THE EFFECT OF HYALURONAN INJECTIONS DURING THE DEVELOPMENT OF OSTEOARTHRITIS

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[Introduction] We previously reported the chondroprotective effect of hyaluronan (HA) (MW=8x10^5, Seikagaku Corp, Tokyo) on the rabbit knee. Chondroprotective effect was observed utilizing a series of 5 weekly injections of HA, as used clinically, following 4 weeks of OA development. The purpose of this study was to compare the effect of the number of injections (1, 3 and 5) of HA on OA induced by ACLT in a rabbit joint.

[Materials and Methods] Sixty New Zealand white rabbits (6 groups of 10 animals) underwent unilateral ACLT. Contralateral knees were used as unoperated control. The groups of animals with 5 injections of HA (5HA), 3 injections (3HA), and 1 injection (1HA) received intraarticular injections of 0.3 ml of HA (10 mg/ml) into the operated knee once a week beginning 4 weeks post surgery as described in Figure 1. The groups of animals with 5 injections of vehicle (sterile phosphate buffer saline) (5VA), 3 injections (3VA), and 1 injection (1VA) received intraarticular injections of 0.3 ml of vehicle into the operated knee once a week as described in Figure 1. All animals were sacrificed at 9 weeks. At sacrifice gross morphology of the femoral condyle and tibial plateau was assessed by the Outerbridge classification modified by Yoshioka (i.e. grade 1 intact; grade 2 min fibrillation, grade 3 overt fibrillation, grade 4a erosion ≤ 2 mm, grade 4b 2 mm < erosion ≤ 5 mm, grade 4c erosion > 5 mm. For histological studies, 5 µm sections were cut and stained with H&E and safranin O/fast green. Cartilage area was assessed histomorphometrically. Biochemically, tissue cellularity of the synovium was assessed by the method described by Amiel et al.3) All results were subjected to statistical analysis using ANOVA with a level of significance of p=0.05.

[Results] Gross morphology: Table 1 illustrates the grading classification of the femoral and tibial articular cartilage following the 5, 3 and 1HA, and the 5, 3 and 1VA injections. For the femoral condyles with 5HA injec, 20% (2 rabbits) showed grade 1, whereas no 5VA rabbits had grade 1. In the tibial plateau with 5HA injections, 30% showed grade 1 as compared to 10% of the 5VA group. In the femoral condyle of the 3HA injection group only one grade 4 was observed, while three 3VA (30%) rabbits showed grade 4c. Two of the rabbits in the femoral 3HA injection group had grade 1, while no 3VA rabbits showed grade 1. In the tibial plateau, 60% of the 3HA showed grade 1, while only 40% of the 3VA did. In the femoral condyle single injection group, grade 4 OA was seen in 40% of the 1HA and 50% of the 1VA. In the tibial group, only one of the 1HA showed grade 4 OA while three (30%) tibiae of the 1VA showed grade 4.

Histomorphometry of the Cartilage Area: In the 5-injection study, the 5HA group showed a significant effect on the cartilage area preservation when compared to the 5VA. The cartilage area of the 3 injections was not statistically different in the HA injections when compared to the vehicle injection group. In the one injection group no statistical differences were found between the 1HA and 1VA.

[Discussion] HA injections, i.e. 5, 3 and 1, have demonstrated a positive effect on the development of OA in the rabbit ACLT model. Inflammation during the development of OA following HA treatment was reduced by decreased cellularity of the synovial lining, as well as by reduced thickening of the synovial layer. The semiquantitative histomorphometric data demonstrated an optimal effect by reduction of degradation of the articular cartilage matrix during the development of OA when a 5-injection HA treatment was performed, unlike the 3 and 1 injections.


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