International Union of Basic and Clinical Pharmacology. C. Nomenclature and Properties of Calcium-Activated and Sodium-Activated Potassium Channels

Leonard K. Kaczmarek, Richard W. Aldrich, K. George Chandy, Stephan Grissmer, Aguan D. Wei, and Heike Wulff

Abstract

A subset of potassium channels is regulated primarily by changes in the cytoplasmic concentration of ions, including calcium, sodium, chloride, and protons. The eight members of this subfamily were originally all designated as calcium-activated channels. More recent studies have clarified the gating mechanisms for these channels and have documented that not all members are sensitive to calcium. This article describes the molecular relationships between these channels and provides an introduction to their functional properties. It also introduces a new nomenclature that differentiates between calcium- and sodium-activated potassium channels.

I. Introduction

The first evidence that elevations of intracellular Ca\textsuperscript{2+} can increase the K\textsuperscript{+} permeability of the plasma membrane was obtained by demonstrating that chelation of Ca\textsuperscript{2+} suppresses the flux of K\textsuperscript{+} out of red blood cells (Gardos, 1958). Intracellular injection of Ca\textsuperscript{2+} into neurons of both invertebrates and vertebrates was subsequently found to activate a K\textsuperscript{+} conductance (Krnjević and Lisiewicz, 1972; Meech, 1972). The discovery of a Drosophila mutant termed slowpoke (slo) (Elkins et al., 1986), in which a Ca\textsuperscript{2+}-dependent component of K\textsuperscript{+} current is lacking in indirect flight muscles, eventually led to the identification of the first gene that encodes a Ca\textsuperscript{2+}-activated K\textsuperscript{+} channel (Atkinson et al., 1991; Adelman et al., 1992) and its mammalian homolog (Butler et al., 1993). The properties of this channel in heterologous expression systems

The authors serve as the Subcommittee on Calcium-Activated and Sodium-Activated Potassium Channels of the Nomenclature Committee of the International Union of Pharmacology (NC-IUPHAR).

Address correspondence to: Leonard K. Kaczmarek, Departments of Pharmacology and Cellular and Molecular Physiology, Yale School of Medicine, New Haven, CT 06520. E-mail: leonard.kaczmarek@yale.edu
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ABBREVIATIONS: AHP, afterhyperpolarization; IUPHAR, International Union of Basic and Clinical Pharmacology; RCK, regulator of conductance for K\textsuperscript{+} ions; Slack, sequence like a calcium-activated K channel; Slick, sequence like an intermediate conductance K channel.
corresponded very closely to a channel that had by then been very well characterized in many types of tissues as well as in lipid bilayers. This K⁺ channel is sensitive to both Ca²⁺ and transmembrane voltage and had been termed the BK or MaxiK channel because of its large unitary conductance (Contreras et al., 2013). This prototypical Ca²⁺-gated channel is termed KCa1.1 in the standardized nomenclature that is used to classify K⁺ channels (Wei et al., 2005). Like all the other channels discussed in this review, a functional K⁺ channel is comprised of a tetramer of these pore-forming proteins.

Genes that encode other Ca²⁺-dependent K⁺ channels were subsequently discovered by screening cDNA libraries for sequences resembling the ion-selective pore of known K⁺ channels, and then testing the expressed channels for sensitivity to changes in internal Ca²⁺ levels. After KCa1.1, the next two classes of channels to be discovered were found to be gated by Ca²⁺ but, unlike KCa1.1, to be insensitive to membrane voltage. These are the KCa2 family, which consists of three members (KCa2.1, KCa2.2, and KCa2.3) (Köhler et al., 1996) and the KCa3 family, which contains only a single member (KCa3.1) (Ishii et al., 1997b; Joiner et al., 1997). These two families have also been termed SK and IK channels, respectively (Contreras et al., 2013). One factor that has contributed to the popularity of this channel is its very large unitary conductance, 200 pS or more in symmetrical K⁺ concentrations (Marty, 1981). Because it is expressed in a wide range of tissues, it is the easiest channel to detect when patch clamping cells and it is readily reconstituted into lipid bilayers (Pallotta et al., 2013; Wrighton et al., 2015). One feature that distinguishes KCa1.1 (and also KCa5.1, see below) from all other K⁺ channels is that it has seven transmembrane domains (Fig. 2). In addition to the canonical six transmembrane domains S1-S6 that are common to all of the voltage-dependent KV family channels, there is an additional domain termed S0, preceding S1 (Meera et al., 1997). As a result, the N-termini of the KCa1.1

![Fig. 1. Phylogenetic relations among members of the Ca²⁺- and Na⁺-dependent K⁺ channel families.](image-url)
and K_{Ca}5.1 proteins are located on the outside rather than the inside of a cell.

The open probability of K_{Ca}1.1 channels is sensitive to membrane voltage, and this sensitivity is conferred by charged residues in the S2, S3, and S4 domains (Stefani et al., 1997; Horrigan and Aldrich, 1999, 2002; Ma et al., 2006). These move in response to changes in transmembrane voltage even in the absence of internal Ca^{2+}. Activation by elevations of Ca^{2+} occurs by shifting the voltage dependence of the channel to progressively more negative potentials. Binding sites for Ca^{2+} ions are located in two regions of the extended cytoplasmic C-terminal termed the RCK1 and RCK2 domains (for regulator of conductance for K^{+} ions) (Jiang et al., 2002; Wu et al., 2010; Yuan et al., 2010). A region termed the Ca^{2+} bowl located in RCK2 contains multiple negatively charged residues (Schreiber and Salkoff, 1997; Bao et al., 2004). The eight RCK domains in each tetrameric K_{Ca}1.1 channel together form a “gating ring” that opens on binding of Ca^{2+}. This conformational change is coupled to the transmembrane regions through a cytoplasmic linker region next to the activation gate in the S6 segment. This allosteric mechanism leads to an increase in open probability following Ca^{2+} binding (Horrigan and Aldrich, 1999; Jiang et al., 2002; Wu et al., 2010; Miranda et al., 2013).

The Ca^{2+}-sensitivity of K_{Ca}1.1 channels, as well as their kinetic behavior, sensitivity to pharmacological agents, and response to activation of protein kinases, varies considerably in different tissues (Contreras et al., 2013). Although there is only one gene that encodes these channels, such diversity is achieved by alternative splicing of mRNAs (Navaratnam et al., 1997; Rosenblatt et al., 1997) and by the fact that the properties of the channel are substantially modified by auxiliary \( \beta \)-subunits. Alternative splicing not only determines the response to Ca^{2+} but also the targeting of the channel to the plasma membrane or to intracellular organelles such as mitochondria (Lagrutta et al., 1994; Xie and McCobb, 1998; Chen et al., 2005; Fodor and Aldrich, 2009; Singh et al., 2012). Binding of the K_{Ca}1.1 \( \alpha \) subunit to one of four different auxiliary subunits (\( \beta1, \beta2, \beta3, \) and \( \beta4 \)) alters voltage dependence, activation rate, sensitivity to a wide range of drugs, and determines whether the channel inactivates during sustained depolarization (Brenner et al., 2000; Uebele et al., 2000; Xia et al., 2000; Contreras et al., 2012). The properties of the channels can be further modified by association with \( \gamma \)-subunits, also known as leucine-rich repeat proteins (Yan and Aldrich, 2012), and by binding to heme, which may allow the channel to respond to changes in redox state of the cell (Tang et al., 2003).

Although K_{Ca}1.1 channels are expressed in many different tissues, their activity is particularly prominent in smooth muscle cells, including those of the vascular endothelium (Contreras et al., 2013). Channel activation, in response to synthesis of nitric oxide (NO) and activation of cGMP-dependent protein kinase, produces muscle relaxation, altering blood vessel diameter and blood pressure. Other tissues in which K_{Ca}1.1 channels have been studied include sensory hair cells of the cochlea of lower vertebrates, where they establish the tonotopic gradient that determines the frequency of sound to which each cell responds (Roberts et al., 1990; Bai et al., 2011). In neurons, as well as in pancreatic \( \beta \)-cells, K_{Ca}1.1 channels regulate action potential firing and secretion (Rajan et al., 1990; Faber and Sah, 2003).

As described above, the pharmacological properties of K_{Ca}1.1 channels depend on their association with auxiliary subunits. The scorpion toxins charybdotoxin, iberiotoxin, and slotoxin are effective pore blockers for these channels (Kaczorowski et al., 1996; Garcia-Valdes et al., 2001). Paxilline is also an effective blocker, widely used in studies in the nervous system (Sanchez and McManus, 1996). A variety of compounds that activate K_{Ca}1.1 channels, such as NS1619, are also commonly used experimental agents (Li et al., 2003). In addition, K_{Ca}1.1 channels are very sensitive to relatively low concentrations of ethanol, which potentiate their...

**Fig. 2.** Schematic representations of the transmembrane topology of K_{Ca} and K_{Na} channels.

(Kaczmarek et al.)
activity (Treistman and Martin, 2009). Both the potentiating effects of ethanol and development of subsequent tolerance to its actions depend on both direct interactions with the KCa1.1 α-subunit, the presence of the β4 subunit, and the lipid environment (Mulolland et al., 2009).

Mutations in either the α- or β-subunits of KCa1.1 channels, as well as changes in expression levels of these proteins, are linked to a variety of clinical conditions including hypertension, diabetes, asthma, and epilepsy (Fernández-Fernández et al., 2004; Du et al., 2005; Seibold et al., 2008; Wang et al., 2012). For example, a mutation of the α-subunit D434G in the cytoplasmic linker that couples the gating ring to the activation gate increases channel open probability, resulting in epilepsy (Yang et al., 2010).

III. The KCa2 Family—Small Conductance Channels Regulated by Calmodulin

This family consists of three members, KCa2.1, KCa2.2, and KCa2.3 (commonly also termed SK1, SK2, and SK3), each of which has a unitary conductance of ~10 pS when measured in symmetrical K+ solutions (Köhler et al., 1996). Their properties and physiologic functions have been reviewed (Stocker, 2004; Adelman et al., 2012). In their transmembrane organization, KCa2 channels resemble the voltage-dependent Kv family of channels with six alpha-helical transmembrane segments, S1-S6, and a consensus K+-selective pore sequence between S5 and S6 (Fig. 2). Unlike the voltage-dependent channels, however, there is only a small number of positively charged residues in the fourth transmembrane segment S4. Consistent with the role of the S4 segment as a voltage sensor in Kv channels, the KCa2 family channels have linear current-voltage relations and are insensitive to changes in transmembrane voltage. With a D for activation by Ca2+ of only several hundred nanomolar, they are substantially more sensitive to small changes in cytoplasmic Ca2+ levels than are the KCa1.1 channels (Köhler et al., 1996). This sensitivity can be attributed to the fact that the α-subunits of these channels form a heteromeric complex with calmodulin and that it is the binding of Ca2+ to this constitutively associated calmodulin rather than to the α-subunit of the channel itself that confers Ca2+ sensitivity (Xia et al., 1998).

In addition to calmodulin, KCa2.2 and KCa2.3 channels in their native state enter into a multiprotein complex containing casein kinase 2 and protein phosphatase 2A (Bildl et al., 2004; Allen et al., 2007). Within this complex, casein kinase phosphorylates a threonine residue in calmodulin, reducing sensitivity of the channel to Ca2+ and increasing the rate at which channels close after transient elevation of cytoplasmic Ca2+ (Bildl et al., 2004). When activated within the complex, protein phosphatase 2A reverses this effect (Allen et al., 2007).

The physiologic role of KCa2 family channels has been investigated most extensively in the nervous system, where they are expressed at high levels in cerebellar Purkinje cells and in pyramidal cells of the hippocampus and cerebral cortex (Stocker and Pedarzani, 2000; Sailer et al., 2002). KCa2.3 channels are also present in nonneuronal tissues, including the vascular endothelium, urinary bladder smooth muscle, and cardiac tissue (Taylor et al., 2003; Chen et al., 2004; Rosenbaum et al., 2012). In many types of neurons, a prominent action of KCa2 channels is to generate a Ca2+-dependent after-hyperpolarization (AHP) of the membrane that follows a burst of action potentials. KCa2 channels thus regulate neuronal firing frequency and spike frequency adaptation (Adelman et al., 2012). There exist multiple components of AHPs, each with a different time course after the burst. Genetic deletion of the KCa2.2 subunit abolishes the medium AHP, which decays over several hundreds of millisecond after a train of action potentials, but does not affect other components of the AHP (Bond et al., 2004).

KCa2 channels may be closely coupled selectively with L-type calcium channels to allow for activation by Ca2+ nanodomains formed by Ca2+ influx through the calcium channels (Marrion and Tavalin, 1998). Neurotransmitter receptors that also elevate cytoplasmic Ca2+ levels, either by Ca2+ influx through the receptor as in the case of NMDA receptors (Oliver et al., 2000; Faber et al., 2005; Ngo-Anh et al., 2005), or by triggering release of Ca2+ from intracellular stores (Power and Sah, 2008; Klement et al., 2010), can also produce neuronal hyperpolarization by activating KCa2 channels. The trafficking of KCa2.2 channel subunits into and out of the spines on the dendrites of hippocampal neurons determines the onset and amplitude of long-term potentiation, a prolonged increase in excitatory synaptic transmission that follows repetitive synaptic stimulation of hippocampal pyramidal cells (Lin et al., 2008; Allen et al., 2011). KCa2 channels thus seem to play an important role in learning and memory (Hammond et al., 2006).

The key pharmacological agent that has been used to characterize the properties of this family of small conductance Ca2+-activated K+ channels, both in native neurons and expression systems, is the neurotoxin apamin, a component of bee venom that appears to be fully selective for this class of channel (Adelman et al., 2012). Although effectively a pore blocker, the selectivity for KCa2 channels is determined by residues outside of the pore region itself (Ishii et al., 1997a; Nolting et al., 2007; Lamy et al., 2010). A variety of compounds that positively modulate these channels by apparently increasing their Ca2+ sensitivity have also been found (Christophersen and Wulff, 2015). The first of these to be characterized was 1-ethyl-2-benzimidazolinone (Devor et al., 1996), a compound that binds at the calmodulin-channel interface (Zhang et al., 2012a). Another activator, NS309, has been shown by X-ray
crystallography to bind to a short region of the channel that links the cytoplasmic domain of KCa2.2 to the S6 segment (Zhang et al., 2013a). This is believed to stabilize the open state of the channel and facilitate gating in the presence of the channel activator.

Based on the physiologic role of KCa2 channels in regulating neuronal excitability, KCa2 activators, which can reduce neuronal firing, are being investigated for the treatment of conditions characterized by hyperexcitability, such as epilepsy, ataxia, and alcohol dependence (Lam et al., 2013). KCa2 channel inhibitors in contrast have been suggested to improve learning and memory (Lam et al., 2013) and are being investigated preclinically for the treatment of atrial fibrillation based on the role of KCa2 channels in modulating action potential duration in the heart (Grunnet et al., 2012).

IV. KCa3.1—A Multifunctional Intermediate-conductance Channel Regulated by Calmodulin

KCa3.1 channels are closely related to the KCa2 family and were originally termed SK4 channels (Ishii et al., 1997b; Joiner et al., 1997). Their unitary conductance is, however, greater than that of KCa2 channels, giving rise to the name IK (for intermediate conductance for K+), which has also been used widely to denote these channels. Like the KCa2 family channels, the sensitivity of KCa3.1 to Ca2+ is determined by its association with calmodulin (Fanger et al., 1999; Joiner et al., 2001) (Fig. 2).

The properties of KCa3.1 closely match those of the first described Ca2+-activated K+ conductance, the “Gardos channel” in red blood cells, where it is expressed at high levels and plays a role in volume regulation (Vandorpe et al., 1998; Hoffman et al., 2003). It is also the major Ca2+-activated K+ channel in human T lymphocytes (Logsdon et al., 1997) and in other immune cells such as B lymphocytes, mast cells, macrophages, and microglia (Cahalan and Chandy, 2009; Feske et al., 2015). KCa3.1 further plays a significant role in many other nonexcitable tissues such as the vascular endothelium, fibroblasts, and secretory epithelia (Wulff and Köhler, 2013). In all these tissues, KCa3.1 regulates Ca2+ signaling and membrane potential. Although two recent studies indicated that KCa3.1 may be expressed in neurons and that it may contribute to the slow afterhyperpolarization that follows a burst of action potentials in neurons of the hippocampus (King et al., 2015; Turner et al., 2015), another study presented data that KCa3.1 does not contribute to this slow AHP (Wang et al., 2016). The potential role of the channel in neurons therefore currently remains unclear.

Many of the pharmacological agents that activate the KCa2 channels are also positive modulators of KCa3.1 (Wulff and Christophersen, 2015) and are being investigated as novel endothelial-targeted antihypertensives because of the role of KCa3.1 and KCa2.3 in the endothelium-derived hyperpolarization vasodilator response (Brähler et al., 2009). The pharmacology of agents that suppress KCa3.1 current, however, differs from that of the KCa2 family. For example, KCa3.1 is insensitive to apamin, but can be blocked by charybdotoxin and the small molecule TRAM-34 (Wulff et al., 2000). Genetic deletion and/or pharmacological treatment with TRAM-34 have been used to demonstrate that KCa3.1 potentially constitutes a novel target for immunosuppression in autoimmune diseases and stroke as well as for fibroproliferative disorders, asthma, and atherosclerosis (Wulff and Castle, 2010). However, the only clinical application of KCa3.1 blockers has so far been for the treatment of sickle cell anemia, where senicapoc failed in Phase-3 clinical trials despite engaging its target as demonstrated by the observed improvements in hematocrit and erythrocyte hydration in the treated patients (Ataga et al., 2011).

V. The KNa1 Family—Channels Regulated by Intracellular Na+ and Cl−

A. KNa1.1

The channel for which we now propose the name KNa1.1 in the official IUPHAR nomenclature was found to be inhibited rather than activated by cytoplasmic Ca2+ in the original study that expressed this channel in oocytes and mammalian cells (Joiner et al., 1998; Budelli et al., 2016). It was named Slack for “Sequence Like A Calcium-Activated K channel” because its unitary conductance (~65 pS in physiologic solutions, ~180 pS in symmetric K+ solutions) is intermediate between that of KCa1.1 channels and that of most other K+ channels. It has also been termed Slo2.2 and, in the earlier IUPHAR nomenclature, KCa4.1, based on the premise that it was likely to resemble KCa1.1 in its properties. It is, however, only 7% identical to KCa1.1, and subsequent work established that it is gated primarily by changes in Na+ and Cl− concentrations (Bhattacharjee et al., 2003; Yuan et al., 2003; Yan et al., 2012). Its properties and physiologic functions have been reviewed recently (Kaczmarek, 2013).

KNa1.1 also differs from KCa1.1 in that it lacks an S0 transmembrane domain and therefore resembles Kv family and KCa2 family channels in its transmembrane topology (Fig. 2). Although KNa1.1 currents are voltage dependent, the protein lacks the motif of repeated basic amino acids in the S4 segment that is characteristic of voltage-dependent channels and KCa1.1. The complete structure of KNa1.1 in its closed state has been determined by cryoelectron microscopy (Hite et al., 2015) and that of the cytoplasmic C-terminal domain, under conditions that are likely to correspond to the open state, by X-ray crystallography (Yuan et al., 2010). As in KCa1.1, there are two RCK domains in the extended C terminus of each KNa1.1 subunit in a tetrameric
channel, and these form a cytoplasmic gating ring that, when it is constricted, closes the ion conduction pore. A potential binding site for Na⁺ ions, which resembles a site conferring Na⁺ dependence on Kᵦ₃ family channels (Sui et al., 1996), has been located in RCK2 and mutations at this site substantially lower Na⁺ sensitivity (Zhang et al., 2010). Confirmation of the site(s) at which Na⁺ binds will, however, require determination of the complete structure in the presence of Na⁺.

Several isoforms of KₐNa.1.1 that differ in their kinetics of activation are produced by alternative splicing of RNA encoding this channel (Brown et al., 2008). One of these, which has been termed Slack-B (KₐNa.1.1B) and has the longest cytoplasmic N-terminal domain of the known isoforms, can form heteromers with KₐNa.1.2 (Slick), whose properties are elaborated in the next section (Chen et al., 2009). The original studies of KₐNa.1.1 expressed in Xenopus oocytes found that coexpression of Slack-B with KₐCa.1.1 gave rise to channels that had properties distinct from either channel alone, suggesting that the two subunits may interact (Joiner et al., 1998). Studies of its regional distribution, however, indicate that this is unlikely to be a major role for KₐNa.1.1 subunits. Unlike the ubiquitous KₐCa.1.1, KₐNa.1.1 is primarily expressed in central and peripheral neurons, with little expression in other tissues except for the testes and kidney (Joiner et al., 1998; Bhattarcharjee et al., 2002; Paulais et al., 2006; Brown et al., 2008; Nuwer et al., 2010; Rizzi et al., 2016).

Phosphorylation by protein kinase C of a serine residue in the cytoplasmic linker region between the RCK domains and the S6 transmembrane domain of KₐNa.1.1 increases current amplitude (Santi et al., 2006; Barcia et al., 2012). The activity of KₐNa.1.1 channels can be also regulated by their interaction with other cellular proteins. These include the transmembrane protein TMEM16C (Huang et al., 2013) and the Fragile X Mental Retardation protein (Brown et al., 2010; Zhang et al., 2012b), both of which enhance KNa1.1 channel currents. In single channel recordings, the probability of opening of a Slack channel is increased if it is present in a cluster with neighboring Slack channels, and such cooperativity is greatly enhanced in the disease-causing mutations (Kim et al., 2014), suggesting that interchannel interactions as well as intrinsic changes in gating account for the observed gain-in-function in the mutants.

No pharmacological agents that act on KₐNa.1.1 selectively have yet been discovered. Quinidine is a very effective blocker of these channels in both expression systems and neurons (Bhattarcharjee et al., 2003; Yang et al., 2006, 2007; Milligan et al., 2014; Rizzo et al., 2016). It has been reported that quinidine ameliorated the symptoms of a patient with malignant migrating partial seizures in infancy, but because this agent blocks a wide variety of other channels, the mechanism is not known (Bearden et al., 2014). Other nonspecific blockers that suppress KₐNa.1.1 channels include bepredil, cloflium, and barium ions (Bhattarcharjee et al., 2003; Yang et al., 2006; de Los Angeles Tejada et al., 2012b). These channels can also be activated by several pharmacological agents including bithionol, loxapine,
and niclosamide (Yang et al., 2006; Biton et al., 2012). Although bithionol also activates KCa1.1 (Li et al., 2003), loxapine appears to be specific for KNa channels over KCa1.1 (Biton et al., 2012).

B. KNa1.2—A Widely Distributed Channel Sensitive to Na⁺ and Cl⁻

The channel for which we now propose the name KNa1.2 in the IUPHAR nomenclature is very closely related to KNa1.1, being ~74% identical in protein sequence, with the greatest divergence from KNa1.1 at the distal C terminus (Bhattacharjee et al., 2003). Because of its similarity in sequence to KNa1.1 (Slack) it was named Slick for "Sequence Like an Intermediate Conductance K channel." It is also referred to as SLO-2 and, in the earlier IUPHAR nomenclature, KCa4.2. As with KNa1.1, however, it has been found to be gated predominantly by Na⁺ and Cl⁻ (Bhattacharjee et al., 2003; Kaczmarek, 2013).

Although there have been no structural studies of KNa1.2 as yet, insights have been provided by mutagenesis (Garg et al., 2013), and its mechanisms of gating are likely to resemble those of KNa1.1. The unitary conductance of KNa1.2 in symmetrical K⁺ solutions is ~140 pS, and it is slightly less sensitive to changes in Na⁺ and more sensitive to alterations in Cl⁻ than is KNa1.1 (Bhattacharjee et al., 2003). As in KNa1.1, there is a Na⁺ coordination motif in RCK2 that determines sensitivity to Na⁺ (Thomson et al., 2015), and channel activity is regulated by binding to phosphatidylinositol 4,5-bisphosphate (de los Angeles Tejada et al., 2012a). Unlike KNa1.1, however, there exists a consensus ATP binding site after the second RCK domain of KNa1.2. The function of this site in cells is, however, not yet clear. Experiments using excised patches demonstrate that application of 5 mM ATP or a nonhydrolyzable ATP analog reduced currents by ~80% and that mutation of this consensus site abolished the effect of ATP (Bhattacharjee et al., 2003). Moreover, ATP and a nonhydrolyzable analog were both found to suppress Na⁺- and Cl⁻-activated K⁺ channels in excised patches from auditory brain stem neurons that express KNa1.2 (Yang et al., 2007). In contrast, experiments with a KNa1.2-GFP fusion construct expressed in HEK cells failed to find an effect of adding ATP to patch pipettes on current amplitude (Berg et al., 2007). ATP was also found not to affect the increase in KNa1.2 currents produced by niflumic acid, a pharmacological activator of these channels (Garg and Sanguinetti, 2014). Thus the conditions under which ATP regulates these channels have yet to be completely established.

Comunoprecipitation and single channel studies have established that KNa1.2 coassembles with B isoform of KNa1.1 (KNa1.1B, Slack-B) to form heteromeric channels with properties distinct from those of either subunit alone (Chen et al., 2009). The formation of such heteromers requires the specific N-terminal domain of Slack-B and apparently does not occur with the shorter N-terminal splice variant KNa1.1A (Slack-A). In contrast to KNa1.1, KNa1.2 currents, as well as those of heteromeric KNa1.2/KNa1.2 channels are suppressed by activation of protein kinase C (Santi et al., 2006; Chen et al., 2009).

KNa1.2 channels are widely distributed throughout the nervous system and are also expressed in a variety of nonneuronal cells including cardiac cells (Bhattacharjee et al., 2003,2005; Yuan et al., 2003; Rizzi et al., 2015). A recent report suggests an important role for KNa1.2 (KCNT2) in controlling the migratory capacity of medulloblastoma cell lines, acting synergistically with Kv10.2 (EAG2) potassium channels (Huang et al., 2015). In the nervous system, coexpression with KNa1.1 can be detected in some but not all types of neurons (Santi et al., 2006; Berg et al., 2007). Based on pharmacological experiments, a proposal has been made that a KNa channel, most likely KNa1.2, regulates K⁺ flux across the inner membrane of mouse heart mitochondria and that channel activation is cardioprotective (Wojtovich et al., 2011).

One major feature that distinguishes KNa1.2 from the closely-related KNa1.1 channel is its sensitivity to small changes in cell volume. When expressed in oocytes, KNa1.2 currents are strongly stimulated by cell swelling and inhibited by a decrease in cell volume (Tejada et al., 2014). There are as yet no reported human mutations in KCNT2, the gene encoding KNa1.2 channels, that could shed light on potential physiologic roles of KNa1.2. Nevertheless, several observations suggest that it serves a protective function during ischemia or tissue injury, the function proposed by the very first publication describing Na⁺-activated K⁺ currents (Kameyama et al., 1984). In neurons, the promoter for the gene is regulated by nuclear factor-κB, which is activated under conditions of hypoxia or injury (Tomasello et al., 2015). In the nematode Caenorhabditis elegans, deletion of the gene for SLO-2, the ortholog to mammalian KNa channels, alters the sensitivity of the animals to hypoxia, although reports differ in the direction of the change in sensitivity (Yuan et al., 2003; Zhang et al., 2013b). Moreover, extrapolation of these findings to mammals is problematic, because, unlike the mammalian KNa channels, nematode SLO-2 is activated by Ca²⁺ rather than Na⁺, an effect that can be attributed to specific glutamic acid residues in RCK1 of the nematode channel (Yuan et al., 2000; Zhang et al., 2013b). The RCK2 domain of SLO-2 more closely resembles those of KNa1.1 and KNa1.2 channels in that the domain corresponding to the Ca²⁺ bowl of KCa1.1 contains repeated positively charged residues rather than the negative charges in KCa1.1.

In common with KNa1.1, there are no known pharmacological agents that selectively target KNa1.2 channels. They are inhibited by quinidine, clofilium, and isoflurane (Bhattacharjee et al., 2003; Berg et al., 2007;
de Los Angeles Tejada et al., 2012b). They are strongly activated by niflumic acid and other fenamates, although with low potency (Dai et al., 2010; Garg and Sanguinetti, 2012). These agents uncouple the channels from modulation by either Na+ or transmembrane voltage and greatly increase current even in the absence of internal Na+ ions. The action of fenamates, which is nonspecific in that they also affect many other channels including KCa1.1 (Gribkoff et al., 1996), is biphasic, suggesting they bind to two distinct sites within KNa1.2 (Garg and Sanguinetti, 2012).

VI. KCa5.1—A Channel Specific to Sperm Cells

The KCa5.1 channel, which is encoded by the KCNUN1 gene, is more commonly referred to as SLO3. It was discovered through its homology to KCa1.1 (Schreiber et al., 1998), and, of all the channels reviewed here, KCa5.1 is most closely related to KCa1.1 in sequence and structure, as determined by X-ray crystallography (Leonetti et al., 2012). Like KCa1.1, KCa5.1 has seven transmembrane segments with an S0 segment before S1 and an extracellular N terminus (Fig. 2). When expressed in oocytes, the mouse channel is voltage dependent and sensitive to pH, being activated by alkalization and suppressed by acidification (Schreiber et al., 1998; Zhang et al., 2006a,b).

KCa5.1 is expressed selectively in sperm cells, and its properties are regulated by LRRC52, a testis-specific accessory subunit (Schreiber et al., 1998; Leonetti et al., 2012; Zeng et al., 2015). Evidence strongly suggests that it corresponds to the large-conductance potassium channel that can be recorded from mature sperm. This native channel is activated by both voltage and internal alkalization and has been termed KSper (Navarro et al., 2007). As sperm encounter the alkaline environment near the ovum in the female reproductive tract, the activation of KSper is believed to produce the membrane hyperpolarization that is observed during the process of sperm capacitation. Normal capacitation is required for sperm to become fully competent for fertilization.

The identification of KCa5.1 with the sperm KSper channel has been well established in mice (Santi et al., 2010; Zeng et al., 2011, 2013). For human sperm, however, one report suggested that the native KSper channels is actually KCa1.1 (Mannowetz et al., 2013). This conclusion was based on recordings of native currents from human sperm, which showed that the human channels have a Ca2+ dependence and pharmacological properties similar to those of KCa1.1. Nevertheless, a more recent study indicated that, as in mice, the human KSper-like current is in fact encoded by human KCa5.1 (Brenker et al., 2014). The explanation for the apparent contradiction offered by this study is that human KCa5.1 channels are functionally different from the mouse channels. In particular, despite their strong sequence similarity, human KCa5.1 channels are significantly more sensitive to activation by internal Ca2+ and less pH sensitive than the mouse channels, which makes them functionally similar to typical KCa1.1 channels.

No pharmacological agents that selectively act on KCa5.1 channels have been found, but they can be blocked by quinidine (Tang et al., 2010; Sánchez-Carranza et al., 2015) as well as by Ba2+ ions and quinine (Wrighton et al., 2015). As the specific factors that regulate the gating of KCa5.1 in different species become established, it may become appropriate to rename this channel.

VII. Summary

Research in the past 15 years has clarified the structural and functional relationships among K+ channels that are gated by changes in intracellular concentrations of Ca2+, Na+, and protons, as well as their biologic roles in different cell types. These are now considered to fall into three distinct groups. The first group contains the ubiquitous large-conductance Ca2+-activated KCa1.1 (BK) channel as well as the sperm-specific KCa5.1 channel, which have seven transmembrane segments in each subunit and an extracellular N terminus. Gating of these channels is controlled by the ion binding directly to two RCK domains located in their extended cytoplasmic C termini. The second group is comprised of two channels activated by internal Na+ and Cl− ions, for which we propose the names KNa1.1 and KNa1.2. These resemble the first group in that they have cytoplasmic C-terminal RCK domains that control gating and have large unitary conductances, but they have only six transmembrane segments in each subunit and their N termini are located intracellularly. The third group of channels is comprised of the small conductance channels, KCa2.1, KCa2.2, and KCa2.3, and the intermediate conductance channel KCa3.1. In contrast to the other channels, these are voltage independent and activation by Ca2+ occurs through the binding of Ca2+ to calmodulin, an integral component of the channel complex, rather than through direct binding of Ca2+ to the α-subunits.

References


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