

Contents

Part I Introduction

1	Molecular Genetic Approaches to Maize Improvement – an Introduction	3
	Robert T. Fraley	
	References	6
2	Maize Tissue Culture and Transformation: The First 20 Years	7
	Todd J. Jones	
2.1	Introduction	7
2.2	Maize Plant Regeneration Systems	8
2.2.1	Somatic Embryogenesis in Maize	8
2.2.2	Organogenesis in Maize	11
2.3	Maize Transformation Systems	12
2.3.1	Protoplast Transformation	13
2.3.2	Particle Bombardment (Biolistics)	13
2.3.3	<i>Agrobacterium</i> -Mediated Transformation	15
2.4	Selectable Marker Systems	16
2.4.1	Herbicidal Selectable Markers	16
2.4.2	Alternative Non-antibiotic, Non-herbicidal Selectable Markers	17
2.5	Marker-Free Transformation	18
2.5.1	Co-transformation and Transgene Segregation	18
2.6	Future Prospects: Bigger and Better	20
2.6.1	Homologous Recombination and Targeted Integration ...	20
2.6.2	High Molecular Weight DNA Transformation	22
	References	23

Part II Transgenic Traits

3 Insect Resistance in Corn Through Biotechnology	31
Graham Head and Dannette Ward	
3.1 Introduction	31
3.2 The Nature of Bt Corn Technologies	32
3.3 Adoption of Bt Corn Technologies and Their Impact on Insecticide Use	33
3.4 The Economic Impact of Bt Corn	35
3.5 The Impact of Bt Corn on Grain Quality	36
3.6 The Environmental Impact of Bt Corn	37
3.7 Conclusions	38
References	39
4 Seed Total Phosphate and Phytic Acid	41
Victor Raboy	
4.1 Introduction	41
4.2 Phytic Acid Synthesis, Breakdown and Storage	43
4.3 Seed Total P	47
4.4 Conclusion	50
References	51
5 Traits and Genes for Plant Drought Tolerance	55
John Mullet	
5.1 Introduction	55
5.2 Prospects for Improving Plant Stress Tolerance Through Genetics	56
5.3 Physiological/Developmental Framework for Assessing the Role and Potential Utility of Genes and Traits for Drought Tolerance	58
5.4 Identification and Testing Gene/Trait Leads for Drought Tolerance	59
5.5 Deployment of Drought Tolerance Genes and Genotype	62
References	63
6 Biotechnology Approaches to Improving Maize Nitrogen Use Efficiency	65
Stephen Moose and Fred E. Below	
6.1 The Importance of Improving Nitrogen Use Efficiency and a Biotechnology Approach	65
6.2 The Biology of Maize NUE	66
6.2.1 N Uptake and Assimilation	67
6.2.2 N Transport	68
6.2.3 N Utilization by Kernels	69
6.2.4 Regulation of N-Associated Processes	70

Contents	ix
6.3 Candidate Genes for Enhancing Maize NUE.....	71
6.3.1 Quantitative Trait Loci	71
6.3.2 RNA Expression Profiling	72
6.3.3 Transgenes for Improving Maize NUE.....	72
6.4 Commercialization of Maize Hybrids with Improved NUE.....	73
References	76
7 Enhancement of Amino Acid Availability in Corn Grain	79
Alan L. Kriz	
7.1 Introduction	79
7.2 Increased Lysine Accumulation Through Deregulation of Metabolic Pathways	80
7.3 Modification of Corn Grain Protein Profiles	81
7.3.1 Distribution of Proteins in the Corn Grain	81
7.3.2 Zein Reduction	81
7.4 Expression of Lysine-rich Proteins in Corn Grain.....	86
References	87
8 Over-expression of Novel Proteins in Maize.....	91
Elizabeth E. Hood and John A. Howard	
8.1 Introduction	91
8.1.1 Why Over-produce Proteins?.....	91
8.1.2 What Do We Want in a Host for Over-production?	91
8.2 Expression Technology	92
8.2.1 What Is the Protein Being Expressed and How Much is Accumulated?	92
8.2.2 Protein Characteristics	92
8.2.3 Molecular and Cellular Characteristics.....	93
8.2.4 General Tools to Effect Accumulation	96
8.3 Production	97
8.3.1 Confinement.....	99
8.4 Examples of Products	100
8.5 Future Prospects	101
References	101
9 Global Regulation of Transgenic Crops	107
Bruce M. Chassy	
9.1 Regulatory Oversight of Transgenic Maize	107
9.1.1 Development of a Regulatory Paradigm and Rationale ..	108
9.1.2 Divergent Regulatory Approaches Around the World ..	109
9.2 Scientific Assessment of Risks Associated with Transgenic Maize	110
9.2.1 Description of the Event and Organisms.....	113
9.2.2 Evaluation of Agricultural Hazards	113

9.2.3	Evaluation of Environmental Hazards	113
9.2.4	Evaluation of Food Safety Hazards	115
9.3	Discussion and Conclusions	120
	References	122

Part III Breeding and Genetics

10	Doubled Haploids	127
	Ming-Tang Chang and Edward H. Coe, Jr	
10.1	Introduction	127
10.2	History	128
10.3	Methods	130
10.3.1	Spontaneous Haploids	130
10.3.2	Genetic Induction	130
10.3.3	Modifications in Handling	131
10.3.4	Artificial Induction	131
10.3.5	Anther Culture, Embryo Culture and Microspore Culture	132
10.3.6	Wide Crosses and Chromosome Elimination	132
10.3.7	Apomixis (Parthenogenesis and/or Androgenesis)	132
10.4	Chromosome Doubling	133
10.4.1	Spontaneous Doubling	133
10.4.2	Selection for Spontaneous Doubling	134
10.4.3	Artificial Doubling	134
10.5	Advantages	134
10.5.1	Genetic Homozygosity	134
10.5.2	Genetic Enrichment	135
10.5.3	Gamete Selection	135
10.5.4	Gene Mutation	136
10.5.5	Molecular Mapping Applications	136
10.6	Future Perspectives	137
	References	138
11	Transposon Tagging and Reverse Genetics	143
	A. Mark Settles	
11.1	Introduction	143
11.2	General Strategies for Transposon Tagging	144
11.3	Directed Tagging	146
11.4	Non-directed Tagging	149
11.5	Reverse Genetics Resources	151
11.5.1	Single-Gene Screening Resources	151
11.5.2	Flanking Sequence Tags and Reverse Genetics	152
11.5.3	An Optimal Reverse Genetics Strategy?	154
11.6	Future Perspectives	155
	References	155

12 EMS Mutagenesis and Point Mutation Discovery	161
Clifford F. Weil and Rita-Ann Monde	
12.1 Introduction	161
12.2 EMS Mutagenesis	161
12.3 TILLING	164
12.3.1 TILLING Mutagenized Lines	164
12.3.2 EcoTILLING	166
12.4 Targeted Resequencing Using Massively Parallel Strategies:	
TRUMPing TILLING	168
12.5 Conclusion	170
References	170
13 Applications of Linkage Disequilibrium and Association Mapping in Maize	173
Elhan S. Ersoz, Jianming Yu, and Edward S. Buckler	
13.1 Introduction	173
13.2 What is Linkage Disequilibrium and How is it Related to Association Mapping Studies	174
13.2.1 How to Estimate LD	175
13.2.2 Interpretation of LD Data	176
13.2.3 LD in Maize	177
13.3 Association Populations and Statistics	177
13.3.1 Population Structure	179
13.3.2 Classic Association Populations	180
13.3.3 Family-Based Association Populations	181
13.3.4 Special Association Populations	182
13.4 False Positives and Power of Association	183
13.5 Phenotyping and Genotyping Strategies for Association Testing	185
13.6 Association Mapping in Crop Plants	187
13.7 Conclusions	189
References	190
14 Maize Genetic Resources	197
Martin M. Sachs	
14.1 Introduction	197
14.2 Genetic Stocks	198
14.2.1 Maize Genetics Cooperation Stock Center (MGCSC; GSZE)	198
14.2.2 The Genetic Stock Collection	199
14.2.3 The Services Provided	201
14.2.4 The Value of the Stocks	202
14.3 Other Maize Germplasm	203
14.3.1 The North Central Regional Plant Introduction Station (NCRPIS; NC7)	203

14.3.2	Centro Internacional de Mejoramiento de Maiz y Trigo (CIMMYT)	204
14.3.3	The National Center for Genetic Resources Preservation (NCGRP)	205
14.4	Conclusions	205
	References	206

Part IV The Corn Genome

15	The Structure of the Maize Genome	213
	Joachim Messing	
15.1	Introduction	213
15.2	The Gold Standard of Genome Sequence	214
15.3	Fractionation Methods of the Maize Genome	215
15.4	Distribution of Methylated and Repetitive DNA in the Maize Genome	216
15.5	One Hundred Random Regions of the Maize Genome	217
15.6	Physical Map of the Maize Genome	218
15.7	Evolution of Maize Chromosome Numbers	220
15.8	Diploidization of the Maize Genome	220
15.9	Retrotransposition	221
15.10	Chromosome Expansion and Contraction	222
15.11	Orthologous and Paralogous Gene Copies	224
15.12	Haplotype Variation	225
	References	227
16	Molecular Markers	231
	Patrick S. Schnable, An-Ping Hsia, Ling Guo, and W. Brad Barbazuk	
16.1	Utility of Molecular Markers	231
16.2	Molecular Markers	232
16.2.1	Detection of Polymorphisms	232
16.2.2	SSRs (Simple Sequence Repeats)	232
16.2.3	IDPs (InDel Polymorphisms)	233
16.2.4	SNPs (Single Nucleotide Polymorphisms)	233
16.3	Maize Mapping Populations	235
16.4	Genetic Maps of Maize	236
16.5	Future Perspectives	237
	References	237
17	Applied Cytogenetics	241
	R. Kelly Dawe	
17.1	Chromosome Analysis on Mitotic Chromosome by FISH	241
17.2	Histones, ChIP, Genes and Histones	242
17.3	Centromere Cytogenetics	243
17.4	Minichromosomes – Using Cytogenetics to Produce a Better Vector	244

17.4.1	Background	244
17.4.2	Efforts in Plants	244
17.4.3	Limitations and Outlook for Engineered Chromosomes	245
	References	246
18	The Wonderland of Global Expression Profiling	251
	David W. Galbraith	
18.1	Introduction	251
18.1.1	Some Definitions	251
18.1.2	Available Platforms for Global Expression Profiling	252
18.1.3	Fractionation and Prepurification Procedures for Complex Systems	255
18.1.4	The Information Content of the Maize Genome	256
18.2	Global Transcript Analysis	257
18.2.1	Affymetrix GeneChips	257
18.2.2	Microarrays Employing PCR Amplicons as Probes	258
18.2.3	Microarrays Employing Long Oligonucleotides as Probes	259
18.2.4	Non-Microarray-Based Profiling	260
18.3	MicroRNA Profiling	261
18.4	Conclusions and Future Prospects	262
	References	262
Part V Molecular Biology and Physiological Studies		
19	Zein Storage Proteins	269
	David R. Holding and Brian A. Larkins	
19.1	Introduction	269
19.2	Storage Proteins in the Maize Kernel	270
19.2.1	Embryo Proteins	270
19.2.2	Endosperm Proteins	271
19.3	High Lysine Corn and the Development of Quality Protein Maize	279
19.4	Future Perspectives	281
	References	282
20	The Complexities of Starch Biosynthesis in Cereal Endosperms	287
	L. Curtis Hannah and Thomas Greene	
20.1	Introduction	287
20.2	The Starch Biosynthetic Pathway	288
20.3	Adenosine Diphosphate Glucose Pyrophosphorylase (AGPase)	289
20.3.1	Subunits of AGPase	289
20.3.2	Subcellular localization of AGPase	290

20.3.3	Allosteric Properties of AGPase	291
20.3.4	Allosteric Properties of AGPase Are Pivotal in Controlling Starch Levels.....	292
20.4	Starch Synthases (SS).....	293
20.4.1	Starch Synthase Isoforms	294
20.5	Starch Branching Enzymes (SBE)	295
20.6	Starch Debranching Enzymes (DBE)	296
20.6.1	Physiological Role of DBEs.....	297
	References	298
21	Development of a High Oil Trait for Maize	303
	Dale L. Val, Steven H. Schwartz, Michael R. Kerns, and Jill Deikman	
21.1	Introduction	303
21.2	Background	304
21.2.1	Kernel Morphology and Lipid Content	304
21.2.2	Effects of Environment and Agricultural Practices on Kernel Oil Content	305
21.2.3	Inheritance of Oil Phenotype	305
21.3	Breeding for High Oil	306
21.3.1	High Oil Sources	306
21.3.2	Oil QTL Analysis	306
21.3.3	TopCross TM Strategy for High Oil Hybrids	308
21.3.4	Contemporary Strategies for High Oil Breeding	308
21.4	Synthesis of Oil in the Kernel	308
21.4.1	Synthesis and Plastid Import of the Carbon Precursor(s) of Acetyl-CoA	310
21.4.2	Synthesis of Acetyl-CoA in the Plastid	312
21.4.3	Plastidial de novo Fatty Acid Synthesis	312
21.4.4	Fatty Acid Termination, Export and Transfer to the ER	313
21.4.5	ER Membrane Glycerolipid Synthesis and Fatty Acid Desaturation	314
21.4.6	Synthesis of TAG from ER Membrane Lipids	314
21.5	Regulation of Oil Biosynthesis	316
21.6	Conclusions	316
	References	318
22	Chloroplasts	325
	Delene J. Oldenburg and Arnold J. Bendich	
22.1	Introduction	325
22.2	Size, Form, and Genomic Map of cpDNA	326
22.2.1	Highlights of the Genome Sequence	326
22.2.2	The Traditional Model: The Circular Form of Maize cpDNA	327
22.2.3	The Revised Model: Linear and Complex Forms of Maize cpDNA	328

22.3	Replication of cpDNA	331
22.3.1	The Traditional Model: D-Loop-to-Theta-to-Rolling Circle Replication	331
22.3.2	The Revised Model: OPaLI-RDR	331
22.4	Plastid Development in Maize	333
22.4.1	Progressive Leaf and Plastid Development	333
22.4.2	Changes in Genome Copy Number and cpDNA Molecular Form	334
22.4.3	Effects of Light on cpDNA	335
22.4.4	Genes that Influence Plastid Development and cpDNA Levels	336
22.4.5	Mesophyll and Bundle Sheath Cell-Specific Processes ..	337
22.5	Strategies for Engineering the Chloroplast	338
22.5.1	Nuclear-Encoded Plastid-Targeted Transgenes	338
22.5.2	Plastid-Encoded Transgenes	338
	References	340

Part VI Biomass and Energy

23	Ethanol Production from Maize	347
	Stefan Schwietzke, Youngmi Kim, Eduardo Ximenes, Nathan Mosier, and Michael Ladisch	
23.1	Introduction	348
23.2	Maize as a Feedstock for Ethanol Production	348
23.3	Ethanol Production from Corn Grain	350
23.3.1	Wet Milling	351
23.3.2	Dry Milling	353
23.3.3	Enzymes	354
23.3.4	DDGS	354
23.4	Ethanol Production from Corn Cob and Corn Stover	355
23.4.1	Cellulolytic Microorganisms	357
23.4.2	Cellulolytic Enzymes	357
23.5	Comparison of Ethanol Yields for Conversion of Starch, By-products, and Corn Stover	359
23.6	Conclusion	362
	References	362
	Index	365