Ion channels and transporters keep ideas flowing

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In biological membranes, channels and transporters underlie essential processes that both mediate and impact critical communications between the intracellular and extracellular milieus. Their functional impairment disrupts cellular signaling and homeostasis, ultimately translating into systemic dysfunction and disease. Similarly, disrupted function befell the collective research endeavor with the onset of the SARS-CoV2 pandemic and its imposition of isolation. For most academic researchers, this effectively ceased not only wet-bench experimentation required to collect research data, but also face-to-face exchange of

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knowledge. Although information famine challenged researchers at all stages, the effects of isolation on junior investigators carried particularly damaging potential.

The virtual Themed Meeting "New Roles for Ion Channels and Transporters in Health and Disease" was conceived as a means to sustain the community and keep it strong. By bridging the communications gap imposed by social isolation, this meeting created a forum for attendees to continue learning from--and be inspired by--colleagues near and far. Perhaps more importantly, its enthusiastic reception revealed and underscored the intangible benefit provided from feeling part of a dynamic community. With this Special Issue, we leverage the momentum gained by the Themed Meeting. It offers symposium reviews as well as topical reviews pertinent to the theme. This issue also comprises primary research papers emerging from groups settling back into experimental mode—the products of inspiration, renewed resolve, and tested resilience.

In an insightful symposium review, Margarida Amaral reflects on strategic patterns employed in generating the recent body of research on Cystic Fibrosis, a disease resulting from impaired Cystic Fibrosis Transmembrane Conductance Regulator (CFTR) function. The relative merits of emergent "omics"-based approaches (transcriptomics, proteomics, functional genomics, interactomics) are weighed against hypothesis-driven approaches driving rational drug design, as well as contrasting high-throughput screening approaches to identify pharmacotherapies (Amaral, 2022). In doing so, Amaral reviews recent strategic approaches based on targeting CFTR, rescuing the fluid secretion defect associated with CF disease, and identifying alternative Cl^- transport pathways. Thus, the example of CF provides an object lesson in strategies for tackling diverse research questions.

In contrast to impaired CFTR function resulting in Cystic Fibrosis disease, aberrantly *enhanced* activity of channels and transporters characterizes certain cancers. In the case of

gastrointestinal cancers, human ether-à-go-go related gene 1 (hERG1; $K_v11.1$) K^+ channels can drive tumour progression by canonical and non-canonical means. Arcangeli and colleagues (2022) discuss how association of hERG1 with β1-integrin subunit can recruit diverse membrane receptors, additional ion channels and transporters *i.e.* Na^+/H^+ exchanger 1 (NHE1) and a neonatal form of voltage-dependent Na^+ channel (nNa_v 1.5), as well as adhesion molecules. Their association into macromolecular signaling complexes ultimately promotes cancer progression (Arcangeli *et al.*, 2022). Interestingly, overexpressed Kv11 channels are capable of conferring resonance and membrane potential oscillations (Matsuoka *et al.*, 2021). This begs a provocative question: do resonance and oscillations modulate function of associated channels and transporters within signaling complexes? Leslie & Brackenbury (2022) further explore the importance of Na^+ homeostasis within breast tumours. Their model interweaves actions of the Na^+/K^+ ATPase, NHE1, the Na^+-HCO_3^- cotransporter 1 (NBCn1), and Na^+ coupled glucose and amino acid transporters, the epithelial Na^+ channel (ENaC), and voltage-gated sodium channels (*i.e.* Na_v 1.5). With the exception of the Na^+/K^+ ATPase, all raise intracellular Na^+. Moreover, NHE1 and NBCn1 concomitantly acidify extracellular pH. Taken together, the emergent model can explain observations of high intracellular Na^+ concentrations and low extracellular pH in breast tumours (Leslie *et al.*, 2022).

In a systematic topical review, Kaulich and coworkers (2022) broadly cover the topic of degenerin (DEG)/ENaC channels. This review bridges work arising from genetic and electrophysiological approaches, highlighting the roles of DEG/ENaC channels in manifold modes of sensation, as well as their potential as therapeutic targets (Kaulich *et al.*, 2022a). Stephan Gründer recounts his symposium presentation, providing a detailed treatment of the structural basis and diversity of neuropeptide gating of invertebrate DEG/ENaC family members (Gründer *et al.*, 2022). Moreover, he presents evidence demonstrating neuropeptide

modulation, rather than gating, of distinct regions within vertebrate ASIC channels. This process likely involves stabilization of either open or closed states. Emergent insights hold implications for understanding how endogenous neuropeptides (including opioid endorphins such as dynorphins) influence diverse physiological processes *via* their actions on ASIC channels. In an additional research paper focusing further on the topic of DEG/ENaC channels, Kaulich and colleagues report functional screening of *Caenorhabditis elegans* DEG/ENaC family members (Kaulich *et al.*, 2022b). These Authors leveraged automated voltage clamp screening, comprehensively profiling pH sensitivity of DEG/ENaC family members, and identifying novel acid-activated as well as acid-inhibited members. Obergrussberger and colleagues recently demonstrated the amenability of other DEG/ENaC family members, specifically ASIC1, to high throughput, automated patch clamp (APC) recording (Obergrussberger *et al.*, 2022). Applying this knowledge back to the observations discussed by Gründer *et al* (2022), these capabilities enable high-throughput pharmacology, to gain detailed insights into ASIC channel modulation kinetics by comprehensive panels of neuropeptides. Summarily, the DEG/ENaC family provides ample opportunities to understand the properties enabling them to behave as successful physiological sensors throughout the animal kingdom.

Endogenous metabolic by-products also can regulate channel and transporter activity. Combining electrophysiological and molecular docking approaches, work from the Rosenbaum lab on TRPV4 cation channels provides emergent evidence for modulation of their single channel conductance and open probability by lysophosphotidylcholine (LPC) and its metabolite, lysophosphotidic acid (LPA) (Benitez-Angeles *et al.*, 2023). TRPV4 expression in epithelial, smooth muscle, and endothelial tissues of organs such as skin and lungs, suggests contributions of LPC and LPA signaling to tissue, organ, and whole animal function.

In the airways, transport proteins play key roles in mediating host responses to airborne stimuli, as summarized by Hollenhorst & Krasteva-Christ (2023). Within the respiratory tract, airborne stimuli and their metabolites signal to the immune system *via* bitter taste receptors specific to distinct cell types within human and mouse upper and lower airways (Hollenhorst, 2023). In addition to initiating immune and inflammatory responses, these signals engage numerous transport proteins (including two pore domain K^+ (K2P) channels, and Trpm5 channels), enhancing mucociliary clearance critical to host defense mechanisms.

Insight into the many physiological roles of Piezo1, a mechanosensitive channel gated by forces such as pressure and membrane tension, is rapidly accumulating. In the eye, Piezo1 in trabecular meshwork cells is critical in sensing intraocular pressure and regulating it by promoting outflow of aqueous humor. Its role in the etiology of glaucoma, a disease characterized by inappropriate development of intraocular pressure, is unfolding steadily (Yarishkin *et al.*, 2021). In our Special Issue, Barnett *et al* (2022) share their discovery that Piezo1 has key functions in a very different tissue--human myometrium. There, it is modulated by PKA/AKT/eNOS-mediated paracrine signaling between the microvascular endothelium and uterine smooth muscle, thereby ensuring quiescence during gestation. It is noteworthy that less Piezo1 is expressed in myometrium from women experiencing pre-term labor. These studies therefore reveal potential targets for the development of therapies to mitigate this condition and its extenuating consequences (Barnett *et al.*, 2022). To complement these insights, an accompanying Journal Club article concisely and thoroughly considers the implications of the Barnett *et al*. study (Majhi *et al.*, 2023).

Finally, in surveying the papers included in this Special Issue, we note that much of what now is known about channels and transporters arises from functional expression studies. Such work can benefit greatly from readily available sequence information drawn from public databases. These resources offer tempting shortcuts—such as gene fragment synthesis--to

"old school" expression cloning, or even PCR cloning from reverse-transcribed cDNA. However, this potential treasure trove can also be a Pandora's box. While convenient, construction of expression plasmids directly from gene fragment synthesis using information extracted from genome sequencing can unleash trouble for downstream application and data interpretation, as discussed in (Maxeiner *et al.*, 2023). This pithy review shares problems the uninitiated might encounter, and a section subtitled "How to avoid nasty surprises" walks the reader through strategic processes to avert disaster. Given the sheer volume of physiological investigations reliant on functional expression, the cautious and rigorous researcher would do well to heed the advice offered.

Taken together, this collection of articles maps potentially fruitful directions for moving forward in understanding how ion channels and transporters function in diverse and equally impactful contexts. What other kinds of physiological, pathophysiological, and environmental signals engage channel and transporter effector machinery? How does the machinery work on different scales and how best can we study each of these? How can we use this information to understand and treat disease? What else can we learn from omics-driven research? What picture emerges from taking different approaches in context? We will know eventually--if we keep communications strong and ideas flowing.

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