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Molecular basis of potassium channels in pancreatic duct epithelial cells

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Abbreviations: AKAP, A-kinase anchoring protein; BK, voltage- and Ca++-dependent maxi-K; BxPC3, human pancreas adenocarcinoma cell line; Capan-1, human pancreas adenocarcinoma cell line; CFPA-C1, human cystic fibrosis pancreatic adenocarcinoma cell line; CFTR, cystic fibrosis transmembrane conductance regulator; DC-EBIO, 5,6-dichloro-1-ethyl-1,3-dihydro-2H-benimidazole-2-one; DHS-I, dehydrosoyasaponin I; DIDS, 4,4'-diisothiocyanatostilbene-2,2'-disulfonic acid; E-4031, N-[4-[1-3-(trifluoromethyl)phenyl]-3-[4-bromo-2-(1-hydroxy-5-chlorophenyl)urea; PD-118057, [2-(4-[2-(3,4-dichloro-phenyl)-ethyl]-phenylamino)-benzoic acid; PKA, cAMP-dependent protein kinase; PKC, protein kinase C; PMA, phorbol 12-myristate 13-acetate; Ro-20-1724, 4-(3-butoxy-4-methoxybenzyl)-[4-[1-[2-(6-methylpyridin-2-yl)ethyl]-piperidine-4-carbonyl]phenyl] methanesulfonamide; EAG, ether-à-go-go gene; 1-EBIO, 1-[(3-(trifluoromethyl)phenyl)-thiourea; NS1608, 3-nitro-1-3,5-bis-trifluoromethyl-phenyl]-3-[4-phenoxyphenyl] benzamide; IK, intermediate-conductance Ca++-activated K; I_k, short-circuit current; K_Na, two-pore domain K channels; K_v inward rectifier potassium channel; L-364,373, (3-R)-1,3-dihydro-5-(2-fluorophenyl)-3-(1H-indol-3-ylmethyl)-1-methyl-2H-1,4-benzodiazepin-2-one; LY97241, N-ethyl-N-[4-(4-nitrophenyl)butyl]heptan-1-amine; NS11021, 1-(3,5-bis-trifluoromethyl-phenyl)-3-[4-bromo-2-(1H-tetrazol-5-yl)-phenyl]-thiourea; NS1608, N-3-(trifluoromethyl)phenyl)-N’-2-hydroxy-5-cholephophenyl)urea; PD-118057, 2-[2-(3,4-dichloro-phenyl)-ethyl]-phenylamino)-benzoic acid; PKA, cAMP-dependent protein kinase; PKC, protein kinase C; PMA, phosphorol 12-myristate 13-acetate; Ro-20-1724, 4-(3-butoxy-4-methoxybenzyl)-2-imidazolidinone; Slack, sequence like a calcium-activated K channel; Slick, sequence like an intermediate conductance K channel; TALK, TWIK-related alkaline pH-activated K channel; TASK, TWIK–related acid-sensitive K channel; TRAM-34, 1-[(2-chlorophenyl)diphenylmethyl]-1H-pyrazole; TWIK, tandem of P-domains in a weak inward rectifying K’ channel; V_{1/2}, half-maximal voltage; XE991, 10,10-bis(4-pyridinylmethyl)-9(10H)-antracenone

Introduction

Potassium channels regulate excitability, epithelial ion transport, proliferation, and apoptosis. In pancreatic ducts, K’ channels hyperpolarize the membrane potential and provide the driving force for anion secretion. This review focuses on the molecular candidates of functional K’ channels in pancreatic duct cells, including KCNQ1 (K’1.1), KCNQ2 (K’1.2) and KCNQ3 (K’1.3). We will give an overview of K’ channels with respect to their electrophysiological and pharmacological characteristics and regulation, which we know from other cell types, preferably in epithelia, and, where known, their identification and functions in pancreatic ducts and in adenocarcinoma cells. We conclude by pointing out some outstanding questions and future directions in pancreatic K’ channel research with respect to the physiology of secretion and pancreatic pathologies, including pancreatitis, cystic fibrosis, and cancer, in which the dysregulation or altered expression of K’ channels may be of importance.

Potential and thereby regulate the excitability of neurons and myocytes and transport of ions and water in epithelia, such as the pancreas and salivary glands. Duct epithelial cells in the pancreas secrete a HCO_3^-rich pancreatic juice that neutralizes acid chyme in the duodenum. Secretin, acetylcholine, and ATP stimulate fluid secretion via signal transduction involving cAMP and Ca++-signaling pathways. The generally accepted model for HCO_3^- secretion involves Cl^-–HCO_3^- exchangers (SLC26A3 and SLC26A6) that operate in parallel with cAMP-activated Cl^- channels (CFTR) or Ca^{2+}-activated Cl^- channels (most likely TMEM16A) on the luminal membrane and Na^+-coupled transporters such Na^+-K^-Cl co-transporter (NKCC1), Na^+-HCO_3^- co-transporter (SLC4A4), and Na^+-H^+ exchanger (SLC9A1) and Na^-K^-pump on the basolateral membrane (Fig. 1). In addition, H^+-K^-pumps are expressed on the luminal and basolateral membranes of pancreatic ducts. K’ channels are clearly important for setting the resting membrane potential and providing the driving force for anion exit and fluid secretion in a stimulated epithelium. K’ channels may also provide the transport partners for H^+-K^-pumps. In addition, certain K’ channels could play an important role in pancreatic pathology, such as cystic fibrosis, pancreatitis, and pancreatic adenocarcinoma. Perhaps surprisingly, there are not so many K’ channels studies performed on pancreatic ducts.

Early electrophysiological studies using microelectrodes and patch-clamp methods indicated that pancreatic ducts expressed voltage- and Ca^{2+}-activated K’ channels, consistent with maxi-K’ channels (BK channels), intermediate-conductance Ca^{2+}-activated
K⁺ channels (IK channels), and pH/HCO₃⁻ sensitive K⁺ channels. Recent studies focusing on molecular candidates have shown that pancreatic ducts express the following channels that could be candidates for above functional channels: Kᵦᵥ3.1 channels coded by the KCNMA1 and KCNB1 genes (α – and β-subunits of the BK channel); the Kₑᵥ3.1 protein (IK channel); the KCNK5 gene (Kᵥ5.1); and they also express: KCNQ1 (Kᵥ7.1, KVLQT1), KCNH2 (Kᵥ11.1, HERG), KCNH5 (Kᵥ10.2, EAG2), KCNT1 (Kᵥ4.1, Slack), and KCNT2 (Kᵥ4.2, Slick), the functions of which remain unclear in duct cells.

It is not known whether many of these candidates are functional in pancreatic ducts or what is their localization and regulation. Therefore, their physiological and possibly pathophysiological functions have not to be confirmed. The aim of this review is to provide an overview of the above mentioned K⁺ channels with respect to their electrophysiological and pharmacological characteristics and functions, as we know from other cell types, preferably in epithelia, and, where known, their identification and functions in pancreatic ducts is given (Table 1). We also address some outstanding questions and future directions in pancreatic K⁺ channel research.

**KCNN4 (Kₑᵥ3.1, IK, SK4)**

Tissue expression

**KCNN4** coding for the Kₑᵥ3.1 protein was cloned from the placenta and pancreas. Functional expression of the **KCNN4** gene has been demonstrated in colonic crypts, salivary acini, and pancreatic ducts. Immunoreactivity of the Kₑᵥ3.1 protein has also been reported in the esophagus, stomach, small intestine, proximal colonic crypts, salivary glands, luminal membrane of lacrimal gland duct cells, and intercalated and intralobular ducts of the pancreas. Interestingly, Kₑᵥ3.1 channel immunoreactivity was shown to be localized in both the basolateral and luminal membranes in pancreatic ducts and monolayer of Capan-1, a human pancreas adenocarcinoma cell line, though its expression appeared to be stronger in the luminal membrane. Consistent with this finding, the short-circuit current (Iₛ) of the Capan-1 cell monolayer was enhanced by the Kₑᵥ3.1 channel activator DC-EBIO in luminal or basolateral bathing solution. Kₑᵥ3.1 could potentially be an important candidate for luminal K⁺ channels in pancreatic ducts. Importantly, equivalent circuit analysis revealed that luminal K⁺ conductance contributed to a minimum of 10% of the total K⁺ conductance in pancreatic duct cells. Moreover, stimulation of the rat pancreas with secretin caused a marked increase in K⁺ concentrations in the pancreatic juice, which was equal to twice that in the plasma, indicating that K⁺ was secreted. K⁺ efflux was also shown to be mediated via mucosal Kₑᵥ3.1 channels in other epithelia, such as the distal colon, and provided, in part, the driving force for agonist-induced anion secretion. Another example is salivary acini, in which both Kₑᵥ3.1 and Kᵥ3.1 were shown to be expressed on the apical membrane and contribute to optimal secretion. Furthermore, H⁺–K⁺–pumps were reported to be expressed on the luminal membranes of pancreatic ducts and their function, such as contributing to local epithelial protection, appeared to depend on the operation of K⁺ channels.

**Channel properties**

Patch-clamp studies using Xenopus oocytes and mammalian expression systems established the basic electrophysiological and pharmacological properties of Kₑᵥ3.1 channels. Single-channel openings were observed at both positive and negative potentials in both Kₑᵥ and Kᵥ channels. The single-channel current–voltage relationship showed weak inward rectification with conductance of 30–54 pS in heterologous expression systems. Interestingly, intermediate-conductance K⁺ channels exhibited a conductance of 80 pS in rat pancreatic duct cells. One explanation for this discrepancy is that unidentified auxiliary proteins for Kₑᵥ3.1 channels or additional KCNN4 genes may exist in rodent cells. Regarding pharmacology, Kₑᵥ3.1 currents were inhibited by charybdotoxin, clotrimazole, TRAM-34, and maurotoxin with Kₑᵥ values of 2–28 nM, 25–150 nM, 20 nM, and 1 nM, respectively. Kₑᵥ3.1 currents were also activated by 1-EBIO and DC-EBIO with Kₑᵥ values of 15–84 μM and 0.8 μM, respectively.

**Regulation**

Regarding regulation, it is well established that Kₑᵥ3.1 channels are activated by the Ca²⁺/calmodulin signaling pathway. For example, heterologously expressed Kₑᵥ3.1 channels were previously shown to be activated by submicromolar free Ca²⁺ concentrations with EC₅₀ values of 0.1–0.3 μM. There is also strong evidence to suggest that the Ca²⁺ sensitivity of Kₑᵥ3.1 channels is mediated by calmodulin and calmodulin...
kinase.\textsuperscript{18,29,34} In addition, ATP/UTP was shown to regulate \(K_{Ca}\) channels via purinergic receptors in pancreatic cell lines and rat pancreatic duct cells.\textsuperscript{10,12,24,35} Both P2Y\(_2\) and P2Y\(_4\) receptors upregulated \(K_{Ca}\) activity in the \textit{Xenopus} oocyte expression system.\textsuperscript{11} Importantly, luminal ATP/UTP, most likely delivered by secret- ing acini,\textsuperscript{36,37} was reported to stimulate ductal secretion.\textsuperscript{24,35,38-41} The physiological role of \(K_{Ca}\) channels in pancreatic secretion could be also investigated with respect to secretin, which acts predominantly via the cAMP/cAMP-dependent protein kinase (PKA) signaling pathway, however, until this becomes available, we need to resort to studies on other cell types. A membrane-associated PKA has been proposed to activate \(K_{Ca}\) channels

<table>
<thead>
<tr>
<th>Gene</th>
<th>Protein</th>
<th>Conductance (pS)</th>
<th>Blockers (K)</th>
<th>Activators (K)</th>
<th>Regulation</th>
</tr>
</thead>
<tbody>
<tr>
<td>(KCN4)</td>
<td>(K_{Ca}) 3.1</td>
<td>30–54\textsuperscript{15,28,29}</td>
<td>charybdotoxin (2–28 nM)\textsuperscript{15,28,29,31} clotrimazole (25–150 nM)\textsuperscript{15,28-31} TRAM-34 (20 nM)\textsuperscript{30} maurotoxin (1 nM)\textsuperscript{32}</td>
<td>1-EBIO (15–84 μM)\textsuperscript{28,29,31,33} DC-EBIO (0.8 μM)\textsuperscript{33}</td>
<td>(Ca^{2+})\textsuperscript{14,15,29,31} calmodulin\textsuperscript{18,29,34} PKA\textsuperscript{19,42,43} extracellular UTP\textsuperscript{11} cell swelling\textsuperscript{7,48}</td>
</tr>
<tr>
<td>(KCN1)</td>
<td>(K_{Ca}) 1.1</td>
<td>100–270\textsuperscript{90,56}</td>
<td>tetraethylammonium (0.14 mM)\textsuperscript{50} charybdotoxin (1–31 nM)\textsuperscript{57,58,62} iberiotoxin (1–9 nM)\textsuperscript{58,61,62} paxilline (2–9 nM)\textsuperscript{58-60}</td>
<td>NS1608 (2 μM)\textsuperscript{50} NS11021 (0.4 μM)\textsuperscript{53}</td>
<td>membrane potential\textsuperscript{97,105,64} (Ca^{2+})\textsuperscript{97,105,64} PKA\textsuperscript{7,56} extracellular UTP\textsuperscript{11}</td>
</tr>
<tr>
<td>(KCNQ1)</td>
<td>(K_{7}) 1</td>
<td>0.7–4\textsuperscript{80,81}</td>
<td>chromanol 293B (10–41 μM)\textsuperscript{68,82,86,87} azimilide (77 μM)\textsuperscript{86} XE991 (0.8 μM)\textsuperscript{88}</td>
<td>L-364,373\textsuperscript{90}</td>
<td>membrane potential\textsuperscript{78-81} cAMP\textsuperscript{97} cytosolic pH\textsuperscript{83}</td>
</tr>
<tr>
<td>(KCNQ1/) (KCNE1)</td>
<td>(K_{7}) 1/ (\text{minK})</td>
<td>4.5–16\textsuperscript{80,81}</td>
<td>chromanol 293B (3–10 μM)\textsuperscript{80,82,86,87} azimilide (5.6 μM)\textsuperscript{86} XE991 (11 μM)\textsuperscript{88} Mefloquine (0.9 μM)\textsuperscript{89}</td>
<td>DIDS\textsuperscript{96} mefenamic acid\textsuperscript{96}</td>
<td>membrane potential\textsuperscript{78-81} cAMP\textsuperscript{97} cytosolic pH\textsuperscript{83}</td>
</tr>
<tr>
<td>(KCNH2)</td>
<td>(K_{11}) 1.1</td>
<td>10–13\textsuperscript{102-104}</td>
<td>E-4031 (7–1250 nM)\textsuperscript{104,105,108-111} BeKm-1 (3–12 nM)\textsuperscript{106-110} ergotoxin (4.5–17 nM)\textsuperscript{107,109} LYS97241 (2.2–19 nM)\textsuperscript{111,112}</td>
<td>mallotoxin (0.5 μM)\textsuperscript{114} PD-118057 (3.1 μM)\textsuperscript{115} ICA-105574 (0.5 μM)\textsuperscript{108}</td>
<td>membrane potential\textsuperscript{116} PKA\textsuperscript{119,120}</td>
</tr>
<tr>
<td>(KCNH5)</td>
<td>(K_{10}) 2</td>
<td></td>
<td></td>
<td></td>
<td>membrane potential\textsuperscript{97} PKC\textsuperscript{97}</td>
</tr>
<tr>
<td>(KCNT1)</td>
<td>(K_{4}) 4.1</td>
<td>180\textsuperscript{122}</td>
<td>bepridil (1 μM)\textsuperscript{125} quinidine (90 μM)\textsuperscript{125}</td>
<td>bithionol (0.8 μM)\textsuperscript{125} niclosamide (2.9 μM)\textsuperscript{126} loxapine (4.4 μM)\textsuperscript{126} niflumic acid (2.7 mM)\textsuperscript{127}</td>
<td>(Ca^{2+})\textsuperscript{121,122} Na\textsuperscript{+}\textsuperscript{122,123} Cl\textsuperscript{−}\textsuperscript{123} PKC\textsuperscript{130}</td>
</tr>
<tr>
<td>(KCNT2)</td>
<td>(K_{4}) 4.2</td>
<td>140\textsuperscript{122}</td>
<td>quinidine\textsuperscript{122} isoflurane\textsuperscript{128}</td>
<td>meclofenamic acid (80 μM)\textsuperscript{127} flufenamic acid (1.1–1.4 mM)\textsuperscript{127,129} niflumic acid (2.1 mM)\textsuperscript{127,129}</td>
<td>membrane potential\textsuperscript{122} Na\textsuperscript{+}\textsuperscript{122,130} Cl\textsuperscript{−}\textsuperscript{122} intracellular ATP\textsuperscript{122} PKC\textsuperscript{130}</td>
</tr>
<tr>
<td>(KCNK5)</td>
<td>(K_{5}) 5.1</td>
<td>50–78\textsuperscript{133,136,137}</td>
<td>quinine (22 μM)\textsuperscript{131} clofilium (25 μM)\textsuperscript{138}</td>
<td>halothane, isoflu- rane, chloroform\textsuperscript{130}</td>
<td>extracellular pH\textsuperscript{133,138,140,141} PKC\textsuperscript{140} osmolality\textsuperscript{38}</td>
</tr>
</tbody>
</table>
in human erythrocytes, the T84 human colonic crypt cell line, and rat submandibular acinar cells. Interestingly, the PKA consensus phosphorylation site at serine 334 in \( K_{\text{Ca}} \)-1.1 channels was not involved in PKA-dependent activation. In contrast to these studies, heterologously expressed \( K_{\text{Ca}} \)-3.1 channels were not affected by PKA activators and/or inhibitors, or were inhibited by the catalytic subunit of PKA. Given these contradictory results, it is tempting to speculate that \( K_{\text{Ca}} \)-3.1 channels may be activated via the phosphorylation of a closely associated protein, the expression of which is tissue-specific. One candidate for this protein is A-kinase anchoring protein (AKAP), which is able to scaffold PKA and components of cAMP signaling pathways, including G protein-coupled receptors and ion channels.

In addition to transepithelial transport, \( K_{\text{Ca}} \)-3.1 channels were also shown to be stimulated by cell swelling, which triggered regulatory volume decreases. Notably, \( KCNN4 \) mRNA levels were upregulated in primary pancreatic tumors, and the growth of ductal adenocarcinoma cell lines in vitro was inhibited by blockers of \( K_{\text{Ca}} \)-3.1 channels, which indicated that these were correlated with the proliferation of pancreatic cancer.

### KCNMA1 (\( K_{\text{Ca}} \)-1.1, Slo1, \( \alpha \)-subunit of BK) and KCNMB (\( \beta \)-subunits)

**Tissue expression**

The \( KCNMA1 \) coding \( K_{\text{Ca}} \)-1.1 (Slo1) protein was cloned from brain and skeletal muscle. Functional expression of the \( KCNMA1 \) gene has been demonstrated in the colon, salivary acini, pancreatic acini, and pancreatic ducts. The \( K_{\text{Ca}} \)-1.1 protein is located in the luminal membrane of colonic epithelia, salivary acini and ducts, and pancreatic ducts. It is noteworthy that there was no labeling of the basolateral membrane of guinea-pig pancreatic duct cells, although the first recordings of maxi-\( K^{+} \) currents were made on the basolateral membrane of rat pancreatic ducts.

**Channel properties**

\( K_{\text{Ca}} \)-1.1 channels have the largest single-channel conductance of all \( K^{+} \) selective channels: 100–270 pS in symmetrical 150 mM KCl. Maxi-\( K^{+} \) currents in isolated rat pancreatic duct cells had a conductance of 170–180 pS. Regarding pharmacology, the \( \alpha \)-subunit of \( K_{\text{Ca}} \)-1.1 was inhibited by tetraethylammonium, charybdotoxin, iberiotoxin, and paxilline, which is able to scaffold PKA and components of cAMP signaling pathways, including G protein-coupled receptors and ion channels.

**Regulation**

Significant diversity has been reported in the functional characteristics of \( K_{\text{Ca}} \)-1.1 channels. It is well established that \( K_{\text{Ca}} \)-1.1 channels are activated by membrane depolarization alone, intracellular \( Ca^{2+} \) alone, or synergistically by depolarization and \( Ca^{2+} \). The single-channel open probability of \( K_{\text{Ca}} \)-1.1 channels markedly increased when the cytoplasmic face of a patch membrane was exposed to 10 \( \mu \)M \( Ca^{2+} \) and voltage was changed over a range of −60 to +80 mV. Under these conditions, the half-maximal voltage (\( V_{1/2} \)) was +23 mV in 10 \( \mu \)M \( Ca^{2+} \); however, these were unphysiological conditions for pancreatic ducts. Importantly, maxi-\( K^{+} \) channels on pancreatic duct cells were activated by much lower \( Ca^{2+} \) concentrations. For example, maxi-\( K^{+} \) channels exposed to 3 \( \mu \)M \( Ca^{2+} \) reached \( V_{1/2} \) at −4 mV. This difference indicated that the \( \beta \)-subunit exists in pancreatic duct cells. Maxi-\( K^{+} \) channels on \( Xeopbus \) oocytes that heterologously expressed both the \( \alpha \) – and \( \beta \)-subunits of \( K_{\text{Ca}} \)-1.1 proteins were about 10-fold more sensitive to activation by voltage and \( Ca^{2+} \) concentration than channels composed of the \( \alpha \)-subunit alone.

Indeed, \( KCNMB1 \) coding the \( \beta \) subunit was detected in isolated pancreatic ducts.

Interestingly, UTP was shown to inhibit \( K_{\text{Ca}} \)-1.1 channels via the P2Y, receptor, and appeared to lead to a decrease in secretion. The basolateral application of ATP/UTP inhibited \( K^{+} \) conductance in rat duct cells and secretion in guinea-pig ducts and human duct cell monolayers. These results collectively indicated that P2Y receptors on the basolateral membrane appeared to downregulate secretion via \( K_{\text{Ca}} \)-1.1 channels in the ductal system.

Regarding the cAMP/PKA signaling pathway, cAMP-dependent phosphorylation can also activate maxi-\( K^{+} \) channels on pancreatic duct cells. The functional response of \( K_{\text{Ca}} \)-1.1 channels to PKA phosphorylation depends on the splice-variant of the \( \alpha \)-subunit. For example, PKA was shown to activate the ZERO variant, whereas PKA inhibited the STREX variant. PKA activation of the ZERO variant requires a conserved C-terminal PKA site. Indeed, the ZERO splice variant has been shown to conduct adrenaline-induced \( K^{+} \) secretion in the distal colon.

### KCNQ1 (\( K_{\text{v}} \)-7.1, KVLQT1) and KCNE1 (minK)

**Tissue expression**

The \( KCNQ1 \) coding \( K_{\text{v}} \)-7.1 (KVLQT1) protein was cloned from the heart. Functional expression of the \( KCNQ1 \) gene has also been demonstrated in the kidney, stomach, small intestine, colon, and pancreatic acini, and pancreatic ducts. Immunoreactivity of the \( K_{\text{v}} \)-7.1 protein was reported in the parietal cells of the stomach, in the basolateral membrane of small intestinal and colonic crypt cells, and in acinar and duct cells of the pancreas. \( K_{\text{v}} \)-7.1 resides in the tubulovesicular and canalicular membranes of gastric parietal cells together with \( H^{+}–K^{+} \)-pumps and participates in gastric acid secretion. \( K_{\text{v}} \)-7.1 was localized in the luminal membrane of pancreatic duct cells, and may be involved in cell volume regulation during purinergic stimulation in epithelial transport, and/or may potentially be associated with \( H^{+}–K^{+} \)-pumps expressed by pancreatic ducts.
The K7.1 protein can assemble with the KCNE family of regulatory β-subunits to fulfill various physiological functions. For example, minK coded by the KCNE1 gene has been shown to modify K7.1 activity by increasing unitary conductance, slowing activation, causing a right shift in the voltage dependence of activation, and modulating pharmacology. Interestingly, it is worth noting that the acidification of cytosolic pH increased K7.1–minK, but decreased K7.1 currents, whereas alkalization decreased K7.1–

The expression and function of KCNE in duct cells has not yet been investigated.

Channel properties

K7.1 channels have very small conductance. Noise analysis revealed estimated single-channel conductances of 0.7–4 pS. Small conductance K+ channels had 1 pS and were inhibited by chromanol 293B, a K7.1 blocker, in the basolateral membrane of rat pancreatic acinar cells. Chromanol 293B inhibited α-subunit of K7.1 with K values of 10–41 μM in Xenopus oocytes and mammalian expression systems.

Importantly, KCNE β-subunits increase the sensitivity of K7.1 to chromanol 293B. K values for K7.1/KCNE1, K7.1/KCNE2 and K7.1/KCNE3 were 3–10 μM, 0.4 μM, and 3–4 μM, respectively. Voltage-gated K+ currents in pancreatic acinar cells were shown to be inhibited by chromanol 293B with a K value of 3 μM. This result supports voltage-gated K+ channels being composed of K7.1 and KCNE1 β-subunit in acinar cells. Azimilide inhibited K7.1 and K7.1/KCNE1 in the same manner as chromanol 293B with K values of 77 μM and 5.6 μM, respectively. In contrast,XE991 inhibited K7.1 and K7.1/KCNE1 with K values of 0.8 μM and 11 μM, respectively. Metloquine inhibited K7.1/KCNE1 with a K value of 0.9 μM. DIDS and mfenamic acid activated K7.1/KCNE1, but not K7.1. On the other hand, L-364,373 activated K7.1, but did not affect K7.1/KCNE1.

Regulation

Regarding regulation, voltage-gated K7.1 channels are known to be regulated by the cAMP signaling pathway. In addition, AKAPs are required for cAMP regulation of recombinant K7.1 channels in mammalian cell lines. Interestingly, a K+ current was elicited by cAMP stimulation in CFTR-transfected, but not untransfected CFPAC-1 cells derived from a cystic fibrosis patient with deletion in Phe-508 in CFTR. AKAPs also mediate PKA compartmentalization with CFTR, therefore, these findings imply that functional CFTR regulates the K7.1 channel, presumably in the luminal membrane of pancreatic duct cells.

KCNH2 (Kv11.1, HERG) and KCNHS (Kv10.2, EAG2)

Tissue expression

The KCNH2 coding Kv11.1 (HERG) protein was isolated from the hippocampal cDNA library. Functional expression of the KCNH2 gene has been demonstrated in colon carcinoma cells. Immunoreactivity of the Kv11.1 protein was also reported in colon carcinoma cells and the luminal membrane
but not KCNT1. This discrepancy indicates that the expression of KCNT1 and KCNT2 channels is in some way associated with the expression of functional CFTR. However, the function of these K⁺ channels in pancreatic duct cells remains to be investigated.

**Channel properties**

K⁺Ca.4.1 and K⁺Ca.4.2 channels have large conductances of 180 pS and 140 pS in symmetrical 130 mM KCl. In the basolateral membrane of the thick ascending limbs of Henle’s loop, Na⁺-activated K⁺ channels had a conductance of 140–180 pS. Regarding pharmacology, K⁺Ca.4.1 was inhibited by bepridil and quinidine with Kᵢ values of 1 μM and 90 μM, respectively. K⁺Ca.4.1 was activated by bithionol, nicosamide, loxapine, and niflumic acid with Kᵢ values of 0.8 μM, 2.9 μM, 4.4 μM, and 2.7 mM, respectively. K⁺Ca.4.2 was inhibited by 1 mM quinidine and isoflurane, and was activated by meclofenamic acid, flufenamic acid, and niflumic acid with Kᵢ values of 80 μM, 1.1–1.4 mM, and 2.1 mM, respectively.

**Regulation**

K⁺Ca.4.1 was shown to be unusually inhibited by intracellular Ca²⁺ at 1 μM. However, K⁺Ca.4.1 may co-assemble with K⁺Ca.1.1 subunits to generate Ca²⁺-activated K⁺ channels. K⁺Ca.4.1 and K⁺Ca.4.2 channels were reported to be activated by intracellular Na⁺ and Kᵢ values of 41 mM and 89 mM in the presence of 30 mM internal Cl⁻, respectively. These channels were also activated by intracellular Cl⁻ or synergistically by Na⁺ and Cl⁻. Intracellular ATP inhibited K⁺Ca.4.2 directly, via the presence of a consensus ATP binding motif. A similar ATP binding motif has not been demonstrated in the K⁺Ca.4.1 sequences. Interestingly, the PKC activator PMA increased K⁺Ca.4.1 currents, but inhibited K⁺Ca.4.2 currents.

**KCNKS (K⁺p.5.1, TASK-2)**

**Tissue expression**

Two-pore domain K⁺ channels (K⁺p.) generate background K⁺ currents over the whole membrane potential range. The pH-sensitive K⁺p.1 subunits (TALK-1, TALK-2 and TASK-2) were shown to be expressed in pancreatic acini. An electrophysiological study indicated that TASK-2 was expressed in HPAF, a human pancreatic ductal adenocarcinoma cell line. KCNK5 coding TASK-2 (K⁺p.5.1) was isolated from the brain cDNA library. KCNK5 is expressed in the kidney, liver, stomach, small intestine, colon, and pancreatic acini. The functional expression of K⁺p.5.1 has been demonstrated in kidney proximal convoluted tubule cells, which could be involved in volume regulation and HCO₃⁻ transport. Clofilium-sensitive K⁺ conductance, possibly K⁺p.5.1, was located in the luminal membrane of the monolayer of HPAF. pH-sensitive K⁺ channels on the luminal membrane of pancreatic duct cells may be physiologically relevant in terms of maintaining the electrical driving force for electrogenic HCO₃⁻ secretion and providing an exit pathway for K⁺ secretion.

**Channel properties**

K⁺p.5.1 channels have an intermediate conductance of 50–78 pS. Regarding pharmacology, K⁺p.5.1 was inhibited by quinine, clofilium, bupivacaine, and ropivacaine with Kᵢ values of 22 μM, 25 μM, 26 μM, and 95 μM, respectively. K⁺p.5.1 was activated by halothane, isoflurane, and chloroform, which are volatile anesthetics.

**Potassium Channels in Pancreatic Cancer**

Ion channels have been associated with the malignant phenotype of cancer cells, as well as contributing to virtually all basic cellular processes, including crucial roles in maintaining tissue homeostasis such as proliferation, differentiation, and apoptosis. Several potassium channels have been suggested as the hallmarks of cancer, including pancreatic duct adenocarcinoma. For example, K⁺Ca.3.1 channels have been correlated with the proliferation of pancreatic cancer. In addition, the expression of G protein-activated inward rectifier potassium channel 1 (K⁺p.1.1) was markedly higher in pancreatic adenocarcinomas than in a normal pancreas, whereas K⁺p.1.3 expression was decreased in pancreatic adenocarcinomas. Downregulation in the expression of K⁺p.1.3 has been associated with metastatic tumors. K⁺p.1.5 was also shown to be highly expressed in pancreatic adenocarcinomas. Furthermore, a specific monoclonal antibody that inhibits the function of K⁺p.10.1 (EAG1) reduced tumor growth of BxPC3, a human pancreas adenocarcinoma cell line, which implicates this channel in cancer progression. Altered pH homeostasis is known to be one of the key hallmarks of cancer. Thus, pH-sensitive K⁺ channels may also play a role in pancreatic adenocarcinoma. The human duct adenocarcinoma cell line, HPAF cells, were reported to express K⁺p.5.1 channels. However, its contribution to cancer progression is still unknown. Although further studies on K⁺ channels in pancreatic cancer must be performed, some candidates, such as K⁺p.10.1, already have the potential to be diagnostic tools and therapeutic targets.

**Concluding Remarks**

This review described the current status on the molecular basis for a number of K⁺ channels found in pancreatic ducts. Electrophysiological studies on ducts and duct cells using microelectrode, patch-clamp, and Ussing chamber methods showed how some of these K⁺ channels contribute to physiological processes in ductal secretion by providing the driving forces for anion transport and as partial accompanying partners in
secretion. Future studies are needed to verify the localization of K⁺ channels to a polular ductal epithelium and afirrn their physiological function in secretion or associated cell processes such as cell volume regulation, as well as their participation in cell proliferation and apoptosis. The pancreas and especially the ductal epithelium are involved in a number of diseases including cystic fibrosis and pancreatitis. Some target therapies should include K⁺ channel openers to maintain or upregulate pancreatic secretion. Our knowledge regarding the role of K⁺ channels in duct cell homeostasis remains relatively sparse. Because some K⁺ channels are being regarded as the hallmark of cancer progression and emerging studies on pancreatic adenocarcinoma foreshadow similar trends, more knowledge is required in this area before specific K⁺ channel openers or inhibitors can be used in the treatment of pancreatic diseases.

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