



*Bradypus variegatus*

# Metabolic Properties of Sloth Muscles (Xenarthra: Pilosa)

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*Choloepus hoffmanni*

## Purpose

Tree sloths have low muscle mass<sup>1</sup>, but they are capable of suspending themselves underneath tree branches for extended periods of time<sup>2</sup>, a behavior that requires both great strength and endurance. This paradox is difficult to reconcile by our current understanding of the physiological properties of mammalian skeletal muscle. This study builds on our previous work that identified only two isoforms of myosin heavy chain (MHC) expressed in sloth forelimb musculature. Sloths have one slow myosin fiber type (MHC-1) and one fast myosin fiber type (MHC-2A)<sup>3</sup>. In addition to these pure fiber types, sloths expressed a small distribution of 1-2A hybrid muscle fibers<sup>4</sup>. Here we investigate metabolic modifications to slow and fast forelimb muscles of two-toed (*Choloepus*) and three-toed (*Bradypus*) sloths that allow for sustained force contractions. This will be achieved by determining the enzymatic (aerobic vs. anaerobic) activities of sloth muscles<sup>5</sup>.

## Hypothesis

Sloths have slow- and fast-twitch muscles that are both highly aerobic in their energy metabolism to sustain force production.

## Methodology

- A total of  $N=6$  animals (3, *B. variegatus*; 3, *C. hoffmanni*)
- Whole muscle tissue samples from slow *m. trapezius* and fast *m. flexor carpi ulnaris* were prepared as homogenates
- Reacted sloth muscle samples in Citrate synthase (CS) and 3-Hydroxyacyl-CoA dehydrogenase (3HAD) incubations for oxidative (aerobic) metabolism<sup>5</sup>
- Phosphofructokinase (PFK) and Lactate dehydrogenase (LDH) incubations for glycolytic (anaerobic) metabolism<sup>5</sup>
- Enzyme assay absorbance measured by spectrophotometry;  $n=3$  trials/muscle/individual
- Determined mean slope of standard curves to quantify enzyme activities using the equation:

$$E = \frac{\Delta \text{Absorbance} \times \text{Dilution Factor}}{[\text{muscle}] \text{ g/L}}$$

E = extinction coefficient

## Results

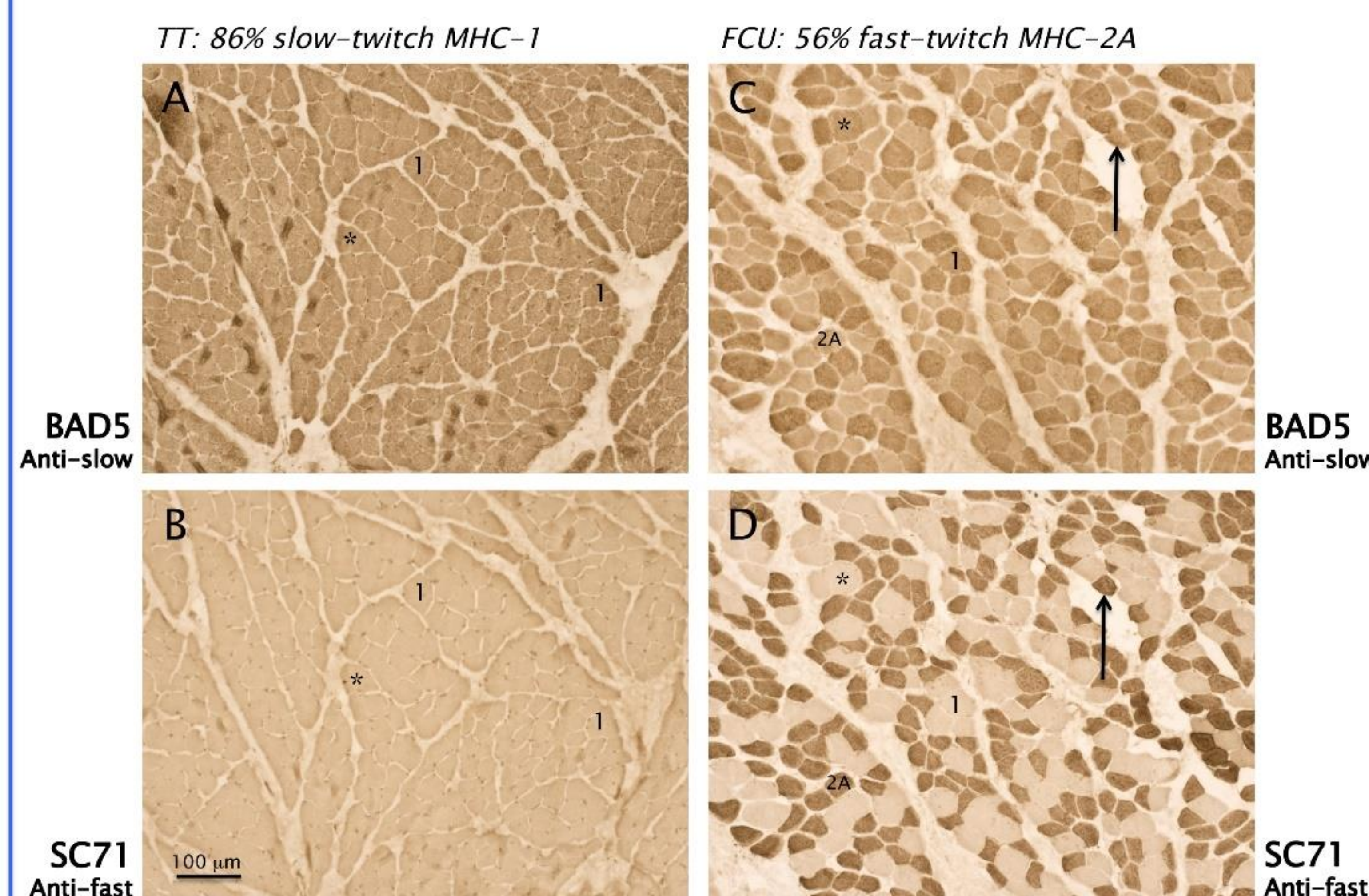


Figure 1. Reactions from *m. trapezius thoracica* (TT: panels A, B). Reactions from *m. flexor carpi ulnaris* (FCU: panels C, D). Panels are labeled with the MHC fiber types identified in our previous study. Individual fibers labeled (1) are slow-twitch MHC-1. Fibers labeled (2A) fast-twitch MHC-2A. The arrow indicates a MHC-1/2A hybrid fiber with contractile properties intermediate to the pure fiber types. The (\*) is the same fiber in each serial section.

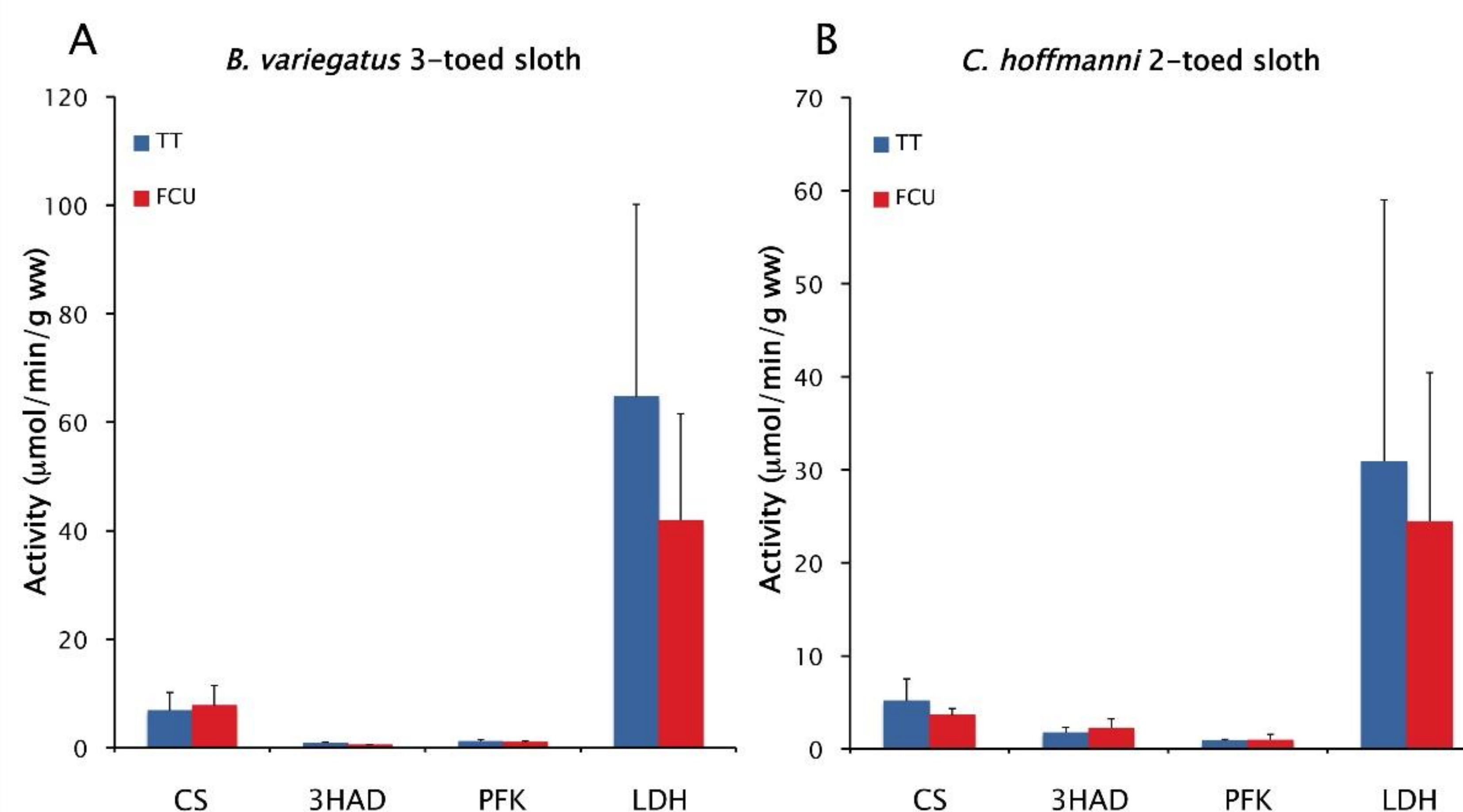


Figure 2. A. Mean  $\pm$  (standard deviation: SD) enzyme activity for two forelimb muscles of *Bradypus variegatus* ( $N=3$ ). B. Mean  $\pm$  SD enzyme activity for the same two forelimb muscles of *Choloepus hoffmanni* ( $N=3$ ). CS, citrate synthase; 3HAD, 3-hydroxyacyl-CoA dehydrogenase; PFK, phosphofructokinase; and LDH, lactate dehydrogenase. CS and 3HAD are markers for oxidative-aerobic muscle metabolism. PFK and LDH are markers for glycolytic-anaerobic muscle metabolism. All activities were calculated by standard energetic formulae. Each assay was run 3x per muscle/individual.

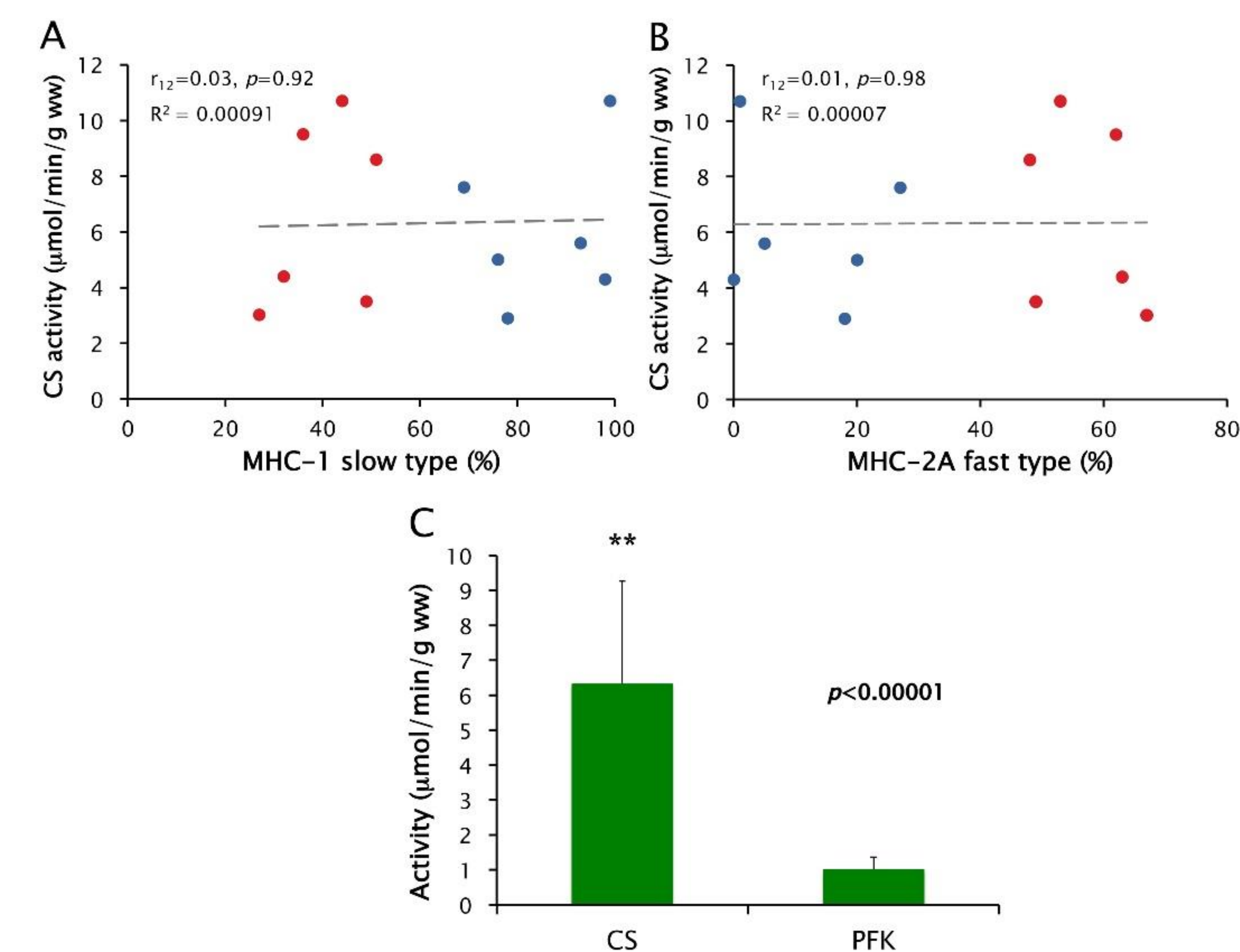


Figure 3. Citrate synthase (CS) activity correlated with slow MHC-1 (A) and fast MHC-2A (B) fiber types. Correlation statistics are Pearson Moment ( $r$ ). C. Mean  $\pm$  SD activities for CS and PFK. All individual muscle data combined and analyzed by  $t$ -test. \*\*Significance accepted at  $p \leq 0.05$ .

## Main Outcomes/Conclusions

- The hypothesis is partially supported
- Enzyme activities not correlated with MHC fiber types
- Muscle and basal metabolism is suppressed in sloths
- Elevated CS and LDH activities suggest recycling of lactic acid with oxidative metabolism for endurance
- Neuromuscular properties also important to sustained force

## References

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