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K2P Potassium Channels, Mysterious and Paradoxically Exciting

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Abstract

New evidence reveals that the common electrolyte disorder hypokalemia can induce K2P1 channels that are normally selective for K^+ to break the rules and conduct Na^+. This defiant behavior leads to paradoxical depolarization of many cells in the heart, increasing the risk for lethal arrhythmia. The new research resolves a mystery uncovered 50 years ago and bestows an array of new riddles. Here, I discuss how K2P1 might achieve this alchemy—through stable residence of the K^+ selectivity filter in a Na^+-conductive state between its open and C-inactive configurations—and predict that other K^+ channels and environmental stimuli will be discovered to produce the same excitatory misconduct.

The human heart gave physiologists a splendid mystery (1, 2) and clinicians a recurrent challenge (3) in 1956, the year Elvis growled “Heartbreak Hotel.” Here it is. When the concentration of K^+ in the bloodstream falls below normal—a condition called hypokalemia—many cells in the heart, including Purkinje fibers in the conduction system, paradoxically depolarize. This disrupts electrical synchrony among cardiac cells, creating a risk for dangerous arrhythmia. Depolarization is unexpected because resting cells are usually selectively permeable to K^+ and, as Nernst advises (4), lowering K^+ concentration in the blood should lead cells to hyperpolarize. Some of the depolarization results from a mysterious inward leak of Na^+, even though known pathways for that ion are closed. Now, a crack detective team—Ma, Zhang and Chen—has resolved both the paradox and the mystery of what gives rise to this leak (5). The answer is exciting: K2P1 K^+ channels—members of a large family of background channels that establish resting membrane potential (RMP)—break the rules by dynamically changing their selectivity for ions when extracellular K^+ concentration falls; under these conditions, Na^+ can pass through K2P1 using a pathway not so very different from those that conduct only K^+.

The Paradox

Mammalian cells spend precious adenosine 5′-triphosphate (ATP) to run the Na^+-K^+ pumps that maintain high concentrations of intracellular K^+ and keep internal Na^+ low. Open K^+ channels allow K^+ ions to exit cells down their concentration gradient. K^+ efflux produces excess negative charge inside the cell until, at equilibrium, the concentration gradient is balanced by electrostatic forces favoring K^+ influx and there is no net K^+ movement. The membrane voltage that yields equilibrium for a particular ion at a given concentration gradient is that ion’s equilibrium reversal potential (E_{ion}). At room temperature and normal conditions, E_K is about −90 mV (Fig. 1A). Most human cells have resting potentials around −70 mV because K^+ is not the only ion that crosses the membranes of quiescent cells. Of greatest relevance here, Na^+ influx moves the membrane potential in the opposite direction,

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toward E_{Na} (around +70 mV). Because more K^+ channels are open at rest than are pathways for other ions, the RMP of both skeletal and cardiac muscle cells depolarizes as external K^+ increases above normal concentrations (6). The conundrum is this: Reducing the external K^+ concentration hyperpolarizes skeletal muscle toward E_K, as expected, whereas the same maneuver causes many cardiac cells to depolarize.

Hypokalemia and Sudden Death

Hypokalemia—low blood K^+—is a common disorder. Present in up to 20% of hospitalized patients, it is usually tolerated in healthy people but can be life-threatening in individuals with cardiovascular disease (7). Normal serum K^+ concentration is 3.5 to 5 mM. Hypokalemia is classified as mild (3.0 to 3.5 mM), moderate (2.5 to 3.0 mM), or severe (<2.5 mM). The condition most often results from urinary or gastrointestinal K^+ loss because of extended use of diuretic medications or severe diarrhea. Less frequently, hypokalemia occurs because of acute shifts of K^+ from the extracellular spaces into skeletal muscle owing to stimulation of Na^+-K^+ pumps by insulin or drugs that stimulate β-adrenergic receptors; therapeutic doses of the latter can abruptly reduce plasma K^+ concentration by 1.1 mM (8). Inadequate dietary intake is rarely a cause of hypokalemia because 95% of K^+ in the body is inside cells, and this serves to maintain serum concentration unless poor intake is prolonged and coupled with substantial loss. Hypokalemia promotes cardiac arrhythmia because myocardial depolarization [due to the mysterious sodium leak considered here and also decreased activity of certain voltage-gated K^+ channels (9)] leads to automaticity—that is, firing of action potentials that are not initiated by the pacemaker of the heart, the sinoatrial node. The risk of sudden cardiac death (which occurs in 3 million people a year worldwide) is increased up to 10-fold in hospitalized patients with hypokalemia as compared with those whose serum K^+ concentrations are normal (10).

K2P Channels

K^+ background currents, long-recognized as essential to nerve and muscle function because of their role in establishing RMP (11, 12), are now largely attributed to K2P channels (Fig. 1B) (13, 14). Humans have 15 genes encoding K2P-family channels. Uncovered by their primary structure of two pore-forming (P) loop domains in each subunit (15) and operation as K^+-selective leak pathways (16–18), K2P subunits have two, nonidentical, K^+ channel signature sequences (P1, T-I/T/V-G-Y/F-G; P2, T-I/VG-F/L-G) (19). Two subunits are required to create a single ion conduction pathway, and native channels consist of either homodimers or mixed subunit complexes (20–22). Subject to a broad array of regulatory influences (23), K2P channels have been implicated in such diverse processes as sensation of oxygen tension, proton concentration, and odors; modulation of blood pressure, cardiac electrophysiology, and the immune response; apoptosis; and action of medications such as general anesthetics (24).

Active K2P channels almost always suppress excitability by stabilizing the RMP of excitable cells below the threshold for firing an action potential and by expediting repolarization to baseline. However, there is a history of defiance in this family. K2P2 (also known as TREK1) has a truncated isoform that shows stable excitatory behavior because it conducts Na^+ (25, 26). K2P1 (also known as TWIK1), the channel under focus here, has now been shown by Ma *et al*. (5) to have the remarkable ability to switch dynamically between suppressive and excitatory function. K2P1 was already known to have exceptional properties that Ma and colleagues had to overcome in order to perform their investigation. Although K2P1 reaches the plasma membrane in tissues throughout the body, it remains well hidden when sought out by electrophysiologists unless a lysine in the cytoplasmic C

terminus (K^{274}) is mutated or a desumoylating enzyme is applied. Clarification of this property revealed that the SUMO (small ubiquitin-like modifier) enzyme cascade operates not only on nuclear transcription factors but at the cell surface, where K2P1 is silenced by covalent linkage of SUMO to the ε-amino group of K^{274} on just one subunit (although both sites can be modified); the channel is activated by mutation of K^{274} because it is no longer subject to sumoylation (27, 28). The SUMO cascade is now also known to regulate the activity of voltage-gated K^+ channels at the surface of hippocampal neurons (29).

Master Sleuths

Ma and colleagues noticed that K2P1 mRNA had been identified in human but not rodent heart and that this distribution correlated with the observation of paradoxical depolarization in man but not mouse. They tested the hypothesis that hypokalemia causes K2P1 to conduct Na^+ as follows. First, they mutated K2P1 K^{274} to E so the channels would not be silenced by the SUMO cascade; then, they expressed the constitutively active channels in Chinese Hamster ovary (CHO) cells and decreased the extracellular K^+ concentration: Na^+ passed inward, depolarizing the membrane. Five other K2P channels did not conduct Na^+ in low extracellular K^+: K2P2, K2P3, K2P9, K2P10, and K2P13. Recognizing that K2P1 had a T in the signature sequence of the P1 loop where the other channels had an I (Fig. 1C), Ma *et al*. created a K2P9 mutant in which the native I was substituted with T (K2P9-I94T) and found that it now conducted Na^+ in low extracellular K^+. The corresponding K2P13 mutant, K2P13-I112T, remained selective for K^+, indicating that the T was important but not sufficient to transfer the phenotype. In an experimental flourish, they analyzed a mouse heart cell line (HL1) and showed that these cells hyperpolarized from −78 to −102 mV when the extracellular K^+ was decreased from 4 mM to 1 mM, unless they expressed human K2P1; in that case, the cells slowly depolarized in low K^+ to −63 mV (Fig. 1, A and D). Moreover, 45% of human primary cardiac myocytes showed paradoxical depolarization from −78 mV to −44 mV when exposed to low concentrations K^+, whereas 55% hyperpolarized to −94 mV—a finding that is consistent with two levels of RMP seen in canine Purkinje fibers (6). Further, when native K2P1 in the human cardiocytes was knocked down with short hairpin RNA (shRNA), paradoxical depolarization was observed in only 15% of the cells, and 85% responded with hyperpolarization.

Changing the Selectivity of K^+ Channels

These results are marvelous and exciting but not surprising. The real questions are, why did it take so long to discover a K^+ channel that alters selectivity in response to a natural environmental challenge, how many more channels are there that do so, and to what stimuli do they react? Changes in the selectivity of K^+ channels have been induced by experimentalists who used point mutations to define the signature sequence and its role (30, 31). Further, natural mutations in K^+ channel filter sequences, identified by their association with diseases in mice or men, have also generated Na^+ conductive pathways (32–34). Although these mutations altered ion selectivity by modifying the filter directly, selectivity has also been changed by mutations distant from the filter in the primary sequence (35, 36) or by co-assembly with accessory subunits that concurrently modify other conduction pathway attributes such as unitary current or the affinity of blockers that bind in the pore (37–40). Moreover, there are families of channels that favor K^+ but are less selective, allowing Na^+ to permeate to varying degrees; these include the hyperpolarization-activated cyclic nucleotide-gated channels (41) and the small-conductance calcium-activated K^+ channels (42). Particularly germane is K2P2, a channel that has two natural forms that vary only in the length of their intracellular N-termini because of alternative translation initiation. Full-length K2P2 and K2P2Δ56 are produced in different amounts in different regions of the rat central nervous system in a manner that varies with development. Whereas full-length

K2P2 is very selective for K^+, the short version lacking 56 residues conducts Na^+, leading to membrane depolarization (25, 26). Thus, K^+ channel signature sequences do not need to be mutated to enable Na^+ conduction but can be persuaded to do so by factors external to the filter.

K2P2Δ56 is an example of a natural, nonpathological structural change that allows Na^+ ions to pass through an otherwise K^+-selective channel under physiological conditions, presumably because the conduction pathway has been stably reconfigured. In contrast, Ma *et al*. (5) demonstrate that K2P1 can change its selectivity in dynamic fashion with the small decreases in external K^+ seen in humans. Although prior observations of induced Na^+ conduction through K^+-selective channels required a nonphysiological experimental manipulation—removal of internal K^+ (43–46)—those studies suggest how K2P1 might operate in vivo. Kv2.1 voltage-gated K^+ channels undergo a natural gating transition mediated by the selectivity filter. In the presence of lowered internal K^+, this transition, called C-type inactivation, moves the channels through three functional states, including one that conducts Na^+. There is an open state that is highly selective for K^+, a transient intermediate state that is more permeable to Na^+ than K^+, and a non-conducting inactivated state (46). The transient state was inferred by functional studies to result from dynamic rearrangement of the selectivity filter (45). Recent computational and structural studies support the idea that there is a Na^+-permeable pore conformation in K^+ channels in between the open and C-type, nonconducting states.

The first crystal structure of a K^+ channel showed that the signature sequence in the bacterial KcsA channel met all expectations (47, 48). It confirmed the proposal that the K^+ filter would be formed by backbone carbonyl oxygens (49), a suggestion based on prior consideration of the Na^+ filter (50), and the conclusion drawn from studies of blockade by barium that there would be at least four K^+-binding sites in the pore (51, 52). The structure also appeared to support the prevailing “Goldilocks hypothesis” for ion selectivity, that the dimensions of the conduction pathway were just right to bind K^+ but too large to coordinate smaller Na^+ and satisfy the energetic cost of its dehydration. However, the snug-fit proposal was argued to be at odds with the natural flexibility of proteins (53), computations used to assess ion-protein and ion-ion interaction energies and a resultant dynamic model for selectivity based on flexible binding sites with fluctuating dipoles (54); indeed, the model anticipated conformational changes within the selectivity filter that would permit Na^+ permeation (55).

Although KcsA is not voltage-gated, it recapitulates the hallmarks of C-type inactivation (56). Moreover, the KcsA selectivity filter can be resolved with x-ray crystallography in three distinct structures (Fig. 1E): one that is open with four ion-binding sites, an intermediate state with three sites, and a nonconducting, C-type inactive state with two sites (57). Supporting the association of ion selectivity and C-type transition, a network of hydrogen bonds behind the KcsA filter that alter C-type inactivation (56) appears to be present in mammalian inward rectifier K^+ channels and to influence their selectivity (58). That four contiguous K^+-binding sites are required for high K^+ selectivity, whereas three allow Na^+ to permeate, is also consistent with structural studies of the NaK channel that conducts both Na^+ and K^+ (59) and the thesis that the same filter residues can coordinate K^+ and Na^+ with different chemistries (60).

The idea that dynamic changes in selectivity can occur is not new (61). The big news is that a highly selective K^+ channel can dynamically respond to small, physiologically relevant changes in the environment (in this case, the concentration of the permeant ion). Why has this only been observed with K2P1? Perhaps because most K^+ channels (including KcsA) have a restrictive activation gate on the intracellular end of the pathway that moves in a

coupled fashion with the filter gate (62) to close the pathway at its intracellular end (so no Na^+ or K^+ can flow) and to destabilize the intermediate state. In contrast, K2P channels open and close through C-type gating at the selectivity filter (18, 63), and the inner portion of the channel moves (in a manner coupled to the filter) but does not restrict ion permeation (64).

I propose that a Na^+-permeable intermediate state that is occluded or short-lived in most K^+ channels is stabilized in K2P1 by hypokalemia and created in K2P2 by an N terminus shortened by 56 residues. Given that filter flexibility and C-type inactivation appear to be shared characteristics of K^+ channels, it seems likely that more channels will be found to show dynamic changes in selectivity. It also seems reasonable to anticipate that other natural environmental stimuli (such as pH, hormones, medications, and second messengers) will mediate changes in selectivity.

More Mysteries

The work of Ma and colleagues provides a delightful array of new riddles. They observe K2P1 to remain selective for 60 s after the extracellular K^+ concentration is decreased, so that cells hyperpolarize briefly and then depolarize unhurriedly, with a time constant of ~6 min (Fig. 1D). Restoration of selectivity on K^+ repletion is even slower, taking 40 to 80 min. However, the changes in selectivity demonstrated in canine Purkinje fibers are essentially immediate (6). Does this reflect different behaviors of the same channel across species or (gasp) different channel types? The slow response of K2P1 to hypokalemia led Ma and friends to posit slow changes in filter structure that become locked in place. Alternatively, events this serene are often due to intracellular regulation, such as control of K2P0 by C-terminal phosphorylation, a process that depends on the conformation of the selectivity filter (18). Why does shRNA knockdown of K2P1 produce all-or-none effects on paradoxical depolarization rather than graded responses, given that the reagents suppress only a portion of the channels? Is there a role for the SUMO cascade in the paradoxical response? Given that K2P2Δ56 has an I in the signature sequence and can pass Na^+, is the T in K2P1 necessary only for dynamic selectivity changes? Does the change in K2P1 selectivity help to stabilize myocardial activity in the face of normal decreases in serum K^+ that might suppress function were the cells to hyperpolarize as expected?

Those old-time physiologists gifted us a paradox. The King gave us “Heartbreak.” The Bee Gees asked, “How can you mend a broken heart?” Ma, Zhang, and Chen have provided answers: Monitor and adjust serum K^+ concentration and work to develop therapies that target K2P1 in order to protect those individuals most at risk of arrhythmia.

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References and Notes

1. Weidmann, S. Elektrophysiologie der Herzmuskelfaser. Huber; Bern: 1956.
2. Adrian RH. The effect of internal and external potassium concentration on the membrane potential of frog muscle. J. Physiol. 1956; 133:631–658. [PubMed: 13368111]
3. Goldstein, SAN.; Levy, DI. Seldin and Giebisch’s, The Kidney: Physiology and Pathophysiology. ed. 4. Alpern, RJ.; Hebert, SC., editors. Elsevier; New York: 2008. p. 1407-1428.
4. Nernst W. Zur Kinetik der in Losung befindlichen Korper. I. Theorie der Diffusion. Z. Phys. Chem. 1888; 2:613–637.

5. Ma L, Zhang X, Chen H. TWIK-1 two-pore domain potassium channels change ion selectivity and conduct inward leak sodium currents in hypokalemia. Sci. Signal. 2011; 4:ra37. [PubMed: 21653227]
6. Gadsby DC, Cranefield PF. Two levels of resting potential in cardiac Purkinje fibers. J. Gen. Physiol. 1977; 70:725–746. [PubMed: 591921]
7. Gennari FJ. Hypokalemia. N. Engl. J. Med. 1998; 339:451–458. [PubMed: 9700180]
8. Scheinin M, Koulu M, Laurikainen E, Allonen H. Hypokalaemia and other non-bronchial effects of inhaled fenoterol and salbutamol: A placebo-controlled dose-response study in healthy volunteers. Br. J. Clin. Pharmacol. 1987; 24:645–653. [PubMed: 2829953]
9. Sanguinetti MC, Jiang C, Curran ME, Keating MT. A mechanistic link between an inherited and an acquired cardiac arrhythmia: HERG encodes the IKr potassium channel. Cell. 1995; 81:299–307. [PubMed: 7736582]
10. Kjeldsen K. Hypokalemia and sudden cardiac death. Exp Clin Cardiol. 2010; 15:e96–e99. [PubMed: 21264075]
11. Goldman DE. Potential, impedance, and rectification in membranes. J. Gen. Physiol. 1943; 27:37–60. [PubMed: 19873371]
12. Hodgkin AL, Katz B. The effect of sodium ions on the electrical activity of giant axon of the squid. J. Physiol. 1949; 108:37–77. [PubMed: 18128147]
13. Goldstein SAN, Bockenhauer D, O'Kelly I, Zilberberg N. Potassium leak channels and the KCNK family of two-P-domain subunits. Nat. Rev. Neurosci. 2001; 2:175–184. [PubMed: 11256078]
14. Plant, LD.; Bayliss, DA.; Kim, D.; Lesage, F.; Goldstein, SAN. Two-P Potassium Channels. 2009. IUPHAR database
15. Ketchum KA, Joiner WJ, Sellers AJ, Kaczmarek LK, Goldstein SAN. A new family of outwardly rectifying potassium channel proteins with two pore domains in tandem. Nature. 1995; 376:690–695. [PubMed: 7651518]
16. Goldstein SAN, Price LA, Rosenthal DN, Pausch MH. ORK1, a potassium-selective leak channel with two pore domains cloned from *Drosophila melanogaster* by expression in *Saccharomyces cerevisiae*. Proc. Natl. Acad. Sci. U.S.A. 1996; 93:13256–13261. [PubMed: 8917578]
17. Ilan N, Goldstein SAN. Kcnkø: Single, cloned potassium leak channels are multi-ion pores. Biophys. J. 2001; 80:241–253. [PubMed: 11159398]
18. Zilberberg N, Ilan N, Goldstein SA. KCNKø: Opening and closing the 2-P-domain potassium leak channel entails "C-type" gating of the outer pore. Neuron. 2001; 32:635–648. [PubMed: 11719204]
19. Single-letter abbreviations for amino acid residues are as follows: Ala A, Cys C, Asp D, Glu E, Phe F, Gly G, His H, Ile I, Lys K, Leu L, Met M, Asn N, Pro P, Gln Q, Arg R, Ser S, Thr T, Val V, Trp W, Tyr Y. X indicates "any amino acid, and the "/" in the P1 loop and P2 loop sequences indicates "or.".
20. Lopes CMB, Zilberberg N, Goldstein SAN. Block of Kcnk3 by protons. Evidence that 2-P-domain potassium channel subunits function as homodimers. J. Biol. Chem. 2001; 276:24449–24452. [PubMed: 11358956]
21. Kollewe A, Lau AY, Sullivan A, Roux B, Goldstein SAN. A structural model for K2P potassium channels based on 23 pairs of interacting sites and continuum electrostatics. J. Gen. Physiol. 2009; 134:53–68. [PubMed: 19564427]
22. Berg AP, Talley EM, Manger JP, Bayliss DA. Motoneurons express heteromeric TWIK-related acid-sensitive K^+ (TASK) channels containing TASK-1 (KCNK3) and TASK-3 (KCNK9) subunits. J. Neurosci. 2004; 24:6693–6702. [PubMed: 15282272]
23. Enyedi P, Czirják G. Molecular background of leak K+ currents: Two-pore domain potassium channels. Physiol. Rev. 2010; 90:559–605. [PubMed: 20393194]
24. Thomas, D.; Goldstein, SAN. Encyclopedia of Neuroscience. Squire, LR., editor. Elsevier, Academic Press; Oxford: 2009. p. 1207-1220.
25. Thomas D, Plant LD, Wilkens CM, McCrossan ZA, Goldstein SAN. Alternative translation initiation in rat brain yields K2P2.1 potassium channels permeable to sodium. Neuron. 2008; 58:859–870. [PubMed: 18579077]

26. Yang S-B, Jan LY. Thrilling moment of an inhibitory channel. Neuron. 2008; 58:823–824. [PubMed: 18579071]
27. Rajan S, Plant LD, Rabin ML, Butler MH, Goldstein SAN. Sumoylation silences the plasma membrane leak K^+ channel K2P1. Cell. 2005; 121:37–47. [PubMed: 15820677]
28. Plant LD, Dementieva IS, Kollewe A, Olikara S, Marks JD, Goldstein SA. One SUMO is sufficient to silence the dimeric potassium channel K2P1. Proc. Natl. Acad. Sci. U.S.A. 2010; 107:10743–10748. [PubMed: 20498050]
29. Plant LD, Dowdell EJ, Dementieva IS, Marks JD, Goldstein SAN. SUMO modification of cell surface Kv2.1 potassium channels regulates the activity of rat hippocampal neurons. J. Gen. Physiol. 2011; 137:441–454. [PubMed: 21518833]
30. MacKinnon R, Miller C. Mutant potassium channels with altered binding of charybdotoxin, a pore-blocking peptide inhibitor. Science. 1989; 245:1382–1385. [PubMed: 2476850]
31. Heginbotham L, Lu Z, Abramson T, MacKinnon R. Mutations in the K^+ channel signature sequence. Biophys. J. 1994; 66:1061–1067. [PubMed: 8038378]
32. Kofuji P, Hofer M, Millen KJ, Millonig JH, Davidson N, Lester HA, Hatten ME. Functional analysis of the weaver mutant GIRK2 K^+ channel and rescue of weaver granule cells. Neuron. 1996; 16:941–952. [PubMed: 8630252]
33. Lees-Miller JP, Duan Y, Teng GQ, Thorstad K, Duff HJ. Novel gain-of-function mechanism in K(+) channel-related long-QT syndrome: Altered gating and selectivity in the HERG1 N629D mutant. Circ. Res. 2000; 86:507–513. [PubMed: 10720411]
34. Choi M, Scholl UI, Yue P, Björklund P, Zhao B, Nelson-Williams C, Ji W, Cho Y, Patel A, Men CJ, Lolis E, Wisgerhof MV, Geller DS, Mane S, Hellman P, Westin G, Åkerström G, Wang W, Carling T, Lifton RP. K^+ channel mutations in adrenal aldosterone-producing adenomas and hereditary hypertension. Science. 2011; 331:768–772. [PubMed: 21311022]
35. Bichet D, Lin Y-F, Ibarra CA, Huang CS, Yi BA, Jan YN, Jan LY. Evolving potassium channels by means of yeast selection reveals structural elements important for selectivity. Proc. Natl. Acad. Sci. U.S.A. 2004; 101:4441–4446. [PubMed: 15070737]
36. Tai K-K, Goldstein SAN. The conduction pore of a cardiac potassium channel. Nature. 1998; 391:605–608. [PubMed: 9468141]
37. Sesti F, Goldstein SAN. Single-channel characteristics of wild-type I_{Ks} channels and channels formed with two minK mutants that cause long QT syndrome. J. Gen. Physiol. 1998; 112:651–663. [PubMed: 9834138]
38. Wollnik B, Schroeder BC, Kubisch C, Esperer HD, Wieacker P, Jentsch TJ. Pathophysiological mechanisms of dominant and recessive KVLQT1 K^+ channel mutations found in inherited cardiac arrhythmias. Hum. Mol. Genet. 1997; 6:1943–1949. [PubMed: 9302275]
39. Abbott GW, Goldstein SAN. A superfamily of small potassium channel subunits: Form and function of the MinK-related peptides (MiRPs). Q. Rev. Biophys. 1998; 31:357–398. [PubMed: 10709243]
40. Goldstein SA, Miller C. Site-specific mutations in a minimal voltage-dependent K^+ channel alter ion selectivity and open-channel block. Neuron. 1991; 7:403–408. [PubMed: 1910787]
41. Kaupp UB, Seifert R. Molecular diversity of pacemaker ion channels. Annu. Rev. Physiol. 2001; 63:235–257. [PubMed: 11181956]
42. Shin N, Soh H, Chang S, Kim DH, Park C-S. Sodium permeability of a cloned small-conductance calcium-activated potassium channel. Biophys. J. 2005; 89:3111–3119. [PubMed: 16143634]
43. Korn SJ, Ikeda SR. Permeation selectivity by competition in a delayed rectifier potassium channel. Science. 1995; 269:410–412. [PubMed: 7618108]
44. Starkus JG, Kuschel L, Rayner MD, Heinemann SH. Ion conduction through C-type inactivated Shaker channels. J. Gen. Physiol. 1997; 110:539–550. [PubMed: 9348326]
45. Wang Z, Zhang X, Fedida D. Regulation of transient Na^+ conductance by intra- and extracellular K^+ in the human delayed rectifier K^+ channel Kv1.5. J. Physiol. 2000; 523:575–591. [PubMed: 10718739]
46. Kiss L, LoTurco J, Korn SJ. Contribution of the selectivity filter to inactivation in potassium channels. Biophys. J. 1999; 76:253–263. [PubMed: 9876139]

47. Doyle DA, Lee A, Lewis J, Kim E, Sheng M, MacKinnon R. Crystal structures of a complexed and peptide-free membrane protein-binding domain: Molecular basis of peptide recognition by PDZ. Cell. 1996; 85:1067–1076. [PubMed: 8674113]
48. Zhou Y, Morais-Cabral JH, Kaufman A, MacKinnon R. Chemistry of ion coordination and hydration revealed by a K^+ channel-Fab complex at 2.0 A resolution. Nature. 2001; 414:43–48. [PubMed: 11689936]
49. Bezanilla F, Armstrong CM. Negative conductance caused by entry of sodium and cesium ions into the potassium channels of squid axons. J. Gen. Physiol. 1972; 60:588–608. [PubMed: 4644327]
50. Hille B. The permeability of the sodium channel to organic cations in myelinated nerve. J. Gen. Physiol. 1971; 58:599–619. [PubMed: 5315827]
51. Neyton J, Miller C. Discrete $Ba2^+$ block as a probe of ion occupancy and pore structure in the high-conductance $Ca2^+$ -activated K^+ channel. J. Gen. Physiol. 1988; 92:569–586. [PubMed: 3235974]
52. Neyton J, Miller C. Potassium blocks barium permeation through a calcium-activated potassium channel. J. Gen. Physiol. 1988; 92:549–567. [PubMed: 3235973]
53. Bernxsèche S, Roux B. Molecular dynamics of the KcsA K^+ channel in a bilayer membrane. Biophys. J. 2000; 78:2900–2917. [PubMed: 10827971]
54. Noskov SY, Bernèche S, Roux B. Control of ion selectivity in potassium channels by electrostatic and dynamic properties of carbonyl ligands. Nature. 2004; 431:830–834. [PubMed: 15483608]
55. Bernèche S, Roux B. A gate in the selectivity filter of potassium channels. Structure. 2005; 13:591–600. [PubMed: 15837197]
56. Cordero-Morales JF, Jogini V, Lewis A, Vásquez V, Cortes DM, Roux B, Perozo E. Molecular driving forces determining potassium channel slow inactivation. Nat. Struct. Mol. Biol. 2007; 14:1062–1069. [PubMed: 17922012]
57. Cuello LG, Jogini V, Cortes DM, Perozo E. Structural mechanism of C-type inactivation in K^+ channels. Nature. 2010; 466:203–208. [PubMed: 20613835]
58. Dibb KM, Rose T, Makary SY, Claydon TW, Enkvetchakul D, Leach R, Nichols CG, Boyett MR. Molecular basis of ion selectivity, block, and rectification of the inward rectifier Kir3.1/Kir3.4 K^+ channel. J. Biol. Chem. 2003; 278:49537–49548. [PubMed: 14504281]
59. Derebe MG, Sauer DB, Zeng W, Alam A, Shi N, Jiang Y. Tuning the ion selectivity of tetrameric cation channels by changing the number of ion binding sites. Proc. Natl. Acad. Sci. U.S.A. 2011; 108:598–602. [PubMed: 21187421]
60. Ye S, Li Y, Jiang Y. Novel insights into K^+ selectivity from high-resolution structures of an open K^+ channel pore. Nat. Struct. Mol. Biol. 2010; 17:1019–1023. [PubMed: 20676101]
61. Khakh BS, Lester HA. Dynamic selectivity filters in ion channels. Neuron. 1999; 23:653–658. [PubMed: 10482233]
62. Cuello LG, Jogini V, Cortes DM, Pan AC, Gagnon DG, Dalmas O, Cordero-Morales JF, Chakrapani S, Roux B, Perozo E. Structural basis for the coupling between activation and inactivation gates in K^+ channels. Nature. 2010; 466:272–275. [PubMed: 20613845]
63. Zilberberg N, Ilan N, Gonzalez-Colaso R, Goldstein SAN. Opening and closing of KCNKø potassium leak channels is tightly regulated. J. Gen. Physiol. 2000; 116:721–734. [PubMed: 11055999]
64. Ben-Abu Y, Zhou Y, Zilberberg N, Yifrach O. Inverse coupling in leak and voltage-activated K^+ channel gates underlies distinct roles in electrical signaling. Nat. Struct. Mol. Biol. 2009; 16:71–79. [PubMed: 19098918]

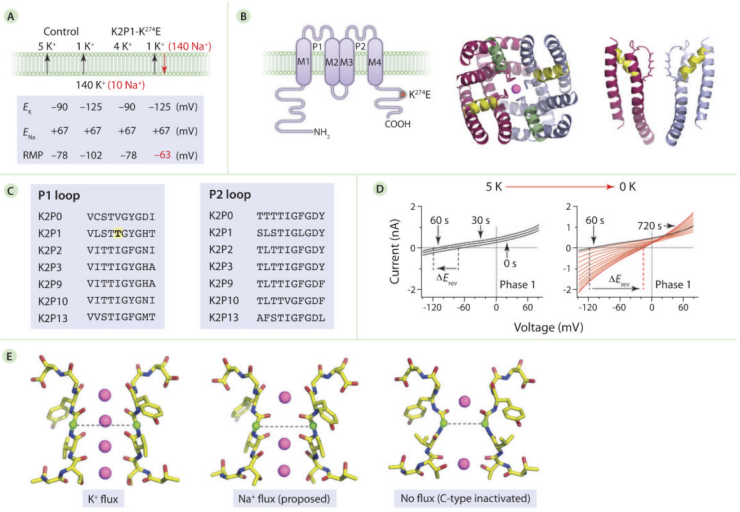

Fig. 1.

(**A**) Reversal potentials for K^+, Na^+, and RMP of CONTROL HL-1 cells and those expressing K2P1-K^{274}E under normal and hypokalemic conditions. E_K and E_{NA} were calculated with 4 mM K^+ and 1 mM K^+ at 20°C, and RMP for mouse HL-1 cells were under control conditions or expressing K2P1-K^{274}E channels from Ma *et al*. (5) showing depolarization to −63 mV, with hypokalemia in the latter case. (**B**) K2P0 subunit and homology model. (Left) Single subunit topology showing M1 to M4 (transmembrane segments 1 to 4) and the two P loops (P1, P2). K^{274}, the residue mutated in K2P1 to avoid sumoylation, is indicated. (Middle) Homology model for the dimeric channel built on the basis of pairs of residues that interact (21) viewed from outside the cell and indicating a K^+ ion (purple) in the pore shows bilateral symmetry with a fourfold symmetric selectivity filter. One subunit is colored in purple, and the other is blue. The P1 pore helices are in yellow, and P2 pore helices are in green. (Right) Side view of domain I of both subunits. (**C**) Signature sequences of P1 loop and P2 loop in K2P0 (dORK1), K2P1, K2P2, K2P3 (TASK1), K2P9 (TASK3), K2P10 (TREK2), and K2P13 (THIK1). Although most K2P channels have an I in the P1 loop, the T in K2P1 (highlighted yellow) is identified by Ma *et al*. as important for its ability to undergo dynamic changes in ion selectivity with hypokalemia. Single-letter abbreviations for the amino acids are standard. (**D**) K2P1-K^{274}E channels are expressed in CHO cells, and external K^+ decreased from 5 mM to nominally no K^+ (0K). Changes in RMP (ΔE_{rev}) reveal rapid hyperpolarization (phase 1) followed by slow paradoxical depolarization (phase 2) [from Fig. 6A in (5)]. (**E**) Three classes of KcsA filter structure determined with the lower activation gate in the open configuration from (57). (Left) The filter with 4 K^+ binding sites conducts K^+ (K flux). (Middle) The intermediate state shows three K^+-binding sites in the filter and is proposed in this essay to be similar to the Na^+-conductive state in K2P1-K^{274}E with hypokalemia (Na^+ flux proposed). (Right) The nonconducting, C-type inactivated filter shows two K^+-binding sites.