C—3 ANALYSIS OF GLYCOSAMINOGLYCANS IN THE CELLULAR MICROENVIRONMENT OF CHONDRON

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Chondrons consist anatomically of a chondrocyte surrounded by a proteoglycan rich pericellular matrix, and encapsulated within fine fibrous collagens (1,2). The proteoglycans in chondrons were unextractable with GuHCl. This study presents a histochemical and a chemical profile of the soluble and the insoluble proteoglycans extracted with 4M GuHCl from chondron rich fractions obtained by homogenization and filtration techniques (2).

MATERIAL AND METHODS

Fresh cartilage sample were resected at the tidemark from the medial and lateral tibial plateaux of mature crossbred dogs. The samples from each dog were diced into chips, and suspended in PBS, then low speed homogenized with a Politron grinder at 4000-6000 rpm. The supernatants were filtered through a series of nylon filters (1,2). Material which was trapped on the 400 to 1000 μm filters was termed the chondron rich fraction (CRF). The soluble proteoglycans (SF) were extracted from CRF with 4M guanidine-HCl. The insoluble materials were studied as the chondron fraction (CF). The glycosaminoglycans in the whole cartilage (WC), SF and CF were isolated by pronase digestion after β-elimination (3). The GAG in these fractions and their composition were analyzed (3).

RESULTS AND DISCUSSION
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The GAGs in each sample were composed of chondroitin sulphate (CS) as a major component, and keratan sulphate (KS) and hyaluronic acid (HA) as minor components. The amounts of CS in CF were smaller than those in WC and in SF. Conversely, the HA content in CF was significantly higher than those in WC and SF as shown in the figure. The isomers of chondroitin sulphate, chondroitin 4-sulphate (C4S) and 6-sulphate (C6S) were found in all fractions and 65% of the CS was the C6S in CF. These results indicate that the chondrocytes in the chondron are surrounded by a specialised microenvironment.

![Diagram showing glycosaminoglycan components](image)

Fig. 1. Glycosaminoglycan components in the whole cartilage, the soluble proteoglycans and the chondron fraction

REFERENCES