Supplementary Methods:

S1. Subject-specific Achilles tendon moment arm:

Subject-specific moment arms were determined using a 38 mm transducer (L14-5W/38; Ultrasonix corp, Richmond, British Columbia), operating at 70 frames per second, placed on the Achilles free tendon, distal to the soleus muscle-tendon junction with a custom orthotic. We collected the positions and orientation of the transducer using three retroreflective markers placed on the custom orthotic. Using previously-published techniques (Manal et al., 2013), we estimated moment arm as the average perpendicular distance between the tendon's line of action, registered in ultrasound images of the free Achilles tendon and the transmalleolar midpoint. Moment arm values were collected across at least 6 joint angles per subject (-20°, -10° , 0° , 10° , 20° , 30°). If a participants' maximal dorsiflexion range of motion (ROM) was greater than 25°, a seventh joint angle was added that was equal to their maximum dorsiflexion ROM. We then fit a curve to those points and used the equation to calculate time series Achilles tendon moment arm as a function of ankle angle.

S2. Subject-specific force-length relation:

To determine each subject's force-length relation and optimal fascicle length (l_o) , medialgastrocnemius muscle fascicle length during isometric contractions was assessed from ultrasound images using an open source MATLAB routine (MathWorks, Natick, Massachusetts), UltraTrack (Farris and Lichtwark, 2016). A second-order polynomial fit was used to derive l_o for each subject.

S3. Quantifying muscle fascicle length and muscle-tendon unit length during walking:

Muscle fascicle length was quantified during walking with a deep learning software package (Cronin et al., 2020). To create the training data for the neural network, muscle fascicle first were manually identified in several frames of various ultrasound videos. The most representative fascicle in the mid-region of a walking ultrasound image was selected and then manually tracked, along with the aponeuroses, in several frames of the ultrasound video using ImageJ. Separate neural networks were trained for each subject using the manually tracked data from each respective subject. These trained neural networks were then used to analyze fascicle length and pennation angle for all ultrasound data from their respective subject. After analysis, the representative fascicle was manually selected from the computer-tracked data on the first frame of the ultrasound video, and this fascicle was isolated in each subsequent frame using a custom MATLAB script (MathWorks, Natick, Massachusetts). Only data from this representative fascicle was used for analysis. In addition, muscle-tendon unit lengths were estimated using standard inverse kinematics procedures derived from a gait2392 model in opensim.

Supplementary Data:

S5. Medial gastrocnemius myotendinous unit (MTU) length:

We first found that MG MTU length decreased during mid-to-late stance when targeting higher than normal TS activity. This outcome is consistent with the shorter fascicle lengths observed during these conditions. We also found that MG MTU lengths during mid-to-late stance were not affected by targeting lesser than normal TS activity. This latter outcome suggests that MTU length change alone cannot explain the decrease in fascicle length for -20 and -40% conditions.



Supplementary Figure 1. Mean medial gastrocnemius myotendinous unit (MTU) length throughout the gait cycle when targeting normal, $\pm 20\%$, and $\pm 40\%$ peak TS activity during walking. Time regions shaded gray indicate a statistically significant (p < 0.05) main effect using statistical parametric mapping.

S6. Joint kinetics and kinematics:

For conditions that prescribed increased TS activity, hip joint power decreased while ankle joint power generally increased during the push-off phase of walking. It is unlikely that this decrease in hip joint power resulted in a greater hip muscle contribution to metabolic cost. Knee joint power absorption generally decreased during leg swing when increasing TS activity, but increased during early stance. However, given the relative economy of negative muscle work, we do not believe these increases at the knee are of sufficient magnitude to explain measured changes in whole-body metabolic energy cost.

For conditions that prescribed decreased TS activity, hip joint power increased while ankle joint power decreased during the push-off phase of walking. Stance phase knee joint power was generally unaffected by condition. These patterns of joint power allude to a proximal redistribution in muscle demand that may contribute to our metabolic outcomes for conditions that prescribed decreased TS activity.



Supplementary Figure 2. Mean joint angle, moment, and power for the hip, knee and ankle joints throughout the gait cycle when targeting normal, $\pm 20\%$, and $\pm 40\%$ peak TS activity during walking. Time regions shaded gray indicate a statistically significant (p<0.05) main effect using statistical parametric mapping.

S7. Time series fascicle length, fascicle velocity, and force:

For our three primary muscle dynamics outcomes (i.e., length, velocity, and force), our selection of the time of peak force for analysis revealed that with increased activation: (*i*) MG fascicle length decreased, (*ii*) fascicle velocity did not change, and (*iii*) GAS force increased. A supplementary SPM analysis supported these conclusions, and demonstrated that increased activation targets similarly yielded: (*i*) decreased MG fascicle length throughout stance, (*ii*) minimal change in fascicle velocity during stance, and (*iii*) increased GAS force during most of stance.

Similarly, our selection of the time of peak force for analysis revealed that with decreased activation: (*i*) MG fascicle length decreased, (*ii*) fascicle velocity did not change, and (*iii*) GAS force decreased. A supplementary SPM analysis also tended to support these conclusions. The main difference was a trend toward modestly higher GAS force during early-to-mid stance. After analyzing our time-series data with SPM, we believe these single-instant measurements are representative of the predominant changes observed throughout the stride.



Supplementary Figure 3. Medial gastrocnemius fascicle length, medial gastrocnemius fascicle velocity, and gastrocnemius force throughout the gait cycle when targeting normal, $\pm 20\%$, and $\pm 40\%$ peak TS activity during walking. Time regions shaded gray indicate a statistically significant (p<0.05) main effect using statistical parametric mapping.