016;24,1753–1762. (10.1007/s00167-016-4068-3)
18. Sharma AR, Jagga S, Lee SS, Nam JS. Interplay between cartilage and subchondral bone contributing to pathogenesis of osteoarthritis. *International Journal of Molecular Sciences* 2013;14,19805–19830. (10.3390/ijms141019805)
19. Abraham AC, Pauly HM, Donahue TL. Deleterious effects of osteoarthritis on the structure and function of the meniscal enthesis. *Osteoarthritis and Cartilage* 2014;22,275–283. (10.1016/j.joca.2013.11.013)
20. Schulze-Tanzil G. Intraarticular ligament degeneration is interrelated with cartilage and bone destruction in osteoarthritis. *Cells* 2019. (10.3390/cells8090990)
21. Shorter E, Sannicandro AJ, Poulet B, Goljanek-Whysall K. Skeletal muscle wasting and its relationship with osteoarthritis: a mini-review of mechanisms and current interventions. *Current Rheumatology Reports* 2019;21,40. (10.1007/s11926-019-0839-4)
22. Bastiaansen-Jenniskens YM, Clockaerts S, Feijt C, et al. Infrapatellar fat pad of patients with end-stage osteoarthritis inhibits catabolic mediators in cartilage. *Annals of the Rheumatic Diseases* 2012;71,288–294. (10.1136/ard.2011.153858)
23. Kapoor M, Martel-Pelletier J, Lajeunesse D, et al. Role of proinflammatory cytokines in the pathophysiology of osteoarthritis. *Nat Rev Rheumatol* 2011; 7:33–42.
24. Su ZQ, Guan WJ, Li SY et al. Evaluation of the Normal Airway Morphology Using Optical Coherence Tomography. *Chest*. 2019;156(5):915-925. doi: 10.1016/j.chest.2019.06.009.
25. Hu Y, Chen X, Wang S et al. Subchondral bone microenvironment in osteoarthritis and pain. *Bone Res*. 2021;9(1):20. doi: 10.1038/s41413-021-00147-z.
26. Hu W, Chen Y, Dou C, Dong S. Microenvironment in subchondral bone: predominant regulator for the treatment of osteoarthritis. *Ann Rheum Dis*. 2021;80(4):413-422. doi: 10.1136/annrheumdis-2020-218089.
27. Singh-Jasuja H, Hilf N, Arnold-Schild D, Schild H. The role of heat shock proteins and their receptors in the activation of the immune system. *Biol Chem*. 2001 Apr;382(4):629-36. doi: 10.1515/BC.2001.074.
28. Kabalyk MA. Age-Related Aspects of the Involvement of Heat Shock Proteins in the Pathogenesis of Osteoarthritis, 01. March. 2018;7,274-280, *Advances in Gerontology*
29. Ngarmukos S, Scaramuzza S, Theerawattanapong N, et al. Circulating and Synovial Fluid Heat Shock Protein 70 Are Correlated with Severity in Knee Osteoarthritis. *Cartilage*. 2020;11(3):323-328. doi: 10.1177/1947603518790075.
30. O'Shea JJ, Schwartz DM, Villarino AV, et al. The JAK-STAT pathway: impact on human disease and therapeutic intervention. *Annu Rev Med* 2015;66:311–28. doi: 10.1146/annurev-med-051113-024537
31. Xin P, Xu X, Deng C, et al. The role of JAK/STAT signaling pathway and its inhibitors in diseases. *Int Immunopharmacol* 2020;80:106210. doi: 10.1016/j.intimp.2020.106210
32. Namba A, Aida Y, Suzuki N, et al. Effects of IL-6 and soluble IL-6 receptor on the expression of cartilage matrix proteins in human chondrocytes. *Connect Tissue Res* 2007;48(5):263–70. doi: 10.1080/03008200701587513
33. Weng LH, Ko JY, Wang CJ, et al. Dkk-1 promotes angiogenic responses and cartilage matrix proteinase secretion in synovial fibroblasts from osteoarthritic joints. *Arthritis Rheum* 2012;64(10):3267–77. doi: 10.1002/art.34602
34. Blasioli DJ, Kaplan DL. The roles of catabolic factors in the development of osteoarthritis. *Tissue Eng Part B Rev* 2014;20(4):355–63. doi: 10.1089/ten.TEB.2013.0377
35. Larsson R, Bernhardsson S, Nordeman L. Effects of eccentric exercise in patients with subacromial impingement syndrome: a systematic review and meta-analysis. *BMC*

- Musculoskelet Disord. 2019 Oct 14;20(1):446. doi: 10.1186/s12891-019-2796-5.
36. Latourte A, Cherifi C, Maillet J, et al. Systemic inhibition of IL-6/Stat3 signalling protects against experimental osteoarthritis. *Ann Rheum Dis* (2017)76(4):748–55. doi: 10.1136/annrheumdis-2016-209757
 37. Liu J, Dong X, Liu Y, et al. The identified hub gene GlcN in osteoarthritis progression and treatment. *Comput Math Methods Med* 2021;2021:5499450. doi: 10.1155/2021/5499450
 38. Legendre F, Duhia J, Pujol JP, Bogdanowicz P. JAK/STAT but not ERK1/ERK2 pathway mediates interleukin (IL)-6/soluble IL-6R down-regulation of type II collagen, aggrecan core, and link protein transcription in articular chondrocytes. association with a down-regulation of SOX9 expression. *J Biol Chem* 2003;278(5):2903–12. doi: 10.1074/jbc.M110773200
 39. Gyurkovska V, Stefanova T, Dimitrova P, et al. Tyrosine kinase inhibitor tyrphostin AG490 retards chronic joint inflammation in mice. *Inflammation* 2014;37(4):995–1005. doi: 10.1007/s10753-014-9820-6
 40. Maldonado M, Nam J. The role of changes in extracellular matrix of cartilage in the presence of inflammation on the pathology of osteoarthritis. *Biomed Res Int* 2013; 2013:284873
 41. Assirelli E, Pulsatelli L, Dolzani P, et al. Human osteoarthritic cartilage shows reduced *in vivo* expression of IL-4, a chondroprotective cytokine that differentially modulates IL-1 β -stimulated production of chemokines and matrix-degrading enzymes *in vitro*. *PloS One* 2014;9(5):e96925. doi: 10.1371/journal.pone.0096925
 42. Leong DJ, Li YH, Gu XI, et al. Physiological loading of joints prevents cartilage degradation through CITED2. *FASEB J* 2011;25(1):182–91. doi: 10.1096/fj.10-164277
 43. He Z, Leong DJ, Xu L, et al. CITED2 mediates the cross-talk between mechanical loading and IL-4 to promote chondroprotection. *Ann N Y Acad Sci* 2019;1442(1):128–37. doi: 10.1111/nyas.14021
 44. Carey AJ, Tan CK, Ulett GC. Infection-induced IL-10 and JAK-STAT: A review of the molecular circuitry controlling immune hyperactivity in response to pathogenic microbes. *Jakstat* 2012;1(3):159–67. doi: 10.4161/jkst.19918
 45. Li X, Kim KW, Cho ML, et al. IL-23 induces receptor activator of NF- κ B ligand expression in fibroblast-like synoviocytes via STAT3 and NF- κ B signal pathways. *Immunol Lett* 2010;127(2):100–7. doi: 10.1016/j.imlet.2009.10.012
 46. Millward-Sadler SJ, Khan NS, Bracher MG, et al. Roles for the interleukin-4 receptor and associated JAK/STAT proteins in human articular chondrocyte mechanotransduction. *Osteoarthritis Cartilage* 2006;14(10):991–1001. doi: 10.1016/j.joca.2006.03.013
 47. Wang L, He C. Nrf2-mediated anti-inflammatory polarization of macrophages as therapeutic targets for osteoarthritis. *Front Immunol* 2022;13:967193. doi: 10.3389/fimmu.2022.967193
 48. Butturini E, Carcereri de Prati A, Mariotto S. Redox regulation of STAT1 and STAT3 signaling. *Int J Mol Sci* 2020;21(19):7034. doi: 10.3390/ijms21197034
 49. Bhattacharjee A, Shukla M, Yakubenko VP, et al. IL-4 and IL-13 employ discrete signaling pathways for target gene expression in alternatively activated monocytes/macrophages. *Free Radic Biol Med* 2013;54:1–16. doi: 10.1016/j.freeradbiomed.2012.10.553
 50. Heinrich PC, Behrmann I, Müller-Newen G, et al. Interleukin-6-type cytokine signalling through the gp130/Jak/STAT pathway. *Biochem J* 1998;334(Pt 2):297–314. doi: 10.1042/bj3340297
 51. Fahy N, de Vries-van Melle ML, Lehmann J, et al. Human osteoarthritic synovium impacts chondrogenic differentiation of mesenchymal stem cells via macrophage polarisation state. *Osteoarthritis Cartilage* 2014;22(8):1167–75. doi: 10.1016/j.joca.2014.05.021
 52. Dai M, Sui B, Xue Y, et al. Cartilage repair in degenerative osteoarthritis mediated by squid type II collagen via immunomodulating activation of M2 macrophages, inhibiting apoptosis and hypertrophy of chondrocytes. *Biomaterials* 2018;180:91–103. doi: 10.1016/j.biomaterials.2018.07.011
 53. Levinger I, Levinger P, Trenerry MK, et al. Increased inflammatory cytokine expression in the vastus lateralis of patients with knee osteoarthritis. *Arthritis Rheum* 2011;63(5):1343–8. doi: 10.1002/art.30287
 54. Huang CY, Lai KY, Hung LF, et al. Advanced glycation end products cause collagen II reduction by activating janus kinase/signal transducer and activator of transcription 3 pathway in porcine chondrocytes. *Rheumatol (Oxford)* 2011;50(8):1379–89. doi: 10.1093/rheumatology/ker134
 55. Lim H, Kim HP. Matrix metalloproteinase-13 expression in IL-1 β -treated chondrocytes by activation of the p38 MAPK/c-Fos/AP-1 and JAK/STAT pathways. *Arch Pharm Res* 2011;34(1):109–17. doi: 10.1007/s12272-011-0113-4
 56. De Andrés MC, Imagawa K, Hashimoto K, et al. Suppressors of cytokine signalling (SOCS) are reduced in osteoarthritis. *Biochem Biophys Res Commun* 2011;407(1):54–9. doi: 10.1016/j.bbrc.2011.02.101
 57. O'Shea JJ, Schwartz DM, Villarino AV, et al. The JAK-STAT pathway: impact on human disease and therapeutic intervention. *Annu Rev Med* 2015;66:311–28. doi: 10.1146/annurev-med-051113-024537
 58. Minegishi Y, Saito M, Tsuchiya S, et al. Dominant-negative mutations in the DNA-binding domain of STAT3 cause hyper-IgE syndrome. *Nature* 2007;448(7157):1058–62. doi: 10.1038/nature06096
 59. Koskinen-Kolasa A, Vuolteenaho K, Korhonen R, et al. Catabolic and proinflammatory effects of leptin in chondrocytes are regulated by suppressor of cytokine signaling-3. *Arthritis Res Ther* 2016;18(1):215. doi: 10.1186/s13075-016-1112-0
 60. Andreas K, Häupl T, Lübke C, et al. Antirheumatic drug response signatures in human chondrocytes: potential molecular targets to stimulate cartilage regeneration. *Arthritis Res Ther* 2009;11(1): R15. doi: 10.1186/ar2605
 61. Van Beuningen HM, de Vries-van Melle ML, Vitters EL, et al. Inhibition of TAK1 and/or JAK can rescue impaired chondrogenic differentiation of human mesenchymal stem cells in osteoarthritis-like conditions. *Tissue Eng Part A* 2014;20(15-16):2243–52. doi: 10.1089/ten.TEA.2013.0553
 62. Li Y, Tang J, Hu Y. Dimethyl fumarate protection against collagen II degradation. *Biochem Biophys Res Commun* 2014;454(2):257–61. doi: 10.1016/j.bbrc.2014.10.005
 63. Maurano MT, Humbert R, Rynes E, et al. Systematic Localization of Common Disease-Associated Variation in Regulatory DNA. *Science* 2012;337,1190–1195.
 64. Barter MJ, Bui C, Young DA. Epigenetic mechanisms in cartilage and osteoarthritis: DNA methylation, histone modifications and microRNAs. *Osteoarthritis Cartilage* 2012;20:339–49.
 65. Allis CD, Jenuwein T. The molecular hallmarks of epigenetic control. *Nature reviews. Genetics*. 2016 Aug;17(8):487–500. DOI: 10.1038/nrg.2016.59.

66. Cai Z, Long T, Zhao Y, et al. Epigenetic Regulation in Knee Osteoarthritis. *Front. Genet.* 2022;13, 942982.
67. Hannon E, Gorrie-Stone TJ, Smart MC, et al. Leveraging DNA-Methylation Quantitative-Trait Loci to Characterize the Relationship between Methyloomic Variation, Gene Expression, and Complex Traits. *Am. J. Hum. Genet.* 2018;103(5), 654–665. doi:10.1016/j.ajhg.2018.09.007
68. Roach HI, Aigner T. DNA methylation in osteoarthritic chondrocytes: A new molecular target. *Osteoarthr. Cartil.* 2007; 15:128–137. doi:10.1016/j.joca.2006.07.002
69. Fernandez-Tajes J, Soto-Hermida A, Vazquez-Mosquera et al. Genome-wide DNA methylation analysis of articular chondrocytes reveals a cluster of osteoarthritic patients. *Ann Rheum Dis* 2014;73:668–77.
70. Rushton MD, Reynard LN, Barter MJ, et al. Characterization of the cartilage DNA methylome in knee and hip osteoarthritis. *Arthritis Rheumatol* 2014;66:2450–60.
71. Roach HI, Aigner T. DNA methylation in osteoarthritic chondrocytes: A new molecular target. *Osteoarthr. Cartil.* 2007; 15:128–137. doi:10.1016/j.joca.2006.07.002
72. Moazedi-Fuerst FC, Hofner M, Gruber G, et al. Epigenetic Differences in Human Cartilage between Mild and Severe OA. *J. Orthop. Res.* 2014, 32(12), 1636–1645. doi:10.1002/jor.22722
73. Kim KI, Park YS, Im GI. Changes in the epigenetic status of the SOX-9 promoter in human osteoarthritic cartilage. *J. Bone Miner. Res.* 2013; 28:1050–1060. doi:10.1002/jbmr.1843
74. Loeser RF, Im HJ, Richardson B, et al. Methylation of the OP-1 promoter: potential role in the age-related decline in OP-1 expression in cartilage. *Osteoarthritis and cartilage* 2009; 17:513–517
75. Miranda-Duarte A. DNA Methylation in Osteoarthritis: Current Status and Therapeutic Implications. *Open Rheumatol. J.* 2018, 12, 37–49. doi:10.2174/1874312901812010037
76. Hannon E, Gorrie-Stone TJ, Smart MC, et al. Leveraging DNA-Methylation Quantitative-Trait Loci to Characterize the Relationship between Methyloomic Variation, Gene Expression, and Complex Traits. *Am. J. Hum. Genet.* 2018, 103(5), 654–665. doi:10.1016/j.ajhg.2018.09.007
77. Moazedi-Fuerst FC, Hofner M, Gruber G, et al. Epigenetic Differences in Human Cartilage between Mild and Severe OA. *J. Orthop. Res.* 2014;32(12), 1636–1645. doi:10.1002/jor.22722
78. Fernández-Tajes J, Soto-Hermida A, Vázquez-Mosquera ME, et al. Genome-wide DNA methylation analysis of articular chondrocytes reveals a cluster of osteoarthritic patients. *Ann. Rheum. Dis.* 2014; 73:668–677. doi:10.1136/annrheumdis-2012-202783.
79. Wang F, Higgins JM. Histone modifications and mitosis: countermarks, landmarks, and bookmarks. *Trends Cell Biol.* 2013;23(4):175–84.
80. Van Rechem C, Whetstone JR. Examining the impact of gene variants on histone lysine methylation. *Biochim. Biophys. Acta* 2014, 1839(12), 1463–76.
81. Wen ZH, Huang JS, Lin YY, et al. Chondroprotective Effects of a Histone Deacetylase Inhibitor, Panobinostat, on Pain Behavior and Cartilage Degradation in Anterior Cruciate Ligament Transection-Induced Experimental Osteoarthritic Rats. *Int. J. Mol. Sci.* 2021;22, 7290
82. Lian WS, Wu RW, Ko JY, et al. Histone H3K27 demethylase UTX compromises articular chondrocyte anabolism and aggravates osteoarthritic degeneration. *Cell Death Dis.* 2022;13, 538.
83. Núñez-Carro C, Blanco-Blanco M, Montoya T, et al. Histone Extraction from Human Articular Cartilage for the Study of Epigenetic Regulation in Osteoarthritis. *Int. J. Mol. Sci.* 2022; 23:3355. doi:10.3390/ijms23063355.
84. Wang P, Mao Z, Pan Q, et al. Histone Deacetylase-4 and Histone Deacetylase-8 Regulate Interleukin-1 β -Induced Cartilage Catabolic Degradation through MAPK/JNK and ERK Pathways. *Int. J. Mol. Med.* 2018, 41(4), 2117–2127. doi:10.3892/ijmm.2018.3410
85. Lee SH, Lee JH, Lee HY, Min KJ. Sirtuin Signaling in Cellular Senescence and Aging. *BMB Rep.* 2019, 52(1), 24–34. doi:10.5483/bmbrep.2019.52.1.290
86. He L, Hannon GJ. MicroRNAs: Small RNAs with a big role in gene regulation. *Nat. Rev. Genet.* 2004; 5:522–531. doi:10.1038/nrg1379.
87. Berezin AE, Eugen I Poplyonkin. Diagnostic and therapeutic value of microRNAs in inflammatory bowel disease. *Biomedical Research and Therapy*, 2020, 7(2):3622–3632.
88. Lindsay MA. microRNAs and the immune response. *Trends Immunol.* 2008; 29:343–51.
89. Fernandes-Silva MM, Carvalho VO, Guimarães GV, Bacal F, Bocchi EA. Physical exercise and microRNAs: new frontiers in heart failure. *Arq Bras Cardiol.* 2012; 98:459–66.
90. Chen CZ, Li L, Lodish HF, Bartel DP. MicroRNAs modulate hematopoietic lineage differentiation. *Science.* 2004; 303:83–86.
91. Zhang C, Zhang Z, Chang Z, et al. miR-193b-5p Regulates Chondrocytes Metabolism by Directly Targeting Histone Deacetylase 7 in Interleukin-1 β -induced Osteoarthritis. *J Cell. Biochem.* 2019, 120(8), 12775–12784. doi:10.1002/jcb.28545
92. Dunne A, O'Neill LA. Adaptor usage and Toll-like receptor signaling specificity. *FEBS Lett.* 2005;579:3330–3335.
93. Ceppi M, Pereira PM, Dunand-Sauthier I, et al. MicroRNA-155 modulates the interleukin-1 signaling pathway in activated human monocyte-derived dendritic cells. *Proc Natl Acad Sci USA.* 2009;106:2735–2740.
94. Wang J, Chen L, Jin S et al. MiR-98 Promotes Chondrocyte Apoptosis by Decreasing Bcl-2 Expression in a Rat Model of Osteoarthritis. *Acta Biochim. Biophys. Sin.* 2016;48(10), 923–929. doi:10.1093/abbs/gmw084
95. Boros E, Nagy I. The Role of MicroRNAs upon Epithelial-to-Mesenchymal Transition in Inflammatory Bowel Disease. *Cells* 2019, 8, 1461; doi:10.3390/cells8111461
96. Wienholds E, Kloosterman WP, Miska E, et al. MicroRNA expression in zebrafish embryonic development. *Science.* 2005; 309:310–311. doi:10.1126/science.1114519.
97. Iliopoulos D, Malizos KN, Oikonomou P, Tsezou A. Integrative MicroRNA and Proteomic Approaches Identify Novel Osteoarthritis Genes and Their Collaborative Metabolic and Inflammatory Networks. *PLoS ONE.* 2008;3: e3740. doi:10.1371/journal.pone.0003740
98. Miyaki S, Nakasa T, Otsuki S, et al. MicroRNA-140 is expressed in differentiated human articular chondrocytes and modulates interleukin-1 responses. *Arthritis Rheum.* 2009; 60:2723–2730. doi:10.1002/art.24745
99. Chelieschi S, Barbarino M, Gallo I, et al. Hydrostatic Pressure Regulates Oxidative Stress through microRNA in Human Osteoarthritic Chondrocytes. *Int. J. Mol. Sci.* 2020; 21:3653. doi:10.3390/ijms21103653
100. Kang D, Shin J, Cho Y, et al. Stress-activated miR-204 governs senescent phenotypes of chondrocytes to promote osteoarthritis development. *Sci. Transl. Med.* 2019;11: 6659. doi:10.1126/6659
101. Huang J, Zhao L, Fan Y, et al. The microRNAs miR-204 and miR-211 maintain joint homeostasis and protect against osteoarthritis progression. *Nat. Commun.* 2019; 10:2876. doi:10.1038/s41467-019-10753-5.
102. Yin CM, Suen WCW, Lin S, et al. Dysregulation of both miR-140-3p and miR-140-5p in synovial fluid correlate with

- osteoarthritis severity. *Bone Jt. Res.* 2017; 6:612–618. doi:10.1302/2046-3758.611.BJR-2017-0090.R1
103. Li HZ, Lin Z, Xu XH, et al. The potential roles of circRNAs in osteoarthritis: a coming journey to find a treasure. *Biosci Rep* (2018)38(5): BSR20180542. doi: 10.1042/bsr20180542
 104. Cai Q, Zheng P, Ma F, et al. MicroRNA-224 enhances the osteoblastic differentiation of hMSCs via Rac1. *Cell Biochem Funct* 2019;37(2):62–71. doi: 10.1002/cbf.3373
 105. Chiu YS, Bamodu OA, Fong IH, et al. The JAK inhibitor tofacitinib inhibits structural damage in osteoarthritis by modulating JAK1/TNF-alpha/IL-6 signaling through mir-149-5p. *Bone* 2021;151:116024. doi: 10.1016/j.bone.2021.116024
 106. Zhang L, Sui C, Zhang Y, et al. Knockdown of hsa_circ_0134111 alleviates the symptom of osteoarthritis via sponging microRNA-224-5p. *Cell Cycle* 2021;20(11):1052–66. doi: 10.1080/15384101.2021.1919838
 107. Du X, Wang X, Cui K, et al. Tanshinone IIA and astragaloside IV inhibit miR-223/JAK2/STAT1 signalling pathway to alleviate lipopolysaccharide-induced damage in nucleus pulposus cells. *Dis Markers* 2021;2021:6554480. doi: 10.1155/2021/6554480
 108. Zhang B, Tian X, Qu Z, et al. Hypoxia-preconditioned extracellular vesicles from mesenchymal stem cells improve cartilage repair in osteoarthritis. *Membranes (Basel)* 2022;12(2):225. doi: 10.3390/membranes12020225
 109. D'Adamo S, Alvarez-Garcia O, Muramatsu Y, et al. MicroRNA-155 Suppresses Autophagy in Chondrocytes by Modulating Expression of Autophagy Proteins. *Osteoarthritis Cartil.* 2016, 24(6), 1082–1091. doi: 10.1016/j.joca.2016.01.005
 110. Lian LP, Xi XY. Long Non-coding RNA XIST Protects Chondrocytes ATDC5 and CHON-001 from IL-1 β -induced Injury via Regulating miR-653-5p/SIRT1 axis. *J. Biol. Regul. Homeost. Agents* 2020,34(2), 379–391. doi: 10.23812/19-549-A-65
 111. Miyaki S, Nakasa T, Otsuki S, et al. MicroRNA-140 is expressed in differentiated human articular chondrocytes and modulates interleukin-1 responses. *Arthritis Rheum.* 2009; 60:2723–2730. doi:10.1002/art.24745
 112. Lian LP, Xi XY. Long Non-coding RNA XIST Protects Chondrocytes ATDC5 and CHON-001 from IL-1 β -induced Injury via Regulating miR-653-5p/SIRT1 axis. *J. Biol. Regul. Homeost. Agents* 2020, 34(2), 379–391. doi: 10.23812/19-549-A-65
 113. Yamasaki K, Nakasa T, Miyaki S, et al. Expression of MicroRNA-146a in osteoarthritis cartilage. *Arthritis Rheum.* 2009; 60:1035–1041. doi:10.1002/art.24404
 114. Bartholomew B. ISWI chromatin remodeling: one primary actor or a coordinated effort? *Curr Opin Struct Biol.* 2014; 24:150–153.
 115. Wang G, He L, Xiang Y, et al. Long noncoding and micro-RNA expression in a model of articular chondrocyte degeneration induced by stromal cell-derived factor-1. *Asian Biomed. Res. Rev. News* 2022,16, 169–179.
 116. Zhang H, Ji L, Yang Y, et al. The therapeutic effects of treadmill exercise on osteoarthritis in rats by inhibiting the HDAC3/NF-kappaB pathway in vivo and in vitro. *Frontiers in Physiology* 2019, 10:1060. doi:10.3389/fphys.2019.01060
 117. Kim D, Song J, Jin EJ. BNIP3-Dependent Mitophagy via PG-C1 α Promotes Cartilage Degradation. *Cells* 2021,10, 1839.
 118. Cheng M, Wang Y. Downregulation of HMGB1 by miR-103a-3p Promotes Cell Proliferation, Alleviates Apoptosis and Inflammation in a Cell Model of Osteoarthritis. *Iran J. Biotechnol.* 2020,18, 24–31.
 119. Endisha H, Datta P, Sharma A, et al. MicroRNA-34a-5p Promotes Joint Destruction During Osteoarthritis. *Arthritis Rheumatol.* 2020,73, 426–439.
 120. Tavallaei G, Lively S, Rockel JS, et al. Contribution of MicroRNA-27b-3p to Synovial Fibrotic Responses in Knee Osteoarthritis. *Arthritis Rheumatol.* 2022,74, 1928–1942.
 121. Jin Y, Xu M, Zhu H, et al. Therapeutic effects of bone marrow mesenchymal stem cells-derived exosomes on osteoarthritis. *J. Cell. Mol. Med.* 2021,25, 9281–9294.
 122. Wang B, Li J, Tian F. Downregulation of lncRNA SNHG14 Attenuates Osteoarthritis by Inhibiting FSTL-1 Mediated NLRP3 and TLR4/NF-Kb Pathway through miR-124-3p. *Life Sci.* 2021,270, 119143. doi: 10.1016/j.lfs.2021.119143
 123. Wang CL, Zuo B, Li D, et al. The Long Noncoding RNA H19 Attenuates Force-Driven Cartilage Degeneration via miR-483-5p/Dusp5. *Biochem. Biophysical Res. Commun.* 2020, 529(2), 210–217. doi: 10.1016/j.bbrc.2020.05.180.
 124. He X, Memczak S, Qu J et al. Single-cell omics in ageing: a young and growing field. *Nat Metab.* 2020;2(4):293–302. doi: 10.1038/s42255-020-0196-7.
 125. Ma S, Chi X, Cai Y et al. Decoding Aging Hallmarks at the Single-Cell Level. *Annu Rev Biomed Data Sci.* 2023;6:129–152. doi: 10.1146/annurev-biodatasci-020722-120642.
 126. Liu Y, Yang Y, Ding L, et al. LncRNA MIR4435-2HG Inhibits the Progression of Osteoarthritis through miR-510-3p Sponging. *Exp. Ther. Med.* 2020a,20(2), 1693–1701. doi: 10.3892/etm.2020.8841
 127. Liu Y, Liu K, Tang C, et al. Long Non-coding RNA XIST Contributes to Osteoarthritis Progression via miR-149-5p/DNMT3A axis. *Biomed. Pharmacother.* 2020b, 128, 110349. doi: 10.1016/j.biopha.2020.110349
 128. Zhang Y, Fukui N, Yahata M, et al. Identification of DNA Methylation Changes Associated with Disease Progression in Subchondral Bone with Site-Matched Cartilage in Knee Osteoarthritis. *Sci. Rep.* 2016,6, 34460. doi: 10.1038/srep34460
 129. Simon TC, Jeffries MA. The Epigenomic Landscape in Osteoarthritis. *Curr. Rheumatol. Rep.* 2017, 19(6), 30. doi: 10.1007/s11926-017-0661-9
 130. Parker E, Hofer IMJ, Rice SJ, et al. Multi-Tissue Epigenetic and Gene Expression Analysis Combined with Epigenome Modulation Identifies RWDD2B as a Target of Osteoarthritis Susceptibility. 2021.
 131. Ramos YF, Meulenbelt I. The role of epigenetics in osteoarthritis: Current perspective. *Curr. Opin. Rheumatol.* 2017,29, 119–129.

Постъпил за печат: 02.07.2025 г.

Submitted: 02.07.2025

✉ Адрес за кореспонденция:
Ирина Момчева
Отделение по Ревматология
УМБАЛ – Бургас

✉ Correspondence address:
Irina Momcheva
Department of Rheumatology
UMHAT – Burgas