Connections Count
Excitation-Contraction Meets Excitation-Transcription Coupling

Mark E. Anderson

It is increasingly clear that cell-signaling systems serve multiple functions. Signaling molecules that play a highly visible functional role in muscle by regulating intracellular Ca\(^{2+}\) ([Ca\(^{2+}\)\)]), homeostasis and contraction are also coupled to the genetic machinery of the cell and thereby shape the repertoire of expressed proteins. Our emerging understanding of excitation-contraction coupling (ECC) follows this theme (Figure). ECC occurs when Ca\(^{2+}\) entry, mostly through L-type Ca\(^{2+}\) channels, activates Ca\(^{2+}\) release from channels guarding the content of [Ca\(^{2+}\)], stores. In vascular smooth muscle, 2 channel types are important: ryanodine receptors that operate by a Ca\(^{2+}\)-induced Ca\(^{2+}\) release mechanism\(^3\) and inositol trisphosphate (IP\(_3\)) receptors that are sensitized by [Ca\(^{2+}\)], but open after binding IP\(_3\).\(^4\) The situation in cardiomyocytes seems to be somewhat simpler, as only the ryanodine receptors have a demonstrated role in ECC. ECC is mainly controlled by short-term signaling events to regulate the continuous ebb and flow of activator [Ca\(^{2+}\)]\(_i\), that is necessary for cycling myofilament crossbridge formation. Nevertheless, the molecular machinery of ECC also regulates the transcriptional activity of the cell over a much longer time scale by a process termed excitation-transcription coupling (ETC).\(^5\)

An important question in signal transduction is how ETC mechanisms in muscle screen out the constant [Ca\(^{2+}\)]\(_i\), fluctuations to deliver cogent instructions to the nucleus. One answer to this question is that transcription factors have distinct response characteristics that may refine the message content of [Ca\(^{2+}\)]\(_i\) oscillations. In B lymphocytes, the transcriptional regulatory proteins nuclear factor-κB, JNK, and NFAT are differentially activated by brief Ca\(^{2+}\) signals of high magnitude compared with prolonged Ca\(^{2+}\) signals of lower magnitude.\(^6\) Location is also important. Regulatory proteins may be anchored (by specific binding proteins) or confined to distinct intracellular domains (eg, the nucleus) that experience very different Ca\(^{2+}\) signals than those measured in the bulk cytoplasm. Connections are essential for success. Ca\(^{2+}\) entry through L-type Ca\(^{2+}\) channels seems to constitute a “privileged” pathway that can selectively couple to the Ca\(^{2+}\)-binding protein calmodulin for signaling Ca\(^{2+}\)-dependent transcriptional events in neurons\(^9\) or for excitation-secretion coupling in chromaffin cells.\(^10\) These possibilities are only now beginning to be explored in the cardiovascular system.

To understand how we might manipulate the connections between ECC and ECT, it is necessary to know where and how these 2 systems interact. Ca\(^{2+}\)-activated kinases and phosphatases are important linking molecules that can coordinate interactions between ECC and ECT. After activation by increased [Ca\(^{2+}\)], these enzymes alter the phosphorylation state of Ca\(^{2+}\)-regulatory protein complexes to directly modulate ECC and act on Ca\(^{2+}\)-dependent transcription factors that “tune” ECC over a longer time frame by affecting expression of ECC regulatory proteins. This proposed linkage has important implications for disease: cardiomyopathy has been associated with disordered expression of several key Ca\(^{2+}\) homeostatic proteins, including the Na\(^+\)-Ca\(^{2+}\) exchanger SR Ca\(^{2+}\) ATPase\(^11,12\) and numerous sarcomeric proteins.\(^13\) Expression of a constitutively active form of the Ca\(^{2+}\) and calmodulin-activated phosphatase calcineurin causes profound hypertrophy and dilated cardiomyopathy.\(^14\) whereas constitutive expression of calmodulin kinase (CaMK) IV results in a hypertrophic phenotype with less pronounced systolic dysfunction (Eric Olson, personal communication, November 1999). Expression of some CaMK isoforms is increased in human heart failure\(^15\) and atrial fibrillation,\(^15\) diseases linked to heart rate and Ca\(^{2+}\)-dependent electrical remodeling. Electrical remodeling in heart failure is associated with action potential prolongation\(^16\) that may itself be an important stimulus for CaMK activation\(^17\) by increasing the duration of the [Ca\(^{2+}\)]\(_i\), transient.\(^18\) CaMK also participates in ECC by modulating Ca\(^{2+}\) entry through L-type Ca\(^{2+}\) channels\(^19\) and by regulating uptake\(^20\) and release of Ca\(^{2+}\) from intracellular stores.\(^21,22\) These examples may illustrate important consequences of pathophysiological ECC-ECT interactions.

The study by Cartin et al\(^24\) in this issue of Circulation Research links the phosphorylation of the cAMP response element binding (CREB) protein and consequent induction of c-fos to CaMK-dependent and Ca\(^{2+}\)-independent signal transduction. In addition to enhancing our conceptual understanding of the ECC-ETC link in vascular smooth muscle, this study nicely highlights one important experimental issue in signaling research: the details of cellular ultrastructure matter. Cartin et al were the first to report the paradoxical effect of suppressing Ca\(^{2+}\) sparks in vascular smooth muscle\(^25\) that results in increased [Ca\(^{2+}\)], because of inactivation of a Ca\(^{2+}\)-activated cell membrane K\(^+\) current. In contrast, the [Ca\(^{2+}\)]\(_i\), transient is virtually ablated in cardiomyocytes under conditions where sparks are eliminated. Thus, the interactions...
Interaction of ECC (solid arrows) and ECT (dashed arrows) may occur at different levels within the cell. The cell membrane (1) is the site of proteins, such as the voltage-gated L-type Ca$^{2+}$ channel, that govern Ca$^{2+}$ entry into the cell. Ca$^{2+}$ entry is further regulated at the cell membrane by other Ca$^{2+}$-activated ion channels (square) and exchangers (pentagon) that help determine cell membrane potential. Adenylate cyclase (AC) is a membrane-associated enzyme that catalyzes the production of cAMP from ATP. AC isoforms are differentially sensitive to membrane-regulated at the cell membrane by other Ca$^{2+}$, and CaMK. The stars indicate ultrastructural regions where CaMK has been localized and demonstrated to affect ECC or ETC. Intracellular Ca$^{2+}$ stores (2) are triggered by Ca$^{2+}$ entry to release Ca$^{2+}$ from ryanodine receptors (diamond) through a Ca$^{2+}$-induced Ca$^{2+}$ release mechanism that further refines ECC in vascular smooth muscle by sensitizing the IP$_{3}$-gated channel (rectangle) to increase Ca$^{2+}$ release. Release of Ca$^{2+}$ from intracellular stores allows contraction (ECC) and activation of Ca$^{2+}$-dependent phosphatases and kinases (ETC) and may regulate the nuclear pore protein complex (ETC) (3). Ca$^{2+}$-dependent regulation of the nuclear pore is likely to be important for determining entry of signaling molecules in transit to the nucleus and regulating egress of mRNA to the cytoplasm. Activation of Ca$^{2+}$-dependent transcription factors in the nucleus (4) initiates production of new proteins for regulation of Ca$^{2+}$ entry and uptake and release of Ca$^{2+}$ from intracellular stores.

of ECC and ETC are dependent on specific cellular environments. One of the important questions that follow from the present work is the identity of the CaMK type responsible for CREB phosphorylation. Cartin et al found that both CaMK II and IV were present in vascular smooth muscle. CaMK IV is thought to be predominantly nuclear, whereas CaMK II may exist in the cytoplasm or nucleus, depending on the isoform mix of the heteromultimerized holoenzyme, and both types can phosphorylate CREB. The consequences of CREB phosphorylation by CaMK II may be inhibitory, because CaMK II can effectively phosphorylate a second negative regulatory site (Ser142). However, both CaMK II and IV can phosphorylate the activating site (Ser133), suggesting that CaMK IV may have determined c-fos levels in these experiments. Details of the probable coregulation of CaMK-mediated CREB phosphorylation by phosphatases in vascular smooth muscle also remain to be elucidated. Finally, linkage of ECC and ETC by kinases and phosphatases offers the possibility of novel therapeutic tools to address cardiovascular disease.

Acknowledgments

This work was supported by grants from the National Institutes of Health (HL03727 and HL62494) and the American Heart Associa-

tion Southeast Affiliate. Drs Jeffrey R. Balser (Departments of Anesthesia and Pharmacology, Vanderbilt University Medical Center), Roger J. Colbran (Department of Molecular Physiology, Vanderbilt University Medical Center), Sabina Kupershmidt (Department of Pharmacology, Vanderbilt University Medical Center), and Dan Roden (Departments of Medicine and Pharmacology, Vanderbilt University Medical Center) provided helpful comments and criticisms.

References


Connections Count: Excitation-Contraction Meets Excitation-Transcription Coupling
Mark E. Anderson

Circ Res. 2000;86:717-719
doi: 10.1161/01.RES.86.7.717

The online version of this article, along with updated information and services, is located on the World Wide Web at:
http://circres.ahajournals.org/content/86/7/717

Permissions: Requests for permissions to reproduce figures, tables, or portions of articles originally published in Circulation Research can be obtained via RightsLink, a service of the Copyright Clearance Center, not the Editorial Office. Once the online version of the published article for which permission is being requested is located, click Request Permissions in the middle column of the Web page under Services. Further information about this process is available in the Permissions and Rights Question and Answer document.

Reprints: Information about reprints can be found online at:
http://www.lww.com/reprints

Subscriptions: Information about subscribing to Circulation Research is online at:
http://circres.ahajournals.org/subscriptions/