Cloning and Expression of a Novel pH-sensitive Two Pore Domain K\(^+\) Channel from Human Kidney*

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A complementary DNA encoding a novel K\(^+\) channel, called TASK-2, was isolated from human kidney and its gene was mapped to chromosome 6p21. TASK-2 has a low sequence similarity to other two pore domain K\(^+\) channels, such as TWIK-1, TREK-1, TASK-1, and TRAAK (18–22% of amino acid identity), but a similar topology consisting of four potential membrane-spanning domains. In transfected cells, TASK-2 produces nonactivating, outwardly rectifying K\(^+\) currents with activation potential thresholds that closely follow the K\(^+\) equilibrium potential. As for the related TASK-1 and TRAAK channels, the outward rectification is lost at high external K\(^+\) concentration. The conductance of TASK-2 was estimated to be 14.5 pS in physiological conditions and 59.9 pS in symmetrical conditions with 155 mM K\(^+\). TASK-2 currents are blocked by quinine (IC\(_{50}\) = 22 \(\mu\)M) and quinidine (65% of inhibition at 100 \(\mu\)M) but not by the other classical K\(^+\) channel blockers tetraethylammonium, 4-aminopyridine, and Cs\(^+\). They are only slightly sensitive to Ba\(^2+\), with less than 17% of inhibition at 1 mM. As TASK-1, TASK-2 is highly sensitive to external pH in the physiological range. 10% of the maximum current was recorded at pH 6.5 and 90% at pH 8.8. Unlike all other cloned channels with two pore-forming domains, TASK-2 is essentially absent in the brain. In human and mouse, TASK-2 is mainly expressed in the kidney, where *in situ* hybridization shows that it is localized in cortical distal tubules and collecting ducts. This localization, as well as its functional properties, suggest that TASK-2 could play an important role in renal K\(^+\) transport.

Potassium channels are present in virtually all living cells. They conduct the flux of potassium ions through the membrane, and in doing so, they are involved in the control of numerous cellular functions, such as neuronal firing, muscle contraction, volume regulation, and hormone secretion (1, 2). In the kidney, they are more particularly involved in the K\(^+\) secretion that is fundamental for K\(^+\) homeostasis (3).

Recently, K\(^+\) channels with unusual structures have been identified. The yeast channel subunit TOK/YKC/YORK/DUK contains two pore-forming (P) domains and eight TMSs \(^1\) in a single subunit (4–7), whereas TWIK-1, the founding member of a new mammalian class, has two P domains and four TMSs (8, 9). The TWIK-related channels TREK-1, TASK (also called cTRAAK), and TRAAK exhibit the same overall structure despite their low similarity in amino acid sequence (10–14). Structurally related channels have also been identified in *Drosophila*, *Caenorhabditis elegans*, and plants (15–17).

In mammals, these 2P domain channels are extraordinarily diverse in terms of both distribution and functional properties. TWIK-1, TREK-1, and TASK are expressed in many tissues with specific patterns, whereas TRAAK is only expressed in neuronal cells (18). From a functional point of view, TWIK-1 expressed baseline weakly inward-rectifying currents that are stimulated by protein kinase C and inhibited by internal acidification (8). TREK-1 produces arachidonic acid-activated mechano-sensitive outwardly rectifying currents that are inhibited by both protein kinase A and protein kinase C (10, 19). On the other hand, TASK and TRAAK currents behave like K\(^+\)-selective “holes,” with no rectification other than that predicted from the constant-field assumptions for an open channel (11–14). Despite this common property, TASK and TRAAK exhibit very different modulations of their activity. TASK is regulated by external pH variations and is inhibited by a small drop of the external pH near the physiological range (11, 13, 14). TRAAK is stimulated by arachidonic acid, as well as other unsaturated fatty acids (12). Despite their different functional properties, all of these 2P domain K\(^+\) channels express quasi-instantaneous and nonactivating currents that do not display voltage-dependent activation thresholds. They are open at the resting potential, and their expression is associated with a strong membrane polarization. All of these properties suggest that the 2P domain K\(^+\) channels are involved in the generation and the modulation of the resting potential of many cell types (18). This paper describes the cloning, the gene localization, the tissue distribution, and the functional characterization of a novel member of this emerging family.

**EXPERIMENTAL PROCEDURES**

Cloning of TASK-2—The sequences of mammalian two P domain K\(^+\) channels were used to search homologs in public DNA data bases by using the TBLASTn alignment program (20) and led to the identification of an expressed sequence tag (GenBank accession number H01932). The 5‘ end of this expressed sequence tag encoded a domain similar to the MIP1 extracellular loop of TWIK-1, but its 3‘-extremity did not encode the P1 and M2 domains of a two P domain channels as expected. We postulated that this expressed sequence tag was a tandem cDNA or was issued from an unspliced mRNA, and we used only the 5‘ part of the

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\(^1\) The abbreviations used are: TMS, transmembrane segments; 2P, two pore-forming; PCR, polymerase chain reaction; RT-PCR, reverse transcription-PCR; pBS, pBluescriptII SK−; pS, pCisociences.

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sequence to design oligonucleotides. These oligonucleotides were further used to carry out 3'-rapid amplification of cDNA ends polymerase chain reaction (PCR) experiments on human brain cDNA by standard methods. A DNA fragment was obtained that extended the region homologous to TWIK-1 and was previously identified in H01927. To test novel sequences deduced from this DNA fragment, a 1.2-kilobase (pBS) and analyzed by restriction analysis and by sequencing of their extremities. The longer cDNA insert (pBS-TASK-2) was completely amplified on both strands by using the dideoxy nucleotide termination method using an automatic sequencer (Applied Biosystems, model 373A).

Analysis of TASK-2 mRNA Distribution—For Northern blot analysis, human multiple tissue Northern blots were purchased from CloneTech and hybridized at 65 °C in ExpressHyb solution with 0.6- and 1.2-kilobase Smal I 32P-labeled fragments from pBS-TASK-2 following the manufacturer’s protocol. For RT-PCR experiments, total RNAs were extracted from adult mouse tissues and from mouse embryos with the SNAP total RNA isolation kit (Invitrogen). After a DNase treatment, 15 μg of reverse-transcribed RNA was used as template for PCR amplification (Life Technologies, Inc.). 1/40 of each sample was used as template for PCR amplification (Tag DNA polymerase, Life Technologies, Inc.) by using pBS-TASK-2 (base positions 358–381 (5'-CTGCTACCTGCCTCATCCTC-3') and 901–924 (5'-GTAGAGGCCCTCCTGATGGTATGATCTC-3') and GAPDH (CloneTech) primers. PCR conditions were 30 cycles of 30 s at 94 °C, 30 s at 60 °C, and 30 s at 72 °C. PCR-amplified fragments were transferred onto nylon membranes and then probed at high stringency with a 32P-labeled Smal I DNA fragment of pBS-TASK-2 (nucleotides 116–1305) (21) on 7-μm paraffin sections of human kidneys fixed in 4% paraformaldehyde. A specific antisense cRNA probe was generated with T7 RNA polymerase (Promega) by in vitro transcription using (32P)-α-UTP from a EcoRI-linearized plasmid containing a 357-base pair NotI/plasmid fragment of the 5'-untranslated sequence of TASK-2 cDNA inserted into pBS. The same plasmid was linearized by XhoI, and T7 RNA polymerase was used for the synthesis of a control sense probe. The probes were hybridized, and then slides were covered with NTB2 emulsion (Kodak) and exposed for 32 days at 20 °C. After development, slides were stained with toluidine blue and photographed. The fragments of human kidney were obtained from surgical ablation of renal cancers (pieces of kidney. The distribution of TASK-2 was analyzed by Northern blot (Fig. 2I) and by RT-PCR from mouse tissues (Fig. 2B). The mouse TASK-2 message was found in the kidney and is present to a lesser extent in the pancreas, liver, the placenta, and the small intestine. The expression of TASK-2 was also analyzed by RT-PCR from mouse tissues (Fig. 2B).

Cloning and Primary Structure of TASK-2—An expressed sequence tag was identified by homology screening with the two P domain K+ channels using the tBLASTn alignment algorithm (20). This sequence was amplified by PCR and used to screen a human kidney cDNA library. A full-length cDNA of 3.5 kilobases was isolated. It contains an extended open reading frame that codes for a polypeptide of 499 residues with a calculated molecular mass of 55.1 kDa. The predicted product displays all the hallmarks of the 2P domain K+ channels (Fig. 1A). Analysis of its hydrophy profile indicates the presence of four TMSSs, designated M1 to M4; the M1 and M2 segments flank the first P domain (P1), and the M3 and M4 flank a second P domain (P2). An extended M1P1 interdomain that is characteristic of this channel family is also found that is expected to be extracellular, as for TWIK-1. This region contains a potential N-linked glycosylation site and a cysteine residue (position 51) that are conserved in TWIK-1, TREK-1, and TRAAK. In TWIK-1, this cysteine residue has been shown to be implicated in the formation of an interchain disulfide bond (25). Despite this overall structural conservation, the novel subunit is only distantly related to the other cloned 2P domain K+ channels (between 18 and 22% of amino acid identity). The dendrogram shown in Fig. 1B also suggests that the novel subunit is more related to TASK than to TWIK-1, TREK-1, or TRAAK. However, it was called TASK-2 to emphasize the fact that it produces K+ currents that are acid-sensitive, like TASK-1, as shown below. For this reason, TASK is now called TASK-1.

Tissue Distribution of TASK-2—The tissue distribution of TASK-2 in adult human was analyzed by Northern blot (Fig. 2A). A 4-kilobase transcript is abundantly expressed in the kidney and is present to a lesser extent in the pancreas, liver, the placenta, and the small intestine. The expression of TASK-2 was also analyzed by RT-PCR from mouse tissues (Fig. 2B). The mouse TASK-2 message was found in the kidney, liver, and the small intestine, in the same relative abundance as in human. As expected, the RT-PCR method is more sensitive than the Northern blot technique, and faint positive signals were also obtained in mouse brain, heart, skeletal muscle and colon. Surprisingly, TASK-2 expression levels in uterus, lung and pancreas are different between human and mouse.

Electrophysiological Measurements in Xenopus Oocytes—The sequence coding for TASK-2 was amplified by PCR using a low error rate polymerase (PWO pol, Boehringer Mannheim) and subcloned into the pEXO vector (22) to give pEXO-TASK. Capped cRNA was synthesized in vitro from the linearized plasmid by using the T7 RNA polymerase (Stratagene). Xenopus laevis oocytes were purchased from CRBM (Montpellier, France). Preparation and cRNA injection of oocytes has been described elsewhere (23). Oocytes were used for electrophysiological studies 2–4 days following injection (20 ng/oocyte). In a 0.3-mM perfusion chamber, a single oocyte was impaled with two standard microelectrodes (1.2-5 MΩ resistance) filled with 3 M KCl and maintained in the holding and superfusing solution by using a micromanipulator, in standard ND96 solution (96 mM NaCl, 2.7 mM KCl, 1.8 mM CaCl2, 2 mM MgCl2, 5 mM HEPES, pH 7.4, with NaOH). Stimulation of the preparation, data acquisition, and analysis were performed using pClamp software (Axon Instruments). Drugs were applied externally by addition to the superfusate (flow rate, 3 μl/min). All experiments were performed at room temperature (21–22 °C).

Patch-Clamp Recordings in Transfected COS Cells—A Not/I EcoRI fragment of 3.2 kilobases was excised from pBS-TASK-2 and subcloned into the pIREs-CD8 vector to give pIREs-CD8-TASK-2. The pIREs-CD8 vector was obtained by replacing the neo gene in the original vector pIREsneo (CLONTECH) by the coding sequence of the marker gene of the T vector lymphoma KOH cells were seeded at a density of 20,000 cells per 35-mm dish, 24 h prior to transfection. Cells were then transiently transfected by the classical DEAE-dextran method with 1 μg of pIREs-CD8-TASK-2 plasmid per 35-mm dish. Transfected cells were visualized 48 h after transfection using the anti-CD8 antibody-coated beads method (24). For whole cell recordings, the internal solution contained 150 mM KCl, 3 mM MgCl2, 5 mM EGTA, and 10 mM HEPES pH 7.4 with NaOH.

RESULTS

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RESULTS
was observed at higher resolution after in situ hybridization. Fig. 3 shows that the expression of TASK-2 is restricted to the distal tubules and the collecting ducts. No specific signal was observed in the proximal tubules or over the glomeruli.

Chromosomal Mapping of TASK-2—The chromosomal assignment of human TASK-2 was carried out by radiation hybrid panel analysis. As shown in Fig. 4, the gene encoding TASK-2 lies on chromosome 6p and is 5.45 cR centromeric to the framework marker WI-4142 (LOD score of 21). Although radiation hybrid maps are not anchored to the cytogenic maps, the most likely localization of the TASK-2 gene is 6p21.31–p21.33.

TWIK-1 has been previously mapped to chromosome 1q42–1q43 (26), and TREK and TASK have been mapped to chromosomes 1q41 and 2p23, respectively (27).

Biophysical and Pharmacological Properties of TASK-2—TASK-2-transfected COS cells display noninactivating currents (Fig. 5A) that are not present in control cells (not shown). The activation kinetics of TASK-2 currents are rapid. They are fitted with a single exponential characterized by time constants of 60.9 ± 150 mV and 62.6 ± 62 mV (n = 8). The current-voltage (I-V) relationship is outwardly rectifying, and almost no inward currents were recorded in an external me
medium containing 5 mM K\( ^+ \) (Fig. 5B). When cells were perfused with a K\( ^+ \)-rich solution (155 mM), the TASK-2 currents presented an almost linear I-V relationship, and the inward currents recorded at very negative potentials were noisy (Fig. 5, A and B). The relationship between the reversal potential and [K\( ^+ \)]\(_o\) is close to the predicted Nernst value (58.9 ± 4.5 mV/decade, \( n = 4 \)), as expected for a highly selective K\( ^+ \) channel (Fig. 5C). TASK-2 was also expressed in *Xenopus* oocytes, where it shows similar properties (Fig. 5D). Activation kinetics of the TASK currents in oocytes are slightly slower, with time constants of 112.6 ± 16.8 ms at \(-50 \) mV and 102.9 ± 7.3 ms at 0 mV (\( n = 6 \)). As previously shown with TWIK-1, TREK-1, TRAAK, and TASK-1 (8, 10–12), the membrane potential of oocytes expressing TASK-2 is strongly polarized (−78.6 ± 2.7 mV, \( n = 9 \)) compared with control oocytes (−42.2 ± 3.1 mV, \( n = 6 \)).

Single channel TASK-2 currents were recorded in outside-out patches from transfected COS cells. They are very flickery and show substates (Fig. 5E). In 5 mM external K\( ^+ \), the single channel I-V relationship is almost linear between –80 and 140 mV and presents a saturation at potentials more positive than +40 mV (Fig. 5F). The slope conductance measured between −60 mV and +20 mV is 14.5 ± 1.4 pS (\( n = 22 \)). In 155 mM external K\( ^+ \), the slope conductance is greatly increased (59.9 ± 3.1 pS, \( n = 19 \)) and saturates both at very negative and positive potentials.

The effects of various pharmacological agents on currents elicited by voltage pulses to +50 mV have been studied in TASK-2-expressing COS cells. The “classical” K\( ^+ \) channels blockers tетraethylammonium (1 mM), 4-aminopyridine (100 μM), and Cs\( ^+ \) (1 mM) were inactive on the recorded currents. Ba\(^{2+}\) only slightly diminished the current at +50 mV (16.9 ± 1.6% at 1 mM, \( n = 3 \)). Quinine induced a dose-dependent inhibition of TASK-2 currents characterized by an IC\(_{50}\) of 22.4 ± 1.8 μM (\( n = 9 \)) (not shown), whereas 100 μM quinidine induced a 65 ± 3.8% inhibition of the current (\( n = 4 \)). A strong effect was observed with lidocaine (1 mM) and bupivacaine (1 mM) with inhibitions of 60.4 ± 1.5% and 80.9 ± 4.5%, respectively (\( n = 4 \)). Zinc (100 μM) was also tested and induced a slight decrease of the current (15.3 ± 2.2%, \( n = 5 \)). Unlike TRAAK (12), TASK-2 is not sensitive to arachidonic acid (10 μM).

Regulation of TASK-2 Activity by External pH—TASK-2 currents were insensitive to the activation of adenyl cyclase obtained by increasing intracellular cAMP with a mixture of isobutylmethylxanthine (1 mM) and forskolin (10 μM) or by perfusion of the permeant 8-chloro-cAMP (500 μM), or to the
activation of protein kinase C obtained by an application of the phorbol ester phorbol 12-myristate 13-acetate (70 nM).

Interestingly, TASK-2 currents are highly sensitive to external pH, like TASK-1 (11). The I-V relationships recorded as in A, with voltage ramps ranging from −150 to +50 mV, 500 ms in duration. C, relationship between the reversal potential measured in COS cells and the external K⁺ concentration; data (mean ± S.E., n = 4) are shown with the linear regression (line). D, current-voltage relationship recorded in a TASK-2-expressing oocyte in 2 mM external K⁺, with voltage ramps ranging from −150 to +50 mV, 500 ms in duration. Inset, currents recorded in 2 mM external K⁺, during voltage pulses ranging from −150 to +50 in 50 mV steps. The holding potential was −80 mV. E, single channel currents recorded in transfected COS cells in outside-out patch at various potentials ranging from −80 mV to +80 mV, 40-mV steps. The dotted lines indicate the zero current level. F, single channel current-potential relationships recorded as in E in 5 mM (n = 19) and 155 mM (n = 19) external K⁺.

Fig. 5. Expression of TASK-2 in COS cells and Xenopus oocytes. A, TASK-2 whole cell currents recorded in transfected COS cells in 5 or 155 mM external K⁺, during voltage pulses ranging from −150 to +50 in 50 mV steps. The holding potential was −80 mV; the dotted lines indicate the zero current level. B, current-voltage relationship recorded as in A, with voltage ramps ranging from −150 to +50 mV, 500 ms in duration. C, relationship between the reversal potential measured in COS cells and the external K⁺ concentration; data (mean ± S.E., n = 4) are shown with the linear regression (line). D, current-voltage relationship recorded in a TASK-2-expressing oocyte in 2 mM external K⁺, with voltage ramps ranging from −150 to +50 mV, 500 ms in duration. Inset, currents recorded in 2 mM external K⁺, during voltage pulses ranging from −150 to +50 in 50 mV steps. The holding potential was −80 mV. E, single channel currents recorded in transfected COS cells in outside-out patch at various potentials ranging from −80 mV to +80 mV, 40-mV steps. The dotted lines indicate the zero current level. F, single channel current-potential relationships recorded as in E in 5 mM (n = 22) and 155 mM (n = 19) external K⁺.

**DISCUSSION**

A New Member in the TWIK Family of K⁺ Channels—TASK-2 is a novel member of the emerging family of 2P domain


**Fig. 6. Sensitivity of TASK-2 currents to external pH.** A, current-voltage relationships deduced from currents elicited by voltage pulses ranging from −150 to +50 mV, in 50-mV steps, measured at three different external pH levels (6.0, 7.4, and 8.6). Steps lasted 500 ms and started from an holding potential of −80 mV. B, whole cell currents elicited in a A, C, relationship between the current measured at −50, 0, and +50 mV and the external pH. Data (mean ± S.E.) were fitted with a Boltzmann relation (pHm = 7.8 ± 0.1, n = 17, at +50 mV). D, effect of pH 6.5, 7.3, and 9.1 on single channel current recorded at 0 mV in the outside-out patch configuration. E, effect of pH 6.5, 7.3, and 9.1 on N.P., calculated from mean single channel currents recorded in outside-out patches at 0 mV during 30 s and from slope conductance between −20 and +20 mV (n = 5). F, effect of pH 6.5, 7.3, and 9.1 on single channel currents recorded at 0 mV (n = 13).

K⁺ channels. Its cloning extends to five the number of these channels identified to date in mammals. Despite an overall structural conservation, TASK-2 does not share more than 18–22% of amino acid identity with the four other cloned channels and does not seem to be more related to any one of them from a phylogenetic point of view.

Both *Shaker* and inwardly rectifying K⁺ channel families of K⁺ channel subunits comprise numerous members corresponding to different genes. Within each of these two superfamilies, different subclasses can be distinguished according to their sequence similarities. Moreover, sequence conservations within each family are associated with similar functional properties. For instance, within the inwardly rectifying K⁺ channel family, the Kir3.x subunits share 55–60% of amino acid identity (28). All these Kir3.x proteins form G-protein-activated K⁺ channels. Kir subunits belonging to the other subgroups are more distant from the point of view of sequences and form inward rectifier channels that are not activated by G proteins (29–31). On the other hand, the *Shaker*-related voltage-dependent Kv1.x K⁺ channels (32–34) or the Ca²⁺-dependent SK channels (35) form subsets of proteins sharing 70–85% of amino acid identity. However, despite a similar structure with six TMSs and one P domain, Kv1.x and SK channels have less than 20% of overall amino acid identity. The question then arises of whether equivalent structural and functional subfamilies can be distinguished in the 2P domain K⁺ channel family. With a low sequence conservation between the different channels cloned up till now (18–38% of identity), the usual criteria of sequence similarity cannot be used. Nevertheless, our work leads us to propose a functional classification. TWIK-1 forms a first functional group because it is the only one to express weakly inward rectifying currents. TREK-1 and TRAAK form a second group of channels that produce outwardly rectifying currents stimulated by arachidonic acid and polyunsaturated fatty acids (10, 12, 19). The fact that both channels share 38% of amino acid identity instead of the 18–22% of identity usually found between the 2P domain K⁺ channels could signify that they have evolved from a common ancestral gene. However, TREK-1 and TRAAK probably have different physiological significance, because they have quite different tissue distributions, as well as different electrophysiological and regulation properties. The quasi-ubiquitous TREK-1 channel is inhibited by cAMP, but the neuronal TRAAK channel is not. TRAAK loses its outward rectification in high external [K⁺], but TREK-1 does not. The last functional group of 2P domain K⁺ channels is composed of TASK-1 and TASK-2. Both channels produce open rectifier K⁺ currents that are inhibited by a drop of external pH in the physiological range (11, 13). Their pharmacological behaviors are also similar. TASK-2, like TASK-1, is relatively insensitive to classical K⁺ channel blockers such as Ba²⁺, Ca²⁺, tetraethylammonium, and 4-aminopyridine, and both TASK-1 and TASK-2 are blocked by the local anesthetics lidocaine and bupivacaine. Their sequences are only distantly related, and for this reason, it is extremely difficult to know whether they have evolved from a common gene coding for an ancestral pH-sensitive K⁺ channel. TASK-1 and TASK-2 have different tissue distributions. TASK-1 is widely expressed in excitable as well as nonexcitable tissues (11), whereas TASK-2 seems to be preferentially present in epithelia. They also show significant differences in terms of electrophysiological and regulation properties. Unlike TASK-1 activity, TASK-2 activity is not inhibited by variations of intracellular cAMP (13), and TASK-2 is the sole 2P domain K⁺ channel cloned to date that displays relatively slow activation kinetics (11, 13, 14).

**TASK-2, a Novel Renal K⁺ Channel**—As discussed previously, TASK-2 has a unique tissue distribution. The TASK-2 message is poorly expressed or absent in the nervous and muscular systems but is present in epithelial tissues, such as lung, colon, intestine, stomach, liver, and particularly in the kidney. In this organ, TASK-2 is more precisely located in the cortical distal tubules and collecting ducts. In these structures, K⁺‐selective currents are postulated to play a major role in the volume regulation and in the control of the negative potential of tubule cells, in the K⁺ recycling across the basolateral membranes in conjunction with the Na-K-ATPase, and in the K⁺ secretion into the tubular lumen in concert with Na⁺ influx throughamiloride-sensitive Na⁺ channels (for reviews, see Refs. 3 and 36). Principal cells of the collecting ducts express at least two types of apical K⁺ currents sharing common properties, such as inhibition by Ba²⁺ and ATP, as well as by internal acidification (3). The first one is a K⁺ channel with a large conductance and a low probability of opening that is activated by membrane depolarization and internal Ca²⁺ and that is inhibited by tetraethylammonium (37, 38). The second one is a small conductance (25 pS) K⁺ channel with a high Po and inward rectification and that is insensitive to tetraethylammonium (39, 40). A cloned K⁺ channel that has the same properties is ROMK2 (41). On the other hand, three other K⁺ currents have been described at the basolateral membrane of the collecting duct cells: a small conductance K⁺ channel (28 pS) up-regulated by protein kinase C, nitric oxide, and cGMP (36, 42, 43); an intermediate conductance (85 pS) K⁺ channel activated by protein kinase A and hyperpolarization (44); and a large conductance (147 pS) K⁺ channel (42). The principal
biophysical and pharmacological properties of TASK-2 do not fit those of these native K⁺ channels. A possibility would be that TASK-2 channels are present in kidney cells but have not yet been recorded, which would not be surprising because of the lack of a specific pharmacology. Another possibility would be that TASK-2 associates with yet unidentified pore-forming subunits or regulatory proteins to produce an active channel in native cells with properties different from those of the cloned channel, as has been observed for some other K⁺ channels (45–47). The inhibition of K⁺ channels by acidification is consistent with the effect of metabolic acidosis, which decreases secretion in distal tubules (36).

New insights into the mechanism of K⁺ secretion have been recently provided by the cloning of several renal K⁺ channels and by fine studies of their distribution and cellular localization. Work presented in this paper opens the possibility of progress in four different directions: (i) the search for a native renal K⁺ channel with the properties of TASK-2, (ii) the search for a potent pharmacology specific of the TASK-2 channel, (iii) the determination of the localization of this channel type, and (iv) the knockout of the TASK-2 gene in mice. On the other hand, genetic diseases associated with channelopathies are now discovered with an increasing frequency (48), and it might turn out that there are human diseases associated with kidney, pancreas, and/or liver dysfunctions corresponding to mutations in the TASK-2 gene.

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