



Discussion/ Explanation of Figure:

The results show that inserting fixed isometric Ca2+ transients into work-loop contractions can shift the work-loop end-systolic curve substantially leftward. The leftward shift is capable of unify isometric and work-loop end-systolic force-length curves, with the magnitude of the shift proportional to the width of the fixed Ca2+ transient.

Contraction-mode differences in intracellular Ca2+, therefore, can sufficiently explain the differing end-systolic curves for isometric vs. work-loop contractions. To explain the underlying mechanism for this phenomenon, we will focus on answering two questions:

1. Why is the intracellular Ca2+ transient contraction-mode dependent?
2. How can an adjustment to the intracellular Ca2+ contraction unify the isometric and work-loop end-systolic curves?

Why is the intracellular Ca2+ transient contraction-mode dependent?

**0ms:** **Figure 8, A** shows that work-loop and isometric contractions start with the accumulation of intracellular Ca2+. At this point isometric and work-loop intracellular Ca2+ transients are essentially equal. Generation of force has not started.

Generating an isometric end-systolic force-length curve requires varying the initial (preset) sarcomere length, while work-loop contractions always start at the same sarcomere length. Thus, unless an isometric contraction occurs at 2.3 (the maximum sarcomere length for this paper), its force-equivalent work-loop will start with a larger sarcomere length.

**18ms:** At 18ms, force generation begins. The rate of Force development is larger for the work-loop contraction than it is in the isometric contraction **(Figure 8, C)**. This is because the rates governing cross-bridge state change from “non-permissive” to “permissive” is dependent on sarcomere length. Thus, the initial value for sarcomere length affects the force profile of a contraction.

The force profile affects the concentration of free, intracellular Ca2+ via apparent Ca2+ binding to troponin.

So the first part of our answer is, the different starting lengths of sarcomeres undergoing work-loop or isometric contractions influences the rate of force generation. The contraction-mode dependent force profile further influences the process of Ca2+ binding to troponin. If the flux of Ca2+ binding to troponin-C increases, there is less free intracellular Ca2+. The result is a steeper recovery phase on the intracellular Ca2+ transient (a “thinner” transient).

The “width” of the intracellular Ca2+ transient is largely dependent on the initial length of the sarcomere undergoing contraction. Since initial sarcomere length is contraction-mode dependent, the shape of the Intracellular Ca2+ expresses behaviour that is contraction-mode dependent.

How can an adjustment to the intracellular Ca2+ contraction unify the isometric and work-loop end-systolic curves?

Our previous question and answer addresses the cause of differing intracellular Ca2+ transients for isometric and work-loop contractions (seen in Figure 5). Our results show that inserting a fixed isometric Ca2+ transient into a work-loop contraction can increase duration of isotonic shortening during the work-loop.

Essentially, we are concerned with understanding the mechanism that allows intracellular Ca2+ concentration to influence the magnitude of cellular force. In the Rice-Tran model, the intracellular Ca2+ concentration affects regulatory troponin. By influencing regulatory troponin, Ca2+ affects the probability of a cross-bridge going from a non-permissive state to a permissive state. For a cross-bridge to achieve a state of force production, it must first pass from the non-permissive to permissive state. Hence, Ca2+ influencing the rates between the “N” and “P” states also influence the probability of a cross-bridge reaching a force-producing state.