
Contents

Preface	v
Contributors	ix

PART I MUTAGENESIS AND RECOMBINATION METHODS

1	Generating Mutant Libraries Using Error-Prone PCR Patrick C. Cirino, Kimberly M. Mayer, and Daisuke Umeno	3
2	Preparing Libraries in <i>Escherichia coli</i> Alexander V. Tobias	11
3	Preparing Libraries in <i>Saccharomyces cerevisiae</i> Thomas Bulter and Miguel Alcalde	17
4	Creating Random Mutagenesis Libraries by Megaprimer PCR of Whole Plasmid (MEGAWHOP) Kentaro Miyazaki	23
5	Construction of Designed Protein Libraries Using Gene Assembly Mutagenesis Paul H. Bessette, Marco A. Mena, Annalee W. Nguyen, and Patrick S. Daugherty	29
6	Production of Randomly Mutated Plasmid Libraries Using Mutator Strains Annalee W. Nguyen and Patrick S. Daugherty	39
7	Evolution of Microorganisms Using Mutator Plasmids Olga Selifonova and Volker Schellenberger	45
8	Random Insertion and Deletion Mutagenesis Hiroshi Murakami, Takahiro Hohsaka, and Masahiko Sisido	53
9	Random Oligonucleotide Mutagenesis Jessica L. Sneeden and Lawrence A. Loeb	65
10	Saturation Mutagenesis Radu Georgescu, Geethani Bandara, and Lianhong Sun	75
11	DNA Shuffling John M. Joern	85
12	Family Shuffling with Single-Stranded DNA Wenjuan Zha, Tongbo Zhu, and Huimin Zhao	91
13	In Vitro DNA Recombination by Random Priming Olga Esteban, Ryan D. Woodyer, and Huimin Zhao	99

14	Staggered Extension Process (StEP) In Vitro Recombination Anna Marie Aguinaldo and Frances H. Arnold	105
15	RACHITT: Gene Family Shuffling by Random Chimeragenesis on Transient Templates Wayne M. Coco	111
16	The Creation of ITCHY Hybrid Protein Libraries Marc Ostermeier and Stefan Lutz	129
17	Preparation of SCRATCHY Hybrid Protein Libraries: Size- and In-Frame Selection of Nucleic Acid Sequences Stefan Lutz and Marc Ostermeier	143
18	Sequence Homology-Independent Protein Recombination (SHIPREC) Andrew K. Udit, Jonathan J. Silberg, and Volker Sieber	153
19	Producing Chimeric Genes by CLERY: In Vitro and In Vivo Recombination Valérie Abécassis, Denis Pompon, and Gilles Truan	165
PART II ANALYSIS OF LIBRARY DIVERSITY		
20	Analysis of Shuffled Libraries by Oligonucleotide Probe Hybridization Peter Meinhold, John M. Joern, and Jonathan J. Silberg	177
21	Sequence Mapping of Combinatorial Libraries on Macro- or Microarrays: Experimental Design of DNA Arrays Valérie Abécassis, Gilles Truan, Loïc Jaffrelo, and Denis Pompon	189
22	Sequence Mapping of Combinatorial Libraries on Macro- and Microarrays: Bioinformatic Treatment of Data Denis Pompon, Gilles Truan, and Valérie Abécassis	199
	Index	213

Directed Evolution Library Creation

Methods and Protocols

Arnold, F.H.; Georgiou, G. (Eds.)

2003, X, 226 p., Hardcover

ISBN: 978-1-58829-285-8

A product of Humana Press