Acute Intervention With Selective Interleukin-1 Inhibitor Therapy May Reduce the Progression of Posttraumatic Osteoarthritis of the Knee: A Systematic Review of Current Evidence

Zachary S. Aman, B.A., Nicholas N. DePhillipo, Ph.D., A.T.C., Filippo Familiari, M.D., LTC Jonathan F. Dickens, M.D., Robert F. LaPrade, M.D., Ph.D., and Maj. Travis J. Dekker, M.D.

Purpose: To evaluate the efficacy of selective interleukin (IL)-1 inhibitor therapy in the reduction of posttraumatic osteoarthritis (PTOA) progression following knee ligament or meniscal injury. **Methods:** A systematic review was conducted evaluating the disease-modifying efficacy of selective IL-1 inhibition in the setting of knee PTOA. **Results:** The literature search identified 364 articles and 11 studies were included (n = 10 preclinical, n = 1 clinical). Drug delivery in preclinical studies was administered using IL-1Ra–encoded helper-dependent adenovirus particles (n = 3), synovial cells transfected with an IL-1Ra–encoded retroviral vector (n = 3), or varying chemical compositions of nonviral microcapsule gene carriers (n = 4). Intervention with selective IL-1 inhibitor therapy within 2 weeks of injury provided the greatest protective benefits in reducing the progression of PTOA regardless of drug delivery methodology in preclinical models. The majority of studies reported significantly better cartilage integrity and reduction in lesion size in animals treated with gene therapy with the greatest effects seen in those treated within 5 to 7 days of injury. **Conclusions:** Early intervention with selective IL-1 inhibitor therapy were effective in reducing proinflammatory IL-1β levels in the acute and subacute phases following traumatic knee injury in preclinical animal model studies, while significantly reducing cartilage damage, lesion size, and PTOA progression at short-term follow-up. However, it was found that the effect of these therapies diminished over time. **Clinical Relevance:** Acute, intra-articular injection of selective IL-1 inhibitors may reduce PTOA progression, supporting the need for additional basic and clinical investigation.

From the Sidney Kimmel Medical College at Thomas Jefferson University, Philadelphia, Pennsylvania, U.S.A. (Z.S.A.); Oslo Sports Trauma Research Center, Oslo, Norway (N.N.D.); Department of Orthopaedics and Trauma Surgery, Magna Graecia University, Catanzaro, Italy (F.F.); Walter Reed National Military Medical Center, Bethesda, Maryland, U.S.A. (J.F.D.); Twin Cities Orthopedics, Edina, Minnesota, U.S.A. (R.F.L.); and Eglin Air Force Base, Eglin, Florida, U.S.A. (T.J.D.).

The authors report the following potential conflicts of interest or sources of funding: J.F.D. reports other from Supreme Orthopedic Systems, Arthrex, and Smith & Nephew, outside the submitted work. R.F.L. receives royalties from Arthrex and Smith & Nephew; is a paid consultant for Arthrex, Ossur, and Smith & Nephew; receives research support from Arthrex, Linvatec, Ossur, and Smith & Nephew; is on the editorial/governing board for AJSM and KSSTA; and has committee/member appointments for AOSSM, ISAKOS, AANA, and ESSKA. Full ICMJE author disclosure forms are available for this article online, as supplementary material.

Received July 23, 2021; accepted February 4, 2022.

Address correspondence to Maj. Travis Dekker, M.D., 307 Boatner Rd., Eglin AFB, USAF, Eglin, FL 32542. E-mail: travisdekker88@gmail.com

0749-8063/211056/$36.00

https://doi.org/10.1016/j.arthro.2022.02.009

Posttraumatic osteoarthritis (PTOA) of the knee accounts for up to 12% of all symptomatic cases of osteoarthritis (OA) in the United States[1] and is estimated to occur in up to 50% of patients 5 to 10 years after anterior cruciate ligament (ACL) tear or reconstruction.[2,3] As patients with ACL tears are, on average, 17 years of age,[2,4,5] early-onset OA following traumatic injuries has significant implications on future disability,[6,7] health care costs, and overall patient health.[8,9] This represents a significant challenge for clinicians because there remains a paucity of recommended treatment options for the prevention of OA progression[10] following ligament and meniscal injury, highlighted by the significantly limited number of clinical trials investigating disease-modifying therapies for OA.[10]

Although numerous studies have sought to investigate pharmacologic treatments for established OA, trials evaluating disease-modifying therapies directed toward preventing PTOA also offer a unique setting to

proactively target acute inflammatory mediators involved in disease progression.[11-14] Specifically, interleukin-1 (IL-1) has been reported to downregulate the synthesis of cartilage extracellular matrix and upregulate metalloproteinase expression involved in cartilage degeneration,[15] suggesting a potential role of IL-1 receptor antagonist (IL-1Ra) therapy in modifying OA-related inflammation. Previous clinical and preclinical investigations have demonstrated that IL-1Ra effectively and safely decreases proinflammatory cytokine concentrations while protecting cartilage integrity in the setting of established OA,[16-19] indicating a potential therapeutic option for delaying PTOA in young patients following ligament or meniscal injury.

Although the long-term efficacy of IL-1Ra in the clinical treatment of established, nontraumatic OA remains controversial,[14] the therapeutic efficacy of IL-1Ra or selective agents inhibiting the production or activity of IL-1 in the setting of PTOA has yet to be systematically reviewed. Thus, the purpose of this systematic review was to evaluate the efficacy of selective IL-1 inhibitor therapy in the reduction of PTOA progression following knee ligament or meniscal injury. It was hypothesized that selective IL-1 inhibitor therapy would effectively reduce IL-1 synovial fluid levels and reduce the progression of gross and microscopic cartilage degeneration in the setting of PTOA.

Methods

Article Identification and Selection

A systematic review of the literature evaluating the disease-modifying efficacy of selective IL-1 inhibitor agents in the setting of PTOA of the knee was performed according to the PRISMA (Preferred Reporting Items for Systematic Review and Meta-Analysis) guidelines.[20] The search query was performed in May 2021 using the Cochrane Database of Systematic Reviews, the Cochrane Central Register of Controlled Trials, PubMed (1980-2021), MEDLINE (1980-2021), and Embase (1980-2021). The search terms used were ["osteoarthritis" AND "interleukin-1 receptor antagonist"] and ["post-traumatic osteoarthritis" AND "interleukin"].

The inclusion criteria were English language and human and animal studies investigating the use of selective IL-1 inhibitor agents specific to the treatment of PTOA of the knee following ligament or meniscal injury. Selective IL-1 inhibitor therapies were considered as those directly inhibiting the production or activity of IL-1, including recombinant IL-1Ra proteins (anakinra), IL-1Ra gene therapy, selective down-regulation of IL-1 gene expression, or monoclonal antibodies against IL-1 (canakinumab).[21] Exclusion criteria were patients or animals with symptomatic or previously diagnosed OA, fractures, in vitro and ex vivo studies, case studies, and review articles.

Two investigators (Z.S.A and N.N.D) independently reviewed the titles and abstracts of all identified articles and were assessed for inclusion and exclusion criteria after removal of duplicates. Full-text articles were then obtained and reviewed for final inclusion and were reconciled between the 2 investigators. Discrepancies between final included articles were reviewed by the senior author (T.J.D) for final decision of article inclusion. References from included articles were also reviewed to ensure no studies meeting inclusion criteria were missed.

Data Collection

Data from both human and animal studies were extracted by a single investigator (Z.S.A.) and reviewed by a second author (N.N.D.). The main variables extracted and recorded were etiology of PTOA, sample size, subject characteristics, dosage, route and frequency of administration, and duration of treatment. Drug type, mechanism, and vehicle (if applicable) also were recorded. Primary outcomes assessed were synovial fluid analysis (enzyme-linked immunosorbent assay), gene expression (reverse transcription polymerase chain reaction, microarray), immunohistochemistry, and cartilage integrity (gross, histology, micro-computed tomography [CT], Osteoarthritis Research Society International [OARSI]). Efficacy of selective Il-1 inhibitors was determined as outcomes showing improvement in cartilage integrity on histologic, gross, or imaging evaluation, reduction in IL-1 synovial fluid concentrations, and significant presence of IL-1ra as an indicator of sustained effect at primary study endpoints. Knee Injury and Osteoarthritis Outcome Scores (KOOS) were assessed for evaluating drug efficacy for the included human trial.

Results

The literature search identified 364 articles with 11 studies meeting inclusion criteria (Fig 1). There were 10 animal studies[22−31] and 1 human study.[12] Five preclinical studies were performed on rabbits,[24,26-29] 3 studies on mice,[22,23,31] 1 study on rats,[31] and 1 study on dogs.[25] The clinical efficacy of IL-1Ra treatment was evaluated in 1 Level I randomized-controlled trial comprising 11 ACL-deficient patients.[12]

Preclinical Studies

PTOA Animal Models

Of the preclinical studies, 3 studies evaluated PTOA models with ACL transection (mouse, n = 1; rat, n = 1, dog, n = 1),[22,25,31] 4 studies with medial collateral ligament (MCL) transection and medial meniscectomy (rabbit, n = 4),[24,27−29] 1 study with transections of the ACL and anterior attachment of the medial meniscotibial ligament (mouse, n = 1),[23] 1 study with ACL and MCL transections with medial meniscectomy (rabbit, n = 1),[26] and 1 with transection of the anterior

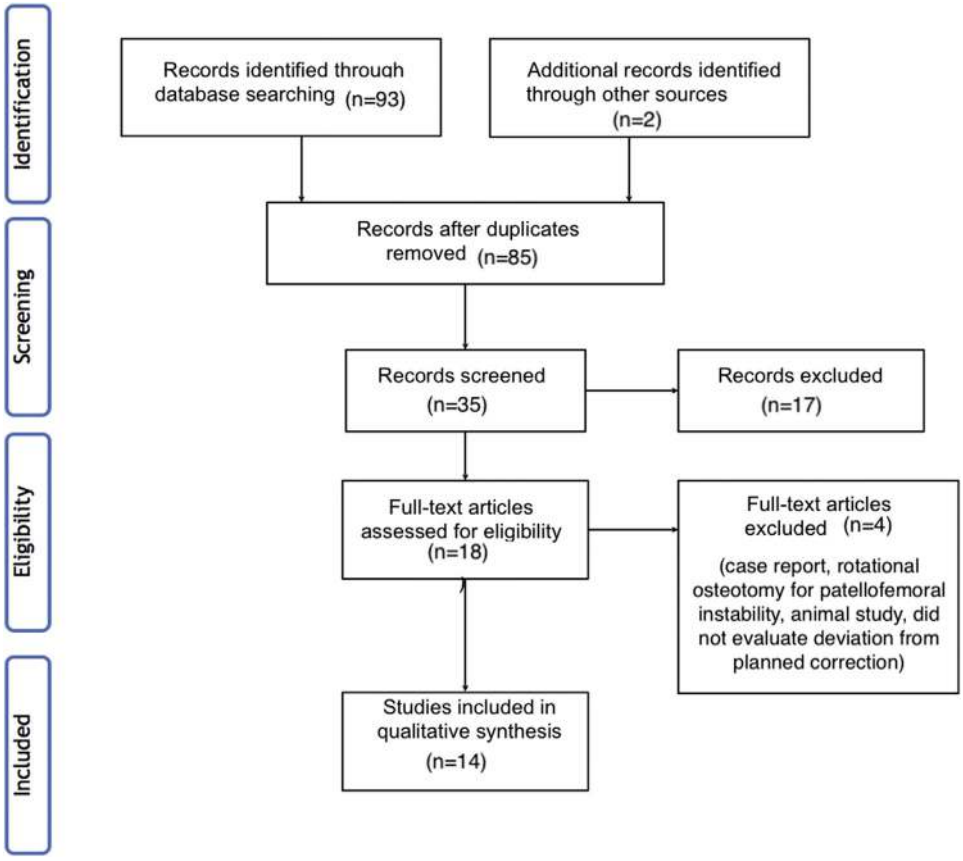

Fig 1. PRISMA (Preferred Reporting Items for Systematic Reviews and Meta-Analyses) flowchart showing the selection criteria used to identify studies with the search strategy.

attachment of the medial meniscotibial ligament only (mouse, n = 1).[30] Preclinical study designs and outcomes are detailed in Table 1.

Selective IL-1 Inhibitor Delivery Systems

Preclinical treatment protocols including dosage, frequency, route of administration, and drug characteristics are summarized in Table 2. Nine preclinical studies administered intra-articular injections[22-29,31] and 1 preclinical study administered oral treatment.[30] Initiation of treatments ranged from 2 days before injury to 1 month after injury. Three studies administered 1 injection of IL-1Ra encoded helper-dependent adenovirus (HDAd) particles,[22–24] 3 studies administered 1 injection of synovial cells transfected with an IL-1Ra–encoded retroviral vector,[25–27] 1 study administered 2 injections of liposome-mediated recombinant IL-1Ra plasmid DNA,[29] 1 study administered 3 injections of chitosan-DNA nanoparticles containing IL-1Ra DNA,[28] and 1 study administered 6 injections of a liposome-mediated miR-144-3p mimic.[31] Only one study administered oral treatment using yeast microcapsule-mediated delivery of IL-β shRNA.[30]

IL-1β Activity in Synovial Fluid

IL-1β activity in synovial fluid was analyzed in 4 preclinical studies.[23,26,29,30] In 3 studies, IL-1β expression was significantly decreased compared with treatment control groups after yeast microcapsule,[30] liposome-mediated,[29] and retrovirus transfected synovial cell delivery of IL-1Ra[26] when evaluated at a range of 7 to 40 days ($P < .05$). IL-1β levels were evaluated in meniscotibial-deficient mice, MCL- and medial meniscus-deficient rabbits, and ACL-MCL-medial meniscus–deficient rabbits, respectively. In ACL-deficient mice treated with 1 injection of IL-1Ra–encoded HDAd particles,[23] there was a mild decrease in IL-1β levels, with no significant difference compared with sham controls at 2.5 months post-injection ($P > .05$).

IL-1Ra Expression

IL-1Ra levels in gene therapy models were analyzed in 4 preclinical studies.[24,25,27,28] Adequately maintained IL-1Ra expression was reported in 3 studies at 1 or 2 weeks, whereas 1 study reported a significant fall in expression after 4 weeks. In 2 studies administering 1 injection of synovial cells transfected with an IL-1Ra encoded retroviral vector,[24,27] IL-1Ra expression was stable after 14 days in comparison with absent levels in control MCL and medial meniscus-deficient rabbit knees,[27] whereas IL-1Ra levels were considerably diminished at 2 and 4 weeks compared with initial injection levels in ACL-deficient dog knees.[25] In rabbits treated with 1

Table 1. Preclinical Study Designs and Outcomes

Study	Drug	Subjects	PTOA Model	Experimental Groups	Results Summary
Nixon et al., 2018[22]	HDAd-mIL-1Ra	FVB/N mice	Cruciate ligament transection	1) HDAd-IL-1ra 2) HDAd-GFP control 3) PBS 4) Sham	Gross morphology (micro-CT): 2 days before CLT: • significantly greater cartilage volume than the HDAd-GFP group, no difference than placebo or sham • Significantly greater bone surface covered by cartilage than HDAd-GFP and PBS groups 2 weeks after CLT: • Significantly greater cartilage volume, surface area, and bone area covered by cartilage compared with HDAD-GFP and PBS, no significant difference with sham Histology (OARSI) 2 days before injury: • Significantly lower OARSI scores and fewer osteophytes than placebo and HDAd-GFP control 3 days after CLT: • No significant difference for mean histologic score for cartilage damage, number of osteophytes between GFP and PBS groups, significantly greater OARSI scores than sham 2 weeks after CLT: • No significant difference in OARSI scores compared with HDAd-GFP and placebo, significantly worse OARSI scores than sham
Stone et al., 2019[23]	HDV- NfkB-Il1ra	Male FVB mice	Cruciate ligament transection OR Meniscotibial ligament transection	1) monotherapy HDAd-NFkB-Il1ra 2) monotherapy HDAd-EF1-Prg4 3) HDAd-empty (control) 4) combination of HDAd- NFkB-Il1ra and HDVEF1-Prg4	IL-1β expression: mild decrease in IL-1β levels, no significant difference compared with sham control Gross morphology (micro-CT): Significantly greater cartilage

(continued)

Table 1. Continued

Study	Drug	Subjects	PTOA Model	Experimental Groups	Results Summary
				5) no injections (sham group)	volume loss compared with sham control at 2.5 months, with no differences in cartilage volume compared with empty HDV IL-1ra treatment retained significantly more covered surface area compared with empty HDAd-treated mice IL-1ra treatment 2 weeks after DMM had no statistical differences in cartilage volume compared with sham at 2.5 months; effect was lost at 3.5 months. Combined treatment with IL-1ra and Prg4 gene therapy had no difference in cartilage volume compared with sham and was significantly greater than IL-1ra or Prg4 monotherapy
Wang et al., 2006[24]	Ad-IL-1Ra	New Zealand white rabbits	MCL tear + medial meniscectomy	1) IL-1ra 2) sTNF-RI 3) Combination 4) Ad-GFP (control)	IL-1ra expression: Significant levels of IL-1ra were detected at both 3 and 7 days in IL-1ra group, undetectable in knees receiving HDAd-GFP control Histology (subjective): Distinct decrease in cartilage lesion size in IL-1ra group compared with HDAd-GFP and sTNF-RI monotherapy
Pelletier et al., 1997[25]	rHuIL-1Ra gene	Mongrel dogs	ACL section only OR ACL section + contralateral knee synovectomy	1) PBS 2) Lac Z 3) IL-1ra	IL-1ra expression: IL-1ra levels were considerably diminished at 2 and 4 weeks compared with levels after initial injection Gross morphology (macroanalysis): Significantly reduced size and grade of tibial plateau cartilage lesions compared with placebo and lac Z groups, no differences in femoral lesions **Histology (subjective):** Severity of microscopic lesions were significant less than lac Z, no significant difference in structural

(continued)

Table 1. Continued

Study	Drug	Subjects	PTOA Model	Experimental Groups	Results Summary
					changes than control and lac Z groups, significantly greater histologic score for the synovial membrane than control and lac Z group due to significantly greater mononuclear cell infiltration ($P < .008$)
Tang et al., 2015[26]	IL-1β siRNA	New Zealand white rabbits	ACL + MCL + medial meniscectomy	1) saline (control) 2) TNF-alpha siRNA 3) IL-1β siRNA 4) Both TNF-alpha and IL-1β siRNA	IL-1β expression: IL-1β expression was significantly lower in IL-1β siRNA group than treatment control group at 1, 2, and 4 weeks Histology (subjective and Mankin score) Reduced cartilage damage at 4 weeks compared with the control, significantly lower Mankin scores at 1, 2, and 4 weeks after injury
Zhang et al., 2004[27]	PLXRN-IL-1Ra	New Zealand white rabbits	MCL tear + medial meniscectomy	1) no treatment control 2) lacZ control 3) IL-1ra gene 4) IL-10 gene 5) IL-1ra and IL-10, 6) naïve control (no surgery)	IL-1ra expression: IL-1ra levels were stable after 14 days, whereas no synovial fluid IL-1ra was detected in control rabbit knees Immunohistochemistry: Expression of IL-1ra was maintained in grafted cells after 14 days Histology (subjective): Distinct reduction in cartilage lesion size compared with the treatment control group
Zhang et al., 2006[28]	PcDNA3.1-IL-1Ra plasmid	New Zealand white rabbits	MCL tear + medial meniscectomy	1) placebo 2) IL-1ra 3) IL-10contralateral knees served as controls	IL-1ra expression: Detectable level of IL-1ra only in knees treated with the chitosan-IL-1Ra nanoparticle Immunohistochemistry: IL-1ra detected in a small group of chondrocytes compared with none in the placebo group after 14 days, IL-1ra was not detected in synovial cells in placebo or IL-10 groups Histology (subjective): less severe cartilage lesions compared with placebo
Zhang et al., 2015[29]	Recombinant IL-1ra plasmid DNA	New Zealand white rabbits	MCL tear + medial meniscectomy	1) control (normal saline) 2) IL-ra transfection	IL-1β expression IL-1β and TNF-α levels in dual and

(continued)

Table 1. Continued

Study	Drug	Subjects	PTOA Model	Experimental Groups	Results Summary
				3) TGF-β1 transfection 4) IL-1ra/TGF-β1 double transfection	single groups significantly lower than control ($P < .05$) IL-1ra expression: Significantly greater IL-1ra levels than the blank control group Immunohistochemistry: Subjectively darker staining of type II collagen than the control group Histology (subjective and Mankin score): IL-1ra—treated group did not develop osteophytes, had darker color, subjectively better safranin O state, and significantly lower Mankin scores compared with the control group.
Lin et al., 2021[31]	miR-144-3p	male Sprague—Dawley rats	ACL transection	1) control (no tear), 2) ACLT 3) ACLT + control mimic (miRNA negative control) 4) ACLT + miR-144-3p	Gross morphology (micro-CT): Significantly reduced changes in trabecular thickness, number, and separation than untreated ACL-deficient and ACL-deficient mice treated with a mimic control Immunohistochemistry: significantly reduced IL-1β—positive synovial cells compared with both untreated ACL deficient and ACL-deficient mice treated with mimic control groups Histology (OARSI): Significantly lower OARSI and cartilage scores than both untreated ACL-deficient and ACL-deficient mice treated with mimic control groups.
Zhang et al., 2021[30]	IL-1β shRNA	Male C57BL/6 mice	DMM	1) yeast only (control)	IL-1β expression: Significantly decreased IL-1β expression compared with the control group in mice. Histology (OARSI): Significantly lower OARSI grades compared with the control group.

ACL, anterior cruciate ligament; ACLT, anterior cruciate ligament transection; CLT, cruciate ligament transection; CT, computed tomography; DMM, destabilization of the medial meniscus (transection of the anterior attachment of the meniscotibial ligament); HDAd, helper-dependent adenovirus; HDAd-GFP, green fluorescence protein encoded helper-dependent adenovirus; IL-1β, interleukin 1β, IL-1Ra; interleukin-1 receptor antagonist; MCL, medial collateral ligament; OARSI, Osteoarthritis Research Society International; PBS, phosphate-buffered saline; PTOA, post-traumatic osteoarthritis; siRNA, small interfering RNA; sTNF-RI, soluble tumor necrosis factor-α receptor type I; TGF-β, transforming growth factor β; TNF-α, tumor necrosis factor α.

injection of IL-1Ra encoded helper-dependent adenovirus particles,[24] significant levels of IL-1Ra were detected at both 3 and 7 days, whereas levels were undetectable in knees receiving green fluorescence protein–encoded HDAd control injections. Rabbits treated with liposome-mediated recombinant IL-1Ra plasmid DNA injections had significantly greater IL-1Ra levels than the blank control group at 14 days ($P < .05$).[29]

Immunohistochemistry Analysis

There were 4 studies that reported IL-1Ra treatment efficacy with immunohistochemistry analysis.[27-29,31] All studies reported favorable immunohistochemical profiles following therapy. Sustained IL-1ra expression was seen in 2 rabbit studies after treatment with chitosan-DNA nanoparticles[28] and retrovirus transfected synovial cells.[27] Significant reduction in IL-beta positive synovial cells with liposome-mediated miR-1443p treatment in rats.[31] Liposome-mediated recombinant IL-1Ra plasmid DNA injections resulted darker type II collagen staining compared with controls.[29]

Gross Morphology

Gross morphology of cartilage was evaluated in 4 studies.[22,23,25,31] Three studies reported gross cartilage changes on micro-CT, whereas 1 study performed macroscopic examination. All 4 studies demonstrated that selective IL-1 inhibitor therapy had protective effects on cartilage or subchondral bone integrity following meniscal or ACL trauma. Cartilage surface area and volume were shown to be significantly greater than control treatment groups ($P < .05$) and were comparable with sham groups in 2 ACL-deficient mouse models following IL-1ra–encoded adenovirus vehicles on micro-CT analysis.[22,23] Significant reductions in dog tibial plateau cartilage lesion size ($P < .04$) and grade ($P < .01$) after synovial cells transfected with IL-1Ra encoded retrovirus compared with placebo and untreated groups.[25] In rats treated with liposome-mediated miR-1443p, there were significantly reduced changes in trabecular thickness, number, and separation than untreated ACL-deficient and ACL-deficient mice treated with a mimic control on micro-CT analysis ($P < .05$).[31]

Histology

Histologic analysis was performed in 9 preclinical studies.[22,24-31] Histologic outcomes were assessed using OARSI grades in 3 studies, Mankin scores in 2 studies, and subjective microscopic examination only in 4 studies. Lower OARSI grades and Mankin scores indicated lower stages of OA. In 7 studies,[24,26-31] histologic and microscopic analyses demonstrated that selective IL-1 inhibitor treatment had a significant role in reducing the progression of OA regardless of drug vehicle, characterized by a significant reduction in OA progression[29-31] and significantly less cartilage damage[26,27] or lesion size[24,28] when compared with control groups. Conflicting histologic evidence of reduced OA progression after treatment with IL-1Ra encoded helper-dependent adenovirus particles was reported in one study, which demonstrated that significant reduction in OA progression was dependent on timing of intervention and skeletal maturity.[22] Compared with placebo and treatment control groups, significant improvements were only seen in skeletally immature mice treated 2 days before ACL transection ($P < .05$), whereas skeletally immature mice treated 2 weeks after ACL transection and skeletally mature mice treated 3 days after injury did not significantly reduce OA progression, while also having significantly worse OA progression than sham controls ($P < .05$).[22] In 1 study in dogs, IL-1ra treatment did not significantly alter microscopic structural changes compared with placebo.[25]

Clinical Study

Patient demographics, treatment protocol, and drug characteristics for the 1 identified clinical trial[12] are summarized in Table 3. Patients with acute ACL tears with or without concomitant MCL or meniscal lesions were treated with one intraarticular injection of recombinant IL-1Ra (Anakinra/Kineret; Sobi). Therapy was initiated at a mean 2 weeks postinjury and outcomes were assessed at a mean 35 days after injury. Compared with the placebo group, patients treated with IL-1Ra injections had significantly improved pain ($P = .0011$), quality of life ($P = .0048$), activities of daily living ($P = .0015$), and sports function ($P = .0026$) KOOS scores 14 days after treatment. Patients treated with IL-1Ra had reduced yet nonsignificant synovial fluid IL-1β activity from baseline ($P = .06$). Final IL-1Ra and IL-1β changes in expression were not significantly different from the placebo group ($P > .05$).

Discussion

The most important findings of this study were that early intervention with selective IL-1 inhibitor therapies in preclinical animal model studies was effective in reducing proinflammatory IL-1β levels in the acute and subacute phases following traumatic knee injury and significantly reduced cartilage damage, lesion size, and PTOA progression at short-term end points. However, it was found that the effect of these therapies diminished over time, indicating the need for further investigations analyzing long-term outcomes and optimization of drug-delivery systems and protocols. Furthermore, there is a significant lack of clinical trials investigating IL-1Ra therapy in the setting of PTOA.

Clinically, PTOA of the knee can result in chronic pain and functional impairment years after a traumatic event secondary to prominent progressive loss of articular cartilage, subchondral bone remodeling, and

Table 2. Preclinical Treatment Protocols and Drug Delivery Characteristics

Study	Drug Type/ Mechanism	Vehicle	Route	Dosage and Frequency	Start Day	Duration	Primary Outcomes
Nixon et al., 2018[22]	Gene therapy/IL-1ra	HDAd-mediated	Intra-articular	1×10^8 vp/once	1) 2 days before CLT 2) Skeletally immature mice: 2 weeks after CLT 3) Skeletally mature mice: 3 days after CLT	1) 6 weeks 2) 8 weeks 3) 2 months	Histology (OARSI), micro-CT, hot plate analysis
Stone et al., 2019[23]	Gene therapy/IL-1ra	HDAd-mediated	Intra-articular	1×10^8 vp in 5microl of PBS/ once	2 weeks' postsurgery	1) 2.5 months 2) 3.5 months	Cartilage volume, surface area, gene expression, noxious stimuli response
Wang et al., 2006[24]	Gene therapy/IL-1ra	HDV-mediated	Intra-articular	1×10^8 pfu of adenovirus in 0.2 ml saline/once	5 days' postsurgery	7 days	IL-1ra and sTNF-RI expression, quantitative histology
Pelletier et al., 1997[25]	Gene therapy/IL-1ra	Retrovirus infected synovial fibroblasts	Intra-articular	60×10^6 cells in 2 mL of PBS/once	2 days after surgery	4 weeks	Macrocartilage analysis, histology, IL-1ra levels
Tang et al., 2015[26]	Gene therapy/IL-1β siRNA (Il-1ra gene)	Recombinant lentivirus (retrovirus) infected synovial cells	Intra-articular	5×10^6 cells in 1 mL of injection/once	7 days' postsurgery	1, 2, or 4 weeks	Histology, Mankin score, IL-1β, and TNF-αexpression (ELISA)
Zhang et al., 2004[27]	Gene therapy/IL-1ra	Retrovirus-infected synovial cells	Intra-articular	5×10^6 cells in 0.5 mL of PBS/once	5 days' postsurgery	14 days	IL-1ra expression, histology, immunohistochemical analysis
Zhang et al., 2006[28]	Gene therapy/IL-1ra	Chitosan-DNA nanoparticles	intra-articular	0.4 mL of chitosan-DNA containing 20 microg PcDNA3.1-IL-1Ra Plasmid/3 total in 48-hour intervals	5 days postsurgery	14 days	IL-1ra expression (ELISA), immunohistochemical analysis, histology
Zhang et al., 2015[29]	Gene therapy/IL-1ra	Liposomes	Intra-articular	100 μg + lipofectamine TM 2000 reagent 50 μL + NS 0.5 mL/ twice	5 and 7 days after surgery	14 days	IL-1ra expression (ELISA), histology, immunohistochemical analysis

(continued)

Table 2. Continued

Study	Drug Type/ Mechanism	Vehicle	Route	Dosage and Frequency	Start Day	Duration	Primary Outcomes
Lin et al., 2021[31]	microRNA/direct IL-1β binding and downregulation	None	Intra-articular	5 nmol in 25 µL of PBS + Lipofectamine 2000 (25 µL) in PBS (50 µL PBS, for a total volume of 100 µl)/once weekly for six weeks	NR	10 weeks	micro-CT analysis, histology, IL-1β and miR-144-3p expression
Zhang et al., 2021[30]	Gene therapy/ recombinant IL-1β shRNA	Yeast microcapsule	Oral	30 mg/kg rYeast/3× per day every 2 days	1 month after surgery	40 days	IL-1β expression in bone marrow and knee joint, inflammatory markers, cartilage (OARSI)

CLT, cruciate ligament transection; CT, computed tomography; ELISA, enzyme-linked immunosorbent assay; HDAd, helper-dependent adenovirus; IL-1Ra, interleukin-1 receptor antagonist; OARSI, Osteoarthritis Research Society International; PBS, phosphate-buffered saline; vp, viral particles; siRNA, small interfering RNA; sTNF-RI, soluble tumor necrosis factor-α receptor type I.

intra-articular inflammation. While OA has historically been understood as a clinical end point resulting from multifactorial processes including age, obesity, and chronic "wear and tear," [23] PTOA manifests after an identifiable cause, resulting in a degenerative cascade involving several inflammatory mediators.[14,32] The acute inflammatory phase following a traumatic event provides the opportunity to initiate treatment targeting specific pathophysiology with minimal confounding contributions. In a comprehensive review of disease markers and implications, Khella et al.[32] reported IL-1β as a credible factor inducing PTOA progression, confirming that an imbalance of IL-1β expression perpetuated prolonged postinjury inflammation contributing to cartilage damage.[32] Furthermore, the authors reported that IL-1β is significantly increased in the acute and subacute phases of PTOA progression for up to 1.5 months,[33,34] emphasizing the need for effective drug-delivery systems to ensure IL-1 antagonism to remain in the joint for this duration. However, in contrast to previous studies evaluating IL-1 antagonist treatment in established OA, it has been reported that IL-1β effects may not be contributory 1.5 months following ACL injury,[32,35] indicating that delayed IL-1β targeted therapies may not be effective in the chronic stages of OA development or once PTOA is diagnosed.[36]

In studies that initiated treatment within 5 to 7 days after injury, IL-1β expression was found to be significantly reduced in the acute and subacute inflammatory phases with correlating reductions in cartilage degeneration. In contrast, Stone et al.[23] reported nonsignificant IL-1β expression reduction and limited long-term efficacy in cartilage integrity following IL-1Ra monotherapy. However, the authors initiated treatment at 2 weeks following surgery and evaluated IL-1β expression at 2.5 and 3.5 months.[23] These findings suggest that early intervention, as aligned with the natural pathologic course, is necessary to provide a protective benefit to reduce the risk PTOA progression, regardless of drug delivery methods.

Drug retention in the knee joint is a well-known obstacle to pharmaceutical efficacy and significantly limits the efficacy of recombinant IL-Ra products such as anakinra for sustained treatment due to a half-life of 4 to 6 hours. Although demonstrating short-term clinical potential in some OA trials[37,38] and the included PTOA trial,[12] the need to investigate long-term release drug delivery systems to reduce repeated intra-articular injections is warranted.[39] Helper-dependent adenoviruses have been recently used for its ability to transduce host synoviocytes and chondrocytes with the ability to provide long-term expression of selected genes within the knee joint.[40] Similarly, retrovirus-mediated vectors introduce genes via ex vivo transfer with harvested cells.[27] However, there is still potential for an immune response directed at the HDAd viral capsid and injected

Table 3. Clinical Study Treatment Protocol, Drug and Study Characteristics, and Demographics

Study	Drug	Sample Size	Age, y	Injury Details	Inclusion Criteria	Drug Type/ Mechanism	Vehicle	Route	Dosage and frequency	Start Day	Evaluation Time Points	Primary Outcomes
Kraus et al., 2012[12]	Anakinra	Anakinra group: n = 6 (2 female) placebo; n = 5 (3 female)	24 ± 4 years	Anakinra treatment group; 2 isolated ACL tears, 2 ACL tears with partial MCL tear, 2 ACL tears with meniscal tear Placebo group; One isolated ACL tear, one ACL tear with partial MCL tear; two ACL tears with meniscal tear, one ACL tear with MCL and meniscal tear	Injury less than 4 weeks before eval, ACL confirmed tear on MRI, age 18-30 years, negative pregnancy, nonobese, no previous injury/ OA. Allowed knee bracing, NSAIDs, analgesics, ice, ROM and quad training permitted (no significant differences between groups)	Recombinant protein/IL-1ra	None	Intra-articular	150 mg/mL (1 mL)/once	mean 2 weeks post injury (15 ± 7 days)	4, 14, and 28 days	SF IL-1a, IL-1β, IL-1Ra and serum HA, KOOS score

ACL, anterior cruciate ligament; HA, hyaluronan; IL-1Ra; interleukin-1 receptor antagonist; KOOS, Knee Injury and Osteoarthritis Outcome Score; MCL, medial collateral ligament; MRI, magnetic resonance imaging; NSAIDs, nonsteroidal anti-inflammatory drugs; OA, osteoarthritis; ROM, range of motion.

cells which has limited higher dosages of therapy,[23] whereas ex vivo cell manipulation requires cumbersome cell manipulation prior to initiating therapy. The immunogenic potential of retrovirus infected cells was observed by Pelletier et al,[25] reporting that IL-1Ra–transduced cells provoked increased synovial inflammation resulting in a loss of IL-1Ra expression. To combat these limitations, drug-delivery systems using nonviral microcapsule gene carriers have been developed and have shown to be safe and promising, although preparation, cellular infectivity, and federal approval remain barriers to becoming convenient and efficacious clinical products.[28,41]

In preclinical studies reporting IL-1Ra expression, the majority detected sustained concentrations up to 14 days following injection. However, in the only study evaluating IL-1Ra concentrations after 14 days, Pelletier et al.[25] reported significantly diminished levels at 4 weeks after injection with retrovirus infected cells. Furthermore, studies investigating long term histologic and gross morphology results reported varying efficacy at 4 weeks to 3.5 months, indicating a diminishing effect of drug delivery systems. Overall, microcapsule-mediated IL-1Ra delivery appeared to provide the most beneficial long-term effect on cartilage integrity at end points of 2 weeks or longer, although there were differences in microcapsule biochemical composition across these studies. Therefore, further investigations of microcapsule mediated delivery should be performed to delineate their efficacy in clinical trials.

In this systematic review, there was a significant lack of identified clinical trials investigating IL-1Ra treatment to reduce PTOA progression, accentuating the recent findings of DePhillipo et al.,[10] who reported a significant lack of clinical trials investigating disease-modifying therapies in the clinical trial pipeline. The pilot clinical study reported that anakinra injections administered 2 weeks after ACL injury had significantly greater KOOS scores compared with the placebo group, although IL-1Ra activity was significantly diminished at a mean 35 days follow-up.[12] Albeit promising preliminary results in a significantly limited sample size, the clinical literature has yet to be established and lacks pertinent value to clearly evaluate the disease-modifying potential of selective IL-1 inhibitor therapy as a viable treatment option to patients at this time. However, although clinical conclusions cannot be drawn, this systematic review best serves to promote the further advancement and optimization of selective IL-1 inhibitor protocols as supported by the promising results in the basic science literature demonstrating its efficacious role in reduction of OA progression, maintenance of cartilage integrity, and direct antagonism of pathologic pathways involved in the development of PTOA.

Given the findings of the included preclinical studies, it should be emphasized that future preclinical selective IL-1 inhibitor therapy trials should be performed in the setting of PTOA, as the findings of the current review suggest that early intervention is necessary to target and modify pathophysiologic pathways involved in early OA progression. As in vivo animal models have been demonstrated to successfully replicate clinical post-traumatic inflammatory conditions, the drug delivery systems summarized in this review may also provide the foundation for future clinical trials.[32] Overall, selective IL-1 inhibitor drug-delivery protocols must be further optimized, evaluated, and compared in homogenous PTOA models to limit potential confounding variables that could influence drug efficacy. Furthermore, it is currently unknown of the effects of selective IL-1 inhibition and ACL graft healing potential and future studies are warranted to be performed in ACL reconstruction models before performing clinical trials.

Limitations

We acknowledge some limitations to this systematic review. Studies that only evaluated the effects selective IL-1 inhibitor therapy in the setting of post-traumatic knee injury were included in this study and the results are not generalizable to established OA treatment. Most importantly, there was only one clinical study included in this systematic reviewing, highlighting a severe lack of evidence to recommend selective IL-1 inhibitor treatment in the clinical setting. PTOA models were significantly heterogenous, varying by ligamentous and meniscal deficiencies performed in a variety of animal models with inconsistent age groups, which limits direct comparisons and implications for clinical treatment. Furthermore, there was significant heterogeneity in drug-delivery vehicles, with several drug delivery systems only investigate in single trials potentially limiting the ability to translate to clinical trials which warrants further investigations. We also acknowledge that the development of PTOA is multifactorial in nature, including the contributions of subchondral bone integrity after injury. As few included studies reported on subchondral bone integrity before and after treatment, direct comparisons on treatment effect on the subchondral bone could not be adequately analyzed. There were no studies investigating the effect selective IL-1 inhibitor agents following ACL reconstruction. It is possible that relevant articles were not identified with our search criteria, as with all systematic reviews. In addition, we selected only literature in the English language, potentially contributing to publication bias.

Conclusions

Early intervention with selective IL-1 inhibitor therapy were effective in reducing proinflammatory IL-1β levels in the acute and subacute phases following traumatic knee injury in preclinical animal model studies, while significantly reducing cartilage damage,

lesion size, and PTOA progression at short-term follow-up. However, it was found that the effect of these therapies diminished over time.

References

1. Suomalainen P, Järvelä T, Paakkala A, Kannus P, Järvinen M. Double-bundle versus single-bundle anterior cruciate ligament reconstruction: A prospective randomized study with 5-year results. *Am J Sports Med* 2012;40:1511-1518. doi:10.1177/0363546512448177.
2. Cinque ME, Dornan GJ, Chahla J, Moatshe G, LaPrade RF. High rates of osteoarthritis develop after anterior cruciate ligament surgery: An analysis of 4108 patients. *Am J Sports Med* 2018;46:2011-2019. doi:10.1177/0363546517730072.
3. Cinque ME, Kunze KN, Williams BT, Moatshe G, LaPrade RF, Chahla J. Higher incidence of radiographic posttraumatic osteoarthritis with transtibial femoral tunnel positioning compared with anteromedial femoral tunnel positioning during anterior cruciate ligament reconstruction: A systematic review and meta-analysis. *Am J Sports Med* 2022;50:255-263.
4. Lohmander LS, Ostenberg A, Englund M, Roos H. High prevalence of knee osteoarthritis, pain, and functional limitations in female soccer players twelve years after anterior cruciate ligament injury. *Arthritis Rheum* 2004;50:3145-3152. doi:10.1002/art.20589.
5. von Porat A, Roos EM, Roos H. High prevalence of osteoarthritis 14 years after an anterior cruciate ligament tear in male soccer players: A study of radiographic and patient relevant outcomes. *Ann Rheum Dis* 2004;63:269-273. doi:10.1136/ard.2003.008136.
6. Menendez MI, Hettlich B, Wei L, Knopp MV. Feasibility of Na18F PET/CT and MRI for noninvasive in vivo quantification of knee pathophysiological bone metabolism in a canine model of post-traumatic osteoarthritis. *Mol Imaging* 2017;16. doi:10.1177/1536012117714575:1536012117714575.
7. Riordan EA, Little C, Hunter D. Pathogenesis of post-traumatic OA with a view to intervention. *Best Pract Res Clin Rheumatol* 2014;28:17-30. doi:10.1016/j.berh.2014.02.001.
8. Centers for Disease Control and Prevention (CDC). National and state medical expenditures and lost earnings attributable to arthritis and other rheumatic condition—United States, 2003. *MMWR Morb Mortal Wkly Rep* 2007;56:4-7.
9. Hunter DJ, Bierma-Zeinstra S. Osteoarthritis. *Lancet Lond Engl* 2019;393:1745-1759. doi:10.1016/S0140-6736(19)30417-9.
10. DePhillipo NN, Aman ZS, Dekker TJ, Moatshe G, Chahla J, LaPrade RF. Preventative and disease-modifying investigations for osteoarthritis management are significantly under-represented in the clinical trial pipeline: A 2020 review. *Arthroscopy* 2021;37:2627-2639. doi:10.1016/j.arthro.2021.03.050.
11. Bajpayee AG, De la Vega RE, Scheu M, et al. Sustained intra-cartilage delivery of low dose dexamethasone using a cationic carrier for treatment of post traumatic osteoarthritis. *Eur Cell Mater* 2017;34:341-364. doi:10.22203/eCM.v034a21.
12. Kraus VB, Birmingham J, Stabler TV, et al. Effects of intraarticular IL1-Ra for acute anterior cruciate ligament knee injury: A randomized controlled pilot trial (NCT00332254). *Osteoarthritis Cartilage* 2012;20:271-278. doi:10.1016/j.joca.2011.12.009.
13. Lieberthal J, Sambamurthy N, Scanzello CR. Inflammation in joint injury and post-traumatic osteoarthritis. *Osteoarthritis Cartilage* 2015;23:1825-1834. doi:10.1016/j.joca.2015.08.015.
14. Wang LJ, Zeng N, Yan ZP, Li JT, Ni GX. Post-traumatic osteoarthritis following ACL injury. *Arthritis Res Ther* 2020;22:57. doi:10.1186/s13075-020-02156-5.
15. Mabey T, Honsawek S. Cytokines as biochemical markers for knee osteoarthritis. *World J Orthop* 2015;6:95-105. doi:10.5312/wjo.v6.i1.95.
16. Ajrawat P, Dwyer T, Chahal J. Autologous interleukin 1 receptor antagonist blood-derived products for knee osteoarthritis: A systematic review. *Arthroscopy* 2019;35:2211-2221. doi:10.1016/j.arthro.2018.12.035.
17. Caron JP, Fernandes JC, Martel-Pelletier J, et al. Chondroprotective effect of intraarticular injections of interleukin-1 receptor antagonist in experimental osteoarthritis. Suppression of collagenase-1 expression. *Arthritis Rheum* 1996;39:1535-1544. doi:10.1002/art.1780390914.
18. Jiang Y, Genant HK, Watt I, et al. A multicenter, double-blind, dose-ranging, randomized, placebo-controlled study of recombinant human interleukin-1 receptor antagonist in patients with rheumatoid arthritis: Radiologic progression and correlation of Genant and Larsen scores. *Arthritis Rheum* 2000;43:1001-1009. doi:10.1002/1529-0131(200005)43:5<1001::AID-ANR7>3.0.CO;2-P.
19. Seckinger P, Klein-Nulend J, Alander C, Thompson RC, Dayer JM, Raisz LG. Natural and recombinant human IL-1 receptor antagonists block the effects of IL-1 on bone resorption and prostaglandin production. *J Immunol* 1990;145:4181-4184.
20. Moher D, Liberati A, Tetzlaff J, Altman DG, PRISMA Group. Preferred reporting items for systematic reviews and meta-analyses: The PRISMA statement. *PLoS Med* 2009;6:e1000097. doi:10.1371/journal.pmed.1000097.
21. Jotanovic Z, Mihelic R, Sestan B, Dembic Z. Role of interleukin-1 inhibitors in osteoarthritis: An evidence-based review. *Drugs Aging* 2012;29:343-358. doi:10.2165/11599350-000000000-00000.
22. Nixon AJ, Grol MW, Lang HM, et al. Disease-modifying osteoarthritis treatment with interleukin-1 receptor antagonist gene therapy in small and large animal models. *Arthritis Rheumatol* 2018;70:1757-1768. doi:10.1002/art.40668.
23. Stone A, Grol MW, Ruan MZC, et al. Combinatorial *Prg4* and *Il-1ra* gene therapy protects against hyperalgesia and cartilage degeneration in post-traumatic osteoarthritis. *Hum Gene Ther* 2019;30:225-235. doi:10.1089/hum.2018.106.
24. Wang H jun, long Yu C, Kishi H, Motoki K, Mao Z bin, Muraguchi A. Suppression of experimental osteoarthritis by adenovirus-mediated double gene transfer. *Chin Med J (Engl)* 2006;119:1365-1373.
25. Pelletier JP, Caron JP, Evans C, et al. In vivo suppression of early experimental osteoarthritis by interleukin-1 receptor antagonist using gene therapy. *Arthritis Rheum* 1997;40:1012-1019. doi:10.1002/art.1780400604.

26. Tang Q, Hao L, Peng Y, et al. RNAi Silencing of IL-1β and TNF-α in the treatment of post-traumatic arthritis in rabbits. *Chem Biol Drug Des* 2015;86:1466-1470. doi:10.1111/cbdd.12611.
27. Zhang X, Mao Z, Yu C. Suppression of early experimental osteoarthritis by gene transfer of interleukin-1 receptor antagonist and interleukin-10. *J Orthop Res* 2004;22:742-750. doi:10.1016/j.orthres.2003.12.007.
28. Zhang X, Yu C, XuShi, Zhang C, Tang T, Dai K. Direct chitosan-mediated gene delivery to the rabbit knee joints in vitro and in vivo. *Biochem Biophys Res Commun* 2006;341:202-208. doi:10.1016/j.bbrc.2005.12.171.
29. Zhang P, Zhong ZH, Yu HT, Liu B. Exogenous expression of *IL-1Ra* and *TGF-β1* promotes in vivo repair in experimental rabbit osteoarthritis. *Scand J Rheumatol* 2015;44:404-411. doi:10.3109/03009742.2015.1009942.
30. Zhang L, Peng H, Feng M, Zhang W, Li Y. Yeast microcapsule-mediated oral delivery of IL-1β shRNA for post-traumatic osteoarthritis therapy. *Mol Ther Nucleic Acids* 2021;23:336-346. doi:10.1016/j.omtn.2020.11.006.
31. Lin Y, Ko C, Liu S, et al. miR-144-3p ameliorates the progression of osteoarthritis by targeting IL-1β: Potential therapeutic implications. *J Cell Physiol* 2021;236:6988-7000.
32. Khella CM, Asgarian R, Horvath JM, Rolauffs B, Hart ML. An evidence-based systematic review of human knee post-traumatic osteoarthritis (PTOA): Timeline of clinical presentation and disease markers, comparison of knee joint PTOA models and early disease implications. *Int J Mol Sci* 2021;22:1996. doi:10.3390/ijms22041996.
33. Bigoni M, Sacerdote P, Turati M, et al. Acute and late changes in intraarticular cytokine levels following anterior cruciate ligament injury. *J Orthop Res* 2013;31:315-321. doi:10.1002/jor.22208.
34. Irie K, Uchiyama E, Iwaso H. Intraarticular inflammatory cytokines in acute anterior cruciate ligament injured knee. *Knee* 2003;10:93-96. doi:10.1016/s0968-0160(02)00083-2.
35. Watt FE, Paterson E, Freidin A, et al. Acute molecular changes in synovial fluid following human knee injury: Association with early clinical outcomes. *Arthritis Rheumatol Hoboken NJ* 2016;68:2129-2140. doi:10.1002/art.39677.
36. Panina SB, Krolevets IV, Milyutina NP, et al. Circulating levels of proinflammatory mediators as potential biomarkers of post-traumatic knee osteoarthritis development. *J Orthop Traumatol* 2017;18:349-357. doi:10.1007/s10195-017-0473-8.
37. Chevalier X, Giraudeau B, Conrozier T, Marliere J, Kiefer P, Goupille P. Safety study of intraarticular injection of interleukin 1 receptor antagonist in patients with painful knee osteoarthritis: A multicenter study. *J Rheumatol* 2005;32:1317-1323.
38. Chevalier X, Goupille P, Beaulieu AD, et al. Intraarticular injection of anakinra in osteoarthritis of the knee: A multicenter, randomized, double-blind, placebo-controlled study. *Arthritis Rheum* 2009;61:344-352. doi:10.1002/art.24096.
39. Martel-Pelletier J, Pelletier JP. Osteoarthritis: A single injection of anakinra for treating knee OA? *Nat Rev Rheumatol* 2009;5:363-364. doi:10.1038/nrrheum.2009.121.
40. Palmer DJ, Ng P. Helper-dependent adenoviral vectors for gene therapy. *Hum Gene Ther* 2005;16:1-16. doi:10.1089/hum.2005.16.1.
41. Pouton CW, Seymour LW. Key issues in non-viral gene delivery. *Adv Drug Deliv Rev* 1998;34:3-19. doi:10.1016/s0169-409x(98)00048-9.