**A Cross-Bridge Based Model of Force Depression:
Can a Single Modification Address both Transient and Steady-State
Behaviors?**

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ABSTRACT

Force depression (FD), the reduction of isometric force following active shortening, is a phenomenon of skeletal muscle that has received significant attention in biomechanical and physiological literature, yet the mechanisms underlying FD remain unknown. Recent experiments identified a slower rate of force redevelopment with increasing amounts of steady-state FD, suggesting that FD may be caused, at least in part, by a decrease in cross-bridge binding rate (Corr and Herzog, 2005; Koppes et al., 2014). Herein, we develop a cross-bridge based model of FD in which the binding rate function, f , decreases with the mechanical work performed during shortening. This modification incorporates a direct relationship between steady-state FD and muscle mechanical work (Corr and Herzog, 2005; Herzog et al., 2000; Kosterina et al., 2008), and is consistent with a proposed mechanism attributing FD to stress-induced inhibition of cross-bridge attachments (Herzog, 1998; Maréchal and Plaghki, 1979). Thus, for an increase in mechanical work, the model should predict a slower force redevelopment (decreased attachment rate) to a more depressed steady-state force (fewer attached cross-bridges), and a reduction in contractile element stiffness (Ford et al., 1981). We hypothesized that since this modification affects the cross-bridge kinetics, a corresponding model would be able to account for both transient and steady-state FD behaviors. Comparisons to prior experiments (Corr and Herzog, 2005; Herzog et al., 2000; Kosterina et al., 2008) show that both steady-state and transient aspects of FD, as well as the relationship of FD with respect to speed and amplitude of shortening, are well captured by this model. Thus, this relatively simple cross-bridge based model of FD lends support to a mechanism involving the inhibition of cross-bridge binding, and indicates that cross-bridge kinetics may play a critical role in FD.

1. INTRODUCTION

Force depression (FD), the reduction of isometric force following activated shortening, is a characteristic behavior of skeletal muscle that has received considerable attention in the literature, yet its underlying mechanisms remain unknown. Studies in single fibers (Edman et al., 1993; Granzier and Pollack, 1989; Julian and Morgan, 1979; Sugi and Tsuchiya, 1988) and whole muscles (Abbott and Aubert, 1952; Herzog and Leonard, 1997; Maréchal and Plaghki, 1979; Meijer et al., 1997; Morgan et al., 2000) have characterized the phenomenon, indicating that the magnitude of FD at steady-state (FD_{ss}) increases with increased shortening amplitudes (Abbott and Aubert, 1952; De Ruiter et al., 1998; Herzog and Leonard, 1997; Maréchal and Plaghki, 1979; Meijer et al., 1997; Morgan et al., 2000; Sugi and Tsuchiya, 1988), with decreased shortening speeds (Abbott and Aubert, 1952; De Ruiter et al., 1998; Herzog and Leonard, 1997; Maréchal and Plaghki, 1979; Meijer et al., 1997; Morgan et al., 2000; Sugi and Tsuchiya, 1988), with increased force during shortening (De Ruiter et al., 1998; Herzog and Leonard, 1997), and with increased mechanical work during shortening (Herzog et al., 2000; Kosterina et al., 2008). A decrease in fiber stiffness, proportional to steady-state FD, has also been observed at steady-state (Sugi and Tsuchiya, 1988).

In addition to steady-state findings, recent experimental studies have addressed the transient nature of FD. Qualitative observations following activated shortening in the human adductor pollicis indicated a slowing of the force redevelopment rate, particularly following slow and large amplitude shortening (De Ruiter et al., 1998). Measurements at discrete time points during force redevelopment indicated that FD was largest immediately following shortening, decreased over time, and approached a steady-state, suggesting that FD is comprised of two separate mechanisms: a long-lasting, steady-state FD, and a transient depression that decreases

with time (Lee and Herzog, 2003). Detailed analyses of the force recovery period, in whole muscle (Corr and Herzog, 2005; Van Noten and Van Leemputte, 2009) and isolated fibers (Koppes et al., 2014), identified a positive correlation between steady-state FD and mechanical work, and a negative correlation between work and rate of force redevelopment; therefore, more mechanical work results in a slower rise to a more depressed steady-state force.

Despite thorough experimental characterization of FD, few attempts have been made to model this phenomenon. One of the first models (Morgan, 1990) attributed FD to a dispersion of sarcomere lengths developed during active shortening, whereby forces in the non-uniform sarcomeres equilibrate to yield a force smaller than that of an isometric contraction with uniform sarcomeres (Edman et al., 1993; Morgan et al., 2000). Morgan's computational model (Morgan, 1990) captured many aspects of FD behavior, and offered one of the first theoretical models of history-dependent behavior. However, there have been conflicting findings regarding the association of FD with sarcomere length non-uniformities. While Edman et al. (Edman et al., 1993) identified a strong correlation between sarcomere non-uniformity and steady-state FD, Granzier and Pollack (Granzier and Pollack, 1989) observed FD in experiments where sarcomere lengths were controlled to remain uniform. Furthermore, FD has been associated with a significant decrease in stiffness, which cannot be explained by this mechanism (Corr and Herzog, 2005; Lee and Herzog, 2003; Sugi and Tsuchiya, 1988).

In response to active shortening, muscle also exhibits a deactivation that is proportional to the mechanical work performed (Josephson and Stokes, 1999). Recent approaches used this work-dependent deactivation to introduce FD into phenomenologic skeletal muscle models (Kosterina et al., 2013; McGowan et al., 2010; Williams, 2010). In an alternate approach, Wu and Herzog incorporated the experimentally observed relationship, between work and steady-

state FD, into a structural model (Wu and Herzog, 1999) in which force was calculated using the classic cross-bridge model (Huxley, 1957), a power law function was subtracted to match the force at steady-state, and a fading memory function was used to slow force redevelopment. These models showed great agreement with experimental data, yet due to their phenomenological nature, offered limited insight into possible mechanisms of FD.

We believe that the mechanism of FD resides, at least in part, within the cross-bridges. Therefore, a model incorporating cross-bridge kinetics should be able to describe the transient and steady-state aspects of FD. To test this idea, we developed a cross-bridge based model of FD, consistent with a previously proposed mechanism that FD is caused by a stress-induced inhibition of cross-bridge attachment (Herzog, 1998; Maréchal and Plaghki, 1979).

We incorporated this mechanism using a single scalar variable: the mechanical work done during shortening. The mechanism was introduced at the site of actin-myosin interaction, thereby affecting the kinetics of force generation, and providing unique possibilities for comparison with transient and steady-state experimental FD data.

We hypothesized that a model relating mechanical work to a change in the cross-bridge binding rate, might provide accurate predictions of experimentally observed changes in force recovery following active shortening and the associated steady-state force depression (Corr and Herzog, 2005; Koppes et al., 2014).

2. MATERIALS AND METHODS

2.1 Modeling Methods

2.1.1 Cross-Bridge Based Model

In Huxley's classic cross-bridge model (Huxley, 1957), the first-order kinetics governing the actin-myosin reaction is expressed as

$$\left(\frac{\partial n(x,t)}{\partial t} \right) - v(t) \left(\frac{\partial n(x,t)}{\partial x} \right) = f(x) - [f(x) + g(x)]n(x,t), \quad (1)$$

where n is the bond-distribution function of attached cross-bridges, i.e., fraction of attached cross bridges with displacement x at time t , $v(t)$ represents the relative myofilament shortening velocity, and f and g represent the forward (bonding) and backward (unbonding) rate functions, respectively. Once n is found, macroscopic quantities for the entire contractile element can be determined by computing the first three moments of the bond distribution to produce the instantaneous stiffness, force, and total elastic energy stored in the contractile tissue, respectively. Our theoretical model, based on that of Cole et al (Cole et al., 1996), is comprised of a contractile element (CE) in series with an elastic spring, representing the contractile muscle and a compliant tendon, respectively.

We model the contractile tissue using the revised Distribution Moment (DM) model (Zahalak and Ma, 1990). Zahalak's original DM model (Zahalak, 1981, 1986), assumes that the distribution of attached cross-bridges, n , is Gaussian, and through integration of the distribution, transforms the governing partial differential equation (Eq. 1) to a system of three coupled first-order ordinary differential equations (ODEs). The muscle's maximal force producing capability is incorporated into the DM model via α , the proportion of cross-bridges capable of interacting with activated actin (Zahalak and Ma, 1990). α represents the muscle's normalized force-length (F - L) relationship, and is a function of actin-myosin myofilament overlap. We represent α by a hyperbolic function of muscle-tendon unit length, L_{MT} , (Woittiez et al., 1984), that enters the DM model through the rate equations (Zahalak and Ma, 1990), rather than the moment equations of

the bond distribution (Zahalak, 1981, 1986). As a result, the number of participating cross-bridges is correctly incorporated into the cross-bridge binding rates prior to integration. The three equations take the form

$$\dot{\mathbf{Q}}(t) = \dot{\mathbf{Q}}[\alpha(L_{MT}), \mathbf{Q}(t), u(t)], \quad (2)$$

where $\mathbf{Q}$ is a vector of the state variables ($= \{Q_0, Q_1, Q_2\}$), $u(t)$ is the myofilament shortening velocity normalized by Huxley's bond-length scaling parameter ($=v(t)/h$), and the superposed dot indicates differentiation with respect to time (Cole et al., 1996; Zahalak and Ma, 1990). An additional equation relates the muscle-tendon unit velocity, $\dot{L}_{MT}$, to the contractile unit shortening velocity,

$$\dot{L}_{MT}(t) = \dot{L}_{MT}[Q_1, \dot{Q}_1, u(t)], \quad (3)$$

resulting in a system (Eqs. 2 and 3) of four coupled first-order ODEs (Cole et al., 1996; Zahalak, 1986). Because n is assumed Gaussian, and not directly determined, the DM model is an approximation, not an exact solution, to the Huxley equation (Eq. 1). Details of the DM model with a compliant tendon have been presented previously (Cole et al., 1996; Zahalak, 1981, 1986; Zahalak and Ma, 1990), and a brief mathematical description of our model has been included in the Appendix.

The tendon was modeled as an elastic structure with non-constant compliance (Cole et al., 1996), to account for its strain-stiffening behavior exhibited at low loads. The cross-bridge bonding and unbonding rate functions, $f(x)$ and $g(x)$, respectively, are those used by Cole et al (Cole et al., 1996), (Fig. 1). Additional modeling parameters for cat soleus were taken from the literature (Cole et al., 1996), and are summarized in Table 1.

2.1.2 FD modifications

The cross-bridge theory, and its DM approximation, do not provide a means of modeling history-dependent behavior of skeletal muscle, and therefore are unable to account for either transient or steady-state aspects of FD (Walcott and Herzog, 2008). To incorporate FD into our model, the forward bonding rate function, f , was assumed to decrease with the mechanical work performed during activated shortening, W , such that

$$f(x) = [f_1 - (\zeta * W)]x, \quad (4)$$

for $0 < x < h$, where ζ is an arbitrary constant relating the mechanical work, W , to the decrease in bonding rate. A value of $\zeta = 10^4 \text{ (m}\cdot\text{s)}^{-1}$ was chosen, based on the specified rate functions (Fig. 1), to maintain a positive forward binding rate for every simulation. This modification incorporates experimental findings that identified a direct relationship between steady-state FD and muscle mechanical work (Corr and Herzog, 2005; Herzog et al., 2000; Koppes et al., 2014; Kosterina et al., 2008), and is based on the idea that FD is caused by a stress-induced inhibition of cross-bridge attachments (Herzog, 1998; Maréchal and Plaghki, 1979). Therefore, an increase in mechanical work decreases the forward bonding rate, which, with all other rate constants fixed, results in a slower force rise (decreased rate of attachment) to a more depressed steady-state force (fewer attached cross-bridges); a result observed experimentally in whole tissue (Corr and Herzog, 2005) and single-fibers (Koppes et al., 2014).

2.1.3 Simulations

Numerical simulations were run for a cat soleus muscle-tendon unit subjected to three shortening amplitudes (3, 6, 9 mm = 3, 6, 9 % $L_{MT(opt)}$) at three shortening speeds (3, 9, 27 mm/s;

= 7, 21, 63 % optimal fiber lengths/s), (Fig. 2). This protocol was chosen to address the dependence of both steady-state and transient FD on shortening speed, shortening amplitude, and mechanical work performed by the muscle-tendon unit, and to allow direct comparison with previous experimental data from cat soleus muscle-tendon units ($N = 8$) undergoing identical protocols (Corr and Herzog, 2005). In each simulation, the muscle started at rest, was immediately stimulated to full activation, and tetanic activation was maintained throughout the entire test (Fig. 2, 3a). All simulated shortenings occurred on the ascending limb of the F - L relationship; starting once the tetanic force was fully developed, and ending at the same final length ($L_f = 90 \% L_{MT(opt)} = 90 \text{ mm}$).

The coupled four-ODE system was solved numerically in MATLAB (version 7.10, The MathWorks, Inc.) using a 4th-5th order Runge-Kutta algorithm (Matlab's ode45 function) over discrete time intervals (corresponding to time steps between temporal data points), and iterated through the entire data set to produce vectors of stiffness, force, and total elastic energy for each simulation. Mechanical work performed by the muscle, W , was calculated as the dot product of force and velocity, integrated with respect to time as approximated by the trapezoidal method. This integral can be computed for any active shortening event (*e.g.*, monotonic, compound, non-constant shortening rate, stepped shortening, etc.), and reset to zero once activation is interrupted, to mimic previous findings that FD is long lasting (Herzog et al., 1998) and is abolished with an interruption in stimulation (Abbott and Aubert, 1952; De Ruiter et al., 1998; Herzog and Leonard, 1997).

2.2 Transient Analysis

To quantify the transient force recovery, $F(t)$, the force-time simulation data following activated shortening were analyzed using an exponential recovery function,

$$F(t) = F_{\text{inf}} - Ae^{-kt}, \quad (5)$$

where F_{inf} represents the depressed force at steady state, A the amount of force recovered (difference between F_{inf} and force immediately following shortening), and k the exponential force recovery rate, using methods previously described (Corr and Herzog, 2005). Briefly, the entire 0-5 s of force-time simulation data following activated shortening was exported from MATLAB and fit with Eq. 5 ($R^2 > 0.994$) using a Levenberg-Marquardt error minimization algorithm in a commercial software package (DeltaGraph 5.0, SPSS Inc.), to obtain estimates of F_{inf} , A , and k . The 5-s recovery period provided ample time for force to reach steady-state in each simulation.

2.3 Data Analyses

We define steady-state force depression (FD_{ss}) as the difference between the depressed force at steady-state (F_{inf}) and the corresponding isometric reference force (F_{iso}) (Fig. 3a). Similarly, we define steady-state stiffness depression (SD_{ss}) as the difference in muscle-tendon stiffness (S) measured at steady-state following activated shortening, and that of the isometric reference contraction at the same final length. To identify trends with respect to shortening amplitude and speed, parameter values (W , A , k , FD_{ss} , S) were plotted against shortening amplitude for the three speeds of shortening, and against speed for the three amplitudes of shortening. Since mechanical work was not directly controlled, but rather a result of the imposed simulation conditions, regression analyses were conducted (SPSS statistical software, 12.0,

SPSS) to observe the correlation between measured quantities (FD_{ss} , SD_{ss} , A , k) and mechanical work (W).

3. RESULTS

Simulations using the cross-bridge based FD model (Fig. 3a) exhibit similar transient and steady-state behaviors to those observed experimentally in cat soleus (Fig. 3b). Analysis of simulation data revealed that an increase in the amplitude of shortening, for a given speed of shortening, resulted in an increase in mechanical work (Fig. 4a) and a decrease in stiffness (Fig. 4c). Conversely, an increase in shortening speed, for a given amplitude of shortening, resulted in a decrease in mechanical work (Fig. 4b), and a smaller stiffness reduction (Fig. 4d).

The exponential recovery function quantified both steady-state and transient variables of interest. An increase in shortening amplitude, for a given speed, resulted in increases in the recoverable force, A , and FD_{ss} , and a decrease in force recovery rate, k (Fig. 5a, c, e). Increases in shortening speed, for given amplitudes of shortening, produced increases in A and k , and decreases in FD_{ss} (Fig. 5b, d, f). This model displays similar steady state (FD_{ss}) and transient (A , k) FD behaviors, and a similar relationship of FD with respect to speed and amplitude of shortening, as that observed experimentally (Fig. 6).

Mechanical work, across all simulations, correlated positively with FD_{ss} (Fig 7a), negatively with k (Fig. 7b), and did not significantly correlate with A ($P > 0.05$); matching prior experimental observations (Corr and Herzog, 2005). There was also a strong positive correlation between SD_{ss} and FD_{ss} (Fig 7c), consistent with results in rabbit single fibers (Joumaa et al., 2012). It therefore follows that stiffness should negatively correlate with mechanical work, as observed (Fig. 7a). However, this contrasts to our previous experimental findings that failed to observe a significant work-stiffness correlation (Corr and Herzog, 2005).

4. DISCUSSION

4.1 Model Justification

Our modeling approach incorporated a work-mediated reduction in the forward binding rate to account for FD within the cross-bridge, in an attempt to capture both steady-state and transient aspects of FD. This is consistent with a previously proposed mechanism in which FD is caused by a stress-induced inhibition of cross-bridge binding (Herzog, 1998; Maréchal and Plaghki, 1979). In this mechanism, strain in the myofilaments causes a decreased probability of cross-bridge binding within the newly formed actin-myosin overlap zone (formed during active shortening), and thus a reduction in force production. Studies support this mechanism, suggesting that thick and thin filament compliance (Huxley et al., 1994), may cause angular distortions of cross-bridge binding sites on actin, and thus may affect cross-bridge kinetics (Daniel et al., 1998). Therefore, an increase in stress (force) may cause greater angular distortion of binding sites, and thus decrease the probability of cross-bridge attachment. Additionally, an increase in muscle shortening would create a larger newly formed actin-myosin overlap zone, and therefore affect more cross-bridges.

We introduced this mechanism into our model using the mechanical work done during shortening. Since an increase in mechanical work, caused either by increased force (stress), increased shortening excursion (larger overlap zone), or a combination of the two, would result in increased FD, both the force- and length-dependent aspects of this FD mechanism could be modeled with a single scalar variable; the mechanical work done during shortening. Furthermore, since the mechanism operates on cross-bridge binding, it affects cross-bridge kinetics, and therefore both transient and steady-state aspects of FD.

4.2 Interpretation of Results

The exponential recovery function (Eq. 5) provides a means of evaluating the entire transient force recovery, and shows excellent agreement with simulation data in all cases ($R^2 > 0.99$). Furthermore, the function parameters identify important quantities of force redevelopment: the amount of recoverable force (A), the depressed force at steady-state (F_{inf}), and the exponential force redevelopment rate (k), allowing for improved insight into force recovery, and direct comparison with prior transient and steady-state FD experiments (Corr and Herzog, 2005).

Our simulations agree with the steady-state findings of previous experimental studies, showing that FD_{ss} increases with shortening amplitude (Abbott and Aubert, 1952; De Ruiter et al., 1998; Herzog and Leonard, 1997; Maréchal and Plaghki, 1979; Meijer et al., 1997; Morgan et al., 2000; Sugi and Tsuchiya, 1988), decreases with larger shortening speeds (Abbott and Aubert, 1952; De Ruiter et al., 1998; Herzog and Leonard, 1997; Maréchal and Plaghki, 1979; Meijer et al., 1997; Morgan et al., 2000; Sugi and Tsuchiya, 1988), and increases with mechanical work done during shortening (De Ruiter et al., 1998; Herzog et al., 2000). Furthermore, since this mechanism attributes FD to a decrease in the number of attached cross-bridges, there should be a proportional decrease in contractile element stiffness (Ford et al., 1981). In our model, all active shortening caused a reduction in contractile element stiffness that, in turn, reduced muscle-tendon unit stiffness. The magnitude of stiffness reduction increased with FD_{ss} (Fig. 7c) and amplitude of shortening (Fig. 4c), supporting previous experiments in which stiffness reductions were proportional to FD_{ss} in frog (Sugi and Tsuchiya, 1988) and rabbit (Joumaa et al., 2012) single muscle fibers, and shortening amplitude in human

muscles (Lee and Herzog, 2003). Therefore, the experimentally-observed decrease in stiffness (Lee and Herzog, 2003; Sugi and Tsuchiya, 1988), and the amplitude- and speed-dependent trends in FD_{ss} , can be reproduced by our model using a work-related reduction in the forward attachment (binding) rate, and associated reduction in the proportion of attached cross-bridges.

The transient aspects of FD are also well described using this model. Regression analyses identified a strong negative correlation between muscle mechanical work performed during shortening and the exponential recovery rate, k (Fig. 7b), and no correlation between work and the amount of force recovered, A : matching the regressions of prior FD experiments (Corr and Herzog, 2005). Furthermore, model simulation trends of both k and A with respect to the amplitude and speed of shortening (Fig. 5a-d) are similar to those observed experimentally (Fig. 6a-d). An increase in the work done during shortening resulted in a slower force recovery, supporting prior qualitative (De Ruiter et al., 1998) and quantitative (Corr and Herzog, 2005; Koppes et al., 2014) observations.

Alternatively, many of these steady-state findings could also be explained by a recent hypothesis that attributes FD to titin; specifically, the combination of Ca^{2+} -dependent binding of titin to actin, and cross-bridge driven winding of titin around actin (Nishikawa et al., 2012). Similar to the sarcomere length non-uniformity theory, this hypothesis uses the same mechanism to explain FD and FE. Although the winding filament hypothesis can predict the steady-state FD behaviors, it does not address the transient aspects of FD.

4.3 Limitations

As with any cross-bridge based model used to represent whole muscle, while structural information of tissue architecture and actin-myosin interactions are incorporated, some aspects of the model remain phenomenological. Ours is not a purely mechanistic model, rather a

phenomenological incorporation of a previously proposed FD mechanism (Herzog, 1998; Maréchal and Plaghki, 1979) into a structural cross-bridge model to test if this mechanism could account for both steady-state and transient FD, as well as their dependence on speed and amplitude of shortening. Our estimate of ζ , the factor relating mechanical work to the decrease in bonding rate, is phenomenological. This value is coupled with the choice of f_1 , and was selected to ensure a positive forward binding rate for all simulated experiments. While this is an arbitrary choice, it would be difficult to experimentally determine a value for ζ . To do so would require precise control of the amount of strain on actin, and observing how variations in actin strain influence cross-bridge kinetics. This is technically challenging, and beyond the scope of this project. To the best of our knowledge, such measurements cannot be achieved using current single-fiber preparations or actin-myosin assays.

This study was focused on FD, and did not include negative work production associated with lengthening (eccentric) contractions. In our model, a negative work value would act to increase the binding rate, which could possibly explain some aspects of force enhancement (FE) following active lengthening. However, we believe, and recent experimental evidence appears to support, that FE is not simply the opposite of FD (Koppes et al., 2013; Koppes et al., 2015), thus we treat these two phenomena separately.

4.4 Previous Cross-Bridge Based FD Models

A previous modified cross-bridge model of FD (Wu and Herzog, 1999) employed two mathematical adaptations: (a) a power function of the work done during shortening to determine steady-state FD, and (b) a fading memory function to slow the rate of force recovery. This model displayed great agreement with experimental data for a variety of test conditions, and seemed to capture many transient aspects of FD. However, the fading memory function was

descriptive, and did not provide insight into potential biological mechanisms for the decreased force recovery rate; unlike the cross-bridge model we present. Furthermore, steady-state FD was achieved by a vertical shift of the force (ΔF) that was fully developed in the shortening phase, and remained constant until steady-state. However, experiments have shown FD differences are greater at the end of shortening than at steady-state, suggesting that two types of FD may exist: a long-lasting steady-state FD, and an additional transient FD that develops during shortening and disappears with time (Lee and Herzog, 2003). Our model supports this finding, predicting a larger amount of FD at the end of shortening than at steady-state (Fig. 8).

This highlights a fundamental difference between these two models. In Wu and Herzog's model (Wu and Herzog, 1999), force is calculated using the classic cross-bridge model, and mathematical adjustments were made to match the depressed force at steady-state, and to slow force redevelopment to provide transient behaviors similar to those observed experimentally. However, our FD mechanism is incorporated *within* the cross-bridge model, so that the mechanical work performed acts to decrease the forward (binding) rate, thereby affecting cross-bridge kinetics, rather than correcting the force post-calculation. This allows the model to predict transient phase behavior while merely fitting it to predict steady-state behavior, or vice versa. For example, in our case, we prescribed the work-FD relation within the cross-bridge model, and obtained the correct steady-state and transient behaviors as a consequence. This lends support to the stress-induced inhibition of cross-bridge binding hypothesis, and demonstrates that the transient and long lasting effects of FD can be accounted for by a work-mediated binding rate within the classic cross-bridge theory. Taken together, these findings indicate that cross-bridge kinetics may play a critical role in the development of FD.

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TABLES

Table 1. Modeling parameters.

$L_{MT\ (opt)}$ (mm)	$L_{CE\ (opt)}$ (mm)	h (nm)	s_o (μ m)	L_{slack} (mm)	U_{max} (*)	F_{max} (*)	Γ (*)	ε_o (*)
100	32.57	27	2.4	41	0.024	1.0	1.7898	1.11

* = dimensionless

$L_{MT(opt)}$ = optimal muscle-tendon unit length; $L_{CE(opt)}$ = contractile element length at optimal muscle-tendon unit length; h = Huxley's bond length scaling parameter (range of bond length over which there is a significant probability of actin-myosin binding); s_o = sarcomere length in standard reference state; L_{slack} = tendon slack length; U_{max} = tendon strain at maximal isometric force; F_{max} = maximal isometric force; Γ = proportionality constant relating force to the first moment of the bond distribution, Q_1 ; ε_o = normalized cross-bridge bond length at point of yield.

FIGURE LEGENDS

Fig. 1. Rates of cross-bridge attachment, f , and detachment, g , as functions of the normalized cross-bridge bond length, ε . For the standard model, $f_1 = 50 \text{ s}^{-1}$, $g_1 = 1 \text{ s}^{-1}$, $g_2 = 200 \text{ s}^{-1}$, $g_3 = 2 \text{ s}^{-1}$, $\varepsilon_0 = 1.11$. For the force depression model, the forward binding rate parameter, f_1 , was replaced with a function that decreased with the mechanical work performed during shortening, $f_1 = (50 - \zeta * W) \text{ s}^{-1}$. For these simulations in which mechanical work was normalized by force at optimal length (F_{opt}), $\zeta = 10^4 \text{ (m*s)}^{-1}$. Adapted from (Cole et al., 1996; Zahalak, 1986).

Fig. 2. Schematic representation of the activated shortening protocol for the modeling simulations. The simulated cat soleus muscle-tendon unit was shortened 3, 6, and 9 mm (3, 6, 9% optimal muscle-tendon unit length ($L_{\text{MT(opt)}}$)) at speeds of 3, 9, and 27 mm/s (7, 21, and 63 % optimal fiber length/s), to the same final length. All simulated shortenings occurred on the upper ascending limb of the force-length relationship of the soleus muscle-tendon unit. For all simulations, muscles were maximally activated at time 0 s, and maintained at tetanic stimulation throughout the entire test period. This protocol was chosen to match that of a prior experimental study (Corr and Herzog, 2005).

Fig. 3. (a) Normalized force (calculated force, F , normalized by the force at optimal length, F_{opt}) vs. time data for modeling simulations of a subset of the entire test battery: three amplitudes of shortening (3, 6, 9 mm) at one constant rate of shortening (9 mm/s). Each simulation begins with a tetanic stimulation at the initial length, followed by an active shortening, and a period of transient force recovery. Steady-state force depression (FD_{ss}), defined as the difference between

the steady-state force following active shortening and the corresponding isometric reference contraction at the same final length, is shown symbolically for the three active shortenings, indicating that FD_{ss} increased with increasing shortening amplitudes. The first 1.4 s of the entire 5-s transient recovery period are shown. (b) Representative experimental data of a cat soleus muscle-tendon unit subjected to similar shortenings. Measured force, F , normalized by the force at optimal length, F_{opt} , shows similar transient and steady-state responses to those obtained by the model. Additionally, the experimental force recovery data and modeling simulations show similar trends with respect to shortening amplitude. Experimental speeds and amplitudes of shortening were identical to those simulated, however experimental shortenings began on the descending limb of the F - L relation and ended at optimal length, whereas simulations were run on the upper end of the ascending limb. Adapted from (Corr and Herzog, 2005).

Fig. 4. Mechanical work (a, b) and stiffness (c, d) with respect to amplitude and speed of shortening. The trends in values for each speed (left column) or amplitude (right column) are represented by lines, and solid bars indicate the average values; for the three speeds for each amplitude (left column), and for three amplitudes at each speed (right column). Mechanical work (a) increased with an increase in shortening amplitude (a), and decreased with faster shortening speeds (b), presumably due to lower force production, in accordance with the muscle's force-velocity relation (Hill, 1938; Leonard and Herzog, 2005). Stiffness was reduced following each activated shortening. Stiffness reductions increased with shortening amplitude (c), and decreased with shortening speeds (d). Mechanical work was defined as the dot product of the normalized force (F / F_{opt}) and velocity, and stiffness was calculated for the entire muscle-tendon unit.

Fig. 5. Trends of recovery parameters (k , A , FD_{ss}) with respect to amplitude (a, c, e) and speed (b, d, f) of shortening. Value trends for each speed (left column) or amplitude (right column) are represented by lines, and solid bars indicate the average values; for the three speeds at each amplitude (left column), and the three amplitudes at each speed (right column). The amount of recovered force, A , increased with increases in shortening amplitude (a) and shortening speed (b). The rate of force redevelopment, k , decreased with shortening amplitude (c) and increased with shortening speed (d). Steady-state FD ($FD_{ss} = F_{iso} - F_{inf}$) increased with shortening amplitude (e) and decreased with shortening speed (f). These findings indicate that k and FD_{ss} have opposite trends with respect to amplitude and speed of shortening, thus an increase in shortening amplitude, or a decrease in shortening speed, would result in a slower rise to a more depressed steady-state force.

Fig. 6. Experimental data (A , k , FD_{ss} , W) with respect to shortening amplitude and shortening speed, for cat soleus muscle-tendon units subjected to similar shortenings as those modeled herein. Left column: data pooled for all speeds of shortening ($N = 24$ per amplitude), indicating that an increase in shortening amplitude produced increases in (a) recoverable force, A (a), steady-state FD, FD_{ss} (e), and mechanical work, W (g), and a decrease in the rate of recovery, k (c). Right column: data pooled for all amplitudes ($N = 24$ per speed), indicating that an increase in the speed of shortening produced increases in A and k (b, d), and decreases in FD_{ss} and W (f, h). Data are estimated marginal means $\pm$ standard error, with significance determined by complete block ANOVA, $*P < 0.05$. Adapted from (Corr and Herzog, 2005).

1 Fig. 7. Regression analyses of stiffness (S), FD_{ss} and k with respect to mechanical work. (a):
 2 Linear regressions indicated an increase in work would decrease stiffness (circles) and increase
 3 steady-state force depression (squares). The FD_{ss} - W regression improved when fit with an
 4 exponential function ($R^2 = 0.98$). (b): Linear regression of k and mechanical work, showed that
 5 an increase in mechanical work is associated with a slower redevelopment of force. The amount
 6 of recoverable force, A , did not significantly correlate with mechanical work ($P = 0.25$, not
 7 shown). (c): A strong linear relationship between percent stiffness reduction (SD %) and
 8 percent force depression (FD %) at steady-state following active shortening, indicating that
 9 muscle stiffness decreases proportionally to the amount of steady-state FD.

10

11 Fig. 8. Comparison of FD model (black) and classic cross-bridge model (grey) subjected to
 12 identical activated shortenings (6 mm at a speed of 9 mm/s). The amount of FD is largest
 13 immediately after shortening, and decreases with time to a steady-state value.

Figure 1

Figure 2

Figure 3

Figure 4

Figure 5 3 mm/s 9 mm/s 27 mm/s

Shortening Amplitude (mm)

3 mm 6 mm 9 mm

Shortening Speed (mm/s)

Figure 6

Figure 7

Figure 8