ENTROPY IN BIOLOGY

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Heat of Shortening and Crossbridge Dynamics in Skeletal Muscle

- Discoveries of A.V. Hill: force-velocity curve and the heat of shortening
- Crossbridge dynamics: solutions of the direct problem and the inverse problem
- Brief comparison with 21st century data
- Project suggestion
- References

Heat of shortening and crossbridge dynamics in skeletal muscle

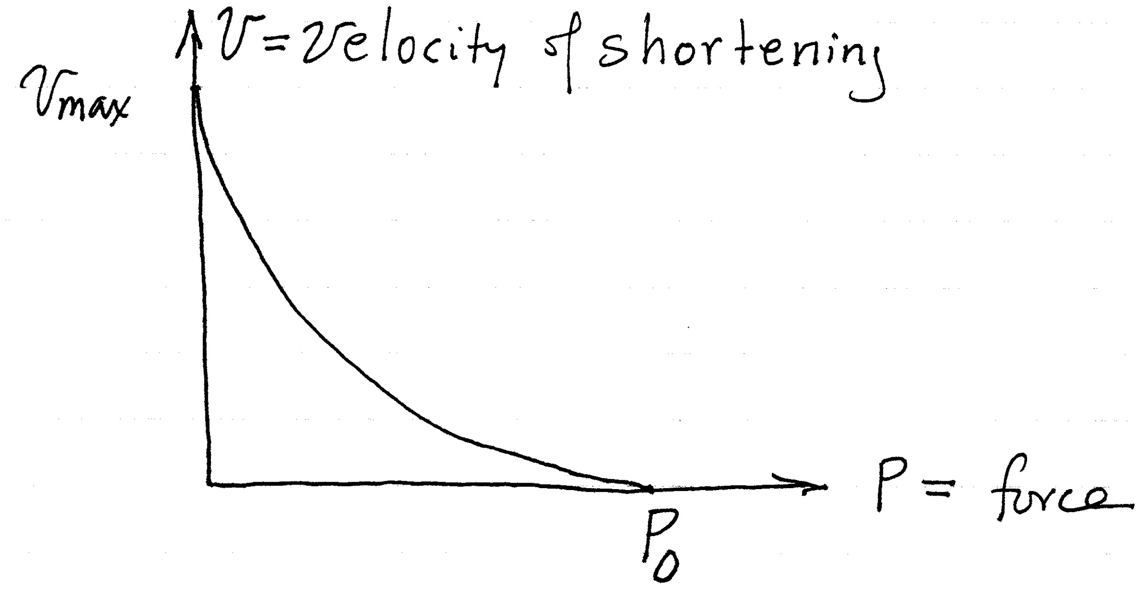
These notes are based on a paper by Lacker & Peskin (1986) in which the observations of A.V. Hill (1938) on macroscopic muscle are applied to the determination of the microscopic properties of myosin crossbridges in skeletal muscle.

We begin by summarizing the findings of A.V. Hill. These are the force-velocity curve and the heat of shortening.

The force-velocity curve describes muscle shortening at a constant velocity v against a constant force P. The relationship discovered by A.V. Hill is

$$(1) \qquad v = b\,\frac{P_0 - P}{P + a}$$

which is sketched on the next page



The three empirically determined constants are b, P_0, a. Note that b has units of velocity, and P_0 and a have units of force.

P_0 is called the isometric force. It is the force that the muscle will develop when it is not allowed to shorten.

At the opposite extreme, $V_{max} = b \frac{P_0}{a}$

is the velocity at which the muscle will shorten when there is no load at all.

An important relationship observed by Hill is that

$$\frac{a}{P_0} = \frac{1}{4} \tag{2}$$

so $v_{max} = 4b$. Two other useful forms of the force-velocity relation are

$$v(P+a) = b(P_0 - P) \tag{3}$$

and

$$P = \frac{bP_0 - av}{b + v} \tag{4}$$

The power generated by the muscle is Pv, and at either end of the force-velocity curve this is zero. If we seek $v = v_*$ to maximize the power, we get the quadratic equation

$$v_*^2 + 2b v_* - \frac{b^2 P_0}{a} = 0 \tag{5}$$

and the positive solution of this equation is

(6) $$v_* = b\left(-1+\sqrt{1+\frac{P_0}{a}}\right)$$

$$= b\left(-1+\sqrt{5}\right)$$

$$\cong \frac{5}{4}b$$

The approximation on the last line is a very good one; if you check it algebraically you will find $80 \overset{?}{=} 81$ after rearranging and squaring both sides. It is also very convenient, since $(P_0+a)/P_0 = 5/4$.

Substituting (6) into (4) we get

(7) $$P_* \cong \frac{1}{3}P_0$$

Next, we consider the heat of shortening.

When a muscle is contracting isometrically, it generates heat at a rate that we shall denote by M_0. This is called the <u>maintenance heat</u>.

When the muscle is allowed to shorten, it generates heat at a higher rate, and the difference, by definition, is the heat of shortening.

What Hill found is that the heat of shortening is proportional to velocity, and that the constant of proportionality is the constant a that appears in the force-velocity relation! Thus, according to Hill (1938) the muscle generates heat at the rate

$$M_0 + av \tag{8}$$

Hill later (1964) proposed a more complicated formula in which the coefficient of v is a linear combination of P and P_0, but we'll adopt the position that equation (8) is too beautiful not to be true!

We will need a formula for M_0, and this can be found from the observation that the heat of shortening is equal to the maintenance heat when the muscle is shortening at the velocity v_*, found above, which is chosen to deliver maximum power to the load. This gives

(9) $$M_0 = \frac{5}{4}ab = \frac{5}{16}P_0 b$$

When the muscle is shortening at velocity v against a load force P, it is doing work at the rate Pv, so the total rate at which it is consuming chemical energy has to be equal to

(10) $$M_0 + (P+a)v$$

$$= \frac{5}{16}bP_0 + \left(\frac{bP_0 - av}{b+v} + a\right)v$$

$$= \frac{5}{16} b P_0 + b(P_0 + a)\frac{v}{b+v}$$

$$= \left(\frac{5}{16} + \frac{5}{4}\frac{v}{b+v}\right) b P_0$$

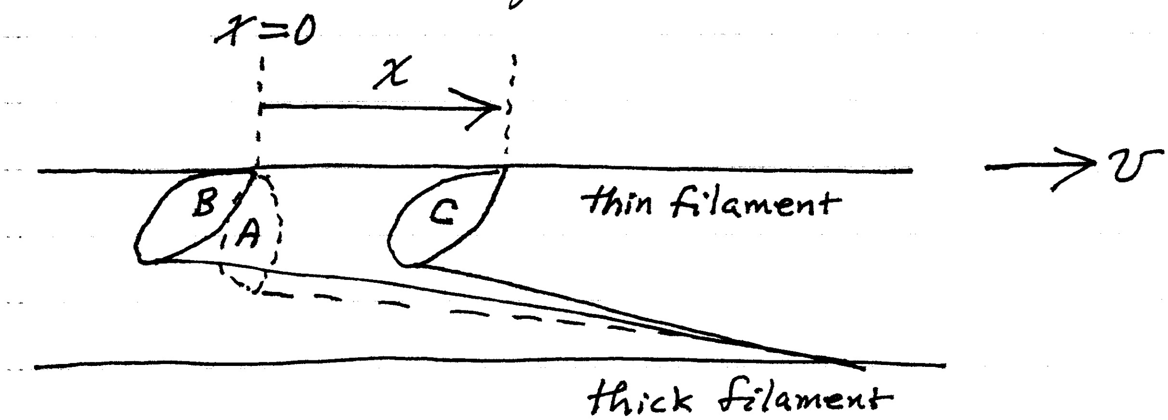
Thus, the total rate of energy consumption is an increasing function of v, and it increases by a factor of approximately 4 as v goes from 0 to $v_{max} = 4b$. (This factor becomes 5 if we let $v \to \infty$, which is unphysical but still relevant to what we will do later.)

Cross-bridge dynamics

We consider a crossbridge model of the kind introduced by Lacker, in which the thin filament is regarded as a continuum, so that myosin heads can attach anywhere along the thin filament. In this kind of model, all unattached myosin heads are equivalent.

We also assume that attachment occurs with the myosin heads in a particular configuration. For purposes of these notes, it is most natural to call this configuration $x=0$, and also to assume that x increases in the direction in which the myosin head is carried by sliding during muscle shortening. In these notes, we consider shortening only.

Considering a myosin head in a "left" half sarcomere, then, we have the following picture (with the mirror image of that picture in a right half-sarcomere):



In the figure, A shows the configuration of the myosin molecule as it attaches to the thin filament. Attachment is immediately followed by rotation of the head into a configuration B in which the tail of the myosin molecule is stretched. The transition $A \to B$ is called the power stroke. Sliding of the thin filament carries the attached myosin head in the direction of increasing x towards a configuration like C, in which the length of the tail is less than it was immediately after the power stroke, and perhaps even less than it was in the original attachment configuration A, as in the example C that is shown. At some point during the sliding process, the myosin head detaches, completing the crossbridge cycle.

In these notes, we scale everything to the level of the individual crossbridge. Thus, from now on we use P to denote the force generated by the muscle divided by the number of myosin heads in a half-sarcomere (whether or not those heads are attached). This means that P is the force that each myosin head feels on average, although individual myosin heads feel different amounts of force — zero if they are unattached, and depending on x if they are attached. Similarly v is the velocity of shortening of the muscle divided by the number of half-sarcomeres that the muscle has along its length, and this means that v is the relative velocity (positive for shortening) between thick and thin filaments in every half-sarcomere. Everything said previously about the laws discovered by A.V. Hill remain valid in terms of these cross-bridge oriented variables. The constants P_0 and a, which have units of force, are scaled to the individual cross-bridge in the same manner as P, and the constant b, which has units of velocity is scaled in the same manner as v.

The steady-state equations for the crossbridge population are as follows:

(11) $$v \frac{du}{dx}(x) = -\beta(x) u(x) \;;\; x > 0,\ v > 0$$

(12) $$v u(0) = \alpha (1 - U)$$

(13) $$U = \int_0^\infty u(x)\, dx$$

(14) $$P = \int_0^\infty p(x)\, u(x)\, dx$$

Here $u(x)$ is the crossbridge population density function with the interpretation that

(15) $$\int_{x_1}^{x_2} u(x)\,dx = \text{fraction of crossbridges which are attached and have } x \in (x_1, x_2)$$

Thus, U is the fraction of attached bridges, and $1-U$ is the fraction of un-attached bridges.

The parameter α is the probability per unit time that any particular unattached crossbridge will attach, and $\beta(x)$ is the probability per unit time that an attached crossbridge with displacement x will detach.

The parameter v is the sliding velocity of a thin filament relative to the thick filaments in its sarcomere. Note that $v = dx/dt$ for any attached crossbridge. Finally, P is the average force per crossbridge, with the average taken over the whole population, including unattached crossbridges. In the equation for P, $p(x)$ is the force generated by an attached crossbridge with displacement x.

Equations (11) & (12) can both be derived from the following steady-state relationship for the fraction of crossbridge which are attached and have displacement in $(0, x)$:

(16) $$\alpha(1-U) = v\,u(x) + \int_0^x \beta(x')\,u(x')\,dx'$$

The left-hand side of this equation describes the rate of increase of this fraction by the formation of newly attached crossbridges, all of which form at $x=0$ and are carried into the interval $(0,x)$ by sliding, and the right-hand side describes the rate of decrease of the fraction of cross-bridges in $(0,x)$ by transport out of the interval and by detachment from within the interval. In a steady state, the two sides must balance.

To derive (11) from (16), differentiate both sides of (16) with respect to x. To derive (12) from (16), just set $x=0$ on both sides of (16).

Another consequence of (16) can be obtained by letting $x \to \infty$ and assuming that $u(\infty) = 0$. We then get

(17) $$\alpha(1-U) = \int_0^\infty \beta(x)\,u(x)\,dx$$

This states that the rate of attachment and the rate of detachment of crossbridges must be equal, which is indeed a requirement that should be satisfied in a steady state.

Note that either side of (17) is the average steady-state rate at which any one crossbridge is cycling. We call this rate R. Because of equations (12) & (17), it can be evaluated in any one of three equivalent ways:

(18) $$R = v\,u(0) = \alpha(1-U) = \int_0^\infty \beta(x)\,u(x)\,dx$$

The direct problem of steady-state crossbridge dynamics can now be stated as follows. Given

$$\alpha, \ \beta(x), \ p(x), \ v$$

solve for $u(x)$, and then evaluate P and R. This is straightforward to do, although some integrals might have to be evaluated numerically.

We are concerned here, however, with the inverse problem of determining $\beta(x)$ and $p(x)$, and perhaps also the parameter α, from experimental data.

As in any inverse problem, the first step is to write out the solution to the direct problem.

From (11), we immediately have

(19) $$u(x) = u(0)\, e^{-\frac{1}{v}\int_0^x \beta(x')\,dx'}$$

Substituting this into (13), we get

(20) $$U = u(0) \int_0^\infty e^{-\frac{1}{v}\int_0^x \beta(x')\,dx'}\, dx$$

and then (12) becomes an equation for $u(0)$:

(21) $$v\, u(0) = \alpha \left(1 - u(0) \int_0^\infty e^{-\frac{1}{v}\beta(x')\,dx'}\, dx \right)$$

for which the solution is

(22) $$u(0) = \frac{\left(\frac{\alpha}{v}\right)}{1 + \left(\frac{\alpha}{v}\right) \int_0^\infty e^{-\frac{1}{v}\beta(x')\,dx'}\, dx}$$

We can now write explicit formulae for all quantities of interest:

(23) $$u(x) = \frac{\left(\frac{\alpha}{v}\right) e^{-\frac{1}{v}\int_0^x \beta(x')dx'}}{1 + \left(\frac{\alpha}{v}\right)\int_0^\infty e^{-\frac{1}{v}\int_0^x \beta(x')dx'} dx}$$

(24) $$U = \frac{\frac{\alpha}{v}\int_0^\infty e^{-\frac{1}{v}\int_0^x \beta(x')dx'} dx}{1 + \frac{\alpha}{v}\int_0^\infty e^{-\frac{1}{v}\int_0^x \beta(x')dx'} dx}$$

(25) $$1 - U = \frac{1}{1 + \frac{\alpha}{v}\int_0^\infty e^{-\frac{1}{v}\int_0^x \beta(x')dx'} dx}$$

(26) $$R = \frac{\alpha}{1 + \frac{\alpha}{v}\int_0^\infty e^{-\frac{1}{v}\int_0^x \beta(x')dx'} dx}$$

$$(27) \qquad P = \frac{\frac{\alpha}{v}\int_0^\infty p(x)\, e^{-\frac{1}{v}\int_0^x \beta(x')dx'} dx}{1 + \frac{\alpha}{v}\int_0^\infty e^{-\frac{1}{v}\int_0^x \beta(x')dx'} dx}$$

This completes the solution of the direct problem. To prepare for doing the inverse problem, we rewrite the above formulae for R and P by making the following changes of variable:

$$(28) \qquad w = \int_0^x \beta(x')dx', \quad dw = \beta(x)dx$$

$$(29) \qquad \gamma(w) = \frac{1}{\beta(x)} \quad \text{at corresponding points}$$

$$(30) \qquad q(w) = p(x) \quad \text{at corresponding points}$$

Note that w has units of velocity.

Because of (29), $dx = \gamma(w)dw$.

From (28), $w=0$ when $x=0$. We

assume that $\beta(x) > 0$ for all $x \geq 0$, so $W(x)$ is strictly increasing and invertible. It is reasonable to assume, moreover, that

(31) $$\int_0^\infty \beta(x)\,dx = +\infty$$

and in that case $W(\infty) = \infty$. It would also happen that there is some finite x_1 such that

(32) $$\int_0^{x_1} \beta(x)\,dx = +\infty$$

In that case, the integrals in the foregoing over $x \in (0, \infty)$ should be replaced by integrals over $x \in (0, x_1)$, but the domain of W is still $(0, \infty)$ since $W(x_1) = \infty$.

After making these changes of variable, the equations for R and P become

$$(33) \qquad R = \frac{\alpha}{1 + \frac{\alpha}{v}\int_0^\infty \gamma(w)\, e^{-\frac{1}{v}w}\, dw}$$

$$(34) \qquad P = \frac{\frac{\alpha}{v}\int_0^\infty g(w)\,\gamma(w)\, e^{-\frac{1}{v}w}\, dw}{1 + \frac{\alpha}{v}\int_0^\infty \gamma(w)\, e^{-\frac{1}{v}w}\, dw}$$

The strategy now is to use data on R as a function of v to determine $\gamma(w)$, and then, with $\gamma(w)$ known, to use data on P as a function of v to determine $g(w)$.

Let $\mathcal{E}_0$ be the amount of chemical energy consumed during each crossbridge cycle. Then $\mathcal{E}_0 R$ is the average rate of consumption of chemical energy per crossbridge. This chemical energy is partly transformed into work and partly into heat, and we already have an expression (10) from the research of A.V. Hill for the sum of the rates at which heat and work are being generated. From this we get

(35)
$$R = \left(\frac{5}{16} + \frac{5}{4} \frac{v}{b+v} \right) \frac{bP_0}{\varepsilon_0}$$

$$= \left(1 + 4 \frac{v}{b+v} \right) \frac{5}{16} \frac{bP_0}{\varepsilon_0}$$

$$= \left(1 + 4 \frac{v}{b+v} \right) R_0$$

where

(36)
$$R_0 = \frac{5}{16} \frac{bP_0}{\varepsilon_0}$$

is the value of R when $v=0$, i.e., the average rate of crossbridge cycling for each crossbridge when the muscle is isometric.

As for P, we have the force-velocity relation (4) which we rewrite here taking into account the relationship (2) that $a = P_0/4$:

(37) $$P = P_0 \frac{b - \frac{1}{4}v}{b + v}$$

Also, by setting

(38) $$v = \frac{1}{s}$$

We convert the integrals in (33) and (34) into Laplace transforms. Putting everything together, we have the following equations:

(39) $$1 + \alpha s \int_0^\infty \gamma(w) e^{-sw}\, dw$$

$$= \frac{\alpha}{R_0\left(1 + \frac{4}{bs+1}\right)} = \frac{\alpha}{R_0} \, \frac{bs+1}{bs+5}$$

(40) $$s \int_0^\infty g(w)\, \gamma(w)\, e^{-sw}\, dw$$

$$= \frac{P}{R} = \frac{P_0\left(\frac{bs - \frac{1}{4}}{bs+1}\right)}{R_0\left(1 + \frac{4}{bs+1}\right)}$$

$$= \frac{P_0}{4R_0}\left(\frac{4bs - 1}{bs+5}\right)$$

From (39),

(41) $$\int_0^\infty \gamma(w) e^{-sw}\, dw = \frac{1}{R_0} \frac{1}{s} \frac{bs+1}{bs+5} - \frac{1}{\alpha} \frac{1}{s}$$

$$= \frac{1}{5R_0} \left(\frac{4b}{bs+5} + \frac{1}{s} \right) - \frac{1}{\alpha} \frac{1}{s}$$

$$= \frac{1}{5\, R_0} \frac{4}{s + \frac{5}{b}} + \left(\frac{1}{5R_0} - \frac{1}{\alpha} \right) \frac{1}{s}$$

and it follows that

(42) $$\gamma(w) = \frac{4}{5\, R_0} e^{-\frac{5w}{b}} + \left(\frac{1}{5R_0} - \frac{1}{\alpha} \right)$$

This puts a restriction on α, since we require $\gamma(w) > 0$ for all $w \in (0, \infty)$:

(43) $$\alpha \geq 5R_0$$

The borderline case is especially simple and especially interesting.* Of all of the cases allowed by (43), it is the only one in which $\gamma(w) \to 0$ as $w \to \infty$, and therefore the only one in which $\beta(x)$ is unbounded from above. As we shall see, in this borderline case, there is a finite value of x, say x_1, such that $\beta(x) \to +\infty$ as $x \to x_1$. This is physically reasonable, since there should obviously be some upper limit to the amount that a crossbridge can be displaced before it detaches. If we impose as a condition that $\beta(x)$ be unbounded from above, then the only possible choice is

(44) $$\alpha = 5 R_0$$

and we assume that this is the case from now on. Then (42) becomes

(45) $$\gamma(w) = \frac{4}{5 R_0} e^{-\frac{5w}{b}}$$

*This case was mentioned in Lacker & Peskin (1986) but not studied in detail

Now recall that $\gamma(w)\,dw = dx$, and that $x = 0$ when $w = 0$. Therefore, the relationship between x and w is

(46) $$x = \int_0^w \frac{4}{5R_0} e^{-5\frac{w'}{b}}\,dw'$$

$$= \frac{4b}{25R_0}\left(1 - e^{-5\frac{w}{b}}\right)$$

and from this we see that as $w \to \infty$, $x \to x_1$, where

(47) $$x_1 = \frac{4b}{25R_0}$$

In terms of x_1, (46) becomes

(48) $$x = x_1\left(1 - e^{-5\frac{w}{b}}\right)$$

which can also be written as

(49) $$e^{5\left(\frac{w}{b}\right)} = \frac{1}{1 - \frac{x}{x_1}}$$

From this, we get

$$\beta(x) = \frac{1}{\gamma(w)} = \frac{5R_0}{4} e^{5\frac{w}{b}} \tag{50}$$

$$= \frac{5R_0}{4} \frac{1}{1 - \frac{x}{x_1}}$$

We still need to determine $g(w)$ and therefore $p(x)$. From (40),

$$\int_0^\infty g(w)\gamma(w) e^{-sw} dw \tag{51}$$

$$= \frac{P_0}{4R_0} \frac{1}{s} \frac{4bs - 1}{bs + 5}$$

$$= \frac{P_0}{20 R_0} \left(\frac{21b}{bs+5} - \frac{1}{s} \right)$$

$$= \frac{P_0}{20 R_0} \left(\frac{21}{s + \frac{5}{b}} - \frac{1}{s} \right)$$

and it follows that

(52) $$g(w)\,\gamma(w) = \frac{P_0}{20R_0}\left(21e^{-5\frac{w}{b}} - 1\right)$$

Then, from (45),

(53) $$g(w) = \frac{P_0}{16}\left(21 - e^{5\frac{w}{b}}\right)$$

and from (49), this implies

(54) $$p(x) = \frac{P_0}{16}\left(21 - \frac{1}{1-\frac{x}{x_1}}\right)$$

$$= \frac{P_0}{16}\left(20 + 1 - \frac{1}{1-\frac{x}{x_1}}\right)$$

$$= P_0\left(\frac{5}{4} - \frac{1}{16}\frac{x}{x_1-x}\right)$$

$$= \frac{5}{4}P_0\left(1 - \frac{1}{20}\frac{x}{x_1-x}\right)$$

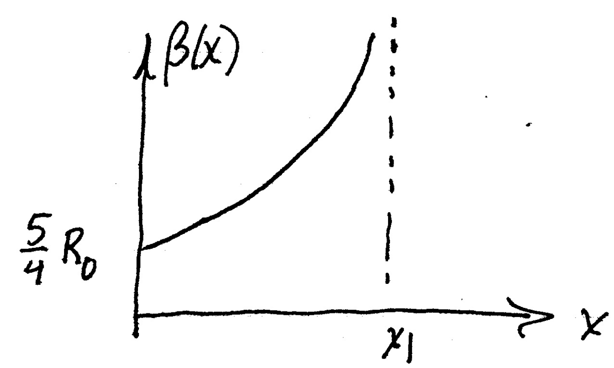
The point x_0 at which $p(x_0) = 0$ is given by

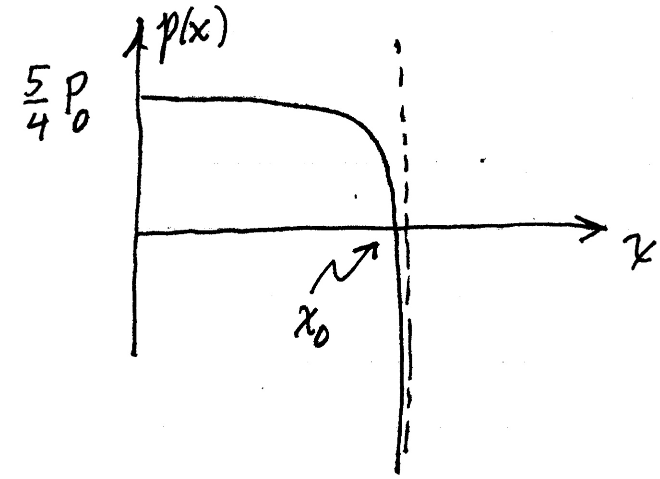
$$21 = \frac{1}{1 - \frac{x_0}{x_1}} \tag{55}$$

and this implies

$$\frac{x_0}{x_1} = \frac{20}{21} \tag{56}$$

<u>Our results can be sketched as follows:</u>





Summary of results:

(57) $$R_0 = \frac{5}{16} \frac{bP_0}{\varepsilon_0}$$

(58) $$\alpha = 5R_0$$

(59) $$x_1 = \frac{4b}{25R_0}$$

(60) $$\beta(x) = \frac{5R_0}{4} \frac{x_1}{x_1 - x}$$

(61) $$p(x) = \frac{5P_0}{4}\left(1 - \frac{1}{20}\frac{x}{x_1 - x}\right)$$

The next step is to check that the data from which we derived the cross-bridge properties are recovered when we solve the direct problem with the crossbridge model derived above.

Note that the integrals that were originally over $x \in (0, \infty)$ should now be replaced by integrals over $x \in (0, x_1)$.

The most important expression that we need to evaluate is

(62) $$e^{-\frac{1}{v}\int_0^x \beta(x')\,dx'}$$

$$= e^{-\frac{1}{v}\frac{5R_0}{4}\int_0^x \frac{x_1}{x_1 - x'}\,dx'}$$

$$= e^{-\frac{1}{v}\frac{5R_0 x_1}{4}\log\frac{x_1}{x_1 - x}}$$

$$= e^{-\frac{b}{5v}\log\frac{x_1}{x_1 - x}} = \left(1 - \frac{x}{x_1}\right)^{\frac{b}{5v}}$$

Integration on both sides over $(0, x_1)$ gives

(63) $$\int_0^{x_1} e^{-\frac{1}{v}\int_0^x \beta(x')dx'}\,dx$$

$$= \frac{-x_1}{\frac{b}{5v}+1}\left(1-\frac{x}{x_1}\right)^{\frac{b}{5v}+1}\Bigg|_0^{x_1}$$

$$= \frac{5v x_1}{b+5v}$$

Then, since $\alpha x_1 = \frac{4}{5}b$,

(64) $$\frac{\alpha}{v}\int_0^{x_1} e^{-\frac{1}{v}\int_0^x \beta(x')dx'}\,dx = \frac{4b}{b+5v}$$

(65) $$1+\frac{\alpha}{v}\int_0^{x_1} e^{-\frac{1}{v}\int_0^x \beta(x')dx'}\,dx = 1+\frac{4b}{b+5v}$$

$$= 5\left(\frac{b+v}{b+5v}\right)$$

Substituting these results into (23-26), we get

(66) $$u(x) = \frac{\frac{\alpha}{v}\left(1-\frac{x}{x_1}\right)^{\frac{b}{5v}}}{5\,\frac{b+v}{b+5v}}$$

$$= \frac{R_0(b+5v)}{v(b+v)}\left(1-\frac{x}{x_1}\right)^{\frac{b}{5v}}$$

(67) $$U = \frac{4b}{5(b+v)}$$

(68) $$1-U = \frac{b+5v}{5(b+v)}$$

(69) $$R = \frac{\alpha}{5\left(\frac{b+v}{b+5v}\right)} = R_0\,\frac{b+5v}{b+v}$$

$$= R_0\left(1+\frac{4v}{b+v}\right)$$

The above formula for R agrees perfectly with equation (35), which was our starting point for the determination of $\beta(x)$, so we have indeed confirmed that $\beta(x)$ was correctly determined.

We turn next to the evaluation of P, see equation (27). The denominator of P is given by (65), and the numerator is

(70) $$\frac{\alpha}{v}\int_0^{x_1} p(x)\, e^{-\frac{1}{v}\int_0^x \beta(x')\,dx'}\, dx =$$

$$\frac{\alpha}{v}\,\frac{5P_0}{4}\int_0^{x_1}\left(1-\frac{1}{20}\,\frac{x}{x_1-x}\right)\left(\frac{x_1-x}{x_1}\right)^{\frac{b}{5v}} dx =$$

$$\frac{\alpha}{v}\,\frac{5P_0}{4}\int_0^{x_1}\left(1-\frac{1}{20}\left(\frac{x_1}{x_1-x}-1\right)\right)\left(\frac{x_1-x}{x_1}\right)^{\frac{b}{5v}} dx =$$

$$\frac{\alpha}{v}\,\frac{5P_0}{4\cdot 20}\int_0^{x_1}\left(21\left(\frac{x_1-x}{x_1}\right)^{\frac{b}{5v}}-\left(\frac{x_1-x}{x_1}\right)^{\frac{b}{5v}-1}\right) dx =$$

$$\frac{\alpha}{v}\,\frac{5P_0 x_1}{4\cdot 20}\left(\frac{21}{\frac{b}{5v}+1}-\frac{1}{\frac{b}{5v}}\right) =$$

$$\frac{25P_0\alpha x_1}{4\cdot 20}\left(\frac{21}{b+5v}-\frac{1}{b}\right)$$

$$=\frac{25P_0\alpha x_1}{4\cdot 20}\left(\frac{20b-5v}{b(b+5v)}\right)$$

$$=\frac{25P_0}{\cancel{4}\cdot 20}\left(\frac{\cancel{4}}{5}\cancel{b}\right)\frac{20b-5v}{\cancel{b}(b+5v)}$$

$$=\frac{5}{4}P_0\,\frac{4b-v}{b+5v}$$

Dividing this by the right-hand side of (65) gives

(71) $$P=P_0\,\frac{b-\frac{1}{4}v}{b+v}=\frac{bP_0-av}{b+v}$$

since $a=P_0/4$, and this is the same as equation (4), so our check is complete.

We can use the above crossbridge theory to evalute some quantities of interest from the point of view of the crossbridge

First, consider the average force per <u>attached</u> crossbridge. This is given by

(72) $$\frac{P}{U} = \frac{P_0 \frac{b - \frac{1}{4}v}{b+v}}{\frac{4b}{5(b+v)}} = \frac{5}{4} P_0 \left(1 - \frac{v}{4b}\right)$$

Next, consider the average step length of a crossbridge from attachment to detachment. The probability of detachment in $(x, x+dx)$ conditioned on survival up to x is

(73) $$\beta(x) \frac{dx}{v}$$

and the probability density for survival up to x is

(74) $$e^{-\frac{1}{v}\int_0^x \beta(x')dx'}$$

so the probability density for detachment at x is

(75) $$\frac{\beta(x)}{v} e^{-\frac{1}{v}\int_0^x \beta(x')dx'}$$

$$= -\frac{d}{dx} e^{-\frac{1}{v}\int_0^x \beta(x')dx'}$$

Therefore, the mean step length is given by

(76) $$\int_0^{x_1} x \left(-\frac{d}{dx} e^{-\frac{1}{v}\int_0^x \beta(x')dx'}\right) dx$$

$$= \int_0^{x_1} e^{-\frac{1}{v}\int_0^x \beta(x')dx'} dx$$

$$= \frac{5v}{b+5v} x_1$$

see (63). Note that this saturates at rather small values of v. For example if $v = b = \frac{1}{4} v_{max}$, the mean step length is already $\frac{5}{6} x_1$.

PIAZZESI DATA WITH MAGENTA LINES FROM OUR THEORY SUPERIMPOSED

Piazzesi G et al. : Skeletal Muscle Performance Determined by Modulation of Number of Myosin Motors Rather Than Motor Force or Stroke Size. Cell 131(2007): 784-795

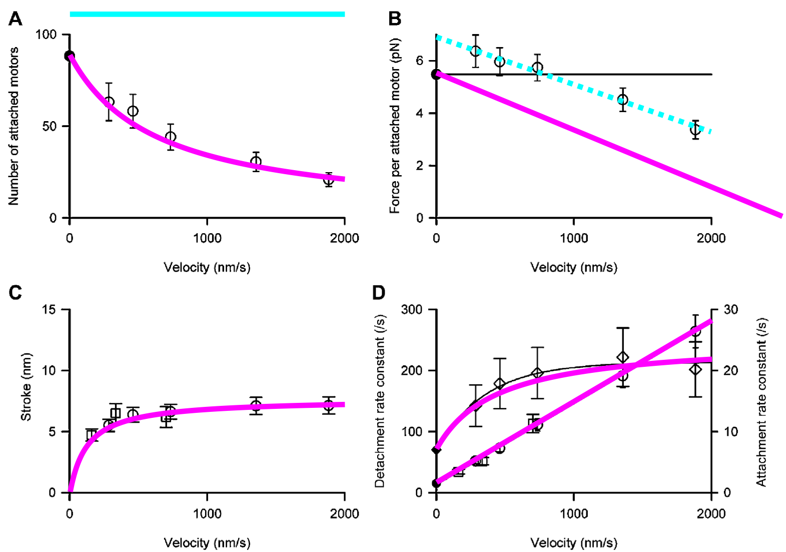


Figure 4. Molecular Basis of the Relationship between Force and Velocity during Steady Shortening

(A) Number of motors attached to actin in each myosin half-filament.

(B) Force per attached motor.

(C) Sliding distance *L* over which a motor remains attached, estimated from X-ray (squares) and mechanical (circles) data.

(D) Apparent attachment rate constants (diamonds) and detachment rate constants (squares, from X-ray data; circles, from mechanics). Error bars denote SE of mean.

Procedure used in fitting data

- Detachment rate (straight line)

in panel D is R/U. From equations (67), (69), and (59)

$$\frac{R}{U} = \frac{R_0 (b + 5v)}{\left(\frac{4}{5} b\right)}$$

$$= \frac{25}{4} R_0 \left(\frac{1}{5} + \frac{v}{b}\right)$$

$$= \frac{1}{x_1}\left(\frac{b}{5} + v\right)$$

so fit to the line determines b and x_1.

- Mean step length (panel C) is now given by equation (76) with no adjustable parameters, since b and x_1 have been determined as above. The formula for the mean step length is

$$\frac{5v}{b+5v}\, x_1$$

- <u>Number of attached bridges</u> (panel A)

Here we plot the fraction of attached bridges U, as given by equation (67):

$$U = \left(\frac{4}{5}\right) \frac{b}{b+v}$$

with b already determined but with the vertical scale chosen for agreement with the data at $v=0$. The cyan line shows the level $U = 1$ on the chosen scale. In the Piazzesi paper, it is stated that the fraction of crossbridges that are attached at $v=0$ is $\frac{3}{10}$, but

our theory gives this fraction as $\frac{4}{5}$.
Since $\frac{3}{10} = \left(\frac{3}{8}\right)\left(\frac{4}{5}\right)$, a possible explanation for the discrepancy would be that only $\frac{3}{8}$ of the crossbridges in the muscle are actually cycling under the conditions of the experiment. Muscle activation is graded and depends on the level of $[Ca^{2+}]$ in the cytoplasm. What $[Ca^{2+}]$ regulates is the availability of actin binding sites on the thin filament to myosin. If availability/non-availability occurs over segments of significant length and persists for significant time, then the

effect of incomplete activation may be equilalent to a reduction in the number of myosin motors that can participate in crossbridge cycling.

- <u>Attachment rate</u> (curve) in panel D

This is a derived quantity, calculated from the detachment rate k_{OFF} by solving

$$k_{ON}(N_0 - N_a) = k_{OFF} N_a$$

Here k_{OFF} is the data on the straight line in panel D, N_a is the number of attached bridges given by the data in panel A, and N_0 is the total number of myosin motors. Thus, the data points that are plotted are calculated according to

$$k_{ON} = k_{OFF} \frac{Na/No}{1 - Na/N_0}$$

For consistency with this, we plot

$$k_{OFF} \frac{\frac{3}{8}U}{1 - \frac{3}{8}U}$$

(see above discussion for explanation of the factor $\frac{3}{8}$.)

- Force per attached motor (panel B)

This is given by equation (72) as

$$\frac{P}{U} = \frac{5}{4} P_0 \left(1 - \frac{v}{4b}\right)$$

To plot this linear function, we simply connect the isometric data point on the vertical axis to the point $v = 4b$ on the horizontal axis, with b determined as above. This determines P_0, since $\frac{5}{4} P_0$ is the isometric value $(v=0)$.

For comparison, we do a least-squares best fit of a straight line to the data points in the figure (excluding the isometric point, which does not appear to lie on the line). The best-fit line is shown in cyan, dotted.

Although the line through the (non-isometric) data points lies above the theoretical line (through the isometric data point), the two lines have nearly the same slope.

The parameters obtained in the above manner are as follows:

$b = 630 \ nm/s \qquad (v_{max} = 2520 \ nm/s)$

$x1 = 7.61 \ nm$

$P_0 = 4.43 \ pN$

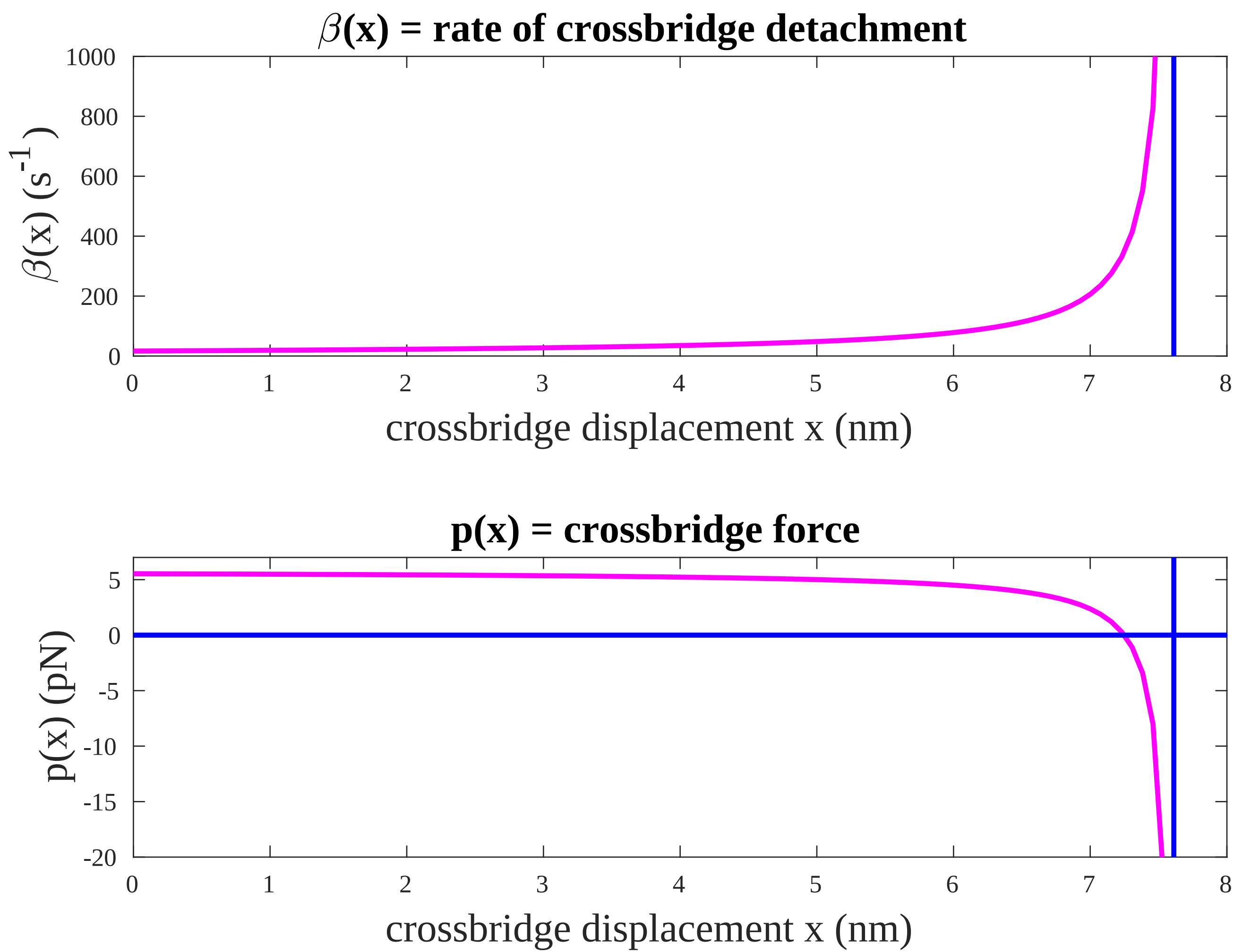
(Note that $\frac{5}{4} P_0 = 5.53 \ pN$)

$R_0 = 13.24 /s \qquad$ (Note that $\beta(0) = \frac{5}{4} R_0$)

$\alpha = 66.2 /s$

$\mathcal{E}_0 = 65.8 \ pN \cdot nm$

With the parameters identified, we get the following quantitative plots of $\beta(x)$ and $p(x)$:



β(x) = rate of crossbridge detachment
β(x) (s^-1)
1000
800
600
400
200
0
0
1
2
3
4
5
6
7
8
crossbridge displacement x (nm)
p(x) = crossbridge force
p(x) (pN)
5
0
-5
-10
-15
-20
0
1
2
3
4
5
6
7
8
crossbridge displacement x (nm)

Project Suggestion:

Simulate quick-release transients as is done in Lacker & Peskin (1986) but for the particular crossbridge model of these notes.

References (with links on CSP website, along with this lecture)

Hill AV (1938)
The heat of shortening and the dynamic constants of muscle.
Proceedings of the Royal Society B 126: 136-195

Hill AV (1964)
The effect of load on the heat of shortening of muscle.
Proceedings of the Royal Society B 159: 297-318

Lacker HM & Peskin CS (1986)
A mathematical method for the unique determination of cross-bridge properties from steady-state mechanical and energetic experiments on macroscopic muscle.
Lectures on Mathematics in the Life Sciences 16: 121-153

Piazzesi G et al (2007)
Skeletal muscle performance determined by modulation of the number of myosin motors rather than motor force or stroke size
Cell 131: 784-795