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OSTEOARTHRITIS and CARTILAGE

Anti-inflammatory activity of chondroitin sulfate

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Summary

The pharmacokinetics of chondroitin sulfate (CS, Condrosulf®, IBSA, Lugano, Switzerland) were investigated in rats and in healthy volunteers using CS tritiated at the reducing end and CS labeled with ^{131}I or ^{99m}Tc respectively. A rapid absorption of orally administered CS is observed in rats and in humans when the drug is dissolved in water. Lower and delayed absorption is observed when CS is administered in gastroresistant capsules. The absolute bio-availability is 15 and 12% for rats and humans respectively. The CS shows a tropism for cartilagineous tissues in rats and for knee tissues in humans as demonstrated by scintigraphic analysis with ^{99m}Tc-CS. Monomers, oligo and polysaccharides produced by enzymatic hydrolysis of CS appear in the blood and tissues together with native CS. The effects of partially depolymerized (m.m. 3 to 15 kD) and desulfated fractions on human leukocytes were investigated. CS and its fractions inhibit the directional chemotaxis induced by zymosan-activated serum, are able to decrease the phagocytosis and the release of lysozyme induced by zymosan and to protect the plasma membrane from oxygen reactive species. In rats the oral administration of CS significantly decreases granuloma formation due to sponge implants and cell migration and lysosomal enzyme release in carrageenan pleurisy. Compared with nonsteroidal anti-inflammatory drugs (indomethacin, ibuprofen), CS appears to be more effective on cellular events of inflammation than on edema formation. It is noteworthy that CS is devoid of dangerous effects on the stomach, platelets and kidneys. In synovial fluid of patients requiring joint aspiration, treated orally for 10 days with CS (800 mg/day) the hyaluronate concentration and the intrinsic viscosity significantly increased, while collagenolytic activity, phospholipase A_2 and *N*-acetylglucosaminidase (NAG) decreased.

These results give an insight into the mechanism of the anti-inflammatory and chondroprotective actions demonstrated by this drug in a number of clinical trials in patients with osteoarthritis.

Key words: Chondroitin sulfate, Osteoarthritis, Pharmacokinetic, Anti-inflammatory drug.

Introduction

Many clinical studies demonstrate the therapeutic effects of orally administered chondroitin sulfate (CS, Condrosulf®, IBSA, Lugano, Switzerland) on osteoarthritic patients with improvement of articular functions and reduction of pain [1–3]. However, some aspects of the pharmacokinetics of orally administered CS and of the mechanisms of its anti-inflammatory and chondroprotective activities require further investigation. We have studied the tissue distribution and the pharmacokinetics of labeled CS in rats, the anti-inflammatory activity in experimental animals and human leukocytes, the pharmacokinetics of oral administration of the labeled drug in humans and its effect on some biochemical parameters of synovial fluid in osteoarthritic patients.

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Materials and Methods

MATERIALS

^3H-sodium borohydride, ^3H-acetic anhydride, ^3H-arachidonate-labeled *Escherichia coli* suspension and tritiated water were from NEN DuPont (Milan, Italy). ^{131}I and ^{99m}Tc were from Sorin (Saluggia, Italy). The CS used in this experimental work was a mixture of chondroitin 4-sulfate (50%) and chondroitin 6-sulfate (50%) with an average molecular mass of 16 kDa, an $SO_3H/COOH$ ratio near to 1, devoid of anticoagulant activity. All chemicals were of analytical grade.

METHODS

The USA National Research Council's guide was followed for all research with animals. The ethical guide lines of the revised Declaration of Helsinki were followed for all research on healthy volunteers or patients.

CS was labeled at its reducing end using tritiated sodium borohydride [4]. The tritiated molecule (80 μCi/kg) was administered to rats

(240–260 g) by the intravenous (i.v.) (3.2 mg/kg) or oral (16 mg/kg) route. Plasma, urine and feces were collected at intervals, and the rats were sacrificed after 6 and 24 h for the collection of tissues and organs. After homogenization and evaluation of the total radioactivity the tissue samples were chromatographed on a Biogel P-100 (1.5×80 cm) column to evaluate the radioactivity with molecular mass (m.m.) similar to that of the administered drug which was utilized to calculate the μg of exogenous CS present in the samples.

CS was also labeled with one residue of hydroxyphenyl propionate per 70 residues of chain sugars. The molecule was then iodinated with ^{131}I [5]. The labeled and unlabeled CS had similar biological and chemical properties. The labeled CS (0.8 g in water; 50 μCi) was administered per os to four healthy volunteers. Four volunteers received 25 mg of CS (50 μCi) in gastroresistant capsules. The plasma radioactivity was fractionated on the basis of m.m. by gel filtration (Biogel P-100, 1.5×80 cm).

SPECT analyses of lower limbs as a function of the time after intravenous administration of CS-labeled ^{99m}Tc to two healthy volunteers were also carried out [6].

A foot edema test was carried out in rats using 1% carrageenan solution [7]. The method described by P. J. Bailey for rats was followed for the sponge implant test (including enzyme determination) [8]. For the pleurisy test carrageenan (0.5 mg) was injected into rats [9]. CS fractions of molecular mass (m.m.) ranging from 3 to 24 kDa were obtained with partial enzymatic hydrolysis by hyaluronidase followed by fractionation on a gel filtration column. Peripheral blood neutrophils were obtained from blood donors. Under agarose chemotaxis, phagocytosis, lysozyme and membrane protection were investigated as reported [10, 11, 12 and 13, respectively].

PATIENTS' TREATMENT

Twenty-four osteo-arthritic patients needing joint aspiration were investigated. In 12 patients (nine females), the synovial fluid parameters were determined before treatment and after 0.8 g of CS once a day for 10 days. Twelve patients (eight females) received the placebo (control sample). The mean age, body weight and height ± S.D. of the two groups of patients were 69.3 ± 7.8 years, 64.0 ± 9.1 years; 57.4 ± 6.1 kg, 55.7 ± 5.8 kg and 162.5 ± 9.4 cm, 164.2 ± 8.9 cm, respectively. The synovial fluids of all the patients were clear or straw-colored, with high viscosity, strong mucine clotting and without fibrin clotting. The osteo-arthritic patients received neither steroidal nor nonsteroidal anti-inflammatory drugs for 20 days before and during treatment. Neither had they received previous intra-articular injections. Protein concentrations were determined with Folin phenol reagent according to Lowry *et al.* [14]. Hyaluronate concentration was determined by the Tengblad method [15]. For the assay of collagenolytic activity exogenous (^3H)-bovine type II collagen was used [16]. Phospholipase A_2 (PLA_2) activity was tested as described by Pruzanski *et al.* [17]. The activity is expressed as units/ml, where one unit of PLA_2 activity is defined as the release of 1% of the radioactivity incorporated into *Escherichia coli* under the assay conditions described, and was equal to the hydrolysis of 90 pmol *Escherichia coli* phospholipid in 30 min at 37°C. *N*-acetylglucosaminidase (NAG) activity was assayed using *p*-nitrophenyl *N*-acetyl-β-D-glucosaminide as substrate [18].

To measure the intrinsic viscosity, the synovial fluids were dialyzed against a solution containing 10 mM sodium phosphate buffer pH 7.3 and 0.2 M. The viscosity was determined with a 2.0 ml Ostwald viscosimeter with a capillary diameter of 0.5 mm at a water flow time of 200 seconds in a water bath at a constant temperature of 20 ± 0.05°C as described by Kragh [19]. Appropriate dilution, depending on the hyaluronate content of the synovial fluids, was made with the buffer described above. The intrinsic viscosity was calculated following Sundblad [20].

STATISTICAL ANALYSIS

Differences in experimental results were analyzed by Student's *t*-test. Data are reported as mean ± standard deviation (S.D.). *P* values less than 0.05 were considered as significant.

Results

Table I reports the distribution in urine, feces and tissues calculated for CS and CS together with its metabolites (M) 6 h after administration. We found that 53% and 19% of the administered drug is excreted, respectively, by the kidneys in the first 24 h after i.v. and oral administration. In Table I the distribution in some tissues 6 h after administration is also reported. When tissue radioactivity is fractionated on the basis of m.m. we observed that tissue radioactivity after i.v. injection is mainly due to CS radioactivity, while after oral administration low m.m. radioactivity due to oligosaccharides, monomer and tritiated water is higher than CS radioactivity. However, with a

Table I
*Distribution and tissue concentration of chondroitin sulfate and its metabolites 6 h after administration to rats**

	Intravenous		Oral	
	CS	CS + M	CS	CS + M
Distribution				
Tissues + blood	280 ± 30	350 ± 30	300 ± 40	1830 ± 200
Urine + bladder content	230 ± 20	380 ± 40	90 ± 10	370 ± 40
Feces + intest. content	–	20 ± 10	–	1560 ± 160
Concentration				
Stomach	1.6 ± 0.5	1.7 ± 0.4	3.8 ± 1.6	12.0 ± 3.7
Small intestine	1.2 ± 0.3	1.6 ± 0.4	3.4 ± 1.6	13.3 ± 4.8
Liver	3.0 ± 0.9	6.2 ± 1.6	2.0 ± 0.8	18.1 ± 6.1
Kidneys	2.9 ± 0.7	53 ± 1.5	3.1 ± 0.9	18.5 ± 4.8
Brain	0.2 ± 0.1	0.5 ± 0.3	0.2 ± 0.1	2.5 ± 0.9
Articular cartilage	3.6 ± 0.8	4.2 ± 1.4	3.3 ± 0.9	11.5 ± 4.1
Synovial fluid	3.4 ± 0.9	3.6 ± 1.0	3.2 ± 1.0	10.7 ± 3.5
Blood	1.9 ± 0.8	2.3 ± 0.7	2.1 ± 0.7	8.4 ± 3.2

The reported values are the means ± S.D.; $n = 10$.
*The amount of chondroitin sulfate (CS) and of chondroitin sulfate plus its metabolites (CS + M) are reported in μg/g for tissues and in μg/ml for biological fluids. The amounts of tritiated chondroitin sulfate administered to rats were 3.2 mg/kg by the intravenous route or 16 mg/kg by the oral route.

dose ratio of 1:5 between i.v. and oral administration, similar quantities of CS are observed in blood, synovia, articular cartilage and many other organs. Differences are observed in the intestinal tract due to the different routes of administration used.

The high content of labeled CS observed in the synovial fluid and cartilage due to the tropism of administered CS for these tissues is noteworthy.

In Table II, the pharmacokinetic parameters determined from total radioactivity values corresponding to CS plus its metabolites are reported. It appears that CS is rapidly adsorbed in the gastro-intestinal tract. From plasma area under the curve (AUC) values vs timing, and from urinary elimination of the drug, the absolute bio-availability turns out to be 15% by oral administration.

Figure 1 reports the plasma levels of CS in humans obtained after fractionation of total radioactivity on gel columns. A rapid increase is observed after administration in water with a maximum at 1 h. The radioactivity is present to a measurable extent after 24 h. After administration in gastroresistant capsules the CS peak is observed at 4 h.

The plasma radioactivity which appears on a gel filtration column in the same position of labeled CS is about 60–70%, while about 30–40% of total radioactivity is observed in the same position of low m.m. degradation products and iodide. Similar results are obtained by the fractionation of plasma after extensive papain digestion of proteins. Table III reports some kinetic parameters of oral administration of ^{131}I-labeled CS. The values are referred to 0.8 g of administered drug.

Table II
Pharmacokinetic parameters of chondroitin sulfate (CS) and its metabolites (M) after administration of the drug to rats

	Intravenous		Oral	
Pharmacokinetic parameters	CS	CS + M	CS	CS + M
Time peak (min)	–	–	46 ± 15	635 ± 91
Peak concentration (μg/ml)	–	–	2.5 ± 0.5	8.9 ± 0.9
$t_{1/2}$ distribution (min)	14.1 ± 4.3	–	–	–
$t_{1/2}$ elimination (min)	182 ± 24	–	297 ± 88	–
Volume of central compartment (ml)	21.4 ± 3.8	38.4 ± 9.5	–	–
Volume of tissue compartment (ml)	82.1 ± 27.0	129 ± 48.3	–	–
$AUC_{0\text{-}1440}$ min administered substance (μg.ml/min)	32.8 ± 4.5	45.7 ± 5.2	28.4 ± 5.1	174.6 ± 23.0

The reported values are the means ± S.D.; $n = 10$.
CS = chondroitin sulfate; CS + M = chondroitin sulfate and its metabolites.
AUC = area under the curve.

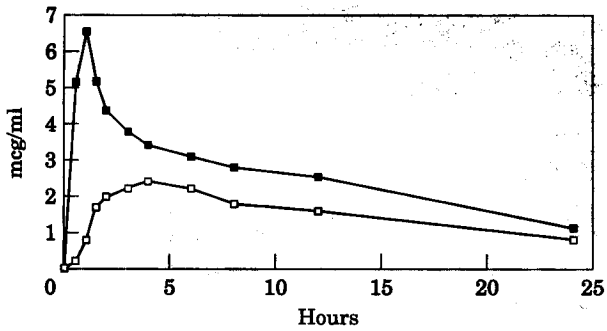

FIG. 1. Chondroitin sulfate concentration in plasma after administration to humans. The data reported are the means of plasma values of four healthy volunteers who received chondroitin sulfate in water (■) or in gastroresistant capsules (□). In both groups the concentration values are referred to 800 mg administration.

Radioactivity observed in urine consists of low m.m. derivatives or iodide. However, high m.m. CS and depolymerized CS derivatives are present in urine collected in the first hours. Total radioactivity excreted in urine after 72 h is about 19% of the administered CS for administration in water and 14% for administration in gastroresistant capsules.

About 90% of administered radioactivity is recovered in urine and feces after 72 h with both administrations.

From plasmatic and urinary values, it appears that the bio-availability as high m.m. CS is about 12% when the drug is administered in water, which is the usual administration route.

SPECT analysis of lower limbs as a function of time after intravenous administration of ^{99m}Tc as sodium pertechnate or CS labeled with ^{99m}Tc shows that radioactivity is higher during the first 40 min in the thigh and in the calf as compared with knee tissues. After this time the radioactivity progressively increases in the knee tissues and becomes much higher than in the adjacent tissues as can be seen in Fig. 2 in which the radioactivity of ^{99m}Tc sodium pertechnate and of CS labeled with ^{99m}Tc as well their difference is reported after 2 h from administration.

Table IV reports some pharmacological activities of CS in rats compared with the anti-inflammatory properties of ibuprofen and indomethacin. It appears that CS administered per os has an anti-inflammatory activity comparable to that of ibuprofen and indomethacin on the development of granuloma due to sponge implants. The drug activity has been measured after 10 h from the implant by the determination of myeloperoxidase (MPO) activity to evaluate the anti-inflammatory activity on polymorphonuclear neutrophil (PMN) infiltration of sponges and after 12 days by determination of NAG activity to evaluate macrophage infiltration. The activities in the untreated controls are 173.7 ± 35.0 and 18.4 ± 5.3 μmol/h/sponge for MPO and NAG, respectively. A comparable activity is also observed in leukocyte migration and *N*-acetylglucosaminidase release in carrageenan pleurisy determined 3 h after carrageenan administration. The leukocyte and the NAG activity in untreated controls were $95.8 \pm 19.1 \times 10^6$ and 12.3 ± 1.8 U/l, respectively. In the development of edema, CS is also active; however, its activity appears to be lower than that of ibuprofen and indomethacin. In the untreated controls the percentage increase of paw volume is 38.5 ± 3.2.

Some activities of CS fractions on human leukocytes are reported in Table V. CS fractions between 3 and 24 kDa and 12 kDa partially desulfated are devoid of chemokinetic and chemotactic activities (data not reported) and have a significant antichemotactic activity on leukocytes using human serum activated by zymosan as

Table III
Some kinetic parameters of chondroitin sulfate (CS) and its metabolites (M) after oral administration of the drug to humans

Administration	t_{max} h	C_{max} μg/ml	AUC μg.h/ml	$t_{1/2}$ h	Vd l
In water					
CS	1.0 ± 0.3	6.6 ± 0.7	86.6 ± 12.6	8.6 ± 0.7	30.3 ± 3.5
CS + M	0.5 ± 0.2	9.4 ± 1.1	121.1 ± 20.2	8.3 ± 0.9	29.7 ± 4.0
In gastroresistant capsules					
CS	4.0 ± 1.0*	2.4 ± 0.5*	47.0 ± 5.5*	9.4 ± 1.0	31.4 ± 3.9
CS + M	4.0 ± 1.1*	3.8 ± 0.6*	71.7 ± 8.2*	8.7 ± 1.1	32.0 ± 4.3

The reported values are the means ± S.D. and are referred to 800 mg administration; $n = 4$; *$P < 0.05$ vs the respective value.
CS = chondroitin sulfate; CS + M = chondroitinsulfate and its metabolites.
AUC = area under the curve.
C_{max} = maximum drug concentration in plasma after a single dose.
t_{max} = time required to reach C_{max}.

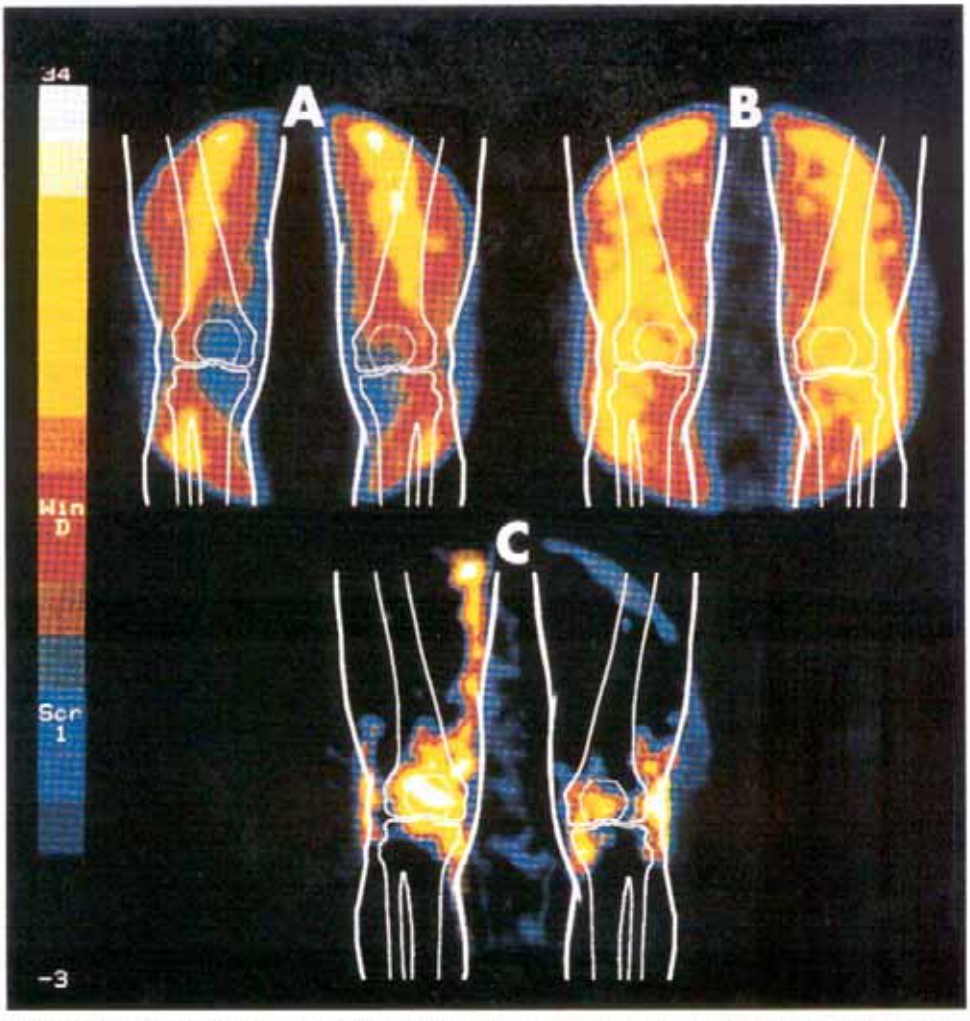

FIG. 2. (A) SPECT analysis of lower limbs 2 h after intravenous administration of ^{99m}Tc sodium pertechnate. (B) SPECT analysis of lower limbs 2 h after intravenous administration of CS labeled with ^{99m}Tc. (C) Difference between B and A.

chemoattractant. All the fractions decrease phagocytosis and lysozyme release and protect the plasma membrane from damage by reactive oxygen species. All fractions show their activity between 1 to 20 μg/ml, and this activity is dose-dependent.

The effect on some synovial fluid parameters of 10 days treatment with CS (800 mg/day) by the oral route on osteo-arthritic patients who needed joint aspiration is reported in Table VI. The concentration of hyaluronate, as well as its m.m. evaluated by viscosity, significantly increased in the synovial fluid of treated patients while protein concentration does not change. Collagenolytic, phospholipase A_2, and NAG activities are significantly decreased after 10 days of CS treatment.

Discussion

The absorption of a glycosaminoglycan drug by the oral route could depend on the length of polysaccharide chains, on the charge density and the presence of clusters with a high number of sulfate groups and on the kind of ions with which the charged groups of the drug have been neutralized.

In preceding papers, it has been shown that orally administered tritiated chondroitin sulfate is absorbed and is found in plasma as high, low and intermediate molecular weight metabolites [4, 21, 22]. Tritiated water derived from metabolization of tritiated chondroitin sulfate is also present. Absorbed radioactivity is distributed to the tissues and, in particular, to the liver, kidneys, synovial fluid and cartilage. We have fractionated the radioactive material by gel filtration columns and have calculated the chondroitin sulfate and its metabolites as a function of time from administration. It should be pointed out that tritiated chondroitin sulfate is labeled at the reducing end. The *in vivo* enzyme degradation of the polymer proceeds, step by step, from the non-reducing end

Table IV
Anti-inflammatory activity of chondroitin sulfate (CS), ibuprofen and indomethacin orally administered to rats

	CS 200 mg/kg (%)	Ibuprofen 50 mg/kg (%)	Indomethacin 5 mg/kg (%)
Oedema formation (paw vol.)			
–by carrageenin	78.7 ± 8.0*	54.0 ± 5.4*	46.7 ± 4.5*
Granuloma formation			
–by sponge			
(a) myeloperoxidase activity (10 h)	62.5 ± 5.4*	70.4 ± 7.9*	66.9 ± 5.8*
(b) *N*-acetyl glucosaminidase activity (12days)	53.6 ± 6.3*	59.0 ± 8.3*	61.0 ± 7.2*
Carrageenan pleurisy			
–Total leukocytes (number)	73.4 ± 6.3*	72.0 ± 7.0*	74.1 ± 7.9*
–*N*-acetylglucosaminidase	72.5 ± 8.3*	68.7 ± 5.4*	76.0 ± 8.0*

The reported values are means ± S.D.; $n = 10$. $*P < 0.05$ with respect to the controls.
The absolute values of the controls are reported in the test.
Myeloperoxidase and N-acetylglucosaminidase activities have been determined 10 hours and 12 days after sponge implant and are respectively proportional to the PMN and macrophages infiltration.
CS = chondroitin sulfate.

toward the reducing end. For this reason, even if enzymatic or chemical fragmentation of chondroitin sulfate takes place inside the chain, the labeled reduced sugar at the end of the polymer chain of the administered tritiated chondroitin sulfate is maintained during all the depolymerization process. We have also observed (data not reported) that only an irrelevant part (less than 1%) of the radioactivity of chondroitin sulfate is released by exchange during 48 hours incubation in rat plasma. For this reason the tritiated water that is formed derives in the greater part from metabolization of the reduced end sugars (e.g. xilitol, galactitol, . . .) only when these sugars are freed, since no enzymes are known at this moment which catalyze the re-oxidation of the sugars labeled at the reducing end of the glycosaminoglycan chains.

After administration of labeled chondroitin sulfate in water by the oral route, the time peak of CS absorption is observed at 46 and 60 min in rats and humans, respectively. These values are similar to the peak time observed in humans in which only the high-molecular weight species were determined after oral administration of unlabeled drug [21]. When CS is administered in gastroresistant capsules, the time peak is observed at 4 h and a lower quantity of CS is absorbed. Our experiments clearly demonstrate that CS is absorbed both at the gastric and the intestinal level. We suggest that the high m.m. CS reaches the blood circulation through the lymphatic system and the thoracic duct together with the lipoproteins and other macromolecules, avoiding in this way the first hepatic passage.

The AUC value and C_{max} are also comparable in rats and humans when the radioactivity is fractionated and the CS concentration is determined.

The radioactive material present in synovial fluid and in the cartilage of rats is exogenous chondroitin sulfate and poly-oligosaccharides and monomers derived from partial or complete depolymerization of the administered drug. The tropism of exogenous CS for articular tissues is also well documented by SPECT analysis of ^{99m}Tc-CS administered to humans.

Table V
Some in vitro activities of chondroitin sulfate and its fractions on human leukocytes

Chondroitin sulfate fractions (molecular mass)	Antichemotactic activity migration (mm)	Phagocytic activity (%)	Lysozyme release (%)	Membrane damage by reactive oxygen species (%)
No addition	8.6 ± 1.4	100 ± 10	100 ± 9	100 ± 8
A (> 24 kDa)	6.3 ± 1.3*	70 ± 7*	61 ± 6*	66 ± 9*
B (20 kDa)	5.7 ± 0.8*	65 ± 10*	55 ± 8*	64 ± 7*
C (12 kDa)	4.9 ± 0.7*	72 ± 9*	55 ± 11*	66 ± 10*
D (6 kDa)	4.3 ± 0.7*	68 ± 5*	60 ± 7*	70 ± 6*
E (3 kDa)	4.5 ± 0.6*	69 ± 8*	63 ± 5*	68 ± 7*
CdS (12 kDa 50% desulfated)	5.3 ± 0.8*	76 ± 6*	59 ± 9*	70 ± 8*

The reported values are means ± S.D.; $n = 6$; $*P < 0.05$ with respect to the controls.

Table VI
Synovial fluid parameters after chondroitin sulfate treatment of gonoarthrosic patients

Patients	Hyaluronate mg/ml	Hyaluronate [η]	Proteins mg/ml	Leukocytes $10^3/\mu l$	Collagenolytic activity dpm/h/ml	PLA_2 nmol/min/ml	NAG U/L
Treated ($n = 12$)							
Day0	1.8 ± 0.7	43.9 ± 5.6	31.3 ± 5.2	2.4 ± 1.5	1329 ± 257	3.1 ± 0.7	36.7 ± 5.3
Day10	2.4 ± 0.7*	49.2 ± 4.5*	30.5 ± 3.9	1.9 ± 1.5	935 ± 197*	2.2 ± 0.5*	24.3 ± 3.1*
Untreated ($n = 12$)							
Day0	1.7 ± 0.9	42.7 ± 6.1	34.2 ± 4.8	2.3 ± 1.3	1374 ± 311	2.9 ± 0.6	32.2 ± 6.2
Day10	1.9 ± 0.7	44.5 ± 5.0	33.7 ± 5.7	2.4 ± 1.4	1215 ± 230	2.8 ± 0.7	30.6 ± 4.4

The reported values are the means ± S.D.; $*P < 0.05$ with respect to day 0.
PLA_2 = phospholipase A_2.
NAG = N-acetylglucosaminidase.

The presence of exogenous chondroitin sulfate and its depolymerized derivatives in synovial fluid and cartilage is very important in explaining the anti-inflammatory and chondroprotective effects of this drug. It is also very important that the mainly CS depolymerized derivatives are active on leukocyte functions and protect cell membranes.

It appears that CS is an effective anti-inflammatory compound, although less active than indomethacin and ibuprofen on the basis of the administered quantities. However, CS is devoid of dangerous effects on stomach, platelets and kidneys, and its use may be maintained for years without relevant collateral effects. It is noteworthy that CS is more active on the cellular component of the inflammatory process, while its activity is lower on the vascular aspect. Anti-inflammatory activity is confirmed by the modification of some biochemical parameters in the synovial fluid of osteo-arthritic patients.

In conclusion, a general picture can be drawn from the data obtained in experimental animals and in humans on the metabolic fate of orally administered chondroitin sulfate. The drug is absorbed as high-molecular-mass polysaccharide together with low-molecular-mass polysaccharide chains resulting from partial depolymerization and/or desulfation.

We have also demonstrated that the orally administered drug reaches synovial fluid and cartilage and modifies some pharmacologic and biochemical markers in experimental animals and osteo-arthritic patients. The therapeutic activity of CS in osteoarthritis may be due to at least three main mechanisms which have been demonstrated up to now: (1) the anti-inflammatory activity which is more evident on the cellular aspect of the inflammation and which is devoid of the collateral effects of other anti-inflammatory nonsteroidal drugs; (2) the metabolic effects on the synthesis of hyaluronate and on the cartilage proteoglycans; (3) the direct antidegradative actions which are realized by the inhibition of some proteolytic activities (collagenase, elastase, proteoglycanase, . . .) and by the decrease of dangerous effects on matrix molecules determined by reactive oxygen species.

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