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REVIEW

Tetracycline use in treating osteoarthritis: a systematic review

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Abstract

Background and aims The purpose of the review was to synthesize the current literature regarding tetracyclines in the treatment of osteoarthritis.

Methods Using multiple databases, a systematic review was performed with customized search terms crafted to identify studies examining doxycycline or minocycline in the treatment of osteoarthritis. Results were classified into basic science mechanistic studies, in vivo animal studies, and human clinical trials. A total of 1446 potentially relevant studies were reviewed, and after exclusion criteria were applied, 23 investigations were included in the final analysis.

Results From 12 basic science mechanistic studies, we report on three main mechanisms by which tetracyclines may exert benefit in osteoarthritis progression: matrix metalloproteinase inhibition, immunomodulation, and nitric oxide synthase inhibition. Seven animal studies showed generally encouraging results. Four articles reported human clinical studies, showing mixed results in the treatment of osteoarthritis, potentially related to the choice of patient population, primary outcomes, and timing of treatment.

Conclusion Tetracyclines have the potential to benefit osteoarthritis patients via multiple mechanisms. Further study is warranted to examine the optimal dose and timing of tetracycline treatment in osteoarthritis.

Keywords Osteoarthritis · Doxycycline · Minocycline · Disease-modifying osteoarthritis drugs · Dmoads

Introduction

Idiopathic osteoarthritis is traditionally considered a disease of mechanical cartilage degradation [1]. Research now characterizes osteoarthritis as a complex disease process involving both mechanical factors and inflammatory contributors [2]. This perspective shift is supported by recent research which has further elucidated the mechanisms by which the immune system perpetuates joint damage.

Multiple studies implicate activated macrophages, neutrophils, and T-cells in the progression of osteoarthritis [3–9]. Inflammatory factors such as cytokines, chemokines, and matrix metalloproteinases (MMPs) produced by joint tissues in response to acute or chronic injury have been found to lead to cartilage and meniscus breakdown [2]. MMPs, in particular MMP-3 and MMP-13, degrade type II collagen leading to progression of osteoarthritis [10, 11]. Posttraumatic arthritis, also traditionally thought of as mechanical in nature, is tied to similar mechanisms of inflammation leading to MMP upregulation. For example, ACL remnants were found to induce MMP-13 expression in the acute phase of an injury, which may explain why posttraumatic arthritis frequently develops despite surgical reconstruction [12]. The advancements in the knowledge of the pathogenesis of osteoarthritis truly characterizes osteoarthritis as a disease of the joint as an organ [13].

Given the complex and interrelated pathways involved in the pathogenesis of osteoarthritis, the potential for a reliable disease-modifying osteoarthritis drug (DMOAD) that targets multiple pathways has been of great interest for decades. While specific MMP inhibitors have been investigated for the treatment or prevention of diseases in multiple clinical trials, their success was limited by nonspecific inhibition, pharmacokinetics, toxicity, instability in vivo, and low oral availability [14]. Nonspecific MMP inhibitors developed as anti-arthritic drugs has led to serious side effects, such as musculoskeletal syndrome, leading to the cessation

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of clinical trials [14]. However, there has recently been a renewed call to investigate MMP inhibitors as tools to mitigate disease progression [14].

Due to renewed interest in MMP inhibition as a mechanism of inflammatory disease treatment, we decided to re-investigate tetracyclines as MMP inhibitors. Initially discovered as a product or actinomyces bacteria, derivatives of tetracycline have been used for decades for multiple infectious indications [15]. Doxycycline and minocycline are tetracyclines that are well studied for their efficaciousness in inhibiting inflammatory mediators that could contribute to the progression of arthritis. Recent reviews have found minocycline to be a promising drug in the treatment of chronic pain [16]. Doxycycline has been investigated in multiple trials for the treatment of osteoarthritis [17] and both doxycycline and minocycline have shown some clinical benefit for rheumatoid arthritis patients [18]. The purpose of this study is to review and synthesize the utility of tetracyclines as anti-inflammatory mediators in the treatment of osteoarthritis in pre-clinical and clinical studies. We hypothesized that tetracyclines have the potential to not only decrease symptoms of osteoarthritis but also inhibit the progression of osteoarthritis via multiple anti-inflammatory pathways.

Methods

A systematic review of the available literature was performed with the use of PRISMA (Preferred Reporting Items for Systematic Review and Meta-Analyses) guidelines and checklist (Fig. 1). The search was completed on April 18, 2020, using an explicit search shown in Table S-1 within PubMed and OVID MEDLINE databases. The search terms were selected to identify any study examining tetracyclines in relation to osteoarthritis. The primary outcome measure of interest was doxycycline or minocycline use in treating osteoarthritis with objective measures of efficacy.

English-language peer-reviewed studies were eligible for inclusion. Basic science, preclinical and clinical trials were included. Exclusion criteria were: Non-English-language studies, non-osteoarthritis-related studies, post-infectious arthritis studies, studies using implantable devices as the method of tetracycline administration, letters to the editor, conference abstracts/proceedings, studies which did not investigate doxycycline or minocycline, and other systematic reviews or meta-analyses. After exporting the citations from the databases and removing duplicate articles, the remaining articles were reviewed for inclusion by title followed by abstract. Manuscripts remaining underwent full-text review leading to the final manuscripts included in the study. Any questions on manuscript inclusion were reviewed by all authors before the final decision.

Studies were grouped into the following categories: basic science mechanistic studies, in vivo animal studies, and human clinical studies. Studies in the basic science category reported on the mechanisms by which tetracyclines may inhibit osteoarthritis progression. In vivo animal studies translated basic science findings into animal models of disease to show objective measures of benefit. Human clinical investigations included studies evaluating the clinical benefit of using doxycycline or minocycline for osteoarthritis in humans.

Results

A total of 1446 studies were reviewed, and after exclusion criteria were applied, 23 studies were included in the final analysis (Fig. 1). Twelve studies reported on basic science mechanisms [19–30] (Table 1), seven reported on outcomes of tetracycline treatment for osteoarthritis in animals [31–37] (Table 2), and four reported on human clinical studies [38–41] (Table 3).

Basic science mechanistic studies

Tetracyclines inhibit potential mediators of disease progression through inhibition of matrix metalloproteinases

The mechanisms by which doxycycline may alter osteoarthritis progression were identified through matrix metalloproteinase (MMP) inhibition. Yu et al. suggested that doxycycline modifies osteoarthritis progression by showing that degradation of human type XI collagen was inhibited in vitro by 10–30 μM doxycycline [20]. Smith et al. expanded upon the known capacity of doxycycline to inhibit MMP-1, 8 and 13 by characterizing the degree to which doxycycline was able to inhibit collagen breakdown by each enzyme in an acellular assay. At a concentration of 50 μM, doxycycline inhibited the activity of MMP-8 and MMP-13 by 64% and 77%, respectively, compared to only 18% for MMP-1 [22].

Shlopov et al. then expanded upon the potential mechanisms of MMP inhibition by doxycycline in two studies. In the first study, isolated human knee osteoarthritic chondrocytes were stimulated with TNF-α in the presence of doxycycline. Doxycycline inhibited collagen degradation through a reduction in MMP-1, MMP-8, and MMP-13 expression and translation in human chondrocytes [23]. The second study demonstrated that in addition to inhibiting translation of MMP-1 and MMP-13, in vitro treatment of human osteoarthritic chondrocytes with doxycycline significantly increased expression of TGF-β while decreasing translation of IL-1α, IL-1β, and IL-6 [24].

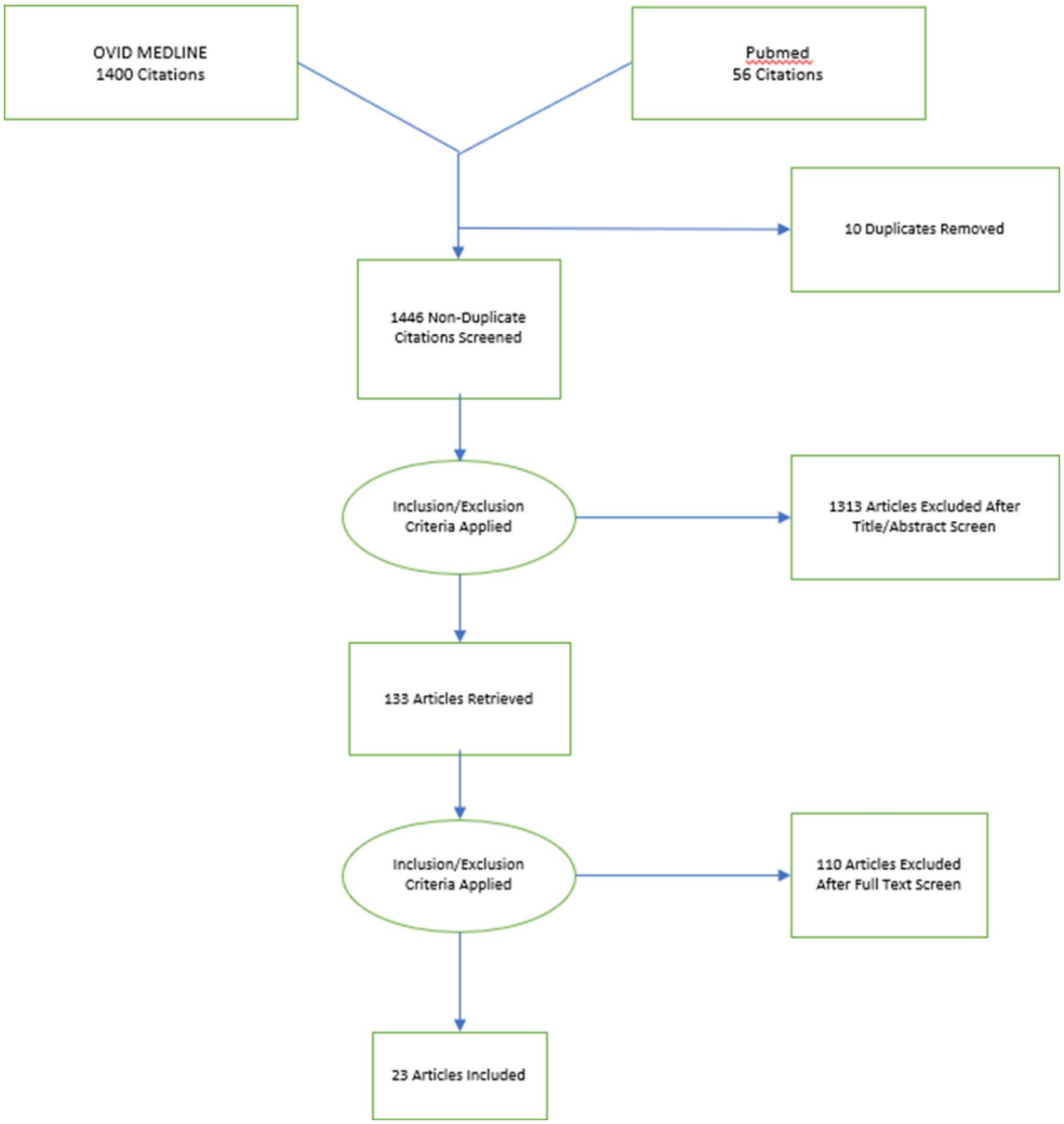

Fig. 1 PRISMA inclusion flowchart

A study from Smith et al. supported doxycycline's ability to inhibit MMPs by observing gelatinase and collagenase activity in the femoral heads of patients undergoing a total hip arthroplasty for osteoarthritis. Patients were either treated with oral doxycycline for 5 days before surgery, in a single dose 3 days before surgery, or with no doxycycline. The femoral head samples from the patients treated with 5 days of doxycycline demonstrated significantly reduced activities of both collagenase and gelatinase [21].

Like doxycycline, minocycline also inhibits MMPs and may be utilized to disrupt degradative pathways. An early trial showed significantly decreased collagenase activity in the synovial fluid of rheumatoid arthritis patients after a 10-day treatment with oral minocycline [19]. Vandooren et al. investigated minocycline inhibition of MMP-9 production and activity. Minocycline was first shown to be a weak inhibitor of MMP-9 in vitro by incubating minocycline with human recombinant MMP-9 and assessing for gelatin degradation. Secondly, minocycline was demonstrated to inhibit the secretion of pro-MMP-9 by myelomonocytic (THP-1) cells. THP-1 cells, which synthesize pro-MMP-9 upon stimulation, were incubated with and without minocycline and stimulated with LPS. Minocycline significantly reduced the secretion of pro-MMP-9 without cytotoxicity [25].

Table 1 Basic science mechanistic studies

References	Drug; concentration/dose	Cell type	Targets	Experimental strategy	Findings
MMP-inhibition					
Greenwald et al. 1987 [19]	Minocycline; 100 mg bid	Synovial cells	Collagenase	Ex vivo enzymatic assay of arthritic knee	Inhibited collagenase activity synovial fluid of RA patients
Yu et al. 1991 [20]	Doxycycline; 10–30 µM	Chondrocytes	Collagenases	Cell culture	Inhibited type XI collagen degradation
Smith et al. 1998 [21]	Doxycycline; 100–200 mg qd	Chondrocytes	Gelatinase, collagenase	Ex vivo assay of arthritic knee	Inhibited gelatinase and collagenase activity
Smith et al. 1999 [22]	Doxycycline; 30–90 µM	–	MMP-1, MMP-8, MMP-13	Acellular enzymatic assay	Inhibited MMP-8 and MMP-13 activity against TII collagen in vitro
Shlopov et al. 1999 [23]	Doxycycline; 1–10 µg/mL	Chondrocytes	MMP-1, MMP-8, MMP-13	Cell culture	Decreased protein and mRNA levels of all MMPs
Shlopov et al. 2001 [24][a]	Doxycycline; 22.5 µM	Chondrocytes	MMP-1, MMP-13	Cell culture	Inhibited translation of MMP-1 and 13
Vandooren et al. 2017 [25]	Minocycline; 2.2–218.6 µM	THP-1 cells	MMP-9	Cell culture	Inhibited pro-MMP-9 secretion and inhibited MMP-9 activity
Anti-inflammatory					
Kloppenburg et al. 1993 [26]	Minocycline; 109.3 µM	T-cells	IFN-y	Cell culture	Inhibited proliferation of T-cells and IFN-y production in vitro
Kloppenburg et al. 1996 [27]	Minocycline; 3.5–109.3 µM	T-cells	IFN-y, TNF-alpha, IL-6	Cell culture	Inhibited IFN-y, TNF-alpha, and IL-6 production by stimulated T-cells
Shlopov et al. 2001 [24][a]	Doxycycline; 22.5 µM	Chondrocytes	MMP-1, MMP-13	Cell culture	Inhibited translation of IL-1, and IL-6 Upregulated TGF-beta
Nitric oxide pathways					
Amin et al. 1996 [28]	Doxycycline and minocycline; 11.3–180 µM, 10.9–174.9 µM	Chondrocytes, macrophages	Nitric oxide synthase	Cell culture	Inhibited nitric oxide production
Borderie et al. 2001 [29]	Doxycycline and minocycline; 1–100 µM	Chondrocytes	Nitric oxide synthase	Cell culture	Inhibited production of nitrosothiols, nitrate, nitrate, and iNOS
Steinmeyer et al. 2010 [30]	Tetracycline, doxycycline, and minocycline; 1–100 µM	Chondrocytes	Aggrecanases ADAMSTS4 and ADAMSTS5	Cell culture	Inhibited aggrecanases and decreased PGE2 and nitric oxide while not decreasing proteoglycan loss

[a]Included in two categories

Tetracyclines have anti-inflammatory properties that may be beneficial for osteoarthritis

Tetracyclines offer an additional mechanism to inhibit osteoarthritis progression through broad anti-inflammatory properties. Doxycycline decreases translation of inflammatory cytokines and increases the production of anti-inflammatory cytokines [24]; however, minocycline is more extensively studied for its anti-inflammatory action in arthritis. Two studies from Kloppenburg et al. demonstrated minocycline's anti-inflammatory potential. In the first, minocycline inhibited the proliferation of cloned synovial T-cells and IFN-γ production by synovial T-cells in vitro [26]. In tandem, minocycline inhibited the production of IFN-γ and TNF-α by activated T-cells in vitro [27]. By limiting T-cell activation, the authors propose that minocycline could reduce the innate immunity reaction in osteoarthritis progression.

Table 2 In vivo animal studies

References	Drug; dose	Organism	Primary experimental outcome	Findings
Idiopathic osteoarthritis models				
Lee et al. 2013 [31]	Doxycycline; 2 mg/mL in drinking water	Rat	Histopathologic examination	MMP-13 deposition decreased in rat osteoarthritic knees
Lu et al. 2013 [32]	Doxycycline hyaluronic acid gels; 17.5 µg	Rabbit	Weight distribution, histopathologic examination	Weight distribution in the affected leg increased. There was histologically less evidence of OA
Aydin et al. 2015 [33]	Doxycycline-chondroitin sulfate microspheres; 5 mg intraarticular injection	Rabbit	Histopathologic examination, radiographic assessment, and mechanical assessment	Treatment was superior radiographically and histologically to control
Posttraumatic arthritis models				
Yu et al. 1992 [34]	Prophylactic doxycycline; 50 mg bid	Canine	Histopathological examination	Significantly less cartilage wear in treatment group
Pardy et al. 2004 [35]	Doxycycline; 50 mg bid	Canine	Histopathologic examination	Subchondral bone loss decreased and bone strain energy density increased
Dinc et al. 2012 [36]	Doxycycline; 1 mg/mL/kg of 50% by volume intraarticular injection	Rabbit	Histopathologic examination	Was inferior to atorvastatin and not significantly different compared to saline in preventing cartilage wear
Pinney et al. 2012 [37]	Doxycycline; 2.2 mg/kg	Rabbit	Histopathologic examination and radiographic assessment	Did not significantly affect the progression of osteoarthritis

Table 3 Human clinical trials

References	Design	No. of groups	Level of evidence	Drug; dose	Primary outcome	Findings
Israel et al. 1998 [38]	Prospective	1	IV	Doxycycline; 50 mg bid	Enzymatic activity in the TMJ	Trend toward decreased collagenase and gelatinase activity in 3 women with TMJ osteoarthritis
Brandt et al. 2005 [39]	Prospective	2	I	Doxycycline; 100 mg bid	Joint space narrowing	Significantly decreased JSN in obese women with unilateral knee osteoarthritis
Snijders et al. 2011 [40]	Prospective	2	I	Doxycycline; 100 mg bid	Clinical response by OMERACT-OARSI criteria	31% in both the treatment group and the control group met responder criteria
Ma et al. 2015 [41]	Retrospective	1	IV	Doxycycline; 100 mg bid	Verbal descriptor scale of pain and functional status	Pain improved or resolved in all patients

The capacity of minocycline to inhibit destructive immune activity as well as overactive metalloproteinase enzymes has been supported in human rheumatoid arthritis studies which demonstrate significant clinical benefit [42–46]. For example in the Minocycline in Rheumatoid Arthritis (MIRA) trial, a group of rheumatoid arthritis patients who received minocycline showed significant improvements in both subjective measures, such as joint tenderness and swelling, and objective measures, such as erythrocyte sedimentation rate and hematocrit, over placebo [42]. Similar results were reproduced in multiple double-blinded clinical trials of minocycline vs. placebo in rheumatoid arthritis patients [43–46].

Tetracyclines inhibit nitric oxide pathways

Tetracyclines may also exert disease modifying effects by limiting nitric oxide pathways in chondrocytes. Amin et al. isolated osteoarthritic cartilage from human total knee arthroplasty patients and assayed for the spontaneous activity of nitric oxide synthase in the presence of doxycycline and minocycline [28]. The results showed dose-dependent

inhibition of nitric oxide synthase (NOS) by both tetracyclines. Minocycline and doxycycline inhibited nitric oxide synthesis by murine macrophages stimulated by IFN-γ or LPS in vitro [28]. Further study into the nitric oxide synthase inhibition in stimulated human osteoarthritic synovial cells by Borderie et al. showed that both doxycycline and minocycline inhibited the production of nitrosothiols, nitrate, and nitrite in addition to inhibiting the synthesis of the inducible NOS [29].

Steinmeyer et al. used human osteoarthritic knee explants to assess the effect of minocycline and doxycycline on aggrecanase activity as well as the amount of nitric oxide and prostaglandin E2 (PGE2) produced by stimulated osteoarthritic knee cartilage. This group also performed a direct assessment of proteoglycan loss in explanted osteoarthritic cartilage upon stimulation with IL-1. Although doxycycline and minocycline were found to inhibit aggrecanases ADAMTS4 and ADAMTS5 as well as decrease the production of PGE2 and nitric oxide, the amount of proteoglycan loss was not significantly decreased by any of the drugs over 11 days of treatment in vitro [30].

In vivo animal studies

In animal studies, doxycycline has shown promise in slowing the progression of osteoarthritis. Medial tibial osteochondral defects were surgically created in rats, and then rats were fed untreated water or water with 2 mg/mL doxycycline for 12-weeks postoperatively [31]. At 12-weeks the rats were euthanized and MMP-13 deposition was assessed in the repair tissues of each group. MMP-13 deposition was significantly lower in the group that received doxycycline. In addition, the International Cartilage Repair Society cartilage repair scores were significantly higher in the group treated with doxycycline indicating a higher degree of cartilage repair [31].

Intraarticular injections of doxycycline were also explored in animal osteoarthritis models. A hyaluronic-acid-doxycycline hydrogel was injected and evaluated in a rabbit surgical knee osteoarthritis model. Rabbits were treated with injections of normal saline, hyaluronic acid, doxycycline, or combined hyaluronic acid-doxycycline. Those treated with hyaluronic acid and hyaluronic acid-doxycycline groups demonstrated decreased pain as determined by about 10% more bodyweight being distributed on the affected leg. On histopathological examination, osteoarthritic progression was significantly decreased in the hyaluronic acid-doxycycline group [32].

In a 2015 study, Aydin et al. used injectable microspheres as the method of drug delivery into osteoarthritic rabbit knee joints [33]. Microspheres contained either doxycycline or doxycycline-chondroitin sulfate. After 8 weeks of treatment, rabbit knees were radiographically superior to control in both doxycycline and doxycycline-chondroitin sulfate groups. The Mankin-Ptizker histological scores of both experimental arms were significantly better compared to control at the end of treatment. The ex vivo hardness scores of the osteoarthritic cartilage showed superior values in the doxycycline-chondroitin sulfate group [33].

Four studies [34–37] evaluated the effect of doxycycline in a posttraumatic model. While posttraumatic arthritis represents only a fraction of osteoarthritis cases, it is a significant cause of pain for patients and loss of function post-injury, with rates of development suggested to be between 21 and 48% after combined anterior cruciate ligament and meniscus injury, even after surgical treatment [47]. Furthermore, osteoarthritis can commonly result from meniscus injuries [47], an injury that is often found incidentally in the aging population [48]. Posttraumatic arthritis models have shown less consistent results regarding doxycycline treatment. Pinney et al. investigated doxycycline treatment in a rabbit model of posttraumatic arthritis [37]. Rabbits were treated with and without the addition of doxycycline to their food for 12 weeks following ACL transection. The rabbits underwent MRI evaluation of the knee every four weeks until euthanasia at 12 weeks. This study did not show conclusive evidence of doxycycline's efficacy in the prevention of posttraumatic arthritis [37]. In another posttraumatic arthritis rabbit model, Dinc et al. transected rabbit ACLs and treated rabbits with either intra-articular saline, doxycycline, or atorvastatin injections once per week for 3 weeks [36]. After 3 weeks, the cartilage wear was scored by the Mankin histological scoring system. Cartilage scores were similar in the control and doxycycline groups, but the atorvastatin group showed significantly less cartilage wear than the control and doxycycline groups [36].

Several animal studies support the potential role of doxycycline for treating posttraumatic osteoarthritis development. In a canine ACL deficient posttraumatic osteoarthritis model, dogs were treated with or without oral doxycycline for 8 weeks immediately following ACL transection. On histopathological examination, doxycycline-treated dogs demonstrated significantly less cartilage wear of the medial femoral condyle compared to untreated controls [34]. In a study from Pardy et al., researchers surgically transected the ACL and the dogs were split into three groups. One group was treated with 24 weeks of oral doxycycline starting 12 weeks after surgery. A second group was treated with 36 weeks of doxycycline starting 36 weeks after surgery. The final group was not treated with doxycycline. In the group treated with 36-weeks of doxycycline, superficial subchondral bone loss was significantly mitigated compared to control and bone strain energy density was increased, though this difference was only 0.01 mJ/mm^3 [35].

Clinical trials

The human studies included in this review consisted of two level IV and two level I studies. The level IV studies had a high risk of bias and inaccurate conclusions due to small sample sizes (5 patients and 17 patients), lack of blinding, and the observational nature of the studies [38, 41]. In contrast, the level 1 studies included are of a particularly high quality due to large sample sizes, meticulous blinding, and prospective definition of outcomes of interest [39, 40].

The two observational cohort studies demonstrated biochemical and patient-reported improvements in patients treated with doxycycline in various joints. Initially, Israel et al. showed a trend toward decreased collagenase and gelatinase activity in osteoarthritic temporomandibular joints of three women treated with doxycycline for three months [38]. In another observational study, Ma et al. performed a retrospective pilot study on 17 patients treated with doxycycline for osteoarthritis of the hand [41]. Clinical response was measured in these patients via a 1–10 verbal descriptor scale of pain as well as a 1–6 scale of the ability to perform tasks related to hand function. The results of this investigation found that all 17 patients had improvements in pain, with most (11/17) having their pain completed resolved. Furthermore, most patients also reported regaining normal functioning of their affected hand, however, this study was underpowered to identify significant improvements and lacked a comparison or control group [41].

The two randomized clinical trials investigated doxycycline treatment for osteoarthritis of the knee with varying results. In the DOXY trial, Brandt et al. treated 431 obese women with primary unilateral knee osteoarthritis with doxycycline 100 mg twice daily or placebo. Radiographic studies were performed at 16 month and 30-month follow-ups. The primary outcome measure was the rate of joint space narrowing (JSN) of the medial tibiofemoral compartment of the arthritic knee by measuring joint space width of the involved knee and the uninvolved knee. Other measures included clinical outcome measures such as the Western Ontario and McMaster Universities Osteoarthritis Index (WOMAC), knee pain after 50-foot walk as measured by the visual analogue scale (VAS), and the global disease activity on a 5-point Likert scale.

The results showed significant decreases in the rate of JSN in the group randomized to doxycycline. While overall clinical outcome scores were not significantly different between the treatment and placebo groups, the frequency of a clinically significant increase in pain scores according to the WOMAC and the VAS were significantly higher in the placebo group. The rate of JSN was also found to significantly correlate with increases in 50-foot walk pain [39].

The second large clinical trial evaluated the effectiveness of doxycycline against placebo with the primary endpoint being clinical pain response. Patients with knee osteoarthritis were treated for 24 weeks and evaluated for clinical responsiveness via the Outcome Measures in Rheumatology Clinical Trials-Osteoarthritis Research Society International (OMERACT–OARSI) criteria, which uses the scales of WOMAC pain, WOMAC function, and VAS-PGA to define clinical effectiveness in a binary fashion. There were 232 total participants in the study with 116 patients in each group. Similar to Brandt et al. [39], the number of responders were similar between placebo and treatment groups according to the OMERACT-OARSI criteria. In addition, there was no significant difference between baseline and week 12 or week 24 scores of WOMAC pain, WOMAC stiffness, VAS-PGA, PCS, and MCS in either group. Interestingly, significantly more patients dropped out of the doxycycline group due to medication side effects, although the adverse events were mild [sun sensitivity (30%) and diarrhea (13%)] [40].

Discussion

The principal findings in the present study are that doxycycline and minocycline limit cartilage degrading enzyme activity in laboratory and animal studies, and the available clinical data offer encouragement for the drugs disease-modifying agents. The basic science studies demonstrated a range of mechanisms by which tetracyclines stem the progression of osteoarthritis including MMP inhibition, anti-inflammatory properties, and nitric oxide pathway inhibition. Such actions may be promising avenues for osteoarthritis treatment, as complex inflammatory pathways have been shown to be responsible for the development of osteoarthritis [13]. In addition to these mechanisms, tetracyclines have demonstrated even more disparate effects that may benefit the osteoarthritic population outside the joint capsule. Our findings overall supported our hypothesis; however, our review identifies the need for further evaluation of the optimal drug delivery method and patient population likely to derive substantial benefit.

The in vivo animal studies consisted of varied results; however, tetracyclines did limit osteoarthritis progression in several studies [31, 33–35]. The difficulty in interpreting the rabbit studies not showing doxycycline efficacy lies in the route of administration and the time points analyzed. These data suggest the option to target inflammation and cartilage degradation immediately following knee injury. Pro-inflammatory responses are present in the knee following both ACL and meniscus injury and lead to increases in pro-inflammatory cytokines, chemokines and matrix-degrading enzymes [49–51]. This pro-inflammatory environment is persistent after injury, so treatment may be more efficacious in altering the posttraumatic osteoarthritis disease process.

As shown in the differing results between the two large randomized clinical trials examining doxycycline in treating osteoarthritis, while doxycycline significantly decreased joint space narrowing over the course of treatment [39], it did not benefit patients with respect to symptoms [40]. While the conclusions differ, the results are not necessarily mutually exclusive. In a 2011 editorial, Brandt argued that cartilage is aneural; therefore, a decrease in pain would not necessarily be expected to occur due to a decrease in cartilage loss [52]. It is possible that while doxycycline does not have significant pain benefit over 24 weeks, it does act as a chondroprotective agent, and may prevent the progression of osteoarthritis over a longer term and have significant benefit for patients early in the disease process.

Along the same lines, a major concern with the doxycycline idiopathic osteoarthritis trial is whether the patient population is the one most likely to benefit from inhibition of enzymatic cartilage degradation. It is possible that a more effective treatment would be administered earlier in the disease, even prior to significant radiographic osteoarthritis development. Magnetic resonance imaging findings of early osteoarthritis are present in both the cartilage and the meniscus prior to radiographic changes [53–55]. Tetracycline administration at this early disease timepoint may be more beneficial due to actual disease modification.

As in the pathogenesis of osteoarthritis, secondary inflammation plays a significant role in a variety of conditions. Tetracyclines have already been used for anti-inflammatory purposes aside from the aforementioned use of minocycline in rheumatoid arthritis [42–46]. For example, minocycline has shown promise in reducing secondary inflammation in the setting of spinal cord injury [56]. Shultz et al. compiled a wide array of anti-inflammatory mechanisms of minocycline that may be of benefit in preventing secondary injury, including scavenging free radicals and inhibiting nitric oxide production [56].

Such central nervous system inflammation has been suggested to play a significant role in chronic osteoarthritis pain in a rat model of osteoarthritis. Sagar et al. showed that osteoarthritic pain behaviors were significantly correlated with activation of spinal astrocytes and glial cells. In the same study, minocycline was shown to inhibit microglial and astrocyte cell activation and attenuate chronic pain behaviors in the rat model of osteoarthritis [57]. This finding demonstrates the potential for tetracyclines to decrease pain even as their chondroprotective mechanism does not afford a pain benefit [52].

Aside from being chondroprotective and having inhibitory effects on potential pain pathways, tetracyclines may have further benefits for osteoarthritis patients. The neuroinflammation associated with chronic osteoarthritis pain highlighted above may also play a role in depressive symptoms [58–60]. Depression is a common comorbidity of osteoarthritis and posttraumatic osteoarthritis patients. A recent meta-analysis of four randomized clinical trials of minocycline treatment for depression found minocycline to be superior to placebo in cases of unipolar depression [61].

In addition, minocycline has been suggested in an in vitro animal study to have the potential to alleviate depression via its effects on the gut microbiome [62]. Rats bred for high anxiety behavior were treated with minocycline. The high-anxiety rats had an altered gut microbiome as well as fewer microglia in their prefrontal cortex. Minocycline increased microglia in the prefrontal cortex, reverted the gut microbiome to a more normal environment, and reduced anxiety-like behavior compared to escitalopram alone [62]. Therefore, the broad anti-inflammatory properties of tetracyclines may benefit osteoarthritis patients in ways not yet explored. Further, the potential mood effects may benefit the patient's functionality and perceived pain even while the chondroprotective effects early in the disease process would not mechanically show function or pain differences.

The matrix-metalloproteinase inhibition by tetracyclines has been suggested to be of benefit for coronary artery disease, which is a common comorbidity of osteoarthritis patients. A large clinical trial demonstrated significant benefit with reductions in infarct size, severity, and left ventricular end-diastolic index in patients taking 1 week of doxycycline after percutaneous intervention in the setting of anterior ST elevation myocardial infarction [63]. An ongoing clinical trial is investigating doxycycline's effectiveness in inhibiting maladaptive heart muscle remodeling following a heart attack (NCT03508232). In the chronic timeframe, MMPs play a role in vascular remodeling and plaque rupture that may lead to a coronary event. Tetracyclines may clinically benefit patients in preventing harmful MMP activity in this setting as well [64].

While tetracyclines may allow for a wide variety of benefits, the risks of tetracycline treatment must be taken into account. As seen in both clinical trials, side effects are relatively common and often include sun sensitivity or gastrointestinal side effects [39, 40]. The side effect profile of tetracyclines, and, more generally, MMP-inhibitors, has led to the limited translation of promising animal study results to human trials [14]. In addition, tetracycline have been shown to increase the prevalence of *Clostridium* and *Bacteroides* families, suggesting an accumulation of resistance genes [65]. While the clinical effects of such a microbiome disturbance are not fully understood, changes in the bacterial community may be immunomodulatory, as *Bacteroides* lipopolysaccharide (LPS) has been shown to neutralize host immune response to E. coli LPS [66]. The result of such an interaction has not been clarified in humans and may be variable depending on the individual patient's microbiome [66]. For example, in high anxiety behavior mice, minocycline was shown to make microbiome changes that may have

ultimately been beneficial for decreasing anxiety symptoms [62].

While tetracyclines offer mechanisms that may benefit osteoarthritis patients, perhaps specific MMP inhibition may offer a more fruitful avenue of treatment. While specific MMP inhibitors have not yet been clinically investigated in the context of osteoarthritis, promising results have been displayed in rheumatoid arthritis with the anti-MMP-9 antibody GS-5745/andecaliximab with minimal side effects [67]. An investigation into specific MMP inhibition in osteoarthritis, particularly inhibition of MMP-8 and/or MMP-13, may provide a DMOAD with minimal side effects.

Our study carries common limitations of systematic reviews. The study designs and resultant data are heterogenous and limit any advanced analysis for laboratory or clinical efficacy. While the limited number of clinical trials permit only a critical review of those studies, we did identify potential benefit in treating the inflammatory aspect of posttraumatic osteoarthritis. The consolidated mechanisms of action in pre-clinical studies support a rationale for further investigation in earlier disease intervention and potential efficacy in altering the progression of posttraumatic osteoarthritis.

Conclusion

Doxycycline and minocycline have a broad spectrum of potential benefits for osteoarthritis patients. Animal studies and human trials have reported both positive and neutral effects on osteoarthritis and posttraumatic arthritis progression. Further study is warranted to examine the benefit of doxycycline early in the osteoarthritic process in stemming progression and secondarily benefiting common comorbidities.

Author contributions CJ, CC, and AS had the original idea for the manuscript. BP performed the literature search and drafted the manuscript. BP, CC, CJ, and AS critically revised the work.

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