Pharmacogn. Res., 2027; 19(2):709-718.
https://www.phcogres.com

Original Article

Camel Milk Powder with *Artemisia sieberi* Extract Inhibit Nuclear Transcription Factor-κB RelA and p50 Activation, Cyclooxygenase-2 Expression and Prostaglandin E2 Production in Interleukin-1β-Stimulated Human Osteoarthritic Chondrocytes

Mona S Almujaydil[1], Waheeba E Ahmed[1], Ruya Mohammed Alshebrami[1], Waleed Al Abdulmonem[2,*]

[1]Department of Food Science and Human Nutrition, College of Agriculture and Food, Qassim University, Buraidah, SAUDI ARABIA.
[2]Department of Pathology, College of Medicine, Qassim University, Buraidah, SAUDI ARABIA.

ABSTRACT

Background: Osteoarthritis is a chronic inflammatory joint disease in which interleukin-1β (IL-1β)-induced Nuclear Factor Kappa B (NF-κB) activation promotes the expression of Cyclooxygenase-2 (COX-2) and Prostaglandin E2 (PGE2), contributing to cartilage degeneration. Camel Milk Powder (CMP) and *Artemisia sieberi* (Shih) possess anti-inflammatory properties, but their combined effects have not been fully investigated. **Objectives:** To evaluate the effects of CMP, Shih, and their Combined Formulation (CMP-Shih) on IL-1β-induced NF-κB activation, COX-2 expression, and PGE2 production in human osteoarthritic chondrocytes. **Materials and Methods:** Primary human osteoarthritic chondrocytes isolated from osteoarthritic articular cartilage and were pretreated with CMP, Shih, or CMP-Shih before IL-1β stimulation. Gene expression was analyzed by TaqMan real-time PCR, protein expression by Western blotting, and NF-κB activity and PGE2 production by ELISA. The involvement of NF-κB signaling was confirmed using the selective NF-κB inhibitor Bay 11-7082. **Results:** CMP and Shih individually inhibited IL-1β-induced NF-κB activation and reduced p65 and p50 expression. CMP-Shih produced significantly greater inhibition than either treatment alone ($p<0.05$). The combination also significantly suppressed COX-2 mRNA and protein expression and decreased PGE2 production. Bay 11-7082 confirmed that these anti-inflammatory effects were mediated, at least in part, through inhibition of NF-κB signaling. **Conclusion:** CMP-Shih effectively attenuated IL-1β-induced inflammatory responses in human osteoarthritic chondrocytes and may represent a promising natural therapeutic candidate for osteoarthritis and other inflammatory joint disorders.

Keywords: Camel milk, Chondrocytes, COX-2, IL-1β, NF-κB, Osteoarthritis, PGE2, Shih.

Correspondence:
Prof. Waleed Al Abdulmonem
Department of Pathology, College of Medicine, Qassim University, Buraidah, SAUDI ARABIA.
Email: dr.waleedmonem@qu.edu.sa
ORCID: 0000-0003-2984-9262

Received: 15-05-2026;
Revised: 18-08-2026;
Accepted: 29-09-2026.

INTRODUCTION

Osteoarthritis (OA), the most common form of arthritis, affects millions of individuals worldwide and is associated with reduced quality of life and substantial healthcare costs (Yan *et al.*, 2026). Although numerous systemic and biomechanical risk factors have been implicated in disease initiation and progression, the precise etiology of this chronic, degenerative joint disorder remains incompletely defined (Yan *et al.*, 2026; Zhang *et al.*, 2022). Currently, no disease-modifying pharmacological therapies are available, and in advanced stages, surgical joint replacement remains the only definitive treatment option (O'Connor *et al.*, 2022). Articular cartilage contains a single specialized cell type, the chondrocyte. Under normal physiological conditions, chondrocytes maintain a dynamic balance between anabolic and catabolic activities, which is essential for preserving the structural and functional integrity of cartilage. These cells constitutively express low levels of inflammatory mediators, including Interleukin (IL)-1β, Cyclooxygenase (COX)-2, and various proteolytic enzymes, supporting controlled matrix turnover and remodeling (Hashemi-Afzal *et al.*, 2024). In OA, however, the production of inflammatory mediators, particularly COX-2 and its downstream product Prostaglandin E2 (PGE2) is markedly increased beyond physiological levels, contributing to cartilage breakdown and pain (Lee *et al.*, 2023; Li *et al.*, 2026). Evidence indicates that excessive PGE2 enhances the expression of metalloproteinases and alters proteoglycan metabolism,

OPEN ACCESS
Manuscript TECHNOMEDIA

DOI: 10.5530/pres.20270268

Copyright Information :
Copyright Author (s) 2027 Distributed under Creative Commons CC-BY 4.0

Publishing Partner : Manuscript Technomedia. [www.mstechnomedia.com]

thereby accelerating extracellular matrix degradation (Huang *et al.*, 2024; Moghbeli *et al.*, 2023). IL-1 is recognized as a central pro-inflammatory and catabolic cytokine in the pathogenesis of OA and represents a potential therapeutic target (Wei *et al.*, 2026; Wen *et al.*, 2025). IL-1β-induced overexpression of COX-2 has been widely documented in OA and plays a significant role in driving inflammation and cartilage destruction (Wen *et al.*, 2025; Wu *et al.*, 2023). Selective suppression of PGE2 production through inhibition of COX-2 activity has therefore emerged as a rational strategy for anti-inflammatory and anti-arthritic therapy (El-Saghier *et al.*, 2026). Consequently, the development of selective COX-2 inhibitors has gained considerable attention, and targeting COX-2 is considered a promising therapeutic approach in OA management. Another important molecular target in OA is the nuclear transcription Factor-Kappa B (NF-κB), which regulates the expression of numerous genes involved in inflammatory responses, including COX-2 (Hoffmann *et al.*, 2025). Given the critical contribution of inflammation to articular tissue damage, both *in vitro* and *in vivo* studies have investigated the role of NF-κB signaling pathways in OA pathogenesis (Hoffmann *et al.*, 2025; Zou *et al.*, 2025). NF-κB is implicated in multiple pathological processes, including inflammation, cartilage degradation, cell proliferation, angiogenesis, and pannus formation (Hoffmann *et al.*, 2025; Feng & Wu, 2022). Therefore, pharmacological modulation or inhibition of NF-κB signaling represents another potentially effective therapeutic strategy for OA.

Artemisia sieberi (Shih) is a greenish-purple perennial shrub characterized by alternate, hairy, pedate leaves and typically reaching 50-150 cm in height. It grows in arid regions, valleys, and open fields and is widely used in traditional Middle Eastern medicine. Shih has been utilized for the management of gangrenous and infected ulcers and is also known for its anthelmintic and antimalarial properties. Several studies have demonstrated hepatoprotective, nephroprotective, antioxidant, and antibacterial effects of its extracts. The ethanolic extract of Shih has been shown to inhibit angiogenesis in both human umbilical vein endothelial cell cultures and chick chorioallantoic membrane models (Jamal *et al.*, 2023). Phytochemical analyses have identified compounds such as cis-davanone, camphor, terpinol, E-nerolidol, and linalool, which exhibit notable antibacterial and antifungal activities (Selim *et al.*, 2026). Camel Milk (CM) constitutes an important dietary component in many arid and semi-arid regions worldwide. It contains a variety of bioactive constituents, including zinc, CM-Shih-related bioactive components, peptides, and mono- and polyunsaturated fatty acids (Behrouz *et al.*, 2022; El-Kattawy *et al.*, 2021). These bioactive substances have been associated with potential therapeutic benefits in conditions such as tuberculosis, asthma, gastrointestinal disorders, and jaundice. Camel milk exhibits greater compositional variability than cow milk, with its content influenced by factors such as age, breed, diet, lactation stage, geographic location, and season. Camel whey proteins contain endogenous proteases, including cathepsin D and chymotrypsin A, along with a broad range of soluble proteins. Depending on its composition, CM has demonstrated anti-inflammatory, antibacterial, and anti-hepatitis activities (Behrouz *et al.*, 2022; Swelum *et al.*, 2021). CM-Shih is considered a principal contributor to the anti-inflammatory and antioxidant properties attributed to camel milk-based formulations (Behrouz *et al.*, 2022; Swelum *et al.*, 2021). In the present study, we demonstrate that CM-Shih significantly suppresses IL-1β-induced COX-2 expression and PGE2 production in primary human OA chondrocytes. These inhibitory effects were associated with reduced activation of the transcription factor NF-κB. Collectively, our findings suggest a novel mechanism of action for this dietary-derived formulation and indicate that CM-Shih or its bioactive constituents may exert cartilage-protective effects in osteoarthritis.

MATERIALS AND METHODS

Specimen Selection and Preparation of Articular Chondrocytes

This *in vitro* experimental study was carried out from February 2025 to April 2026 using primary chondrocytes isolated from osteoarthritic articular cartilage. Cells were pretreated with CMP, Shih, or CMP-Shih before IL-1β stimulation. This study was conducted in compliance with the ethical principles outlined in the Declaration of Helsinki of the World Medical Association. Ethical approval for the use of discarded cartilage samples from Osteoarthritis (OA) patients was obtained (IRB LOG #14-252). Cartilage specimens were collected from knee joints of patients undergoing total joint replacement surgery. Macroscopic assessment of cartilage degeneration was performed by staining femoral head specimens with India ink. Areas exhibiting a smooth articular surface (designated as "unaffected cartilage") were carefully excised. Inclusion criteria: Cartilage specimens were obtained from patients clinically diagnosed with knee OA who underwent total knee replacement surgery. Only discarded articular cartilage samples from patients with confirmed OA diagnosis and adequate macroscopically preserved cartilage areas suitable for chondrocyte isolation were included in this study. Exclusion criteria: Cartilage specimens were excluded from patients with inflammatory joint diseases (such as rheumatoid arthritis or autoimmune arthritis), infectious arthritis, previous joint infection, malignancy, or cartilage samples showing extensive degradation, contamination, insufficient tissue quantity, or poor cell viability following isolation. Primary chondrocytes were isolated from these regions using enzymatic digestion as previously reported (Rellmann *et al.*, 2021). The isolated cells were seeded at a density of 1.2×10^6 cells/mL in six-well culture plates (Millipore, Darmstadt, Germany) and maintained in complete DMEM medium following established protocols (Lu *et al.*, 2023). The preparation of chondrocytes has been summarized in Figure 1.

Preparation of Camel Milk and *Artemisia sieberi* Extract

Camel Milk Powder (CMP) was procured from a local supplier in Saudi Arabia. *Artemisia sieberi* plant material was collected in January 2025 from the Al-Shimasiyah governorate in the Qassim region, Kingdom of Saudi Arabia. The extract was prepared according to previously described methods (Mohammed *et al.*, 2022; Wei *et al.*, 2024). Briefly, aerial parts (shoots) of the plant were washed thoroughly, shade-dried, finely powdered, and stored at room temperature. Dried leaves and stems were ground separately into coarse powder. Extraction was carried out by soaking the plant material (1:10 w/v) in distilled water at 90°C using a tea-bag method. The mixture was then kept at room temperature for 20-24 hr in a glass container. Following extraction, the suspension was centrifuged at 4000 rpm for 10 min using a Hettich UNIVERSAL 320R centrifuge. The supernatant was concentrated by evaporating the solvent at 60-80°C under reduced pressure with a BÜCHI Rotavapor R-200 rotary evaporator. The resulting crude aqueous extract (CM-shih) was stored in opaque containers at -20°C until further use.

Cytotoxicity Assessment of OA Chondrocytes Treated with CM-shih or IL-1β

Human OA chondrocytes (1.2×10^6 cells/well) cultured in complete DMEM-F12 medium were serum-starved overnight at approximately 70-80% confluence prior to treatment. Cells were exposed to CM-shih under various experimental conditions. Cell viability was evaluated using the CellTiter-Glo® Luminescent Cell Viability Assay (Promega, Fitchburg, WI, USA; catalog #G7573), following the manufacturer's instructions. In selected experiments, serum-deprived OA chondrocytes were pretreated with CM-shih (10-100 mg/mL) for 3 hr and subsequently stimulated with 5-10 ng/mL IL-1β (Sigma-Aldrich, St. Louis, MO, USA) for 12-48 hr. Untreated cells were included as negative controls.

Treatment of Primary Human OA Chondrocytes

Primary OA chondrocytes were pre-incubated with different concentrations of CM-shih for 2 hr prior to stimulation with IL-1β, as described previously (Sundar *et al.*, 2024). Cells cultured without CM-shih or IL-1β served as controls. All experiments were performed within 4-6 days of primary culture to minimize the risk of chondrocyte dedifferentiation.

Quantitative Real-Time PCR (qRT-PCR)

Gene expression analysis was performed using quantitative real-time polymerase chain reaction (qRT-PCR) to determine mRNA levels of COX-2, Collagen type II (COL2A1), collagen type X (COL10A1), Aggrecan (ACAN), and SRY-box transcription factor 9 (SOX-9). GAPDH served as the internal reference gene. Total RNA was extracted from OA chondrocytes using a total RNA isolation kit (Ambion, CA, USA; catalog #AM1560). First-strand cDNA synthesis was carried out from 1 µg of total RNA using the SuperScript First-Strand cDNA synthesis kit (Affymetrix Inc., OH, USA; catalog #75780). Primer sequences used for amplification were for human COX-2 (PTGS2): F 5′CAGCCATACAGCAAATCCTTG3′, R5′CAAATGTGATCTGGATGTCAAC3′ and for human GAPDH: F5′ GAAGGTGAAGGTCGGAGTCA, R5′GAAGATGGTGATGGGATTTC3′. PCR amplification was performed using SYBR Green or TaqMan reagents (Applied Biosystems, Foster City, CA, USA) on a StepOne Real-Time PCR System (Applied Biosystems). Thermal cycling conditions consisted of an initial denaturation step at 95°C for 10 min, followed by 40 cycles of 95°C for 15 sec and 60°C for 60 sec. Melting curve analysis was conducted to confirm amplification specificity. Relative gene expression was calculated by normalizing target gene expression to GAPDH and comparing treated samples with untreated controls (Khan *et al.*, 2025; Rasheed *et al.*, 2019).

Western Blot Analysis

Protein expression in treated and untreated primary human OA chondrocytes was analyzed by western blotting as previously described (Hamdan *et al.*, 2026; Rasheed *et al.*, 2019). Total protein lysates were prepared using Pierce RIPA buffer (Thermo Scientific, LI, USA; catalog #89901). Equal amounts of protein (35 µg per lane) were separated by SDS-PAGE using 10% resolving and 4% stacking gels and transferred onto PVDF membranes (Millipore, Bedford, MA, USA; catalog #IPVH00010). Membranes were blocked with non-fat dry milk prepared in Tris-buffered saline containing 0.1% Tween-20 (TBS-T) and incubated with primary antibodies (dilution range 1:200-1:1000) specific to the proteins of interest. After washing, membranes were incubated with HRP-conjugated secondary antibodies (1:1000 dilution). Protein bands were detected using Enhanced Chemiluminescence (ECL) reagents (GE Healthcare, Milwaukee, WI, USA). The resulting images were analyzed with UN-SCAN-IT software (Silk Scientific Corporation, Orem, UT). Each band was measured five independent times, and the average intensity (pixels per band) was calculated. The values were then normalized against appropriate loading controls and presented as mean±Standard Deviation (SD), followed by suitable statistical analysis.

Prostaglandin E_2 (PGE_2) Enzyme-Linked Immunosorbent Assay (ELISA)

PGE_2 levels in the culture supernatants of treated and untreated OA chondrocytes were measured using a commercially available ELISA kit (Cayman Chemical Company, MI, USA; catalogue #514010) in accordance with the manufacturer's protocol. Absorbance values were recorded using an Anthos Zenyth 3100 multimode microplate reader (Salzburg, Austria) (Abdellatif *et al.*, 2020).

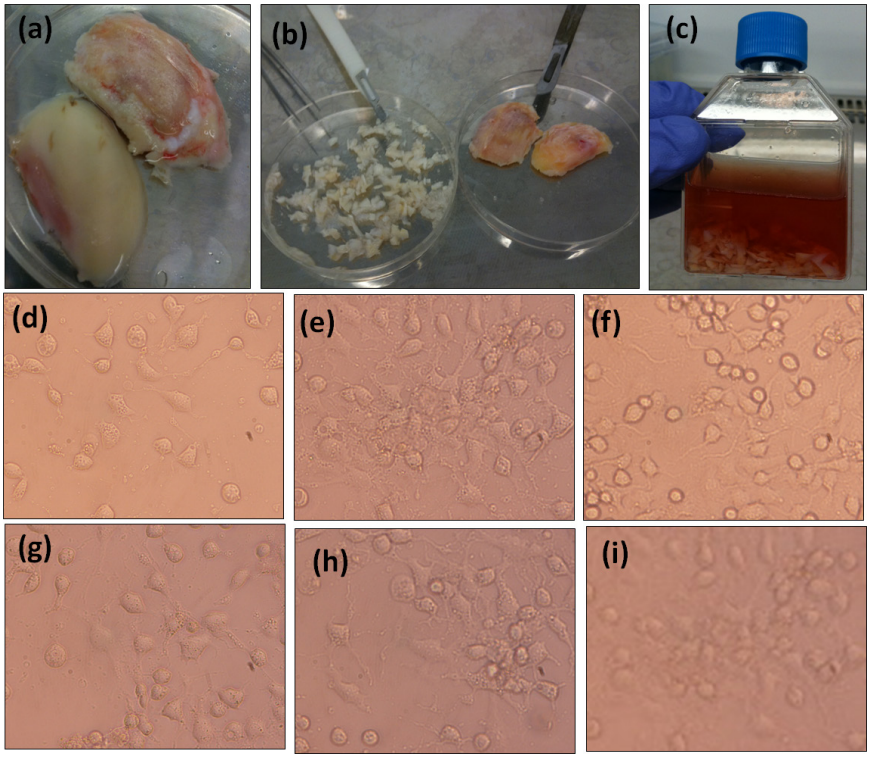


Figure 1: Isolation and experimental treatment of primary human osteoarthritic (OA) chondrocytes. (a) Discarded knee joint specimens obtained from OA patients. (b) Cartilage tissue carefully dissected from OA knee samples. (c) Enzymatic digestion of cartilage fragments using pronase and collagenase to release chondrocytes. (d) Primary human OA chondrocytes following 24 hr of culture. (e) Serum-deprived primary OA chondrocytes serving as untreated controls. (f) Cells stimulated with IL-1β alone. (g) Cells pre-incubated with camel milk powder (CMP) for 2 hr prior to IL-1β stimulation. (h) Cells pretreated with a combination of CMP and *Artemisia sieberi* extract (shih) (CMP-shih) followed by IL-1β stimulation. (i) Cells pretreated with shih extract alone before IL-1β exposure. Concentrations of all treatments are indicated within the respective panels. All experimental groups were subjected to 12 hr serum starvation before treatment.

NF-κB Activity Assay

The inhibitory effect of CM-shih on NF-κB p65 activity in IL-1β-stimulated OA chondrocytes was evaluated in nuclear extracts using a transcription factor ELISA kit (Abcam, MA, USA; catalog #ab133128), following the manufacturer's instructions. Optical density was measured using an Anthos Zenyth 3100 multimode microplate reader (Salzburg, Austria). Nuclear extracts were prepared according to our previously published method (Rasheed *et al*., 2016; Bonder *et al*., 1999).

Statistical Analysis

All experiments were conducted in duplicate and independently repeated a minimum of three times using primary chondrocytes isolated from Osteoarthritis (OA) cartilage obtained from age- and sex-matched donors. Data are presented as the mean±Standard Error of the Mean (SEM), unless otherwise specified. Statistical analyses were carried out using GraphPad Prism version 5 (GraphPad Software, San Diego, CA, USA). Comparisons among multiple groups were evaluated using One-Way Analysis of Variance (ANOVA) followed by Tukey's multiple comparison test, while experiments involving two independent variables were analyzed using two-way ANOVA with Bonferroni's *post hoc* test. A *p*-value of less than 0.05 was considered statistically significant.

RESULTS

Preparation and characterization of chondrocytes from osteoarthritic patients

Primary human Osteoarthritic (OA) chondrocytes were isolated from knee articular cartilage using an enzymatic digestion protocol involving sequential treatment with pronase and collagenase. A schematic overview of the isolation procedure is presented in Figures 1a-1c. To verify the preservation of the chondrogenic phenotype following isolation, we evaluated the expression of established cartilage-specific markers, including Type II collagen (COL2A1), aggrecan, and SOX-9 mRNA, which are widely recognized indicators of differentiated chondrocytes. Primary OA chondrocytes were cultured in monolayer at a density of 1.2×10^6 cells/mL in complete DMEM supplemented with 10% fetal calf serum and maintained at 37°C in a humidified atmosphere containing 5% CO_2. Gene expression analysis demonstrated sustained expression of COL2A1, aggrecan, and SOX-9 mRNA, whereas COL10A1 mRNA, a marker of hypertrophic differentiation, was not detected. These findings confirmed maintenance of the chondrocyte phenotype under the applied culture conditions. To minimize the risk of dedifferentiation, experiments were conducted using cells between 3 and 6 days after plating. Representative micrographs

of isolated chondrocytes before and after exposure to CMP, shih, or CMP-Shih (40× magnification) are shown in Figures 1d-1i. No noticeable changes in cell number or morphology were observed following treatment, suggesting that these agents were not cytotoxic under the tested conditions.

Effect of CMP, shih, and CMP-Shih on viability of human OA chondrocytes, alone and in combination with IL-1β

Cell viability was evaluated following 24-hr treatment with increasing concentrations of CMP (0-50 μg/mL), shih (0-80 μg/mL), or CMP-Shih (50 μg/mL) (Figures 2a-2c). Serum-starved OA chondrocytes were treated as indicated, and viability was quantified using the CellTiter-Glo Luminescent Cell Viability Assay (Promega). No significant differences in cell viability were detected across the tested concentrations of CMP (Figure 2a) or shih (Figure 2b) after 24 hr (p>0.05). Furthermore, stimulation with IL-1β alone or in combination with CMP-Shih (50 μg/mL) did not significantly affect chondrocyte viability compared with untreated controls (Figure 2c). These results indicate that the concentrations used in subsequent experiments did not compromise cell survival.

CMP-Shih suppresses IL-1β-induced COX-2 mRNA and protein expression in primary human OA chondrocytes

To determine whether CMP-Shih modulates inflammatory gene expression, primary OA chondrocytes (70-80% confluence) were pretreated with CMP-Shih (50 μg/mL) for 2 hr, followed by stimulation with IL-1β (10 ng/mL) for 24 hr. COX-2 mRNA expression was quantified by sensitive and specific quantitative RT-PCR and compared with untreated controls.

IL-1β stimulation significantly increased COX-2 mRNA levels relative to unstimulated cells. Pretreatment with CMP-Shih markedly attenuated this IL-1β-induced upregulation (Figure 3a). Although CMP or shih alone also reduced COX-2 mRNA expression, the inhibitory effect was substantially greater when both agents were combined. To determine whether these transcriptional changes were reflected at the protein level, total cellular proteins were analyzed by Western blotting. IL-1β markedly increased COX-2 protein expression, whereas pretreatment with CMP-Shih (50 μg/mL) significantly reduced this induction (p<0.01; Figure 3b). Consistent with mRNA findings, combined treatment exerted a stronger inhibitory effect than either compound alone.

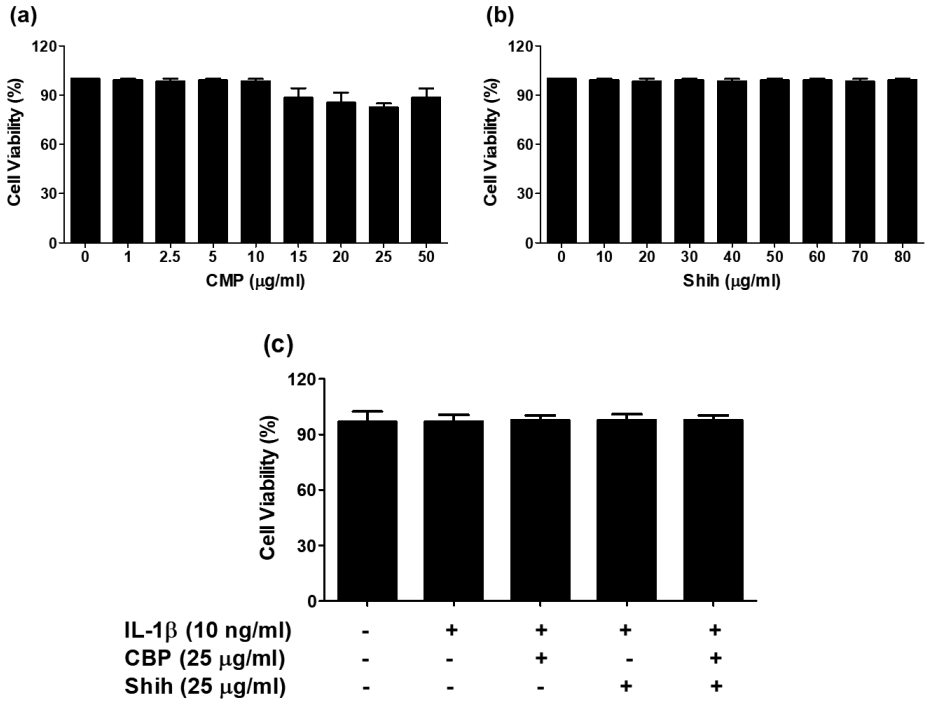


Figure 2: Influence of CMP-shih on the viability of human OA chondrocytes. (a) Primary human OA chondrocytes were incubated with graded concentrations of CMP (0-50 μg/mL) for 24 hr, and cell viability was measured. (b) Cells were treated with increasing concentrations of shih extract (0-80 μg/mL) for 24 hr, followed by viability assessment. (c) Cells were pretreated with CMP, shih, or CMP-shih for 2 hr and subsequently stimulated with IL-1β for 24 hr before determining percentage viability. Serum deprivation was performed for 12 hr prior to all treatments. Viability was quantified using the CellTiter-Glo® Luminescent Cell Viability Assay (Promega).

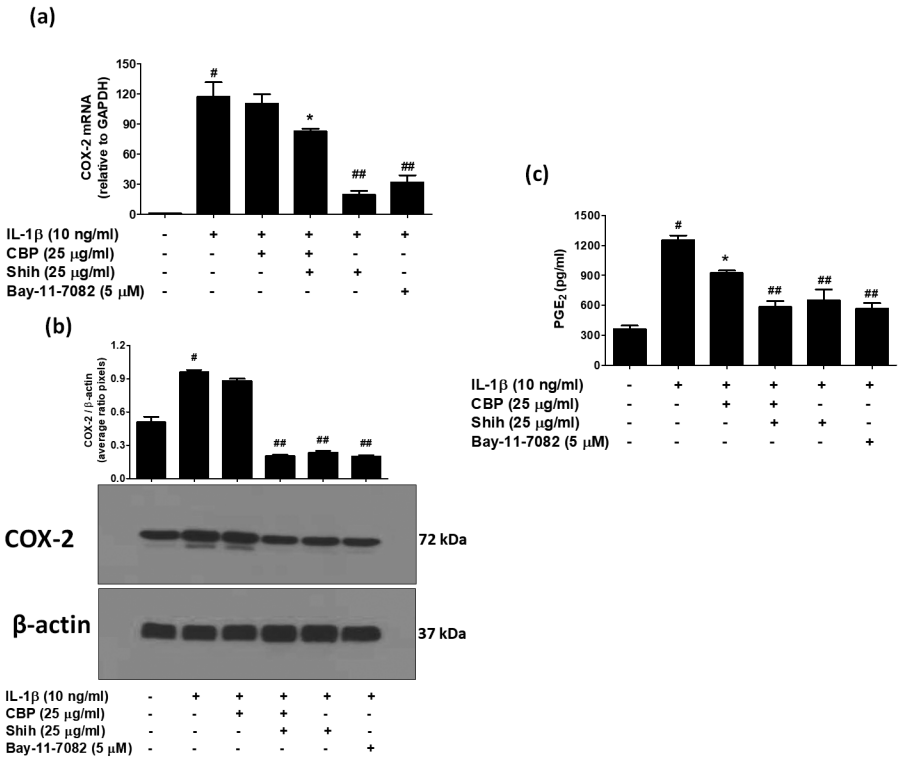


Figure 3: CMP-shih reduces COX-2 expression and PGE_2 production in IL-1β-activated human OA chondrocytes. (a) Quantitative RT-PCR analysis of COX-2 mRNA expression following pretreatment with CMP (25 μg/mL), shih (25 μg/mL), CMP-shih (50 μg/mL), or the NF-κB inhibitor Bay 11-7082 (5 μM) for 2 hr, followed by IL-1β (10 ng/mL) stimulation for 24 hr. COX-2 transcript levels were normalized to GAPDH and expressed relative to untreated controls. Data are presented as mean±SEM from duplicate experiments using chondrocytes isolated from OA donors. (b) Western blot analysis of COX-2 protein levels under identical treatment conditions. Protein expression was normalized to β-actin. Band intensities were quantified using Un-Scan-It software and expressed as mean pixel density derived from five independent scans. (c) Measurement of PGE_2 release into culture supernatants after pretreatment with CMP, shih, or CMP-shih for 2 hr and stimulation with IL-1β (10 ng/mL) for 24 hr. PGE_2 concentrations were determined using a specific ELISA kit. Results represent pooled data from three independent experiments.

CMP-Shih reduces IL-1β-induced PGE_2 production in primary human OA chondrocytes

Cyclooxygenase-2 (COX-2) catalyzes the formation of Prostaglandin E_2 (PGE_2), and increased COX-2 expression is directly associated with elevated PGE_2 synthesis during inflammatory responses. Given this functional relationship, we measured PGE_2 levels in culture supernatants of primary OA chondrocytes pretreated with CMP-Shih (50 μg/mL) for 2 hr and subsequently stimulated with IL-1β (10 ng/mL) for 24 hr. PGE_2 concentrations were determined using a specific ELISA assay. IL-1β stimulation significantly increased PGE_2 production compared with untreated controls ($p<0.01$). Importantly, pretreatment with CMP-Shih significantly suppressed IL-1β-induced PGE_2 secretion ($p<0.01$; Figure 3c).

CMP-Shih inhibits IL-1β-induced NF-κB activation and nuclear translocation in primary human OA chondrocytes

Nuclear Factor Kappa B (NF-κB) is a central regulator of inflammatory gene transcription and plays a pivotal role in mediating immune and inflammatory responses. To elucidate the mechanism underlying CMP-Shih-mediated inhibition of COX-2 expression, we examined NF-κB activation by assessing nuclear translocation and DNA-binding activity of the NF-κB p65 and p50 subunits using transcription factor-specific assays. IL-1β exposure significantly enhanced NF-κB p65 DNA-binding activity compared with control cells ($p<0.0001$). Treatment with CMP, shih, or their combination significantly reduced IL-1β-induced NF-κB p65 activation ($p<0.05$), with CMP-Shih demonstrating the most pronounced inhibitory effect (Figure 4a). Similar trends were observed for NF-κB p50 activity (Figure 4b). To further confirm the involvement of NF-κB signaling in COX-2 regulation, chondrocytes were treated with Bay-11-7082 (10 μM), a well-characterized NF-κB inhibitor. Bay-11-7082 significantly suppressed IL-1β-induced COX-2 expression at both mRNA and protein levels ($p<0.05$; Figures 3a and 3b) and reduced PGE_2 production ($p<0.05$; Figure 3c). Additionally, Bay-11-7082 markedly inhibited IL-1β-induced NF-κB p65 and p50 activation (Figures 4a and 4b). Collectively, these findings indicate that CMP-Shih attenuates IL-1β-induced COX-2 expression and PGE_2 production primarily through inhibition of NF-κB activation and its DNA-binding activity. Figure 5 presents a schematic overview of the proposed mechanism identified in this study, illustrating the pathway from IL-1β stimulation to the development of inflammation.

DISCUSSION

In the present study, we demonstrate for the first time that camel milk in combination with *Artemisia sieberi* (shih) extract (CMP-Shih) markedly attenuates IL-1β-induced inflammatory responses in primary human Osteoarthritic (OA) chondrocytes. The inhibitory effect was characterized by suppression of COX-2 expression, reduced PGE_2 production, and decreased activation of NF-κB subunits p65 (RelA) and p50. Because COX-2 transcription is largely regulated by NF-κB signaling, blockade of NF-κB activation is known to reduce COX-2 mRNA and protein expression as well as downstream PGE_2 synthesis (Mohamed *et al*., 2026; Yang *et al*., 2026). Our findings therefore indicate that CMP-Shih interferes with a central inflammatory signaling axis in OA chondrocytes. OA is a chronic degenerative disease of synovial joints characterized by progressive cartilage erosion, subchondral bone remodeling, osteophyte formation, mild synovitis, and capsular thickening (Oka *et al*., 2026). Clinically, patients commonly experience joint pain, stiffness, reduced mobility, and functional limitation, with disease severity varying substantially among individuals (Iwata *et al*., 2026). Increasing evidence supports the concept that OA is not merely a "wear and tear" disorder but involves sustained low-grade inflammation within the joint microenvironment. In particular, elevated levels of pro-inflammatory cytokines, including IL-1β, have been strongly implicated in OA pathogenesis (Yun *et al*., 2024). IL-1β plays a pivotal role in cartilage degradation by stimulating the production of matrix-degrading enzymes such as Matrix Metalloproteinases (MMPs) and aggrecanases, while simultaneously suppressing synthesis of extracellular matrix components including type II collagen and proteoglycans (Kim *et al*., 2022). In addition, IL-1β upregulates COX-2 expression, a key inducible enzyme responsible for PGE_2 biosynthesis and a well-recognized mediator of chronic inflammation. Although physiological levels of PGE_2 may exert anabolic or homeostatic effects in cartilage, excessive PGE_2 production in OA shifts the balance toward catabolism, promoting proteoglycan depletion, enhanced protease activity, and accelerated type II collagen breakdown. Elevated PGE_2 concentrations have been consistently detected in synovial fluid from OA patients and experimental OA models, highlighting its relevance as a therapeutic target (Yao *et al*., 2023). Current pharmacological management of OA primarily relies on Non-Steroidal Anti-Inflammatory Drugs (NSAIDs), corticosteroids, and certain plant-derived products (Maqbool *et al*., 2021). While these agents may alleviate pain and inflammation, none has convincingly demonstrated disease-modifying effects or the ability to halt cartilage degeneration. Moreover, long-term use is frequently associated with adverse effects, including gastrointestinal, renal, and cardiovascular complications (Maqbool *et al*., 2021). These limitations underscore the importance of identifying safer agents capable of targeting key inflammatory pathways involved in cartilage destruction.

Camel milk and *Artemisia sieberi* (shih) extract have each been reported to possess multiple biological activities, including antibacterial, antiviral, antiparasitic, antiallergic, antioxidant, and anticancer effects (Selim *et al*., 2026). Many of these properties are linked to their immunomodulatory and anti-inflammatory capacities. Camel milk contains bioactive components such as lactoferrin, immunoglobulins, and antioxidant enzymes, which contribute to modulation of inflammatory signaling and regulation of immune responses (Behrouz *et al*., 2022). Likewise, Artemisia species are rich in flavonoids, sesquiterpene lactones, and phenolic compounds that have been shown to inhibit pro-inflammatory mediators and oxidative stress in various experimental systems. However, despite these recognized anti-inflammatory properties, the potential chondroprotective effects of camel milk and shih-particularly in combination-have not previously been evaluated in human OA chondrocytes.

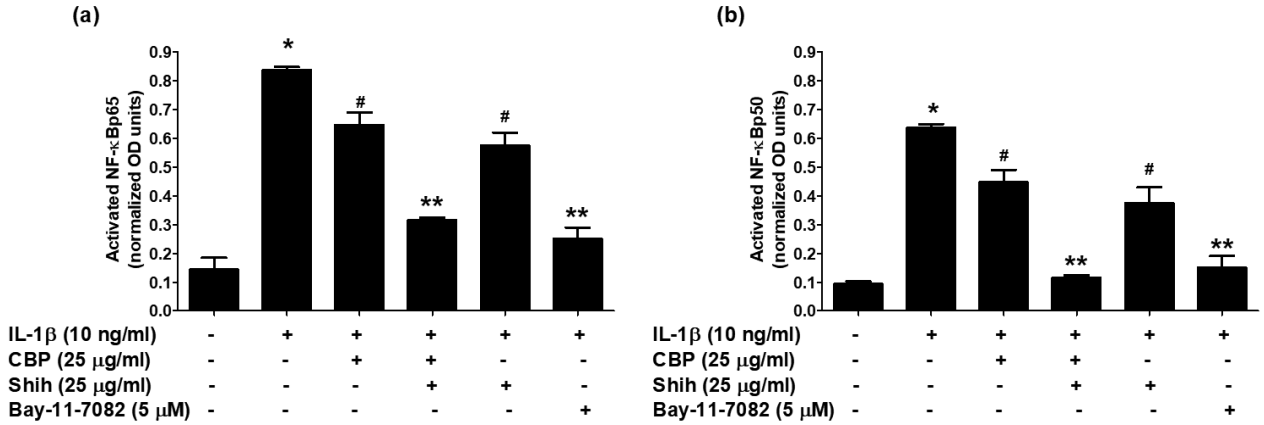


Figure 4: CMP-shih inhibits IL-1β-induced activation of NF-κB p65 and p50 in human OA chondrocytes. Primary human OA chondrocytes were pretreated with CMP, shih, CMP-shih, or Bay 11-7082 for 2 hr, followed by IL-1β (10 ng/mL) stimulation for 30 min. Nuclear translocation and activation of NF-κB subunits p65 (a) and p50 (b) were evaluated using a transcription factor-specific ELISA kit (Abcam). Bay 11-7082-treated nuclear extracts served as positive controls. Data are expressed as mean±SEM from three independent experiments.

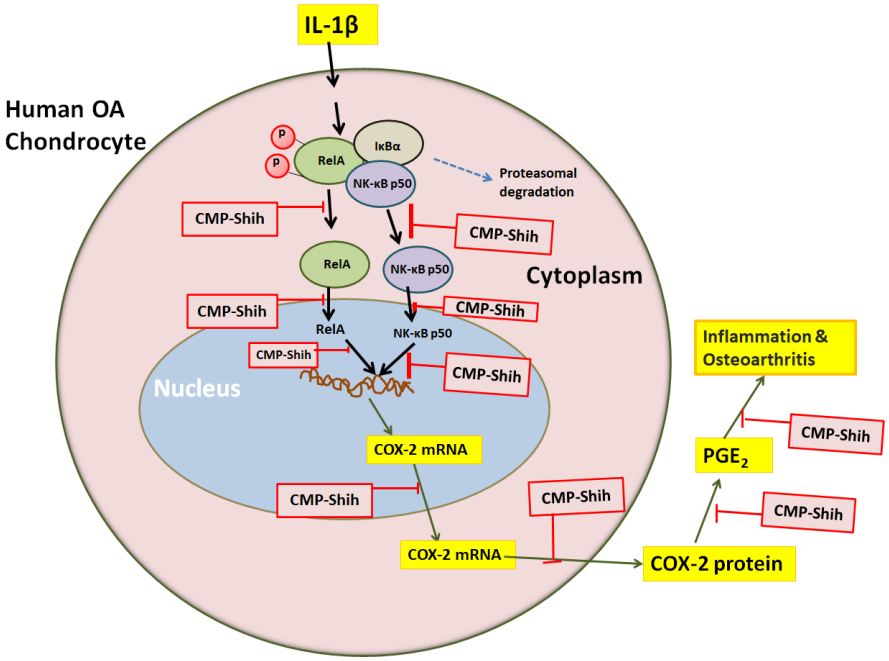


Figure 5: Schematic illustration of the proposed anti-inflammatory mechanism of CMP-shih in human OA chondrocytes. Diagram depicting the inhibitory action of CMP-shih on IL-1β-induced inflammatory signaling pathways in human OA chondrocytes. Binding of IL-1β to its receptor (IL-1R) triggers intracellular signaling cascades that activate NF-κB, promoting nuclear translocation of p65 and p50 subunits and subsequent upregulation of COX-2 expression and PGE_2 synthesis. The proposed sites of CMP-shih intervention within this signaling pathway are indicated.

Our results demonstrate that CMP-Shih significantly suppresses IL-1β-induced COX-2 gene and protein expression and reduces PGE_2 production. Although camel milk and shih individually exerted inhibitory effects, the combined formulation produced a markedly greater response, suggesting additive or synergistic interactions between their bioactive constituents. Such combinatorial approaches may enhance therapeutic efficacy by targeting multiple molecular pathways simultaneously, an approach increasingly recognized as beneficial in complex multifactorial diseases such as OA.

NF-κB is a master transcription factor regulating the expression of numerous genes involved in inflammation, including cytokines, chemokines, adhesion molecules, and enzymes such as COX-2 (Zou *et al.*, 2025). Under resting conditions, NF-κB remains sequestered in the cytoplasm through association with inhibitory IκB proteins. Upon stimulation, such as by IL-1β-IκB undergoes phosphorylation and proteasomal degradation, allowing NF-κB dimers (notably p65/p50) to translocate to the nucleus and initiate transcription of pro-inflammatory genes [13]. NF-κB activation is a well-established mediator of IL-1β signaling, and COX-2 expression is critically dependent on NF-κB transcriptional activity (Hoffmann *et al.*, 2025). In the present study, CMP-Shih inhibited IL-1β-induced nuclear translocation and DNA-binding activity of NF-κB p65 and p50 subunits in OA chondrocytes. These findings confirm that suppression of NF-κB signaling is a central mechanism underlying the observed reduction in COX-2 expression and PGE_2 synthesis. Furthermore, pharmacological inhibition of NF-κB using Bay 11-7082 similarly reduced IL-1β-stimulated COX-2 expression and PGE_2 production, reinforcing the pivotal role of NF-κB in mediating these responses. Collectively, these results support the conclusion that CMP-Shih exerts its anti-inflammatory effects, at least in part, through inhibition of NF-κB activation. Importantly, the inhibitory activity of CMP-Shih was observed at a physiologically achievable concentration (50 μg/mL), suggesting potential translational relevance. By attenuating IL-1β-induced production of the cartilage-degrading mediator PGE_2 via suppression of COX-2 expression, CMP-Shih may contribute to preservation of cartilage homeostasis. This study was limited to an *in vitro* chondrocyte model, and the effects of CMP-Shih need validation in animal models and clinical studies. Second, additional molecular pathways should be investigated to further understand its chondroprotective mechanisms. In short, this study provides novel mechanistic evidence that combined camel milk and shih extract effectively modulate key inflammatory pathways in human OA chondrocytes. Given the limitations of current OA therapies, these findings highlight the potential of CMP-Shih or its bioactive constituents as a basis for developing safer, multi-targeted therapeutic strategies aimed at preventing or slowing OA progression. Further *in vivo* studies and clinical investigations are warranted to validate these chondroprotective effects and to define optimal dosing and safety profiles.

CONCLUSION

The present study provides novel mechanistic insight into the chondroprotective and anti-inflammatory properties of camel milk, *Artemisia sieberi* extract (shih), and particularly their combined formulation. For the first time, we demonstrate that co-administration of camel milk and *Artemisia sieberi* extract significantly attenuates IL-1β-induced inflammatory activation in primary human osteoarthritic chondrocytes. The combined treatment effectively suppressed NF-κB signaling, as evidenced by reduced activation of key NF-κB subunits. In parallel, it markedly decreased IL-1β-induced COX-2 mRNA and protein expression, leading to a substantial reduction in the production of the cartilage-degrading mediator PGE_2. These findings indicate that inhibition of the NF-κB/COX-2/PGE_2 axis represents a central mechanism underlying the observed anti-inflammatory and chondroprotective effects. Overall, our results expand the current understanding of how camel milk and *Artemisia sieberi* extract modulate inflammatory pathways relevant to osteoarthritis. The enhanced efficacy observed with the combined formulation suggests potential synergistic interactions between their bioactive components. These data support further preclinical and clinical investigations to evaluate the therapeutic potential of this combination as a safe, multi-targeted strategy for the prevention or management of osteoarthritis.

ACKNOWLEDGEMENT

The authors gratefully acknowledge Qassim University, represented by the Deanship of Graduate Studies and Scientific Research, for the financial support provided for this research under grant number (QU-J-PG-2-2025-54101) during the academic year 1446 AH / 2024 AD.

ABBREVIATIONS

ACAN: Aggrecan; **AI:** Artificial Intelligence; **ANOVA:** Analysis of Variance; **CM:** Camel Milk; **CMP:** Camel Milk Powder; **CMP-Shih:** Camel Milk Powder-*Artemisia sieberi* extract combination; **CO_2:** Carbon Dioxide; **COX-2:** Cyclooxygenase-2; **COL2A1:** Collagen type II alpha 1 chain; **COL10A1:** Collagen type X alpha 1 chain; **DMEM:** Dulbecco's Modified Eagle Medium; **DMEM/F12:** Dulbecco's Modified Eagle Medium/ Nutrient Mixture F-12; **DNA:** Deoxyribonucleic Acid; **ECL:** Enhanced Chemiluminescence; **ECM:** Extracellular Matrix; **ELISA:** Enzyme-Linked Immunosorbent Assay; **GAPDH:** Glyceraldehyde-3-Phosphate Dehydrogenase; **HRP:** Horseradish Peroxidase; **IL:** Interleukin; **IL-1R:** Interleukin-1 receptor; **IL-1β:** Interleukin-1 beta; **IRB:** Institutional Review Board; **mRNA:** Messenger Ribonucleic Acid; **NF-κB:** Nuclear Factor kappa B; **OA:** Osteoarthritis; **PCR:** Polymerase Chain Reaction; **PGE_2:** Prostaglandin E_2; **PVDF:** Polyvinylidene Difluoride; **qRT-PCR:** Quantitative Real-Time Polymerase Chain Reaction; **RelA:** v-rel avian reticuloendotheliosis viral oncogene homolog A; **RIPA:** Radioimmunoprecipitation Assay; **RNA:** Ribonucleic Acid; **rpm:** Revolutions per minute; **RT-PCR:** Reverse Transcription Polymerase Chain Reaction; **SD:** Standard Deviation; **SDS-PAGE:** Sodium Dodecyl Sulfate-Polyacrylamide Gel Electrophoresis; **SEM:** Standard Error of the Mean; **SOX-9:** SRY-box transcription factor 9; **TBS-T:** Tris-Buffered Saline with Tween-20; **TaqMan:** Fluorogenic probe-based real-time PCR assay; **µM:** Micromolar.

CONFLICT OF INTEREST

The authors declare that there is no conflict of interest.

REFERENCES

Abdellatif, A. A., Rasheed, Z., Alhowail, A. H., Alqasoumi, A., Alsharidah, M., Khan, R. A., Aljohani, A. S., Aldubayan, M. A., & Faisal, W. (2020). Silver citrate nanoparticles inhibit PMA-Induced TNFA expression via deactivation of NF-KB activity in human cancer Cell-Lines, MCF-7. International Journal of Nanomedicine, Volume 15, 8479–8493. https://doi.org/10.2147/ijn.s274098

Behrouz, S., Saadat, S., Memarzia, A., Sarir, H., Folkerts, G., & Boskabady, M. H. (2022). The antioxidant, Anti-Inflammatory and immunomodulatory effects of camel milk. Frontiers in Immunology, 13, 855342. https://doi.org/10.3389/fimmu.2022.855342

Bonder, Finlay-Jones, & Hart. (1999). Interleukin-4 regulation of human monocyte and macrophage interleukin-10 and interleukin-12 production. Role of a functional interleukin-2 receptor γ-chain. *Immunology*, *96*(4), 529-536. https://doi.org/10.1046/j.1365-2567.1999.00711.x

El-Kattawy, A. M., Algezawy, O., Alfaifi, M. Y., Noseer, E. A., Hawsawi, Y. M., Alzahrani, O. R., Algarni, A., Kahilo, K. A., & El-Magd, M. A. (2021). Therapeutic potential of camel milk exosomes against HepaRG cells with potent apoptotic, anti-inflammatory, and anti-angiogenesis effects for colostrum exosomes. Biomedicine & pharmacotherapy = Biomedecine & pharmacotherapie, 143, 112220. https://doi.org/10.1016/j.biopha.2021.112220

El-Saghier, A. M., Atia, F. A. M., Enaili, S. S., Mohamed, M. A. A., Shapep, A. M. A., Salah, H., Roushdy, N., & Hamed, A. M. (2026). Design, synthesis, and biological evaluation of novel Spirosulfamethoxazole-1,3,4-thiadiazole-2-carboxamide derivatives as dual COX-2/5-LOX inhibitors with promising anti-inflammatory and anti-osteoarthritic potential. Bioorganic chemistry, 179, 109986. https://doi.org/10.1016/j.bioorg.2026.109986

Hashemi-Afzal, F., Fallahi, H., Bagheri, F., Collins, M. N., Eslaminejad, M. B., & Seitz, H. (2024). Advancements in hydrogel design for articular cartilage regeneration: A comprehensive review. Bioactive materials, 43, 1–31. https://doi.org/10.1016/j.bioactmat.2024.09.005

Feng, T., & Wu, Q. F. (2022). A review of non-coding RNA related to NF-κB signaling pathway in the pathogenesis of osteoarthritis. International immunopharmacology, 106, 108607. https://doi.org/10.1016/j.intimp.2022.108607

Hamdan, A. M. E., Alahdal, H., Al-Zubaidy, H. F. S., Abu-Elfotuh, K., Reda, E., Mahran, Y. F., Alghamdi, A., Aboelsoud, H. A., Ismael, S. A., Mohamed, E. K., Amer, A. A., Nazeam, J. A., El-Haddad, A. E., Abdelhakim, K. R., Kozman, M. R., Mohamed, N. M., & Gowifel, A. M. H. (2026). Unveiling the neuroprotective effects of pomegranate extract and/or physical activity against aluminum chloride-induced Alzheimer's disease in rats: modulation of multitude pathways. Frontiers in pharmacology, 17, 1793211. https://doi.org/10.3389/fphar.2026.1793211

Hoffmann, A., Cheng, G., & Baltimore, D. (2025). NF-κB: master regulator of cellular responses in health and disease. *Immunity & Inflammation*, *1*(1), 2. https://doi.org/10.1007/s44466-025-00014-0

Huang, L., Kim, J. H., You, L., Park, S. H., Zhang, J., Shin, C. Y., Sutopo, N. C., Byun, H. W., Omaliss, K., Masphal, K., Son, J., Kim, G. R., Lee, B. H., Kim, J. H., Lee, J., & Cho, J. Y. (2024). Anti-oxidative, anti-apoptotic, and anti-inflammatory activities of Connarus semidecandrus Jack ethanol extract in UVB-irradiated human keratinocytes. Journal of ethnopharmacology, 321, 117574. https://doi.org/10.1016/j.jep.2023.117574

Iwata, H., Sato, S., Somemura, S., Takemoto, M., Takahashi-Suzuki, Y., *et al.* (2026). Effects of Mechanical Stress on Bone and Cartilage Metabolism: How Mechanical Stress Affects Energy Metabolism in Bone and Cartilage Tissues (Our Research Overview): Mini Review. *International journal of molecular sciences*, *27*(3), 1380. https://doi.org/10.3390/ijms27031380

Jamal, K., Al-Taweel, A., Bukhari, S. I., Orfali, R., Moubayed, N. M. S., *et al.* (2023). Isochlorogenic Acid Glucosides from the Arabian Medicinal Plant *Artemisia sieberi* and Their Antimicrobial Activities. *Molecules*, *28*(22), 7460. https://doi.org/10.3390/molecules28227460

Khan, Y. S., Farhana, A., Kuddus, M., Shahid, S. M. A., Alsrhani, A., *et al.* (2025). MicroRNA-125b-5p Drives MMP-2 Expression via Activation of RAGE-38MAPK-p65/p50NF-κB Axis: A Novel Mechanism in Human Lung Cancer Cells. *International Journal of Molecular Sciences*, *26*(20), 9983. https://doi.org/10.3390/ijms26209983

Kim, S. E., Choi, S. J., Park, K., Kim, H.-J., Song, G. G., & Jung, J. H. (2022). Intra-Articular Injection of Rebamipide-Loaded Nanoparticles Attenuates Disease Progression

and Joint Destruction in Osteoarthritis Rat Model: A Pilot Study. *Cartilage*, *13*(1),19476035211069250. https://doi.org/10.1177/19476035211069250

Lee, H., Nam, J., Jang, H., Park, Y. S., Son, M. H., Lee, I. H., Eyun, S. I., Jeon, J., & Yang, S. (2023). Novel molecule BBC0901 inhibits BRD4 and acts as a catabolic regulator in the pathogenesis of osteoarthritis. Biomedicine & pharmacotherapy = Biomedecine & pharmacotherapie, 166, 115426. https://doi.org/10.1016/j.biopha.2023.115426

Li, R., Cao, R., Kang, J., Li, Y., Lin, X., & Zhang, Z. (2026). Advances in chemoprevention of familial adenomatous polyposis. Frontiers in oncology, 16, 1815387. https://doi.org/10.3389/fonc.2026.1815387

Lu, J., Yu, M., & Li, J. (2023). PKC-δ Promotes IL-1β-Induced Apoptosis of Rat Chondrocytes via Activating JNK and P38 MAPK Pathways. *Cartilage*, *15*(3), 315-327. https://doi.org/10.1177/19476035231181446

Maqbool, M., Fekadu, G., Jiang, X., Bekele, F., Tolossa, T., Turi, E., Fetensa, G., & Fanta, K. (2021). An up-to-date on clinical prospects and management of osteoarthritis. *Annals of Medicine and Surgery*, *72*(72), 103077. https://doi.org/10.1016/j.amsu.2021.103077

Moghbeli, M., Taghehchian, N., Akhlaghipour, I., Samsami, Y., & Maharati, A. (2023). Role of forkhead box proteins in regulation of doxorubicin and paclitaxel responses in tumor cells: A comprehensive review. International journal of biological macromolecules, 248, 125995. https://doi.org/10.1016/j.ijbiomac.2023.125995

Mohamed, W. A., Hassanen, E. I., Khalefa, H. S., Ibrahim, M. A., Hassan, N. H., & Mahmoud, M. A. (2026). Mechanistic approach on the protective role of rutin against ammonia-induced hepatic encephalopathy in Nile tilapia (*Oreochromis niloticus*) focusing on Cox-2/NF-κB pathway. *Fish Physiology and Biochemistry*, *52*(3), 90. https://doi.org/10.1007/s10695-026-01711-2

Mohammed, H. A., Qureshi, K. A., Ali, H. M., Al-Omar, M. S., Khan, O., & Mohammed, S. A. A. (2022). Bio-Evaluation of the Wound Healing Activity of *Artemisia judaica* L. as Part of the Plant's Use in Traditional Medicine; Phytochemical, Antioxidant, Anti-Inflammatory, and Antibiofilm Properties of the Plant's Essential Oils. *Antioxidants*, *11*(2), 332. https://doi.org/10.3390/antiox11020332

O'Connor, D., Johnston, R. V., Brignardello-Petersen, R., Poolman, R. W., Cyril, S., Vandvik, P. O., & Buchbinder, R. (2022). Arthroscopic surgery for degenerative knee disease (osteoarthritis including degenerative meniscal tears). The Cochrane database of systematic reviews, 3(3), CD014328. https://doi.org/10.1002/14651858.CD014328

Oka, T., Kitagawa, T., Nasu, T., Kurasawa, Y., Kunihara, H., Suzuki, K., & Nakamura, N. (2026). Clinical outcomes of biologic therapies for knee osteoarthritis: A systematic review and meta-analysis of studies with ≥12-month follow-up. Modern rheumatology, 36(5), 696–704. https://doi.org/10.1093/mr/roag010

Rasheed, N., Alghasham, A., & Rasheed, Z. (2016). Lactoferrin from *Camelus dromedarius* Inhibits Nuclear Transcription Factor-kappa B Activation, Cyclooxygenase-2 Expression and Prostaglandin E2 Production in Stimulated Human Chondrocytes. Pharmacognosy Research, *8*(2), 135-141. https://doi.org/10.4103/0974-8490.175612

Rasheed, Z., Rasheed, N., Abdulmonem, W. Al, & Khan, M. I. (2019). Author Correction: MicroRNA-125b-5p regulates IL-1β induced inflammatory genes via targeting TRAF6-mediated MAPKs and NF-κB signaling in human osteoarthritic chondrocytes. *Scientific Reports*, *9*(1), 6882. https://doi.org/10.1038/s41598-019-50844-3

Rasheed, Z., Rasheed, N., Abdulmonem, W. Al, & Khan, M. I. (2019). MicroRNA-125b-5p regulates IL-1β induced inflammatory genes via targeting TRAF6-mediated MAPKs and NF-κB signaling in human osteoarthritic chondrocytes. *Scientific Reports*, *9*(1), 6882. https://doi.org/10.1038/s41598-019-42601-3

Rellmann, Y., Eidhof, E., & Dreier, R. (2021). Review: ER stress-induced cell death in osteoarthritic cartilage. Cellular signalling, 78, 109880. https://doi.org/10.1016/j.cellsig.2020.109880

Selim, S., Harun-Ur-Rashid, M., & Al Jaouni, S. K. (2026). Unveiling the multifaceted potential of Artemisia: Cutting-edge insights into nutritional benefits and emerging therapeutic applications. *Journal of Pharmaceutical Analysis*, *16*(5), 101531. https://doi.org/10.1016/j.jpha.2025.101531

Sundar, R., Sundar, G., John, A., & Abraham, A. (2024). *Butea monosperma* bark extract: a natural boost for osteogenesis via Wnt/β-catenin pathway activation in adipose-derived mesenchymal stem cells. *Biotechnology Letters*, *47*(1), 3. https://doi.org/10.1007/s10529-024-03545-5

Swelum, A. A., El-Saadony, M. T., Abdo, M., Ombarak, R. A., Hussein, E. O. S., Suliman, G., *et al.* (2021). Nutritional, antimicrobial and medicinal properties of Camel's milk: A review. *Saudi Journal of Biological Science*s, 28(5), 3126-3136. https://doi.org/10.1016/j.sjbs.2021.02.057

Wei, J., Zhang, Y., Xie, C., Lei, G., & Zeng, C. (2026). The gut-joint axis in osteoarthritis. *Nature Reviews Rheumatology*, *22*(6), 345-361. https://doi.org/10.1038/s41584-026-01378-2

Wei, W. L., Wang, W. J., Chen, H., Lin, S. Y., Luo, Q. S., Li, J. M., Yan, J., & Chen, L. L. (2024). A promising Artemisia capillaris Thunb. Leaf proteins with high nutrition, applicable function and excellent antioxidant activity. Food chemistry: X, 21, 101153. https://doi.org/10.1016/j.fochx.2024.101153

Wen, L., Grad, S., Creemers, L. B., & Stoddart, M. J. (2025). Establishment of an *ex vivo* cartilage damage model by combined collagenase treatment and mechanical loading. *Arthritis Research & Therapy*, *27*(1), 30. https://doi.org/10.1186/s13075-025-03499-7

Wu, Y., Ying, J., Zhu, X., Xu, C., & Wu, L. (2023). Pachymic acid suppresses the inflammatory response of chondrocytes and alleviates the progression of osteoarthritis via regulating the Sirtuin 6/NF-κB signal axis. *International Immunopharmacology*, *124*, 110854. https://doi.org/10.1016/j.intimp.2023.110854

Xu, C., Cui, X., Shi, Y., Zhang, T., Ni, Z., Li, K., Chen, X., & Wang, F. (2025). Natural products in the treatment of osteoarthritis: Current status and prospects. *Journal of orthopaedic translation*, *55*, 94-120. https://doi.org/10.1016/j.jot.2025.07.007

Yan, M., Zhang, X., Liu, H., Kang, Q., Li, J., Zhang, B., Lin, D., & Li, X. (2026). From mechanical triggering to metabolic-inflammatory driving: a new paradigm of knee osteoarthritis pathogenesis. *Frontiers in Immunology*, *17*. https://doi.org/10.3389/fimmu.2026.1833313

Yang, M., Feng, K., Li, Y., Zeng, S., Ma, H., & Feng, H. (2026). Research Progress on Anti-Inflammatory and Antioxidant Mechanism of Artemether Based on MAPK/NF-κB Signaling Pathway. *International Journal of Molecular Sciences*, *27*(10), 4607. https://doi.org/10.3390/ijms27104607

Yao, Q., Wu, X., Tao, C., Gong, W., Chen, M., Qu, M., Zhong, Y., He, T., Chen, S., & Xiao, G. (2023). Osteoarthritis: pathogenic signaling pathways and therapeutic targets. *Signal Transduction and Targeted Therapy*, *8*(1), 56. https://doi.org/10.1038/s41392-023-01330-w

Yun, H., Liu, Q., Zeng, Z., & Lian, L. (2024). Experimental study on water-exit of cylinder. *Ocean Engineering*, *293*, 116585. https://doi.org/10.1016/j.oceaneng.2023.116585

Zhang, M., Hu, W., Cai, C., Wu, Y., Li, J., & Dong, S. (2022). Advanced application of stimuli-responsive drug delivery system for inflammatory arthritis treatment. *Materials Today Bio*, *14*, 100223. https://doi.org/10.1016/j.mtbio.2022.100223

Zou, Z., Pan, S., Sun, C., Wei, J., Xu, Y., Xiao, K., Zhao, J., & Gu, R. (2025). AM1241 inhibits chondrocyte inflammation and ECM degradation through the Nrf2/HO-1 and NF-κB pathways and alleviates osteoarthritis in mice. *Molecular Medicine*, *31*(1), 9. https://doi.org/10.1186/s10020-024-01012-5

Cite this article: Almujaydil MS, Ahmed WE, Alshebrami RM, Abdulmonem WA. Camel Milk Powder with *Artemisia sieberi* Extract Inhibit Nuclear Transcription Factor-κB RelA and p50 Activation, Cyclooxygenase-2 Expression and Prostaglandin E2 Production in Interleukin-1β-Stimulated Human Osteoarthritic Chondrocytes. Pharmacog Res. 2027;19(2):709-18.