Contents

Preface ................................................................. v
Contributors .......................................................... ix

PART I  IDENTIFICATION OF REGULATORY RNAs

1 Bioinformatic Discovery of Bacterial Regulatory RNAs Using SIPHT ........ 3
Jonathan Livny

2 How to Identify CRISPRs in Sequencing Data ............................... 15
Christine Drevet and Christine Pourcel

3 A Strategy for Identifying Noncoding RNAs Using Whole-Genome Tiling Arrays ................................................................. 29
Stephen G. Landt and Eduardo Abeliuk

4 Genetic Screens to Identify Bacterial sRNA Regulators .............. 41
Pierre Mandin

PART II  FUNCTION AND EXPRESSION OF REGULATORY RNAs

5 Competition Assays Using Barcoded Deletion Strains to Gain Insight into Small RNA Function ......................................................... 63
Errett C. Hobbs and Gisela Storz

6 Use of Semi-quantitative Northern Blot Analysis to Determine Relative Quantities of Bacterial CRISPR Transcripts ..................... 73
Ksenia Pougach and Konstantin Severinov

7 RNA Visualization in Bacteria by Fluorescence In Situ Hybridization ..... 87
Jay H. Russell and Kenneth C. Keiler

PART III  RNA STRUCTURE AND BIOPHYSICAL MEASUREMENTS

8 RNA Structure Prediction: An Overview of Methods ...................... 99
Matthew G. Seetin and David H. Mathews

9 Crystallization of RNA–Protein Complexes: From Synthesis and Purification of Individual Components to Crystals ..................... 123
Anna Perederina and Andrey S. Krasilnikov

10 Analysis of RNA Folding and Ligand Binding by Conventional and High-Throughput Calorimetry ...................................... 145
Joshua E. Sokoloski and Philip C. Bevilacqua
PART IV RNA-PROTEIN INTERACTIONS

11 Use of Aptamer Tagging to Identify In Vivo Protein Binding Partners of Small Regulatory RNAs

Colin P. Corcoran, Renate Rieder, Dimitri Podkaminski, Benjamin Hofmann, and Jörg Vogel

12 Gel Mobility Shift Assays to Detect Protein–RNA Interactions

Alexander V. Yakhnin, Helen Yakhnin, and Paul Babitzke

13 RNase Footprinting of Protein Binding Sites on an mRNA Target of Small RNAs

Yi Peng, Toby J. Soper, and Sarah A. Woodson

PART V RNA–RNA INTERACTIONS

14 Computational Identification of sRNA Targets

Brian Tjaden

15 Detection of sRNA–mRNA Interactions by Electrophoretic Mobility Shift Assay

Teppei Morita, Kimika Maki, and Hiroji Aiba

16 Activity of Small RNAs on the Stability of Targeted mRNAs In Vivo

Guillaume Desnoyers and Eric Massé

PART VI INTERACTIONS OF REGULATORY RNA WITH RNA POLYMERASE AND THE RIBOSOME

17 Native Gel Electrophoresis to Study the Binding and Release of RNA Polymerase by 6S RNA

Karen M. Wassarman

18 Ribosome Purification Approaches for Studying Interactions of Regulatory Proteins and RNAs with the Ribosome

Preeti Mehta, Perry Woo, Krithika Venkataraman, and A. Wali Karzai

19 Analysis of Aminoacyl- and Peptidyl-tRNAs by Gel Electrophoresis

Brian D. Janssen, Elie J. Diner, and Christopher S. Hayes

20 In Vitro Trans-Translation Assays

Daisuke Kurita, Akira Muto, and Hyouta Himeno

Index