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The Role of Acidosis in Fatigue: Pro Perspective

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The etiology of muscle fatigue defined as a loss of force, velocity, and power in response to contractile activity has been studied and debated for over a hundred years, yet important questions remain either unresolved or controversial.[2,7,10] As clearly shown in published reviews by Fitts[7] and later by Allen et al.[2] and Kent-Braun et al.,[10] the causes of muscle fatigue are complex and involve multiple factors within the central nervous system and the active muscles. The factors central to the debate on the importance of low cell pH as a causative fatiguing agent are the definition of muscle fatigue, the extent of intracellular acidosis, and the changes in the cellular environment that occur in conjunction with the increase in H^+. The most pertinent established facts are that: 1) high-intensity contractile activity can cause muscle pH to decline from 7.0 to as low

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as 6.3–6.2 in multiple species, including frogs, cats, rats, and humans;[7] 2) acidosis induced by contractile activity can be correlated with muscle fatigue;[7] and 3) the fatigue-inducing effects of low pH are exacerbated by other cell changes with fatigue, especially the increase in inorganic phosphate (P_i) and the reduced amplitude of the Ca^{2+} transient.[10] Clearly, fatigue during endurance activities, such as marathon running, are not caused by low muscle pH.[7]

The primary argument against a role for high H^+ in fatigue at physiological temperatures centers on studies where only peak isometric force and velocity were evaluated, acidosis was modest (pH > 6.7), and/or intracellular Ca^{2+} was saturating.[2,17,21] Under those conditions, acidosis plays a reduced albeit still significant role in fatigue. Single fiber studies support the argument that at physiological muscle temperatures, H^+ does not have a significant role in fatigue if the degree of acidosis is limited (cell pH 6.7–7.0), P_i remains below 5 mM, and fatigue is defined as a decline in peak isometric force under conditions of saturating Ca^{2+}.[2] However, those conditions do not reflect the fatigue state that occurs in vivo, where cell pH values < 6.4 are accompanied by increases in P_i to as high as 30 mM, and the amplitude of the Ca^{2+} transient declines to μM (pCa 6.0).[1,7] Furthermore, the more important indicator of fatigue and one's ability to perform work is peak power not isometric force, and high H^+ significantly inhibits peak power even in the presence of saturating Ca^{2+} and physiological muscle temperatures.[9,11] Here, I will review the key evidence that low pH is an important causative agent in muscle fatigue during intense contractile activity.

Relationship Between Cell pH And Isometric Force

To defend a role for low pH as a causative agent in fatigue, one has to document that considerable acidosis occurs and that it shows some relationship with fatigue. The extent of the decline in cell pH is well documented and dependent on the intensity of the contractile activity and is higher in fast fibers compared with slow fibers.[7] For example, there are a number of studies showing muscle pH to decline to values as low as 6.2, which includes human studies by Hermansen and Osnes,[8] Taylor et al.,[18] and Wilson et al.[23] Additionally, a common observation is that the degree of muscle fatigue (i.e., force decline) and the rate of recovery are correlated with the extent of acidosis.[7,12]

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The correlation is not perfect because factors other than low pH contribute to fatigue, in particular depolarization of the sarcolemma due to increases in extracellular K^+ and reductions in SR Ca^{2+} release.[2,7] Nonetheless, even these factors often have a pH component due to H^+ inhibition of cell ATPases (see below).

From the late 1800s until mid-1980s, multiple studies implicated low pH as an important fatigue agent.[7] Important among them were single skinned fiber studies demonstrating high H^+ to significantly depress peak force.[7] Those studies have been questioned because they were conducted at nonphysiological temperatures (12°C–15°C), and more recent experiments conducted at 30°C (including ours) have demonstrated low pH to have a more modest effect on peak isometric force.[2,11,15] However, from these data, one cannot conclude that low pH is an unimportant fatigue agent, but only that it has a mitigated effect on peak force under saturating Ca^{2+} conditions. Saturating Ca^{2+} conditions do not mimic those that occur during fatigue, and further, these data do not consider the effect H^+ has on fiber shortening velocity and peak power. Importantly, low pH has been shown to depress velocity and power at 30°C by 30% and 35% and even peak isometric force in suboptimal Ca^{2+},[11,14] with the effects exacerbated in the presence of high P_i.

The Interaction Between Low pH, High P_i, And Depressed Ca^{2+}

In 1988, Cooke at al.[4] showed the depressive effect of low pH to be exacerbated by 20 mM P_i, and recently, we found this to be true even at 30°C and saturating Ca^{2+}.[14] Although both low pH and high P_i have a more modest (albeit still significant) effect on peak isometric force at 30°C, in combination (pH 6.2 and 30 mM P_i), these ions inhibit peak force by approximately 35% and 50% in slow and fast fiber types, respectively.[14] In addition, H^+ and P_i both individually reduce the myofibrillar Ca^{2+} sensitivity to a greater extent at 30°C compared with 15°C, with the greatest effect occurring in the presence of both ions.[15] Because Allen et al.[1] showed intracellular Ca^{2+} declines with fatigue, the effect of low cell pH on peak force is considerably worse in fatigue conditions where intracellular Ca^{2+} is suboptimal and P_i approaches 30 mM.

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The Inhibiting Effect Of Low pH On Fiber Velocity And Power

Arguably, the strongest case for the importance of low pH as a fatigue agent is the observation that high H^+ significantly depresses peak power.[11] Although low pH but not P_i significantly depressed V_{max}, both ions individually reduced type II fiber peak power by 18%.[5,11] In combination, these ions depressed fiber V_{max} by 31% and peak power by 59% (Fig. 1).[14] These results agree with those of Karatzaferi et al.[9] who reported a 55% decrease in fast fiber peak power under similar conditions. Interestingly, when myosin light chain 2 was phosphorylated, a condition that likely exists in fatigued fibers, the inhibition of peak power was increased to 70%.[9] Additionally, similar to the findings on peak isometric force, the possibility exists that the pH depression in fiber shortening velocity and peak power would be magnified by suboptimal Ca^{2+} conditions that exist in fatigued fibers. For example, with fiber shortening > 10%, low Ca^{2+} is known to depress fiber shortening velocity.[13]

Multiple Contributing Factors For A Role Of Low pH In Fatigue

The inhibiting effects of low pH on Ca^{2+} sensitivity, velocity and peak power alone support the conclusion of acidosis being an important fatiguing agent. However, these are not the only reasons H^+ induces fatigue; other factors of importance include inhibition of fiber ATPases, including the myofibril, SR, and Na^+–K^+ ATPases. Specifically, inhibition of the myofibril ATPase contributes to the reduced fiber V_{max}, whereas SR and Na^+–K^+ pump inhibition contribute to a reduced SR Ca^{2+} uptake and surface membrane depolarization, respectively.[2,10] The former reduces the SR lumen $[Ca^{2+}]$ and hence Ca^{2+} release with activation, whereas the latter contributes to excitation–contraction coupling failure. Finally, low pH reduces fast fiber efficiency causing the cell to use more ATP for a given force production.[10] This would accelerate the development and extent of fatigue.[10]

In summary, low muscle pH is an important fatigue agent during high-intensity exercise where cell pH falls below 6.6, and the effects are exacerbated by a parallel decline in intracellular Ca^{2+} and increased P_i. Although these effects have been documented in skinned

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fibers, it will be important for future studies to confirm their importance in human studies involving in vivo fatigue paradigms.

Responses and rebuttal

In his article Westerblad[19] argues that any fatigue inducing agent should change in parallel with the decline in contractile function, and that this is not always the case for acidosis. However, this criterion would only be true if an increased hydrogen ion concentration was the only cause of fatigue, which is not the case.[2,7] Other factors, such as sarcolemma depolarization and reduced SR Ca^{2+} release, contribute to fatigue and reduce the correlation between pH with force and/or power. In the Degroot et al.[6] article cited by Dr. Westerblad,[19] pH decreased immediately postexercise while force partially recovered. Clearly, the initial recovery was independent of any direct effect of pH. Thus, as the authors concluded in this particular paradigm, pH was not the only cause of fatigue. In another example of the multifaceted nature of fatigue, we showed that skeletal muscle force recovers from high-frequency stimulation in two phases, an initial fast phase unrelated to pH, and a second slower phase highly correlated ($P = 0.94$) to pH recovery.[12] Another important consideration is that pH-induced inhibition of the Na^+–K^+ and SR ATPases that contribute to fatigue are no longer relevant during recovery where even low pump function can maintain the Na^+ and K^+ gradients across the cell and SR Ca^{2+} uptake. Thus, in fatigue paradigms, where these pumps are inhibited, some early recovery in force would be expected despite no recovery in pH.

Dr. Westerblad[19] cites two articles in which fatigue was produced in isolated single fibers without any significant change in pH or in which it was not accelerated when the fibers were acidified as evidence against low pH as a fatigue agent.[3,20] However, the protocol used to induce fatigue in these studies has been shown to disrupt excitation–contraction coupling and lead to loss of force at low frequencies.[22] These studies demonstrate that fatigue can result from factors other than an elevated $[H^+]$ but do not disprove an important role for H^+ in exercise conditions where significant acidosis occurs.[7]

Dr. Westerblad notes that low pH has less of a depressive effect on peak isometric force at near physiological temperature going from approximately 50% at 10°C to approximately 20% at 30°C.[17] Nonetheless, the loss of force is still significant and under conditions

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existing with fatigue becomes considerably more important. For example, the effect of low pH on force and power are exaggerated by high inorganic phosphate (P_i) for which we and Karatzaferi et al.[9] have shown power to be depressed by approximately 60%. The latter study showed an even greater 70% depression in power in the pH 6.2, 30 mM P_i condition when the myosin light chain 2 was phosphorylated, an effect not observed with high P_i and pH 7 condition, which demonstrates the importance of low pH in depressing peak power.

In considering the importance of acidosis in fatigue, it is important to consider the effect H^+ has on depressing myofibrillar Ca^{2+} sensitivity. Recently, we found the H^+ induced depression of Ca^{2+} sensitivity to be considerably greater at 30°C compared with 15°C.[15] This effect takes on added importance in fatigue where the amplitude of the Ca^{2+} transient is reduced, and fibers are operating on the steep portion of the force–$[Ca^{2+}]_i$ relationship.[2] This means that the inhibition of force by H^+ is considerably greater than observed under saturating Ca^{2+} conditions as pH 6.2 has reduced the pCa_{50} from 6.58 to 5.36 in fast type IIx fibers.[15] Dr. Westerblad questions our results by noting that the Ca^{2+} concentration required to achieve pCa_{50} increased approximately 5-fold at 30°C, an increase much larger than the 3.6-fold increase reported by Palmer and Kentish.[16] However, the main difference between the two studies was the Ca^{2+} sensitivity in the pH 7.0 condition for which pCa_{50} was 6.58 and 5.86, and this difference can be attributed at least in part to the higher temperature used in our study as temperature increases myofibrillar Ca^{2+} sensitivity.[15] Importantly, the pCa_{50} values reported at pH 6.2, which is the Ca^{2+} required to elicit 50% of peak force, were essentially identical for the fast psoas fibers in the Palmer and Kentish[16] study and our fast rat gastrocnemius fibers being 5.30 and 5.36, respectively.[15] Additionally, the increase in free Ca^{2+} required to obtain pCa_{50} under fatigue conditions in both studies was similar at 3.63 and 4.36 μM, and the difference can be attributed to the higher Ca^{2+} sensitivity at pH 7 at 30°C compared with 25°C.[15] Dr. Westerblad suggests that our approximately 15-fold increase in Ca^{2+} required to reach pCa_{50} under fatigue conditions would require an approximately 7.5-fold increase in tetanic Ca^{2+} which is incompatible with the measured decline of tetanic Ca^{2+} in the fatigued state.[2] His reasoning for this statement is not clear, because the important

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criteria is not the delta change in pCa_{50} between pH 7 and 6.2 but the actual Ca^{2+} concentration required under fatigue conditions to reach a given % of peak force. In our study and the article of Palmer and Kentish,[16] the $[Ca^{2+}]$ required to reach pCa_{50} at pH 6.2 are essentially identical. In our work and that of Palmer and Kentish,[16] the H^+-induced decline in myofibrillar Ca^{2+} sensitivity would cause force to decline below 50% of that observed in the nonfatigued state.

Conclusions

Acidosis induces fatigue due to the direct effects on cross-bridge force, velocity, and power and by suppressing myofibrillar Ca^{2+} sensitivity. The latter is particularly important in fatigue for which the amplitude of the Ca^{2+} transient is reduced and fibers are operating on the steep portion of the force–$[Ca^{2+}]_i$ relationship. The force and power inhibiting effects of low pH are greater than that observed under saturating Ca^{2+} conditions and are further exacerbated by the associated increase in P_i.

The present discussion does not constitute endorsement by ACSM.

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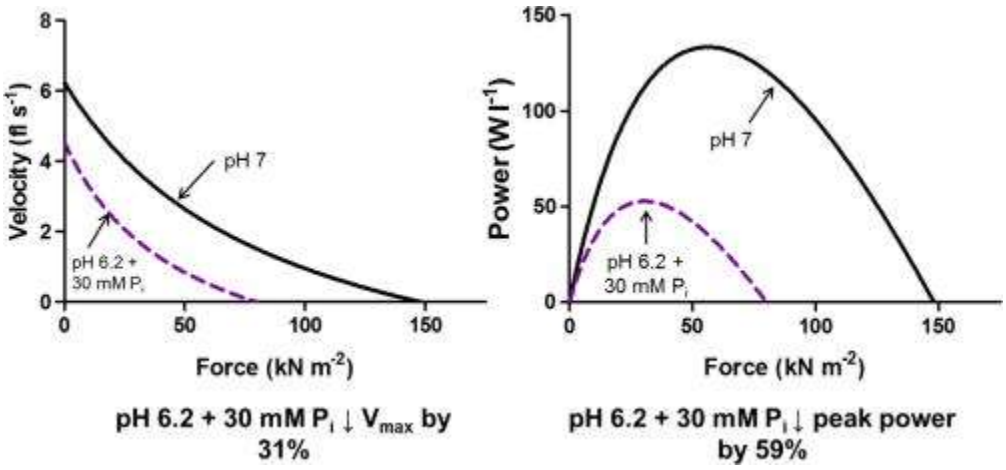

Figure 1. Force–velocity and force–power curves obtained from rat type II fibers in control pH 7.0 and pH 6.2 + 30 mM P_i conditions at 30°C. Shortening velocity (fiber lengths per second) and power (watts per liter) are plotted as a function of the force expressed relative to the fiber cross-sectional area ($kN \cdot m^{-2}$). Data were adapted and reprinted from Nelson et al. Am J Physiol Cell Physiol 2014;307:C939–50.

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