

Ion Channels

Methods and Protocols

Edited by

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It is now clear that ion channels play essential roles in cell biology and physiology and that their dysfunction is the root cause of many human diseases. In *Ion Channels: Methods and Protocols*, internationally recognized biomedical scientists describe recent technological breakthroughs and demonstrate their use in successful experimental designs. The diverse applications range from the study of allosteric regulation of ion channel activity using a classic mutagenesis approach, to the study of channel subunit stoichiometry using a novel biophysical approach based on fluorescence resonance energy transfer. Highlights include methods for heterologous expression of ion channels in cells, for determining channel structure-function, for studying channel regulation and physiological function, and for genetic screening and investigating channelopathies. The protocols follow the successful *Methods in Molecular Biology*™ series format, each offering step-by-step laboratory instructions, an introduction outlining the principles behind the technique, lists of the necessary equipment and reagents, and tips on troubleshooting and avoiding known pitfalls.

Comprehensive and highly practical, *Ion Channels: Methods and Protocols* offer both novice and experienced researchers a versatile collection of proven techniques for illuminating the structure, function, and regulation of ion channel proteins and finding suitable drug targets when they cause disease.

FEATURES

- Proven techniques for studying the structure, function, and regulation of ion channels
- Cutting-edge methods for genetic screening and studying channelopathies
- Readily reproducible techniques for heterologous expression of ion channels in cells
- Discussion of recent technological breakthroughs and advanced experimental designs
- Step-by-step instructions to ensure rapid implementation and successful results
- Tricks of the trade and notes on troubleshooting and avoiding known pitfalls

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Part I. Methods for Exogenous Expression of Ion Channels in Cells. Functional Reconstitution of the Human Epithelial Na⁺ Channel in a Mammalian Expression System. Overexpression of Proteins in Neurons Using Replication-Deficient Virus. Exogenous Expression of Proteins in Neurons Using the Biolistic Particle Delivery System. **Part II. Methods for Studying Channel Structure-Function.** Tertiary and Quaternary Structure Formation of Voltage-Gated Potassium Channels. Biophysical Approach to Determine the Subunit Stoichiometry of the Epithelial Sodium Channel Using the *Xenopus laevis* Oocyte Expression System. Spectroscopy-Based Quantitative Fluorescence Resonance Energy Transfer Analysis. **Part III. Methods for Studying Channel Regulation and Physiological Function.** Probing the Effects of Phosphoinositides on Ion Channels. Epithelial Sodium Channel in Planar Lipid Bilayers. A Simple In Vivo Method for Assessing Changes

of Membrane-Bound Ion Channel Density in *Xenopus* Oocytes. Preparation of Cortical Brain Slices for Electrophysiological Recording. Juxtacellular Labeling and Chemical Phenotyping of Extracellularly Recorded Neurons In Vivo. Carbon Fiber Amperometry in the Study of Ion Channels and Secretion. **Part IV. Methods for Studying Channelopathies, Genetic Screening, and Molecular Biology.** Genetic Screening for Functionality of Bacterial Potassium Channel Mutants Using K⁺ Uptake-Deficient *Escherichia coli*. KCNQ1 K⁺ Channel-Mediated Cardiac Channelopathies. Tissue-Specific Transgenic and Knockout Mice. Index.

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