SYMPOSIUM REPORT

Two-pore-domain potassium channels in smooth muscles: new components of myogenic regulation

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Gastrointestinal (GI) smooth muscles are influenced by many levels of regulation, including those provided by enteric motor neurones, hormones and paracrine substances. The integrated contractile responses to these regulatory mechanisms depend heavily on the state of excitability of smooth muscle cells. Resting ionic conductances and myogenic responses to agonists and physical parameters, such as stretch, are important in establishing basal excitability. This review discusses the role of 2-pore-domain K^+ channels in contributing to background conductances and in mediating responses of GI muscles to enteric inhibitory nerve stimulation and stretch. Murine GI muscles express TREK-1 channels and display a stretch-dependent K^+ (SDK) conductance that is also activated by nitric oxide via a cGMP-dependent mechanism. Cloning and expression of mTREK-1 produced an SDK conductance that was activated by cGMP-dependent phosphorylation at serine-351. GI muscle cells also express TASK-1 and TASK-2 channels that are inhibited by lidocaine and external acidification. These conductances appear to provide significant background K^+ permeability that contributes to the negative resting potentials of GI muscles.

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Gastrointestinal smooth muscles are composed of excitable cells, and the excitability mechanisms regulate their contractile behaviour. Many GI muscles have intrinsic pacemaker activity (slow waves) that is supplied by interstitial cells of Cajal (ICC) that are electrically coupled to the smooth muscle syncytium. Slow waves conduct into smooth muscle cells, cause depolarization, and activate voltage-dependent Ca^{2+} channels. Inward current through these channels can result in action potentials or summate with the slow wave depolarization. Ca^{2+} entry via voltage-dependent Ca^{2+} channels initiates excitation–contraction coupling. Smooth muscle cells have negative membrane potentials that participate in regulating the level of excitability and determining responses to the many stimuli that affect contractility. The negative membrane potentials are set by K^+ conductances in the plasma membrane. GI smooth muscles vary greatly in the pattern and types of excitable events from tissue to tissue and from species to species. Much of this variability is due to differences in K^+ channel expression. Regulation of K^+ channel open probability also affects the frequency, amplitude and duration of excitable events. Many K^+ channel subtypes have been identified in smooth muscles. Most recently an important role for 2-pore-domain K^+ channels has been found in setting membrane potential and regulating responses to stretch and nitrergic stimulation. This brief review discusses some of the observations linking 2-pore-domain K^+ channels to the regulation of GI motility.

Two-pore-domain K^+ channels

There are three main classes of K^+ channels in animal cells, and classification of these channels is based primarily on structural and functional criteria. The voltage-gated K^+ channels have six transmembrane domains and one pore-forming domain. Four subunits of these channel proteins form a functional channel. Inward rectifier K^+ channels have only two transmembrane domains and a single pore domain. The final group is the two-pore-domain K^+ (K2P or KCNK) channels. These channels have four transmembrane domains and two pore domains. Two subunits of these channels form functional channels. In mammals there are six distinct subfamilies of

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the K2P channels and 15 genes that encode K2P channels (Fig. 1).

K2P channels are found in neuronal and non-neuronal tissues, and they provide a wide variety of important functions, including sensing of oxygen and pH (Patel & Honore, 2001), setting of resting membrane potentials (Kim *et al.* 1998; Millar *et al.* 2000), sensing of changes in $[K^+]$, responses to agonists (Kim, 2003), neuroprotection (Buckler & Honore, 2005), and mechanosensitivity (Bang *et al.* 2000; Kim, 2003; Chemin *et al.* 2005). These channels are also candidates for the action of volatile anaesthetics on neural excitability (Buckingham *et al.* 2005). Selective pharmacology for the K2P channels is lacking, and most detailed information about the function of these channels comes from studies on cloned channels expressed heterologously in model cells.

There is considerable evidence suggesting that K2P channels form specific and important conductances in excitable cells. For example, K2P channels provide the 'standing outward' K^+ current (I_{KSO}) in cerebellar granule cells (Millar *et al.* 2000). This conductance may be composed of TASK-1 (KCNK3), TASK-3 (KCNK9) and TREK-2 (KCNK10) channels (Millar *et al.* 2000; Han *et al.* 2002; Lauritzen *et al.* 2003). K2P channels may be involved in important cerebellar functions, such as spatial determination and accuracy, motor coordination, balance, muscle tone, and learning of motor skills. In other regions of the brain K2P channels, such as TASK-1 and TASK-3, are expressed and may function in behaviour such as sleep and wakefulness, contribute to background conductances, and provide acid and halothane sensitivity (Meuthw *et al.* 2003). Hypothalamic neurosecretory cells also express discrete background currents that may result from K2P channels (Han *et al.* 2003).

Cardiac myocytes display a background K^+ current, $I_{K,P}$, that influences the amplitude and duration of action potentials (Backx & Marban, 1993). Kim and colleagues have suggested that this conductance is due to a splice variant of TASK-1 (Kim *et al.* 1998). There is also a component of background K^+ in cardiac myocytes that is activated by arachidonic acid and stretch. These are signatures for the TREK family of K2P channels, and TREK-1 has been suggested to encode this conductance (Terrenoire *et al.* 2001). However, a search for other

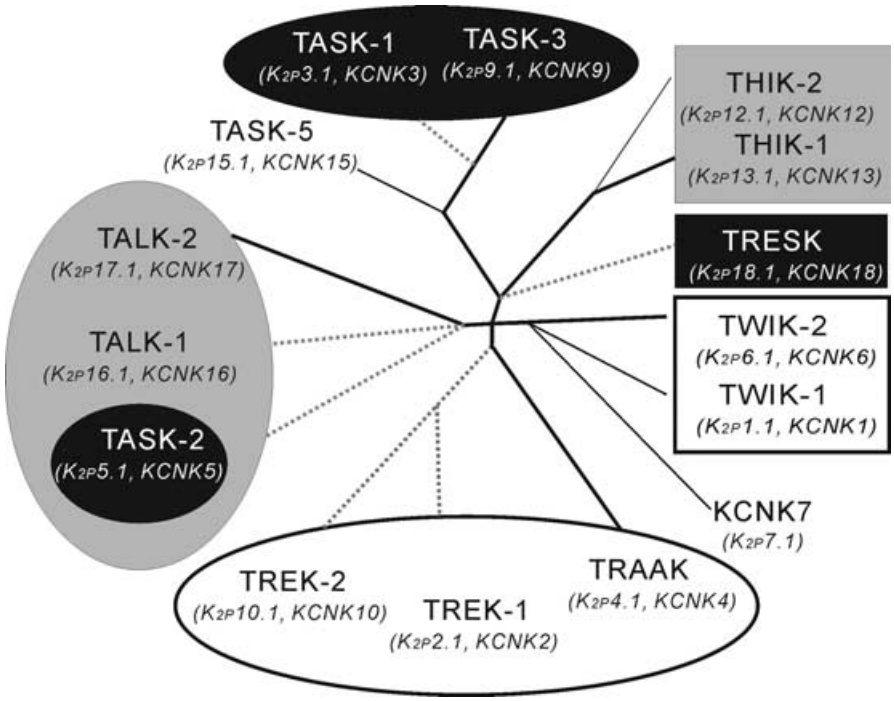

Figure 1. Structural and functional subclasses of two-pore-domain K^+ (K2P) channels
A phylogenic tree of the K2P channel family is shown with different nomenclature indicated (IUPHAR, HUGO). Six functional subfamilies (TWIK, TREK, TASK, TASK-2, THIK and TRESK) of the K2P channel can be classified based on contained functional domain. The TREK subfamily (TREK-1, TREK-2 and TRAAK, white oval) is activated by arachidonic acid, unsaturated fatty acids and mechanical stretch. Two subfamilies of extracellular pH-dependent K2P channels are identified. The TASK subfamily (TASK-1 and TASK-3, black oval) are inhibited by extracellular acidic pH. In contrast the TASK-2 family (TASK-2, TALK-1 and TALK-2, grey oval) are activated by alkaline pH. TASK-2 can also be inhibited by acidosis. Volatile anaesthetics (e.g. halothane, isoflurane) inhibit the THIK subfamily but activate TREK-1, TREK-2, TASK-1, TASK-2, TALK-1 and TRESK (dotted lines). Several K2P channels are not functionally expressed (thin lines).

K2P isoforms throughout the heart has revealed at least nine types of K2P channels distributed heterogeneously in tissues of the four chambers (Liu & Saint, 2004). Thus, the contributions and functions of these channels in cardiac muscles is likely to be complex and specific native conductances may be due to mixtures of various channel subtypes or heterodimers.

There have been limited reports of K2P channels in smooth muscle tissues. With immunohistochemical techniques, Type I cells of the carotid body were shown to have TASK-1-, TASK-2-, TASK-3- and TRAAK-like immunoreactivity (Yamamoto *et al.* 2002). Smooth muscle cells in the carotid body expressed TASK-3. Gurney and colleagues have shown TASK-1 expression in smooth muscle cells of pulmonary artery (Gurney *et al.* 2003). These authors have also demonstrated functional expression of a conductance in these cells that is inhibited by extracellular pH, Zn^{2+} and anandamide, but the conductance was insensitive to intracellular Ca^{2+}, 4-aminopyridine and quinine. These are properties that closely mimic the pharmacology of heterologously expressed TASK-1 channels, and the authors concluded that TASK-1 channels may play an important role in regulating resting membrane potential in pulmonary arteries and eliciting vasoconstriction responses during hypoxia. Another group has reported expression of TASK and TREK isoforms in human myometrium with RT-PCR, and Western analysis showed expression of TASK-1 and TREK-1 (Bai *et al.* 2005). We have found expression and function of TASK and TREK channels in GI muscles, as discussed below.

TREK-like currents in GI muscles

Most investigators remember the landmark work of Edith Bulbring during the 1950s describing the 'myogenic response' in smooth muscles (Bulbring, 1954). She showed that stretching taenia coli muscles caused depolarization. This response can result in contraction as voltage-dependent Ca^{2+} channels are activated. More recently stretch-dependent non-selective cation conductances have been identified that may underlie the depolarization of the myogenic effect in smooth muscles (e.g. Kirber *et al.* 1988; Hisada *et al.* 1993). Many regions of the GI tract would be poorly served by the dominance of such a mechanism. Several portions of GI organs, such as the gastric fundus and proximal colon, serve a reservoir function during digestion. The muscles of these regions are greatly stretched during eating and during the recovery of water and electrolytes. The reservoir function would be defeated if the myogenic response described by Bulbring dominated. In fact, the response to stretch in some regions is either no depolarization or even hyperpolarization (S. D. Koh, unpublished observation). Thus, stretch of muscles leads to stabilization of membrane potential. We reasoned that these effects could be due to the expression of stretch-dependent K^+ channels.

Some time ago we identified K^+ channels in canine colonic smooth muscle cells that had a unitary conductance of 80–90 pS and were activated by nitric oxide (Koh *et al.* 1995). We later found that channels with a similar conductance were expressed in murine colonic myocytes, and in addition to being activated by nitrergic and cGMP-dependent pathways, the channels were activated by membrane stretch or by cell elongation (Koh & Sanders, 2001). The stretch-dependent channels were determined to be K^+ channels (i.e. SDK channels) with a unitary conductance of 95 pS (Fig. 2). These channels had very low open probabilities during on-cell recording, but the channels were activated when negative pressure was applied to the outer surface of membrane patches during on-cell, single channel recordings. The effects of negative pressure were graded and release of suction restored low open probability (Fig. 3*A*). SDK channels with the same conductance were found in canine colonic myocytes as well. Excision of patches caused a large increase in channel openings, suggesting that the channels are negatively gated in cells via interactions with the cytoskeleton. Stretching the cells activated K^+ channels with the same unitary conductance as those activated by negative pipette pressure (Fig. 3*B*). The SDK channels were not blocked by tetraethylammonium and 4-aminopyridine or by reducing $[Ca^{2+}]_i$. Channels with the same conductance were also identified in other regions of the GI tract, such as bladder smooth muscle cells (S. D. Koh, unpublished observations).

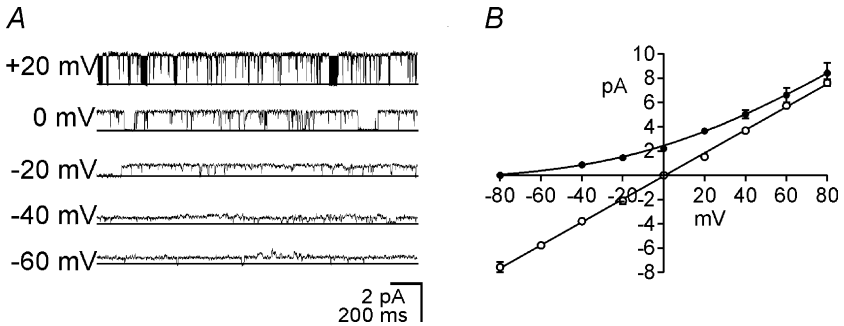

Figure 2. Current–voltage relationship of SDK channels in excised patch
A, representative traces of SDK channels showing channel activity recorded from holding potentials between −60 and +20 mV in an excised patch under asymmetrical K^+ gradients. *B*, relationship between current amplitude and voltage in asymmetrical K^+ (5 mM/140 mM) gradient was fitted by the GHK equation (●). Similar experiments were also performed in symmetrical K^+ (140 mM/140 mM) gradients (○). The conductance of SDK channels under these conditions was 95 pS.

SDK channels are similar in properties to the TREK subgroup of K2P channels. (i.e. TREK-1 (KCNK2), TREK-2 (KCNK10) and TRAAK (KCNK4); see Fig. 1). TREK channels are activated by stretch and by certain polyunsaturated fatty acids, such as arachidonic acid and lysophosphatidylcholine, and volatile anaesthetics. The channels are insensitive to intracellular [Ca^{2+}] and classical K^+ channel blockers with a unitary conductance of ~100 pS in symmetrical K^+ gradients (Patel & Honore, 2001; Franks & Honore, 2004). The channels are open over the entire range of physiological voltages, and thus may provide a background K^+ conductance that can contribute to resting membrane potential. The fact that they are activated by stretch and, as described below, by nitrergic stimulation, also suggests these channels may be a major mechanistic factor in important motility patterns in GI muscles, such as receptive relaxation.

Molecular analysis of GI smooth muscle myocytes revealed expression of 2-pore K^+ channel family genes, including TREK-1 and -2 (Koh *et al.* 2001). TRAAK was not expressed in any of the cells. The properties of mTREK-1 cloned from murine smooth muscle were compared with the native SDK currents observed in smooth muscle cells by expressing mTREK-1 in *Xenopus* oocytes and COS cells. In oocytes mTREK-1 was manifest as a rapidly activating and non-inactivating current. The resting potentials of cells expressing TREK-1 were 30 mV more negative than membrane potentials of non-transfected control oocytes. Studies to determine ion selectivity verified that mTREK-1 was a K^+ conductance, and current via these channels was poorly sensitive to classical K^+ channel-blocking drugs (Koh *et al.* 2001). Expression of mTREK-1 in COS cells resulted in a stretch-activated K^+ conductance that was also activated by 8-Br-cGMP or sodium nitroprusside. These properties match the native SDK channels in GI smooth muscle cells.

Regulation of TREK-1 channels by protein kinase A (PKA)-dependent phosphorylation of Ser-333 causes closure of channels (Patel *et al.* 1998). We confirmed this effect with mTREK-1 and showed that the activating influence of cGMP-dependent phosphorylation was due to phosphorylation at Ser-351. The amino acid sequence of mTREK-1 has two consensus sequences for PKG phosphorylation. Mutation of the protein kinase G (PKG) consensus sequence at Ser-351 (i.e. Ser-351A) blocked the stimulatory effect of sodium nitroprusside and 8-Br-cGMP (Koh *et al.* 2001). A small degree of inhibition of current occurred when cells with the

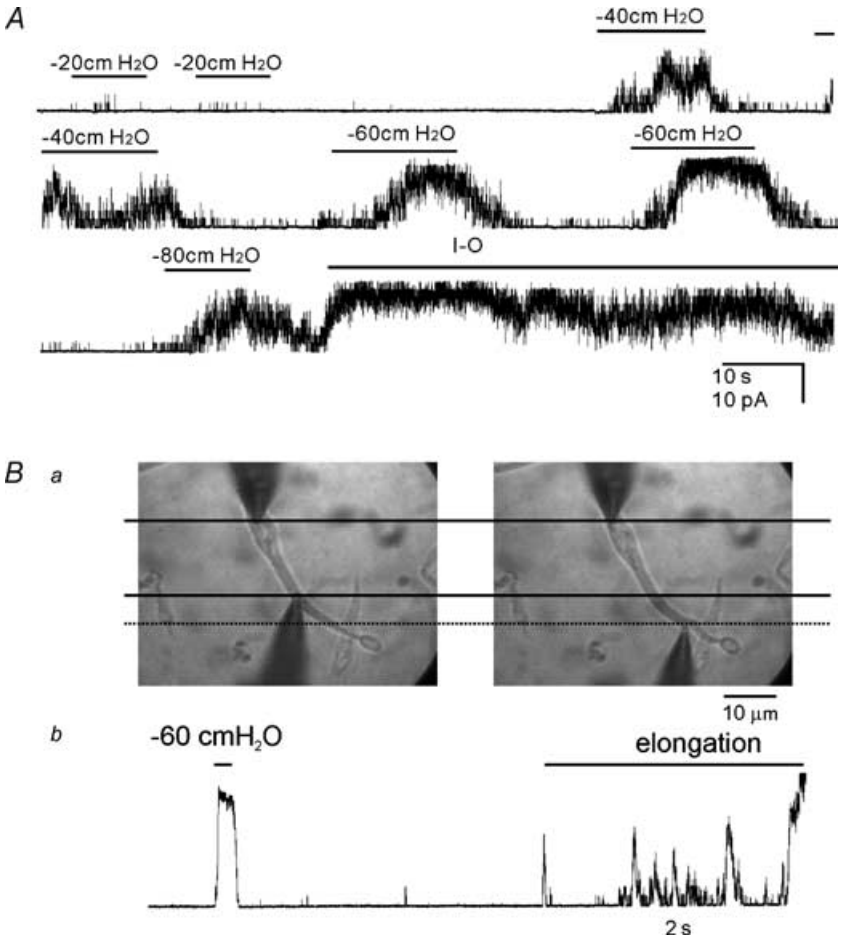

Figure 3. Mechanosensitivity of SDK channels
A, the effect of negative pressure on open probability of SDK channels. A negative pressure of −20 cmH_2O had little effect on channel activity. However, greater negative pressures (−40 cmH_2O) applied to the same patch increased NP_o (i.e. channel number × open probability) to 6.2. Further negative pressure (−60 and −80 cmH_2O) increased NP_o to the maximal level. After removal of negative pressure in each step, the open probability returned to near zero. After application of pressure pulses, the patch was excised. This caused maximal activation of channels in the patch. *B*, activation of stretch-dependent K^+ (SDK) channels via cell elongation in murine colonic myocytes. *Ba*, two patch pipettes were sealed to the same cell. Single channel currents were measured via one pipette, and the other pipette was used to stretch the cell. *Bb*, after confirming that negative patch pressure (−60 cmH_2O) activated SDK channels in this patch, the cells were elongated (in this example by 8 μm). Cell elongation caused activation of channels with the same properties as negative pressure.

Ser-351A mutant channels were exposed to 8-Br-cGMP, suggesting that cGMP-dependent mechanisms (i.e. either activation of PKG or cross-over activation of PKA) may result in phosphorylation of Ser-333. Application of 8-Br-cAMP to cells with the mutant mTREK-1 channels marked and sustained suppression of currents. Thus, the major regulatory effects of cGMP-dependent and cAMP-dependent mechanisms occur via different sites of phosphorylation.

The studies described above suggest that TREK-1 encodes stretch-dependent K^+ channels in GI muscles and these channels are potently activated by nitrergic stimulation. We have also found that interstitial cells of Cajal (S. D. Koh, unpublished observations) in the gastric fundus and proximal colon also express SDK channels. Thus, these channels may mediate a portion of the enteric inhibitory response to nitrergic nerve stimulation in GI muscles. An obstacle in determining the physiological importance of 2-pore-domain K^+ channels in native cells is the poor specificity of channel blocking substances for these conductances. Several years ago we noted that some amino acids are capable of blocking hyperpolarization responses to NO in GI muscles. Thus, we conducted a survey of several amino acids and found that sulphur-containing amino acids, such as L-cysteine, L-methionine or DL-homocysteine, can block TREK-1 channels and SDK channels in colonic myocytes (Park *et al.* 2005). Among this series L-methionine was the most selective blocking drug, and it had little or no effect on the other K^+ conductances expressed in colonic myocytes. The sulphur-containing amino acids depolarized colonic muscles and inhibited nitrergic inhibitory junction potentials. These data show the importance of SDK (i.e. TREK-1) channels in nitrergic responses in GI muscles.

It is interesting to note that many regions of the GI tract, in spite of common expression of TREK channels, do not respond to stretch in a similar manner. For example, stretch of jejunal muscles resulted in net depolarization (S. D. Koh, unpublished observations). Segmentation is the most common motility pattern in the small intestine, and this is characterized by localized contraction of one region, movement of contents and distension of adjacent segments, and then contraction of the distended segments. This contraction–distension cycle repeats itself over and over during the intestinal phase of digestion. Since the distended segment must contract soon after being distended, a dominant SDK conductance might be counterproductive. The dominant mechanosensitive conductance in small intestinal muscles may be a stretch-dependent non-selective cation conductance. It is likely that the compliment of mechanosensitive channels in GI muscles is tuned for the specific functional needs of the region or organ. The integrated responses of GI muscles will be a function of whether there is net inward or outward current activated by stretch.

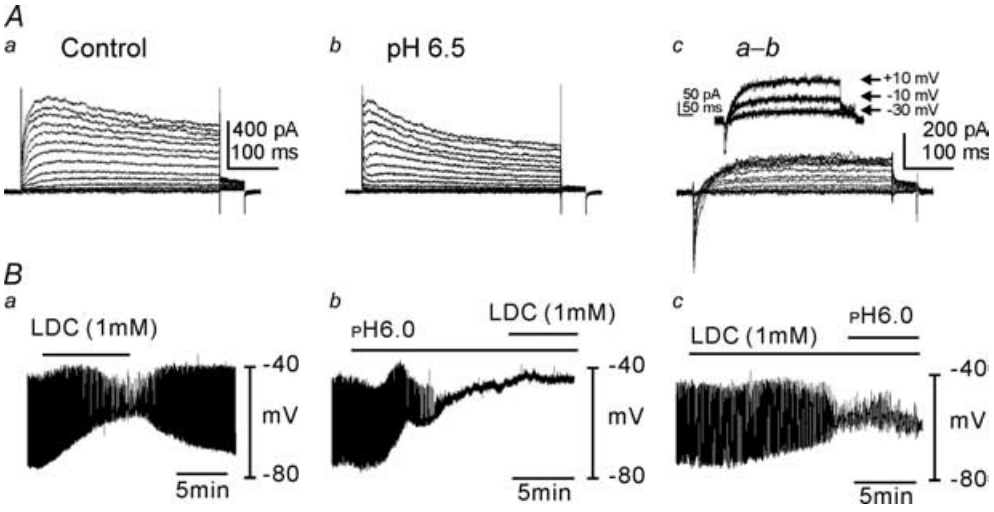

Figure 4. Effects of extracellular acidic pH on outward currents in the presence of 4-aminopyridine and TEA in colonic myocytes (*A*) and effects of lidocaine (LDC) on membrane potential of murine ileum (*B*)
Aa, control traces from colonic myocytes are shown in the presence of 4-aminopyridine (5 mM) and TEA (10 mM). *Ab*, exposure to pH 6.5 solution decreased outward current. *Ac*, difference currents obtained by subtracting currents obtained at pH 6.5 (*Ab*) from control currents (*Aa*). Inset shows exponential fits (continuous lines) of outward currents generated by steps to −10, 0 and +10 mV to obtain time constants of activation. *Ba,* lidocaine (10^{-4} M) depolarized murine ileal tissue. *Bb*, exposure of ileal muscles to pH 6.0 caused depolarization. Application of lidocaine (1 mM) after pH 6.0 solution did not induce further depolarization. *Bc*, in reverse order, lidocaine caused depolarization, but addition of acidic pH (6.0) in the continued presence of lidocaine had no further effect on membrane potential.

TASK-like currents in GI muscles

The mechanism for the dominant permeability of GI muscle cell membranes to K^+ ions, and thus the negative resting potentials of these cells, remains unclear. Expression of a variety of K^+ channel genes, including various combinations of K_V 1.2, 1.5 and 2.2, K_v11.1, K_{Ca} (BK, IK and SK) and K_{IR} 2, 3 and 6 have been reported over the past decade (see Sanders *et al.* 2006 for review). However, blockers of all of these channels only yield partial depolarization of GI smooth muscle cells. Thus, a significant background conductance is likely to exist to explain the relatively high permeability to K^+.

We have recently considered the possibility that a pH-sensitive K^+ conductance might contribute to the resting membrane potential in GI muscles. Acidification of the solution bathing gastrointestinal muscles produces significant depolarization, and hyperpolarization results with alkaline pH (Cho *et al.* 2005). Bipolar regulation by pH is a unique property of TASK-1 and TASK-2 among known ion channels. These channels can be regulated by either acidic or alkaline changes from physiological pH (Duprat *et al.* 1997; Lesage & Lazdunski, 2000). TASK channels are 'open-rectifier' background K^+ conductances with current–voltage properties predicted by the Goldman-Hodgkin-Katz (GHK) current equation (Lesage, 2003). We have investigated the expression and function of such a conductance in GI muscle cells.

RT-PCR using specific primer sets showed that TASK-1 and TASK-2 mRNAs are expressed in muscles and smooth muscle cells of the small and large intestines of mice. TASK-2 was also identified in smooth muscle cells of both the circular and longitudinal muscle layers by immunohistochemical techniques. Antibodies for TASK-1 were not of sufficient quality to provide specific resolution of this protein. TASK-2 was cloned from murine colon, and the channels were expressed in *Xenopus* oocytes. Expression of TASK-2 yielded a rapidly activating, non-inactivating K^+ current that was inhibited by acidification (i.e. pH 7.0). Native murine colonic and ileal smooth muscle cells expressed a similar current (Cho *et al.* 2005). Under conditions that blocked known voltage- and Ca^{2+}-dependent conductances, a substantial outward current was observed under whole-cell voltage clamp. This current was blocked by reducing pH to 6.5 (Fig. 4*A*) and by lidocaine (lignocaine). Pretreatment with lidocaine blocked the pH-sensitive current. We also tested the effects of local anaesthetics and reduced pH on intact intestinal muscles. Lidocaine and bupivacaine depolarized muscles and reduced slow wave amplitude. The effects of lidocaine were not blocked by pretreatment with TEA (10 mM), 4-aminopyridine (5 mM), glibenclamide (10^{-5} M), apamin (300 nM), or MK499 (a blocker of KCNH2 channels). But the depolarization effects of reduced pH were blocked by pretreatment with lidocaine (Fig. 4*B*). These experiments demonstrated the presence of TASK-1 and -2 channels in intestinal smooth muscles and documented a functional conductance in myocytes and intact muscles with properties similar to cloned TASK channels. These data suggest that TASK family channels may contribute significantly to the relatively high background K^+ permeability of GI smooth muscle cells.

Conclusions

Our data support the conclusion that 2-pore-domain K^+ channels play important roles in regulating responses of GI smooth muscles. All neural, hormonal, and paracrine regulation of smooth muscle contractions is superimposed upon the basal excitability state of smooth muscle cells. TREK and TASK channels appear to participate in establishing the moment-to-moment state of excitability in GI muscles. Additional regulation of TREK channels appears to be important in tuning responses of GI muscles to stretch and enteric inhibitory nerve stimulation. These channels contribute to receptive relaxation in GI muscles. There may also be contributions from other 2-pore-domain K^+ channels that are as yet unrecognized in GI muscles, and little is known about the function of these channels in the smooth muscles of other hollow organs of the body. Further investigation will reveal the contributions of these channels to the integrated responses of smooth muscles.

References

Backx PH & Marban E (1993). Background potassium current active during the plateau of the action potential in guinea pig ventricular myocytes. *Circ Res* **72**, 890–900.

Bai X, Bugg GJ, Greenwood SL, Glazier JD, Sibley CP, Baker PN, Taggart MJ & Fyfe GK (2005). Expression of TASK and TREK, two-pore domain K^+ channels, in human myometrium. *Reproduction* **129**, 525–530.

Bang H, Kim Y & Kim D (2000). TREK-2, a new member of the mechanosensitive tandem-pore K^+ channel family. *J Biol Chem* **275**, 17412–17419.

Buckingham SD, Kidd JF, Law RJ, Franks CJ & Sattelle DB (2005). Structure and function of two-pore-domain K^+ channels: contributions from genetic model organisms. *Trends Pharmacol Sci* **26**, 361–367.

Buckler KJ & Honore E (2005). The lipid-activated two-pore domain K^+ channel TREK-1 is resistant to hypoxia: implication for ischaemic neuroprotection. *J Physiol* **562**, 213–222.

Bulbring E (1954). Smooth muscle potentials recorded in the taenia coli of the guinea pig. *J Physiol* **123**, 55*P*–56*P*.

Chemin J, Patel AJ, Duprat F, Lauritzen I, Lazdunski M & Honore E (2005). A phospholipid sensor controls mechanogating of the K^+ channel TREK-1. *EMBO J* **24**, 44–53.

Cho SY, Beckett EA, Baker SA, Han I, Park KJ, Monaghan K, Ward SM, Sanders KM & Koh SD (2005). A pH-sensitive potassium conductance (TASK) and its function in the murine gastrointestinal tract. *J Physiol* **565**, 243–259.

Duprat F, Lesage F, Fink M, Reyes R, Heurteaux C & Lazdunski M (1997). TASK, a human background K^+ channel to sense external pH variations near physiological pH. *EMBO J* **16**, 5464–5471.

Franks NP & Honore E (2004). The TREK K2P channels and their role in general anaesthesia and neuroprotection. *Trends Pharmacol Sci* **25**, 601–608.

Gurney AM, Osipenko ON, MacMillan D, McFarlane KM, Tate RJ & Kempsill FE (2003). Two-pore domain K channel, TASK-1, in pulmonary artery smooth muscle cells. *Circ Res* **93**, 957–964.

Han J, Gnatenco C, Sladek CD & Kim D (2003). Background and tandem-pore potassium channels in magnocellular neurosecretory cells of the rat supraoptic nucleus. *J Physiol* **546**, 625–639.

Han J, Truell J, Gnatenco C & Kim D (2002). Characterization of four types of background potassium channels in rat cerebellar granule neurons. *J Physiol* **542**, 431–444.

Hisada T, Walsh JV Jr & Singer JJ (1993). Stretch-inactivated cationic channels in single smooth muscle cells. *Pflugers Arch* **422**, 393–396.

Kim D (2003). Fatty acid-sensitive two-pore domain K^+ channels. *Trends Pharmacol Sci* **24**, 648–654.

Kim D, Fujita A, Horio Y & Kurachi Y (1998). Cloning and functional expression of a novel cardiac two-pore background K^+ channel (cTBAK-1). *Circ Res* **82**, 513–518.

Kirber MT, Walsh JV Jr & Singer JJ (1988). Stretch-activated ion channels in smooth muscle: a mechanism for the initiation of stretch-induced contraction. *Pflugers Arch* **412**, 339–345.

Koh SD, Campbell JD, Carl A & Sanders KM (1995). Nitric oxide activates multiple potassium channels in canine colonic smooth muscle. *J Physiol* **489**, 735–743.

Koh SD, Monaghan K, Sergeant GP, Ro S, Walker RL, Sanders KM & Horowitz B (2001). TREK-1 regulation by nitric oxide and cGMP-dependent protein kinase. An essential role in smooth muscle inhibitory neurotransmission. *J Biol Chem* **276**, 44338–44346.

Koh SD & Sanders KM (2001). Stretch-dependent potassium channels in murine colonic smooth muscle cells. *J Physiol* **533**, 155–163.

Lauritzen I, Zanzouri M, Honore E, Duprat F, Ehrengruber MU, Lazdunski M & Patel AJ (2003). K^+-dependent cerebellar granule neuron apoptosis. Role of task leak K^+ channels. *J Biol Chem* **278**, 32068–32076.

Lesage F (2003). Pharmacology of neuronal background potassium channels. *Neuropharmacology* **44**, 1–7.

Lesage F & Lazdunski M (2000). Molecular and functional properties of two-pore-domain potassium channels. *Am J Physiol Renal Physiol* **279**, F793–F801.

Liu W & Saint DA (2004). Heterogeneous expression of tandem-pore K^+ channel genes in adult and embryonic rat heart quantified by real-time polymerase chain reaction. *Clin Exp Pharmacol Physiol* **31**, 174–178.

Meuthw SG, Budde T, Kanyshkova T, Broicher T, Munsch T & Pape HC (2003). Contribution of TWIK-related acid-sensitive K^+ channel 1 (TASK1) and TASK3 channels to the control of activity modes in thalamocortical neurons. *J Neurosci* **23**, 6460–6469.

Millar JA, Barratt L, Southan AP, Page KM, Fyffe RE, Robertson B & Mathie A (2000). A functional role for the two-pore domain potassium channel TASK-1 in cerebellar granule neurons. *Proc Natl Acad Sci U S A* **97**, 3614–3618.

Park KJ, Baker SA, Cho SY, Sanders KM & Koh SD (2005). Sulfur-containing amino acids block stretch-dependent K^+ channels and nitrergic responses in the murine colon. *Br J Pharmacol* **144**, 1126–1137.

Patel AJ & Honore E (2001). Properties and modulation of mammalian 2P domain K^+ channels. *Trends Neurosci* **24**, 339–346.

Patel AJ, Honore E, Maingret F, Lesage F, Fink M, Duprat F & Lazdunski M (1998). A mammalian two pore domain mechano-gated S-like K^+ channel. *EMBO J* **17**, 4283–4290.

Sanders KM, Koh SD & Ward SM (2006). Organization and electrophysiology of interstitial cells of Cajal and smooth muscle cells in the gastrointestinal tract. In *Physiology of the Gastrointestinal Tract*, 4th edn, ed. Johnson LR. Elsevier Press, San Diego (in press).

Terrenoire C, Lauritzen I, Lesage F, Romey G & Lazdunski M (2001). A TREK-1-like potassium channel in atrial cells inhibited by beta-adrenergic stimulation and activated by volatile anesthetics. *Circ Res* **89**, 336–342.

Yamamoto Y, Kummer W, Atoji Y & Suzuki Y (2002). TASK-1, TASK-2, TASK-3 and TRAAK immunoreactivities in the rat carotid body. *Brain Res* **950**, 304–307.