Calcium Channel Diversity in the Cardiovascular System

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The flux of calcium ions (Ca2+) into the cytosol, where they serve as intracellular messengers, is regulated by two distinct families of Ca2+ channel proteins. These are the intracellular Ca2+ release channels, which allow Ca2+ to enter the cytosol from intracellular stores, and the plasma membrane Ca2+ channels, which control Ca2+ entry from the extracellular space. Each of these two families of channel proteins contains several subgroups. The intracellular channels include the large Ca2+ channels ("ryanodine receptors") that participate in cardiac and skeletal muscle excitation-contraction coupling, and smaller inositol trisphosphate (InsP3)-activated Ca2+ channels. The latter serve several functions, including the pharmacomechanical coupling that activates smooth muscle contraction, and possibly regulation of diastolic tone in the heart. The InsP3-activated Ca2+ channels may also participate in signal transduction systems that regulate cell growth. The family of plasma membrane Ca2+ channels includes L-type channels, which respond to membrane depolarization by generating a signal that opens the intracellular Ca2+ release channels. Calcium ion entry through L-type Ca2+ channels in the sinoatrial (SA) node contributes to pacemaker activity, whereas L-type Ca2+ channels in the atrioventricular (AV) node are essential for AV conduction. The T-type Ca2+ channels, another member of the family of plasma membrane Ca2+ channels, participate in pharmacomechanical coupling in smooth muscle. Opening of these channels in response to membrane depolarization participates in SA node pacemaker currents, but their role in the working cells of the atria and ventricle is less clear. Like the InsP3-activated intracellular Ca2+ release channels, T-type plasma membrane channels may regulate cell growth. Because most of the familiar Ca2+ channel blocking agents currently used in cardiology, such as nifedipine, verapamil and diltiazem, are selective for L-type Ca2+ channels, the recent development of drugs that selectively block T-type Ca2+ channels offers promise of new approaches to cardiovascular therapy.

Calcium ions (Ca2+), which can be viewed as the most important of nature’s signal transducers, participate in excitation and contraction of cardiac and vascular smooth muscle (for review see ref. 1-3). This critical signaling role reflects both the electric and chemical properties of Ca2+. Because it is a cation, Ca2+ carries an electric charge when it crosses the high resistance plasma membrane that separates the extracellular and intracellular spaces. As a result, Ca2+ generates depolarizing electric currents when it enters the negatively charged cytosol of cells at rest. The participation of Ca2+ in chemical signaling is made possible by its ability to bind tightly, and with considerable specificity, to high affinity Ca2+-binding sites on specialized proteins within the cell.

The passage of Ca2+ from the extracellular space into the cell interior is made possible by the opening of members of an extended family of ion-selective channels, which allows this ion to diffuse passively across the plasma membrane. The driving force for this downhill process is a large electrochemical gradient, which reflects the fact that extracellular Ca2+ concentration is in the millimolar range, whereas cytosolic Ca2+ concentration in most cells at rest is about 10,000 times lower, less than 1 μmol/liter. This Ca2+ concentration gradient, along with the electrical gradient that favors Ca2+ fluxes into the negatively charged cell interior, provides a powerful electrochemical force that drives Ca2+ through the open plasma membrane channels.

The present review focuses on a newly recognized diversity in the plasma membrane Ca2+ channels that participate in signal transduction in the cardiovascular system. Different responses of the various classes of Ca2+ channels to newly discovered Ca2+ channel blocking drugs offer novel mechanisms for the treatment of cardiovascular disease.

Extracellular and Intracellular Calcium Cycles

Two distinct Ca2+ cycles, both of which control the entry and removal of Ca2+ from the cytosol, can participate in cell signaling (Fig. 1). The most primitive is the extracellular cycle, in which Ca2+ enters and leaves the cytosol by crossing the plasma membrane from what is, in effect, an unlimited store of Ca2+ in the extracellular space. A second Ca2+ cycle is seen in more specialized cells, such as adult cardiac myocytes, where Ca2+ is pumped into and out of limited stores contained within an intracellular membrane system. The latter, called the sar-
Figure 1. Schematic diagram showing (top) key structures and (bottom) major calcium ion (Ca\(^{2+}\)) fluxes involved in cardiac excitation-contraction coupling in adult cardiac myocytes. The thickness of the arrows indicates the magnitude of the Ca\(^{2+}\) fluxes; their direction describes the "energetics" of the Ca\(^{2+}\) fluxes (downward arrows describe passive Ca\(^{2+}\) fluxes, and upward arrows describe energy-dependent Ca\(^{2+}\) transport). Ca\(^{2+}\) enters the cell from the extracellular fluid by way of plasma membrane (plasmalemmal) Ca\(^{2+}\) channels (A); although most of this Ca\(^{2+}\) triggers calcium release from the sarcoplasmic reticulum, a small portion directly activates the contractile proteins (A\(_1\)). Ca\(^{2+}\) transport back into the extracellular fluid involves two plasma membrane systems: Na\(^+\)/Ca\(^{2+}\) exchange (B\(_1\)) and the plasmalemmal Ca\(^{2+}\) pump (B\(_2\)). The sarcoplasmic reticulum membrane regulates two Ca\(^{2+}\) fluxes: Ca\(^{2+}\) release from the subsarcolemmal cisternae by way of the intracellular Ca\(^{2+}\) release channels ("ryanodine receptors") (C) and active Ca\(^{2+}\) uptake from the subsarcolemmal cisternae by way of the intracellular Ca\(^{2+}\) release channels. Coupling of the Action Potential to Ca\(^{2+}\) Release in Cardiac and Skeletal Muscle

All muscle cells recognize a rise in cytosolic Ca\(^{2+}\) concentration as a signal to contract, but the source of this activator Ca\(^{2+}\) differs among various muscle types. In more primitive myocytes, such as are found in vascular smooth muscle and the embryonic heart, contraction depends on Ca\(^{2+}\) that enters the cytosol from the extracellular space (the extracellular Ca\(^{2+}\) cycle). In contrast, the highly specialized myocytes of skeletal muscle and the adult heart derive most of their activator Ca\(^{2+}\) from intracellular stores in the sarcoplasmic reticulum (the intracellular Ca\(^{2+}\) cycle). Dependence on Ca\(^{2+}\) release from intracellular stores requires a functional link between depolarization of the plasma membrane (the action potential) and the opening of intracellular Ca\(^{2+}\) release channels in the sarcoplasmic reticulum. In skeletal muscle this link is effected by a mechanical coupling, in which plasma membrane depolariza-
called pharmacomechanical coupling. As noted later, the alpha-adrenergic agonists and angiotensin II, this process is systems that are activated by such chemical mediators as contraction. Because InsP3 is generated by signal transduction in delivering the activator Ca2+ that initiates smooth muscle nels, which bind to and are opened by InsP3, play a major role coupling. The InsP 3 receptor intracellular Ca2+ release chan-
nels, which bind to and are opened by InsP3, play a major role in initiating the slow contractile movement removes a “plug” that, in the muscle at rest, occludes the intracellular Ca2+ release channel. The plug itself is a portion of an L-type plasma membrane Ca2+ channel (see later) that opens and closes the Ca2+ release channel in the sarco
cystem in response to changes in the electric potential across the plasma membrane. A different member of the family of L-type plasma membrane Ca2+ channels couples a change in membrane potential to Ca2+ release from the cardiac sarco
cystem. In the heart, the action potential initiates contraction when the opening of a limited number of plasma membrane L-type Ca2+ channels results in localized increases in cytosolic Ca2+ concentration, called “Ca2+ sparks” (4). The latter cause a much greater Ca2+ release from the sarco
cystem by opening adjacent intracellular Ca2+ release channels in a process often referred to as “Ca2+-induced Ca2+ release” (5,6).

**Intracellular Ca2+ Release Channels**

Intracellular Ca2+ release channels, whose structure differs considerably from that of the Ca2+ channels in the plasma membrane, are members of a single family that includes at least two classes of related proteins (Table 1, for review see ref. 7–10). The ryanodine receptor Ca2+ channels, which predominate in skeletal and cardiac muscle, deliver the large amount of Ca2+ that mediates the final step in excitation-contraction coupling. The InsP3 receptor intracellular Ca2+ release channels, which bind to and are opened by InsP3, play a major role in delivering the activator Ca2+ that initiates smooth muscle contraction. Because InsP3 is generated by signal transduction systems that are activated by such chemical mediators as alpha-adrenergic agonists and angiotensin II, this process is called pharmacomechanical coupling. As noted later, the InsP3-gated Ca2+ channels often participate in signal transduction systems other than those that activate muscle contraction.

Both the ryanodine receptors and InsP3 receptors are tetrameric structures, each subunit of which includes a large cytosolic domain and two alpha-helical transmembrane segments. The ryanodine receptors of cardiac muscle are slightly smaller than those of skeletal muscle, and there is only ~66% identity between the amino acid sequences of these proteins. Although the InsP3 receptors are only about half the size of the ryanodine receptors, there is considerable homology between these two classes of intracellular Ca2+ release channel. Their three-dimensional structures are quite similar in that the four subunits in both ryanodine receptors and InsP3 receptors are believed to surround a central pore through which activator Ca2+ is released when the channel is opened.

The InsP3 receptors, whose single-channel conductance is less than that of the larger ryanodine receptors (11), are the predominant Ca2+ channels that initiate the slow contractile responses in smooth muscle (12). The more explosive contractile responses of cardiac and skeletal muscle are initiated by the opening of the higher conductance ryanodine receptors. Although the function of the small numbers of InsP3-regulated Ca2+ channels found in the heart is not clearly understood, evidence that they slowly release only a small amount of Ca2+ suggests that they do not participate in the Ca2+ cycles that trigger excitation-contraction coupling. They may, instead, participate in the regulation of rest tension (diastolic “tone”). An intriguing possibility is that the InsP3 receptors, like the T-type Ca2+ channels discussed later, also regulate cell growth. This hypothesis is supported by evidence that InsP3, along with other inositol phosphates, participates in a number of signaling cascades, including some that are activated when tyrosine kinase–linked receptors bind to a variety of growth factors and

<table>
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<th>Channel Type</th>
<th>Location</th>
<th>Opened By</th>
<th>Major Function</th>
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<td>L-type Ca2+ channel “plug”</td>
<td>Intracellular Ca2+ cycle</td>
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<td>L-type Ca2+ channel Ca2+ entry</td>
<td>Excitation-contraction coupling</td>
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<tr>
<td>Inositol triphosphate receptors</td>
<td></td>
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<tr>
<td>Cardiac muscle</td>
<td>Inositol triphosphate</td>
<td>Pharmacomechanical coupling</td>
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<tr>
<td>Smooth muscle</td>
<td>Inositol triphosphate</td>
<td>? Growth regulation</td>
<td></td>
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<tr>
<td>Plasma membrane Ca2+ channels</td>
<td>Smooth muscle</td>
<td></td>
<td></td>
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<tr>
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<td>SA node</td>
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*See also Table 2. AV = atrioventricular; Ca2+ = calcium ion; SA = sinoatrial.
Table 2. Plasma Membrane Ion Channels That Participate in Electrical Signaling (the action potential)

<table>
<thead>
<tr>
<th>Channel Type</th>
<th>Major Function</th>
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<tr>
<td>Na⁺</td>
<td>Action potential upstroke</td>
</tr>
<tr>
<td>Ca²⁺</td>
<td>Action potential plateau (cardiac muscle)</td>
</tr>
<tr>
<td>K⁺</td>
<td>Repolarization</td>
</tr>
<tr>
<td>Cl⁻</td>
<td>Repolarization</td>
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Ca²⁺ = calcium ion; Cl⁻ = chloride ion; K⁺ = potassium ion; Na⁺ = sodium ion.

cytokines. Because the InsP₃-regulated Ca²⁺ channels are opened by signaling cascades that activate nuclear transcription factors, this class of intracellular Ca²⁺ release channels may play an important role in regulating cell growth, differentiation and perhaps programmed cell death (apoptosis).

**Plasma Membrane Ion Channels**

The ion-selective channels in the plasma membrane, which are generally named for the ions that they carry (Table 2), are complex proteins made up of as many as five subunits, called alpha₁, alpha₂, beta, gamma and delta (for reviews, see ref. 13–17). Central to their physiologic function are the large proteins (called alpha or alpha₁ subunits in different channels) that make up and surround the channel pore (Fig. 2). These subunits, which probably arose from a single primitive ancestor through gene duplication and divergence, determine ion specificity and contain both the activation and inactivation gates that allow changes in membrane voltage to open, close and inactivate the channels. The alpha and alpha₁ subunits of plasma membrane channels, like the major proteins in the intracellular Ca²⁺ release channels, are tetramers. However, their structures differ considerably. Unlike the four protein subunits that make up the intracellular Ca²⁺ channels, which contain two alpha-helical transmembrane segments (see earlier), each of the four domains in the alpha or alpha₁ subunits of most plasma membrane ion channels contains six alpha-helical transmembrane segments (Fig. 2). Moreover, the N-terminal regions of the intracellular Ca²⁺ release channels, which project into the cytosol as electronmicroscopically visible “feet,” are much larger than the N-terminal regions of the plasma membrane channels.

The four domains of the alpha and alpha₁ subunits of most plasma membrane Na⁺ and Ca²⁺ channels are encoded by a single gene, as are adjacent regions of a single large membrane protein (Fig. 3). In contrast, the four domains in most K⁺ channels are encoded by separate genes and so not linked covalently, which allows exchanges among these domains to provide for a large number of channel subtypes. In some K⁺ channels, the major subunits are smaller peptides that consist of only a portion of the ancestral channel domain (18,19).

A number of homologies are found among the plasma membrane ion channels, notably in the S₁ alpha-helical transmembrane segment, which is rich in charged amino acids and so serves as the “voltage sensor” that mediates the voltage-dependent conformational change that activates the channel. Other homologies among the plasma membrane ion channels are found in the “pore” region, which is made up of the S₅ and

**Figure 2.** Four members of the family of ion channel proteins. The major subunits of the calcium (Ca²⁺) and sodium (Na⁺) conductance channels are tetramers made up of our four covalently linked domains numbered I to IV. The voltage sensor that initiates contraction in depolarized mammalian skeletal muscle resembles the Ca²⁺ channel, but it is smaller because it lacks a portion of the C-terminal amino acid sequence of the latter. Potassium (K⁺) conductance channels also contain four domains, but unlike the domains of the Ca²⁺ and Na⁺ channels, domains I to IV of the K⁺ channels are not covalently linked through a continuous peptide chain. C = carbon terminal; N = nitrogen terminal. Modified from Katz (1) with permission.

**Figure 3.** An ion channel domain. The ion channels depicted in Figure 2 are tetramers made up of four domains, each of which contains six alpha-helical transmembrane segments. Known ion channel domains are believed to have evolved from a common ancestral protein. The S₁ segment, which is rich in positively charged amino acids, is believed to open the channel in response to membrane depolarization. The transmembrane segments S₅ and S₆, along with the intervening peptide chain, probably surround the pore through which ions cross the lipid barrier in the core of the membrane bilayer. C = carbon terminal; N = nitrogen terminal. Adapted from Katz AM, N Engl J Med 1993;328:1244–51. Copyright 1993, Massachusetts Medical Society. All rights reserved.
S, alpha-helical transmembrane segments and intervening sequence of amino acids.

The pore region in these ion channels is of considerable pharmacologic interest because variations among the amino acids in this region alter such variables as the specificity and voltage-dependence of drug binding to the channels. Organic molecules are now being designed to interact with specific amino acid sequences in this key region of a number of ion channels (for review, see ref. 20). This structural approach to drug development offers exciting prospects for new therapies to treat diseases in which ion channels play a pathogenic role. In view of the central role of Ca\(^{2+}\) in signal transduction, new approaches to modifying Ca\(^{2+}\) channel function seem especially promising.

**Plasma Membrane Ca\(^{2+}\) Channels**

The plasma membrane Ca\(^{2+}\) channels provide the major pathways for Ca\(^{2+}\) entry into myocardial and most smooth muscle cells (for review see ref. 8 and 21–26). The major subunits, called α\(_1\), are a single large protein that contains four covalently linked domains (Fig. 2 and 3). However, not all members of this family respond in the same way to a change in membrane potential. Whereas membrane depolarization opens a pore in most Ca\(^{2+}\) channels, in the case of the skeletal muscle protein the most important effect of depolarization is to initiate a conformational change that removes a “plug” that, in the muscle at rest, occludes the pore of an adjacent intracellular Ca\(^{2+}\) release channel. Thus, even though Ca\(^{2+}\) entry through the skeletal muscle plasma membrane Ca\(^{2+}\) channel is too slow to provide an effective signal for excitation–contraction coupling, by opening intracellular Ca\(^{2+}\) release channels this protein still plays a central role in Ca\(^{2+}\) signal transduction.

From a clinical standpoint, the most important members of this family are the L-type Ca\(^{2+}\) channels (Table 3), which bind the familiar classes of Ca\(^{2+}\) channel blockers (dihydropyridines such as nifedipine, phenylalkylamines such as verapamil, and benzothiazepines such as diltiazem). Closely related is the voltage sensor of skeletal muscle, which is also an L-type Ca\(^{2+}\) channel. Other classes of plasma membrane Ca\(^{2+}\) channels are referred to as T, N, P, Q and R (8,27) (Table 3). Complete structural information is not available for all members of this family of Ca\(^{2+}\) channels, but as far as is now known, these membrane proteins are homologous both to each other and to the extended family of plasma membrane ion channel proteins listed in Table 2 (14,27,28).

**L-type calcium channels.** The most abundant of the plasma membrane Ca\(^{2+}\) channels in skeletal and cardiac muscle cells are the L-type Ca\(^{2+}\) channels which, as discussed earlier, play a key role in the intracellular Ca\(^{2+}\) cycle by opening the intracellular Ca\(^{2+}\) release channels. Ca\(^{2+}\) entry through the L-type Ca\(^{2+}\) channels in the working cells of the atria and ventricles contributes to the plateau of the cardiac action potential as well as providing the Ca\(^{2+}\) that opens the intracellular Ca\(^{2+}\) channels. In the SA node, Ca\(^{2+}\) entry through L-type Ca\(^{2+}\) channels contributes to pacemaker activity, whereas members of this class of Ca\(^{2+}\) channels in the AV node are essential for AV conduction. In smooth muscle, other L-type Ca\(^{2+}\) channels participate in the extracellular Ca\(^{2+}\) cycle by controlling the Ca\(^{2+}\) entry that binds to and so directly activates the contractile apparatus. L-type Ca\(^{2+}\) channels are also found in endocrine cells, where they regulate the Ca\(^{2+}\) entry responsible for excitation–secretion coupling, and in some neurons, where they control transmitter release. As noted earlier, it is the L-type Ca\(^{2+}\) channels whose openings are inhibited by the familiar classes of clinically available Ca\(^{2+}\) channel blockers.

**T-type Ca\(^{2+}\) channels.** The T-type Ca\(^{2+}\) channels are found in a variety of tissues, including the heart and vascular smooth muscle. Like most L-type Ca\(^{2+}\) channels, T-type Ca\(^{2+}\) channels are opened by membrane depolarization; however, studies of single-channel openings using patch clamp methods have shown several differences between the gating of L- and T-type Ca\(^{2+}\) channels. One of these is the more rapid inactivation of the T-type than of the L-type Ca\(^{2+}\) channels, which means that once the membrane is depolarized, the T-type Ca\(^{2+}\) channels close and become refractory (inactivate) more quickly. This feature is responsible for the transient openings of the T-type Ca\(^{2+}\) channels, and thus their name (T = transient, L = long-lasting).

The threshold of the T-type Ca\(^{2+}\) channels is lower than that of the L-type channels, which allows the former to be opened by smaller depolarizations than are needed to open the L-type Ca\(^{2+}\) channels. Because their low threshold allows depolarization to begin when membrane potential has deviated only slightly from the resting potential, this property is well suited for the participation of T-type Ca\(^{2+}\) channels in pace-
The relatively high ratio of T-type Ca$^{2+}$ to L-type channels in the rabbit SA node (31) is also consistent with a role for the former in pacemaker activity. The importance of the depolarizing current carried by T-type Ca$^{2+}$ channels in generating the upstroke of the action potential is less clear in the working cells of the atria and ventricles, where large inward currents are carried by more abundant and more rapidly opening Na$^+$ channels (see later).

The distribution of L-type and T-type Ca$^{2+}$ channels is quite different in various types of cardiac muscle. The highest density of T-type Ca$^{2+}$ channels is found in cells, like nodal and embryonic cardiac myocytes, that lack a prominent transverse tubular system, whereas the ratio of L-type to T-type Ca$^{2+}$ channels is much higher in cells containing these structures (32). The transverse tubules, which facilitate transmission of the depolarizing signal generated by an action potential into large cardiac myocytes that are specialized for contraction (1), are rich in the specialized junctions that provide the functional link between the plasma membrane Ca$^{2+}$ channels and the intracellular Ca$^{2+}$ release channels. The low density of T-type Ca$^{2+}$ channels in these structures therefore suggests that these channels do not play a primary role in excitation-contraction coupling. The finding that dihydropyridine Ca$^{2+}$ channel blockers, which inhibit L-type but not T-type Ca$^{2+}$ channels (32), prevent Ca$^{2+}$ release from the sarcoplasmic reticulum (33) also supports the view that the heart's T-type Ca$^{2+}$ channels play little role in cardiac excitation-contraction coupling. Further evidence that T-type Ca$^{2+}$ channels are not important for cardiac excitation-contraction coupling is their very low density in cardiac myocytes that are specialized for contraction. In the guinea pig heart, they are <10% of the density of the L-type Ca$^{2+}$ channels and 1% of that of Na$^+$ channels (34). However, in smooth muscle cells that lack abundant Na$^+$ channels, the T-type Ca$^{2+}$ channels can play an important role in activating contraction.

The finding that the depolarizing current carried by the T-type Ca$^{2+}$ channels in guinea pig ventricular myocytes is concurrent with the much larger Na$^+$ current during the upstroke of the cardiac action potential (33) argues against an important role for the T-type Ca$^{2+}$ channels in generating the action potential upstroke. Unlike the brief depolarizing current carried by the T-type Ca$^{2+}$-channels, which is almost entirely "buried" in the much larger Na$^+$ currents, the currents carried by the L-type Ca$^{2+}$ channels are more long-lasting. For this reason, although L-type Ca$^{2+}$ channels play only a minor role in the initial depolarizing current during the action potential upstroke in cells that contain large numbers of Na$^+$ channels, they make an important contribution to the slow inward Ca$^{2+}$ current that occurs later, during the plateau of the action potential.

The data just reviewed suggest that the T-type Ca$^{2+}$ channels in the heart are not of primary importance either as charge carriers or as mediators of excitation-contraction coupling, and so play a quite different role in signal transduction than do the L-type Ca$^{2+}$ channels. Several lines of evidence indicate that the cardiac T-type Ca$^{2+}$ channels participate in signal transduction pathways that regulate cell growth and proliferation. For example, T-type Ca$^{2+}$ channels are abundant in the fetal heart (35), and the densities of T- and L-type Ca$^{2+}$ channels are regulated differently during maturation, when the rate of growth slows markedly. Whereas L-type channel density remains fairly constant throughout early postnatal development, the number of T-type channels is greatest during periods of rapid growth, decreasing after growth ceases in the adult (36-40). Evidence for a growth-promoting effect of T-type Ca$^{2+}$ channel activation is found in reports that expression of this class of Ca$^{2+}$ channels increases when growth factors impinge on the heart; for example, when atrial myocytes are stimulated by growth hormone (41), and in hypertrophied myocytes isolated from both the pressure-overloaded left ventricle (42) and a genetic cardiomyopathy (43). Ca$^{2+}$ influx through channels whose threshold resembles that of T-type Ca$^{2+}$ channels has also been implicated in the proliferative response mediated by platelet-derived growth factor (44). The finding that Ca$^{2+}$ entry through T-type Ca$^{2+}$ channels is enhanced by endothelin-1, which induces myocardial hypertrophy and the expression of muscle specific genes (45), is also consistent with a role for these channels in regulating protein synthesis. Further evidence that activation of T-type Ca$^{2+}$ channels promotes cell growth is provided by recent evidence (46) that a specific blocker of these channels has an antiproliferative effect on damaged blood vessels.

A clue as to how T-type Ca$^{2+}$ channels might play a role in growth-regulating signal cascades is found in a report (47) that these channels can be activated by diacyglycerol, an intracellular signalling molecule that is released by phospholipase C. The latter is a lipolytic enzyme that plays a prominent role in the regulation of protein synthesis and cell proliferation. The finding that phorbol esters, which mimic the growth-promoting effects of diacyglycerol, also activate cardiac T-type Ca$^{2+}$ channels (47) provides additional evidence that these channels regulate cell growth. Furthermore, angiotensin II, an extracellular messenger that, when bound to AT$\text{}_{1}$ receptors, initiates a growth response that is mediated in part by diacyglycerol (48), also increases the opening of T-type Ca$^{2+}$ channels in adrenal cells (49). Conversely, stimulation of AT$\text{}_{2}$ receptors—which, when bound to angiotensin II, have been reported to inhibit growth (50)—decreases the current carried by T-type Ca$^{2+}$ channels (51). Similarly, atrial natriuretic peptide, which inhibits adrenal T-type Ca$^{2+}$ channels, blocks aldosterone secretion (52). Further evidence that Ca$^{2+}$ entry through T-type Ca$^{2+}$ channels can stimulate protein synthesis is suggested by a report that tetradrine, an antihypertensive plant alkaloid that blocks T-type Ca$^{2+}$ channels, inhibits steroidogenesis by adrenal cells (53), and that mibebradil (Ro 40-5967), another Ca$^{2+}$ channel blocker that under physiologic conditions is selective for T-type Ca$^{2+}$ channels, inhibits neointima formation after vascular injury (46).

The availability of selective T-type Ca$^{2+}$ channel blockers is helping to define other physiologic roles of these ion channels in the cardiovascular system. One such drug, U-88779E, has a neuroprotective effect after cerebral ischemia (54). Vasodila-
tor (55,56) and antihypertensive effects (57,58) seen after administration of the T-type Ca\textsuperscript{2+} channel blocker mibefradil indicate that the Ca\textsuperscript{2+} that enters arteriolar smooth muscle through T-type Ca\textsuperscript{2+} channels helps to maintain tone in these resistance vessels. In the heart, as expected of a T-type Ca\textsuperscript{2+} channel blocker, mibefradil inhibits the sinus pacemaker but has neither a significant inhibitory effect on AV node conduction (59) nor a negative inotropic effect (59-62). Mibefradil has also been reported (63) to stimulate release of endothelial-derived relaxing factor. Whereas mibefradil can block L-type Ca\textsuperscript{2+} channels in partially depolarized cells (64), this effect is attenuated at the normal resting potential, so that in the heart under physiologic conditions this drug is probably specific for T-type Ca\textsuperscript{2+} channels, which are blocked at all levels of membrane potential (57).

Although much remains to be learned about the pathophysiological role of T-type Ca\textsuperscript{2+} channels, selective blockade of these channels holds promise for new advances in the treatment of cardiovascular disease. Furthermore, although details as to differences between the L-type and T-type Ca\textsuperscript{2+} channels are still emerging, it is already clear that the extensive clinical experience with the familiar L-type channel blocker mibefradil is of considerable potential clinical value because of several of these regulatory proteins, notably the plasma membrane Ca\textsuperscript{2+} specializations (4,6), due in large part to the buffering effect of the localized (4,6), due in large part to the buffering effect of the

Conclusions

This review of the Ca\textsuperscript{2+} channels that regulate Ca\textsuperscript{2+} flux into the cytosol illustrates how variations both between and among members of different families of channel proteins has contributed to the remarkable signaling diversity seen in the cardiovascular system. Duplication and divergence of genes that encoded common ancestral proteins has provided a dazzling array of specialized proteins able to mediate the many different signal transduction pathways that utilize messenger Ca\textsuperscript{2+} to modify cell function. Understanding the roles of these specializations is of considerable potential clinical value because several of these regulatory proteins, notably the plasma membrane Ca\textsuperscript{2+} channels, can be modified selectively by drugs. New knowledge of the structure and biology of the Ca\textsuperscript{2+} channel proteins could allow the design of drug molecules able to modify specific signal transduction cascades, thereby providing new agents for the treatment of cardiovascular disease.

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