



Preface

The concept for this manual grew out of our experiences as instructors in the early to mid 1990s for the Cold Spring Harbor Laboratory course on Molecular Embryology of the Mouse, or “The Mouse Course,” as it was commonly known. That course, which continues to this day as the course “Mouse Development, Stem Cells, and Cancer,” has been, for 40 years, the premier place to learn techniques associated with the use of the mouse in developmental, physiological, and genetic studies, and in particular for making and analyzing genetic alterations through transgenesis and gene targeting.

We found, however, that after three intense weeks of learning the language of mouse embryology and being instructed in sophisticated, cutting-edge techniques of gene manipulation by experts in the field, the students still had questions at the end of the course about how to put it all together in order to analyze a mutant. Their consternation when faced with a complex phenotype resulting from a change brought about by gene manipulation or spontaneous mutation was our biggest challenge as teachers, and there was little in the way of reference books we could point to for guidance. Our original handbook (see below), and now this up-to-date laboratory manual, is our attempt to provide a strategic guide, a road map through the entire procedure of analyzing the phenotype of a mutant mouse strain, starting with the planning stages of obtaining or producing a mutation by mutagenesis experiments, by homologous recombination gene targeting in ES cells, or by gene editing by CRISPR–Cas technology, to the complete analysis of complex phenotypes that might have their effects at any stage of development.

This edition, now in the form of a laboratory manual, has been adapted, expanded, and updated from our book, *Mouse Phenotypes: A Handbook of Mutation Analysis* (Papaioannou and Behringer 2004). The manual provides the concepts and tools needed to make genetic alterations in the mouse and to analyze the resulting mutant phenotypes, with an emphasis on analyzing mutations that affect embryonic development. Advice on strategy and practical hints are provided, to help make the analysis of mutant phenotypes a more efficient and productive enterprise. The book takes the form of a branching pathway of analysis as illustrated in Chapter 1. By answering simple questions about the particular mutation or phenotype, the reader is directed to the relevant chapters of the book, skipping over chapters that do not apply. Specific examples of mutations, protocols, and situations are fully referenced, often based on our own experiences, both in the text and in Appendix 2. Whatever your starting point and whether you are a novice or an experienced embryologist, this book will guide you through a fundamental phenotypic analysis to study gene function in the context of the organism as a whole.

We would like to thank Cold Spring Harbor Laboratory Press for the opportunity to update and expand our original book with a second edition. In the years since its first publication, new technologies have brought genetic manipulations within the reach of virtually any laboratory, rendering the principles of phenotypic analysis even more relevant. We again thank our students in the Cold Spring Harbor course on Molecular Embryology of the Mouse, 1995 and 1996, for the inspiration for this book as well as our co-instructors on the course, Peter Koopman, Terry Magnuson, and Andras Nagy, and course assistants, Debbie Chapman, Yuji Mishina, Jenny Nichols, and Bill Shawlot.

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Reference

Papaioannou VE, Behringer RR. 2004. *Mouse phenotypes: a handbook of mutation analysis*. Cold Spring Harbor Laboratory Press, Cold Spring Harbor, NY.

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