

Functional Genomics


Methods and Protocols

Second Edition

Edited by

Michael Kaufmann and Claudia Klinger

Private Universität, Witten/Herdecke gGmbH, Witten, Germany

 **Humana Press**

Contents

<i>Preface</i>	v
<i>Contributors</i>	xi
PART I BIOINFORMATICS	
1 Prediction of Protein Tertiary Structures Using MUFOLD	3
<i>Jingfen Zhang, Zhiquan He, Qingguo Wang, Bogdan Barz, Ioan Kosztin, Yi Shang, and Dong Xu</i>	
2 Prediction of Protein Functions	15
<i>Roy D. Sleator</i>	
3 Genome-Wide Screens for Expressed Hypothetical Proteins	25
<i>Claus Desler, Jon Ambæk Durhuus, and Lene Juel Rasmussen</i>	
4 Self-Custom-Made SFP Arrays for Nonmodel Organisms	39
<i>Ron Ophir and Amir Sherman</i>	
PART II DNA ANALYSIS	
5 Construction and Analysis of Full-Length and Normalized cDNA Libraries from Citrus	51
<i>M. Carmen Marques and Miguel A. Perez-Amador</i>	
6 Assembling Linear DNA Templates for In Vitro Transcription and Translation	67
<i>Viktor Stein, Miriam Kaltenbach, and Florian Hollfelder</i>	
7 Automated Computational Analysis of Genome-Wide DNA Methylation Profiling Data from HELP-Tagging Assays	79
<i>Qiang Jing, Andrew McLellan, John M. Grealley, and Masako Suzuki</i>	
PART III RNA ANALYSIS	
8 Detection of RNA Editing Events in Human Cells Using High-Throughput Sequencing	91
<i>Iouri Chepelev</i>	
9 Comparative Study of Differential Gene Expression in Closely Related Bacterial Species by Comparative Hybridization	103
<i>Ruisheng An and Parwinder S. Grewal</i>	
10 Whole-Genome RT-qPCR MicroRNA Expression Profiling	121
<i>Pieter Mestdagh, Stefaan Derveaux, and Jo Vandesompele</i>	
11 Using Quantitative Real-Time Reverse Transcriptase Polymerase Chain Reaction to Validate Gene Regulation by PTTG	131
<i>Siva Kumar Panguluri and Sham S. Kakar</i>	

- 12 FRET-Based Real-Time DNA Microarrays 147
*Arjang Hassibi, Haris Vikalo, José Luis Riechmann,
 and Babak Hassibi*

PART IV PROTEIN ANALYSIS I: QUANTIFICATION AND IDENTIFICATION

- 13 2-D Gel Electrophoresis: Constructing 2D-Gel
 Proteome Reference Maps 163
*Maria Paola Simula, Agata Notarpietro, Giuseppe Toffoli,
 and Valli De Re*
- 14 The Use of Antigen Microarrays in Antibody Profiling 175
Krisztián Papp and József Prechl
- 15 Limited Proteolysis in Proteomics Using
 Protease-Immobilized Microreactors. 187
Hiroshi Yamaguchi, Masaya Miyazaki, and Hideaki Maeda
- 16 Mass Spectrometry for Protein Quantification in Biomarker Discovery 199
Mu Wang and Jinsam You

PART V PROTEIN ANALYSIS II: FUNCTIONAL CHARACTERIZATION

- 17 High-Throughput Microtitre Plate-Based Assay
 for DNA Topoisomerases 229
James A. Taylor, Nicolas P. Burton, and Anthony Maxwell
- 18 Microscale Thermophoresis as a Sensitive Method to Quantify Protein:
 Nucleic Acid Interactions in Solution 241
*Karina Zillner, Moran Jerabek-Willemsen, Stefan Dubr,
 Dieter Braun, Gernot Längst, and Philipp Baaske*
- 19 Bioluminescence Resonance Energy Transfer: An Emerging Tool
 for the Detection of Protein-Protein Interaction in Living Cells 253
Søren W. Gersting, Amelie S. Lotz-Havla, and Ania C. Muntau
- 20 LuMPIS: Luciferase-Based MBP-Pull-Down Protein Interaction
 Screening System 265
Maria G. Vizoso Pinto and Armin Baiker
- 21 Yeast Two-Hybrid Screens: Improvement of Array-Based Screening
 Results by N- and C-terminally Tagged Fusion Proteins 277
*Thorsten Stellberger, Roman Häuser, Peter Uetz,
 and Albrecht von Brunn*
- 22 Inducible microRNA-Mediated Knockdown of the Endogenous
 Human Lamin A/C Gene 289
Ina Weidenfeld
- 23 Multiple-Gene Silencing Using Antisense RNAs in *Escherichia coli* 307
Nobutaka Nakashima, Shan Goh, Liam Good, and Tomohiro Tamura
- 24 Functional Screen of Zebrafish Deubiquitylating Enzymes
 by Morpholino Knockdown and In Situ Hybridization 321
William Ka Fai Tse and Yun-Jin Jiang

25	Silencing of Gene Expression by Gymnotic Delivery of Antisense Oligonucleotides	333
	<i>Harris S. Soifer, Troels Koch, Johnathan Lai, Bo Hansen, Anja Hoeg, Henrik Oerum, and C.A. Stein</i>	
26	Polycistronic Expression of Interfering RNAs from RNA Polymerase III Promoters	347
	<i>Laura F. Steel and Viraj R. Sanghvi</i>	
PART VI METABOLITE ANALYSIS		
27	Metabolite Analysis of <i>Cannabis sativa</i> L. by NMR Spectroscopy	363
	<i>Isvett Josefina Flores-Sanchez, Young Hae Choi, and Robert Verpoorte</i>	
28	Metabolome Analysis of Gram-Positive Bacteria such as <i>Staphylococcus aureus</i> by GC-MS and LC-MS	377
	<i>Manuel Liebeke, Kirsten Dörries, Hanna Meyer, and Michael Lalk</i>	
29	Metabolic Fingerprinting Using Comprehensive Two-Dimensional Gas Chromatography – Time-of-Flight Mass Spectrometry	399
	<i>Martin F. Almstetter, Peter J. Oefner, and Katja Dettmer</i>	
	<i>Index</i>	413