

CONTENTS

1. NUCLEAR TRANSFORMATION OF EUKARYOTIC MICROALGAE: HISTORICAL OVERVIEW, ACHIEVEMENTS AND PROBLEMS..... 1

Rosa León and Emilio Fernández

Abstract	1
Introduction	1
Microalgae Groups Transformed	2
Methods for Microalgae Transformation	4
Characteristics of the Transformation Process	6
DNA Constructions Used in Transformation	6
Difficulties for Stable Expression of the Transgenes	8
Concluding Remarks	8

2. TRANSFORMATION OF CYANOBACTERIA 12

Agustín Vioque

Abstract	12
Introduction	12
Transformation of Cyanobacteria	13
Applications	14

3. MOLECULAR BIOLOGY AND THE BIOTECHNOLOGICAL POTENTIAL OF DIATOMS 23

Peter Kroth

Abstract	23
Diatom Biology	23
Genetic Manipulation of Diatoms	25
Biochemistry of Diatoms and Technological Applications	29
Synthesis of Fatty Acids	29
Biomining 30	
Concluding Remarks	31

4. TOOLS AND TECHNIQUES FOR CHLOROPLAST TRANSFORMATION OF *CHLAMYDOMONAS* 34

Saul Purton

Abstract	34
Introduction	34
Delivery of DNA into the Chloroplast Compartment	36
Integration of Transforming DNA	37
Polyploidy and the Problems of Heteroplasmy	39
Selection Strategies	41
Reverse-Genetic Studies of the <i>Chlamydomonas</i> Plastome	42
Expression of Foreign Genes in the <i>Chlamydomonas</i> Chloroplast	42
Future Prospects	43

5. INFLUENCE OF CODON BIAS ON THE EXPRESSION OF FOREIGN GENES IN MICROALGAE 46

Markus Heitzer, Almut Eckert, Markus Fuhrmann and Christoph Griesbeck

Abstract	46
General Aspects of Codon Bias in Pro- and Eukaryotic Expression Hosts	46
<i>Phaeodactylum tricornutum</i>	47
<i>Chlamydomonas reinhardtii</i> —Expression from Chloroplast and Nucleus	48
Concluding Remarks	52

6. IN THE GRIP OF ALGAL GENOMICS 54

Arthur R. Grossman

Abstract	54
Introduction	54
Which Organisms Should Have Their Genomes Sequenced?	56
Full Genome Sequences	56
cDNA and Partial Genome Sequences	64
Viral Genomes	65
Concluding Remarks	67

7. INSERTIONAL MUTAGENESIS AS A TOOL TO STUDY GENES/FUNCTIONS IN *CHLAMYDOMONAS* 77

Aurora Galván, David González-Ballester and Emilio Fernández

Abstract	77
<i>Chlamydomonas</i> as a Model for Translational Biology	77
Mutants as a Tool for Functional Genomics	78
Future Perspectives	86

8. OPTIMIZATION OF RECOMBINANT PROTEIN EXPRESSION IN THE CHLOROPLASTS OF GREEN ALGAE 90

Samuel P. Fletcher, Machiko Muto and Stephen P. Mayfield

Abstract	90
Introduction	90
Expression of Recombinant Proteins in the <i>Chlamydomonas</i> Chloroplast	92
Strategies for Increasing Recombinant Protein Expression in Algal Chloroplast	94
Conclusion and Prospectus	96

9. PHYCOREMEDIATION OF HEAVY METALS USING TRANSGENIC MICROALGAE 99

Sathish Rajamani, Surasak Siripornadulsil, Vanessa Falcao, Moacir Torres,
Pio Colepicolo and Richard Sayre

Abstract	99
Metals in the Environment	99
The Role of the Algal Cell Wall in Heavy Metal Binding and Tolerance	100
The Plasma Membrane and Heavy Metal Flux	101
Heavy Metal Metabolism in the Cytoplasm of Algae	102
Algal Heavy Metal Biosensors	103
Application of Engineered Algae for Bioremediation: The Risks and Benefits	106

10. HYDROGEN FUEL PRODUCTION BY TRANSGENIC MICROALGAE 110

Anastasios Melis, Michael Seibert and Maria L. Ghirardi

Abstract	110
Overview	110
Sulfur-Nutrient Deprivation Attenuates Photosystem-II Repair and Promotes H ₂ -Production in Unicellular Green Algae	111
Genetic Engineering of Sulfate Uptake in Microalgae for H ₂ -Production	113
Application of the Hydrogenase Assembly Genes in Conferring H ₂ -Production Capacity in a Variety of Organisms	113
Engineering O ₂ Tolerance to the Green Algal Hydrogenase	115
Engineering Starch Accumulation in Microalgae for H ₂ -Production	116
Engineering Optimal Light Utilization in Microalgae for H ₂ -Production	117
Future Directions	118

11. MICROALGAL VACCINES 122

Surasak Siripornadulsil, Konrad Dabrowski and Richard Sayre

Abstract	122
Introduction	122
Oral Vaccines	123
Microalgal Vaccines	123
Recent Progress	124

INDEX 129