Modulation of BK$_{\text{Ca}}$ Channel Gating by Endogenous Signaling Molecules
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Large-conductance Ca\(^{2+}\)- and voltage-activated K\(^+\) (BK\(_\text{Ca}\), MaxK, or Slo1) channels are well known for their roles in regulation of membrane excitability, partly owing to their large conductance, 250–300 pS in symmetrical 150 mM K\(^+\), and to the synergistic activation mechanism encompassing membrane depolarization and intracellular Ca\(^{2+}\)/Mg\(^{2+}\) (98). The interdependent activation mechanism enables BK\(_\text{Ca}\) channels to typically exert a negative feedback influence on cellular excitability (98, 118). The importance of BK\(_\text{Ca}\) channels in regulation of vascular tone, determination of action potential duration and frequency, and neurotransmitter release has been well documented (98). Consistent with the functional importance, notable phenotypes, such as hypertension, erectile dysfunction, and urinary incontinence, are associated with inhibition or downregulation of BK\(_\text{Ca}\) channel activity (98, 118, 157) forms a basis of the following model of BK\(_\text{Ca}\) channel activation. The VSDs transduce changes in membrane potential, and their movements are electromechanically coupled to the permeation gate within the pore domain. Extensive mutagenesis studies suggest that the activation by Ca\(^{2+}\) probably involves two distinct high-affinity Ca\(^{2+}\) sensors per subunit: the RCK1 sensor (130) and the Ca\(^{2+}\) bowl sensor (102). Activation of the Ca\(^{2+}\) sensors induces conformational changes in the gating ring and leads to opening of the permeation gate near/within S6 via the linkers connecting the gating ring and S6 (78, 88, 130). The interactions among the gate, VSDs, and divergent cation sensors are allosteric and reciprocal (78). Ca\(^{2+}\)-binding facilitates opening of the gate, and, conversely, opening of the gate facilitates binding of Ca\(^{2+}\) to its sensors by increasing the binding affinity. However, the coupling between the gate and the voltage/intracellular ligand sensors is such that, even without VSD activation and Ca\(^{2+}\), the gate may open once every few minutes on the average (47). Furthermore, open probability (P\(_o\)) of the channel approaches unity at sufficiently depolarized voltages without Ca\(^{2+}\), and the channel opens frequently at negative voltages without VSD activation with high concentrations of Ca\(^{2+}\). Only one gene (KCNMA1) (23) codes for the pore-forming Slo1 subunit, but the functional properties of the BK\(_\text{Ca}\) channel are exceptionally diverse, encompassing many mechanisms from transcriptional regulation (67), microRNA-mediated regulation (111), and alternative splicing (1) to acute modulation of gating by signaling molecules. Alternative splicing of the RNA produces numerous transcripts, translating to diverse Slo3 proteins with distinct functional properties in different tissues and in different hormonal states (105, 115, 131). To further increase the diversity, native BK\(_\text{Ca}\) channel complex (KCNMB) in a single channel assembly, each of the 14 transmembrane segments containing both NH\(_2\)- and COOH-terminal tails, each of which contains multiple phosphorylation sites (89). Functional characterization also shows that the molecular signaling and local and spatial regulation, such as neuronal excitability, vascular tone regulation, and neurotransmitter release.

The functional versatility of BK\(_\text{Ca}\) channels is expressed in almost every tissue in our body and participate in many critical functions. In this review, we focus on modulation of BK\(_\text{Ca}\) channels by small endogenous molecules, emphasizing their molecular mechanisms. The mechanistic information available from studies on the small naturally occurring modulators is expected to contribute to our understanding of the physiological and pathophysiological roles of BK\(_\text{Ca}\) channels.
channel complexes often contain auxiliary β subunits (KCNMB) in a tissue-specific manner (66). Four β subunits, β-1, β-2, β-3, and β-4 (10, 21, 65, 66, 89, 119), have been identified, each of which contains two transmembrane segments connected by a large extracellular loop, leaving both NH2 and COOH termini in the intracellular side (89). Functionally, the presence of β subunits markedly alters the channel’s gating and pharmacological characteristics (10, 84, 89, 117). Recent evidence also shows that native BKCa channels are part of macromolecular signaling complexes that include enzymes (4, 76, 124) and/or ion channels (13, 37) to mediate local and spatially directed signaling (FIGURE 1). Once assembled, the BKCa channel activity is subject to modulation by a wide spectrum of biologically relevant factors such as serine/threonine/tyrosine phosphorylation, cysteine/methionine oxidation, steroid hormones, and gases [oxygen, nitric oxide (NO), and carbon monoxide (CO)] (52, 69, 120, 123, 124). Of the modulatory factors, phosphorylation of BKCa channels has been studied extensively and summarized (183). This review primarily focuses on examples of regulation of BKCa channels by other molecules: H+, heme, CO, reactive oxygen and nitrogen species, and lipids.

**Protons, H+**

H+ is a vitally important ion, and its intracellular concentration is under a tight control. However, some fluctuations in pHi may occur as a consequence of normal cell function (26). Depolarization by action of Kv1.2/2.1 channels (56, 135) changes in extracellular Ca2+ probably induces conformational changes in the S6 segments, which are connected to the NH2 terminus of the neighboring α-subunit (78, 88, 130). Ca2+ also induces conformational changes in the α-subunit, probably via an effect on pHi (78, 88, 130). Ca2+ induces conformational changes in the α-subunit, probably via an effect on pHi.

**Additional Text**

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It is not surprising that treatment of cells/membrane patches containing BKCa channels with reactive species such as H2O2 has been reported to produce a myriad of effects.

By contrast, BKCa channels are robustly activated by intracellular H+, a characteristic shared only by a few others among the K+ channel family (80). Early studies, often using native BKCa channels, reported that intracellular H+ decreased ionic currents through BKCa channels (27). Recent studies utilizing heterologously expressed Slo1 channels as well as native BKCa channels now show that H+ stimulates opening of the channel (8, 44, 51, 92). It is uncertain what accounts for the seemingly contradictory results.

Intracellular H+ increases ionic currents through BKCa channels in the absence of Ca2+ without altering the single-channel current size (i) (8, 44, 51, 92). The increase in open probability (P o) caused by H+ is accompanied by a 40- to 50-nm shift in the macroscopic conductance-voltage curve (GV) to the negative direction with an EC50 value of pH i 6.5 and a Hill coefficient of >2 such that the nearly full effect is observed between pH i = 6.0 and 7.2 (8), a physiologically feasible range. A mutagenesis study identified His365 and His394 (using NP 002238 numbering) in the RCK1 sensor domain, which is important in the Ca2+-dependent activation (130), as the necessary residues, with His365 and His394 accounting for 2/3 and 1/3 of the shift in GV, respectively. Mutation of His365/His394 to neutral Ala approximated the voltage dependence at pH 7.2, whereas that to positively charged Arg produced a voltage dependence similar to that at pH 6.2. Combined with the finding that the stimulatory effect of H+ diminishes with increasing ionic strength, it has been suggested that the prototatic imidazole side chains of His365 and His394 electrostatically interact with nearby electronegative elements. One of the electrostatic interaction partners appears to be Asp367, which is a critical component in high-affinity Ca2+ sensing (130). In summary, it is envisioned that intracellular H+ protonates the side chains of His365 and His394 located within the high-affinity Ca2+-sensor site in the RCK1 domain and then the positively charged side chains interact with Asp367. This interaction in turn mimics the action of Ca2+, expanding the gating ring and promoting opening of the gate.

The RCK1 sensor in Slo1 responds to both Ca2+ and H+. This multi-ligand nature of the RCK1 sensor with regard to H+ and Ca2+ is probably physiologically significant since the intracellular concentrations of these two ions are also reciprocally regulated (7, 134). The H+ sensitivity of the BKCa channel may also play an important role in pathophysiological conditions, such as in cerebral ischemia during which significant increases in both intracellular H+ and Ca2+ concentrations are observed (71).

Heme

Like H+, heme is a fundamentally important molecule, typically as a stable protein prosthetic group. Emerging evidence suggests that free intracellular heme may function as a non-genomic signaling molecule, acutely modulating BKCa channels (50, 114). Bioinformatic analysis of the Slo1 primary sequence suggested that the sequence CKACH (114, 125) in the linker region between the cytoplasmic RCK1 and RCK2 segments might be capable of coordinating heme. Indeed, heme applied to the cytoplasmic side decreased P o in cell-free membrane patches with a high affinity (IC50 = 10 nM) without altering I (114). The modulatory effect of heme is independent of the redox status of the iron center, but substitution of the iron with other metals generally interferes with its modulatory ability. Mutations in the sequence CKACH disrupted the sensitivity of the channel to heme (55, 114, 123), suggesting that the sequence may be part of the heme binding site.

This idea was further corroborated by UV-vis/electrospray ionization mass spectroscopy assays (55) performed on a model peptide whose sequence corresponds to the sequence CKACH (114, 125). The modulatory effect of heme is not dependent on the CO redox state of the iron center, which is expected to be fixed in the CO-protected CO-modulated BKCa channel proteoliposomes (120, 123). These results demonstrated that CO modulates the BKCa channel through those elements of the channel proteoliposome that contain a histidine/histidine/histidine sensor (80).

Although the molecular mechanism of the heme action of the BKCa channel is relatively clear, its physiological significance is less certain. If heme is in fact a signaling molecule, a mechanism to activate the signaling cascade and a mechanism to terminate the signal should be envisioned that couples the heme signal to other signaling pathways. The influence of heme is relatively low, less than Ca2+, and produces a native signal that is limited to a certain cellular distribution.

Carbon Monoxide

One of the most ancient signaling molecules (CO) is released by phosphodiesterases (PDEs) as a stable metabolite of CO-derived guanosine monophosphate (PKG) (96). The consensus Ser-Thr-Tyr motif involved in phosphorylation often localized in the CO-modulated BKCa channel proteoliposomes (120, 123), but CO may exert distinct effects through those elements of the channel that contain a histidine/histidine/histidine sensor (80).

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membrane using heme transporters (95, 104). The heme signal can be terminated by the action of heme oxygenase (HMOX) (79), thus removing any direct influence of heme on BKCa channels. HMOX, however, produces a number of heme degradation products, including CO, a putative gaseous messenger, which may exert distinct effects on the channels.

**Carbon Monoxide**

One of the well known effects of carbon monoxide (CO) is to relax blood vessels (70). CO, like another gaseous messenger NO, binds to the heme iron center in soluble guanylyl cyclase (sGC) and increases its activity, leading to an increased level of cGMP and of phosphorylation by cGMP-dependent protein kinase (PKG) (96). Experimental phosphorylation of the PKG-consensus Ser residues in Slo1 located near the Ca2+-bowl sensor (87) and at the distal COOH terminus (38) increases P in a few fold. The underlying mechanism of the PKG-mediated regulation is not yet known and neither is whether the aforementioned residues are dynamically phosphorylated in vivo in response to CO.

In addition to the PKG-dependent mechanism, electrophysiological studies suggest CO directly stimulates BKCa channels, implicating that the channels themselves are gas sensors. CO, applied as a gas or using CO-releasing molecules (CORMs) (38), increases P51, 54, 122, 129 even in cell-free membrane patches (120, 123, 124, 126), suggestive of the possibility that CO mediates the channel directly or indirectly through those entities intimately associated with the channel proteins, possibly in the same macromolecular complex. Typically, CO-sensitive proteins, such as sGC, are heme proteins in which the reduced iron center (Fe2+) interacts with CO (16). Thus Jaggar et al. (54) postulated that the heme bound to the BKCa channel acts as a sensor for CO. Formation of the channel activity by CO reflects the ability of CO to remove the inhibitory influence of heme. However, this hypothesis is not consistent with more recent results. For example, BKCa channels treated with the oxidant H2O2, which is expected to oxidize the heme iron and disrupt the CO-protein interaction (16), remain sensitive to CO (52). Furthermore, modifications that render the BKCa channel insensitive to heme fail to disrupt the CO sensitivity (52, 123); the effects of heme and CO are mediated by different molecular loci. A recent study has shown that the RCK1 sensor in Slo1 is required for the stimulatory action of CO (52); mutation of His365, His394, or Asp367, the residues in the RCK1 sensor involved in the H+ and Ca2+ sensitivity, also eliminates the CO sensitivity (52). This finding suggests that both CO and H+ increase P1 by mimicking the action of Ca2+ on the RCK1 sensor (52). The essential roles of His365 and His394 in the CO sensitivity are consistent with the earlier observations that low pH and the histidine modifier DEPC antagonize the CO action (52, 120).

The results summarized above collectively show that the RCK1 sensor encompassing His365, His394, and Asp367 is essential for the high sensitivity of the BKCa channel to multiple ligands: Ca2+, H+, and CO. This finding has been interpreted to indicate that the RCK1 sensor is multi-ligand in nature and accommodates Ca2+, H+, or CO (51, 52). Alternatively, it may be postulated that a separate sensor for each ligand exists and that the binding information converges on the aforementioned His and Asp residues and is transmitted to the channel’s gate. Several lines of results, including that the effects of mutations affecting the Ca2+/H+ sensitivity are generally additive (51, 54), favor the former multi-ligand postulate.

The physicochemical mechanism by which CO interacts with the RCK1 sensor remains elusive. It is conventionally believed that CO-sensing proteins require a metal or heme cofactor (16). The available structures of the bacterial channel MthK, which shows a reasonable degree of primary sequence similarity to the BKCa channel, do not suggest that the Slo1 RCK1 sensor harbors any metal or heme cofactor (56, 57). As an alternative idea, an electrostatic interaction between the weak dipole moment of CO and the RCK1 ligand sensor pocket comprised of His365, His394, and Asp367 has been suggested to contribute (52). In addition to the RCK sensor, an additional CO interaction site may exist in the Slo1 channel (123). CO may increase P even at saturating concentrations of Ca2+ (121), which cannot be easily explained by the effect of CO as a Ca2+-mimetic for the RCK1 sensor (52).

In many cell types, hypoxia inhibits BKCa channels, and the direct stimulation of the BKCa channel by CO represents one of the cellular mechanisms of oxygen sensing (124). In oxygen-sensing carotid body glomus cells, Slo1 proteins are found closely associated with HMOX2 (124), which catalyzes heme to produce CO in an O2-dependent manner (61). It has been postulated that, under normoxia, CO generated by HMOX2 continuously stimulates the BKCa channel, and that hypoxia inhibits HMOX2 and removes the stimulatory influence of CO on the BKCa channel (124). The overall scheme is supported by several lines of evidence, including the results of HMOX2 gene-knockdown experiments (124). However, the HMOX2-mediated oxygen-sensing mechanism may not be the dominant one in vivo because mice with the HMOX2 gene constitutively disrupted show relatively normal hypoxic responses (82, 90).

**Reactive Oxygen/Nitrogen Species**

The aerobic existence inevitably creates reactive molecules capable of oxidizing cellular constituents including proteins (45). Although excess concentrations of the reactive molecules are clearly deleterious,
causing oxidative stress, cells utilize some of the reactive molecules at low concentrations as vital signaling molecules (53). Redox modulation of cell function has been difficult to study in part because the experimental tools available to manipulate the levels of reactive species often lack desired specificity and because reactive species are capable of readily modifying multiple amino acid residues, including cysteine, methionine, histidine, tryptophan, and tyrosine. Furthermore, some oxidation reactions are critically dependent on multivalent cations, such as Fe²⁺/Fe³⁺, which may present as contaminating species. It is not surprising then that treatment of cells/membrane patches containing BKCa channels with reactive species such as H₂O₂ has been reported to produce a myriad of effects (9, 19, 24, 68, 121, 128, 136). A consensus effect of H₂O₂, a physiological oxidant produced during normal oxygen metabolism, applied to heterologously expressed BKCa channels is inhibitory, attributed largely to a decrease in P₀ (19, 33, 77, 106, 112, 113) and, to a lesser extent, a decrease in the number of channels available to open (N) (106, 137). Typically, the diminished P₀ by H₂O₂ persists after wash, indicative of amino acid modification, but the gating change is reversed by reducing agents such as the physiological reducing agent glutathione (GSH) (19, 33) as well as DTT (33, 77, 106, 113), all of which are capable of regenerating a free sulfydryl group (-SH) in cysteine from the oxidized side chain sulfenic acid (-SOH). A variety of thiol modifying agents, such as NEM, DTNB, and MTSEA (33, 113, 121), also decrease P₀, thus implicating that the absence of free sulfydryl groups in BKCa channel is critical. A systematic Cys-to-Ala mutagenesis of Slo1 showed that oxidation of Cys911 near the Ca²⁺ bowl sensor in the distal COOH terminus decreased the energetic contribution of the Ca²⁺ bowl sensor to the channel activation (113). The proximity of Cys911 to the Ca²⁺ bowl sensor is in line with the Ca²⁺ dependence of the inhibitory effect; in the absence of Ca²⁺, the redox status of Cys911 plays little role, but the effect becomes greater with increasing concentrations of Ca²⁺ while leaving that Ca²⁺ dependence mediated by the RCK1 sensor intact (113). In addition to Cys911 at the distal COOH terminus, Cys430 in the RCK1 domain also contributes to the oxidation sensitivity of the Ca²⁺ dependence of the BKCa channel (137). Both Cys430 and Cys911 are susceptible to air oxidation and account for the rundown phenomena following patch excision (137). Additional Cys residues, whose oxidation alters the channel gating, are also present in the channel. One interesting example is the biologically modulating inclusion of the STREX exon in the cytoplasmic region of Slo1 (131). The STREX inclusion introduces additional Cys residues and potentiates the inhibitory effect of oxidation (36).

Reactive molecules other than H₂O₂ have been reported to affect BKCa channels: O₂⁻ (72, 113), NO (2, 18, 19, 69), and peroxynitrite (ONOO⁻) (72, 77, 113). Many of the effects are inhibitory in nature, and some of the inhibitory effects are mediated by Cys911 (77, 113). In addition, nitrothiosylation (for review, see Ref 81) of yet an unidentified cysteine residue induced by NO-releasing compounds may increase the channel activity (2, 18, 69). It may be noted that application of a NO-releasing compound to heterologously expressed BKCa channels failed to increase P₀ (52).

Direct biochemical/proteomic evidence that any of the Cys residues in the BKCa channels are dynamically oxidized and reduced under physiological conditions is not yet available. Pathophysiologically, Cys oxidation in the BKCa channel is likely to be a contributing factor in those disease states where oxidative stress is implicated. One such condition is diabetes-induced vascular dysfunction where oxidative stress is an important contributing factor and suggested to induce oxidation of Cys911, leading to impaired vasorelaxation (77).

Methionine is another amino acid readily susceptible to oxidation. Oxidation of methionine to methionine sulfoxide by the addition of an oxygen atom to its reactive sulfur atom changes its flexible and nonpolar side chain to a rigid and polar one, roughly equivalent to the side chain of lysine (15). Methionine oxidation has marked functional effects in many proteins, including calmodulin (14), calcium/calmodulin-dependent kinase (35), and ion channels (28, 49, 60) and is implicated in many phenomena including aging and neurodegenerative disorders (49, 107). In BKCa channels, oxidation of any of one of the three Met residues MeS63, MeT12, and MeT39 located in the RCK1 and RCK2 domains by chloramines increases P₀ in the absence of Ca²⁺ in part by shifting GV to the negative direction by ~50 mV (100). The overall shift in the voltage dependence is caused by stabilization of the activated state of VSD and of the open state of the gate (100). Interestingly, the stimulatory effect of methionine oxidation in Slo1 is drastically potentiated by conformational changes in VSD (59) but a mechanistic interpretation of the finding remains to be developed.

**Lipids and Metabolites**

Lipids are a structurally diverse group of molecules that include fatty acids, phospholipids, and sterols, and many lipids and lipid-related metabolites are recognized as cellular signaling molecules involved in regulation of a variety of physiological and pathophysiological processes including gating of BKCa channels (17). The interest in modulation of BKCa channels by lipids is further stimulated by the recent finding that voltage-dependent gating of ion channels critically depends on lipids (13).

**Fatty acids: its metabolism**

Anchidonic acid is synthesized from dietary arachidonic acid as well as eicosatetraenoic acid (EET), dihydriodole (DHETs) exert effects (32, 127) and heterolologously expressed BKCa channels has in P₀ [3, 29, 32] and changes in P₀. A functional domain of the gating ring is impaired vasorelaxation (77).

Despite the advancement in understanding the structural, functional, and pathophysiologic roles that the fatty acids may play, more is learned about the effects of fatty acids on the gating ring of BKCa channels.
channels critically depends on membrane phospholipids (133).

**Fatty acids: arachidonic acid and its metabolites**

Arachidonic acid, a fatty acid, initially synthesized from dietary sources in select cells, is stored in cell membranes and released to the cytoplasm by the action of phospholipases. Once released, arachidonic acid as well as its metabolites such as hydroxyeicosatetraenoic acids (HETEs), epoxyeicosatrienoic acid (EET), and dihydroxyeicosatrienoic acids (DHETs) exert a variety of effects (93, 97), including stimulation of BKCa channels in pituitary tumor cells (32, 127), artery smooth muscle cells (3, 6, 29, 46, 63), and heterologous expression systems (39, 110). The stimulatory action of various fatty acids on the BKCa channel has been typically attributed to an increase in $P_o$ (3, 29, 32) and/or to an increase in N (3). How the changes in $P_o$ correlate with the changes in the functional domains of the channel, the gate, the VSDs, and the gating ring has not been fully explored.

Despite the large number of fatty acids capable of enhancing the BKCa channel activity, considerable structural specificity has been reported, suggesting that the fatty acid effector, presumably the channel itself, has specific interaction sites. To effectively increase $P_o$, fatty acids should have a cis conformation (32), a relatively long tail group (C > 8), and a negatively charged head group (29). For example, oleic acid (C18), arachidonic acid (C20), and eicosapentaenoic acid (C20) meet these structural requirements and increase BKCa $P_o$ by several fold (29, 32). The location of the double bonds in fatty acids may be also important (39, 127).

The biophysical mechanism and the molecular components necessary for the stimulatory action of the fatty acids are not yet clear. Because fatty acids may "flip" across cell membranes (39), it has been difficult to determine whether the fatty acid interaction sites face the intracellular side or extracellular side. Additionally, there is no clear consensus whether the auxiliary subunits of the BKCa channel, β1–4, are required for the fatty acid action. The predominantly vascular auxiliary subunit β1 may not be required for 17,18-epoxyeicosatetraenoic acid, one metabolite of arachidonic acid, to activate the BKCa channel, suggesting that the pore-forming Slo1 subunit is sufficient (46). In contrast, arachidonic acid increases $P_o$ when Slo1 is expressed with β2 or β3 but not when expressed alone or coexpressed with β4 (110). One likely but complicating possibility is that fatty acids differ in their β-subunit requirement.

**Phospholipids**

Phospholipids are an important constituent of cell membranes and have been known to alter the functions of BKCa channels in many ways (40, 85, 91). Phosphatidylinositol 4,5-bisphosphate (PIP$_2$) in particular has been a subject of intense investigation because this negatively charged phospholipid influences numerous ion channels and because it serves as a precursor for inositol 1,4,5-triphosphosphate (IP$_3$) and diacylglycerol (DAG), both of which in turn modulate
many ion channels. This multifunctional nature of PIP₂ is physiologically noteworthy but has hindered execution of well controlled experiments (109). Nevertheless, an emerging paradigm is that PIP₂, especially when phosphorylated by lipid kinases, directly affects many ion channels (109), and BKₐ channels appear to be no exception. This is illustrated in a recent study by Vainhanathan et al. (116), who showed that application of exogenous PIP₂ to the cytoplasmic side at a physiological concentration (-10 μM) (83) increases currents through both native vascular and heterologously expressed BKₐ channels. The current enhancing effect of PIP₂ depends on its negative phospho-phate group and the inositol moiety, and the washout kinetics is influenced by the acyl chain length. The importance of the negative phosphate group in PIP₂ prompted Vainhanathan et al. to mutate a cluster of three positively charged residues (BBK) in the Slo1 BKₐ linker segment. Neutralization of the charged residues noticeably diminished the overall effect of PIP₂, perhaps suggesting that the sequence BBK is a PIP₂ interaction site. The involvement of the Slo6-BCK1 linker in the PIP₂ action is reminiscent of the results obtained in KCNQ, another voltage-gated K⁺ channel (130). Biophysically, PIP₂ increases P_in the BKₐ channel by shifting G/V to the negative direction by ~15 mV at an intermediate concentration of Ca²⁺. It is interesting to note that a similar shift in the voltage dependence is observed when the Slo6-BCK1 linker is shortened (86); binding of PIP₂ to the linker region may affect the coupling process between the gate in the pore module and the cytoplasmic gating structure.

Steroid hormones
Steroid hormones are well known for their genomic effects, but their acute, non-genomic mode of action involving direct binding to membrane-bound effectors, including BKₐ channels, is starting to be appreciated (86). Multiple steroid hormones, including estrogens (117), testosterone (43), dehydroepiandrosterone (62), and glucocorticoids (75), have been reported to acutely affect BKCa channels. Acute modulation of BKCa channels greatly expands their functional repertoires, allowing the channels to contribute to multitudes of physiological and pathophysiological phenomena. Important mechanistic insights into the channel regulation by small molecules such as Ca²⁺, Mg²⁺, and H⁺ are now available, and the mechanisms of action of other modulatory agents on the BKₐ channel gating should become better elucidated in the near future. The BKₐ channel gating is allosterically mediated by three major domains of the channel, the pore, VSDs, and the gating ring, all of which undergo rapid and marked conformational changes, and any modifications of the energetics and kinetics of the functional domains could change the channel current size. Thus additional modulatory phenomena of BKₐ channels are certainly waiting to be discovered. Such future studies will incorporate the realization that BKₐ channels form macromolecular complexes with other signaling molecules, as voltage-dependent Ca²⁺ channels (12, 13), HMOX2 (124), and protein kinases/phosphatases (78). The macromolecular assembly formation itself may be dynamic and subject to modulation. The knowledge obtained from studies of the modulator action could contribute to rational design of therapeutically useful low-molecular-weight compounds targeting BKₐ channels. Dysfunction of a modulatory pathway may underlie a disease state, and synthetic compounds could be designed to regulate the pathway in a predictable manner. Much work and excitement lie ahead.

Concluding Remarks

Acute modulation of BKₐ channels greatly expands their functional repertoires, allowing the channels to contribute to multitudes of physiological and pathophysiological phenomena. Important mechanistic insights into the channel regulation by small molecules such as Ca²⁺, Mg²⁺, and H⁺ are now available, and the mechanisms of action of other modulatory agents on the BKₐ channel gating should become better elucidated in the near future. The BKₐ channel gating is allosterically mediated by three major domains of the channel, the pore, VSDs, and the gating ring, all of which undergo rapid and marked conformational changes, and any modifications of the energetics and kinetics of the functional domains could change the channel current size. Thus additional modulatory phenomena of BKₐ channels are certainly waiting to be discovered. Such future studies will incorporate the realization that BKₐ channels form macromolecular complexes with other signaling molecules, as voltage-dependent Ca²⁺ channels (12, 13), HMOX2 (124), and protein kinases/phosphatases (78). The macromolecular assembly formation itself may be dynamic and subject to modulation. The knowledge obtained from studies of the modulator action could contribute to rational design of therapeutically useful low-molecular-weight compounds targeting BKₐ channels. Dysfunction of a modulatory pathway may underlie a disease state, and synthetic compounds could be designed to regulate the pathway in a predictable manner. Much work and excitement lie ahead.

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