CHANGES WITH AGING AND OSSIFICATION IN THE PROTEOGLYCAN FROM HUMAN LUMBAR YELLOW LIGAMENT

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INTRODUCTION

Focusing on changes in proteoglycan in the ligament matrix during ligamentous ossification, we used yellow ligaments to biochemically analyze glycosaminoglycan (GAG) or components of proteoglycan. This paper presents the analysis of changes in GAG arising from aging and degeneration and comparative evaluation of our analytical findings from cases of ossified yellow ligaments.

MATERIALS AND METHODS

A total of 72 yellow ligament specimens were collected from patients (31 male, 20 female) who had undergone spinal surgery and autopsy. The age of the patients ranged from 15 to 81 years, with 45.9 the average. Removed specimens were divided into two blocks. In one, sagittal pathologic tissue preparations were stained with H.E. and toluidine blue (pH 4.1) to observe the extent of ligament degeneration. In the other block, GAS was biochemically analyzed.

For the biochemical analysis, the materials was digested with protease to become dissoluble and treated with alkali and then TCA. After centrifugation the supernatant was partially purified by gel filtration and crude GAG was extracted.

Ethanol precipitation was followed by chondroitinase ABC and chondroitinase AC II digestion and the unsaturated disaccharide was purified. On an ion-exchange column YMC/PA-03 for high-performance liquid chromatography (HPLC), NaH₂PO₄ was eluted at a linear concentration gradient of 16–800 mM.

RESULTS AND DISCUSSION

Based on the histopathological findings, ligament specimens were divided into three groups: group A - the normal aging group, in which ligamentous tissue remained practically normal or fiber tissue had degenerated slightly; group B, in which fiber tissue had markedly degenerated and chondroid cells emerged; and group C, in which ossification was apparent. Thirty-four cases (47 specimens) were group A, 12 cases (17 specimens) group B, and 5 (8 specimens) group C. Graphs of the age-based averages of the rates of unsaturated disaccharide content in group A, the normal aging group (Fig. 1),
revealed that dermatan sulfate (DS) exceed 40% in all the younger patients and tended to decrease with age as chondroitin 6-sulfate (Ch-6S), hyaluronic acid (HA), and chondroitin (OS) increased. An inter-group comparison of the unsaturated disaccharide content rate based on ligamentous degeneration and ossification progress in those aged 50 or higher is made in the graph (Fig. 2).

Group B and C had higher Ch-6S and chondroitin 4-sulfate (Ch-4S) content than did group A but lower DS content. This tendency was most pronounced in group C.

As noted above, a marked percent increase in Ch-6S and Ch-4S content was observed in cases where chondroid cells appeared in the ligaments, especially when the ligament had ossified. As the chemical composition of the ligament in these cases differed significantly from that in the normal aging group, it is possible that ligamentous ossification is closely associated with the emergence of chondroid cells and the resulting accelerated synthesis of proteoglycan that is mainly composed of Ch-6S and Ch-4S.

REFERENCE