

Preface

Nearly fourteen years separates the publication of these volumes from the original *Plant Molecular Biology—A Practical Approach*. The original book, edited by Charlie Shaw, represented a milestone in the Practical Approach series as the first book in the series dedicated to the application of molecular biology to plant biology. In 1988, the field of Plant Molecular Biology was burgeoning. A number of recombinant DNA techniques had been adapted for the analysis of plant genes, and several dozen genes had been isolated and characterised from plants. The techniques for transformation of a few species using *Agrobacterium tumefaciens* mediated gene transfer had been developed although there were comparatively few laboratories with practical experience of producing transgenic plants. The methods for using endogenous plant transposons for gene tagging had been established and the first steps towards using transposons as tools for heterologous gene tagging had been taken. Several techniques for the analysis of plant organelles were also available, including the post-translational transport of proteins into chloroplasts. Approaches for analysis of the sub-cellular localization of macromolecules included light, fluorescent and electron microscopy. In addition to the growing use of recombinant DNA approaches to the study of higher plants, these techniques were also being applied to plant viruses as well as other models including *Chlamydomonas* and *Cyanobacteria*. All these approaches were featured in *Plant Molecular Biology—A Practical Approach*, and an appendix reported the use of the 'recently developed reporter gene' β -glucuronidase.

The advances since 1988 are astounding. There have been an unimaginable number of technical developments that have revolutionised the tools available and the scale of experiments that are possible. A large number of Practical Approach Books have been published since 1988 and several of these have focused on aspects of plant science or included approaches applicable to the analysis of plants, but there was never a second edition of *Plant Molecular Biology—A Practical Approach*. In compiling this all-new two-volume book, *Molecular Plant Biology*, we were mindful that this was not a second edition, but a sequel. Many of the approaches and applications described in the initial book are as relevant and important today as they were in 1988. However, as technologies

have advanced there are now a myriad of approaches that build on the original techniques, as well as entirely new approaches based on technologies that did not exist at the time of publication of the original book.

The development and wide-spread use of PCR, the advent of automated DNA sequencing, the availability of entire plant genome sequences, the discovery of new reporter genes, the technological advances in bio-robotics and imaging that have resulted in DNA micro-arrays, and the ability to routinely transform a wide range of plant species, including all our major crops, highlight just a few of these developments. Not only have these advances revolutionised the way that fundamental experimental biology can be done, but they have also been applied commercially with the appearance, and in some cases subsequent removal, of products from transgenic plants on our supermarket shelves.

These tremendous advances have occurred in a relatively short time period, and we have both been fortunate enough to have witnessed all these advances during our own research careers. As editors of this new two-volume book, *Molecular Plant Biology* we have drawn on our combined research experiences and contacts to identify and persuade an international array of authors with particular expertise to contribute the chapters that we feel represent the breadth and scope of the field today. We would like to express our extreme gratitude to these authors for contributing to these books. The range of approaches covered in these two volumes is immense. Not only would it be impossible to find an individual with hands-on experience and expertise in all the topics covered in these two books, but there will surely be few laboratories anywhere where all these approaches are in routine use. We would like to thank all the contributors for sharing their expertise and making these books possible.

In editing these books we were keen to incorporate practical approaches as applied to a range of plant species, and not just the common models. Although much can be gained from the focused analysis of one species, there are many aspects of biology that are unique to non-model organisms. Some approaches covered in the original book are not covered in these new volumes. This is not meant to signify that they are no longer relevant or important, it is merely a reflection of the breadth of the field and a lack of space to incorporate and update approaches for specific organisms such as *Chlamydomonas* and *Cyanobacteria*. An exception to this has been the inclusion of a new chapter dealing with the moss *Physcomitrella patens*, which as a relatively new molecular model and represents the only plant in which homologous recombination is routine.

The two volumes are used to divide the topics into two themes, gene identification and isolation (Volume 1) and gene expression and gene product analysis (Volume 2). Each volume is divided into three sections and the rationale for the organisation of topics within these Sections is described below. However, many of the approaches have multiple applications, and others may therefore see alternative logic.

In Volume 1, the chapters in Section 1 (Gene Identification) provide methods and considerations for gene identification by classical mutagenesis. Also included here is a chapter on plant transformation, because it is an essential tool

for the following two chapters which cover gene tagging strategies in transgenic plants. The final chapter in Section 1 focuses on genomic subtraction for gene identification. In Section 2 (Gene Organization) we have included a chapter on approaches for gene mapping, and a further chapter on the techniques for construction and screening of YAC, BAC and cosmid libraries. Also covered are approaches for chromosome *in situ* analysis. Section 3 (Library Screening and cDNA Isolation) there are three chapters that describe PCR cloning strategies, western and South-western library screens and complementation cloning.

In Volume 2, the first Section (Gene Expression) covers a range of methodologies for the analysis of gene expression, with chapters on transcript analysis, *in situ* RNA hybridisation and DNA micro-arrays. Approaches for the *in vitro* analysis of DNA:protein interactions are also covered, along with a final chapter on inducible gene expression in plants. In Section 2 (Gene Product Analysis) there are six chapters covering heterologous expression of recombinant proteins, analysis of proteins and protein targeting to chloroplasts, and biochemical approaches for the analysis of plant tissues. The application of the yeast two-hybrid system for analysis of protein:protein interactions is also included with the final two chapters in this Section devoted to antibody techniques and the construction and application of phage display libraries. The final Section of the book is devoted to functional analysis *in vivo*. In this Section we have included three chapters where the approaches primarily permit analyses in living cells. The first of these focuses on calcium imaging, the second describes a range of approaches using various reporter genes and the third is devoted to *Physcomitrella patens* as a model organism in which homologous recombination provides an exciting new *in vivo* approach. As the final chapter in the two volumes, this brings us full circle back to the first chapter in Volume 1 that is dedicated to mutagenesis.

We hope that this organisation illustrates that the approaches described in these volumes are not a linear progression through genetic, biochemical and molecular techniques, but that integration of these approaches and reiteration of analyses are essential to generate a global perspective of plant biology. In compiling the indexes for these two volumes we have provided one combined index for both books to facilitate full cross-referencing between the two volumes and we hope that this proves to be useful.

In 1988 when we first used *Plant Molecular biology—A Practical Approach*, one of us had just started a PhD, the other had just started a postdoc. We hope that the readers of these new books will find them as useful as we found the original book during our own time at the bench.

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P. M. G.

C. B.