EXCITATION-CONTRACTION COUPLING IN SKELETAL MUSCLE

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Summary

In striated muscle contraction is under the tight control of myoplasmic calcium concentration ($[Ca^{2+}]_i$). The elevation in $[Ca^{2+}]_i$ and the consequent binding of calcium to intracellular regulatory proteins leads to the shortening of the muscle fibers. During relaxation calcium ions are removed from the myoplasmic space.

Calcium ions at rest are stored in the sarcoplasmic reticulum (SR) from which they are rapidly released upon the depolarization of the sarcolemmal and transverse (t-) tubular membranes of the muscle cell. The proteins responsible for this controlled and fast release of calcium are the dihydropyridine receptor (DHPR) and the calcium release channel found in the membranes of the t-tubules and of the terminal cisternae of the SR, respectively.

This chapter draws an up-to-date picture of the events that occur between T-tubular depolarization and the release of calcium from the SR.

1. Introduction

In skeletal muscle fibers, as in all muscle cells of our body, the interaction of contractile proteins, organized into the thin and thick filaments, is responsible for the shortening and the production of force. Skeletal muscle fibers use the changes in intracellular calcium concentration ($[Ca^{2+}]_i$) to regulate the interaction of the contractile proteins. An increase in $[Ca^{2+}]_i$ will result in shortening, while the decrease of $[Ca^{2+}]_i$ to its resting level will relax the muscle fiber. Skeletal muscle fibers have developed an intricate process to couple information arriving from the central nervous system to cell shortening. These steps include the electrical excitation of the muscle fiber, the increase in $[Ca^{2+}]_i$ and the contraction, and were collectively termed *excitation-contraction coupling* (ECC) in the middle of the last century.

Fifty years of intense research have identified and characterized almost every step in ECC. Neuronal impulses are transmitted from the axon terminal onto the muscle fiber at highly specialized regions of the surface membrane, the neuro-muscular junction (see *Muscle Energy Metabolism*). Here acetylcholine released from the axon terminal causes the opening of ionic channels (nicotinic acetylcholine receptors) and, consequently, the depolarization of the surface membrane.

Skeletal muscle fibers resemble other excitable cells in the structure and function of their surface membrane outside the neuro-muscular junction. The surface membrane contains, among others, specialized ion channels (voltage gated sodium, potassium calcium and chloride channels) and pumps (Na^+-K^+-ATP-ase) to maintain a resting membrane potential of –90 mV and to produce action potentials if stimulated. The depolarization produced in the neuro-muscular junction thus initiates propagating action potentials that travel along the surface membrane. At certain points (at the connection of the A and I bands in mammals) the surface membrane invaginates and forms the delicate network of Transverse- (T-) tubules that ramify the entire muscle fiber. As we will see later, T-tubules play an essential role in the transmission of electrical information onto intracellular processes. Since the T-tubules contain all necessary ion channels to produce an action potential, the electrical signal enters the T-tubules and reaches the innermost parts of muscle fiber much faster than diffusion would allow. Most likely T-tubules have evolved for exactly this purpose, i.e. to ensure rapid activation of the entire volume of the muscle fiber.

The depolarization of the surface and T-tubular membranes plays a crucial role in muscle function. Both the amplitude and the time necessary to develop a contraction depend strongly on the membrane potential. Experimental evidence clearly shows that depolarizing the cell to, or beyond –50 mV from the resting value of –90 mV (using voltage clamp techniques or simply by increasing the concentration of extracellular potassium) will initiate force production. Increasing the size of the depolarization will result in larger contractions that develop faster. Maximal force is reached around -10 mV, but the time to reach the peak is further decreased if depolarization is taken beyond –10 mV.

Comparing the time course of an action potential, the depolarization, with the evoked increase in force (or tension, which is force divided by cross section) reveals a 2-4 ms

delay, the so-called latency, in the latter process. This is the time necessary to convert the electrical signal into an increase in $[Ca^{2+}]_i$, for the calcium ions to reach, by simple diffusion, the regulatory binding sites on the protein troponin C and for the initiation of the interaction of the contractile proteins.

The source of calcium ions that participate in the regulation of contraction is different in different muscle types. In cardiac muscle calcium enters the intracellular space from the external environment through voltage gated calcium channels (see *Heart*). Skeletal muscle, on the other hand, will continue to contract if calcium is removed from the extracellular solution. The source of calcium is an intracellular organelle, the *sarcoplasmic reticulum* (SR). The SR surrounds the thin and thick filament and forms an intracellular network. Two morphologically and functionally distinct parts can be identified on the SR, the longitudinal tubules (LT) and the terminal cisternae (TC).
As the actual steps and the underlying molecular mechanisms in ECC became more and more clear, the meaning of the term itself changed. It was originally used, as mentioned before, to refer to all steps between muscle excitation and contraction. At present, as the molecular interactions within the thin and thick filaments have been described, ECC refers to the events that link membrane depolarization to the increase in $[Ca^{2+}]_i$. As such ECC is specialized signal transduction that links external stimuli to changes in $[Ca^{2+}]_i$.

Throughout the following sections the term ECC will be used in this, more restricted sense. We will, therefore, focus on the molecular events that connect T-tubular depolarization to the release of calcium from the SR. Two key molecules, the voltage sensor of ECC and the calcium release channel, and their interaction will be discussed.

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Biographical Sketches

László Kovács obtained his medical degree in Debrecen in 1963. He became a faculty member of the Department of Physiology at the University Medical School of Debrecen. He completed his postdoctoral training at the University of Rochester (N.Y.); later he was a visiting professor at the University of Maryland in Baltimore. He has been a full professor since 1988, and he was the chairman of the Department from 1991 to 2005. He has studied the membrane properties of excitable and non-excitable cells, the mechanism of signal transmission through membranes, and intracellular pathways. Together with his colleagues he gave the first, quantitative description of the voltage dependence and kinetic properties of the changes in intracellular calcium concentration during activation of skeletal muscle fibres. He characterized the properties of intracellular calcium binding sites, and that of the calcium pump in the sarcoplasmic reticulum membrane, analysed the feed-back processes between the voltage sensor in the surface membrane and the calcium release channels. Later he extended his studies to clinical problems by learning the molecular details of different diseases where the alterations of ionic channels or the intracellular signalization pathway are the most important elements of the pathomechanism. Therefore, he investigated the properties of cultured skeletal muscle cells derived from biopsies of patients suffering in hereditary diseases, and the significance of the intracellular protein kinase C izosymes in the proliferation and differentiation of skeletal muscle fibres and of human skin derived cell line. His scientific achievement was appreciated by electing him member of the Hungarian Academy of Sciences in 1998 and member of the Academia Europaea in 2004. In 2006 he became the head of the Research Centre for Molecular Medicine in the University of Debrecen.

László Csernoch obtained his MSc in physics from the Kossuth Lajos University of Debrecen in 1985. He then joined the Department of Physiology of the University Medical School of Debrecen where he received his PhD in 1990. As a postdoctoral fellow of the Muscular Dystrophy Association he spent two years at the Department of Biological Chemistry, University of Maryland, Baltimore. He was twice invited as a visiting professor to the Department of Physiology, Claude Bernard University, Lyon, to the Department of Molecular Biophysics and Physiology, Rush Medical University, Chicago and to the Department of Physiology and Biophysics, Robert Wood Johnson Medical School, Piscataway. Since 2005 he has been a professor and the chair of the Department of Physiology at the University of Debrecen. His major scientific interest is studying the excitation-contraction coupling of skeletal muscle fibres, focusing on the regulation of calcium release from the sarcoplasmic reticulum. Together with his colleagues he gave a quantitative description of the delayed component of intra-membrane charge movement. He was the first to detect voltage-evoked elementary calcium release events in mammalian skeletal muscle. He has also characterized the purinergic signalling and its role in the regulation of the calcium homeostasis in developing skeletal muscle cells. He extended his studies to other excitable and to non-excitable cells and described the role of purinergic signalling on keratinocytes, on the outer hair cells of the inner ear and on melanoma cells, among others. He was the first to report the over-expression of the ryanodine receptor/calcium release channel in melanoma cells and to describe its interaction with the purinergic signalling pathway and possible role in tumour-genesis. He is the member of the Hungarian Physiological Society, The Physiological Society and the Biophysical Society. His achievements were recognized with the Széchenyi István Award.