The Influence of In Vivo vs. Ex Vivo Testing Environment on Sheep Medial Gastrocnemius Tendon Hysteresis

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Summary

In this study, direct measurements of muscle force and tendon length were performed *in vivo* during locomotion and *ex vivo* using a material testing machine. *Ex vivo* testing replicated the *in vivo* force-time curves of representative step cycles. Tendon hysteresis (energy dissipation during unloading) was greater *in vivo* than *ex vivo* conditions. Our results suggest that differences in the testing environment largely account for the variance in tendon hysteresis between *in vivo* and *ex vivo* conditions.

Introduction

Ex vivo tendon hystereses range between 3% and 20% across tendons and species [1]. *In vivo* hystereses exhibit large variability (2-45%), with mean values up to 55% [2,3]. These variations have been attributed to differences in estimating both tendon elongations and forces *in vivo* and differences in the testing environment (*ex vivo* vs. *in vivo*) [2,3]. Comparing direct measurements of tendon hystereses *in vivo* and *ex vivo* for the same tendon may help determine the cause(s) for the differences reported in the literature. This study aimed to investigate the differences in tendon hystereses measured *in vivo* during sheep locomotion and *ex vivo* using a material testing machine. We hypothesized that hystereses measured *in vivo* would be greater than those measured *ex vivo*.

Methods

We present exemplar *in vivo* and *ex vivo* medial gastrocnemius (MG) tendon data from one sheep walking on a treadmill at varying speeds (0.7, 1.3, and 2 m/s) and uphill inclinations (0°, 3°, and 6°). *In vivo* MG forces were measured using a buckle-type force transducer [4], and tendon lengths were measured using a silastic tubing length sensor [5]. *Ex vivo* experiments were conducted with an Instron machine, which recorded force and length simultaneously. The *in vivo* force-time curves of a representative step cycle for each locomotion condition were replicated during the *ex vivo* testing, enabling comparisons for near-identical loading conditions between the *in vivo* and *ex vivo* experiments. Tendon hystereses were calculated based on the loading and unloading phases and averaged over five consecutive step cycles *in vivo* and *ex vivo*.

Results and Discussion

Across all conditions, our results suggest that *in vivo* tendon hysteresis was greater (44±8%) than *ex vivo* (14±1%). The greater hysteresis values measured *in vivo* compared to *ex vivo* remain unexplained. We speculate that differences in the fluid environment between the *in vivo* and *ex vivo* conditions (e.g., different temperatures, osmolarities, viscosities), as well as differences in boundary conditions, might result in changes in the tendon mechanical properties. Additionally, we measured hysteresis in the free tendon *ex vivo* vs. the tendon *in vivo* surrounded by other tendons and the epitenon. Other potential biological differences (blood supply, continuous regeneration and protein turnover of the tendon *in vivo* but not *ex vivo*), might also explain these differences. These results raise the question: what are the consequences of deriving our understanding (e.g., muscle mechanical models) of tendon hystereses from *ex vivo* measurements?

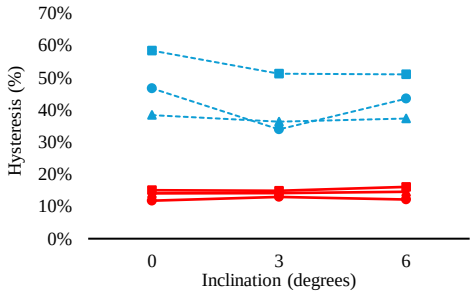

Figure 1: Blue dashed lines and red solid lines show *in vivo* and *ex vivo* hysteresis measurements, respectively. Circles, triangles, and squares indicate 0.7, 1.3, and 2 m/s, respectively.

Conclusions

We conclude that sheep MG tendon has greater hystereses (more energy loss) *in vivo* than *ex vivo*, highlighting the importance of accounting for *in vivo* conditions when evaluating tendon properties during dynamic activities and in developing musculoskeletal models to avoid underestimating energy dissipation during loading/unloading cycles.

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References

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