K⁺ channels in the secretory membrane of the parietal cell.
Focus on “Gastric parietal cell secretory membrane contains PKA- and acid-activated Kir2.1 K⁺ channels”

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Physiologists generally accept the H⁺-K⁺-ATPase as the principal gastric proton pump, and a mechanism of membrane recruitment and recycling of the pump as the primary means for regulating the activity of gastric HCl secretion. One of the major challenges in gastric HCl secretory physiology is the identification and characterization of the K⁺ efflux pathway across the apical membrane that must accompany operation of the H⁺/K⁺ pump.

When the H⁺-K⁺-ATPase was first identified (2) and shown to function as an electroneutral one-for-one H⁺ for K⁺ exchange pump (6, 13), this left a major void in the information and interpretations that were available from studies on intact gastric preparations. In particular, Warren Rehm (11, 12), who had championed a hypothesis of electrogenic HCl secretion, and others (5) had demonstrated that the gastric epithelium could generate a current of H⁺, especially in the absence of Cl⁻. Even with normal Cl⁻ solutions, electric current opposing HCl secretion (mucosa to serosa) diminished H⁺ output while enhancing Cl⁻ flux, and current passed in the direction of HCl secretion (serosa to mucosa) enhanced H⁺ output and reduced net Cl⁻ flux (11). Although the original proposals to account for these observations have been challenged, any comprehensive mechanism of the acid secretory process must be consistent with and provide rational explanation for the observations. In fact, demonstration that the apical secretory membrane of parietal cells had conductive pathways for Cl⁻ (3) demonstrated that several members of the inward rectifying (Kir) channel family, including Kir4.1, 4.2, and 7.2, are expressed as a mathematical model (6). The proposed K⁺ channel provides a path for K⁺ to enter the secretory lumen of the parietal cell and be recycled back to the cytoplasm as a substrate for the H⁺/K⁺ pump exchange. A Cl⁻ channel provides a means for Cl⁻ to enter the secretory lumen, representing a counter ion for K⁺ current across the apical membrane and balancing the charge for electroneutral HCl secretion. In the absence of Cl⁻, the apical membrane becomes a potential electrogenic H⁺ pump. Current passed serosa to mucosa would be carried by K⁺, and the resulting delivery of substrate to the H⁺/K⁺ exchange pump would deliver a current of H⁺. Without external current, the K⁺ channel together with the H⁺/K⁺ exchange pump provide an equivalent electromotive force.

Thus the existence of K⁺ and Cl⁻ channels in the stimulated apical membrane, as implied from functional studies of the membrane, could account for observations on purified gastric vesicles as well as electrophysiological data in situ. It remained to specifically identify the K⁺ and Cl⁻ channel proteins as well as to discover how they were regulated with respect to acid secretion. A CIC-2-type Cl⁻ channel, able to function at very low extracellular pH, has been identified in H⁺-K⁺-ATPase-rich membrane vesicles (7). These channels are regulated by protein kinase A (PKA) and low extracellular pH, consistent with regulation of gastric HCl secretion by cAMP. Identification of the K⁺ channel has been somewhat more elusive.

Several authors have recently identified candidate channels for the apical K⁺ conductance associated with acid secretion. Grahammer et al. (4) and Dedek and Waldegger (1) independently found that the KCNQ1 K⁺ channel (also known as KvLQT1) is abundantly expressed, and apparently colocalizes with H⁺-K⁺-ATPase, in human and mouse mucous cells. Inhibition of acid secretion in three model systems—perfused rat stomach, Heidenhain pouches in dog, and rabbit gastric glands—by chromanol 293B, a presumed “specific” KCNQ1 channel inhibitor (synthesized by Aventis Pharma), prompted the proposal that KCNQ1 and its functional subunits, KCNE2 or KCNE3, comprise the critical K⁺ channel at the apical membrane of the parietal cell (4). Using RT-PCR, Fujita et al. (3) demonstrated that several members of the inward rectifying K⁺ (Kir) channel family, including Kir4.1, 4.2, and 7.2, are also expressed in gastric tissue. Immunofluorescence and immunogold microscopy revealed that, of these family members, only Kir 4.1 was present in parietal cells and that it was in the apical microvillar regions of those cells.

In the current article in focus (Ref. 8, see page C495 in this issue), Malinowska et al. offer evidence for yet another K⁺ channel candidate to facilitate gastric secretion. They used RT-PCR and in situ hybridization to demonstrate an abundance of mRNA for Kir 2.1 in rabbit gastric mucosa, with lesser levels of Kir 4.1 and Kir 7.1. Immunofluorescence microscopy confirmed Kir 2.1 expression in parietal cells, apparently colocalizing with H⁺-K⁺-ATPase and CIC-2 Cl⁻ channels. Western blots showed that Kir 2.1 was present in H⁺-K⁺-ATPase-rich membrane vesicles isolated from stomach in either the resting or stimulated state. Channels from vesicles incorporated into planar lipid bilayers did not show rectification, but instead had linear current-voltage relationships with relatively low slope conductance (11 pS). There was a greatly increased open probability for membranes derived from stimulated, compared with resting, stomach. Treatment of resting vesicles with PKA and/or reduction of pH tended to increase open probability of incorporated channels toward that of stimulated vesicles, thus prompting the suggestion that the channel is regulated in a manner parallel to gastric CIC-2 channels.
that more than one apical K⁺ channel properties (14, 15). It is not unreasonable to suspect conductive channels for K⁺ have revealed a variety of K⁺ species) and at locations consistent with functional participation in the secretory process. They all have the interesting property of sustained activity, and possibly activation, at low pH, which is necessary for participation in acid secretion. However, the electrophysiological measurements on Kir4.1 and KCNQ1 were done with channels expressed in heterologous cells (1, 3, 4). Only Malinowska et al. (8) measured the electrical properties of channels taken from gastric membranes, but at this point it is not possible to say that the properties were indeed those of homotetrameric Kir 2.1, and not Kir4.1 or some oligomeric Kir assembly, or even KCNQ1. Patch-clamp studies on basolateral membrane of gastric acid-secreting cells have revealed a variety of K⁺ channels, each with distinctive channel properties (14, 15). It is not unreasonable to suspect that more than one apical K⁺ channel may be operating to serve the acid secretory function. Moreover, channel properties have been shown to vary widely depending on subunit assembly, with distinctive electrophysiology for homooligomeric assembly of four Kir subunits compared to heterooligomers. Properties of KCNQ1 will also vary depending on which member of the KCNE gene family it is associated with (9). The problem is now clearly focused, and further work on the signature channel properties of the apical membrane will surely resolve these questions for the parietal cell.

REFERENCES