
Preface

The nucleus provides eukaryotic cells with a unique functional compartment to protect and process the genetic information, offering both versatility and robustness to fine-tune gene expression and genome function. Inside the nucleus, the DNA is wrapped around histone proteins in a highly organized manner to form chromatin, which provides packaging and regulatory functions to control gene expression, genome replication, repair, and recombination. First discovered as a stainable substance in the cell during the nineteenth century, chromatin is now recognized as a highly dynamic macromolecular assembly that integrates developmental and environmental cues to adjust the transcriptional program, to respond to damage, and to facilitate DNA replication. Chromatin dynamics are driven by a complex interplay of events at the microscopic, nanoscopic, molecular, and biochemical levels. There is a growing interest in understanding how dynamic changes in chromatin organization and the epigenetic landscape direct and respond to physiological and developmental processes, and with this, the variety and power of methods to analyze plant chromatin have increased accordingly. For example, the rise of high-throughput techniques to analyze chromatin fractions has made it possible to elucidate the genome-wide composition and distribution of nucleosomes, histone variants, histone modifications and DNA modifications in relation to gene accessibility, local chromatin structure, gene expression, and DNA replication and repair activities. In addition, novel experimental designs have facilitated the identification of spatial interactions between genomic regions within the nuclear space, generating models for interchromosomal associations and functional, subnuclear chromatin domains.

Considerable progress has been made in obtaining genome-wide snapshots of chromatin states, but the effect of these different states on processes such as transcription and replication and on the 3D organization of chromatin is largely unknown. Extensive effort is therefore being spent on understanding these processes by using a combination of classical biochemistry and genetics, together with state-of-the-art protein complex identification methods. Recent progress in methods to assess 3D chromatin architecture has fuelled the emerging view that the spatial organization of chromatin has a major role in the regulation of gene expression. Studies on inter- and intrachromosomal chromatin contacts, gene positions relative to distinct chromatin compartments, and the functional compartmentalization of chromatin domains in different cell types are valuable approaches to elucidate the functional relevance of chromatin organization.

Understanding chromatin dynamics and functionality in plants requires the development of optimized protocols that solve or circumvent the technical challenges posed by the complexity of plant genomes, the plant cell wall, and the relative inaccessibility of specific cell types inside complex tissues and organs. In this edition, we provide a comprehensive collection of protocols that can be exploited to study plant chromatin structure and composition (Section I), to investigate the regulation of chromatin features in relation to the epigenetic regulation of gene expression and function (Section II), and to explore the interaction between chromatin modifications, gene regulation, and the 3D spatial organization of the chromatin inside the nucleus (Section III). In addition to presenting a large number of state-of-the-art protocols, each section is prefaced by a Review summarizing the current state of knowledge on the topic and the various approaches that can be used to study the specific aspects of plant chromatin dynamics. Moreover, three Technical Reviews offer an

overview of specific methods together with a balanced discussion of their benefits and limitations, with the aim of providing practical advice to the reader.

This volume contains a comprehensive collection of detailed protocols and (technical) reviews that will assist the plant chromatin community in understanding the relationships between 3D chromatin architecture, local chromatin structure, epigenetic modifications, and functional genomics. We are very grateful to all authors for sharing their knowledge, practical experience, and theoretical considerations by providing detailed protocols presented in a pragmatic and accessible manner. We believe that this collective effort will greatly contribute to exciting discoveries in the field of plant chromatin research.

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