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Efficacy of Turmeric for Treatment of Osteoarthritis: A Systematic Review and Meta-Analysis of Experimental Animal Model Studies

Stacey Sheahan VMD, MS-TCVM

ABSTRACT

The objective of this systematic review was to determine the efficacy of using turmeric (*Curcuma longa*) in veterinary patients for control of osteoarthritis (OA). The literature search was conducted using three electronic databases and a university library. Keywords searched included "turmeric", "curcumin" and "osteoarthritis". There were 24 experimental model studies in laboratory species that used turmeric as a treatment for OA and met inclusion criteria. These studies compared a turmeric/curcumin treated group to placebo (or no treatment) control group. The meta-analysis based on the reported statistical significance (*p*-values) concluded an overall *p*-value of 3.0×10^{-28}, which demonstrated turmeric/curcumin was highly effective in improving OA in these studies for the parameters measured. Meta-analysis using random-effects model on the four most reported outcome measurements (TNF-α, IL-1β, NF-kβ, MMG) also concluded effectiveness of turmeric/curcumin in improving OA conditions (all *p*-values < 0.001). The positive results in animal model studies from this systematic review and meta-analysis suggest that turmeric/curcumin may be of benefit for companion animals with OA, and supports further investigation of its use as part of food therapy and/or herbal formulations with randomized controlled clinical studies.

Keywords: canine osteoarthritis, traditional Chinese medicine, turmeric, curcumin, meta-analysis, Chinese herbal medicine, cytokines, food therapy

*Address correspondence to Dr. Stacey Sheahan (dr.staceygilles@gmail.com).

ABBREVIATIONS

ACL	Anterior cruciate ligament
Bax	Bcl-2 associated X protein
CAT	Catalase
COAST	Canine Osteoarthritis Staging Tool
COX-1	Cyclooxygenase-1
COX-2	Cyclooxygenase-2
IL	Interleukin
IL-1β	Interleukin-1beta
IL-4	Interleukin-4
IL-6	Interleukin-6
IL-10	Interleukin-10
IL-13	Interleukin-13
MCP-1	Monocyte-chemotactic protein-1
MDA	Malondialdehyde
MIA	Monosodium iodacetate
MMG	Modified Mankins Grading
MMP	Matrix metalloproteinases
MMP-13	Matrix metalloproteinase-13
NF-kB	Nuclear factor-kappa B
NSAIDs	Non-steroidal anti-inflammatory drugs
OA	Osteoarthritis
OARSI	Osteoarthritis Research Society International
PGE_2	Prostaglandin E_2
TCM	Traditional Chinese medicine
TCVM	Traditional Chinese veterinary medicine
TEO	Turmeric essential oil
TNF-α	Tumor necrosis factor-alpha

From: Animal Medical Center of Somerset County, Somerset, Pennsylvania USA

Professional Certifications: CTPEP, CVA, CVCH, CVFT, CVTP

Osteoarthritis (OA) is an inflammatory, degenerative process that leads to progressive loss of articular cartilage, damage to subchondral bone, synovitis and is a potentially irreversible disease.[1,2] In human patients, companion animal species and laboratory species, OA is characterized by inflammation, pain and decreased mobility. As severity and chronicity escalates, poor quality of life also occurs. In dogs, OA affects 20% of animals older than 1 year and 80% of dogs over age 8.[3,4]

There are numerous and varied etiologies associated with development of the disease. These may include but are not limited to genetic predisposition, aging, obesity, trauma, infectious and systemic diseases.[1,4-6]Although there are diverse causes with overlapping factors, processes occur over time at the tissue and cellular level

that gradually take on a similar nature and result in the typical phenotypic image of OA (Figure 1).[2]

The participation of the immune system in the pathogenesis of the disease is one of the key elements. Current research continues to focus attention on the role of cytokines that impact the severity and duration of the disease and factors that turn them off and on [e.g. NF-kB (nuclear factor kappa β)].[2] Cytokines play a key role influencing catabolic and anabolic processes in joint tissue that is under high mechanical load.[2] As an imbalance occurs between these two processes, normal joint homeostasis changes from healthy to an increase in inflammation and degradation that lead to gradual loss of joint function and pain. Cytokines can be divided into inflammatory types, such as tumor necrosis factor (TNF-α), interleukin-1 beta (IL-1β) and interleukin-6 (IL-6); or anti-inflammatory types, such as IL-4, IL-10, and IL-13.[2] Both IL-1β and TNF-α are considered 2 of the most important OA associated inflammatory cytokines. Increased concentrations of these are found in OA affected synovial fluid, synovial membranes, cartilage and the subchondral bone layer. They are responsible for marked catabolic effects with subsequent destruction of articular cartilage. The anti-inflammatory cytokines mainly involve inhibiting the synthesis of inflammatory cytokines, particularly IL-1β and TNF-α. Observed effects include increased proteoglycan synthesis, inhibition of apoptosis of chondrocytes, decreased synthesis/secretion of the destructive metalloproteinases (MMP), and decreased levels of prostaglandin E_2 (PGE2) and cyclooxygenase-2 (COX-2).[2,7]

Current pharmaceutical treatment options for the management of OA include analgesics, steroids and non-steroidal anti-inflammatories (NSAIDs) which reduce inflammation and pain. Their long-term use, however, sometimes can't be sustained due to inadequate pain relief, immune disturbances (steroids), liver toxicity and serious gastrointestinal events.[8] Today's health-conscious society has an increased interest in herbal and natural remedies for treating diseases. The use of Chinese herbal medicine for diseases including osteoarthritis has been common in China for centuries. While the biological and molecular impact of herbal medicine is not well understood, positive clinical treatment outcomes have led to further use in veterinary medicine.[6]

Turmeric (common name for *Curcuma longa*), is an Indian spice that has been used in Ayurvedic and traditional Chinese medicine (TCM) to treat inflammatory processes for centuries. Turmeric belongs to the ginger family (Zingiberacceae). Much of its pharmacological activity has been mainly attributed to the curcuminoids which are primarily curcumin (77%), followed by desmethoxycurcumin (17%) and bisdemethoxycurcumin (3%).[9] Traditional Chinese medicine uses the rhizomes of turmeric with its golden yellow color, called *Jiang Huang*. The taste and energetics are warm, pungent and bitter with a fragrant smell. It invigorates Blood, activates *Qi*, reduces swelling and treats Wind-Damp obstruction. The channels *Jiang Huang* influences are the Spleen, Stomach and Liver. Contraindications include a caution when used during pregnancy and caution in Blood Deficiency without Blood Stagnation. When used as an alternative medicine or dietary supplement, turmeric is typically used as an extract that is standardized to 80-95% curcuminoids, primarily curcumin.[8] No oral administration side effects have been reported and recommended species doses include: horses and cattle (25-45 grams (g); llamas, alpacas, goats, sheep and pigs (5-10 g); dogs (1-3 g); cats and rabbits (0.2-0.5 g); birds (0.1-1 g).[10]

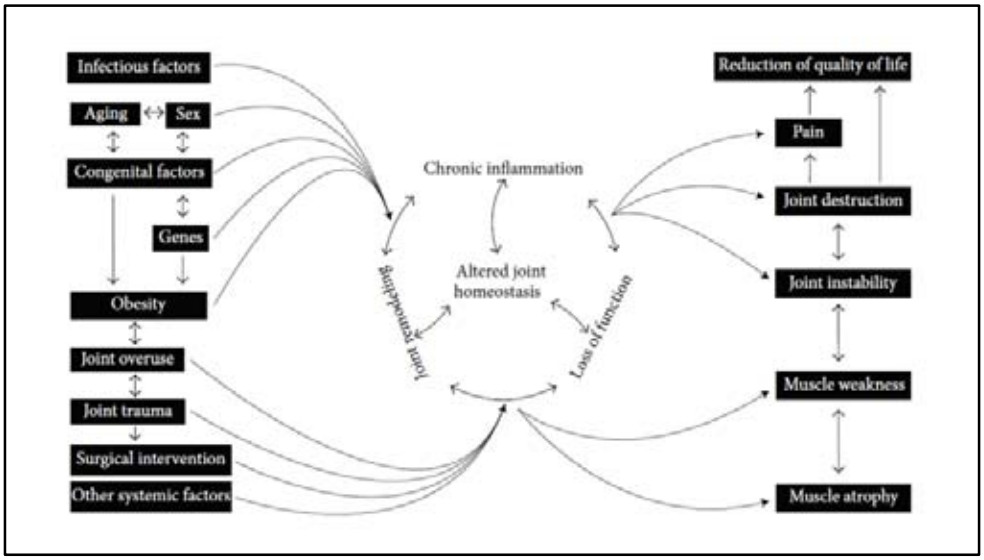

Figure 1: Schematic of a closed disease circle, comprising the disease progression of osteoarthritis, taking into account its causes and consequences. Adapted from Wojdasiewicz P et al. The role of inflammatory and anti-inflammatory cytokines in the pathogenesis of osteoarthritis.[2]

Turmeric and its major constituent, curcumin, have been widely studied in human medicine, laboratory animals and companion animals with the goal of understanding its diverse beneficial properties. Unlike ginger, turmeric does not modulate cyclooxygenase-1 (COX-1) activity but does modify NF-κβ signaling, inflammatory cytokines and COX-2 activity.[8] A remarkable list of medicinal effects have been documented such as beneficial use in arthritis, neurodegenerative diseases, allergies, cancer, inflammatory bowel disease, nephrotoxicity, diabetes and cardiovascular disease. The diversity of diseases it treats relates to its range of effects that include anti-inflammatory, antioxidant, antimicrobial, anti-proliferative, and anti-atherosclerotic.[5]

Several systematic review studies regarding the use of turmeric and curcumin in human OA studies have been reported.[8,11-14] A review of 15 human clinical studies concluded turmeric/curcumin demonstrated potential for treatment of osteoarthritis.[6] A meta-analysis of 8 controlled randomized human clinical trials found there was compelling evidence for its use as a dietary adjunct to conventional therapy, although, there was an insufficient number of subjects to permit definitive recommendations.[9] Henrotin et al. reviewed 86 human clinical trials and concluded that "curcumin represents a new paradigm since it is not yet a recommended intervention in OA but should be considered based on its safety and efficacy."[15]

The purpose of this systematic review and meta-analysis was to review literature evidence of turmeric and/or curcuminoid extracts efficacy to provide beneficial treatment of inflammatory and degenerative changes present in osteoarthritis. It was hypothesized that the body of research studies would support turmeric's potential to treat OA with a statistically significant meta-analysis. This could then support conducting clinical trials in companion animals that at present are virtually non-existent.

MATERIALS AND METHODS

Search engine tools Google Scholar, PubMed, Science Direct and literature within the Chi University Library (Reddick, Florida, USA) were accessed using the keywords turmeric, curcumin and osteoarthritis. The articles listed under the search engine web pages were screened by the author and independent reviewer with selection based on review of the titles. The literature search included peer-reviewed articles reporting osteoarthritis, turmeric, curcumin in laboratory species, small animal companion species and human patients. The studies could be retrospective or prospective controlled studies that included turmeric/curcumin as part of the OA treatment. Turmeric/curcumin use in the studies could be as a single agent (compared to a control group/placebo or conventional medicine). It could also be in combination with conventional treatment (compared to a conventional medicine control group). Veterinary research studies should use lameness scoring, pain scoring, or ground forces to determine treatment efficacy and listed with statistical evidence of outcome. Studies selected were from the years 2000 to 2020 and language restrictions were based on full article translations (English) being available. Exclusion criteria included review articles, systematic review/meta-analyses, studies lacking control groups or studies containing a mixture of ingredients where turmeric/curcumin could not be isolated from other ingredients to investigate its effects only. All articles selected from the initial search underwent assessment based on the inclusion/exclusion criteria, with ultimately all studies that fit the criteria included in the statistical meta-analysis.

For each of the articles included in the meta-analysis, the quality of the study was assessed based on the Jadad Scale.[16-17] The scale, ranging from 0 to 5, is widely used in systematic review studies for assessing the quality of randomized controlled trials. The assessment is based on the following three criteria: (1) randomization: score 1 if mentioned, score 2 if method described; (2) blinding: score 1 if mentioned, score 2 if method described; and (3) data outcome: score 1 if the fate of all subjects in the trial is known (e.g. complete the trial, drop-out, excluded due to protocol deviation).

The statistical meta-analyses included two approaches. The first employed Stouffer's Z-score analysis, which combines statistical significance (*p*-values) of all qualified studies to derive an overall *p*-value to test the hypothesis. The second approach applied the random effects model on studies that reported a common outcome measurement. For either approach, an overall *p*-value less than 0.05 would support the hypothesis that laboratory species, companion animals and humans can benefit from the use of turmeric/curcumin in treating symptoms of osteoarthritis. For studies that only reported open intervals for *p*-values such as "$p < 0.01$" or "$p > 0.05$", a conservative approach was applied to determine exact *p*-values for the meta-analysis. For instance, when a study reported a *p*-value < 0.05, 0.05 was used, and when a study reported a *p*-value > 0.05, 0.99 was used in the meta-analysis. This approach helped to ensure that the statistical significance of each study was not falsely overestimated. All graphical presentation and statistical analysis were performed using commercial software[a].

RESULTS

An initial search and screening based on the keywords and search engines defined in the protocol were performed on 270 articles. A total of 77 articles were selected by the investigator and an independent reviewer for detailed review and assessment for study inclusion. The assessment resulted in 26 articles that met the inclusion criteria: 24 experimental animal model studies, 1 human clinical study, and 1 veterinary clinical study.[14,18-41] The meta-analysis was performed only on experimental model studies due to the limited number of human and veterinary studies (Table 1).

Table 1: The 24 experimental animal model studies that were included in the systematic review and meta-analysis along with each study's Z-score.

Reference	Experimental Treatment	Z-score Turmeric Efficacy*
Aborehab (2017)[18]	Chondroprotective effect of ginger/turmeric treatment for MIA-induced OA rat model; 4 groups (n=10/group), ginger/turmeric treatment (3 experimental groups) vs. untreated controls	-3.89
da Silva Campos (2017)[19]	Synthesis and characterization of gold nanoparticles combined with curcumin, effects on experimental DMM-induced OA in mice; 4 groups (n=8/group), curcumin and/or gold nanoparticles (3 dose groups) vs. untreated controls	-3.89
El-senosi (2017)[20]	Biochemical studies on the effect of curcumin in experimentally induced MIA-induced osteoarthritis in rats; 4 groups (n=9/group), curcumin (2 groups) vs. controls (positive and negative)	-3.89
Feng (2019)[21]	Curcumin inhibition of the PERK-eIF2α-CHOP pathway through promoting SIRT1 expression in oxidative stress-induced rat chondrocytes and ameliorates OA progression in a transected ACL rat model; rat origin chondrocyte cell line study with curcumin treatment of cells (5 dose groups) vs. untreated control	-1.96
Funk (2006)[22]	Efficacy and mechanisms of action of turmeric supplements in treatment of streptococcal cell wall induced OA in rats; turmeric extract injection vs. vehicle control, inject before, during, after max joint inflammation; n=8-12 per group	-1.96
Funk (2010)[23]	Anti-arthritic effects of streptococcal cell wall induced OA in rats and toxicity of essential oils of turmeric (curcuma longa l.); 4 groups (n=3/group), turmeric essential oil extractions inject or oral vs. vehicle inject or oral control	-2.58
Zhang (2018)[24]	Curcumin improves age-related spontaneous and DMM-induced OA in mice by promoting autophagy; 5 groups (10/group), 12 month curcumin treated spontaneous aging or surgery induced OA vs. aging control, sham surgery control	-1.96
Horcajada (2015)[25]	Oleuropein or rutin consumption decreases the spontaneous development of OA in the hartley guinea pig; 4 groups (n=15/group), untreated controls vs 3 treated groups: Oleo vs Oleo+Rutin vs Oleo + Rutin +Curcumin	-0.01
Khorsandi (2014)[26]	Combination of curcumin and piperine improved MIA-induced OA in rat model; 5 groups (n=8/group), oral curcumin, oral piperine, oral curmin+piperine, positive/negative controls	-1.96
Kim (2019)[27]	Effects of curcuma longa rhizoma on MIA-induced OA in rat model; 5 groups (n=6/group), curcumin high dose treated, curcumin low dose treated, NSAID treated vs positive/negative controls	-1.96
Li (2016)[28]	JAK2/STAT3 signal pathway mediating curcumin in cartilage cell metabolism of DMM-induced OA in rats; 3 groups (n=5/group), curcumin treatment group vs positive/negative controls	-1.96
Liu (2016)[29]	Inflammatory cytokines and oxidative stress markers in the inhibition of OA in mice by curcumin; 4 groups (n=5/group), curcumin low dose or curcumin high dose vs positive/negative controls	-1.96
Nakahata (2020)[30]	Periodic curcumin monoglucuronide injections ameliorate structural DMM-induced OA changes in rats; 8 groups (n=8/group), IA inject curcumin analyzed at 4 time-points vs saline control inject same 4 timepoints	-2.33
Niazvand (2017)[31]	Curcumin-loaded poly lactic-co-glycolic acid nanoparticles effects on MIA-induced OA in rats; 4 groups (n=10/group), oral curcumin or nano-curcumin to treat OA vs positive/negative controls	-2.58
Nicoliche (2020)[32]	Evaluation of the articular cartilage in rats with IA zymosan injection induced arthritis treated with curcumin in rats; 3 groups (n=5/group), oral curcumin vs positive/negative controls	-2.86
Nixon (2017)[33]	Evaluation of TRB-n0224, a chemically modified curcumin for the treatment of surgical ACL transection induced OA in rabbits; 4 groups (n=10/group), oral curcumin low dose, or curcumin high dose vs positive/ negative controls	-0.01
Park (2020)[34]	Highly bioavailable curcumin powder suppresses articular cartilage damage in rats with MIA-induced OA in rats; 6 groups (n=13/ group, oral saline n=12), oral Theracurmin® low dose, mid-dose or high dose vs. oral saline control (no OA induced), MIA-induced OA control, NSAID treated OA control	-2.58
Park (2016)[35]	Curcumin and tetrahydrocurcumin both prevent OA symptoms and decrease the expressions of pro-inflammatory cytokines in estrogen-deficient rats (MIA-induced OA, ovariectomized); 5 groups (n=10/group), oral curcumin or tetrahydrocurcumin vs. normal control, positive control, or placebo control	-1.96
Sun (2010)[36]	Effects of curcumin on expressions of CL-2, Mmp9, Cox-2, IKKβ, NF-Kb and tissue changes in rabbit stifle cartilage OA (papain injection); 3 groups (n=10/group), IA inject curcumin for OA group, saline IA for OA versus untreated control	-1.96
Sun (2017)[37]	Curcumin prevents DMM-induced OA in mice by inhibiting the activation of inflammasome NLRP3; 5 groups (n=8/group), IP inject curcumin for OA, DMSO for OA versus no surgery control. sham control, OA with no treatment	-2.58

Table 1 Cont.

Yan (2019)[38]	Involvement of TLR4 in the protective effect of intra-articular administration of curcumin on rat surgical ACL transection induced OA; 5 groups (n=10/group); IA curcumin, IA curcumin + lipopolysacchaide (LPS) inject vs positive/negative controls, LPS inject only	-1.96
Zhang (2019)[39]	Curcumin reduces inflammation in MIA-induced OA in rats through blocking TLR4/MyD88/NK- κB signal pathway; 4 groups (n=12/group); curcumin, curcumin + PBS vs positive/negative controls	-1.96
Zhang (2016)[40]	Curcumin slows OA progression and relieves DMM-induced OA-associated pain symptoms in a post-traumatic OA mouse model and cell line studies; In-vitro: cell line human OA affected chondrocytes treated 1) with curcumin 2) with nanoparticle curcumin; Mouse: 3 groups (n=8/group); oral curcumin, topical curcumin nanoparticle vs vehicle control	-1.96
Zhou (2020)[41]	Chemically modified curcumin (CMC2.24) alleviates ACL transection induced OA progression in rats by restoring cartilage homeostasis and inhibiting chondrocyte apoptosis via the NF-kB/HIF-2alpha axis; 5 groups (n=5/group), IA low dose modified curcumin, same but high dose vs sham with IA saline, OA with IA saline, high dose curcumin not modified	-3.29
Overall Treatment Efficacy (*p*-value)		$p = 3.03 \times 10^{-28}$

OA=osteoarthritis, MIA= monosodium iodacetate, DMM= destablization of medial meniscus, ACL= anterior cruciate ligament, IA=intra-articular, IP=intra-peritoneal; *Z score of -1.96/-2.58/-3.29/-3.89 was used when the *p*-value was reported as $p < 0.05/0.01/0.001/0.0001$; and a Z score of -0.01 was used when the *p*-value was reported as $p > 0.05$

Among the 24 experimental model studies qualified for inclusion, the majority reported results on multiple outcome measurements in several laboratory animal species (Table 2). The investigator selected 11 measurements that appeared in at least 2 studies: TNF-α, IL-1β, NF-κβ, MMG, IL-6, cartilage lesion scoring (Osteoarthritis Research Society International, OARSI), cartilage destructive matrix metalloproteinase-13 (MMP-13), monocyte chemotactic protein-1 (MCP-1), malondialdehyde (MDA) marker for oxidative stress, prostaglandin E_2 (PGE_2) and Bcl-2 associated X protein (Bax) apoptosis regulator. These measurements covered all 24 studies. For studies reporting statistical significance (*p*-values) on more than one of these 11 measurements, the one that appeared in the greatest number of studies was used for meta-analysis. When both TNFα and IL-1β were reported in a study, TNFα was used.

There were 5 separate meta-analyses performed on data collected from the 24 studies included in the systematic review. One meta-analysis assessed statistical significance of the overall OA treatment efficacy of turmeric/curcumin. The next 4 meta-analyses evaluated statistical significance of the 4 most common measurements (TNF-α, IL-1β, NF-κβ, MMG) used in studies to assess turmeric/curcumin's beneficial effects on treating arthritis.

Assessment of study quality was judged through assignment of Jadad scores (scale 0-5) based on randomization, blinding and fate of all subjects known. Of the 24 studies evaluated, 15 included a statement that randomization was performed for group assignments, but none described the method of randomization (score=1). None of the studies mentioned blinding during the study, nor described the fate of the subjects at the end of the study (score=0). This resulted in 15 studies with a Jadad Scale of 1 and the remaining 9 scored 0.

Overall Treatment Efficacy of Turmeric/Curcumin for OA

The overall efficacy of turmeric/curcumin in treating model-induced OA was investigated by meta-analysis of all 24 studies. The overall statistical significance was established by using Stouffer's Z-score method that converts each observed *p*-value to a Z score (Table 1). All 24 studies reported statistically significant efficacy for turmeric/curcumin treated OA in animal models. The overall *p*-value from this meta-analysis was statistically significant (3.03×10^{-28}).

Turmeric/Curcumin Treatment Effect on: TNF-α, IL-1β, MMG, NF-κβ

The second meta-analysis used the TNF-α data (mean±SD, sample size) reported in 8 studies (Table 2).[22,27,34,35,37-40] A random-effects model (between study variance) was applied and confidence intervals determined. For each of the studies, the animal group treated with turmeric/curcumin had smaller mean TNF-α outcomes (less joint inflammation) when compared to the untreated control group. The difference between control and treated animals was statistically significant in 7 of 8 studies and the meta-analysis revealed a statistically significant overall effect on the reduction of TNF-α ($p = 8.1 \times 10^{-7}$) in turmeric/curcumin treated animals when compared to controls. Significant ($p < 0.0001$) heterogeneity (variation) between this group of studies was documented. Data were not available for articles 30 and 33 (reported statistical significance, *p*-value, without providing mean+/-SD). These studies can be included in

the Stouffer's Z-score meta-analysis, but not effect size meta-analysis. It can be concluded that, based on the TNF-α outcomes, the evidence reported in these 8 animal model studies suggested that turmeric/curcumin could be effective in improving OA conditions.

In a third meta-analysis, the same protocol was performed on the IL-1β statistics (mean±SD, sample size). There were 8 studies used (Table 2).[18,23,27,35,37-40] The same meta-analysis model was used for each study with the standardized group mean difference and its 95% confidence interval determined. Similar to TNF-α, all studies observed smaller mean IL-1β outcomes in the curcumin treated group (less joint inflammation) compared to untreated controls. Six out of these 8 studies demonstrated statistical significance on group mean difference. The test of heterogeneity also suggested that significant heterogeneity existed among studies (p < 0.0001). The meta-analysis model revealed a significant overall effect (p = 0.0004). Data were not available for references 24 and 30 (reported statistical significance, p-value, without providing mean+/-SD).

The fourth meta-analysis used the same model as reported previously on the MMG statistics from 6 studies (Table 2).[24,26,31,34,39,41] Similarly, by looking at each mean group difference and the associated 95% confidence interval, all studies observed significantly smaller mean MMG outcomes in the curcumin treated group (less histologic damage) compared to untreated controls. The test of heterogeneity again suggested that significant heterogeneity existed among studies (p < 0.003). The meta-analysis model revealed a significant overall effect ($p = 9.6\times10^{-8}$). Data (mean+/-SD) were not available in reference 30.

In the fifth meta-analysis, there were 5 studies where NF-κβ statistics were extractable from the articles (Table 2).[27,34,36,38,39] The differences for group means and their 95% confidence for all studies reported smaller NF-κβ means in the curcumin treated animals (less joint inflammation) when compared to the control animals with 4 studies (article 38 not significant) able to conclude

Table 2: Measurements of inflammation and cartilage injury reported in each article used in the systematic review and meta-analysis.

Article Reference Number	TNF-α	IL-1β	MMG	NF-κβ	IL-6	MMP-13	OARSI	MCP-1	PGE2	MDA	Bax
18		x								x	
19							x				
20										x	
21							x				
22	x							x	x		
23		x						x			
24		x	x								x
25									x		
26			x								
27	x	x		x	x				x		
28											x
29								x			
30	x	x	x		x		x				
31			x								
32						x					
33	x				x	x					
34	x		x	x							
35	x	x			x	x					
36				x							
37	x	x									
38	x	x		x							
39	x	x	x	x	x						
40	x	x				x	x				
41			x	x		x					
Article Total	**10**	**10**	**7**	**6**	**5**	**5**	**4**	**3**	**3**	**2**	**2**

TNF-α=tumor necrosis factor-α; IL-1β=interleukin-1β; MMG=modified Mankins cartilage lesion grading; NF-κβ=nuclear factor-κβ; IL-6=interleukin-1; MMP-13=matrix metalloproteinases-13; OARSI cartilage lesion grading; MPC-1=monocyte chemotactic protein 1; PGE_2=prostaglandin E_2; MDA=malondialdehyde; Bax=Bcl-2 associated X protein

statistical significance. Significant heterogeneity among studies was again concluded by the heterogeneity test ($p < 0.001$). The meta-analysis model suggested a significant overall effect ($p = 0.0003$). Data were not available in article 41 (reported statistical significance, *p*-value, without providing mean+/-SD).

DISCUSSION

The goal of this systematic review was to determine turmeric/curcumin's therapeutic effect on OA disease based on scientific evidence in the literature reported over the past 20 years. The extensive literature search conducted by the investigator identified 24 controlled OA animal model studies satisfying inclusion criteria. A separate meta-analysis was constructed that evaluated each of the most common outcome measurements of joint injury (TNF-α, IL-1β, MMG, NF-κβ) for the research studies along with a meta-analysis investigating overall statistical significance for turmeric's efficacy in treating model-induced OA. All individual outcome meta-analyses were statistically significant with the overall *p*-value (all 24 studies) markedly significant at 3.03×10^{-28}. It can be concluded from this systematic review and meta-analysis that turmeric/curcumin has beneficial treatment effects on OA disease in animal model studies and should be investigated in companion animal clinical studies.

Modern research has demonstrated that curcumin is a highly pleiotropic molecule that interacts and regulates numerous molecular targets.[9] Both in-vitro and in-vivo studies have identified at least some of the underlying mechanisms that involve inflammatory cytokines (e.g. TNF-α, IL-1β), transcription factors (NF-κβ), growth factors, protein kinases and other enzymes (e.g. Cox-2).[9] The inflammatory cytokines are particularly important in OA disease and were the most common outcome (TNF-α, IL-1β) investigated for beneficial effects associated with turmeric administration in this systematic review and meta-analysis.

Findings from the meta-analysis demonstrated measurement of TNF-α and IL-1β levels in OA affected animals had a significant reduction ($p = 8.1\times10^{-7}$ and $p = 0.0004$ respectively) when treated with turmeric/curcumin as compared to controls. The reduction associated with turmeric is particularly remarkable in that inflammatory cytokines such as TNF-α, IL-6 and IL-1β, consistently increase after injury. The reduction of these cytokines is an important step in controlling the articular degradation process as these start a cascade of intracellular events that activate proteinases, promote extracellular matrix protein destruction, create a pro-destructive articular milieu, suppress anabolic pathways and synergize with other inflammatory cytokines to amplify cartilage destruction.[8,9,42]

Of interest and different from the studies looking at joint production of inflammatory cytokines are the 7 studies that collected and submitted cartilage for histologic evaluation of turmeric/curcumin treatment effects (Table 2). The modified Mankins histologic grading (MMG) is a scoring system which evaluates cartilage/subchondral bone injury. The scoring system (0-6) organized microscopic observation from 0 (normal) to 6 (severe disorganization/destruction) of affected cartilage.[31] Meta-analysis demonstrated a statistically significant smaller mean MMG in the turmeric/curcumin treatment groups as compared to controls ($p = 9.6\text{x}10^{-8}$). This suggests the potential effectiveness of turmeric/curcumin in helping to protect the cartilage surfaces from the degenerative process that leads to osteoarthritis. An additional histologic grading system (OARSI) was used to evaluate curcumin-treated OA disease.[19,21,30,40] Scores were also lower for test groups as compared to controls along with reduced synovitis and subchondral plate thickening. It was concluded that these findings provide evidence that curcumin significantly slows OA disease progression and exerts a palliative effect in an OA mouse model.

There were 5 studies that evaluated levels of NF-κβ, a pro-inflammatory mediator that stimulates the COX-2 inflammatory cascade (Table 2). Turmeric/curcumin not only demonstrated the ability to reduce inflammation by reducing the pro-inflammatory cytokines IL-1β and TNF-α but also able to affect more basic root causes such as NF-κβ expression of genes that induce inflammation. All studies analyzed in this systematic review demonstrated reduction in NF-κβ ($p = 0.0003$) as compared to the osteoarthritis control models. In human studies, patients with arthroscopic manifestations of early OA and knee pain but normal radiographs exhibited significantly higher immune-histological measures of inflammation, such as TNF-α, IL-1β, NF-κβ and COX-2 in synovial tissues.[11] The statistically significant reduction of these inflammatory mediators, as demonstrated in the present study, suggests benefit from the medicinal administration of turmeric/curcumin, particularly in slowing inflammatory destruction of joints.

From a TCM perspective, there is a long history of using functional foods in food therapy and for medicinal purposes.[43] Turmeric, in particular, is an important functional food with a wide variety of uses and formulations (Table 3). It is extensively cultured in the tropical areas of Asia and a number of global communities such as China, India, Iran and Indonesia use it to formulate certain traditional medications to cure ailments. The nutritional composition of the herb reveals it to be a rich source of carbohydrates and fiber.[43] It also contains some proteins and fats (no cholesterol) as well as pyridoxine, vitamin C, potassium, calcium, magnesium and phosphorus in appropriate amounts making it a nutritionally rich natural food product.[43] Turmeric has a variety of biological activities (326 have been identified) and has profound anti-inflammatory, antioxidant and anti-proliferative beneficial health effects.[43]

Many of the studies reviewed suggest that the bioavailability of turmeric/curcumin may be a limiting factor in its effectiveness as a treatment for arthritis. Curcumin and bisdemethoxycurcumin are highly hydrophobic molecules. This characteristic results in low

oral bioavailability, therefore, researchers have sought delivery systems to improve absorption for medicinal effects.[7,15,43] Formulations designed to enhance bioavailability can be divided broadly into 3 groups: 1) addition of lipid and/or piperine, 2) absorption and dispersion on matrices[b] and/or 3) decrease of particle size[c].[44] Curcumin is fat soluble, therefore, its consumption with a fatty meal improves absorption and piperine stimulates the gastrointestinal system to prevent efflux of curcumin. Early formulations describe incremental bioavailability increases with the addition of turmeric oil[d] and a small amount of piperine[e].[44] In addition to commercial formulations, a wide range of micellar and nano-particle formulations have been prepared which are being investigated in research studies similar to studies in this review.[19,44] More research needs to be done to determine how to make turmeric more bioavailable while keeping the product safe.[15]

Limitations to this systematic review included the necessity to use studies that had species variability (rats, mice, guinea pigs, rabbits), dissimilar experimental designs and numerous outcomes which led to statistically significant heterogeneity among studies. The multiple outcomes measured were particularly challenging in that of the 77 initial articles considered, 82 different measurements were reported. This systematic review selected and used 11 measurements that appeared in most studies. Other limitations to consider were the low Jadad scores assigned to experimental studies used in this investigation. The low scores predominately relate to using a scoring system that is designed for assessing quality in clinical trials with poor relevance to preclinical laboratory animal model studies.

It is worth mentioning the reasons why the majority of articles on human or veterinary subjects reviewed initially were excluded from the meta-analysis despite valuable information contained in these articles. First, many of these articles were review or systematic review articles, not clinical studies.[4,8,11-13,45-47] Second, 4 studies were excluded because the test treatments contained multiple ingredients (mixed with turmeric/curcumin) and hence these studies could not conclude effects on only turmeric/curcumin extracts.[48-51] Other studies were disqualified due to lack of standardized treatment or control groups on which to base conclusions on efficacy of turmeric/curcumin extracts.[1,6,52] Finally the systematic review and meta-analysis was only performed on laboratory animal model studies due to the limited number of human and veterinary clinical studies (1 human clinical study, 1 veterinary clinical study) that met the inclusion criteria for this study.[14,53]

Research in companion animals overall lacks systematic review studies, especially regarding supplements and Chinese herbal medicine. This is not unexpected due to the lack of quality randomized controlled clinical trials. In order to validate turmeric's efficacy, studies using larger sample sizes need to be conducted into dose and administration routes with canine pharmacokinetic data to document bioavailability of turmeric formulations. In addition, documentation of pain reduction in canine studies needs to be better defined. Studies reviewed noted changes in gait or ground reactive forces but the use of standardized grading systems for canine OA was lacking.[45] The safety of using turmeric/curcumin in OA canine patients also needs further study. Dejonckheere listed several contraindications in certain breeds and co-morbidities.[47]

Table 3: The main products of turmeric, their descriptions, and uses.[43]

Product	Description	Uses
Whole rhizome (dried)	- Appearance: orange-brown, red-yellow, or pale yellow - Chemical composition: it may contain 3-15% curcuminoids, and 1.5 to 5% essential oils	Medicinal purposes
Ground turmeric	- Appearance: either yellow or red-yellow in color - Chemical composition: the main active ingredients (i.e., curcuminoids and essential oils) may decrease during processing and also by exposure to light. It is necessary to pack the powder in a UV protective container	Used as a spice, dye, medicine, dietary supplement
Turmeric oil	- Appearance: yellow to brown oil - Chemical composition: essential oils from the leaves are usually dominated by monoterpenes. Rhizomes oil mainly contains sesquiterpenes	Used as a spice, medicine, dietary supplement
Turmeric oleoresins	- Appearance: dark yellow, reddish-brown viscous fluid - Chemical composition: they consist of up to 25% essential oil and 37-55% curcuminoids	Used as food coloring, medicine, dietary supplement
Curcumin	- Appearance: crystalline powder yellow to orange-red color - Chemical composition: a mixture of curcumin and its bisdemethoxy- and demethoxy-derivatives (no fixed proportions). The three major curcuminoids may occupy 90% of the whole proportion. Oils and resins may be the minority of composition	Used as medicine and dietary supplement

Adapted from Ahmad R et al. Biochemistry, safety, pharmacological activities and clinical application of tumeric: A mechanistic review.[43]

In conclusion, turmeric/curcumin is a promising treatment for osteoarthritis and was supported by this systematic review and meta-analysis. This suggests further investigation of its beneficial effects on canine OA in clinical studies would be justified. From a TCVM perspective, turmeric with its major component, curcumin, may be useful as part of food therapy and Chinese herbal medicine formulations. Current recommendations following this review would be to use turmeric in combination with other treatments for the prevention and treatment of osteoarthritis while monitoring the patient for safety.

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FOOTNOTES

a. R version 3.5.2. The R Foundation for Statistical Computing, Vienna Austria; http://www.R-project.org
b. Cureit®, Aurea Biolabs, Kerala, India
c. Biocurc®, Boston BioPharm Inc, Boston, MA, USA
d. Biocurcumax®, Arjuna Natural Ltd, Alura, India
e. Curcumin C^3 complex®, Sabinsa Corporation, East Windsor, NJ, USA

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