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Review

Chondroitin sulfate in osteoarthritis management among diabetic patients: molecular mechanisms and clinical potential

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Abstract

Osteoarthritis and type 2 diabetes mellitus are two prevalent chronic conditions that impose a substantial burden on healthcare systems worldwide. The comorbidity of type 2 diabetes mellitus and osteoarthritis (especially in individuals with overweight/obesity) presents a formidable challenge due to the potential synergistic effects of these conditions on metabolism and joint health. Chronic low-grade systemic inflammation plays a pivotal role in the development of both type 2 diabetes mellitus and osteoarthritis and is a common feature they share. Chondroitin sulfate is a naturally occurring glycosaminoglycan integral to the structure and function of articular cartilage. It has gained attention as a potential therapeutic agent for osteoarthritis due to its multifaceted mechanisms of action, including an effect on transcriptional factors and signaling pathways associated with chronic low-grade systemic inflammation. Chondroitin sulfate originated from natural sources, including standardized small marine fish extract, and can be regarded as a promising means in the therapy of osteoarthritis and type 2 diabetes mellitus. It can serve as an illustrative medication able to alleviate NF-κB-mediated inflammatory and metabolic disorders. Standardized small marine fish extract demonstrates anti-inflammatory, antioxidant, antidiabetic, anti-cancer, antimicrobial, anti-hypertensive, and wound healing properties.

Keywords: osteoarthritis, type 2 diabetes mellitus, chronic low-grade systemic inflammation, standardized small marine fish extract, chondroitin sulfate.

Introduction

Osteoarthritis (OA) and type 2 diabetes mellitus (T2DM) are two prevalent chronic conditions that impose a substantial burden on healthcare systems worldwide. The coexistence of T2DM and OA (especially in individuals with overweight/obesity) presents a formidable challenge due to the potential synergistic effects of these conditions on metabolism and joint health. T2DM has a detrimental effect on the results of arthroplasty and is a risk factor for the advancement of OA [1]. 47.3 percent of T2DM patients have some degree of OA [2]. It is still unclear what is behind the significant OA prevalence among T2DM individuals diagnosed with type 2 diabetes mellitus.

Overweight/obesity and advanced age are two risk factors for both OA and T2DM that, to some extent, may contribute to the explanation of why OA is more common in individuals with diabetes. T2DM is expected to affect 4.6 million people in the US between the ages of 18 and 44, 14.3 million between the ages of 45 and 64, and 12.0 million over the age of 65 [3]. Radiographically characterized knee OA substantially increases with age, affecting 14% of adults over 25 and 37% of those over 60 [4].

Recent research has shown that OA is linked to systemic metabolic abnormalities frequently observed in T2DM. This may indicate that diabetes affects the OA pathophysiology regardless of body fat or the aging process per se [5]. It has now been suggested that there is an underlying connection between OA and T2DM stemming from the metabolic changes.

In recent years, there has been T2DM considerable interest in searching for effective management of osteoarthritis in T2DM individuals, with a growing focus on the role of metabolic agents, which are able to impact transcriptional factors and signaling pathways regulating inflammation and oxidative processes in the human body.

Chondroitin sulfate (CS) is a naturally occurring glycosaminoglycan integral to the structure and function of articular cartilage. It has gained attention as a potential therapeutic agent for osteoarthritis due to its multifaceted mechanisms of action, including an effect on transcriptional factors and signaling pathways associated with nuclear factor kappa-light-chain-enhancer of activated B cells (NF-κB), signal transducer and activator of transcription (STAT) 3, mitogen-activated protein kinases (MAPKs), wingless-like (Wnt)/β-catenin, as well as nuclear factor erythroid 2-related factor (Nrf) 2.

This review aims to explore the current literature on the molecular pathways and therapeutic capability of CS in managing osteoarthritis among diabetic individuals.

Pathogenic overlaps: exploring shared pathways in the development of osteoarthritis and diabetes

Chronic low-grade inflammation (CLGI) is a central characteristic shared by both OA and T2DM. On the one hand, inflammation induced by such factors as biomechanical stress plays a role in OA development [6]. On the other hand, the OA progression occurs through the development of a systemic inflammatory response resulting from a number of conditions, including infectious diseases (*e.g.*, COVID-19) [7, 8], systemic metabolic alterations ("metainflammation") [9–11], and haemocoagulation ("thromboinflammation") [12], disruptions in gut microbiota composition [13–15], disorders of central and peripheral circadian oscillators [16, 17], inflammatory responses within the brain and spinal cord ("neuroinflammation") [18], and age-related inflammation ("inflammaging") [19, 20].

Both direct biomechanical joint impact and systemic inflammatory response factors induce the release of early-stage pro-inflammatory cytokines such as interleukins (ILs) 1β and 18 [21]. Damage-associated molecular patterns (DAMPs) caused by mechanical stress and released from damaged cellular organelles, dying cells, or the extracellular matrix (*e.g.*, adenosine triphosphate, uric acid, pore-forming toxins, and metabolites), hyperlipidemia, reactive oxygen and nitrogen species (ROS/RNS), and crystals, activate toll-like receptors (TLRs) and contribute to the assembly of the NLRP3 inflammasome [21, 22]. Caspase-1 is activated when the inflammasome is formed, and it cleaves gasdermin D (GSMD), pro-IL-1 and 18. The N-terminal GSMD leads to the formation of a transmembrane pore, which is a pathway for bioactive ILs 1 and 18 to enter extracellular space and trigger pyroptosis.

Additionally, TLRs cause the NF-κB to become active, which promotes the transcription of transmembrane pro-TNFα, which the cell releases as TNFα. Further signals from IL-1β, IL-18, and TNFα enhance chondrocyte death and spread the process to the cells in the synovial membrane [21].

Subsequent activation of signals originating from IL-1β, IL-18, and TNFα further intensifies chondrocyte apoptosis. The initiation of the NF-κB signaling pathway commences when IκB kinase (IKK) is activated, leading to the phosphorylation and subsequent breakdown of IκBα mediated by the proteasome. Consequently, the p65 protein is released, then undergoes phosphorylation and relocates from the cytoplasm to the nucleus. These occurrences trigger the activation of the transcription of various genes, including additional pro-inflammatory cytokines, *e.g.*, IL-6, chemokines, cathepsins, complement proteins, matrix metalloproteinases (MMPs) 1, 2, 3, 7, 8, 9, and 13, as well as aggrecanases, specifically a disintegrin and metalloproteinase with thrombospondin motifs (ADAMTS) 4 and 5 [23–25]. As a result, an elevation in the phosphorylated p65 level has been observed in osteoarthritis, while the level of IκBα was found to be diminished [6].

NF-κB has the potential to heighten joint injury by triggering the production of prostaglandin E2, cyclooxygenase 2 (COX2), inducible nitric oxide synthase (iNOS), and nitric oxide, thus facilitating tissue inflammation, the synthesis of catabolic factors, and the apoptosis of articular chondrocytes. In turn, this leads to the activation of additional transcription factors, such as hypoxia-inducible factor 2α (HIF-2α), E74-like factor 3 (ELF3), and runt-related transcription factor 2 (RUNX2) [25]. These transcription factors subsequently

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stimulate the expression of ADAMTS5 and MMP13 proteases that contribute to the differentiation process of pre-hypertrophic articular chondrocytes into fully differentiated chondrocytes.

All of this promotes cartilage degradation and an NF-κB-dependent systemic inflammatory response [26]. The ROS/RNS formation, which contributes to oxidative/nitrosative stress, is also promoted by NF-κB activation. Specific NF-κB inhibitors [27, 28] and polyphenols [29–33] can be used to address this issue.

Moreover, ROS/RNS can function as secondary signaling molecules, leading to heightened activation of redox-sensitive pathways linked to MAPKs, alongside STAT3 [34]. These pathways play a pivotal role in the development of oxidative/nitrosative stress, along with the generation of pro-inflammatory cytokines [35, 36], while also participating in cartilage degradation [24, 37].

Enhanced activity of MAPKs, including the p38 cascade, extracellular signal-regulated kinase (ERK) ½, and c-Jun N-terminal kinases (JNK), regulates the downstream expression of pro-inflammatory cytokines and MMPs, as well as the perception of pain, in osteoarthritis [38, 39]. The process initiates with the binding of pro-inflammatory cytokines and growth factors to their respective receptors on the cell membrane. This serves as the starting point, triggering intracellular MAPKs (MKKs) to phosphorylate specific MAPKs. While MKK1 and 2 activate ERK1 and 2, MKK3 and 6 are responsible for p38 phosphorylation, and MKK4 and 7 phosphorylate JNK1 and 2. These activated MAP kinases subsequently trigger the activation of additional protein kinases and transcriptional regulatory proteins. This, in turn, leads to the elevation of certain inflammatory genes such as MMPs, IL-1, and TNF-α. Then, these cytokines can perpetuate JNK activation, resulting in increased cytokine and MMP production [39]. An intriguing discovery involves the potential suppression of chondrocyte inflammation and OA improvement through the inhibition of p38MAPK. This highlights the therapeutic potential of targeting p38MAPK in the OA treatment [40].

The existing body of evidence indicates that the Janus kinase 2 (JAK2)/STAT3 signaling pathway holds a significant function in cytogenesis and participates in the OA progression [41]. Based on current research, directing interventions toward the JAK2/STAT3 pathway emerges as a viable approach for OA therapy [41, 42].

The activation of phosphatidylinositol 3-kinase (PI3K)-AKT signaling occurs through cytokines such as IL-1β when it binds to its cell surface receptor. Following stimulation, the membrane protein PI3K triggers the AKT phosphorylation, a process that has been shown to have a synergistic effect on NF-κB signaling. The activation of the PI3K/AKT pathway leads to an elevated production of MMPs within cells, including chondrocytes [43].

All the previously mentioned factors initiate a self-sustaining (vicious) cycle between the cartilage and synovial membrane, playing an important role in the OA progression. This sequence could be counteracted by activating a redox-sensitive transcription factor, which functions as an antagonist to NF-κB – Nrf2 [44, 45]. The activation of Nrf2 holds the capability to impede M1 polarisation while encouraging M2 polarisation via various signaling pathways, encompassing TLR/NF-κB, MAPK, and JAK/STAT [46]. It has been demonstrated that the ability of Nrf2 to suppress NLRP3 inflammasome in chondrocytes [47], as well as to limit apoptosis, extracellular matrix degradation, inflammation, and the activation of NF-κB signaling in chondrocytes and in OA mice, is associated with the suppression of high-mobility group protein B1 (HMGB1) expression [48].

The canonical and non-canonical Wnt signaling pathways are another route that becomes more active in response to OA [25]. Maintaining the metabolic equilibrium of chondrocytes relies on the baseline Wnt activity, but heightened Wnt activity, observed in OA, can lead to the deterioration of the cartilage matrix through associated proteases that degrade the extracellular matrix [49]. The levels of MMP-13, ADAMTS 4 and 5 were significantly increased in animals exhibiting an amplified expression of β-catenin, which is essential for the functioning of the canonical Wnt signaling pathway [50]. In mice, inhibiting the canonical Wnt pathway demonstrated a reduction in OA severity by stimulating anticatabolic effects on chondrocytes and anti-fibrotic effects on synovial fibroblasts [51].

T2DM is also known as a condition linked to CLGI. Contributors to persistent inflammation in T2DM include continuous activation signals originating from various sources. These sources encompass dysfunctional adipose tissue, the abnormal metabolic activation of leukocytes, DAMPs released from injured tissue and endothelium, and excessive cytokines discharged by senescent cells [52]. The principal instigator of meta-inflammation in individuals with T2DM is perceived to be the enlargement of adipose tissue, followed by the onset of hyperglycemia prompted by insulin resistance. The well-documented association between malfunction in visceral adipose tissue and the development of insulin resistance is firmly established [53].

Persistent hyperglycemia leads to the formation of covalent linkages between glucose and diverse plasma proteins (albumin, globulins, fibrinogen, collagen), lipids, and nucleic acids. These connections result in the formation of distinct advanced glycation end products (AGEs), which hold a leading position in the pathophysiology of both diabetic complications [54, 55] and OA [56]. The interaction between AGEs and their primary cellular receptor (advanced glycation end product receptor, RAGE) triggers a multitude of signaling pathways, including NF-κB, STAT3, Wnt/β-Catenin, MAPK/ERK, JNK etc. [55–58]. These cascades increase oxidative/nitrosative stress and inflammation. The convergence of these effects underscores the importance of AGEs in mediating the crosstalk between diabetes and OA. The subsequent effects of the AGEs/RAGE interaction encompass compromised insulin signaling, disturbances in metabolic balance, RAGE-induced toxicity in pancreatic β cells, and alterations in epigenetic patterns [56, 59].

AGE accumulation, a hallmark of diabetes, has been implicated in both OA and diabetes-related joint complications [60]. AGEs contribute to matrix alterations in cartilage and promote oxidative/nitrosative stress, leading to accelerated joint degeneration in OA. Furthermore, an insulin-resistant state and obesity are also associated with elevated levels of free fatty acids, which can potentially induce CLGI by activating the TLR4/NF-κB signaling pathway [61], thereby potentially impacting the progression of OA [62]. Leptin, a significant adipokine predominantly released by adipose tissue, is able to stimulate chondrocyte apoptosis while also intensifying the production of cytokines and MMPs by chondrocytes [63].

Recent studies have identified that the induction of the transcription factor Nrf2 serves as a key mechanism for antioxidant defense in the OA cartilage of T2DM patients [64, 65]. Experiments conducted *ex vivo* have indicated a reduction in Nrf2 expression in OA cartilage from T2DM patients compared to non-T2DM cases [65].

Emerging research has revealed shared molecular mechanisms linked to the involvement of specific transcription factors and signaling pathways that contribute to the development and progression of both T2DM and OA. This makes them promising targets for the prevention and treatment of these conditions.

Thus, the progression of both OA and T2DM involves the emergence of CLGI and oxidative/nitrosative stress, especially driven by the interaction between AGEs and their receptor (RAGE). This interaction triggers cascades of pro-inflammatory, pro-oxidant, pro-histolytic, and pro-diabetic signaling pathways linked to NF-κB, STAT3, Wnt, MAPK/ERK, and JNK. These pathways, operating within positive feedback loops, further intensify the systemic inflammatory response and disruptions in metabolic processes.

Potential of chondroitin sulphate in pathogenetic therapy for chronic low-grade inflammation in osteoarthritis and diabetes

A class of long linear polysaccharides known as sulfated glycosaminoglycan (GAG) chains are covalently joined to many core proteins to create proteoglycans (PGs). Nearly all mammalian cell surfaces are surrounded by PGs, which are important pericellular and extracellular matrix components. PGs help to produce favorable microenvironments for a variety of crucial cellular processes, including cell adhesion, proliferation, differentiation, and cell fate decisions [66, 67]. The several GAG moieties present in PGs, such as chondroitin sulfate (CS), heparan sulfate, and keratan sulfate chains, primarily mediate the multifunctional capabilities of PGs.

The most common GAG in the human body is CS, and in particular, chondroitin-4-sulfate and chondroitin-6-sulfate, which are essential components of aggrecan, the main representative of the PGs in the cartilage matrix. These compounds play a role in controlling cell adhesion, proliferation, and differentiation, as well as in the transport of water, amino acids, and lipids within hyaline cartilage. They also contribute to determining the viscosity of synovial fluid, along with the crucial biomechanical characteristics of cartilage tissue, such as its elasticity [68]. Several clinical investigations and meta-analytic assessments have presented evidence supporting the suggestion to employ CS-containing agents as symptomatic slow-acting drugs for osteoarthritis (SYSADOA). These medications exert significant beneficial effects on bone and cartilage tissue structure, as well as on joint function in cases of OA, resulting in a noticeable reduction of chronic pain [69, 70].

According to reports, CS possesses anti-inflammatory properties that prevent the formation of atherosclerotic plaques [71], has effects on lowering blood lipids [72], modulates the immune system [73], demonstrates anti-psoriasis capabilities [74], and shows anti-tumor effects [75]. Additionally, it serves as a preventive measure against complications of diabetes mellitus such as diabetic nephropathy [76], diabetic osteoporosis [77, 78], and diabetic wounds [79, 80].

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The application of CS in the OA treatment appears to be more favorable when compared to glucosamine sulfate, which is known as the most prevalent form of glucosamine. The use of the latter entails certain risks for individuals with diabetes mellitus [1].

It has been observed that patients with diabetes demonstrate a significant increase in PGs depolymerization. When compared to healthy control individuals, the urine of the patients with diabetes contained less CS and more low-sulfated chondroitin sulphate-protein complex [81]. In addition, chondroitin sulfate has a higher 6-sulfated disaccharide content and a lower 4-sulfated disaccharide content in patients with diabetes.

H. Pang *et al*. conducted a comparison of the structural characteristics of CS sourced from various origins, examining its impact on both α-amylase activity and blood glucose levels [82]. Oral administration of CS derived from sharks and pigs effectively lowers postprandial blood glucose levels in both healthy and diabetic mice. The research discovered that CS obtained from various sources exhibited distinct biological effects despite their closely similar molecular weights and disaccharide subunit compositions. This study highlighted the potential of shark and pig CS as novel functional food components for T2DM management, functioning as α-amylase inhibitors.

Notably, obese mice that received PGs from salmon cartilage per os displayed beneficial effects on hyperglycemia and insulin sensitivity. These improvements were linked to a decrease in the expression of critical inflammatory regulators such as TNF-α, IL-6, and C-X-C motif chemokine ligand 2 in adipose tissue [83].

H.X. Zheng *et al*. demonstrated that administering CS for eight weeks led to an amelioration in diabetes symptoms among rats with streptozotocin-induced diabetes. The group that received CS treatment exhibited increased body weight, reduced water and food intake, lowered blood glucose levels, increased bone mineral density, improved bone structure, and decreased femoral osteoclasts and tibia adipocyte counts [77]. Following the CS administration, bone histomorphometric parameters returned to their normal state. There was a notable reduction in the levels of serum pro-inflammatory cytokines (IL-1β, IL-6, and TNF-α) and a significant increase in the activities of serum superoxide dismutase, glutathione peroxidase, and catalase [77, 78]. In the CS-treated group, the levels of serum alkaline phosphatase, cross-linked C-telopeptide of type I collagen, tartrate-resistant acid phosphatase 5b, osteocalcin and receptor activator of NF-κB ligand (RANKL) decreased, while the serum RUNX 2 and osteoprotegerin levels increased. Bone immunohistochemistry results showed that CS can effectively enhance the expression of osteoprotegerin and RUNX2 and reduce the expression of RANKL in rats with diabetes. All of these indicate that CS could prevent diabetic osteoporosis – mainly through decreasing blood glucose, antioxidative and anti-inflammatory properties, as well as regulation of osteoprotegerin/RANKL expression. CS can, therefore, effectively prevent bone loss caused by diabetes.

Some publications elucidate the molecular mechanisms of such CS action, which substantiate the feasibility of using CS as a means of pathogenetic therapy for OA and T2DM. It has been demonstrated that CS is able to mitigate CLGI by impacting the nuclear NF-κB translocation, a process intricately linked to CLGI markers such as serum levels of IL-1, IL-6, and C-reactive protein [84, 85].

Among the mechanisms contributing to the development of CLGI and oxidative/nitrosative stress, the CS capability to mitigate the AGEs generation and RAGE activation assumes a noteworthy role [86, 87]. The AGEs/RAGE mechanism is recognized for triggering a series of signaling pathways, encompassing NF-κB, STAT3, Wnt/β-Catenin, MAPK/ERK, and JNK cascades (Figure 1) [56–58]. In contrast to heparin and hyaluronic acid, CS has been shown to inhibit AGE3-induced upregulation of class A receptor 1 [87].

Moreover, CS has demonstrated its ability to not only inhibit NF-κB activation but also to impact distinct pathways that lead to the generation of pro-inflammatory and pro-oxidant proteins [88–90]. According to recent literature, CS is able to activate the ERK½ and p38 MAPK signaling pathways [91–93] and interfere with the transcriptional activity of STAT3 through a mechanism separate from STAT3 phosphorylation [90].

Combining these effects results in a decrease in the expression of a number of pro-inflammatory enzymes, including phospholipase A2, COX2, and iNOS, as well as cytokines, in particular IL-1 and TNF-α. Furthermore, it has been shown that Wnt3a signaling in fibroblasts can be inhibited by exogenous CS-E [94]. The CS capability to reduce the susceptibility of rabbit chondrocytes to apoptosis is also associated with its ability to suppress NF-κB translocation and the MAPK signaling cascade via p38 and Erk1/2 [95].

CS also possesses anti-inflammatory and antioxidant characteristics, as it can up-regulate Nrf2. These anti-inflammatory, antioxidant, and anticatabolic properties enhance the structural and functional conditions of cartilage, synovial membrane, and subchondral bone [96].

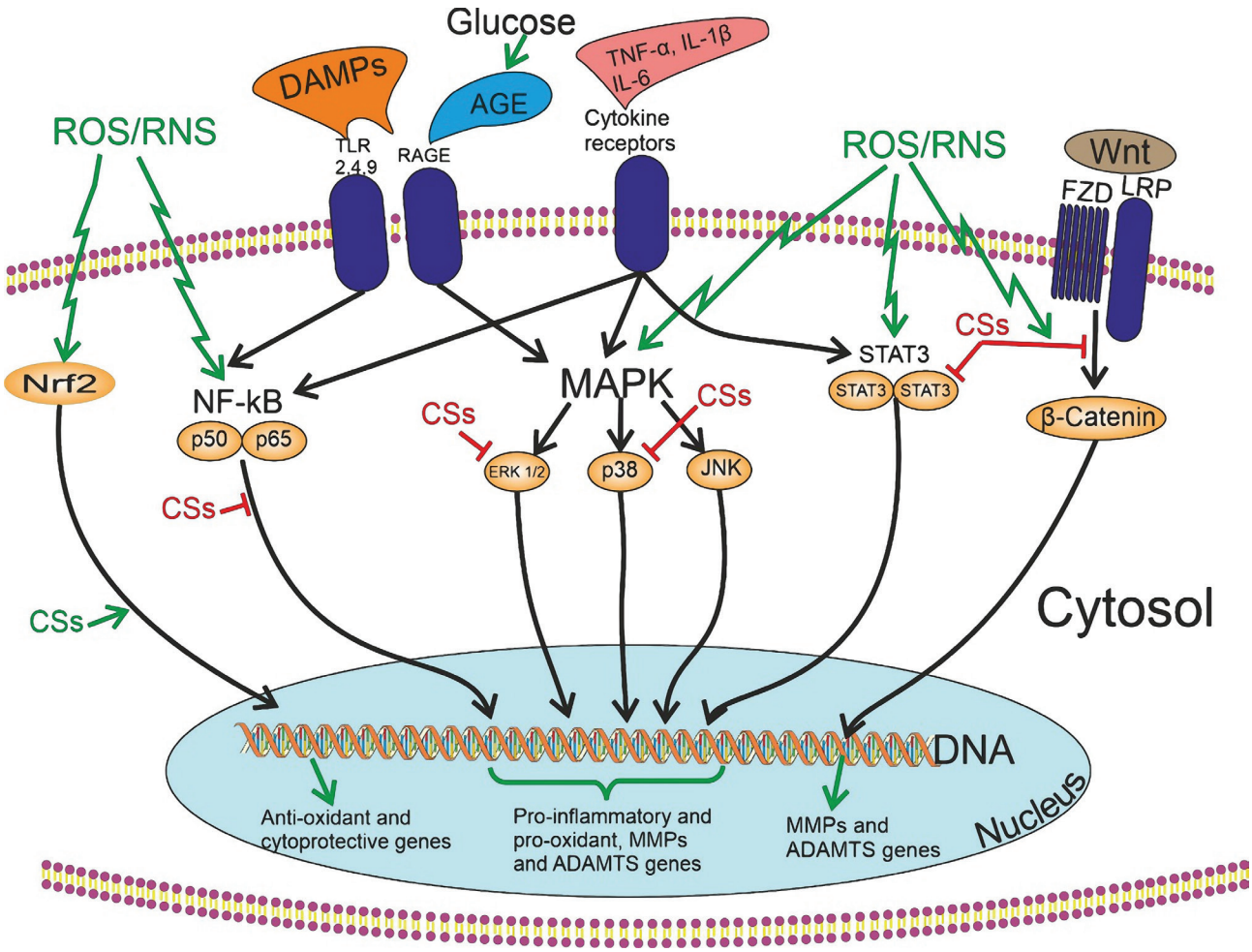

Figure 1: The molecular mechanisms of the anti-inflammatory, antioxidant, and anticatabolic effects of chondroitin sulfates, which are beneficial for the treatment of osteoarthritis in diabetic patients. CSs significantly reduce the nuclear translocation of NF-κB, decrease the phosphorylation of ERK½, abrogate p38 MAPK phosphorylation, and hinder the movement of STAT3 into the nucleus, resulting in a great decrease in their ability to regulate pro-oxidant, pro-inflammatory, MMPs, and ADAMTS gene expression. Additionally, chondroitin sulfates impair Wnt/β-catenin signaling in fibroblasts and demonstrate anti-inflammatory and antioxidant effects by up-regulating Nrf2. Note: ADAMTS – A Disintegrin and Metalloproteinase with Thrombospondin Motifs; AGE –Advanced Glycosylation End Product; CSs – chondroitin sulfates; DAMPs – Damage-Associated Molecular Patterns; ERK½ – Extracellular Signal-Regulated Kinase ½; FZD – Frizzled, receptor in the Wnt signaling pathway; IL – Interleukin; JNK – c-Jun N-terminal Kinase; LRP – Low-Density Lipoprotein Receptor-related Protein; MAPK – Mitogen-Activated Protein Kinase; MMPs – Matrix Metalloproteinases; NF-κB – Nuclear Factor κB; Nrf2 – Nuclear Factor Erythroid 2-Related Factor 2; p38 – p38 Mitogen-Activated Protein Kinase; RAGE – Receptor for Advanced Glycosylation End Products; ROS – Reactive Oxygen Species; RNS – Reactive Nitrogen Species; STAT3 – Signal Transducer and Activator of Transcription 3; TLR – Toll-Like Receptor; TNF –Tumor Necrosis Factor.

Moreover, CS can impact the development of diabetes by shifting the balance between Th1 and Th2 pathways during the progression of diabetes [97].

Some meta-analyses reveal inconsistencies in the outcomes of clinical trials, which are linked to potential biases, brand influences, and variations in sample sizes [69, 70]. In some instances, the CS application failed to enhance pain relief or the functional condition of joints affected by OA [98]. The uncertain outcomes of these clinical trials might be attributed to the diverse forms of CS given to patients (*e.g.*, oral or parenteral), varied raw materials used to produce CS, and the presence or absence of specific physiologically active compounds (PGs compounds, amino acids, minerals etc.).

It has been found that the molecular mass characteristics are relatively comparable among land-derived CS samples (from cows, pigs, and chickens) but notably distinct from aquatic CS samples (derived from sharks, rays, and squids), which exhibit higher molecular mass measurements than the former group [99]. Additionally, the aquatic CS samples demonstrate distinct charge density values exceeding 1.0, attributed to the presence of disulfated disaccharides, in contrast to terrestrial samples with charge density values below 1.0

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due to the absence of such disulphated disaccharides. Furthermore, the fact that CS is derived from animals gives rise to potential safety concerns for consumers due to the potential existence of transmissible infectious agents like prions. These proteins can lead to the development of spongiform encephalopathies in cattle and are also implicated as causal factors in Creutzfeldt-Jakob disease in humans. Alternatively, there could be limitations on its use due to religious reasons.

In a randomized, double-blind, placebo-controlled pilot study, 60 overweight adults with symptomatic OA were assigned to a daily intake of 600 mg of non-animal-derived CS over a period of 12 consecutive weeks [100]. The assessment of knee pain, quality of life, and relevant inflammation markers (levels of C-reactive protein and erythrocyte sedimentation rate) evidences the efficacy of supplementing with non-animal-derived CS in overweight individuals with knee OA. This supplementation demonstrated improvements in knee function, pain mitigation, and inflammation markers.

Artificially produced CS is distinguished by the existence of three- and four-sulfate groups that are absent in CS occurring naturally. This distinction is linked to the reduced biocompatibility of these medications. Conversely, CS sourced from marine organisms has a diminished amount of sulfate groups in the fourth position but shows a notable abundance of negatively charged particles. This characteristic aids in the binding of CS to hyaluronic acid [99, 101, 102].

However, employing CS for the treatment of underlying causes of CLGI also presents inherent risks. These include the possibility of undesirable reactions, such as gastrointestinal disturbances, as well as the potential for interactions with concurrent medications. Furthermore, the consistency and purity of commercially accessible CS preparations can vary, influencing their effectiveness and safety. Some CS forms can exhibit unfavorable effects associated with the risk of thrombosis, as observed in sodium salts of CS A and C, which makes them unsuitable in the treatment for thrombophlebitis [12].

Hence, considering the intricacies of the pathogenesis of systemic inflammatory response and its complications within the context of SYSADOA, a preference should be given to parenteral drugs that do not intensify the blood's tendency to clot.

To sum up, the anti-inflammatory, antioxidant, anti-osteoarthritic and hypoglycaemic characteristics of CS offer a dual advantage by tackling both cartilage damage associated with OA and inflammation linked to diabetes. Although CS seems to be promising as a potential therapeutic means for managing CLGI, additional research is needed to provide a clear understanding of its mode of operation and to evaluate its safety and efficacy in clinical contexts.

So, the experimental investigations have demonstrated that CS possesses the capability to attenuate the generation of AGEs and subsequent activation of RAGE, thereby inhibiting signaling pathways like NF-κB, STAT3, Wnt/β-Catenin, MAPK/ERK, and JNK. As a result, there is a restriction of CLGI and oxidative/nitrosative stress, leading to enhancements in the structural and functional status of cartilage, synovial membrane, and subchondral bone. CS-dependent up-regulation of Nrf2 is accompanied by anti-inflammatory, antioxidant, antidiabetic, and anticatabolic actions, which collectively improve the conditions of cartilage, synovial membrane, and subchondral bone. Moreover, it alters the balance between Th1 and Th2 pathways during the progression of diabetes. The efficacy of CS application in OA is substantiated by numerous clinical studies. However, at present, the antidiabetic effect of CS is mainly supported by experimental research, emphasizing the need to elucidate this effect in clinical settings.

Purified bioactive extract of small marine fish as a promising approach in the management of osteoarthritis and diabetes mellitus

The efficacy of the CS obtained from marine organisms such as salmon, shark, thornback skate, small sea fish, and others has been documented in various studies [103–107]. Standardized small marine fish extract (SSMFE) has demonstrated anti-inflammatory and antioxidant properties and the ability to stimulate the synthesis of new cartilage. These effects are attributed to the composition and manufacturing process of the medications. For instance, the medication Alflutop (manufactured by Biotehnos S.A., Romania) is composed of 0.01 g of a standardized and purified bioactive extract sourced from four species of small marine fish: Black Sea merlin (Merlangius euxinus), Black Sea pufferfish (Alosa tanaica nordmanni), Black Sea sprats (Sprattus phalericus), and Black Sea anchovy (Engraulis encrassicholus ponticus). The medication incorporates chondroitin-4-sulfate and chondroitin-6-sulfate, as well as other glycosaminoglycans such as hyaluronic acid, dermatan sulfate, and keratan sulfate. Furthermore, it contains low molecular weight polypeptides (with molecular weights up to 50 kDa),

a variety of amino acids (including alanine, arginine, proline, hydroxyproline, methionine, glutamate, valine, leucine, isoleucine, serine, threonine, lysine, asparagine, phenylalanine, and tyrosine), and minerals (sodium, potassium, calcium, zinc, magnesium, iron, copper, and manganese) that play exceptional roles in connective tissue metabolism [12]. Alflutop is intended for parenteral administration (both intramuscular and intra-articular) to patients dealing with primary and secondary OA in various areas and with degenerative-dystrophic conditions of the spine and soft tissue pathologies as well.

During clinical trials, SSMFE exhibited various beneficial effects, including chondroprotective qualities (slowing OA progression and osteophyte growth, minimizing articular cartilage matrix degradation), anti-inflammatory and antioxidant properties, as well as analgesic effects. It is noteworthy that significant improvements were observed in joint and spinal function, leading to an improved quality of life [12]. The medication also contributed to reducing the time required for the healing of ulcerative and erosive lesions triggered by non-steroidal anti-inflammatory drugs (NSAID) intake. As a result, researchers recommended its use for patients with OA who are susceptible to NSAID-related gastrointestinal issues [108]. When integrated into the initial treatment for musculoskeletal conditions alongside NSAIDs, SSMFE (Alflutop) accelerated the onset of analgesic effects and reduced the reliance on NSAIDs [109].

Furthermore, SSMFE has been found to be an effective means of comprehensive pharmacotherapy of back pain. This can be attributed to its capability to alleviate afferent nociceptive activity by mitigating inflammation in spinal structures and diminishing central sensitization [110, 111]. Moreover, the positive impact of this preparation includes a prolonged analgesic effect lasting up to a month following the completion of the treatment course.

The mechanisms underlying SSMFE's impact on inflammation and regeneration have been extensively investigated in various experimental studies, which have unveiled its ability to suppress the expression and release of pro-inflammatory cytokines (such as IL-1β, IL-6, and IL-8). Furthermore, it can hinder the activity of aggressive proteolytic enzymes, including MMPs and ADAMTS4. This inhibition results in a decreased occurrence of alterations and apoptosis in chondrocytes and osteocytes [112–115]. A pivotal facet of SSMFE transcriptional repression of pro-inflammatory cytokines involves the diminishment of NF-κB expression, particularly its p50 subunit (NF-κB1) [115].

In the presence of IL-1β and TNF-α stimulation, SSMFE effectively countered the decline in chondrocyte numbers through a dual approach: by impeding apoptosis and enhancing their proliferation rate [116].

In our opinion, the SSMFE's ability to suppress NF-κB represents a valuable discovery that not only elucidates the efficacy of SSMFE in treating not only OA but also offers promise for its potential antidiabetic properties, given that NF-κB plays an important role in the pathogenesis of T2DM.

Previously, it has been reported that the consumption of lean sea fish at a daily rate of 75–100 g had a favorable impact on T2DM [117]. Recently, it has been revealed that enzymatically derived fish protein hydrolysates serve as a rich reservoir of biologically active peptides endowed with anti-inflammatory, antioxidant, antidiabetic, anti-cancer, antimicrobial, anti-hypertensive, and wound healing activities [118, 119]. The hydrolysates were subjected to characterization and evaluation of their capacity to hinder enzymes relevant to conditions related to metabolic syndrome. The hydrolysates produced were tested for their capability to inhibit enzymes such as angiotensin-1-converting enzyme (associated with blood pressure regulation), acetylcholinesterase (linked to nervous system maintenance), and dipeptidyl peptidase 4 (related to T2DM development) [120]. Among the observed amino acids in the fish protein hydrolysate samples, glutamate and aspartate were the most prevalent.

An intriguing aspect that underscores the superiority of SSMFE when compared to other constituents of the SYSADOA for patients with both OA and T2DM at risk of developing CLGI is its diminished immunogenicity. Conversely, formulations containing GAG-peptide complexes sourced from cattle's bone marrow and intercostal cartilage carry the potential to trigger the production of autoantibodies. Prior to initiating treatment with such medications, patients are advised to consult a physician to rule out the presence of systemic autoimmune disorders. It is noteworthy that these conditions are recognized to coincide with the CLGI progression [12].

In our view, the advantages outlined in relation to SSMFE-derived formulations can likely be attributed not solely to their manufacturing process involving degreasing and deproteinisation but also to the incorporation of various other biologically active components (such as amino acids and minerals) present in this intricate formulation, alongside CS.

Cartilage damage is known to result in impaired amino acid metabolism involving processes such as arginine biosynthesis as well as metabolism of amino acids such as serine, threonine and proline [121], which are

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indeed present in SSMFE. Moreover, Alflutop contains 7 amino acids (phenylalanine, tyrosine, alanine, isoleucine, leucine, valine, and glutamate), the deficiency of which is significantly correlated with decreased insulin secretion and elevated fasting or 2-hour glucose levels [122]. Deficiency of tyrosine, alanine, isoleucine, and glutamate is also significantly associated with an increased risk of developing T2DM after accounting for potential comorbidities.

An additional significant aspect that distinguishes SSMFE from other SYSADOAs is its incorporation of essential minerals crucial for tissue restoration. As an illustration, magnesium demonstrates anti-inflammatory and antioxidative properties, contributing to the enhancement of cartilage matrix formation and the promotion of chondrocyte proliferation, both of which effectively counter osteoarthritis. Conversely, zinc plays a key role in antioxidant defense and the suppression of prostaglandin synthesis. It also stimulates the growth and development of cartilage while assisting in the differentiation of mesenchymal stem cells into chondrocytes within osteochondral defects [123]. Additionally, potassium, magnesium, and zinc are essential for restoring mineral deficits in individuals with uncontrolled hyperglycemia, particularly those undergoing chronic diuretic therapy. Deficiencies in these minerals, as is known, can potentially lead to carbohydrate intolerance [124, 125].

To summarise, the effects of SSMFE observed under experimental conditions are corroborated by clinical studies, validating its capacity to curtail joint inflammation, deter cartilage degradation, enhance joint functionality, and alleviate pain and disability among patients with OA. The presence of certain amino acids and minerals (such as potassium, magnesium, and zinc) in the formulation of SSMFE-based preparations (like Alflutop) might not only amplify the effectiveness of OA treatment but also limit carbohydrate metabolic disorders.

Conclusions

OA and T2DM share a common pathogenetic mechanism, manifested through the involvement of CLGI and oxidative/nitrosative stress pathways. These mechanisms are closely linked to the functional state of pro-inflammatory, pro-oxidant, pro-histolytic, and pro-diabetic signaling pathways.

SSMFE can be regarded as a promising means in the therapy of OA and T2DM. It serves as an illustration of medications with the ability to alleviate NF-κB-mediated inflammatory and metabolic disorders. SSMFE demonstrated anti-inflammatory, antioxidant, antidiabetic, anti-cancer, antimicrobial, anti-hypertensive, and wound healing properties. The biologically active substances found in SSMFE (CS, amino acids and minerals) are characterized by a high safety profile, thus promoting a more effective correction of carbohydrate metabolism.

Conflict of interest

The authors declare no conflict of interest.

References

1. Veronese N, Cooper C, Reginster JY, Hochberg M, Branco J, Bruyère O, Chapurlat R, Al-Daghri N, Dennison E, Herrero-Beaumont G, Kaux JF, Maheu E, Rizzoli R, Roth R, Rovati LC, Uebelhart D, Vlaskovska M, Scheen A. Type 2 diabetes mellitus and osteoarthritis. Semin Arthritis Rheum. 2019 Aug;49(1):9-19. doi: 10.1016/j.semarthrit.2019.01.005.
2. Centers for Disease Control and Prevention (CDC). Prevalence of doctor-diagnosed arthritis and arthritis-attributable activity limitation--United States, 2010-2012. MMWR Morb Mortal Wkly Rep. 2013 Nov 8;62(44):869-73.
3. Centers for Disease Control and Prevention (CDC). National Diabetes Statistics Report. Atlanta: GA Centers for Disease Control and Prevention, U.S. Dept of Health and Human Services; 2017.
4. Lawrence RC, Felson DT, Helmick CG, Arnold LM, Choi H, Deyo RA, Gabriel S, Hirsch R, Hochberg MC, Hunder GG, Jordan JM, Katz JN, Kremers HM, Wolfe F; National Arthritis Data Workgroup. Estimates of the prevalence of arthritis and other rheumatic conditions in the United States. Part II. Arthritis Rheum. 2008 Jan;58(1):26-35. doi: 10.1002/art.23176.
5. Piva SR, Susko AM, Khoja SS, Josbeno DA, Fitzgerald GK, Toledo FG. Links between osteoarthritis and diabetes: implications for management from a physical activity perspective. Clin Geriatr Med. 2015 Feb;31(1):67-87.
6. Chow YY, Chin KY. The Role of Inflammation in the Pathogenesis of Osteoarthritis. Mediators Inflamm. 2020 Mar 3;2020:8293921. doi: 10.1155/2020/8293921.
7. Lauwers M, Au M, Yuan S, Wen C. COVID-19 in Joint Ageing and Osteoarthritis: Current Status and Perspectives. Int J Mol Sci. 2022 Jan 10;23(2):720. doi: 10.3390/ijms23020720.
8. Rose MJ, LaValley MP, Jafarzadeh SR, Costello KE, Shah N, Lee S, Borrelli B, Messier SP, Neogi T, Kumar D. Impact of COVID-19 Pandemic on Physical Activity, Pain, Mood, and Sleep in Adults with Knee Osteoarthritis. J Meas Phys Behav. 2022 Dec;5(4):294-298. doi: 10.1123/jmpb.2022-0019.
9. Wang X, Hunter D, Xu J, Ding C. Metabolic triggered inflammation in osteoarthritis. Osteoarthritis Cartilage. 2015 Jan;23(1):22-30. doi: 10.1016/j.joca.2014.10.002.
10. Gratal P, Lamuedra A, Medina JP, Bermejo-Álvarez I, Largo R, Herrero-Beaumont G and Mediero A Purinergic System Signaling in Metainflammation-Associated Osteoarthritis. Front. Med. 2020;7:506. doi: 10.3389/fmed.2020.00506

11. Warmink K, Vinod P, Korthagen NM, Weinans H, Rios JL. Macrophage-Driven Inflammation in Metabolic Osteoarthritis: Implications for Biomarker and Therapy Development. International Journal of Molecular Sciences. 2023; 24(7):6112. https://doi.org/10.3390/ijms24076112
12. Rekalov DH, Golovach IY, Kostenko VO, Shevchenko MV, Kulynych RL. Pharmacotherapy of Diseases of Musculoskeletal System in Patients with Risk of COVID-19-Associated Thromboinflammation. Ukr Rheumatol J. 2022;(1):1-9. doi:10.32471/rheumatology.2707-6970.87.16832
13. Chisari E, Wouthuyzen-Bakker M, Friedrich AW, Parvizi J. The relation between the gut microbiome and osteoarthritis: A systematic review of literature. PLoS One. 2021 Dec 16;16(12):e0261353. doi: 10.1371/journal.pone.0261353.
14. Ramires LC, Santos GS, Ramires RP, da Fonseca LF, Jeyaraman M, Muthu S, Lana AV, Azzini G, Smith CS, Lana JF. The Association between Gut Microbiota and Osteoarthritis: Does the Disease Begin in the Gut? Int J Mol Sci. 2022 Jan 27;23(3):1494. doi: 10.3390/ijms23031494.
15. Wei Z, Li F, Pi G. Association Between Gut Microbiota and Osteoarthritis: A Review of Evidence for Potential Mechanisms and Therapeutics. Front. Cell. Infect. Microbiol. 2022;12:812596. doi: 10.3389/fcimb.2022.812596
16. Du Z, You X, Wu D, Huang S, Zhou Z. Rhythm disturbance in osteoarthritis. Cell Commun Signal. 2022 May 24;20(1):70. doi: 10.1186/s12964-022-00891-7.
17. Rogers N, Meng QJ. Tick tock, the cartilage clock. Osteoarthritis Cartilage. 2023 May 23:S1063-4584(23)00800-2. doi: 10.1016/j.joca.2023.05.010.
18. Guida F, Rocco M, Luongo L, Persiani P, Vulpiani MC, Nusca SM, Maione S, Coluzzi F. Targeting Neuroinflammation in Osteoarthritis with Intra-Articular Adelmidrol. Biomolecules. 2022 Oct 11;12(10):1453. doi: 10.3390/biom12101453.
19. Rezuș E, Cardoneanu A, Burlui A, Luca A, Codreanu C, Tamba BI, Stanciu GD, Dima N, Bădescu C, Rezuș C. The Link Between Inflammaging and Degenerative Joint Diseases. Int J Mol Sci. 2019 Jan 31;20(3):614. doi: 10.3390/ijms20030614.
20. Motta F, Barone E, Sica A, Selmi C. Inflammaging and Osteoarthritis. Clin Rev Allergy Immunol. 2023 Apr;64(2):222-238. doi: 10.1007/s12016-022-08941-1.
21. Roškar S, Hafner-Bratkovič I. The Role of Inflammasomes in Osteoarthritis and Secondary Joint Degeneration Diseases. Life (Basel). 2022 May 13;12(5):731. doi: 10.3390/life12050731.
22. Lee KH, Kang TB. The Molecular Links between Cell Death and Inflammasome. Cells. 2019 Sep 10;8(9):1057. doi: 10.3390/cells8091057.
23. Lambert C, Zappia J, Sanchez C, Florin A, Dubuc JE, Henrotin Y. The Damage-Associated Molecular Patterns (DAMPs) as Potential Targets to Treat Osteoarthritis: Perspectives From a Review of the Literature. Front Med (Lausanne). 2021 Jan 18;7:607186. doi: 10.3389/fmed.2020.607186.
24. Riegger J, Brenner RE. Pathomechanisms of Posttraumatic Osteoarthritis: Chondrocyte Behavior and Fate in a Precarious Environment. Int J Mol Sci. 2020; 21(5):1560. doi: 10.3390/ijms21051560.
25. Yao Q, Wu X, Tao C, Gong W, Chen M, Qu M, Zhong Y, He T, Chen S, Xiao G. Osteoarthritis: pathogenic signaling pathways and therapeutic targets. Signal Transduct Target Ther. 2023 Feb 3;8(1):56. doi: 10.1038/s41392-023-01330-w.
26. Gauci S, Stanton H, Little C, Fosang A. Proteoglycan and collagen degradation in osteoarthritis. 2017. doi: 10.1007/978-3-319-45803-8_3.
27. Akimov OY, Kostenko VO. Role of NF-κB transcriptional factor activation during chronic fluoride intoxication in development of oxidative-nitrosative stress in rat's gastric mucosa. J Trace Elem Med Biol. 2020 Apr 19;61:126535. doi: 10.1016/j.jtemb.2020.126535.
28. Frenkel' YD, Cherno VS, Kostenko VO. Effect of NF-κB and Nrf2 transcription factor modulators on indicators of oxidative-nitrosative stress in skeletal muscles of rats under chronic hypomelatoninemia and carbohydrate-lipid diet. Fiziol Zh. 2023;69(2):11-18. doi: 10.15407/fz69.02.011.
29. Sa L, Wei X, Huang Q, Cai Y, Lu D, Mei R, Hu X. Contribution of salidroside to the relieve of symptom and sign in the early acute stage of osteoarthritis in rat model. J Ethnopharmacol. 2020 Sep 15;259:112883. doi: 10.1016/j.jep.2020.112883.
30. Yelins'ka AM, Liashenko LI, Kostenko VO. Quercetin potentiates antiradical properties of epigallocatechin-3-gallate in periodontium of rats under systemic and local administration of lipopolisaccharide of Salmonella typhi. Wiad Lek. 2019 Aug 31;72(8):1499-1503.
31. Yavtushenko IV, Nazarenko SM, Katrushov OV, Kostenko VO. Quercetin limits the progression of oxidative and nitrosative stress in the rats' tissues after experimental traumatic brain injury. Wiad Lek. 2020;73(10):2127-2132.
32. Frenkel' YD, Zyuzin VO, Cherno VS, Kostenko VO. Effect of epigallocatechin-3-gallate and quercetin on the production of reactive oxygen and nitrogen species in liver of rats exposed to round-the-clock light and kept on carbohydrate-lipid diet. Fiziol Zh. 2022;68(1):20-27. doi: 10.15407/fz68.01.020.
33. Frenkel Y, Cherno V, Kostenko H, Kostenko V. Resveratrol attenuates the development of nitro-oxidative stress in the liver of rats under a round-the-clock lighting and high-carbohydrate-lipid diet. Romanian Journal of Diabetes, Nutrition and Metabolic Diseases. 2023;30(1):48-54. doi: 10.46389/rjd-2023-1217.
34. Kostenko V, Akimov O, Gutnik O, Kostenko H, Kostenko V, Romantseva T, Morhun Y, Nazarenko S, Taran O. Modulation of redox-sensitive transcription factors with polyphenols as pathogenetically grounded approach in therapy of systemic inflammatory response. Heliyon. 2023 Apr;9(4):e15551. doi: 10.1016/j.heliyon.2023.e15551.
35. Ansari MY, Ahmad N, Haqqi TM. Oxidative stress and inflammation in osteoarthritis pathogenesis: Role of polyphenols. Biomed Pharmacother. 2020 Sep;129:110452. doi: 10.1016/j.biopha.2020.110452.
36. Yelins'ka AM, Akimov OYe, Kostenko VO. Role of AP-1 transcriptional factor in development of oxidative and nitrosative stress in periodontal tissues during systemic inflammatory response. Ukr Biochim J. 2019;91(1):80-85. doi: 10.15407/ubj91.01.080.
37. Wang F, Guo Z, Yuan Y. STAT3 speeds up progression of osteoarthritis through NF-κB signaling pathway. Exp Ther Med. 2020;19(1):722-728. doi:10.3892/etm.2019.8268.
38. Peng HZ, Yun Z, Wang W, Ma BA. Dual specificity phosphatase 1 has a protective role in osteoarthritis fibroblast-like synoviocytes via inhibition of the MAPK signaling pathway. Mol Med Rep. 2017 Dec;16(6):8441-8447. doi: 10.3892/mmr.2017.7617.
39. Loeser RF, Erickson EA, Long DL. Mitogen-activated protein kinases as therapeutic targets in osteoarthritis. Curr Opin Rheumatol. 2008 Sep;20(5):581-6. doi: 10.1097/BOR.0b013e3283090463.
40. Li Z, Dai A, Yang M, Chen S, Deng Z, Li L. p38MAPK Signaling Pathway in Osteoarthritis: Pathological and Therapeutic Aspects. J Inflamm Res. 2022 Feb 3;15:723-734. doi: 10.2147/JIR.S348491.
41. Chen B, Ning K, Sun ML, Zhang XA. Regulation and therapy, the role of JAK2/STAT3 signaling pathway in OA: a systematic

review. Cell Commun Signal. 2023 Apr 3;21(1):67. doi: 10.1186/s12964-023-01094-4.
42. Lee SY, Lee SH, Na HS, Kwon JY, Kim GY, Jung KA, Cho KH, Kim SA, Go EJ, Park MJ, Baek JA, Choi SY, Jhun JY, Park SH, Kim SJ and Cho ML. The Therapeutic Effect of STAT3 Signaling-Suppressed MSC on Pain and Articular Cartilage Damage in a Rat Model of Monosodium Iodoacetate-Induced Osteoarthritis. Front. Immunol. 2018;9:2881. doi: 10.3389/fimmu.2018.02881
43. Xie L, Xie H, Chen C, Tao Z, Zhang C, Cai L. Inhibiting the PI3K/AKT/NF-κB signal pathway with nobiletin for attenuating the development of osteoarthritis: in vitro and in vivo studies. Food Funct. 2019 Apr 17;10(4):2161-2175. doi: 10.1039/c8fo01786g.
44. Song Y, Hao D, Jiang H, Huang M, Du Q, Lin Y, Liu F, Chen B. Nrf2 Regulates CHI3L1 to Suppress Inflammation and Improve Post-Traumatic Osteoarthritis. J Inflamm Res. 2021 Aug 24;14:4079-4088. doi: 10.2147/JIR.S310831.
45. Yuan T, Wang X, Cai D, Liao M, Qin J. Anti-arthritic and cartilage damage prevention via regulation of Nrf2/HO-1 signaling by glabridin on osteoarthritis. Arab J Chem. 2021;14(3):103207. doi: 10.1016/j.arabjc.2021.103207.
46. 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.
47. Yan Z, Qi W, Zhan J, Lin Z, Lin J, Xue X, Pan X, Zhou Y. Activating Nrf2 signalling alleviates osteoarthritis development by inhibiting inflammasome activation. J Cell Mol Med. 2020 Nov;24(22):13046-13057. doi: 10.1111/jcmm.15905.
48. Li W, Tao C, Mao M, Zhu K. The Nrf2/HMGB1/NF-κB axis modulates chondrocyte apoptosis and extracellular matrix degradation in osteoarthritis. Acta Biochim Biophys Sin (Shanghai). 2023 May 26;55(5):818-830. doi: 10.3724/abbs.2023078.
49. Yasuhara R, Ohta Y, Yuasa T, Kondo N, Hoang T, Addya S, Fortina P, Pacifici M, Iwamoto M, Enomoto-Iwamoto M. Roles of β-catenin signaling in phenotypic expression and proliferation of articular cartilage superficial zone cells. Lab Invest. 2011 Dec;91(12):1739-52. doi: 10.1038/labinvest.2011.144.
50. Zhou Y, Wang T, Hamilton JL, Chen D. Wnt/β-catenin Signaling in Osteoarthritis and in Other Forms of Arthritis. Curr Rheumatol Rep. 2017 Sep;19(9):53. doi: 10.1007/s11926-017-0679-z.
51. Lietman C, Wu B, Lechner S, Shinar A, Sehgal M, Rossomacha E, Datta P, Sharma A, Gandhi R, Kapoor M, Young PP. Inhibition of Wnt/β-catenin signaling ameliorates osteoarthritis in a murine model of experimental osteoarthritis. JCI Insight. 2018 Feb 8;3(3):e96308. doi: 10.1172/jci.insight.96308.
52. van de Vyver M. Immunology of chronic low-grade inflammation: relationship with metabolic function. J Endocrinol. 2023 Feb 23;257(1):e220271. doi: 10.1530/JOE-22-0271.
53. Calderón-DuPont D, Torre-Villalvazo I, Díaz-Villaseñor A. Is insulin resistance tissue-dependent and substrate-specific? The role of white adipose tissue and skeletal muscle. Biochimie. 2023 Jan;204:48-68. doi: 10.1016/j.biochi.2022.08.021.
54. Singh VP, Bali A, Singh N, Jaggi AS. Advanced glycation end products and diabetic complications. Korean J Physiol Pharmacol. 2014 Feb;18(1):1-14. doi: 10.4196/kjpp.2014.18.1.1.
55. Khalid M, Petroianu G, Adem A. Advanced Glycation End Products and Diabetes Mellitus: Mechanisms and Perspectives. Biomolecules. 2022 Apr 4;12(4):542. doi: 10.3390/biom12040542.
56. He CP, Chen C, Jiang XC, Li H, Zhu LX, Wang PX, Xiao T. The role of AGEs in pathogenesis of cartilage destruction in osteoarthritis. Bone Joint Res. 2022 May;11(5):292-300. doi: 10.1302/2046-3758.115.BJR-2021-0334.R1.
57. Wu X, Shi X, Chen X, Yin Z. Advanced glycation end products regulate the receptor of AGEs epigenetically. Front Cell Dev Biol. 2023 Feb 14;11:1062229. doi: 10.3389/fcell.2023.1062229.
58. Martín-Carro B, Martín-Vírgala J, Fernández-Villabrille S, Fernández-Fernández A, Pérez-Basterrechea M, Navarro-González JF, Donate-Correa J, Mora-Fernández C, Dusso AS, Carrillo-López N, Panizo S, Naves-Díaz M, Fernández-Martín JL, Cannata-Andía JB, Alonso-Montes C. Role of Klotho and AGE/RAGE-Wnt/β-Catenin Signalling Pathway on the Development of Cardiac and Renal Fibrosis in Diabetes. Int J Mol Sci. 2023 Mar 9;24(6):5241. doi: 10.3390/ijms24065241.
59. Lee J, Yun JS, Ko SH. Advanced Glycation End Products and Their Effect on Vascular Complications in Type 2 Diabetes Mellitus. Nutrients. 2022 Jul 27;14(15):3086. doi: 10.3390/nu14153086.
60. Courties A, Sellam J. Osteoarthritis and type 2 diabetes mellitus: What are the links? Diabetes Res Clin Pract. 2016 Dec;122:198-206. doi: 10.1016/j.diabres.2016.10.021.
61. Rogero MM, Calder PC. Obesity, Inflammation, Toll-Like Receptor 4 and Fatty Acids. Nutrients. 2018 Mar 30;10(4):432. doi: 10.3390/nu10040432.
62. Wang Y, Wluka AE, Hodge AM, English DR, Giles GG, O'Sullivan R, Cicuttini FM. Effect of fatty acids on bone marrow lesions and knee cartilage in healthy, middle-aged subjects without clinical knee osteoarthritis. Osteoarthritis Cartilage. 2008 May;16(5):579-83. doi: 10.1016/j.joca.2007.09.007.
63. Courties A, Gualillo O, Berenbaum F, Sellam J. Metabolic stress-induced joint inflammation and osteoarthritis. Osteoarthritis Cartilage. 2015 Nov;23(11):1955-65. doi: 10.1016/j.joca.2015.05.016.
64. Dodson M, Shakya A, Anandhan A, Chen J, Garcia JGN, Zhang DD. NRF2 and Diabetes: The Good, the Bad, and the Complex. Diabetes. 2022 Dec 1;71(12):2463-2476. doi: 10.2337/db22-0623.
65. Vaamonde-Garcia C, Courties A, Pigenet A, Laiguillon MC, Sautet A, Houard X, Kerdine-Römer S, Meijide R, Berenbaum F, Sellam J. The nuclear factor-erythroid 2-related factor/heme oxygenase-1 axis is critical for the inflammatory features of type 2 diabetes-associated osteoarthritis. J Biol Chem. 2017 Sep 1;292(35):14505-14515. doi: 10.1074/jbc.M117.802157.
66. Mikami T, Kitagawa H. Sulfated glycosaminoglycans: their distinct roles in stem cell biology. Glycoconj J. 2017 Dec;34(6):725-735. doi: 10.1007/s10719-016-9732-9.
67. Volpi N. Chondroitin Sulfate Safety and Quality. Molecules. 2019 Apr 12;24(8):1447. doi: 10.3390/molecules24081447.
68. Messina OD, Vidal Wilman M, Vidal Neira LF. Nutrition, osteoarthritis and cartilage metabolism. Aging Clin Exp Res. 2019 Jun;31(6):807-813. doi: 10.1007/s40520-019-01191-w.
69. Honvo G, Bruyère O, Geerinck A, *et al*. Efficacy of Chondroitin Sulfate in Patients with Knee Osteoarthritis: A Comprehensive Meta-Analysis Exploring Inconsistencies in Randomized, Placebo-Controlled Trials. Adv Ther. 2019 May;36(5):1085-1099. doi: 10.1007/s12325-019-00921-w.
70. Honvo G, Reginster JY, Rabenda V, *et al*. Safety of Symptomatic Slow-Acting Drugs for Osteoarthritis: Outcomes of a Systematic Review and Meta-Analysis. Drugs Aging. 2019 Apr;36(Suppl 1):65-99. doi: 10.1007/s40266-019-00662-z.
71. Melgar-Lesmes P, Garcia-Polite F, Del-Rey-Puech P, Rosas E, Dreyfuss JL, Montell E, Vergés J, Edelman ER, Balcells M. Treatment with chondroitin sulfate to modulate inflammation and

atherogenesis in obesity. Atherosclerosis. 2016 Feb;245:82-7. doi: 10.1016/j.atherosclerosis.2015.12.016.

72. Seol BG, Kim JH, Woo M, Song YO, Choi YH, Noh JS, Cho EJ. Skate cartilage extracts containing chondroitin sulfate ameliorates hyperlipidemia-induced inflammation and oxidative stress in high cholesterol diet-fed LDL receptor knockout mice in comparison with shark chondroitin sulfate. Nutr Res Pract. 2020 Jun;14(3):175-187. doi: 10.4162/nrp.2020.14.3.175.
73. Hatano S, Watanabe H. Regulation of Macrophage and Dendritic Cell Function by Chondroitin Sulfate in Innate to Antigen-Specific Adaptive Immunity. Front Immunol. 2020 Mar 3;11:232. doi: 10.3389/fimmu.2020.00232.
74. Vergés J, Montell E, Herrero M, Perna C, Cuevas J, Pérez M, Möller I. Clinical and histopathological improvement of psoriasis with oral chondroitin sulfate: a serendipitous finding. Dermatol Online J. 2005 Mar 1;11(1):31.
75. Pudełko A, Wisowski G, Olczyk K, Koźma EM. The dual role of the glycosaminoglycan chondroitin-6-sulfate in the development, progression and metastasis of cancer. FEBS J. 2019 May;286(10):1815-1837. doi: 10.1111/febs.14748.
76. Gomes CL, Leão CL, Venturotti C, Barreira AL, Guimarães G, Fonseca RJ, Fortunato RS, Mourão PA, Delgado AG, Takiya CM, Leite M Jr. The protective role of fucosylated chondroitin sulfate, a distinct glycosaminoglycan, in a murine model of streptozotocin-induced diabetic nephropathy. PLoS One. 2014 Sep 5;9(9):e106929. doi: 10.1371/journal.pone.0106929.
77. Zheng HX, Chen J, Zu YX, Wang EZ, Qi SS. Chondroitin Sulfate Prevents STZ Induced Diabetic Osteoporosis through Decreasing Blood Glucose, AntiOxidative Stress, Anti-Inflammation and OPG/RANKL Expression Regulation. Int J Mol Sci. 2020 Jul 26;21(15):5303. doi: 10.3390/ijms21155303.
78. Qi SS, Shao ML, Sun Z, Chen SM, Hu YJ, Li XS, Chen J, Zheng HX, Yue TL. Chondroitin Sulfate Alleviates Diabetic Osteoporosis and Repairs Bone Microstructure via Anti-Oxidation, Anti-Inflammation, and Regulating Bone Metabolism. Front Endocrinol (Lausanne). 2021 Oct 27;12:759843. doi: 10.3389/fendo.2021.759843.
79. Shah SA, Sohail M, Khan SA, Kousar M. Improved drug delivery and accelerated diabetic wound healing by chondroitin sulfate grafted alginate-based thermoreversible hydrogels. Mater Sci Eng C Mater Biol Appl. 2021 Jul;126:112169. doi: 10.1016/j.msec.2021.112169.
80. Zhang Y, Chen W, Feng W, Fang W, Han X, Cheng C. Multifunctional chondroitin sulfate based hydrogels for promoting infected diabetic wounds healing by chemo-photothermal antibacterial and cytokine modulation. Carbohydr Polym. 2023 Aug 15;314:120937. doi: 10.1016/j.carbpol.2023.120937.
81. Zhao T, Lu X, Davies NM, Gong Y, Guo J, Zhang H, Li Z, Hong J, Fu G, Li P. Diabetes results in structural alteration of chondroitin sulfate in the urine. J Pharm Pharm Sci. 2013;16(3):486-93. doi: 10.18433/j3gs3c.
82. Pang H, Li J, Miao Z, Li SJ. Inhibitory effects of chondroitin sulfate on alpha-amylase activity: A potential hypoglycemic agent. Int J Biol Macromol. 2021 Aug 1;184:289-296. doi: 10.1016/j.ijbiomac.2021.06.062.
83. Hirose S, Asano K, Nakane A. Attenuation of obesity-induced inflammation in mice orally administered with salmon cartilage proteoglycan, a prophylactic agent. Biochem Biophys Res Commun. 2017 Mar 11;484(3):480-485. doi: 10.1016/j.bbrc.2017.01.056.
84. Kim BH, Lee KH, Chung EY, Chang YS, Lee H, Lee CK, Min KR, Kim Y. Inhibitory effect of chroman carboxamide on interleukin-6 expression in response to lipopolysaccharide by preventing nuclear factor-kappaB activation in macrophages. Eur J Pharmacol. 2006 Aug 14;543(1-3):158-65. doi: 10.1016/j.ejphar.2006.05.042.
85. Stabler TV, Huang Z, Montell E, Vergés J, Kraus VB. Chondroitin sulphate inhibits NF-κB activity induced by interaction of pathogenic and damage associated molecules. Osteoarthritis Cartilage. 2017 Jan;25(1):166-174. doi: 10.1016/j.joca.2016.08.012.
86. Iannuzzi C, Borriello M, D'Agostino A, Cimini D, Schiraldi C, Sirangelo I. Protective effect of extractive and biotechnological chondroitin in insulin amyloid and advanced glycation end product-induced toxicity. J Cell Physiol. 2019 Apr;234(4):3814-3828. doi: 10.1002/jcp.27153.
87. Nishinaka T, Mori S, Yamazaki Y, Niwa A, Wake H, Yoshino T, Nishibori M, Takahashi H. A comparative study of sulphated polysaccharide effects on advanced glycation end-product uptake and scavenger receptor class A level in macrophages. Diab Vasc Dis Res. 2020 Jan-Feb;17(1):1479164119896975. doi: 10.1177/1479164119896975.
88. Jomphe C, Gabriac M, Hale TM, *et al*. Chondroitin sulfate inhibits the nuclear translocation of nuclear factor-kappaB in interleukin-1beta-stimulated chondrocytes. Basic Clin Pharmacol Toxicol. 2008;102(1):59-65. doi:10.1111/j.1742-7843.2007.00158.x
89. du Souich P, García AG, Vergés J, Montell E. Immunomodulatory and anti-inflammatory effects of chondroitin sulphate. J Cell Mol Med. 2009 Aug;13(8A):1451-63. doi: 10.1111/j.1582-4934.2009.00826.x.
90. Andrés RM, Payá M, Montesinos MC, *et al*. Potential antipsoriatic effect of chondroitin sulfate through inhibition of NF-κB and STAT3 in human keratinocytes. Pharmacol Res. 2013;70(1):20-26. doi:10.1016/j.phrs.2012.12.004
91. Holzmann J, Brandl N, Zemann A, Schabus R, Marlovits S, Cowburn R, Huettinger M. Assorted effects of TGFbeta and chondroitinsulfate on p38 and ERK1/2 activation levels in human articular chondrocytes stimulated with LPS. Osteoarthritis Cartilage. 2006 Jun;14(6):519-25. doi: 10.1016/j.joca.2005.12.005.
92. Martín-de-Saavedra MD, del Barrio L, Cañas N, Egea J, Lorrio S, Montell E, Vergés J, García AG, López MG. Chondroitin sulfate reduces cell death of rat hippocampal slices subjected to oxygen and glucose deprivation by inhibiting p38, NFκB and iNOS. Neurochem Int. 2011 May;58(6):676-83. doi: 10.1016/j.neuint.2011.02.006.
93. Yamagishi Y, Someya A, Imai K, Nagao J, Nagaoka I. Evaluation of the anti-inflammatory actions of various functional food materials including glucosamine on synovial cells. Mol Med Rep. 2017;16(2):1353-1359. doi:10.3892/mmr.2017.6691
94. Willis CM, Klüppel M. Inhibition by chondroitin sulfate E can specify functional Wnt/β-catenin signaling thresholds in NIH3T3 fibroblasts. J Biol Chem. 2012 Oct 26;287(44):37042-56. doi: 10.1074/jbc.M112.391490.
95. Jomphe C, Gabriac M, Hale TM, Héroux L, Trudeau LE, Deblois D, Montell E, Vergés J, du Souich P. Chondroitin sulfate inhibits the nuclear translocation of nuclear factor-kappaB in interleukin-1beta-stimulated chondrocytes. Basic Clin Pharmacol Toxicol. 2008 Jan;102(1):59-65. doi: 10.1111/j.1742-7843.2007.00158.x.
96. Ju C, Hou L, Sun F, Zhang L, Zhang Z, Gao H, Wang L, Wang D, Lv Y, Zhao X. Anti-oxidation and Antiapoptotic Effects of Chondroitin Sulfate on 6-Hydroxydopamine-Induced Injury Through the Up-Regulation of Nrf2 and Inhibition of Mitochondria-Mediated Pathway. Neurochem Res. 2015 Jul;40(7):1509-19. doi: 10.1007/s11064-015-1628-8.
97. Sireesh D, Dhamodharan U, Ezhilarasi K, Vijay V, Ramkumar KM. Association of NF-E2 Related Factor 2 (Nrf2) and inflammatory cytokines in recent onset Type 2 Diabetes Mellitus. Sci Rep. 2018 Mar 23;8(1):5126. doi: 10.1038/s41598-018-22913-6.

 https://doi.org/10.46389/rjd-2023-1425

98. Rojas-Briones V, Harrison-Muñoz S, Irarrázaval S. Is chondroitin sulfate effective for osteoarthritis? Medwave. 2017 Apr 26;17(Suppl2):e6929. Spanish, English. doi: 10.5867/medwave.2017.6929.
99. Valoti E, Miraglia N, Bianchi D, *et al*. Shark-like chondroitin sulphate and process for the preparation thereof. United States Patent US 8,664,196 B2. March 4, 2014.
100. Rondanelli M, Braschi V, Gasparri C, Nichetti M, Faliva MA, Peroni G, Naso M, Iannello G, Spadaccini D, Miraglia N, Putignano P, Alalwan TA, Perna S. Effectiveness of Non-Animal Chondroitin Sulfate Supplementation in the Treatment of Moderate Knee Osteoarthritis in a Group of Overweight Subjects: A Randomized, Double-Blind, Placebo-Controlled Pilot Study. Nutrients. 2019 Aug 29;11(9):2027. doi: 10.3390/nu11092027.
101. Valcarcel J, Novoa-Carballal R, Pérez-Martín RI, Reis RL, Vázquez JA. Glycosaminoglycans from marine sources as therapeutic agents. Biotechnol Adv. 2017 Nov 1;35(6):711-725. doi: 10.1016/j.biotechadv.2017.07.008.
102. Abdallah MM, Fernández N, Matias AA, Bronze MDR. Hyaluronic acid and Chondroitin sulfate from marine and terrestrial sources: Extraction and purification methods. Carbohydr Polym. 2020 Sep 1;243:116441. doi: 10.1016/j.carbpol.2020.116441.
103. Michelle NY, Shanthi G, Loqman MY. Effect of orally administered Channa striatus extract against experimentally-induced osteoarthritis in rabbits. Int J Appl Res Vet Med. 2004;2(3):171-175.
104. López-Senra E, Casal-Beiroa P, López-Álvarez M, Serra J, González P, Valcarcel J, Vázquez JA, Burguera EF, Blanco FJ, Magalhães J. Impact of Prevalence Ratios of Chondroitin Sulfate (CS)- 4 and -6 Isomers Derived from Marine Sources in Cell Proliferation and Chondrogenic Differentiation Processes. Mar Drugs. 2020 Jan 31;18(2):94. doi: 10.3390/md18020094.
105. Eckert T, Jährling-Butkus M, Louton H, Burg-Roderfeld M, Zhang R, Zhang N, Hesse K, Petridis AK, Kožár T, Steinmeyer J, Schauer R, Engelhard P, Kozarova A, Hudson JW, Siebert HC. Efficacy of Chondroprotective Food Supplements Based on Collagen Hydrolysate and Compounds Isolated from Marine Organisms. Mar Drugs. 2021 Sep 26;19(10):542. doi: 10.3390/md19100542.
106. Henrotin Y, Antoine C, Zwerts E, Neutelings T, Bouvret E. Oral supplementation with fish cartilage hydrolysate accelerates joint function recovery in rat model of traumatic knee osteoarthritis. Food Sci Nutr. 2021 Apr 10;9(6):2877-2885. doi: 10.1002/fsn3.2244.
107. Vassallo V, Stellavato A, Russo R, Cimini D, Valletta M, Alfano A, Pedone PV, Chambery A, Schiraldi C. Molecular Fingerprint of Human Pathological Synoviocytes in Response to Extractive Sulfated and Biofermentative Unsulfated Chondroitins. Int J Mol Sci. 2022 Dec 14;23(24):15865. doi: 10.3390/ijms232415865.
108. Drozdov VN, Shikh EV, Serebrova SY, Abrosimov AG, Starodubtsev AK. Alflutop – in modern symptom-modifying osteoarthritis therapy. Ter Arkh. 2019 May 15;91(5):134-140. doi: 10.26442/00403660.2019.05.000169.
109. Alexeeva LI, Sharapova EP, Taskina EA, *et al*. Multicenter double-blind randomized placebo-controlled trial of the symptom- and structure-modifying effect of alflutop in patients with knee osteoarthrosis. Communication 1. Evaluation of the symptom-modifying effect of the drug. Rheumatology Science and Practice. 2013;51(5):532-538. doi: 10.14412/1995-4484-2013-1545.
110. Samartsev IN, Zhivolupov SA, Barantsevich ER, Danilov AB. The evaluation of the efficacy of Alflutop in the complex treatment of patients with chronic lower back pain (the observational study ZEITNOT). Zh Nevrol Psikhiatr Im S S Korsakova. 2021;121(2):24-30. doi: 10.17116/jnevro202112102124.
111. Sarvilina IV, Danilov AB. Comparative analysis of the use of symptomatic slow acting drugs for osteoarthritis containing chondroitin sulfate or affecting its biosynthesis in patients with non-specific low back pain. Zh Nevrol Psikhiatr Im S S Korsakova. 2023;123(1):81-96. doi: 10.17116/jnevro202312301181.
112. Olariu L., Dumitriu B., Buse E., Rosoiu N. The in vitro effect of Alflutop product on some extracellular signaling factors involved in the osteoarticular pathology inflammation. Analele AOSR. 2015;4(2):7–18.
113. Olariu L, Pyatigorskaya N, Dumitriu B, *et al*. "In vitro" chondro-restitutive capacity of Alflutop proved on chondrocytes cultures. Rom Biotechnol Lett. 2016;22(16):12047-12053.
114. Olariu L, Dumitriu B, Ene DM, *et al*. Alflutop modulates "in vitro" relevant mechanisms of osteoarthritic pathology. Academy of Romanian Scientists Annals – Series on Biological Sciences. 2017;6(1):100-113.
115. Buse E, Dumitriu B, Olariu L, *et al*. Cellular and molecular activity of a standardized small fish extract in an experimental model of primary human cartilage cells. Rom J Rheumatol. 2018;27:23-31.
116. Olariu L, Dumitriu B, Craciun L, Buse E, Rosoiu N, Bojinca M, Papacocea T. The in vitro influence of a pharmaceutically active small sea fish extract on apoptosis and proliferation mechanisms amplified by inflammatory conditions. Farmacia. 2018;66(3):524-529. doi: 10.31925/farmacia.2018.3.19.
117. Rylander C, Sandanger TM, Engeset D, Lund E. Consumption of lean fish reduces the risk of type 2 diabetes mellitus: a prospective population based cohort study of Norwegian women. PLoS One. 2014 Feb 24;9(2):e89845. doi: 10.1371/journal.pone.0089845.
118. Kumar L, Shakila R, Jeyasekaran G. In vitro anti-cancer, anti-diabetic, anti-inflammation and wound healing properties of collagen peptides derived from unicorn leatherjacket (Aluterus monoceros) at different hydrolysis. Turkish Journal of Fisheries and Aquatic Sciences. 2019; 19:551-560. doi: 10.4194/1303-2712-v19_7_02.
119. Yathisha UG, Ishani B, Iddya K, Mamatha BS. Antihypertensive activity of fish protein hydrolysates and its peptides. Crit Rev Food Sci Nutr. 2019;59(15):2363-2374. doi: 10.1080/10408398.2018.1452182.
120. Naik AS, Whitaker RD, Albrektsen S, Solstad RG, Thoresen L and Hayes M Mesopelagic Fish Protein Hydrolysates and Extracts: A Source of Novel Anti-Hypertensive and Anti-Diabetic Peptides. Front. Mar. Sci. 2021;8:719608. doi: 10.3389/fmars.2021.719608.
121. Southan J, McHugh E, Walker H, Ismail HM. Metabolic Signature of Articular Cartilage Following Mechanical Injury: An Integrated Transcriptomics and Metabolomics Analysis. Front Mol Biosci. 2020;7:592905.doi:10.3389/fmolb.2020.592905.
122. Vangipurapu J, Stancáková A, Smith U, Kuusisto J, Laakso M. Nine Amino Acids Are Associated With Decreased Insulin Secretion and Elevated Glucose Levels in a 7.4-Year Follow-up Study of 5,181 Finnish Men. Diabetes. 2019;68(6):1353-1358. doi:10.2337/db18-1076.
123. Li G, Cheng T, Yu X. The Impact of Trace Elements on Osteoarthritis. Front Med (Lausanne). 2021;8:771297. doi:10.3389/fmed. 2021.771297.
124. Joe MC, Mae SA, Arshag DM. The Role of Micronutrients in Managing Diabetes. Diabetes. 2009;22(4):214-218. https://doi.org/10.2337/diaspect.22.4.214.
125. Dubey P, Thakur V, Chattopadhyay M. Role of Minerals and Trace Elements in Diabetes and Insulin Resistance. Nutrients. 2020 Jun 23;12(6):1864. doi: 10.3390/nu12061864.