DYNAMIC POLARIZATION MICROSCOPY FOR IN-SITU MEASUREMENTS OF COLLAGEN FIBER REALIGNMENT DURING IMPACT

Xianyu Wu, Hsiao-Ying Shadow Huang, Mark Pankow, Kara Peters

North Carolina State University, Raleigh, NC, USA

email: xwu13@ncsu.edu

INTRODUCTION

Tendon tissue is flexible but strong fibrous connective tissue which attaches muscle to bone. The transition from tendon to bone is a complex region that transfers the load effectively to prevent injury and provide proper joint function [1]. The structure and composition of the tendon-to-bone insertion is functionally graded along its longitudinal direction. These tissues are subjected to physiologic loading on a daily basis; however, these highly specialized tissues are often damaged from onetime events such as car accidents or sports activities.

The insertion region contains collagen fibers that align with the loading direction and the architecture of collagen fibers affects the mechanical properties of the tissue. Since the collagen fibers realign as a function of time the insertion region is loading-rate sensitive and most injuries occur during high speed dynamic events. Therefore, understanding the high loading rate behavior and failure mechanisms of the insertion region will lead to better clinical repair techniques.

Many tissues (e.g. tendon-to-bone insertion) have a birefringence response that has different refractive indices in different principal axes of loading. The polarized light microscopy (PLM) technique can be used to image the collagen fiber alignment angles [2] due to the birefringence response allowing us to visualize the collagen fibers indirectly.

The long-term goal of this work is to better understand the tendon-to-bone insertion injury due to medium strain rate impact (e.g. sports activity). Specifically, we imaged collagen fiber realignment during impact, to investigate the ability of the tendon-to-bone insertion to survive harsh impact events and understand the realignment process.

METHODS

A PLM setup was built in the lab and quasi-static testing (Fig. 1) was used to monitor the birefringence property changes of a known material under

changing stress conditions. Initially polycarbonate dogbone specimens were tested to validate the setup and analysis algorithm. A MATLAB script has been written which can generate fiber alignment and retardation maps from the image sequence.



Monochromatic filter (546.1 nm) Specimen Camera **Figure 1**: Quasi-static test setup and self-built polarized light microscopy experimental setup.

To perform dynamic experiments a drop weight tower was modified for medium strain rate testing (10%/sec - 100%/sec). Experiments are performed on porcine tendon-to-bone insertions to investigate their dynamic response and deformation. The PLM setup has been modified to operate at high frequency and synchronized with a high-speed camera, see Fig. 2, allowing for dynamic visualization of the tissue during dynamic deformation. The dynamic alignment maps will be correlated with the drop tower impacting load, which is measured using an accelerometer.



Figure 2: Dynamic Polarized light microscopy setup with optical elements incorporated into drop tower.

RESULTS AND DISCUSSION

A 0.381 mm thick polycarbonate dogbone specimen (38.1 mm wide) was cut to test. A 1.651 mm diameter hole was cut at the center of the polycarbonate specimen to create a non-uniform stress field when the specimen was loaded on the test instrument. The specimen was mounted on the quasi-static testing instrument and loaded. A series of images were recorded as the sample was incrementally loaded. Fig. 3 shows the alignment and retardation maps generated from one stage of loading. The vectors overlapped on top of the alignment map were generated by averaging the corresponding alignment map over 10×10 pixels sub areas. Based on the stress field induced by uniform tension on a plate with a hole and the optic stress law, the experiment results matched the analytical expectation, validating the experimental setup and analysis code.



Figure 3: Polarized light (a) alignment image and (b) retardation image from measurements of a polycarbonate dogbone specimen. The center hole area was covered using a mask in the data processing.

Several dynamic experiments have been conducted using this modified drop tower on porcine tendon specimens. Fig. 4 shows images of high-speed deformation of a tendon tissue sample. The sequence of images shows how the sample deforms with respect to time.



Figure 4: An image captured using high-speed camera during an impact test of a tendon tissue specimen (11 mm wide, marked by dashed boxes) mounted in the modified drop tower. Black marks on the tendon specimen were made by graphite as reference. High speed camera frame rate: 12500 FPS.

CONCLUSIONS

The presented work has built the foundation for understanding dynamic deformation of collagen fiber realignment. In the future work, the PLM setup will be integrated with a high-speed camera for monitoring the tendon-to-bone insertion deformation in-situ under different strain rate loadings.

Studies conducted in tendon-to-bone insertion dynamic tests will provide important information regarding insertion failures and associated collagen fiber realignments. Better understanding of the tendon-to-bone dynamic behavior would help to improve the clinical repairing techniques and design of engineered tissue replacement. The study on dynamic behavior of tendon-to-bone insertions will also help in designing better implants that can potentially resist against failure.

REFERENCES

- 1. Thomopoulos S, et al. *J Orthopaedic Research*, **21(3)**, 413-419, 2003
- 2. Tower TT, et al. *Biophysical J*, **81(5)**, 2964-2971, 2001
- 3. Tower TT, et al. *Annuals of Biomedical Engineering*, **30(10)**, 1221-1233, 2002 Nov 1