

EDITORIAL

Shared and unique aspects of ligand- and voltage-gated ion-channel gatingDerek Bowie *Department of Pharmacology and Therapeutics, McGill University, Montréal, Québec H3G 1Y6, Canada*

Email: derek.bowie@mcgill.ca

Edited by: Ole Petersen

Ligand- and voltage-gated ion channels form a large superfamily of membrane-bound signalling proteins that fulfil many important roles in health and disease (Hille, 2001). Voltage-gated ion channels are primarily responsible for the generation and propagation of action potentials in excitable tissue (Stuart *et al.* 1997; Catterall *et al.* 2005; Jan & Jan, 2012) whereas ligand-gated ion channels constitute the hardwiring of chemical synapses – though they also fine tune synaptic strength during periods of sustained patterned activity and altered homeostasis (Turrigiano, 2008; Nicoll, 2017). Together, the combined activity of ligand- and voltage-gated ion channels gives rise to many complex physiological processes from cardiac and skeletal muscle contraction to the more enigmatic behaviours of the CNS such as cognition and memory.

The study of ion channels has undergone unprecedented advances in recent years with the convergence of several scientific disciplines on this important research topic. Structural biology has emerged as a leading approach to understand ion channel function as well as drug action (Gouaux & Mackinnon, 2005). Furthermore, recent advances in genetic manipulations, particularly in rodents, have permitted ion channel families to be assigned distinct roles in health and disease. To explore this emerging area of physiology, *The Journal of Physiology* sponsored a symposium at the 2017 International Union of Physiological Sciences meeting in Rio de Janeiro, Brazil entitled ‘Shared and unique aspects of the gating mechanisms of ligand- and voltage-gated ion-channels’. Chaired by *Journal* editors Drs Yoshihiro Kubo and Derek Bowie, it brought together five researchers (see Fig. 1) whose work is at the forefront of this rapidly developing field of study. This

issue of *The Journal of Physiology* brings together three timely review articles and an original paper that capture some of the ideas, discussions and debates that arose during the symposium.

Dr Yoshihiro Kubo from Japan’s National Institute for Physiological Sciences opened the symposium by presenting recent data from his lab where they looked at the unexpected effect of the broad-spectrum antiparasitic agent ivermectin on the G-protein-gated inwardly rectifying K⁺ (or GIRK) channel (Chen *et al.* 2017). Since its discovery in soil samples of bacteria taken near a Japanese golf course in the 1970s (Laing *et al.* 2017), ivermectin has proven to be one of the most effective drugs used by veterinarians and health professionals to combat parasite infection. Although its anthelmintic action is primarily mediated by targeting the activity of nematode glutamate-gated chloride channels to curtail motility, feeding and reproductive behaviour (Yates *et al.* 2003), ivermectin has also been shown to affect other ion channel families such as GABA_A, glycine and nicotinic acetylcholine receptors (Wolstenholme & Rogers, 2005). This multifaceted nature of ivermectin pharmacology is the main topic of the comprehensive review by Chen and Kubo where they explore shared and unique modulatory properties of ivermectin on a variety of ligand-gated ion channels (Chen & Kubo, 2018). Insight into ivermectin’s action on each ion channel target is beautifully illustrated by the authors’ effective use of X-ray crystallographic data to compare and contrast the structural determinants of each binding pocket. Given its hydrophobicity, ivermectin binds to residues lining the transmembrane (TM) regions of Cys-loop ligand-gated ion channels, such as the nematode glutamate-gated chloride channels and mammalian glycine receptors. The binding of ivermectin near the extracellular surface of the plasma membrane is critical as it induces the rotation of TM regions to facilitate channel opening. Surprisingly, however, ivermectin binds to GIRK channels on a different site, which lies at the interface of the TM region and intracellular domains (Chen *et al.* 2017). The authors conclude that although ivermectin binds to many ion channel targets, its clinical value as a drug

with few side effects is afforded by its preferred high-affinity binding to nematode glutamate-gated chloride channels.

Dr Cecilia Bouzat from the National University of the South (UNS), Bahia Blanca, Argentina, presented recent work from her lab on the functional and pharmacological properties of $\alpha 7$ subunit-containing nicotinic acetylcholine receptors (nAChRs) (daCosta *et al.* 2011; Andersen *et al.* 2013, 2016; Nielsen *et al.* 2018). Although nAChRs are expressed throughout the body, $\alpha 7$ receptors are especially concentrated in the mammalian brain where they exhibit both ionotropic and metabotropic functions (Wu *et al.* 2016; Kabbani & Nichols, 2018). They are also implicated in several CNS disorders (Dineley *et al.* 2015) and consequently there has been a need to develop selective drugs. The review article by Bouzat and colleagues (Bouzat *et al.* 2018) highlights these recent advances in our understanding of the $\alpha 7$ nAChR. Potentiation of the $\alpha 7$ receptor has been identified as a novel therapeutic strategy to treat several neurodegenerative disorders, including Alzheimer’s and Parkinson’s diseases, and inflammatory disorders, whereas drugs which reduce receptor activity may be beneficial in the treatment of cancer cell proliferation (Bouzat *et al.* 2018).

Dr Frank Bosmans from Johns Hopkins University in Baltimore, MD, USA talked about the pioneering work that he and his colleagues have performed using animal toxins to probe the functional behaviour of voltage-gated ion channels, particularly voltage-gated Na⁺ channels (Nav) (Bosmans & Swartz, 2010; Kalia *et al.* 2015). Voltage-gated Nav channels are primarily responsible for the rapid upstroke of the action potential in cardiac and skeletal muscle as well as being integral to the complex firing properties of central neurons. Not surprisingly, therefore, Nav channels are involved in many physiological processes and have also been implicated in numerous pathologies including cardiac dysfunction, neuropathic pain and genetic forms of epilepsy, such as Dravet syndrome (Abriel, 2010; Catterall, 2012; Waxman, 2013; Chen-Izu *et al.* 2015). Given all of this, the Nav channel community has been developing a comprehensive understanding of how the structural architecture

of Nav channels relates to their functional behaviour (Ahern *et al.* 2016). In the current issue of *The Journal*, Gilchrist and Bosmans present an original research article detailing how animal toxins can be used to understand the slow gating kinetics of Nav1.8 (Gilchrist & Bosmans, 2018), a Nav channel implicated in pain mechanisms (Han *et al.* 2016). To do this, the authors introduced toxin sensitivity into each of the four voltage sensor domains of Nav1.8 by exchanging their sequences with those of Nav1.2 to probe their role in channel gating and inactivation. Using this chimera approach, Gilchrist and Bosmans conclude that the voltage sensor domains I–III participate in channel opening, whereas voltage sensor domain IV regulates channel opening as well as the onset of fast inactivation (Gilchrist & Bosmans, 2018).

Dr Marc Gielen presented a compelling account of his postdoctoral work with Dr Trevor Smart at University College London where he performed a comprehensive analysis of Cys-loop GABA_A and glycine receptor desensitization (Gielen *et al.* 2015). The review article co-authored with his colleague, Dr Pierre-Jean Corringer, from the Institut Pasteur in Paris, is a *tour de force* treatise of the structural biology of Cys-loop receptors (Gielen & Corringer, 2018). The authors argue that although the mechanism of Cys-loop receptor activation

has been studied extensively at both the functional (Lape *et al.* 2008; Mukhtasimova *et al.* 2009; Purohit *et al.* 2013) and structural (Corringer *et al.* 2010; Althoff *et al.* 2014; Sauguet *et al.* 2014) level, an understanding of the structural basis of desensitization is only beginning to emerge (Miller & Aricescu, 2014). Functional work had already argued that desensitization of Cys-loop nicotinic acetylcholine receptors involves two distinct, but inter-related gates (Auerbach & Akk, 1998), though their location within the ion channel pore remained to be established. The authors propose that the activation and desensitization gates are structurally distinct and located at each end of the ion channel pore. This proposal is consistent with the ‘foot in the door’ mechanism assigned to the channel blocker picrotoxin (Gielen *et al.* 2015) and Markov modelling of channel activation (Gielen & Corringer, 2018). Gielen and Corringer conclude by emphasizing the importance of the dual gate mechanism of activation and desensitization as a common explanation for the behaviour of several structurally unrelated ion channels.

Dr Derek Bowie from McGill University in Montréal, Canada closed the symposium with a presentation focusing on the structural and functional mechanisms that define the distinct families of the ionotropic

glutamate receptor (iGluR). iGluRs are widely expressed in the vertebrate brain where they mediate the vast majority of fast excitatory transmission at central synapses (Dingledine *et al.* 1999; Traynelis *et al.* 2010). Not surprisingly, iGluRs are also implicated in many debilitating CNS disorders (Bowie, 2008). Despite their similar and overlapping tetrameric architecture (Sobolevsky, 2015), Bowie argued that the distinct gating behaviour of different iGluR subfamilies can be linked to differences in residues that line the interface between two subunit binding pockets that are arranged in a back-to-back formation (Dawe *et al.* 2015). This region of the protein is often referred to as the ligand-binding domain (LBD) dimer interface and was recognized from early structural studies as an important determinant of channel gating (Horning & Mayer, 2004; Bowie, 2010). Recent work from the Bowie lab has shown that the apex of the LBD dimer interface is critically important in defining the rapid millisecond gating behaviour of AMPA- and kainate-type iGluRs as well as their regulation by auxiliary proteins (Daniels *et al.* 2013; Dawe *et al.* 2013, 2016). He concluded that ongoing work from his lab suggested that the tardier gating of NMDA-type iGluRs was also determined by residues in the LBD dimer interface but from a different site. Taken together, this work



Figure 1. The speakers and colleagues who attended the symposium on ‘Shared and unique aspects of the gating mechanisms of ligand- and voltage-gated ion-channels’ at the 2017 International Union of Physiological Sciences meeting in Rio de Janeiro, Brazil

Top row, left to right: Drs Marc Gielen, Filip Van Petegem and Frank Bosmans; bottom, left to right: Drs Yoshihiro Kubo, Cecilia Bouzat and Derek Bowie.

highlights how subtle yet critical changes to the amino-acid sequence that encodes the LBD dimer interface may have contributed to the emergence of different iGluR classes during evolution.

References

- Abriel H (2010). Cardiac sodium channel $\text{Na}_v1.5$ and interacting proteins: physiology and pathophysiology. *J Mol Cell Cardiol* **48**, 2–11.
- Ahern CA, Payandeh J, Bosmans F & Chanda B (2016). The hitchhiker's guide to the voltage-gated sodium channel galaxy. *J Gen Physiol* **147**, 1–24.
- Althoff T, Hibbs RE, Banerjee S & Gouaux E (2014). X-ray structures of GluCl1 in apo states reveal a gating mechanism of Cys-loop receptors. *Nature* **512**, 333–337.
- Andersen N, Corradi J, Sine SM & Bouzat C (2013). Stoichiometry for activation of neuronal $\alpha 7$ nicotinic receptors. *Proc Natl Acad Sci USA* **110**, 20819–20824.
- Andersen ND, Nielsen BE, Corradi J, Tolosa MF, Feuerbach D, Arias HR & Bouzat C (2016). Exploring the positive allosteric modulation of human $\alpha 7$ nicotinic receptors from a single-channel perspective. *Neuropharmacology* **107**, 189–200.
- Auerbach A & Akk G (1998). Desensitization of mouse nicotinic acetylcholine receptor channels. A two-gate mechanism. *J Gen Physiol* **112**, 181–197.
- Bosmans F & Swartz KJ (2010). Targeting voltage sensors in sodium channels with spider toxins. *Trends Pharmacol Sci* **31**, 175–182.
- Bouzat C, Lasala M, Nielsen BE, Corradi J & Esandi MDC (2018). Molecular function of $\alpha 7$ nicotinic receptors as drug targets. *J Physiol* **596**, 1847–1861.
- Bowie D (2008). Ionotropic glutamate receptors & CNS disorders. *CNS Neurol Disord Drug Targets* **7**, 129–143.
- Bowie D (2010). Ion-dependent gating of kainate receptors. *J Physiol* **588**, 67–81.
- Catterall WA (2012). Voltage-gated sodium channels at 60: structure, function and pathophysiology. *J Physiol* **590**, 2577–2589.
- Catterall WA, Goldin AL & Waxman SG (2005). International Union of Pharmacology. XLVII. Nomenclature and structure-function relationships of voltage-gated sodium channels. *Pharmacol Rev* **57**, 397–409.
- Chen IS & Kubo Y (2018). Ivermectin and its target molecules: shared and unique modulation mechanisms of ion channels and receptors by ivermectin. *J Physiol* **596**, 1833–1845.
- Chen IS, Tateyama M, Fukata Y, Uesugi M & Kubo Y (2017). Ivermectin activates GIRK channels in a PIP_2 -dependent, $\text{G}_{\beta\gamma}$ -independent manner and an amino acid residue at the slide helix governs the activation. *J Physiol* **595**, 5895–5912.
- Chen-Izu Y, Shaw RM, Pitt GS, Yarov-Yarovoy V, Sack JT, Abriel H, Aldrich RW, Belardinelli L, Cannell MB, Catterall WA, Chazin WJ, Chiamvimonvat N, Deschenes I, Grandi E, Hund TJ, Izu LT, Maier LS, Maltsev VA, Marionneau C, Mohler PJ, Rajamani S, Rasmuson RL, Sobie EA, Clancy CE & Bers DM (2015). Na^+ channel function, regulation, structure, trafficking and sequestration. *J Physiol* **593**, 1347–1360.
- Corringer PJ, Baaden M, Bocquet N, Delarue M, Dufresne V, Nury H, Prevost M & Van Renterghem C (2010). Atomic structure and dynamics of pentameric ligand-gated ion channels: new insight from bacterial homologues. *J Physiol* **588**, 565–572.
- daCosta CJ, Free CR, Corradi J, Bouzat C & Sine SM (2011). Single-channel and structural foundations of neuronal $\alpha 7$ acetylcholine receptor potentiation. *J Neurosci* **31**, 13870–13879.
- Daniels BA, Andrews ED, Arousseau MR, Accardi MV & Bowie D (2013). Crosslinking the ligand-binding domain dimer interface locks kainate receptors out of the main open state. *J Physiol* **591**, 3873–3885.
- Dawe GB, Arousseau MR, Daniels BA & Bowie D (2015). Retour aux sources: defining the structural basis of glutamate receptor activation. *J Physiol* **593**, 97–110.
- Dawe GB, Musgaard M, Andrews ED, Daniels BA, Arousseau MR, Biggin PC & Bowie D (2013). Defining the structural relationship between kainate-receptor deactivation and desensitization. *Nat Struct Mol Biol* **20**, 1054–1061.
- Dawe GB, Musgaard M, Arousseau MRP, Nayeem N, Green T, Biggin PC & Bowie D (2016). Distinct structural pathways coordinate the activation of AMPA receptor-auxiliary subunit complexes. *Neuron* **89**, 1264–1276.
- Dineley KT, Pandya AA & Yakel JL (2015). Nicotinic ACh receptors as therapeutic targets in CNS disorders. *Trends Pharmacol Sci* **36**, 96–108.
- Dingledine R, Borges K, Bowie D & Traynelis SF (1999). The glutamate receptor ion channels. *Pharmacol Rev* **51**, 7–61.
- Gielen M & Corringer P-J (2018). The dual-gate model for pentameric ligand-gated ion channels activation and desensitization. *J Physiol* **596**, 1873–1902.
- Gielen M, Thomas P & Smart TG (2015). The desensitization gate of inhibitory Cys-loop receptors. *Nat Commun* **6**, 6829.
- Gilchrist J & Bosmans F (2018). Using voltage-sensor toxins and their molecular targets to investigate $\text{NaV} 1.8$ gating. *J Physiol* **596**, 1863–1872.
- Gouaux E & Mackinnon R (2005). Principles of selective ion transport in channels and pumps. *Science* **310**, 1461–1465.
- Han C, Huang J & Waxman SG (2016). Sodium channel $\text{Nav}1.8$: emerging links to human disease. *Neurology* **86**, 473–483.
- Hille B (2001). *Ion Channels of Excitable Membranes*, 3rd edn, Sinauer Associates, Inc., Sunderland, MA, USA.
- Horning MS & Mayer ML (2004). Regulation of AMPA receptor gating by ligand binding core dimers. *Neuron* **41**, 379–388.
- Jan LY & Jan YN (2012). Voltage-gated potassium channels and the diversity of electrical signalling. *J Physiol* **590**, 2591–2599.
- Kabbani N & Nichols RA (2018). Beyond the channel: metabotropic signaling by nicotinic receptors. *Trends Pharmacol Sci* **39**, 354–366.
- Kalia J, Milesu M, Salvatierra J, Wagner J, Klint JK, King GF, Olivera BM & Bosmans F (2015). From foe to friend: using animal toxins to investigate ion channel function. *J Mol Biol* **427**, 158–175.
- Laing R, Gillan V & Devaney E (2017). Ivermectin – old drug, new tricks? *Trends Parasitol* **33**, 463–472.
- Lape R, Colquhoun D & Sivillotti LG (2008). On the nature of partial agonism in the nicotinic receptor superfamily. *Nature* **454**, 722–727.
- Miller PS & Aricescu AR (2014). Crystal structure of a human GABA_A receptor. *Nature* **512**, 270–275.
- Mukhtasimova N, Lee WY, Wang HL & Sine SM (2009). Detection and trapping of intermediate states priming nicotinic receptor channel opening. *Nature* **459**, 451–454.
- Nicoll RA (2017). A brief history of long-term potentiation. *Neuron* **93**, 281–290.
- Nielsen BE, Minguez T, Bermudez I & Bouzat C (2018). Molecular function of the novel $\alpha 7\beta 2$ nicotinic receptor. *Cell Mol Life Sci* (in press; <https://doi.org/10.1007/s00018-017-2741-4>).
- Purohit P, Gupta S, Jadey S & Auerbach A (2013). Functional anatomy of an allosteric protein. *Nat Commun* **4**, 2984.
- Sauguet L, Shahsavari A, Poitevin F, Huon C, Menny A, Nemezc A, Haouz A, Changeux JP, Corringer PJ & Delarue M (2014). Crystal structures of a pentameric ligand-gated ion channel provide a mechanism for activation. *Proc Natl Acad Sci USA* **111**, 966–971.
- Sobolevsky AI (2015). Structure and gating of tetrameric glutamate receptors. *J Physiol* **593**, 29–38.
- Stuart G, Spruston N, Sakmann B & Häusser M (1997). Action potential initiation and backpropagation in neurons of the mammalian CNS. *Trends Neurosci* **20**, 125–131.
- Traynelis SF, Wollmuth LP, McBain CJ, Menniti FS, Vance KM, Ogden KK, Hansen KB, Yuan H, Myers SJ & Dingledine R (2010). Glutamate receptor ion channels: structure, regulation, and function. *Pharmacol Rev* **62**, 405–496.
- Turrigiano GG (2008). The self-tuning neuron: synaptic scaling of excitatory synapses. *Cell* **135**, 422–435.
- Waxman SG (2013). Painful Na^+ -channelopathies: an expanding universe. *Trends Mol Med* **19**, 406–409.

- Wolstenholme AJ & Rogers AT (2005). Glutamate-gated chloride channels and the mode of action of the avermectin/milbemycin anthelmintics. *Parasitology* **131**(Suppl), S85–95.
- Wu J, Liu Q, Tang P, Mikkelsen JD, Shen J, Whiteaker P & Yakel JL (2016). Heteromeric $\alpha 7\beta 2$ nicotinic acetylcholine receptors in the brain. *Trends Pharmacol Sci* **37**, 562–574.
- Yates DM, Portillo V & Wolstenholme AJ (2003). The avermectin receptors of *Haemonchus contortus* and *Caenorhabditis elegans*. *Int J Parasitol* **33**, 1183–1193.

Additional information**Competing interests**

None declared.