**Introduction:**

Rotator cuff tendon (RCT) is the primary dynamic stabilizer of the shoulder joint and is placed under significant stress during overhead and contact sport. Rupture of the RCT results in significant pain and difficulty with overhead activities. In clinical practice, the tendon rarely heals without surgical reposition. RCT degenerative change is a major cause of RCT rupture. However, the process of tendon degeneration is poorly understood. Little attention has been paid to the pathogenesis of tendonopathy from a cellular and molecular perspective. Apoptosis, or programmed cell death, is a physiological process contributing to the control of cell population. However, excessive apoptosis can promote tissue degeneration. The presence or absence of apoptosis has not been evaluated in tendonopathies. The aim of this study, therefore, was to investigate the involvement of apoptosis in the pathogenesis of RCT disorders.

**Materials and methods:**

Twenty-five RCT samples were collected from the surgery of RCT repair for patients suffering from RCT rupture. Six subscapularis tendon tissues were collected from patients without RCT rupture history as controls. Patients’ medical history were collected and analyzed. DNA fragmentation assay was undertaken on tendon paraffin sections by the DNA End Labelling method. With this method apoptotic cells stain dark brown in nuclei due to intensive DNA fragmentation. The percentage of apoptotic cells was assessed by blinded investigators. Statistical analysis was performed using Students t-tests.

DNA laddering assay was performed to detect the DNA laddering pattern characteristic for apoptosis. DNA was extracted from tendon tissue, labeled with 32P-dATP, fractionated by agarose gel electrophoresis, and exposed to X-ray film. Cell markers for macrophages and fibroblasts were used to identify cell types in tendon tissue.

**Results:**

A large number of apoptotic cells (arrows) were found on sections of ruptured RCT tissue as shown in Fig.1-a & b, where degenerative changes were obvious. In control tendon tissue, cells were not stained (Fig. 1-c). The percentage of apoptotic cells in degenerative RCT tissue was significant higher than that in control tendon (34% vs 13%, p<0.001, Fig.2). Excessive apoptosis detected in degenerative RCT was confirmed by DNA laddering assay. Tendon tissue with excessive apoptosis showed clear DNA laddering on agarose gel (Fig.3, lane2,3,4). DNA extracted from control tendon did not show this pattern (Fig.3, lane1).

Immunohistochemical staining was performed to identify cell types undergoing apoptosis using macrophage and fibroblast markers. It was clearly shown that the majority of apoptotic cells were fibroblasts (Fig.1-d, arrows).

**Discussion:**

1. We are the first to reveal that excessive apoptosis is present in rotator cuff tendon disorders.
2. Cells undergoing apoptosis in rotator cuff tendon are mainly fibroblasts. Excessive apoptosis may play a role in the pathogenesis of tendon degeneration.

3. Further studies are required to elucidate if the excessive apoptosis of tendon fibroblasts affects the functions of the cells (e.g. collagen synthesis), and if inhibition of apoptosis would prevent or inhibit tendon degeneration.

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**Reference:**