



Calcium Techniques: A Laboratory Manual

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Life begins with a surge of calcium ions (Ca^{2+}) at fertilization, and thereafter, Ca^{2+} signaling influences nearly every aspect of mammalian development and physiology, from gene expression and cell proliferation to muscle contraction and nerve impulses. To create spatiotemporally distinct Ca^{2+} signals, cells use a variety of mechanisms to recognize, transport, and buffer Ca^{2+} . Thus, a diverse range of reliable experimental techniques is necessary to study the movement of Ca^{2+} and the various effectors involved.

This laboratory manual provides step-by-step protocols for studying many facets of Ca^{2+} signaling, as well as background information on the principles and applications of the techniques. Contributors discuss how to use fluorescent, luminescent, and genetically encoded Ca^{2+} probes in conjunction with state-of-the-art imaging modalities to characterize Ca^{2+} signals. Electrophysiological measurements of Ca^{2+} channel activity are described, as are radioactive Ca^{2+} flux assays and methods to investigate signaling mediated by specific Ca^{2+} -mobilizing messengers (IP_3 , cADPR, and NAADP). Techniques to modulate and suppress intra- and intercellular signals are also provided. Each protocol is complete with a list of required materials, detailed recipes for media and reagents, and troubleshooting advice.

Specific chapters are devoted to Ca^{2+} signaling techniques in non-mammalian systems, such as plants, yeast, zebrafish, and *Xenopus*. Methods for assessing Ca^{2+} -binding kinetics and strategies for developing mathematical models of Ca^{2+} signaling are also included. Thus, this manual is a comprehensive laboratory resource for biochemists, cell and developmental biologists, and physiologists who are using or looking to expand their repertoire of Ca^{2+} techniques.

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