Maintenance of the synthesis of large proteoglycans in anatomically intact murine articular cartilage by steroids and insulin-like growth factor I

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Abstract

Objectives—The exact regulation of the synthesis of cartilage specific molecules, such as collagen type II and aggrecan, by articular chondrocytes is unknown, but growth factors and hormones probably play an important part. The effects of glucocorticosteroids (prednisolone and triamcinolone), in combination with insulin-like growth factor I (IGF-I), on the synthesis and hydrodynamic volume of proteoglycans from murine patellar cartilage were investigated.

Methods—The in vitro effect of IGF-I and steroids on proteoglycan synthesis in murine patellar cartilage was evaluated by [³⁵S]sulphate incorporation in combination with dissociative gel chromatography using a Sephacryl S-1000 column. The impact of in vivo prednisolone (0–5 mg/kg) on proteoglycan synthesis in murine patellar cartilage was analysed by [³⁵S]sulphate incorporation immediately after dissection from the knee joint.

Results—Prednisolone stimulated proteoglycan synthesis in murine patellar cartilage from normal knees and in cartilage from knees injected with papain in vitro in the absence and presence of IGF-I. Moreover, oral administration of prednisolone for seven days to C57B110 mice resulted in enhanced proteoglycan synthesis in patellar cartilage. The incubation of patellar cartilage for 48 hours without serum or growth factors led to the synthesis of proteoglycans with a smaller hydrodynamic volume than those synthesised immediately after dissection of the patellae. This could either be circumvented by the addition of IGF-I or by the addition of glucocorticosteroids (prednisolone or triamcinolone) to the culture medium.

Conclusions—These results show that in a dose range of 0.0003–0.3 mmol/l, glucocorticosteroids, like IGF-I, stimulate proteoglycan synthesis and maintain the synthesis of hydrodynamically large proteoglycans by chondrocytes from murine articular cartilage. This indicates that glucocorticosteroids might play a part in the preservation of matrix integrity in articular cartilage.

Articular cartilage consists of chondrocytes embedded in an extracellular matrix rich in collagen type II and the hyaluronic acid binding, cartilage specific proteoglycan aggrecan. Aggrecan gives articular cartilage its unique ability to resist compression and to undergo reversible deformation, whereas collagen provides tensile strength. Control of chondrocyte biosynthetic function is essential in the maintenance and integrity of this tissue. Disturbances in the metabolism of the articular chondrocytes, as in osteoarthritis and arthritis, will lead to the deterioration of articular cartilage and eventually to joint failure. Insulin-like growth factor I (IGF-I) is considered to be a cardinal factor in the maintenance of chondrocyte biosynthetic function. This factor appears to be as effective as serum in stimulating proteoglycan synthesis in bovine, human, and murine chondrocytes.^{1–4} Although it has been reported that IGF-I has no effect on proteoglycan degradation in bovine explants, other workers have observed considerable inhibition of proteoglycan breakdown by IGF-I in bovine and murine cartilages.^{1 5–7}

Glucocorticosteroid receptors have been shown to be present on articular cartilage chondrocytes, indicating that chondrocyte metabolism can be influenced directly by physiological and therapeutic glucocorticoids.^{8 9} The specific effects of glucocorticosteroids on chondrocyte proteoglycan synthesis are conflicting, however, and remain controversial. An inhibitory effect of glucocorticosteroids on proteoglycan synthesis has been reported for normal rat, bovine, and porcine chondrocytes^{10–12} and human osteoarthritic cartilage.¹³ In contrast, the maintenance of differentiated chondrocyte phenotype and proteoglycan synthesis are stimulated in rabbit chondrocytes by glucocorticosteroids.^{14–16} Moreover, there appears to be a synergistic action of glucocorticosteroids and IGF-I on proteoglycan synthesis in rabbit chondrocytes.¹⁷

In this study we examined the effect of steroids, in the presence and absence of IGF-I, on proteoglycan synthesis in murine patellar cartilage. Patellar cartilage from normal knees and knees injected with papain was used. The injection of papain leads to a repair response in patellar cartilage mimicking

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the repair response observed in osteoarthritic cartilage. During *in vitro* culture of normal cartilage and of cartilage from knees injected with papain prednisolone stimulated the synthesis of proteoglycans at low and moderate concentrations (0.0003–0.3 mmol/l) in the presence and absence of IGF-I. Administration of prednisolone by mouth resulted in increased proteoglycan synthesis in patellar cartilage, as measured by [³⁵S]sulphate incorporation *ex vivo*. Furthermore, steroids and IGF-I were able to maintain the synthesis of normal proteoglycans during the *in vitro* culture of patellar cartilage in the absence of serum factors.

Materials and methods

INDUCTION OF A REPAIR RESPONSE IN ARTICULAR CARTILAGE

C57B110 mice (20–25 g, 10–16 weeks old) were used in all experiments. They were fed a commercial pellet diet (RHM, Hope Farms, Linschoten, The Netherlands) and given acidified tap water *ad libitum*. The animals were kept in boxes with sawdust bedding in an air conditioned room.

The induction of an articular cartilage repair response was performed as described previously.¹⁸ In brief, the right knee joints of mice were injected once, intra-articularly, with 6 µl 5 mg/ml papain (type IV, double crystallised, 15 U/mg; Sigma, St Louis, MO, USA) in a solution of 0.03 M L-cysteine hydrochloride (Sigma) to activate the papain. Three days after the injection of papain the patellae were dissected and used in the various experiments. This protocol results in a moderate depletion of patellar cartilage proteoglycans resulting in a stimulation of proteoglycan synthesis of approximately 50% above normal values on the third day after injection.¹⁸

EFFECT OF PREDNISOLONE ON PROTEOGLYCAN SYNTHESIS *IN VITRO*

Mice were killed by cervical dislocation and the whole patellae, with a standard amount of surrounding tissue, were dissected from normal knee joints and knee joints injected with papain according to the method of van den Berg *et al.*¹⁹ Patellae were incubated in the absence and presence of a for mice physiological concentration of IGF-I (0.25 µg/ml; Boehringer, Mannheim, Germany) in RPMI 1640 DM medium (Flow Laboratories, Irvine, UK) supplemented with various concentrations of prednisolone disodium phosphate (0–3 mmol/l; Sigma). All incubations were performed for 24 hours in 24 well cluster dishes at 37°C in a humidified atmosphere containing 5% carbon dioxide. The last two hours of the culture period were carried out in the presence of 1.48 MBq [³⁵S]sulphate (Na₂SO₄; Du Pont de Nemours, Den Bosch, The Netherlands).

After incubation the patellae were washed three times with physiological saline to remove non-incorporated radiolabel and were subsequently fixed in 96% ethanol. Decalcification of the patellae with 5% formic acid was followed by

stripping of the articular cartilage layer from the remainder of the patella. Patellar cartilage was digested in Lumasolve (Perstorp Analytical, Oud-Beijerland, The Netherlands) at 60°C. The amount of incorporated [³⁵S]sulphate was measured by liquid scintillation analysis.

EFFECT OF IGF-I AND STEROIDS ON HYDRODYNAMIC VOLUME OF NEWLY SYNTHESISED PROTEOGLYCAN

Murine patellae were dissected from normal knee joints and knee joints injected with papain. The patellae were incubated for 48 hours in the presence and absence of IGF-I (0.25 µg/ml) and various concentrations of prednisolone or triamcinolone acetonide (Sigma; 0, 0.03, and 0.3 mmol/l). After incubation the patellae were cultured for four hours in RPMI 1640 DM medium containing 6 MBq [³⁵S]sulphate followed by a thorough rinsing in physiological saline. In additional experiments the labelling interval was followed by a 24 hour chase period in medium containing similar supplements to the original incubation medium.

Subsequently, the patellae were decalcified in 3.5% Na₂-EDTA (Merck, Darmstadt, Germany) for four hours and thereafter the cartilage layer was stripped from the bone. The stripped cartilage was ground with a glass mortar and pestle followed by proteoglycan extraction at 4°C for 24 hours with 4 M guanidinium hydrochloride (Merck) in the presence of protease inhibitors (0.01 M EDTA, 0.1 M 6-aminocaproic acid (Sigma), 0.005 M benzamidine hydrochloride (Sigma), 5 mg/ml trypsin inhibitor (Sigma), 0.005 M iodoacetate (Sigma)). This extraction procedure resulted in the extraction of over 90% of the incorporated radiolabel.

Extracted proteoglycans were supplemented with 1 mg of an unlabelled bovine proteoglycan mixture as a carrier and applied to a Sephacryl S-1000 column (1.6 × 100 cm, Pharmacia, Uppsala, Sweden). The column was equilibrated and eluted with 4 M guanidinium chloride (dissociative). The flow rate was 6 ml/h and fractions of 3 ml were collected. The fractions were assayed for radioactively labelled proteoglycans using a liquid scintillation counter. All runs were checked for reproducibility by comparison of the elution pattern of the unlabelled bovine carrier proteoglycans of different runs. All runs of one particular experiment were performed on the same column.

EFFECT OF *IN VIVO* PREDNISOLONE ADMINISTRATION ON PROTEOGLYCAN SYNTHESIS

Male C57Bl mice (12–16 weeks) were injected in the right knee joint with 6 µl of 5 mg/ml papain as described earlier. The left knee was injected with a similar volume of physiological saline. Prednisolone (0–5 mg/kg) dissolved in tap water was administered by mouth daily to the mice for a period of seven days, starting one day after the injection of papain. Control animals received a comparable volume of tap

water only. Groups of mice were matched for weight at the start of the experiment.

Mice were killed by cervical dislocation and the patellae were dissected. Patellae were cultured for two hours in medium containing 1.48 MBq [35 S]sulphate. After incubation the patellae were washed three times with physiological saline to remove non-incorporated [35 S]sulphate and were fixed in 96% ethanol. Decalcification of the patellae with 5% formic acid was followed by stripping of the articular cartilage layer. Patellar cartilage was digested in Lumasolve at 60°C. The amount of incorporated [35 S]sulphate was measured by liquid scintillation analysis.

STATISTICS

Statistical evaluation of the *in vitro* experiments was tested by one-way analysis of variance in combination with Student's *t* test. The *in vivo* experiments were evaluated by the Kruskal-Wallis test in combination with the Wilcoxon test. A *p* value less than 5% was considered significant.

Figure 1 Effect of prednisolone on the incorporation of [35 S]sulphate in patellar cartilage from normal joints. Patellae were incubated in the absence and presence of insulin-like growth factor I (IGF-I) (0.25 μ g/ml) with various concentrations of prednisolone for 24 hours. The last two hours of the culture period were carried out in the presence of [35 S]sulphate. Results are the mean (SEM) of 21 samples (**p* < 0.05). Statistical significance was analysed with respect to the control group after one way analysis of variance.

Results

EFFECT OF PREDNISOLONE ON PROTEOGLYCAN SYNTHESIS *IN VITRO*

Immediately after dissection the proteoglycan synthesis in patellar cartilage of knee joints injected with papain, measured by [35 S]sulphate incorporation, was 55% higher than in the patellar cartilage of the control joints injected with saline. *In vitro* this enhanced proteoglycan synthesis was lost within 24 hours. The addition of IGF-I to the culture medium resulted in a significantly increased proteoglycan synthesis in normal cartilage and cartilage from knee joints injected with papain compared with proteoglycan synthesis without IGF-I (approximately 90%). The proteoglycan synthesis in the presence of IGF-I was comparable with the *in vivo* level of proteoglycan synthesis (data not shown).

Prednisolone had a significant stimulating effect on patellar proteoglycan synthesis in the absence and presence of IGF-I during a 24 hour incubation period (figs 1 and 2). A linear dose-response relation was not observed, but prednisolone had a Gaussian-like effect on the synthesis of proteoglycans. In the absence of IGF-I, concentrations ranging from 0.0003 to 0.3 mmol/l had a significant stimulating effect on proteoglycan synthesis in patellar cartilage from normal knees and knees injected with papain (maximum 47%). In the presence of IGF-I, a stimulating effect of prednisolone was observed at 0.003 and 0.03 mmol/l with normal patellae and at 0.0003, 0.003, and 0.3 mmol/l with patellae from murine knee joints injected with papain. A concentration of 3 mmol/l led to neither a significant stimulation nor inhibition of proteoglycan synthesis.

EFFECT OF IGF-I AND STEROIDS ON HYDRODYNAMIC VOLUME OF NEWLY SYNTHESISED PROTEOGLYCANS

The culture of patellae from normal knee joints for 48 hours in medium free of serum and growth factor resulted in the synthesis of proteoglycan monomers with a smaller hydrodynamic volume than the proteoglycans synthesised immediately after dissection (*ex vivo* labelling; fig 3). The addition of IGF-I to the medium maintained the synthesis of proteoglycans with a hydrodynamic volume similar to those synthesised directly after dissection. These results indicate that the addition of IGF-I to the culture medium results in the synthesis of 'normal' patellar cartilage proteoglycans during *in vitro* incubation. In this context normal means similar to the proteoglycans synthesised *ex vivo*. The murine proteoglycan monomer (Kav 0.43) synthesised immediately after dissection appeared to be smaller than monomers from bovine articular cartilage (Kav 0.25, data not shown).

The effect of prednisolone mimicked the effect of IGF-I on the hydrodynamic volume of newly synthesised proteoglycans (fig 4). In cultures with no IGF-I the addition of 0.03 or 0.3 mM prednisolone led to the production of

Figure 2 Effect of prednisolone on the incorporation of [35 S]sulphate in patellar cartilage from knee joints injected with papain. Patellae were incubated in the absence and presence of insulin-like growth factor I (IGF-I) (0.25 μ g/ml) with various concentrations of prednisolone for 24 hours. The last two hours of the culture period were carried out in the presence of [35 S]sulphate. Results are the mean (SEM) of 21 samples (* p <0.05). Statistical significance was analysed with respect to the control group after one way analysis of variance.

proteoglycans with a hydrodynamic volume comparable with those synthesised *ex vivo*. In the presence of 0.03 mmol/l, however, some of the synthesised proteoglycans were still smaller than the proteoglycans produced normally. Prednisolone had no effect on the hydrodynamic volume of the proteoglycans in the presence of IGF-I (fig 4).

As expected, the effect of prednisolone on the dimension of newly synthesised proteoglycans did not appear to be specific to prednisolone. Triamcinolone acetate had effects analogous to prednisolone on the hydrodynamic volume of newly synthesised proteoglycans (fig 5). Concentrations of 0.03 and 0.3 mM triamcinolone enhanced the synthesis of hydrodynamically large proteoglycans by patellar cartilage cultured in the absence of IGF-I (fig 5A). In the presence of IGF-I, no effects of triamcinolone were detectable on the proteoglycan hydrodynamic volume (fig 5C).

During a 24 hour chase period approximately 50% of the proteoglycans was lost in the absence of IGF-I, whereas this was only 20% in the presence of IGF-I. Triamcinolone had no effect on proteoglycan loss during the 24 hour chase period. The

Figure 3 Effect of *in vitro* culture on hydrodynamic volume of newly synthesised proteoglycans of murine patellar cartilage in the presence and absence of insulin-like growth factor I (IGF-I). Comparison with proteoglycans synthesised *ex vivo*. Proteoglycans were radiolabelled with [35 S]sulphate for four hours immediately after dissection or after 48 hours of incubation with or without IGF-I. Proteoglycans were applied to a Sephacryl S-1000 column and eluted under dissociative conditions. * = *Ex vivo*; + = with IGF-I; • = without IGF-I. One representative experiment of four is shown.

Figure 4 Effect of prednisolone on hydrodynamic volume of newly synthesised proteoglycans of normal patellar cartilage, in the presence and absence of insulin-like growth factor I (IGF-I). Proteoglycans were radiolabelled with [35 S]sulphate for four hours after 48 hours of incubation with prednisolone. Proteoglycans were applied to a Sephacryl S-1000 column and eluted under dissociative conditions. • = 0 mmol/l; + = 0.03 mmol/l; and * = 0.3 mmol/l prednisolone. One representative experiment of four is shown.

chromatographic profiles obtained immediately after labelling and after a 24 hour chase period were essentially similar (fig 5B and D). Only a slight shift to a larger hydrodynamic volume of the proteoglycans was observed.

Figure 5 Effect of triamcinolone acetate on hydrodynamic volume of newly synthesised proteoglycans of normal patellar cartilage in the presence and absence of insulin-like growth factor I (IGF-I). Proteoglycans were radiolabelled with [35 S]sulphate for four hours after 48 hours of incubation with triamcinolone. Part of the patellae was subsequently incubated for another 24 hour (chase). Proteoglycans were applied to a Sephacryl S-1000 column and eluted under dissociative conditions. ● = 0 mmol/L; + = 0.03 mmol/L; and * = 0.3 mmol/L triamcinolone acetate.

This is probably caused by a preferential loss of relatively small proteoglycans to the culture medium. During the chase period the chromatographic pattern was not changed from an 'IGF-like' to a 'non-IGF-like' pattern, indicating that the effects of steroids and IGF-I on the production of the two different proteoglycan populations was an effect on proteoglycan synthesis and not to be mediated by a decreased proteoglycan breakdown. In a separate set of experiments of patellae incubated without IGF-I and steroids and without a chase period, variation of the proteoglycan labelling interval from one to four hours resulted in similar chromatographic patterns, illustrating that the effect of steroids and IGF-I was on newly synthesised proteoglycans instead of being caused by steroid and IGF-I induced alterations in proteoglycan degradation (data not shown). If proteoglycan breakdown affected the differences observed between IGF-I/steroid and non-IGF/steroid chromatographic patterns a change in the elution profile dependent on the labelling period would be expected in the absence of IGF/steroids.

Patellar cartilage from knee joints injected with papain responded in a similar manner to IGF-I and steroids as cartilage from normal knee joints (fig 6). Insulin growth factor I and triamcinolone maintained the synthesis of hydrodynamically large proteoglycans during a 48 hour culture period. In the presence of IGF-I triamcinolone had no additional effects on the dimensions of the newly synthesised proteoglycans.

Figure 6 Effect of triamcinolone acetate on hydrodynamic volume of newly synthesised proteoglycans of patellar cartilage from knee joints injected with papain in the presence and absence of insulin-like growth factor I (IGF-I). Proteoglycans were radiolabelled with [35 S]sulphate for four hours after 48 hours of incubation with triamcinolone. Proteoglycans were applied to a Sephacryl S-1000 column and eluted under dissociative conditions. ● = 0 mmol/L; + = 0.03 mmol/L; and * = 0.3 mmol/L triamcinolone acetate.

EFFECT OF IN VIVO PREDNISOLONE ADMINISTRATION ON PROTEOGLYCAN SYNTHESIS

Knee joints injected with papain showed a significantly enhanced (36%) incorporation of radiolabelled sulphate in the patellar cartilage eight days after injection. Administration of prednisolone by mouth resulted in a significantly increased synthesis of proteoglycans at all doses (0.5–5.0 mg/kg) in normal cartilage and in cartilage with a repair response (fig 7). A dose-response effect of prednisolone on proteoglycan synthesis could not be shown in the dose range studied.

Discussion

Insulin-like growth factor I stimulated the synthesis of proteoglycans in murine patellar cartilage, as measured by [³⁵S]sulphate incorporation, and maintained the synthesis of normal (ex vivo-like) proteoglycans during in vitro culture. No differences were detected between normal cartilage and cartilage from knee joints injected with papain, indicating that cartilage with a subnormal proteoglycan

concentration and an enhanced proteoglycan synthesis rate was as responsive to IGF-I as normal cartilage. The hydrodynamic volume of the subpopulation of large proteoglycans of murine patellar cartilage appeared to be smaller than that of proteoglycans from bovine articular cartilage (our results, data not shown). This appears to be a general phenomenon. A smaller size of murine monomers compared with monomers from comparable tissues of larger species has been reported for proteoglycans from murine articular cartilage and from murine intervertebral disc.^{20, 21} The exact nature of the small molecular weight proteoglycans synthesised by patellar cartilage cultured without IFG-I, probably biglycan or decorin, is under study.

Other workers have reported that IGF-I preferentially stimulates the synthesis of large proteoglycans by chondrocytes. In experiments with rat rib growth plate chondrocytes, incubation with IGF-I resulted in an increased size of the newly synthesised proteoglycan monomers.²² In contrast, other growth factors such as epidermal growth factor and fibroblast growth factor favoured the synthesis of small proteoglycans.²² In bovine articular cartilage explants, IGF-I only stimulated the synthesis of the large proteoglycan subpopulation, but had no effect on the synthesis of small proteoglycans.²³ These observations suggest that IGF-I is able to prevent the de-differentiating effect of prolonged in vitro incubation when judged by the synthesis of proteoglycans.

Incubation of patellar cartilage for 24 hours with rather high concentrations of prednisolone (0.0003–0.3 mmol/l) resulted in a stimulation of [³⁵S]sulphate incorporation and only at a concentration of 3 mmol/l was no stimulation or only a slight, insignificant inhibition seen. With respect to the rate of proteoglycan synthesis, no significant difference in prednisolone effects was observed between cultures with or without IGF-I, neither with normal cartilage nor with cartilage from knees injected with papain. Moreover, in vivo administration of prednisolone up to the highest dose tested (5 mg/kg) led to a significant stimulation of ex vivo proteoglycan synthesis in cartilage from normal knees and knees injected with papain. This is in agreement with the observation that proteoglycan synthesis in isolated rabbit chondrocytes was stimulated by glucocorticosteroids.^{14–16} In earlier studies with anatomically intact murine cartilage it was found that 0.3 mM prednisolone had no significant effect on proteoglycan synthesis in normal cartilage, but stimulated proteoglycan synthesis in trypsin depleted cartilage and in cartilage from knee joints injected with zymosan.²⁴

In contrast with these observations, an inhibiting effect of glucocorticosteroids on proteoglycan synthesis in articular cartilage has been shown by others. Dekel *et al* observed an inhibitory effect of hydrocortisone, at a concentration of 3 mmol/l and higher, on proteoglycan synthesis in cultured pig costal cartilage discs.¹⁰ In a minority of experiments,

Figure 7 Effect of oral administration of prednisolone for seven days on the ex vivo proteoglycan synthesis of patellar cartilage from normal knee joints and knee joints injected with papain. The right knee joint was injected with 6 μ l 5 mg/ml papain whereas the left knee joint was injected with a similar volume of physiological saline. Prednisolone (0–5 mg/kg) dissolved in tap water was given by mouth to the mice daily for a period of seven days, starting one day after the injection of papain. Control animals received a comparable volume of tap water only. After dissection patellae were incubated for two hours in [³⁵S]sulphate. Results are means (SEM) of 27–35 samples. (A) Normal joints; (B) joints injected with papain. **p* < 0.05. Statistical significance was analysed with respect to the control group after one way analysis of variance.

however, a stimulating effect of hydrocortisone was seen.¹⁰ In vitro culture with hydrocortisone decreased [³⁵S]sulphate incorporation in rat rib cartilage and human osteoarthritic cartilage, whereas dexamethasone had similar effects on pig cartilage.^{12 13 25} Administration of prednisolone in rabbits and intra-articular injection of methylprednisolone in horses led to a decreased proteoglycan content and synthesis in articular cartilage.^{26 27} In mice, intra-articular injection of rimexolone or triamcinolone hexacetone suppressed proteoglycan synthesis in normal cartilage but, remarkably, increased proteoglycan synthesis in cartilage from arthritic knee joints.²⁸⁻³⁰

The background to these controversial effects remains obscure, but differences in steroid concentrations, low concentrations stimulating proteoglycan synthesis and high concentrations inhibiting proteoglycan synthesis, could be of major importance. In our hands moderate concentrations (3 mM prednisolone in vitro, 5 mg/kg by mouth) had no deleterious effects on proteoglycan synthesis. Species differences appear not to be the determining factor as stimulating effects of steroids have been observed in murine, porcine, and rabbit cartilage. Steroid concentration in combination with the species studied could be of major importance, however. Also, the environment of the cartilage, normal or arthritic knee joint, seems to influence the effect of steroids on chondrocyte metabolism.²⁸⁻³⁰

Surprisingly, in anatomically intact patellar cartilage glucocorticosteroids not only had an increasing effect on proteoglycan synthesis but also preferentially stimulated the synthesis of proteoglycans with a large hydrodynamic volume. Corresponding effects have been observed by Kato and Gospodarowicz with isolated rabbit costal chondrocytes.¹⁵ Rabbit chondrocytes maintained in glucocorticoid-free medium produced high and low molecular weight proteoglycans, whereas chondrocytes in hydrocortisone supplemented medium synthesised almost only proteoglycans with a high molecular weight.¹⁵ Our results show that glucocorticosteroids, like IGF-I, preserve the differentiated chondrocyte phenotype, not only in cultures of isolated chondrocytes but also in anatomically intact cartilage.

Steroids appear, like IGF-I, to be able to maintain the synthesis of normal proteoglycans in cartilage from normal knees and in cartilage in a repair phase. No deleterious effects on proteoglycan synthesis in murine cartilage could be shown in this study, neither in vitro nor in vivo. These results indicate that glucocorticosteroids could play a part in the maintenance of the homeostasis and integrity of normal and disease affected articular cartilage. Full understanding of glucocorticosteroid effects on articular cartilage demands further research, however.

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