Chondroitin sulfate in the treatment of osteoarthritis: from *in vitro* studies to clinical recommendations

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Yves Henrotin, Mariane Mathy, Christelle Sanchez and Cecile Lambert

***Abstract*:** Chondroitin sulfate (CS) is recommended as a therapeutic intervention in the multimodal approach of osteoarthritis (OA) management. CS has been studied extensively to describe its pharmacology (pharmacokinetic, *in vitro* and *in vivo* effects) and its clinical efficacy. Various results have been reported depending on the system of evaluation (model, dosage and duration) and the source of CS (origin and quality). The purpose of this review was to gather most of the available information about CS and to discuss its potency in OA management.

***Keywords*:** cartilage, chondroitin sulfate, osteoarthritis, mechanism of action, pharmacokinetic

Correspondence to:
Yves Henrotin, PhD
Bone and Cartilage Research Unit, Institute of pathology, level 5, CHU Sart-Tilman, 4000 Liège, Belgium
yhenrotin@ulg.ac.be

Mariane Mathy, PhD
Christelle Sanchez, PhD
Cecile Lambert, PhD
Bone and Cartilage Research Unit, Institute of Pathology, level +5, CHU Sart-Tilman, 4000 Liège, Belgium

Introduction

Chondroitin sulfate (CS) is a major component of the extracellular matrix (ECM) of many connective tissues, including cartilage, bone, skin, ligaments and tendons. Osteoarthritis (OA) is characterized by progressive structural and metabolic changes in joint tissues, mainly cartilage degradation, subchondral bone sclerosis and inflammation of synovial membrane. OA management involves multimodal therapeutic intervention since no cure has been found to date. OA management requires drugs that could slow down, stop or even avoid joint degradation. Many of the recommended interventions present only symptom-modifying effects and a few structure-modifying effects.

CS and other compounds, such as glucosamine, have been used for medicinal purposes for over 40 years. CS is sold as over the counter dietary supplement in North America and is a prescription drug under the regulation of the European Medicine Agency (EMEA) in Europe. CS has raised many interests over the past decades as a potential therapeutic against OA. CS is part of the Osteoarthritis Research Society International (OARSI) recommendations for the management of knee OA [Zhang *et al.* 2010, 2008] and of the European League Against Rheumatism (EULAR) recommendations for the management hip and knee OA [Zhang *et al.* 2005; Jordan *et al.* 2003]. CS and glucosamine could represent a great alternative for OA patients.

CS, as a natural component of the ECM, is a sulfated glycosaminoglycan (GAG) composed of a long unbranched polysaccharide chain with a repeating disaccharide structure of N-acetylgalactosamine and glucuronic acid. Most of the N-acetylgalactosamine residues (Figure 1) are sulfated, particularly in the 4- and 6-position, making CS a strongly charged polyanion. CS is responsible for many of the important biomechanical properties of cartilage, such as resistance and elasticity. Its high content in the aggrecan plays a major role in allowing cartilage to resist pressure stresses during various loading conditions. Chondroitin sulfatation profile has been described in cartilage [Mourao, 1988]. Changes in the structure of CS have been described in OA tissues [Caterson *et al.* 1990] and a diminished ratio of CS-6:CS-4 have been found in synovial fluid and cartilage of OA patients [Shinmei *et al.* 1992]. More than three decades ago, it was proposed [Schwartz and Dorfman, 1975] that CS supply, as a therapeutic intervention to cartilage damage, could provide building blocks for the synthesis of new matrix component, since increasing CS concentration could act in favour of matrix regeneration and account for its beneficial effects.

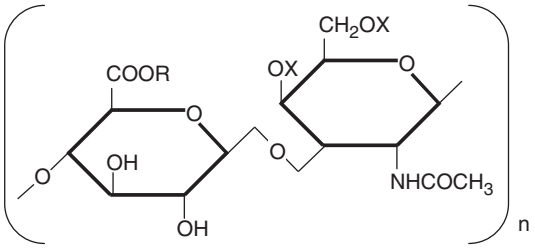

Figure 1. Chemical structure of chondroitin sulfate, where R is Na or H and X is SO_3R or H.

However, since then, the mechanism of action of CS, as a therapeutic intervention, is still to be elucidated and its efficacy is under debate.

The overall goal of this review was to gather most of the available information about CS, its pharmacokinetics, its *in vitro* and *in vivo* effects and its clinical efficacy. The aim on this review is to discuss the clinical usefulness of CS and the perspectives for CS use in OA management.

Pharmacokinetics

Natural CS has a molecular weight of 50–100 kDa. After the extraction process, the molecular weight is 10–40 kDa depending on the raw material. CS is mainly obtained from bovine, porcine or marine (shark) cartilage.

CS is mostly administered orally at doses ranging from 800 to 1200 mg/day. CS is rapidly absorbed by the gastrointestinal tract. The absorbed CS reaches the blood compartment as 10% CS and 90% depolymerized low-molecular-weight derivatives.

Different bioavailability and pharmacokinetic variables have been reported depending on the CS characteristics and origin [Malavaki *et al.* 2008]. Some of them have been reported in Table 1. The bioavailability of CS is under debate. The data depends on the system used for evaluation, the structure and characteristic of CS such as molecular mass, charge density and cluster of disulfated disaccharides, the concentration used and the animal species used to perform the study [Volpi, 2003]. Most of the studies used CS from bovine trachea (95% purity) which is the same material used in clinical trials [Barnhill *et al.* 2006].

The bioavailability of CS ranges from 10% to 20% following oral administration [Lauder, 2009]. CS absorption could depend on the sulfatation status. The position and percentage of sulfate groups vary generally in relation to specific animal sources [Volpi, 2006a, 2006b]. Indeed, desulfated chondroitin in mouse [Kusano *et al.* 2007] has a very rapid uptake, with a peak occurring within 15 min and a very rapid clearance returning to baseline after 3 h, whereas shark CS in male healthy volunteers has a slower uptake (peak at 8.7 h) and slower clearance (levels maintained until 16 h postadministration) [Volpi, 2003]. CS from bovine trachea presents a rapid increase in plasma of male healthy volunteers with a peak within 1–5 h after administration and a return to baseline after 10 h [Volpi, 2002].

The pharmacokinetic parameters of CS determined in healthy male volunteers [Volpi, 2002] after oral administration showed a significant increase in CS plasma levels (more than 200%) compared with predose levels over a 24-h period. The peak concentrations were observed 2 h after dosage and the increase is significant from 2 to 6 h after dosage. First-order kinetics was observed for doses up to 3000 mg. Multiple doses of 800 mg in patients with OA do not alter CS kinetics. Recently, using fluorophore-assisted carbohydrate electrophoresis and superpose 6 chromatography, Jackson and colleagues have observed no modification of the endogenous concentration (20 µg/ml) and CS disaccharide composition after oral ingestion of a single dose of 1200 mg CS. One explanation could be that little, if any, of the ingested CS reaches the circulation in a form which is unchanged or composed of disaccharides of larger fragments [Jackson *et al.* 2010]; alternatively, it could be that the assay was not sensitive enough to detect lower concentrations. This remains to be demonstrated by other methods of analysis. Another characteristic of CS is its capacity to accumulate in joint tissue. A high content of labelled CS has been found in joint tissues, including synovial fluid and cartilage after oral administration in humans [Ronca et al. 1998].

It is difficult to assess the relevancy of the maximal concentration attained in the blood compartment (C_{max}) for this kind of drug, since *in vivo* the response can be the result of the effects of CS and disaccharides. What is more, CS is a slow-acting drug, resulting in a slow onset of action with a maximal effect attained

Table 1. Different pharmacokinetic parameters depending on the system of study and the source of CS.

Study	Chondroitin source	System	Results	
Conte *et al.* [1995]	Bovine trachea 16 mg/kg orally	Wistar rats	$C_{max} = 2.3 \pm 0.4$ µg/ml $t_{max} = 1.6 \pm 0.4$ h	
	Bovine trachea 16 mg/kg orally	Young beagle dogs	$C_{max} = 1.9 \pm 0.3$ µg/ml $t_{max} = 2.1 \pm 0.4$ h	
	Bovine trachea Single dose of 0.8 g or two doses of 0.4 g	Healthy volunteers ($N = 12$; 6 males and 6 females)	Single dose: $C_{max} = 2.6 \pm 0.5$ µg/ml $t_{max} = 5.0 \pm 1.0$ h	Two doses: $C_{max} = 1.2 \pm 0.2$ µg/ml $t_{max} = 5.2 \pm 1.0$ h
	Bovine trachea Single dose 0.8 g/day	OA patients ($N = 18$)	Plasma concentration: D5: 1.80 ± 0.69 µg/ml D15: 1.70 ± 0.0.68 µg/ml D30: 1.89 ± 0.74 µg/ml	
Ronca *et al.* [1998]	CS-6 and CS-4 mix (1:1) Oral administration 16 mg/kg (4 mg/rat)	Rats ($N = 10$)	CS: $C_{max} = 2.5 \pm 0.5$ µg/ml $t_{max} = 46 \pm 15$ min CS + metabolites: $C_{max} = 8.9 \pm 0.9$ µg/ml $t_{max} = 635 \pm 91$ min	
Volpi [2002]	Chondrosulf ® Single dose (4 g)	Male healthy volunteers ($N = 20$)	$C_{max} = 3.88 \pm 0.85$ µg/ml $t_{max} = 21.7 \pm 14.1$ h	
Volpi [2003]	Shark CS Single dose (4 g)	Male healthy volunteers ($N = 20$)	$C_{max} = 4.87 \pm 2.05$ µg/ml $t_{max} = 8.7 \pm 4.5$ h	

CS, chondroitin sulfate; C_{max}, maximal plasmatic concentration; t_{max}, time to reach maximal plasmatic concentration; D, day of treatment.

after several months. In addition, CS is a drug of biological origin meaning that its measurement in biological fluids does not discriminate the drug from endogenous molecules. Maximal effect (E_{max}) has been predicted using an alternative method [du Souich and Vergès, 2001]. The effect E_{max} was calculated based on clinical efficacy. It was calculated that 50% of E_{max} is reached in 35 days in patients with mild OA. The approximate half-life of CS and its derivatives in plasma in humans is 15 h. The steady state is attained after 3–4 days and 3–6 months of treatment may be needed to obtain the maximal effect.

CS is not metabolized by cytochrome P450. This is in favour of a very low risk of interaction with other drugs.

Finally, a carry-over effect has been described for CS. Indeed after a delayed onset of action, the maximal effect persists after the cessation of therapy.

Mechanism of action

Many effects have been described for CS in different *in vitro* and *in vivo* studies using different types and concentrations of CS and different models. The exact mechanism of action still remains to be detailed. *In vitro* effects on joint cells and *in vivo* effects in arthritic models have been reported in Tables 2 and 3, respectively.

Anti-inflammatory effect

CS has been reported to have anti-inflammatory effects. It was shown to inhibit *in vitro* the synthesis of various inflammatory intermediates, such as nitric oxide (NO) synthase, cyclooxygenase (COX)-2, microsomal prostaglandin synthase (mPGES)-1 and prostaglandin (PG) E_2 [Campo *et al.* 2009a; Legendre *et al.* 2008; Chan *et al.* 2005b; Bassleer *et al.* 1998]. It was also shown that CS could act on the toll-like receptor (TLR)-4 to inhibit the inflammatory cytokines, MyD88 and tumour necrosis factor (TNF) receptor associated factor (TRAF)-6, through the inhibition of nuclear factor (NF)-κB activation [Campo *et al.* 2009b].

The anti-inflammatory effect of CS was also demonstrated *in vivo*. CS was shown to reduce synovitis in collagen-induced arthritis in mouse [Omata *et al.* 2000]. It was also able to reduce the pro-inflammatory cytokine, interleukin (IL)-6, in the same model [Cho *et al.* 2004]. CS also reduced IL-1β in joint tissue in Freund's adjuvant arthritis when administered as a dietary supplement [Chou *et al.* 2005] and TNF-α and myeloperoxidase in the plasma of rat with collagen-induced arthritis and after oral administration [Campo *et al.* 2003].

Table 2. *In vitro* effects of chondroitin sulfate.

Study	Chondroitin	System and duration	Results
Verbruggen *et al.* [1977]	10–400 μg/ml	Monolayer synovial cells	Stimulation by 11% of hyaluronate synthesis during the growth phase Increase by 88% during the stationary phase
Bassleer *et al.* [1998]	Bovine 100–1000 μg/ml	Human articular chondrocytes in clusters stimulated with IL-1β Up to 32 days	Significant reduction of IL-1β-stimulated production of PGE_2 Increase of PG and type II collagen production
Wang *et al.* [2002]	C poly S 10 μg/ml	Human articular chondrocytes in agarose gel stimulated with IL-1β 1 week	Stimulation of aggrecan, hyaluronan and type II collagen due to MMPs downregulation
Campo *et al.* [2004]	CS-4 1.0 and 2.0 mg/ml	Human skin fibroblast	Anti-oxidant action of CS-4 by increased cell viability and inhibition of DNA fragmentation
Chan *et al.* [2005a]	20 μg/ml	Bovine articular cartilage explants stimulated with IL-1β 24 to 48 h	Reduction of IL-1β-induced MMP-13
Chan *et al.* [2005b]	20 μg/ml	Bovine articular cartilage explants stimulated with IL-1β 24 to 48 h	Suppression of IL-1β-induced NO synthase, COX-2 and mPGES-1 gene expression
Monfort *et al.* [2005]	10–1000 μg/ml	Human OA chondrocytes stimulated with IL-1β 48 h	Inhibition of MMP-3
Holzmann *et al.* [2006]	25 μg/ml	Human articular chondrocytes stimulated with LPS Up to 72 h	Inhibition of MMP-13 by the reduction of p38 and Erk1/2 activation
Tat *et al.* [2007]	200 μg/ml	Human OA subchondral bone osteoblasts Up to 48 h	Reduction of proresorptive agents Reversion of vitamin D3 effect on OPG:RANKL ratio
Tahiri *et al.* [2008]	Chondrosulf® or porcine 10 or 100 μg/ml	Rabbit articular chondrocytes stimulated with IL-1β 20 h	Increase PG synthesis Inhibition of ADAMTS-5 expression
Jomphe *et al.* [2008]	Purified bovine CS-4 and CS-6 200 μg/ml	Rabbit chondrocytes stimulated with IL-1β 48 h	Reduction of NF-κB nuclear translocation Prevention of Erk1/2 and p38 activation Reduction of nitroprusside-induced apoptosis
Legendre *et al.* [2008]	Avian CS 1–100 μg/ml	Bovine articular chondrocytes in hypoxic alginate beads 24–48 h	Decrease of IL-1β-induced MMP-1, -3, -13 and ADAMTS-4 and -5 Reduction of IL-1β-induced COX-2 and iNOS Reduction of PGE_2 production but not NO Slight increase of GAG synthesis Counteract IL-1β suppression of TGFβ receptors
Katta *et al.* [2009]	Mix of CS A, CS B and CS C 10–50 mg/ml	Bovine cartilage explants 24 h	Effective at joint lubricant Diffusion into cartilage
David-Raoudi *et al.* [2009]	Avian CS 100 μg/ml	Human OA fibroblast-like synviocytes 48 h with pretreatment	Regulation of hyaluronan synthase
Campo *et al.* [2009a]	Bovine CS-4 and shark CS-6 0.5 and 1.0 mg/ml	Mouse articular chondrocytes stimulated with LPS 24 h	Anti-apoptotic effect of both CS Inhibition of inflammatory mediators, iNOS and MMPs more evident with CS-4 than with CS-6
Campo *et al.* [2009b]	Bovine CS-4 and shark CS-6 25 and 50 μg/ml	Mouse articular chondrocytes stimulated with LPS 24 h	Inhibition of inflammatory cytokines (MyD88 and TRAF-6), inhibition of activation of NF-kB and iNOS by both CS Effect through TLR4 (confirmed with specific antibody experiment)

(continued)

Table 2. Continued.

Study	Chondroitin	System and duration	Results
Bian *et al.* [2009]	CS-6 and CS-4 from shark origin 10–100 mg/ml	Bovine cartilage explants 4 weeks	Reduction of GAG content
Imada *et al.* [2010]	Shark (mainly CS-6) and porcine (mainly CS-4) 1–100 μg/ml	Human articular chondrocytes in alginate beads and synovial fibroblast stimulated with IL-1β 6 or 4 days	Different effect of both CS Suppression of IL-1β-induced ADAMTS-4 and -5 in both cell types Suppression of IL-1β-induced MMP-13 and increase expression of aggrecan in chondrocytes Recovery of TIMP-3 in chondrocytes and TIMP-1 in synoviocytes by porcine CS

CS, chondroitin sulfate; IL, interleukin; PGE_2, prostaglandin E_2; PG, proteoglycan; MMP, matrix metalloproteinase; NO, nitric oxide; OPG, osteoprotegerin; RANKL, receptor activator for NF-κB ligand; ADAMTS, a disintegrin and metalloprotease with thrombospondine motifs; NF-κB, nuclear factor κB; GAG, glycosaminoglycan; TGF, transforming growth factor; TLR, Toll-like receptor; LPS, lipopolysaccharide.

Anticatabolic and anabolic effects

The first anabolic effect of CS was described more than 30 years ago. CS increased the synthesis of hyaluronate in synovial cells [Verbruggen and Veys, 1977]. Since then, it was also demonstrated to increase type II collagen and proteoglycan (PG) synthesis in human chondrocytes [Wang *et al.* 2002; Bassleer *et al.* 1998] and GAG in bovine chondrocytes [Legendre *et al.* 2008]. This effect can also be secondary to matrix metalloproteinase (MMP) downregulation [Wang *et al.* 2002]. CS was also shown to inhibit MMP-1, -3 and -13, and ADAMTS-4 and -5 (aggrecanases) in human, porcine and bovine chondrocytes [Imada *et al.* 2010; Legendre *et al.* 2008; Tahiri *et al.* 2008; Chan *et al.* 2005a; Monfort *et al.* 2005]. MMP-13 inhibition can happen through the inhibition of p38 and Erk1/2 activation [Holzmann *et al.* 2006]. Different inhibitory potencies were demonstrated for CS-4 and CS-6 against MMPs in murine chondrocytes [Campo *et al.* 2009a]. Differences between CS-6 and CS-4 were also recently shown in human cells. Indeed, CS-6 (shark origin) can counteract IL-1β inhibition of tissue inhibitor of metalloproteases (TIMP)-3 in human chondrocytes and TIMP-1 in synovial fibroblast, whereas CS-4 (porcine trachea origin) did not produce the same effect [Imada *et al.* 2010]. CS can upregulate hyaluronan synthase in fibroblast-like cells [David-Raoudi *et al.* 2009] and could be efficient as a joint lubricant as was shown in bovine cartilage explants [Katta *et al.* 2009]. It was also reported that CS could have an influence on the resorption process that takes place in the subchondral bone during OA. CS could indeed increase the osteoprotegerin (OPG):receptor activator for NF-κB ligand (RANKL) ratio in OA osteoblasts in favour of subchondral bone homeostasis [Tat *et al.* 2007]. CS was demonstrated to act on most of the joint tissues involved in OA pathophysiology. Finally, a recent *in vitro* study in bovine cartilage explants treated with CS-4 and CS-6 for 4 weeks have revealed a negative effect of CS showing that it reduced the GAG content [Bian *et al.* 2009]. It is important to note that this study was performed with the highest concentration of CS ever tested (10–100 mg/ml).

The anabolic and anticatabolic effect of CS has also been shown *in vivo*. CS can increase PG production in a rabbit model of cartilage degradation [Uebelhart *et al.* 1998], prevent the increase of MMP-9 in Freund's adjuvant arthritis in rat when administered as a dietary supplement [Chou *et al.* 2005] or inhibit MMP-13 in collagen-induced arthritis in mouse [Campo *et al.* 2008].

Anti-apoptotic effect

CS has been demonstrated to have anti-apoptotic properties both *in vitro* and *in vivo*. It has been shown that CS (200 μg/ml) reduced the sensibility of rabbit chondrocytes to apoptosis [Jomphe *et al.* 2008]. This effect was associated with the reduction of NF-κB translocation and with the reduction of the MAP kinase signalling pathway through p38 and Erk1/2. Another study has pointed to the anti-apoptotic effect of both CS-4 and CS-6 in mouse articular chondrocytes [Campo *et al.* 2009a]. *In vivo*, it was shown in a

Table 3. *In vivo* effects of chondroitin sulfate.

Study	Chondroitin	Model	Results
Uebelhart *et al.* [1998]	80 mg/day oral or i.m.	Rabbit model of cartilage injury induced by chymopapain	Significant increase of PG production
Omata *et al.* [2000]	1000 mg/kg	Collagen-induced arthritis in mouse	Reduction of synovitis parameters: cell infiltration, fibrosis and proliferation of lining cells
Campo *et al.* [2003]	1 ml/kg (i.p.) 25 mg/kg (orally)	Collagen-induced arthritis in rat	Treatment began after arthritis onset (day 10) Protection against oxidative stress Reduction of plasma levels of TNF-α and myeloperoxidase
Cho *et al.* [2004]	1200 mg/kg	Collagen-induced arthritis in rat	Reduction of IL-6 levels
Caraglia *et al.* [2005]	0.3 mg/day	Spontaneous OA in mouse 12 days	Significant reduction of apoptotic chondrocytes after 30 days
Chou *et al.* [2005]	Dietary supplement (18 mg/g)	Freund's adjuvant-induced arthritis in rat	Prevention of MMP-9 increase Reduction of IL-1β in joint tissues but not in serum
Campo *et al.* [2008]	CS-4 (bovine trachea) 30–120 mg/kg (i.p.)	Collagen-induced arthritis in mouse For 25 days	Inhibition of NF-κB activation Inhibition of MMP-13 Inhibition of caspases-3 and -7 activation

TNF, tumour necrosis factor; i.m., intramuscularly; i.p., intraperitoneally; CS, chondroitin sulfate; TNF, tumour necrosis factor; MMP, matrix metalloproteinase; IL, interleukin; OA, osteoarthritis; NF-κB, nuclear factor κB; PG, proteoglycan.

mouse model of spontaneous OA that a 12-day treatment with CS (0.3 mg/day) reduce the number of apoptotic chondrocytes when assessed after 30 days [Caraglia *et al.* 2005]. It was also demonstrated to be able to inhibit caspase-3 and -7 activation, in collagen-induced arthritis in mouse [Campo *et al.* 2008]. These results are in favour of a chondroprotective effect of CS.

Anti-oxidant effect

The anti-oxidant effects have been reviewed [Campo *et al.* 2006a, 2006b]. CS was shown to have anti-oxidant properties *in vitro* in human skin fibroblasts [Campo *et al.* 2004] and *in vivo* in collagen-induced arthritis in rat [Campo *et al.* 2003]. CS provides protection against hydrogen peroxide and superoxide anions. Indeed, these studies demonstrated that it could limit cell death, reduce DNA fragmentation and protein oxidation, decrease the generation of free radicals and act as a free radical scavenger. It reduces lipid peroxidation and improves anti-oxidant defence by restoring endogenous anti-oxidants reduced glutathione (GSH) and superoxide dismutase (SOD).

Cell signalling pathway regulation

CS has been found to act on major cell signalling pathways. It was shown to inhibit the activation of the MAP kinase pathway, p38 and Erk ½ and the inhibition of NF-κB transactivation in rabbit chondrocytes [Jomphe *et al.* 2008; Vergés *et al.* 2004]. CS was also shown to act on other cell signalling intermediates [Egea et al. 2010]. CS was able to induce the protein kinase C (PKC)/PI3K/Akt pathway in neuroblastoma cells. Many effects described for CS on OA pathophysiology could be due to its action on cell signalling.

It is important to keep in mind that most of the *in vitro* effects have been obtained using high concentrations of CS, which are not in concordance with the expected plasmatic concentrations. What is more, considering the slow acting effect of the drug in humans, the *in vitro* and *in vivo* systems were used for quite short durations. Different authors have suggested that the high concentration used *in vitro* was used to compensate these concerns [Tat *et al.* 2010; Monfort *et al.* 2008b]. Differences in the purity and properties of CS could be responsible for the disparity of the effects. This was addressed recently in two comprehensive reviews [Rainsford, 2009; Volpi, 2009]. This matter was even confirmed in articular chondrocytes [Tat *et al.* 2010] when three different types of CS were tested in the same system and produced different effects on key OA mediators. This study demonstrated that CS from bovine origin was much more efficient at inhibiting catabolic mediators than that of porcine origin. This is also important to note that the *in vivo* effects of CS have been mostly observed in animal models of arthritis. Only one study has been conducted in a spontaneous OA model [Caraglia *et al.* 2005].

The exact mechanism of action of CS still remains to be clarified. Taken altogether, these data suggest the intervention of CS at different levels of pathophysiology of OA, as summarized in Figure 2.

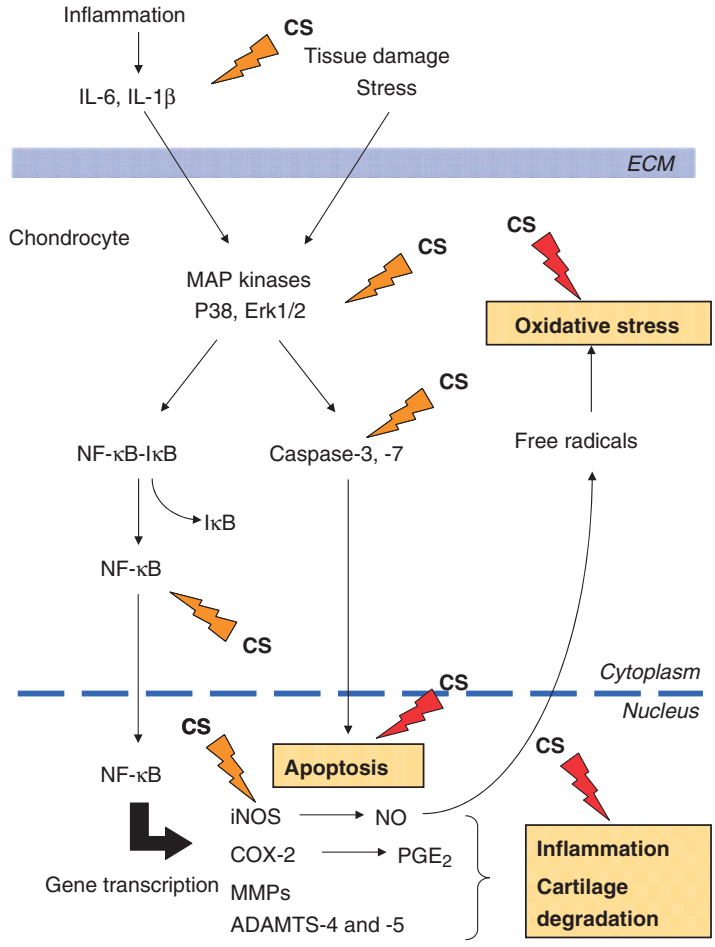

Figure 2. Schematic representation of chondroitin sulfate (CS) targets in chondrocytes during osteoarthritis as suggested by *in vitro* studies: CS acts on cell signalling, inflammatory and catabolic pathways and on oxidative stress (as shown with black arrows). CS targets different intermediates in these pathways (as shown with grey arrows). CS, chondroitin sulfate; IL, interleukin; ECM, extracellular matrix; PGE_2, prostaglandin E_2; PG, proteoglycan; MMP, matrix metalloproteinase; NO, nitric oxide; ADAMTS, a disintegrin and metalloprotease with thrombospondine motifs; NF-kB, nuclear factor kB; COX, cycloossigenase, iNOS, inducible Nitric Oxide Synthase.

Clinical effect of chondroitin sulfate

The therapeutic efficacy of CS has been studied and reported in different clinical trials in OA patients. The effect of CS on OA patients has been evaluated either on OA symptoms (pain and function) to determine its symptomatic slow-acting drug for OA (SYSADOA) effect or on disease modification (structure effect, joint space narrowing (JSN)) to determine its disease-modifying OA drug (DMOAD) effect.

Symptomatic slow-acting drug for OA effects

Various meta-analyses [Reichenbach *et al.* 2007; Richy *et al.* 2003; Leeb *et al.* 2000; McAlindon *et al.* 2000] have reported a different effect size (ES) on pain and function for CS ranging from 0.9 to as low as 0.3.

Recently, three studies have been designed and published regarding the SYSADOA effect of CS, i.e. the GAIT study [Hochberg and Clegg, 2008; Clegg *et al.* 2006], the STOPP trial [Kahan *et al.* 2009] and one other randomized controlled trial (RCT) [Maziéres *et al.* 2007; Maziéres *et al.* 2006] (Table 4). More recently, the Osteoarthritis Research Society International (OARSI) recommendations for OA management determined a moderate to large ES (0.75, 95% CI 0.50–0.99) [Zhang *et al.* 2010] but emphasized the publication bias, the heterogeneity of the results and a cumulative reduction of the ES (1.85, 95% CI 1.47–2.22, in 1992 to 0.75, 95% CI 0.50–1.01, in 2006).

Clinical trials have reported globally a beneficial effect of CS on pain and function (Table 4).

Table 4. Clinical efficacy of CS.

Study	Outcome	Intervention	Population and duration	Outcome/ effect size
Global assessment				
Maziéres *et al.* [2007]	Investigators global assessment of clinical improvement (knee)	CS *vs.* placebo	RCT ($n=307$) Knee OA 6 months (end of treatment)	3.1 (CS) and 2.5 (Placebo), $p=0.044$ Favours CS
	Patient global assessment of clinical improvement (knee)	CS *vs.* placebo	RCT ($n=307$) Knee OA 6 months (end of treatment)	NS
Quality of Life				
Maziéres *et al.* [2007]	SF-12 physical component (change from baseline)	CS *vs.* placebo	RCT ($n=307$) Knee OA 6 months (end of treatment)	5.8 (CS) and 3.8 (placebo), $p=0.021$ Favours CS
	SF-12 mental component	CS *vs.* placebo	RCT ($n=307$) Knee OA 6 months (end of treatment)	NS
Pain				
Clegg *et al.* [2006]	Pain	CS *vs.* placebo	RCT, $N=1583$, 24 weeks	CS not better than placebo
Maziéres *et al.* [2007]	Pain during activity, VAS (change from baseline)	CS *vs.* placebo	RCT ($n=307$) Knee OA 6 months (end of treatment)	-41% (CS) and -32% (Placebo), $p=0.029$ Favours CS
	Pain at rest, VAS (change from baseline)	CS *vs.* placebo	RCT ($n=307$) Knee OA 6 months (end of treatment)	NS
Reichenbach *et al.* [2007]	Pain related outcomes	CS *vs.* placebo/no treatment	MA, 20 RCTs, $n=3846$, knee or hip OA, trials length 6 to 132 weeks	Significant heterogeneity
Kahan *et al.* [2009]	VAS and WOMAC subscale	CS *vs.* placebo	RCT ($N=622$), knee OA, follow up 2 years	Significantly faster improvement in pain in target knee with CS
Function				
Clegg *et al.* [2006]	Joint swelling, effusion or both on clinical examination	CS *vs.* placebo	RCT, $N=1583$, 24 weeks	CS significantly better
Maziéres *et al.* [2007]	Lequesne's index (change from baseline)	CS *vs.* placebo	RCT ($n=307$) Knee OA 6 months (end of treatment)	NS
Structure modification				
Reichenbach *et al.* [2007]	Minimum JSW	CS *vs.* placebo/ no treatment	MA, 5 RCTs, $N=1192$, knee and hip OA, studies length range 6 to 132 weeks	ES mean difference 0.16 mm, 95% CI 0.08–0.24 Favours chondroitin
	Mean JSW	CS *vs.* placebo/no treatment	MA, 5 RCTs, $N=1192$, knee and hip OA, studies length range 6 to 132 weeks	ES mean difference 0.23, 95% CI 0.09–0.37 Favours chondroitin

(continued)

Table 4. Continued.

Study	Outcome	Intervention	Population and duration	Outcome/ effect size
Kahan *et al.* [2009]	Minimum JSW	CS *vs.* placebo	RCT ($N=622$), knee OA, follow up 2 years	Increased effect of treatment with time CS significantly better for JSW
Lee *et al.* [2010]	Minimum JSN	CS *vs.* placebo	MA, 4 RCTs, $N=1088$, Knee OA, follow up of 1 to 2 years	Small but significant protective effect of CS at 2 years on minimum JSN
Hochberg [2010]	Minimum JSW		MA, 3 RCTs, $N=1179$, Knee OA, follow up 2 years	Significant reduction in the rate of decline in joint space width with CS ES 0.23 (95% CI 0.11–0.35)
Adverse events				
Maziéres *et al.* [2007]	Number of patients with at least one treatment-related AEs	CS *vs.* placebo	RCT, $N=307$, knee OA, 24 weeks	NS
	Total number of AEs	CS *vs.* placebo	RCT, $N=307$, knee OA, 24 weeks	$N=18$ (CS) and $N=20$ (placebo) NS
Reichenbach *et al.* [2007]	Patients experiencing AEs	CS *vs.* placebo/no treatment	MA, 12 RCTs, $N=1929$, knee and hip OA, studies length range 6 to 132 weeks	NS
	Withdrawals due to AEs	Chondroitin *vs.* placebo/no treatment	MA, 9 RCTs, $N=1781$, knee and hip OA, studies length range 6 to 132 weeks	NS
	Patients experiencing severe AEs	Chondroitin *vs.* placebo/no treatment	MA, 2 RCTs, $N=217$, knee and hip OA, studies length range 6 to 132 weeks	NS
Kahan *et al.* [2009]	Withdrawals due to AEs	CS *vs.* placebo	RCT ($N=622$), knee OA, follow up 2 years	NS

CS, chondroitin sulfate; AE, adverse event; RCT, randomized controlled trial; NS, nonsignificant; OA, osteoarthritis; SF-12, Short Form 12-item health survey; MA, meta-analysis; ES, effect size; JSW, joint space width; JSN, joint space narrowing; VAS, visual analogue scale; WOMAC, Western Ontario and McMaster Universities Arthritis Index.

However, the largest trial, the GAIT study, reported no effect on these endpoints.

Structure-modifying effects

The structure-modifying effects of CS have been reported and analysed in two recent meta-analyses [Hochberg, 2010; Lee *et al.* 2010]. It has previously been reported that there was no heterogeneity in the results of RCTs on structure-modifying effects [Hochberg *et al.* 2008]. The results showed a small but significant reduction in the worsening of JSN (ES = 0.26, 95% CI 0.16–0.36). Recently, the follow-up of patients for 2 years revealed a significant effect of CS on JSN in CS-treated patients compared with placebo [Kahan *et al.* 2009].

Safety and tolerability

Most of the clinical trials reported a great safety profile and a good tolerability of CS. The frequency of side effects and the drop-off rate were equivalent in CS and placebo groups. No significant severe side effects were observed with CS. Rescue medication (acetaminophen or non-steroidal anti-inflammatory drugs [NSAIDs]) are permitted during clinical trials. This fact could diminish the difference between CS and placebo groups [Monfort *et al.* 2008a] (Table 4).

Chondroitin sulfate in guidelines

CS has been recommended in the guidelines published by OARSI and the European League Against Rheumatism (EULAR) for the management of knee OA [Zhang *et al.* 2008; Jordan *et al.* 2003]. The OARSI recommends CS for its symptomatic and structure-modifying effect in knee OA, but also recommends discontinuing it after 6 months if no symptomatic response is apparent. Concerning hip and hand OA, EULAR recommends the SYSADOA, including CS, for their symptomatic effects and low toxicity, but underlines the facts that the ESs are small, suitable patients are not well defined, and clinically relevant structure modification and pharmacoeconomic aspect are not well established.

Discussion

CS has been used for decades now and it was important to describe its pharmacokinetic, mechanism of action and to describe its clinical efficacy.

CS was shown to have various effects from anti-inflammatory and anticatabolic to anti-apoptotic, and also anti-oxidant properties. All of these results were obtained in different systems with different dosages. Many reviews have addressed the importance of CS source and purity [Lauder, 2009; Rainsford, 2009; Volpi, 2009; Lauder *et al.* 2000]. In addition, CS sulfated at different positions (e.g. 4 or 6) could have different effects. As an example, CS-4 is more effective as anti-oxidant than CS-6 [Albertini *et al.* 2000; Volpi and Tarugi, 1999]. *In vitro* and *in vivo* studies have limitations, mostly due to the concentration used and the duration of the study. CS is employed at concentrations which are largely superior to the plasmatic concentration after an oral administration of a therapeutic dose of CS. Indeed CS reached a plasma concentration of approximately 2.0 μg/ml in OA patients after oral administration of 0.8 g of CS [Conte *et al.* 1995]. This study also showed that CS can attain 2.7 μg/ml in synovial fluid of OA patients. Another experiment in rat after oral administration (16 mg/kg) showed a concentration of 3.3 μg/g in cartilage [Ronca *et al.* 1998]. Most *in vitro* studies used at least 10 μg/ml of CS and up to 1000 μg/ml (see Table 2). One has to keep in mind the conditions of the study when interpreting the data. Finally, the efficacy of CS in inflammatory conditions other than OA and mostly more severe than OA, have recently been reviewed [Vallières and du Souich, 2010; du Souich *et al.* 2009]. CS was shown to be effective in pathology including psoriasis [Möller *et al.* 2010; Verges *et al.* 2005]. This suggests a strong anti-inflammatory potency of CS. Unfortunately, the lack of *in vivo* demonstration of the effect of CS on OA progression. Only one study has investigated the effect of chondroitin on an OA model. Further research investigating the effects of CS on cartilage degradation, subchondral bone remodelling and synovium inflammation are required to better understand the clinical efficacy of CS.

CS is recommended as SYSADOA intervention for OA management by the OARSI [Zhang *et al.* 2010, 2008] and EULAR [Jordan *et al.* 2003]. Both organizations recognized its symptomatic and structure-modifying effects. OARSI recommends the discontinuation of treatment after 6 months in the absence of efficacy [Zhang *et al.* 2008]. At the time that EULAR published its guidelines, they suggested growing evidence for CS efficacy. However, by the time of the latter OARSI update [Zhang *et al.* 2010] was published, the ES has decreased, suggesting an instability of efficacy, which may be attributable, at least in part, to an increase in the quality of studies. The inconsistency of efficacy as shown in the several clinical trials is of importance. This meta-analysis

[Zhang *et al.* 2010] has also shown evidence of important publication bias. These observations are also made for other compounds of the SYSADOA class, including glucosamine sulfate (GS). In contrast, the ES for acetaminophen, although very small, had apparently stabilized with time, suggesting that further RCT to test its analgesic efficacy were not required. Altogether, this review indicates that there is a need for clinical trials of good quality with a duration and number of patients involved appropriate to this kind of compound. It is also relevant to compare the ES for pain of the main pharmacological modalities commonly used in the treatment of OA. By comparison, the ES of CS is one of the most important. For example, the ES pain (2010) for acetaminophen was 0.14 (95% 0.05–0.23), for NSAIDs 0.29 (95% 0.22–0.35) and for CS 0.75 (95% 0.50–1.01). This suggests the clinical relevance of using CS in the treatment of OA, with regards with its low toxicity. OA is a long-term progressive disease for which an appraisal of the risk/benefit ratio of each treatment should be considered before its application.

Some studies have evaluated the potency of the use of CS in combination as with GS. This was done *in vitro* [Chan *et al.* 2006, 2007, 2005a, 2005b] and also in clinical trials [Vangsness *et al.* 2009; Sawitzke *et al.* 2008; Clegg *et al.* 2006, 2005]. The synergistic effect of both compounds still has to be demonstrated but many authors recommend the use of the combination.

One reason to be optimistic about the use of CS alone or in combination with a related product such as GS is the safety profile. CS does not interact with other drugs and does not present serious adverse events. This is in favour of a positive risk/benefit ratio.

Conclusion

Despite the moderate effects of CS on pain and function, CS is an interesting product for the management of knee OA. Clinical evidence is in favour of a slow-acting effect on symptoms in moderate knee OA. CS is recommended by the most popular guidelines. Its safety profile is surely one of its main benefits for the treatment of aging patient with some comorbidity. There is then no limitation to its use in OA patients, if we ignore the economical impact. Nevertheless, caution should be exercise with regards to the type and the formulation of CS. Of course, some questions remain regarding its mechanism of action. The effect of CS on subchondral bone and synovium inflammation could be better documented.

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Conflict of interest statement

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