Control of skeletal RyR channels by proteins in the SR lumen

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Excitation contraction (EC) coupling is the mechanism linking depolarization of the surface membrane with Ca^{2+} release from the sarcoplasmic reticulum (SR) and is the process which initiates skeletal and cardiac muscle contraction. Ca^{2+} release during EC coupling is triggered by a transverse-tubule action potential. The SR Ca^{2+} release channel the ryanodine receptor (RyR) - has recruited a surface membrane Ca^{2+} channel (the dihydropyridine receptor) as a voltage sensor to detect the action potential, and Ca^{2+} binding protein calsequestrin (CSQ) to sense the environment inside the SR. Both the skeletal (RyR1) and cardiac (RyR2) isoforms of the RyR form the hub of giant macromolecular complexes with transmembrane, luminal and cytoplasmic proteins. The activity of the channels is set by the integrated effects of associated proteins, ligands, covalent modification by redox reactions and phosphorylation. Luminal interactions, such as that with CSQ, are essential for communicating the $[Ca^{2+}]$ in the store to the RyR, but are not well understood. We have identified several key luminal mechanisms which control RyR1 activity and communication of store load to the RyR. CSQ regulates the RyR1 via two distinct processes, physically coupling either directly to the RyR or to the channel via anchoring proteins triadin and junctin. CSQ can inhibit native RyR1s (via triadin and junctin) under physiological Ca^{2+} conditions (1 mM), but it directly activates the purified RyR1. Additionally, we have found that CSQ regulation of RyR1 channel activity allows CSQ to communicate the SR store Ca^{2+} load to the RyR in a phosphorylation-dependent manner. Phosphorylation of CSQ is important not only for RyR1 regulation, but it also influences the Ca^{2+} binding capacity of CSQ (which in turn, influences the size of the SR Ca^{2+} store). We have defined junctin as the key anchoring protein required in mediating the inhibition imposed by CSQ on the RyR1. In addition, we have shown that CSQ is able to regulate the way in which RyR1 responds to changes in luminal Ca^{2+} concentration. CSQ is now defined as a luminal Ca^{2+} sensor for RyR1. These different mechanisms of CSQ regulation of the RyR1 allow for a complex regulation system that is sensitive to changes in the cellular environment. The formation of a luminal protein complex between triadin, junctin, CSQ and the RyR1 allows enhancement of Ca^{2+} release from the SR when the store is fully loaded but protects the SR from Ca^{2+} depletion when the Ca^{2+} load falls. The results of current research highlight the complex nature of luminal regulation of the RyR1 and the importance of luminal SR proteins in maintaining store load and in controlling processes leading to muscle contraction.