Role of endoplasmic reticulum stress and the interaction between the endoplasmic reticulum and mitochondria in the development of tendinopathy

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Introduction: The most common sites of tendinopathy are the rotator cuff, Achilles tendon, and patellar tendon. Symptoms include local pain and joint dysfunction. Pathological changes include aseptic tendon inflammation, collagen structure degradation, and weakening of biomechanical properties. Research on tendinopathy has progressed, but a comprehensive understanding of its pathological process is still limited. Supraspinatus tendinopathy (SST) is often associated with rotator cuff impingement syndrome. The mouse model recently established by Cong et al. simulates this condition more accurately than traditional collagenase injection. Oxidative stress is crucial in the development of tendinopathy, which is characterized by increased reactive oxygen species (ROS) and decreased superoxide dismutase (SOD), leading to mitochondrial damage.

Mitochondria and endoplasmic reticulum (ER) are key organelles for ATP and protein synthesis, and they are interconnected in metabolism, apoptosis, and autophagy. The ER regulates calcium storage and cooperates with mitochondria to affect apoptosis. This study explored the role of ER stress and the coupling of ER and mitochondrial calcium in the pathogenesis of rotator cuff tendinopathy.

Methods: A total of 65 male wild-type C57BL/6 mice (n=110 shoulders) were used in this study. This study was approved by IACUC. Fifty-two mice were used to establish a supraspinatus tendinopathy model induced by subacromial impingement. After anesthesia, the subjects underwent bilateral shoulder surgery. Titanium microchips were implanted under the bilateral subacromial process and firmly fixed to the periosteum on the acromion surface. The subjects in the supraspinatus tendon (SST) impingement model were divided into four groups according to the time interval after impingement: 2 weeks, 4 weeks, 6 weeks, and 8 weeks. Sixteen normal mice were also set up as a normal SST control group. The mice in each group were sacrificed at the predetermined time points, and the supraspinatus tendon specimens were subsequently obtained. These specimens were then fully analyzed and thoroughly scientifically evaluated using multiple imaging techniques, including biomechanics, histology, immunohistochemistry, quantitative polymerase chain reaction (qPCR), and electron microscopy.

Results: The mean ultimate tensile strength (UTS) and stiffness of the tendinopathy specimens exhibited a considerable decrease of approximately 50% ($P<0.001$) when compared to normal tendons (Figure 1). Histological analysis revealed a significant elevation in Bonar scores following impact (Figure 1). In the surgical group, there was a notable increase in the positive area of endoplasmic reticulum stress (ERS)-related molecules, demonstrating a consistent trend post-impact. Furthermore, genes associated with ERS were upregulated in the tendinopathy group, with a significant increase observed in the expression of the inositol triphosphate receptor (IP3R). Immunofluorescence staining indicated heightened levels of molecules involved in ER-mitochondrial calcium coupling (Figure 3). Transmission electron microscopy (TEM) provided evidence of substantial alterations in ERS morphology across all surgical groups (see Figure 4). Additionally, semiquantitative analysis corroborated these morphological changes in ERS and underscored the augmented contacts between the endoplasmic reticulum and mitochondria in tendinopathy specimens.

Discussion: The endoplasmic reticulum (ER) and mitochondria play crucial roles in the progression of tendinopathy. ER stress in tendons and the molecules involved in the coupling of ER and mitochondrial calcium (Ca2+) are essential for regulating cellular metabolism.

Significance/Clinical Relevance: The endoplasmic reticulum (ER) and mitochondria play pivotal roles in tendinopathy's progression and underlying mechanisms. These organelles are involved in regulating excessive apoptosis and autophagy in tenocytes. Notably, the stress response of the ER within tendons, along with the molecules implicated in ER-mitochondrial calcium (Ca2+) coupling, is critical for regulating cellular metabolism. Therefore, these components are positioned as significant targets for effective intervention and treatment strategies for tendinopathy in future research endeavors.