The Compliance of Individual Collagen Fibrils of Osteoarthritic Cartilage Is Inferior To Intact Cartilage

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INTRODUCTION

It was found that collagen fibrils' disruption of articular cartilage occurs ahead of proteoglycan loss and cartilage erosion in the initiation of osteoarthritis (OA) [1, 2]. As previously reported in *Nature Nanotechnology*, the breakdown of collagen fibrils' meshwork in OA cartilage was detected by atomic force microscopy (AFM). Yet few attentions were paid to mechanical properties of individual collagen fibril, which was essential for the mechanical properties of collagen fibrils' meshwork. This study aimed to apply indentation-type AFM to compare the Young's modulus of individual collagen fibrils from osteoarthritic articular cartilages with intact articular cartilage from hip fracture.

MATERIALS AND METHODS

Sample preparation

The fresh specimens of articular cartilage were collected with informed consent from postmenopausal women, who underwent arthroplasty for either the end-stage of osteoarthritis (OA) or hip fracture. The articular cartilage from hip fracture served as the age-match control. The fresh specimens were cryo-sectioned with 500μm for each section from articular surface to the calcified zone. Afterward, all the specimens were dehydrated with alcohol with same protocol and store at 4°C before indentation tests.

Experimental details

The atomic force microscope (AFM) (Solver Pro47 SPM; NT-MDT company, Russia) was used in this study. The NSG20 indentation tips were used for indentation test, which tip radius was less than 10 nm. The collagen fibrils were located by AFM imaging under tapping mode, and the nanoindentation tests were performed on the overlap region of collagen fibrils. A total of twenty indentations were performed for each section of the specimens under ambient condition with indentation depth around 20 nm.

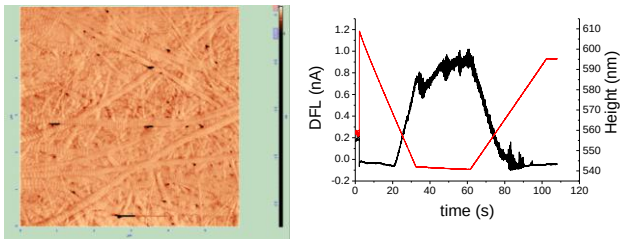

Fig. 1 Typical AFM image of collagen fibrils (left) and Typical DFL and Height curves obtained from AFM indentation tests performed on single collagen fibril (right).

Analysis method

A newly proposed protocol proposed recently[3], which enable the tip-cantilever properties to be calibrated and the viscoelastic effects during the indentation tests to be well addressed, was employed to analyze the DFL and height data obtained from the AFM indentation tests. The key equation is

$$\frac{\Delta\dot{\delta}}{\Delta\dot{D}} = A(1+\frac{a}{E_r}) \quad (1)$$

Where $\Delta\dot{\delta}$ and $\Delta\dot{D}$ are the height rate jump and the DFL signal rate jump respectively. E_r is the reduced modulus, A is the cantilever sensitivity and a is a constant represent the tip-cantilever property. By analyzing the DFL-Height curves of AFM indentation tests performed on two samples with known reduced modulus (in this study, a PP and a fused quartz sample were used), the cantilever sensitivity A and the tip-cantilever constant can be determined. The calibrated A and then can be used to calculate the reduced modulus of collagen fibril based on the experimental data of performed indentation tests.

RESULTS

(1) With the increase of the diameter of collagen fibrils, the reduced modulus of individual collagen fibril also increased from the superficial zone (2.26±0.31GPa), transition zone (2.62±0.14 GPa) to deep zone (3.44±0.21 GPa) of intact cartilage;

(2) The collagen fibrils between remaining cartilage from OA appeared thicker and stiffer (4.20±0.31 GPa) than those from the deep zone of intact cartilage (Fig.2.)

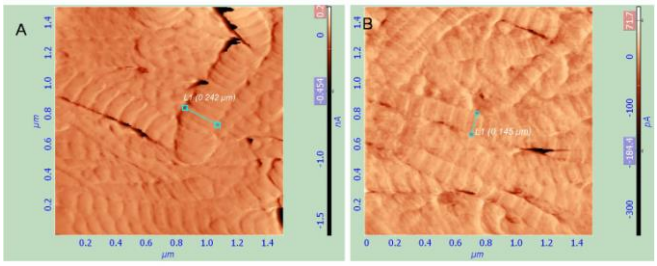

Fig.2. the diameter of individual collagen fibril of remaining cartilage in OA (A) was larger than that from the deep zone of intact cartilage.

CONCLUSION

Atomic force microscopy enabled us to address the disparity in the mechanical properties of individual collagen fibrils between OA cartilage and intact cartilage. The drop in the compliance of collagen fibrils contributed to the deterioration of articular cartilage degeneration in OA.

REFERENCE

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