Contents

Preface ............................................................................................................. v
Contributors ................................................................................................... xiii

PART I. OVERVIEW
1 Structural Basis of Protein–Protein Interactions
   Robert C. Liddington ................................................................. 3
2 Quantitative Analysis of Protein–Protein Interactions
   Keith D. Wilkinson ................................................................. 15

PART II. IN VITRO TECHNIQUES
3 Characterization of Protein–Protein Interactions
   by Isothermal Titration Calorimetry
   Adrian Velazquez-Campoy, Stephanie A. Leavitt,
   and Ernesto Freire ................................................................. 35
4 Circular Dichroism Analysis for Protein–Protein Interactions
   Norma J. Greenfield ................................................................. 55
5 Protein–Protein Interaction Analysis by Nuclear Magnetic
   Resonance Spectroscopy
   Guanghua Gao, Jason G. Williams, and Sharon L. Campbell .......... 79
6 Measuring Rhodopsin–G-Protein Interactions
   by Surface Plasmon Resonance
   John Northup ............................................................................ 93
7 Using Light Scattering to Determine the Stoichiometry
   of Protein Complexes
   Jeremy Mogridge ........................................................................ 113
8 Sedimentation Equilibrium Studies
   Ian A. Taylor, John F. Eccleston, and Katrin Rittinger ................. 119
9 Analysis of Protein–Protein Interactions by Simulation
   of Small-Zone Gel Filtration Chromatography
   Rosemarie Wilton, Elizabeth A. Myatt, and Fred J. Stevens .......... 137
10 Fluorescence Gel Retardation Assay to Detect
    Protein–Protein Interactions
    Sang-Hyun Park and Ronald T. Raines .................................. 155
## Contents

<table>
<thead>
<tr>
<th>Chapter</th>
<th>Title</th>
<th>Authors</th>
<th>Page</th>
</tr>
</thead>
<tbody>
<tr>
<td>11</td>
<td>Fluorescence Polarization Assay to Quantify Protein–Protein Interactions</td>
<td>Sang-Hyun Park and Ronald T. Raines</td>
<td>161</td>
</tr>
<tr>
<td>12</td>
<td>Studying Protein–Protein Interactions via Blot Overlay or Far Western Blot</td>
<td>Randy A. Hall</td>
<td>167</td>
</tr>
<tr>
<td>13</td>
<td>Glutathione-S-Transferase–Fusion Based Assays for Studying Protein–Protein Interactions</td>
<td>Haris G. Vikis and Kun-Liang Guan</td>
<td>175</td>
</tr>
<tr>
<td>14</td>
<td>Affinity Capillary Electrophoresis Analyses of Protein–Protein Interactions in Target-Directed Drug Discovery</td>
<td>William E. Pierceall, Lixin Zhang, and Dallas E. Hughes</td>
<td>187</td>
</tr>
<tr>
<td>15</td>
<td>Mapping Protein–Ligand Interactions by Hydroxyl-Radical Protein Footprinting</td>
<td>Nick Loizos</td>
<td>199</td>
</tr>
<tr>
<td>16</td>
<td>Use of Phage Display and Polyvalency to Design Inhibitors of Protein–Protein Interactions</td>
<td>Michael Mourez and R. John Collier</td>
<td>213</td>
</tr>
</tbody>
</table>

**PART III. DETECTING PROTEIN–PROTEIN INTERACTIONS IN HETEROLOGOUS SYSTEMS**

<table>
<thead>
<tr>
<th>Chapter</th>
<th>Title</th>
<th>Authors</th>
</tr>
</thead>
<tbody>
<tr>
<td>17</td>
<td>A Bacterial Two-Hybrid System Based on Transcriptional Activation</td>
<td>Simon L. Dove and Ann Hochschild</td>
</tr>
<tr>
<td>18</td>
<td>Using the Yeast Two-Hybrid System to Identify Interacting Proteins</td>
<td>John Miller and Igor Stagljar</td>
</tr>
<tr>
<td>19</td>
<td>Analysis of Protein–Protein Interactions Utilizing Dual Bait Yeast Two-Hybrid System</td>
<td>Ilya G. Serebriiskii and Elena Kolova</td>
</tr>
<tr>
<td>20</td>
<td>The Split-Ubiquitin Membrane-Based Yeast Two-Hybrid System</td>
<td>Safia Thaminy, John Miller, and Igor Stagljar</td>
</tr>
<tr>
<td>21</td>
<td>Reverse Two-Hybrid Techniques in the Yeast Saccharomyces cerevisiae</td>
<td>Matthew A. Bennett, Jack F. Shern, and Richard A. Kahn</td>
</tr>
<tr>
<td>22</td>
<td>Mammalian Two-Hybrid Assay for Detecting Protein–Protein Interactions In Vivo</td>
<td>Jae Woon Lee and Soo-Kyung Lee</td>
</tr>
<tr>
<td>23</td>
<td>Co-Immunoprecipitation from Transfected Cells</td>
<td>Shane C. Masters</td>
</tr>
</tbody>
</table>
Contents

PART IV: PROBING PROTEIN–PROTEIN INTERACTIONS IN LIVING CELLS

24 Microscopic Analysis of Fluorescence Resonance Energy Transfer (FRET)
   Brian Herman, R. Venkata Krishnan, and Victoria E. Centonze ........................................ 351

25 Monitoring Molecular Interactions in Living Cells Using Flow Cytometric Analysis of Fluorescence Resonance Energy Transfer
   Francis Ka-Ming Chan ................................................................................................. 371

26 Fluorescence Correlation Spectroscopy: A New Tool for Quantification of Molecular Interactions
   Keith M. Berland ........................................................................................................ 383

27 Confocal Microscopy for Intracellular Co-Localization of Proteins
   Toshiyuki Miyashita .................................................................................................... 399

28 Mapping Biochemical Networks with Protein-Fragment Complementation Assays
   Ingrid Remy and Stephen W. Michnick ...................................................................... 411

29 In Vivo Protein Cross-Linking
   Fabrice Agou, Fei Ye, and Michel Véron .................................................................. 427

PART V: PROTEOMICS-BASED APPROACHES

30 Computational Prediction of Protein–Protein Interactions
   John C. Obenauer and Michael B. Yaffe .................................................................... 445

31 Affinity Methods for Phosphorylation-Dependent Interactions
   Greg Moorhead and Carol MacKintosh ...................................................................... 469

32 Two-Dimensional Gel Electrophoresis for Analysis of Protein Complexes
   Karin Barnouin ......................................................................................................... 479

33 Sample Preparation of Gel Electrophoretically Separated Protein Binding Partners for Analysis by Mass Spectrometry
   Rainer Cramer, Malcolm Saxton, and Karin Barnouin ............................................. 499

34 Quantitative Protein Analysis by Solid Phase Isotope Tagging and Mass Spectrometry
   Huilin Zhou, Rosemary Boyle, and Ruedi Aebersold .............................................. 511

35 Internet Resources for Studying Protein–Protein Interactions
   Shane C. Masters ........................................................................................................ 519

Index .......................................................................................................................... 525
Protein-Protein Interactions
Methods and Applications
Fu, H. (Ed.)
2004, XVI, 532 p., Hardcover
A product of Humana Press