PLANT BIOTECHNOLOGY AND FOOD SECURITY

Adoption, Farming Systems, and Sustainable Development

Edited by Jyoti Prakash Sahoo and
Kailash Chandra Samal

CRC Press
Taylor & Francis Group

Plant Biotechnology and Food Security

The global agricultural sector faces mounting pressures from climate change, population growth, and the urgent need for sustainable food production. As the world moves toward a population of nearly 10 billion by 2050, innovative solutions are essential to ensure food security. *Plant Biotechnology and Food Security: Adoption, Farming Systems, and Sustainable Development* provides a comprehensive examination of how biotechnological advancements can transform agriculture, enhance productivity, and promote environmental sustainability. This book bridges the gap between traditional farming practices and cutting-edge technologies, offering a balanced perspective on the role of biotechnology in modern agriculture.

Key Features:

- *Agricultural Evolution and Biotech Integration*: Examines the development of agriculture and the need for biotechnological advances.
- *GM Crops and RNAi Technology*: Explores genetically modified crops and the use of RNA interference for creating stress-resistant crops.
- *Genome Editing and CRISPR*: Discusses CRISPR/Cas9 and HI-Edit technology as tools for enhancing climate resilience in agriculture.
- *Genomics and Molecular Breeding*: Focuses on next-generation sequencing and molecular breeding techniques for crop improvement.
- *Precision Agriculture and Biofortification*: Highlights digital tools in agriculture, biotech for plant protection, and the role of biofortified crops in addressing malnutrition.

This book is an essential resource for researchers, policymakers, agricultural professionals, and students seeking to understand how biotechnology can drive food security while balancing economic, environmental, and ethical considerations. By presenting scientific advancements alongside real-world applications, it aims to foster collaboration in building a resilient and sustainable agricultural future.

Taylor & Francis
Taylor & Francis Group
http://taylorandfrancis.com

Plant Biotechnology and Food Security

Adoption, Farming Systems, and Sustainable Development

Edited by
Jyoti Prakash Sahoo

and
Kailash Chandra Samal

CRC Press
Taylor & Francis Group
Boca Raton London New York

CRC Press is an imprint of the
Taylor & Francis Group, an **informa** business

Designed cover image: Getty Images

First edition published 2026
by CRC Press
2385 NW Executive Center Drive, Suite 320, Boca Raton FL 33431

and by CRC Press
4 Park Square, Milton Park, Abingdon, Oxon, OX14 4RN

CRC Press is an imprint of Taylor & Francis Group, LLC

© 2026 selection and editorial matter, Jyoti Prakash Sahoo and Kailash Chandra Samal; individual chapters, the contributors

Reasonable efforts have been made to publish reliable data and information, but the author and publisher cannot assume responsibility for the validity of all materials or the consequences of their use. The authors and publishers have attempted to trace the copyright holders of all material reproduced in this publication and apologize to copyright holders if permission to publish in this form has not been obtained. If any copyright material has not been acknowledged please write and let us know so we may rectify in any future reprint.

Except as permitted under U.S. Copyright Law, no part of this book may be reprinted, reproduced, transmitted, or utilized in any form by any electronic, mechanical, or other means, now known or hereafter invented, including photocopying, microfilming, and recording, or in any information storage or retrieval system, without written permission from the publishers.

For permission to photocopy or use material electronically from this work, access www.copyright.com or contact the Copyright Clearance Center, Inc. (CCC), 222 Rosewood Drive, Danvers, MA 01923, 978-750-8400. For works that are not available on CCC please contact mpkbookspermissions@tandf.co.uk

Trademark notice: Product or corporate names may be trademarks or registered trademarks and are used only for identification and explanation without intent to infringe.

ISBN: 978-1-032-98262-5 (hbk)
ISBN: 978-1-032-98265-6 (pbk)
ISBN: 978-1-003-59779-7 (ebk)

DOI: 10.1201/9781003597797

Typeset in Times
by KnowledgeWorks Global Ltd.

Contents

Preface

The global agricultural landscape is undergoing unprecedented challenges, driven by climate change, population growth, and the urgent need for sustainable development. As the world strives to achieve food security for a projected population of nearly 10 billion by 2050, the role of plant biotechnology has become increasingly pivotal. This book, *Plant Biotechnology and Food Security: Adoption, Farming Systems, and Sustainable Development*, seeks to explore the transformative potential of biotechnology in addressing these issues. The chapters in this book are meticulously designed to cover a wide spectrum of topics, ranging from the foundational principles of traditional farming to the latest advancements in genome editing and digital agriculture.

Chapter 1 sets the stage by examining the historical context of agriculture and the need for integrating modern biotechnological tools to enhance productivity and sustainability. The focus then shifts to the transformative potential of genetically modified crops (Chapter 2) and the revolutionary role of RNA interference (Chapter 3) in strengthening crop resilience against biotic and abiotic stresses. Furthermore, the book explores the ground-breaking applications of genome editing (Chapter 4) and haploid inducer-mediated genome editing (HI-Edit) technology (Chapter 5) in developing climate-resilient crops. The integration of next-generation sequencing and crop genomics (Chapter 6) with molecular breeding approaches (Chapter 7) is another focal point of this book. The book also explores the collaborative potential of biotechnology and precision agriculture (Chapter 8) in enhancing plant disease management, alongside the vital role of biofortification (Chapter 9) in tackling global nutritional security challenges. Chapter 10 focuses on the essential role, bioinformatics plays in crop development. Chapter 11 examines the positive environmental impacts of genetically engineered crops, including reductions in pesticide use, more efficient water utilization, and the promotion of biodiversity. Chapter 12 explores how biotechnology can offer solutions to smallholder farmers, especially in developing countries where food insecurity is a major concern. Chapter 13 looks at the global debate surrounding CRISPR/Cas9-based genome editing technologies. Chapter 14 emphasizes the importance of integrating biotechnological innovations into broader sustainable agricultural practices. Chapter 15 explores the need for inclusive policies that ensure equitable access to biotech advancements for farmers. Chapter 16 tackles the controversies and opposition that still surround the global acceptance of biotech crops. With the rise of technologies such as big data, artificial intelligence, and the Internet of Things (IoT), Chapter 17 explores how digital tools can complement biotechnological innovations to create more precise, efficient, and sustainable agricultural practices. Finally, Chapter 18 explores the emerging field of nanobiotechnology and its potential role in addressing the challenges posed by climate change.

This book serves as a comprehensive resource that highlights how plant biotechnology can revolutionize agriculture and contribute to food security. By blending scientific research with economic considerations and environmental protection, it demonstrates how biotechnology can offer solutions to some of the most critical challenges facing the world today. Intended as a valuable guide for researchers, policymakers, farmers, and students, this book aims to inspire collaborative efforts to harness the power of biotechnology in building a more food-secure and sustainable future.

Jyoti Prakash Sahoo, PhD
Kailash Chandra Samal, PhD

About the Editors

Jyoti Prakash Sahoo is a dynamic researcher and academician currently serving as Assistant Professor in the Faculty of Agriculture and Allied Sciences at C.V. Raman Global University, Bhubaneswar, India. He completed his M.Sc. (2016) and PhD (2023) in Agricultural Biotechnology from Assam Agricultural University, Jorhat, India and Odisha University of Agriculture and Technology (OUAT), Bhubaneswar, India, respectively. His notable works include dissecting genetic architecture for *Cercospora* leaf spot and charcoal rot disease resistance in mung bean landraces, molecular breeding strategies for stress resilience in crops, sustainable management of viral pathogens in okra, and drought tolerance genetics in rice using microsatellite markers. He has guided ten M.Sc. (Biotechnology) students. He regularly publishes in high-impact journals indexed by Scopus, UGC-CARE, and Clarivate, while also authoring important chapters for widely referenced agricultural biotechnology books. He actively contributes as a peer reviewer for prestigious journals, including *Frontiers in Plant Science*, *BMC Genomics*, and *Scientific Reports*, and holds life memberships in professional societies, including the Society of Biotechnology and Bioinformatics and Indian Society of Plant Breeders. His outstanding contributions to plant molecular biology and biotechnology have been recognized with prestigious honors, including the young scientist award (2021), the university best PhD thesis award of OUAT (2023), and multiple best oral presentation awards (2023) at national and international conferences.

Kailash Chandra Samal was the former Dean of the College of Horticulture at Odisha University of Agriculture and Technology (OUAT), Chiplima, India. He holds an M.Sc. in Agricultural Biotechnology from Tamil Nadu Agricultural University, Coimbatore, India (1991), and a PhD in Biotechnology from Utkal University, Bhubaneswar, India (2011). With 32 years of academic and research experience at OUAT, he has taught undergraduate, postgraduate, and PhD students while conducting pioneering research in plant molecular biology, plant tissue culture, microbial technology, DNA fingerprinting, and computational biology. He has developed cost-effective protocols for the large-scale micropropagation of several crops, including banana, ginger, turmeric, sugarcane, stevia, and sarpagandha. His work includes the development of molecular marker techniques for assessing the clonal fidelity of micropropagated plants and the creation of a DNA fingerprint database for promising rice varieties in Odisha, India. He has studied the phenotypic, biochemical, and molecular characteristics of indigenous aromatic rice varieties from Odisha, India and molecular fingerprints of other crops such as ragi, wheat, rapeseed, sesame, black gram, green gram, eggplant, and tomato. He has guided 40 M.Sc. (Ag) and PhD students in molecular biology and biotechnology. He has also secured ten externally funded projects from Department of Biotechnology (DBT), Rashtriya Krishi Vikas Yojana (RKVY), and the Ministry of Agriculture, Government of India. A member of several professional bodies, he has published ten books and over 128 research papers. His research contributions also include the isolation and characterization of abiotic stress-related genes such as DREB, deposited in GenBank (NCBI), USA.

Contributors

Manish Kumar Agrawal
Department of Plant Pathology
Uttar Banga Krishi Viswavidyalaya
Cooch Behar, India

Shilpa Bahubalendra
Department of Agricultural Extension Education
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Solanki Bal
School of Smart Agriculture
Adamas University
Barrackpore-Barasat Rd, Barbaria, Kolkata, India

Tanya Barpanda
Department of Plant Breeding and Genetics
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Jeebanjyoti Behera
Department of Agricultural Extension Education
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Laxmipreeya Behera
Department of Agricultural Biotechnology
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Aninda Chakraborty
Centurion University of Technology and Management
Paralakhemundi, India

Surender Singh Chandel
Department of Agricultural Biotechnology
College of Agriculture
CSK Himachal Pradesh Agricultural University
Palampur, India

Manasi Dash
Department of Plant Breeding and Genetics
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Hema Deupa
Department of Food and Nutrition
School of Home Science
Babasaheb Bhimrao Ambedkar University
Lucknow, India

Shipa Rani Dey
Faculty of Agricultural Sciences
GLA University
Chaumuhan, India

Haragopal Dutta
Division of Genetics
ICAR-Indian Agricultural Research Institute
Pusa, New Delhi, India

Suman Dutta
Department of Genetics and Plant Breeding
Ramakrishna Mission Vivekananda Educational and Research Institute
Kolkata, India

Kanivalan Iwar
National Agrobiodiversity Centre
National Institute of Agricultural Sciences, Rural Development Administration
Jeonju, Republic of Korea

Desika J
Department of Genetics and Plant Breeding
V.O. Chidambaranar Agricultural College and Research Institute
Tamil Nadu Agricultural University
Tuticorin, India

Biswajit Jena
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Sonika Kalia
Department of Biotechnology
School of Applied and Life Sciences
Uttaranchal University
Dehradun, India

Dhanapati Keerthana
Department of Genetics and Plant Breeding
Rani Lakshmi Bai Central Agricultural University
Jhansi, India

Prasann Kumar
Department of Agronomy
School of Agriculture
Lovely Professional University
Phagwara, India

Renu Kumari
ICAR-National Institute for Plant Biotechnology
Pusa Campus, New Delhi, India

Kousalya Loganathan
Department of Botany
Nirmala College for Women
Red Fields, Coimbatore, Tamil Nadu, India

Binod Kumar Mahapatra
Department of Botany
Utkal University
Bhubaneswar, India

Abinash Mishra
Department of Plant Breeding and Genetics
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Amlan Mohanty
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Akankshya Mohapatra
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Deepali Mohapatra
Department of Plant Pathology
College of Agriculture
Odisha University of Agriculture and Technology,
Bhubaneswar, India

Sourav Ranjan Nanda
Department of Plant Breeding and Genetics
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Debarati Nandi
Department of Agricultural and Food Engineering
Indian Institute of Technology
Kharagpur, India

Sudhanya Nath
Fisheries and Animal Resources Development (F and ARD) Department
Block Veterinary Dispensary
Hemgir, Sundargarh, Odisha, India

Amruta Panda
Department of Botany
Utkal University
Bhubaneswar – 751004, India

Angelina Patro
Department of Agricultural Extension Education
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Subrat Pattanaik
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Samikshya Sankalini Pradhan
Department of Plant Pathology
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Jannila Praveena
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Arati Priyadarshini
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Sanghamitra Rout
Centurion University of Technology and Management
Paralakhemundi, India

Juliet Hepziba S
Department of Genetics and Plant Breeding
V.O. Chidambaranar Agricultural College and Research Institute
Tamil Nadu Agricultural University
Tuticorin, India

A Sheena Sabatina
ICAR-National Institute for Plant Biotechnology
Pusa Campus, New Delhi, India

Sree Vathsa Sagar US
Department of Genetics and Plant Breeding
Tamil Nadu Agricultural University
Coimbatore, India

Durgadatta Sahoo
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Jyoti Prakash Sahoo
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Lipikant Sahoo
Department of Plant Pathology
College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Pavitra Saini
Amity Institute of Forensic Sciences
Amity University
Noida, Uttar Pradesh, India

Basanta Kumar Samal
Department of Nematology, College of Agriculture
Odisha University of Agriculture and Technology
Bhubaneswar, India

Parul Pratyasa Samal
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Manikandan Selvarasu
Department of Botany, Govt. Arts and Science College
Tittagudi, Tamil Nadu, India

Barsharani Sethi
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Priyanka Shankar
Department of Human Development and Family Studies
School of Home Science
Babasaheb Bhimrao Ambedkar University
Lucknow, India

Gaurav Sharma
Department of Floriculture
Rani Lakshmi Bai Central Agricultural University
Jhansi, India

Isha Sharma
Department of Agricultural Biotechnology
College of Agriculture
CSK Himachal Pradesh Agricultural University
Palampur, India

Shivali Sharma
Department of Fruit Science
Rani Lakshmi Bai Central Agricultural University
Jhansi, India

Siddhartha Shankar Sharma
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Sunny Sharma
School of Agriculture
Lovely Professional University
Jalandhar, India

Vinay Sharma
Center of Excellence in Genomics and Systems Biology
International Crops Research Institute for the Semi-Arid Tropics (ICRISAT)
Patancheru, Hyderabad, India

Chintan Singh
Amity Institute of Forensic Sciences
Amity University, Noida, Uttar Pradesh, India

Jyoti Singh
Amity Institute of Forensic Sciences
Amity University
Noida, Uttar Pradesh, India

D. Swapna
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Swarnalata Tripathy
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Sachin Tripura
Department of ILFC (Animal Nutrition)
College of Veterinary Science and Animal Husbandry
Birsa Agricultural University
Ranchi, Jharkhand, India

Deepa Dharsini V
Department of Genetics and Plant Breeding
Tamil Nadu Agricultural University
Coimbatore, India

Dhaarani Vijayakumar
Faculty of Agriculture and Allied Sciences
C.V. Raman Global University
Bhubaneswar, India

Abbreviations

µM	micromolar
2,4-D	2,4-Dichlorophenoxyacetic acid (synthetic herbicide)
ABA	abscisic acid
ABM	advanced breeding methods
ABS	African Bio-fortified Sorghum Project
ACMV	African cassava mosaic virus
AFLP	amplified fragment length polymorphism
Ag NPs	silver nanoparticles
AGO	Argonaute
AGRA	Alliance for a Green Revolution in Africa
AHTEG-SEC	Ad Hoc Technical Expert Group on Socio-Economic Considerations
AI	artificial intelligence
Al_2O_3	aluminum oxide
AM fungi	arbuscular mycorrhizal fungi
amiRNA	artificial microRNA
APR	adult plant resistance
Au NPs	gold nanoparticles
Avr gene	avirulence gene (pathogen-derived)
B. amyloliquefaciens	*Bacillus amyloliquefaciens*
B. subtilis	*Bacillus subtilis*
BE	base editing
Bio-MnNPs	bio-functionalized manganese nanoparticles
BLAST	basic local alignment search tool
BPKP	Bharatiya Prakritik Krishi Paddhati
Bt	*Bacillus thuringiensis*
Ca^{2+}	calcium ion
Cas	CRISPR-associated protein
CAZyme	carbohydrate-active enzymes
CBD	Convention on Biological Diversity
CCP4	Collaborative Computational Project Number 4 (for crystallography)
CDs	carbon dots
CENH3	centromeric histone H3
CeO_2 NPs	cerium oxide nanoparticles
CGIAR	Consultative Group on International Agricultural Research
CH_4	methane
CjCas9	*Campylobacter jejuni* Cas9
CMV	cucumber mosaic virus
CNN	convolutional neural network
CO_2	carbon dioxide

CP	coat protein
CPB	Cartagena Protocol on Biosafety
CRISPR	clustered regularly interspaced short palindromic repeats
crRNA	CRISPR RNA
CrtB	phytoene synthase
crtI	carotene desaturase
Cry proteins	crystal proteins
CSA	climate-smart agriculture
CSPs	cold shock proteins
Cu-CNFs	copper carbon nanofertilizers
CuO	copper oxide
CYP6AE14	cytochrome P450 enzyme
DALYs	disability-adjusted life years
DArT	diversity arrays technology
dbEST	Database of Expressed Sequence Tags
DBT	Direct Benefit Transfer
dCAPS	derived Cleaved Amplified Polymorphic Sequence
DCL (Dicer-like)	Dicer enzyme family
DCL3	Dicer-like 3
DDBJ	DNA Data Bank of Japan
DFS	digital financial services
DGAT1	diacylglycerol acyltransferase 1
DH	doubled haploid
DHA	docosahexaenoic acid
DMR6	Downy Mildew Resistance 6
DMSP	dimethylsulfoniopropionate
DNA	deoxyribonucleic acid
DOX	DMSP-amine oxidase
DRO1	Deeper Rooting 1
DSB	double-strand break
dsRNA	double-stranded RNA
EBI	European Bioinformatics Institute
EFSA	European Food Safety Authority
eIF4E	eukaryotic translation initiation factor 4E
EMBL	European Molecular Biology Laboratory
ENA	European Nucleotide Archive
e-NAM	electronic national agricultural market
ENMs	engineered nanomaterials
ENPs	engineered nanoparticles
EPA	eicosapentaenoic acid
EPSPS	5-enolpyruvylshikimate-3-phosphate synthase
EU	European Union
F1, F2	filial generations (first and second generations in plant breeding)
FAAH	fatty acid amide hydrolase
FAO	Food and Agricultural Organization

FDA	Food and Drug Administration
Fe	iron
Fe_3O_4 NPs	iron oxide nanoparticles
Fe_3O_4	iron oxide
FnCas9	*Francisella novicida* Cas9
FRET	fluorescence resonance energy transfer
FT	FLOWERING LOCUS T
G × E × M	Genotype × Environment × Management
GAB	genomics-assisted breeding
GAFSP	Global Agriculture and Food Security Program
GAT	glyphosate acetyltransferase
GBLUP	genomic best linear unbiased prediction
GBS	genotyping by sequencing
GCG	Genetics Computer Group
GEAC	Genetic Engineering Appraisal Committee (India)
GFP	green fluorescent protein
GHG	greenhouse gas
GIS	geographic information system
GM Crops	genetically modified crops
GMO	genetically modified organism
GMP	good manufacturing practice
GNSS	Global Navigation Satellite System
GO	Gene Ontology
GOX	glyphosate oxidoreductase
GPS	Global Positioning System
gRNA	guide RNA
GS	genomic selection
GWAS	genome-wide association studies
H_2O_2	hydrogen peroxide
HA	humic acid
HDR	homology-directed repair (DNA repair pathway)
Hidden Hunger	micronutrient deficiencies
HI-Edit	haploid induction-assisted genome editing
HIGE	haploid induction via gametic embryogenesis
HIGS	host-induced gene silencing
HIR	haploid induction rate
HLB	huanglongbing (citrus greening disease)
HMGR	3-hydroxy-3-methylglutaryl-CoA reductase
HR	homologous recombination
HSP70	heat shock protein 70
HT Crops	herbicide-tolerant crops
ICT	Information and Communication Technology
IIRR	Indian Institute of Rice Research
IMGE	inducer-mediated genome editing
Indels	insertions/deletions
IoT	Internet of Things

IP	intellectual property
IPCC	Intergovernmental Panel on Climate Change
IPDM	integrated plant disease management
IPM	integrated pest management
IPR	intellectual property rights
IR	insect-resistant
ISAAA	International Service for the Acquisition of Agri-biotech Applications
ISSR	inter-simple sequence repeat
IVR	interactive voice response
JAM	Jan Dhan, Aadhaar, and Mobile
JRC	Joint Research Centre (EU)
K	potassium
KEGG	Kyoto Encyclopedia of Genes and Genomes
KOG	eukaryotic orthologous groups
KPL	kinesin protein-like
LCYB	lycopene beta-cyclase (gene)
LDH	layered double hydroxide
LHCII	light-harvesting complex II
LiDAR	Light Detection and Ranging
LLP	low-level presence
LMIC	low- and middle-income countries
LshCas13a	*Leptotrichia shahii* Cas13a
MAS	marker-assisted selection
Mg	magnesium
miRNA	microRNA
ML	machine learning
MMT	methionine S-methyltransferase
MMT	montmorillonite clay
Mn	manganese
mRNA	messenger RNA
MSNs	mesoporous silica nanoparticles
MWCNTs	multi-walled carbon nanotubes
N	nitrogen
N_2O	nitrous oxide
NCBI	National Center for Biotechnology Information
NCONF	National Centre for Organic and Natural Farming
NDVI	normalized difference vegetation index
NF-Y	nuclear transcription factor Y
NGO	non-governmental organization
NGS	next-generation sequencing
NHEJ	non-homologous end joining
NHX	Na+/H+ Exchanger
NILs	near-isogenic lines
NmeCas9	*Neisseria meningitidis* Cas9
NMNF	National Mission on Natural Farming

NMs	nanomaterials
NPR	nonexpressor of pathogenesis-related genes
NPs	nanoparticles
nptII	neomycin phosphotransferase II gene
NUE	nitrogen use efficiency
n-ZnO	zinc oxide nanoparticles
ODM	oligonucleotide-directed mutagenesis
OECD	Organisation for Economic Co-operation and Development
OFSP	orange-fleshed sweet potato
ONT	Oxford Nanopore Technologies
P	phosphorus
PA	phytic acid
PA	polyamines
PacBio	Pacific Biosciences
PAM	protospacer adjacent motif
PAT	phosphinothricin acetyltransferase
PCR	polymerase chain reaction
PDB	Protein Data Bank
PDH	proline dehydrogenase
PDR	pathogen-derived resistance
PE	prime editing
PEGRNA	prime editing guide RNA
PGPR	plant growth-promoting rhizobacteria
Phl	phenazine-1-carboxylate
PHS	pre-harvest Sprouting
PIR	Protein Information Resource
PKVY	Paramparagat Krishi Vikas Yojana
PpalEPIC8	*Phytophthora palmivora* cysteine protease inhibitor 8
ppm	parts per million
PPO	polyphenol oxidase
PPP	public-private partnership
pre-miRNA	Precursor microRNA
PRSV	*Papaya ringspot virus*
PsbS	Photosystem II Subunit S
PSTVd	potato spindle tuber viroid
PSY	phytoene synthase gene
PTGS	post-transcriptional gene silencing
PUFAs	polyunsaturated fatty acids
PYL	pyrabactin resistance-like
QPM	quality protein maize
QTL	quantitative trait loci
R&D	Research and Development
RACK1	receptor for activated C kinase 1
RAPD	random amplified polymorphic DNA
RdDM	RNA-directed DNA methylation

rDNA	recombinant DNA
RDR2	RNA-dependent RNA polymerase 2
RdRP	RNA-dependent RNA polymerase
RFLP	restriction fragment length polymorphism
R-genes	resistance genes
RISC	RNA-induced silencing complex
RNA	ribonucleic acid
RNAi	RNA interference
RNA-Seq	RNA sequencing
RNP	ribonucleoprotein
ROS	reactive oxygen species
RTF	restricted taxonomic functionality
Rubisco	ribulose-1,5-bisphosphate carboxylase/oxygenase
SaCas9	*Staphylococcus aureus* Cas9
SAR	synthetic aperture RADAR
SCAR	sequence-characterized amplified region
ScCas9	*Streptococcus canis* Cas9
SDC	S-methylmethionine decarboxylase
SDGs	sustainable development goals
SDN-1/2/3	site-directed nuclease (types 1, 2, and 3)
Se	selenium
SECs	socio-economic considerations
SES	socio-ecological systems
sgRNA	single guide RNA
SGS	second-generation sequencing
SINE	short interspersed nuclear element
SiO_2 NPs	silicon dioxide nanoparticles
siRNA	small interfering RNA
SmNAC	stress-responsive NAC transcription factor
SNP	single nucleotide polymorphism
snRNA	small non-coding RNA
SOS	salt overly sensitive
SpCas9	*Streptococcus pyogenes* Cas9
ssDNA	single-stranded DNA
SSNs	site-specific nucleases
SSR	simple sequence repeat
ssRNA	single-stranded RNA
SUN	scaling up nutrition (movement)
SWEET	sugars will eventually be exported transporters genes
SWIR	short-wave infrared
TALEN	transcription activator-like effector nuclease
TE	transposable element
TF	transcription factor
TGS	transcriptional gene silencing
TiNPs	titanium nanoparticles
TiO_2	titanium dioxide

TIR1	transport inhibitor response 1
tracrRNA	trans-activating CRISPR RNA
TRAP	target region amplification polymorphism
UAV	unmanned aerial vehicle
UB2	UBIQUITIN2
UN	United Nations
UniProt	Universal Protein Resource
USDA	United States Department of Agriculture
UTR	untranslated region
UV	ultraviolet
VAD	Vitamin A deficiency
V-ATPase	vacuolar-type ATPase
Vip proteins	vegetative insecticidal proteins
VIS	visible spectrum
VIS-SWIR	visible and short-wave infrared
VLP	virus-like particles
VRT	variable rate technology
WEMA	water efficient maize for Africa
WGR	whole-genome regression
WHO	World Health Organization
WRI1	WRINKLED1
ZBNF	zero budget natural farming
ZFNs	zinc finger nucleases
Zn NPs	zinc nanoparticles
ZNCPCs	zincated nanoclay polymer composites
ZnNPs	zinc nanoparticles

Taylor & Francis
Taylor & Francis Group
http://taylorandfrancis.com

1 Traditional Farming and Modern Biotechnology

Bridging the Gap for Sustainable Agriculture

Pavitra Saini, Jyoti Singh, and Chintan Singh

1.1 INTRODUCTION

Sustainable agriculture requires integrating modern biotechnology with traditional farming methods to address global food security and environmental challenges (Singh et al., 2024). Traditional practices such as crop rotation, intercropping, and organic composting enhance soil fertility, biodiversity, and carbon sequestration, reducing reliance on synthetic inputs (Shikha et al., 2024). However, with the global population projected to reach 9.7 billion by 2050, traditional methods alone are insufficient (Berners-Lee et al., 2018). Modern biotechnologies like CRISPR-Cas9, molecular breeding, and nanotechnology improve crop resilience, yield, and resource efficiency by minimizing environmental harm (Photos, 2023).

Combining these approaches can create sustainable agroecosystems that meet current and future food demands while preserving ecological balance (Anderson & Gipmans, 2024). A holistic framework merging traditional knowledge with biotechnology is essential for addressing climate change, food insecurity, and biodiversity loss. Biofortification and precision agriculture enhance crop nutrition and reduce waste, supporting global sustainability goals like the UN Sustainable Development Goals (SDGs) (Photos, 2023). Collaboration between farmers and scientists ensures culturally appropriate, adaptive solutions, empowering local communities while advancing equitable food systems (Photos, 2023). This integrated approach fosters resilience, sustainability, and equity, benefiting both local and global stakeholders (Singh et al., 2020). This chapter explores how bridging the gap between traditional farming knowledge and modern biotechnological advancements can pave the way for a sustainable agricultural future.

1.2 THE ESSENTIALS OF SUSTAINABILITY

Sustainability in agriculture ensures long-term food production while preserving ecosystems, minimizing resource depletion, and supporting farmer livelihoods. It balances environmental health, economic viability, and social equity through practices like soil conservation, efficient water use, biodiversity preservation, and responsible technology adoption. As the globe faces the problems of climate

DOI: 10.1201/9781003597797-1

change and dwindling resources, it is becoming increasingly necessary to adopt farming practices that not only increase output but also protect ecosystems and promote social fairness (Anderson & Gipmans, 2024). This complete approach includes a commitment to reducing greenhouse gas emissions, saving water, and increasing biodiversity in agricultural systems. Adopting a holistic approach to sustainable agriculture would ultimately result in a healthier planet and better livelihoods for everyone involved in the food production process (Liu et al., 2023).

1.2.1 Economic Resilience: The Foundations for a Sustainable Future

Economic sustainability in agriculture refers to farming practices that are both profitable and give farmers and food producers with enough revenue to support their families. Furthermore, financial success might encourage a wider adoption of sustainable practices (Campos et al., 2023). Diversification, which entails farming a variety of crops or livestock to lessen risks from disease, poor weather, and changing market conditions, is an important aspect in agricultural economic sustainability. Efficiency is also important, and by using effective agricultural practices like precision agriculture, producers can enhance earnings while decreasing costs (Nia et al., 2020). By fostering an environment in which farmers can succeed financially, it is not only improve the individual livelihoods but also the general health of the agricultural economy and sustainability (Anderson et al., 2016).

1.2.2 Sustainable Agriculture for Protecting Natural Resources

Various essential approaches help to achieve environmental sustainability in agriculture. Soil health is critical, and measures such as cover cropping and conservation tillage can efficiently preserve soil quality and reduce erosion. Cover crops are seeded during the off-season to improve soil structure, increase nutrient levels, and inhibit weed growth (He et al., 2020). Conservation tillage lowers soil disruption, improves moisture retention, and further reduces erosion. Water preservation is another crucial element of sustainable farming. Drip and precision irrigation minimize water waste by providing water directly to the roots of plants in a controlled way (Nia et al., 2020). Agroforestry involves integrating trees, crops, and livestock to establish varied ecosystems that promote biodiversity of plants and animals, while improving soil quality and carbon absorption. Furthermore, using fewer synthetic pesticides and fertilizers is vital for decreasing pollution and safeguarding natural resources. Integrating renewable energy sources like solar and wind power into farming practices decreases carbon emissions (He et al., 2020).

1.3 CONVENTIONAL FARMING VS. SUSTAINABLE AGRICULTURE

Conventional farming relies on synthetic fertilizers, pesticides, and intensive monoculture practices to maximize short-term yields, often leading to soil degradation, water pollution, and biodiversity loss. In contrast, sustainable agriculture emphasizes crop rotation, organic fertilizers, and agroecological methods to maintain soil health, conserve water, and reduce environmental harm. While conventional farming prioritizes high productivity and economic efficiency, sustainable agriculture focuses on long-term ecological balance, resilience, and food security. Studies show that sustainable practices can improve soil fertility, reduce greenhouse gas emissions, and enhance ecosystem services compared to conventional methods (Gomiero et al., 2011; Pretty, 2018).

Conventional agriculture, driven by economic principles, prioritizes high yields and financial profits through agrochemicals, large-scale tillage, and mechanization, often at the expense of soil health and ecosystem services (He et al., 2020). In contrast, sustainable agriculture focuses on ecological balance, integrating environment friendly practices like crop rotation, conservation tillage, and organic farming to enhance biodiversity and resource conservation. While conventional farming offers immediate high yields, it risks long-term soil degradation, nutrient loss, and pollution (Campos et al., 2023). Achieving true sustainability requires integrating technological advancements while maintaining ecological balance to support both present and future agricultural needs (Nia et al., 2020).

1.4 BIOTECHNOLOGY FOR SUSTAINABLE FARMING PRACTICES

Agricultural scientists face challenges like population growth, resource limitations, land loss, climate change, and environmental degradation. Biotechnology offers sustainable solutions by reducing pesticide and fertilizer use, enhancing high-yield and stress-tolerant crops, and improving soil, air, and water quality (Campos et al., 2023). It modifies plants, animals, and microbes to optimize agricultural sustainability, minimize artificial inputs, and maximize food production on limited land. It also aids in resource conservation, phytoremediation, and nutrient efficiency. Breakthroughs like genetically engineered rainbow papayas have saved Hawaii's papaya industry from collapse in the 1990s. These papayas are resistant to the devastating ringspot virus, preserving the fruit's natural taste and nutritional value while offering farmers with a disease-resistant, sustainable crop option (Anderson & Gipmans, 2024). Ongoing research aims to develop similar resistances in crops like tomatoes and potatoes (Nia et al., 2020).

1.4.1 Enhancement of Yield

Food security depends on increasing crop yields as the world's population rises. Key yield characteristics like photosynthesis, biomass distribution, inflorescence shape, stomatal regulation, nutrient efficiency, and resilience to environmental stressors can all be improved through genetic manipulation (Yu et al., 2024). A key enzyme in photosynthesis that transforms CO_2 into biomass i.e., Rubisco, has a big impact on output. But it also produces a harmful byproduct when it combines with O_2, which can cut yields by 20%–50% (Wei et al., 2022). Production can be increased by incorporating carbon-concentrating mechanisms from cyanobacteria into crops and increasing Rubisco activity through enhanced carboxylation capacity (Wei et al., 2022). Modifying the expression of genes implicated in non-photochemical quenching has been shown to improve yield and photoprotection (Singh et al., 2020). Additionally, transgenic plants have improved yields; for example, *zmm28* gene overexpression has raised maize yields without causing any harm (Yu et al., 2024).

1.4.2 NUTRIENT ASSIMILATION EFFICIENCY

Nutrient use efficiency (NUE) measures crop yield relative to nitrogen inputs, including fertilizers and soil nitrogen, and is crucial for sustainable agriculture by boosting productivity while reducing environmental harm. Enhancing NUE involves optimizing plant nitrogen uptake and metabolism through biotechnology. The genes *GS (Glutamine synthetase)*, *GOGAT (Glutamate synthase)*, *NRT (Nitrate transporter)*, and *AMT (Ammonium transporter)* play pivotal roles in nitrogen metabolism (Shikha et al., 2024). *GS* converts toxic ammonium into glutamine, while *GOGAT* recycles it into glutamate, forming the core of nitrogen assimilation. Meanwhile, *NRT* genes facilitate nitrate uptake from the soil, and *AMT* genes transport ammonium, ensuring efficient nitrogen acquisition. Beyond NUE, research focuses on enabling crops to fix atmospheric nitrogen, reducing reliance on synthetic fertilizers. Strategies include engineering symbiotic relationships between cereals and nitrogen-fixing bacteria (NFB) or directly transferring bacterial *nif* genes found in NFB like *Rhizobium* and *Azotobacter*, encoding the nitrogenase enzyme complex that converts atmospheric nitrogen (N_2) into ammonia (NH_3), enabling biological nitrogen fixation and reducing reliance on synthetic fertilizers in plants (Yu et al., 2024).

1.4.3 RESILIENCE AGAINST ABIOTIC AND BIOTIC CHALLENGES

1.4.3.1 Parasite Deterrence

The creation of insect-resistant transgenic plants marks a major advancement in agricultural biotechnology, resulting from extensive research conducted by both the public and private sectors (Sahoo et al., 2020). The most widely used transgenic plant includes cry genes derived from the *Bacillus thuringiensis* (Bt) bacterium. Several other genes, such as *betaine aldehyde dehydrogenase (BADH)*, *Vitreoscilla haemoglobin* (*Vgb*), *levansucrase gene (SaccB)*, *arrowhead proteinase inhibitor (API)*, *cysteine proteinase inhibitor (OC-I)*, *Nicotiana tabacum histidine kinase-1 (NTHK1)*, and *jasmonic ethylene responsive factor (JERF-36)*, have also been introduced to a range of crops (Yu et al., 2024). Moreover, transgenic cotton and maize have shown resistance to lepidopteran and coleopteran insect larvae, leading to increased crop yields, reduced production costs, and a decreased need for pesticides (Zheng et al., 2025).

1.4.3.2 Immunity to Viruses

Plant biotechnology has made significant strides in enhancing viral immunity in crops. One approach involves the overexpression of RNA-dependent RNA polymerase (RdRP) and RNA silencing genes (*DCL, AGO, RDR*), which strengthen the plant's RNA interference (RNAi) pathway to degrade viral RNA. Another strategy utilizes pathogen-derived resistance, where introducing viral coat protein genes (e.g., CP genes from *TMV* or *PRSV*) triggers an immune response without infection. Additionally, CRISPR-Cas9 genome editing has been employed to disrupt host susceptibility genes like *eIF4E* or *eIF(iso)4E*, which many viruses hijack for replication. Some genetically modified crops, such as virus-resistant rainbow papaya (expressing *PRSV CP* gene) and bean golden mosaic virus-resistant beans (using RNAi), demonstrate the success of these biotechnological interventions (Wang et al., 2023).

1.4.3.3 Endurance of Non-Biological Stress

About 70% of global crop production is affected by severe abiotic stressors, limiting productivity and leading to financial losses (Chen et al., 2021). To combat these challenges, biotechnology has identified and manipulated key stress-responsive genes, including osmoprotectant biosynthesis genes (*proline biosynthesis (P5CS), BADH*) for drought and salinity tolerance, heat shock proteins (*HSPs*) for thermotolerance, and ion transporters (*SOS1, NHX (Na+/H+ antiporters)*) for salt stress resilience (Ruuskanen et al., 2023). Additionally, transcription factors like *DREB*, *NAC*, and *WRKY* regulate multiple stress-responsive pathways, while antioxidant genes (*SOD, CAT, APX*) mitigate oxidative damage caused by abiotic stress (Wang et al., 2023). Biotechnological techniques, including tissue culture, marker-assisted selection, *in vitro* mutagenesis, and genetic transformation, have developed several plant varieties that withstand abiotic stress (Wang et al., 2023).

1.5 SUGARCANE BREEDING FOR STRESS TOLERANCE: CASE STUDY

Modern sugarcane breeding leverages molecular markers to enhance sucrose accumulation and stress resilience by targeting key genes and quantitative trait loci (QTL). Markers linked to *sucrose synthase (SuSy)*, *sucrose phosphate synthase (SPS)*, and *invertase (INV)* genes help to select high-sugar genotypes, while stress-responsive genes (*DREB, NAC, SOD*) are used to improve drought and salinity tolerance. Techniques like QTL mapping, GWAS (genome-wide association studies), and marker-assisted backcrossing (MAB) accelerate the development of elite cultivars with optimized yield and stress adaptation. For instance, markers associated with *P5CS* and *NHX* enhance salt tolerance, while *ERD (early dehydration-inducible)* genes improve drought resilience. Researchers can use techniques like genetic transformation and functional genomics to identify key regulatory genes and develop strategies to improve resilience to environmental challenges like drought and pests (Chavan et al., 2022). *Saccharum* germplasm has been extensively studied with markers like RAPDs, RFLPs, AFLPs, SNPs, TRAPs, SSRs, and ISSRs for genetic mapping, phylogenetics, and molecular-assisted selection (MAS) (Duan et al., 2021).

Microsatellite DNA markers have effectively identified genetic variations, aiding breeding strategies (Sahoo et al., 2019). Studies using SSR and TRAP markers have revealed key genetic similarities, such as a 70% similarity across accessions. Recent advancements, including fluorescence-labeled SSR markers with high-performance capillary electrophoresis, improve parental germplasm management (Zheng et al., 2025). Genetic mapping of *Saccharum* sp. using SSR, AFLP, and DArT markers has

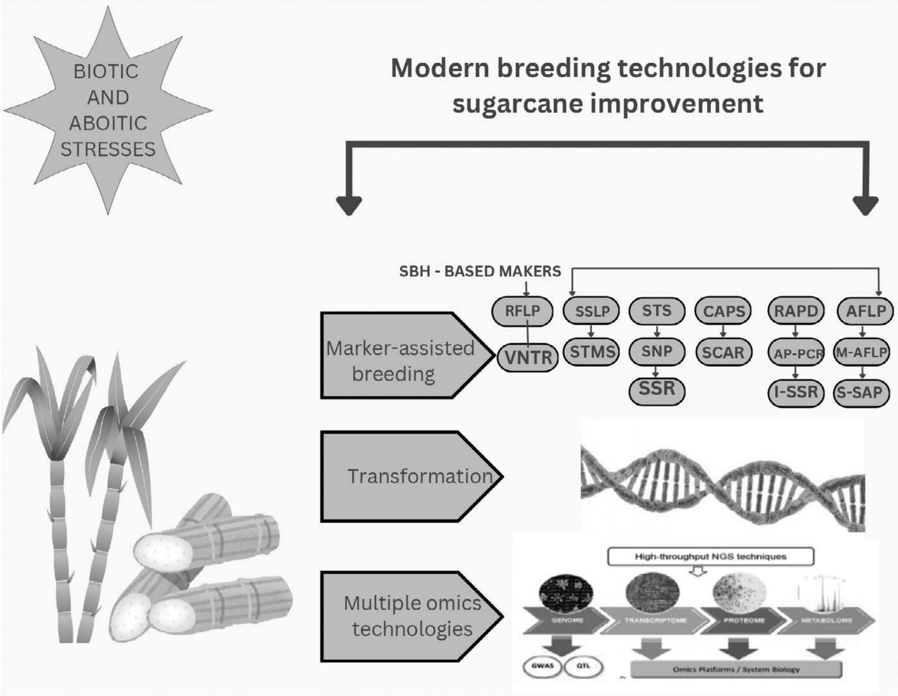


FIGURE 1.1 Contemporary breeding techniques for enhancing the genetic resilience of sugarcane to environmental stresses.

identified crucial genetic linkages and QTLs, enhancing crop genetic structure analysis and aiding cultivar improvement (Zheng et al., 2025). A contemporary breeding strategy for enhancing the genetic resilience of sugarcane to environmental stresses is indexed in Figure 1.1.

1.5.1 GENOMIC MARKERS FOR BIOTIC STRESS ADAPTATION

Several loci linked to disease resistance and yield have been successfully found by QTL mapping. Researchers have identified numerous QTLs associated with disease resistance in sugarcane, including 18 QTLs for yellow leaf virus resistance, 5 for Fiji leaf gall, and several others for leaf scald, pachymetra root-rot, and brown rust (through the *Bru1* gene) (Zheng et al., 2025). Additionally, 11 DNA markers have been linked to resistance against smut disease. QTL mapping using AFLP and SSR markers has also pinpointed a major QTL for resistance to the yellow leaf virus and a connection to yellow leaf spot disease resistance (Duan et al., 2021). For pest control, nine QTLs associated with resistance to the spotted-stem borer (*Chilo sacchariphagus*) in sugarcane were identified using AFLP, RFLP, and SSR markers, accounting for 6%–10% of observed trait variation (Wang et al., 2023).

1.5.2 GENETIC MARKERS PERTAINING TO ABIOTIC STRESS MANAGEMENT

Molecular markers play a vital role in identifying sugarcane genotypes resistant to abiotic stresses like drought and salinity, with techniques such as AFLP, RAPD, SSR, and SCAR markers proving effective in screening for tolerance. For instance, a SCAR marker derived from RAPD sequence *OPAK-12724* accurately identified 12 out of 23 drought-tolerant genotypes, while R-ISSR markers outperformed other methods in evaluating eight sugarcane varieties, revealing Co-997 as the most drought-resistant and Co-775 as the most susceptible. Additionally, AFLP primers further validated these findings, highlighting the challenges sugarcane faces as a glycophyte, where salt stress significantly hampers growth and productivity (Wang et al., 2023).

By effectively tracking genetic loci linked to salt resistance, molecular markers can lessen the need for laborious phenotypic analyses. PCR-based markers have been applied to evaluate the genetic diversity of sugarcane cultivars for salt tolerance (Wang et al., 2023). For example, by combining RAPD markers with *in vitro* mutagenesis, researchers identified salt-tolerant lines in *Saccharum officinarum*, highlighting genetic differences between these tolerant lines and control groups (Wang et al., 2023). Their ability to assess genetic variation among sugarcane cultivars was validated by screening with 15 ISSR markers. RAPD markers were used to characterize parent and mutant lines in tissue culture in order to find tolerant lines following exposure to salt stress and drought. Using five TRAP markers, molecular profiling of 18 sugarcane genotypes under salt stress revealed genetic heterogeneity (Duan et al., 2021).

1.6 NANOTECHNOLOGY-DRIVEN APPROACHES FOR CROP IMPROVEMENT

Crop breeding improves genetic traits through conventional and molecular methods, including whole-genome sequencing, while nanotechnology enables precise biochemical and nucleotide delivery into plant cells (Wang et al., 2023). Nanomaterials such as silica nanoparticles and mesoporous silica nanoparticles (MSNs) efficiently transfer DNA in crops like tobacco and maize, and modified techniques like particle bombardment enhance transgene expression with reduced cell damage (Samal et al., 2021). Magnetic nanoparticles deliver genes to cotton pollen without compromising viability, and carbon nanotubes facilitate robust Green Fluorescent Protein (GFP) expression in wheat and cotton, demonstrating their versatility in genetic engineering (Wang et al., 2023). Although CRISPR/Cas9 has advanced gene editing, challenges in delivery efficiency and homology-directed repair persist, prompting the use of nanomaterials like cationic gold nanoparticles and MSNs to improve precision, despite limitations in species adaptability and regeneration requirements (Wang et al., 2023). However, Figure 1.2 shows how nanotechnology aids crop breeding by delivering Cas and sgRNA genes into plant cells using methods like *Agrobacterium* or particle bombardment.

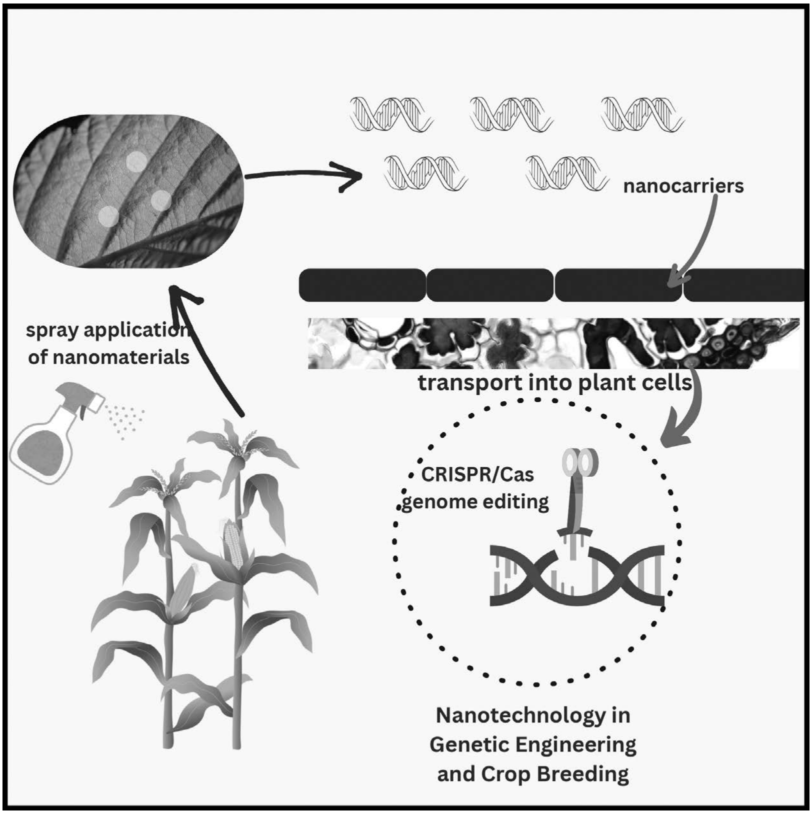


FIGURE 1.2 A diagram shows how nanotechnology aids crop breeding by delivering Cas and sgRNA genes into plant cells using methods like *Agrobacterium* or particle bombardment.

1.6.1 NANOTECHNOLOGY IN AGROCHEMICALS FOR ENHANCED CROP YIELD AND PROTECTION

The use of nanotechnology in agrochemicals is transforming crop protection and agricultural sustainability. Agrochemicals, which include fertilizers, insecticides, and plant growth regulators, are critical for increasing agricultural yield and quality. Farmers can obtain more precise delivery of pesticides by encapsulating them in nano-carriers, increasing the efficiency of active substances while reducing the overall quantity required (Duan et al., 2020). This focused method enhances pest management while simultaneously minimizing harm to non-target creatures, resulting in a healthier ecosystem (Duan et al., 2021).

1.6.1.1 Nanotechnology in Fertilizers: A New Era for Agricultural Efficiency

Nanotechnology in fertilizers enhances agricultural efficiency by improving nutrient delivery, reducing environmental impact, and boosting crop yields through targeted, nano-scale formulations (e.g., nano-encapsulated nutrients and slow-release nanoparticles) (Chen et al., 2021). Nano fertilizers enhance agricultural productivity and soil fertility by improving NUE and addressing eutrophication (Wang et al., 2023). They are classified into controlled-release, controlled-loss, and nanocomposite fertilizers, integrating essential nutrients for better crop absorption and soil health. Studies show that ZnO nanoparticles increased barley yield by 91%, compared to 31% with conventional fertilizers, while TiO_2 nanoparticles boosted phosphorus and potassium uptake by 34% and 35% in cucumber seedlings (Duan et al., 2021). Controlled-release fertilizers improve efficiency, increasing wheat yield by 6% and residual soil nitrogen by 10%, while reducing nitrogen leaching by 25% and runoff losses by 22% (Chen et al., 2021).

1.6.1.2 Nano Pesticides: Enhancing Efficacy and Sustainability in Agriculture

Nanotechnology has revolutionized crop protection by addressing the inefficiencies and environmental harm caused by traditional pesticide overuse, with approximately 90% of conventional pesticides failing to effectively target pests or contaminating ecosystems (Chen et al., 2021). Nano-encapsulation and nano-formulation technologies enable precise delivery of active pesticidal compounds, ensuring controlled release and sustained effectiveness while minimizing non-target impacts and pest resistance. These advanced formulations enhance key properties such as thermal stability, solubility, and biodegradability, as demonstrated by nanofiber pheromones for the oriental fruit moth, which maintain long-term efficacy without compromising safety (Chen et al., 2021).

1.6.1.3 Herbicides: Essential Tools for Modern Agriculture

Weeds spread aggressively beyond their natural habitat, requiring herbicides synthetic or biological compounds for control. While effective, excessive herbicide use can harm crops and pose health risks. Nanotechnology enhances herbicide efficiency by using nanoparticles to improve targeting, absorption, and controlled release. Encapsulation of herbicides like triazine and atrazine allows precise application, addressing both surface weeds and subsurface structures like rhizomes. Nano herbicides optimize biodegradability, solubility, and thermal stability and reduce losses (Chen et al., 2021).

1.6.1.4 Innovative Bactericides: Strategies for Effective Microbial Management

Bactericides are chemical substances, both synthetic and biological, that can kill bacteria. The incorrect use of bactericides has contributed to the creation of multi drug-resistant bacteria, which pose a large global danger and a serious challenge for agriculture. Nanotechnology-driven innovations hold promise for resolving this challenge (Srivastava et al., 2025). The efficiency of nanoparticles (NPs) is determined by their interactions with microbes, demanding a full understanding of both microbial biology and NP physicochemical qualities for successful development (Srivastava et al., 2025). Soil microbial communities' size and composition are significantly influenced by metallic oxide nanoparticles, such as CuO and Fe_3O_4 (Chen et al., 2021). An overview of nanotechnology applications in modern agriculture is indexed in Figure 1.3.

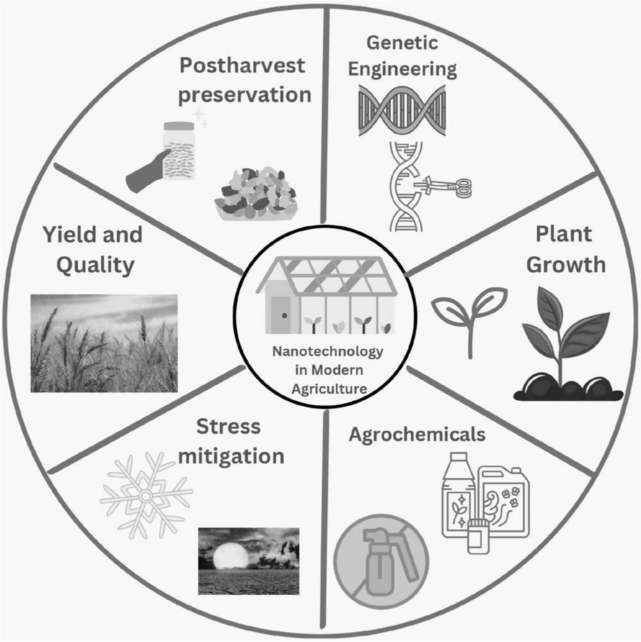


FIGURE 1.3 An overview of nanotechnology applications in modern agriculture, highlighting its role in enhancing yield, quality, stress resistance, and postharvest preservation.

1.6.2 ENHANCING CROP LIFE CYCLES WITH PHYTONANOTECHNOLOGY INNOVATIONS

Nanomaterials like MSNs, gold nanoparticles (Au NPs), silica nanoparticles (SiO_2 NPs), and chitosan nanoparticles have shown great potential for boosting crop growth and development. Studies have found these materials to be effective in supporting the growth of crops such as soybean, rice, wheat, peanut, tomato, potato, and onion, from the initial stages of seed germination right through to full maturity. The effectiveness of the used nanomaterials for plant growth depends on their concentration, content, size, and physical and chemical characteristics (Srivastava et al., 2025).

1.6.2.1 Seed Sprouting

Seed germination is the first and most important phase in a plant's life cycle. Zinc nanoparticles (Zn NPs) have been found to promote seed germination in various crops, including wheat, soybean, onion, and peanut. Likewise, treating seeds with metal oxide nanoparticles, such as silica (SiO_2) and titanium dioxide (TiO_2), has been shown to considerably enhance germination in select crops (Srivastava et al., 2025). Despite an increasing number of research, indicating the beneficial impacts of nanomaterials (NMs) on seed germination, the processes that make NMs more effective than standard materials are unknown (Chintan et al., 2024). However, Figure 1.4 illustrates how nanomaterials penetrate plant cell walls and membranes, facilitating targeted delivery of agrochemicals to cellular organelles.

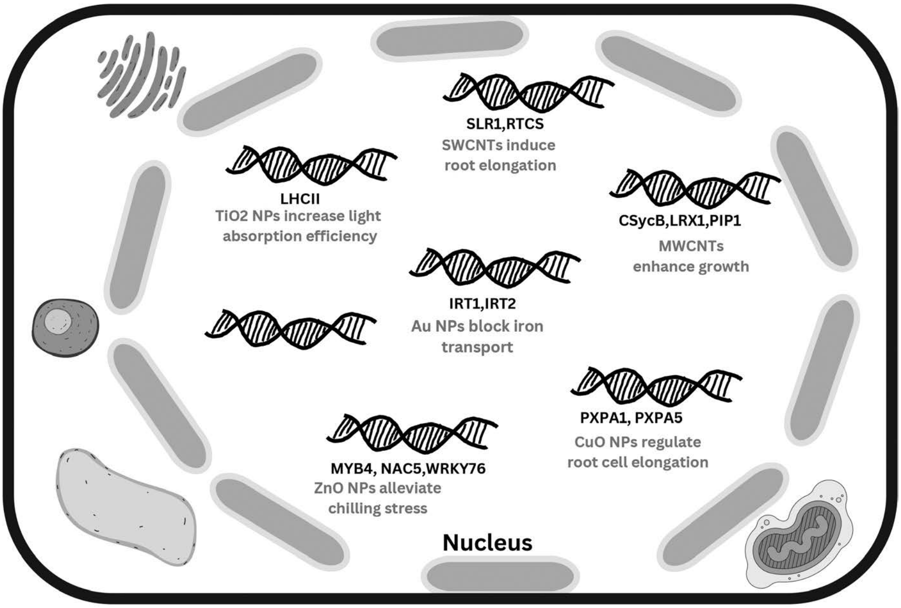


FIGURE 1.4 Nanomaterials interact at the molecular level within plant cells.

1.6.2.2 The Process of Photosynthesis

Nanomaterials can enhance photosynthesis by improving chlorophyll content, enzyme activity (such as Rubisco), and metabolic processes, with titanium dioxide and silica nanoparticles demonstrating significant potential in boosting plant growth and productivity (Srivastava et al., 2025). These advancements in nanotechnology offer promising solutions for increasing photosynthetic efficiency and crop yields (Chintan et al., 2024). Wheat seedlings treated with 0–5 g/L of SiO_2 nanoparticles showed the highest chlorophyll content and photosynthetic rates (Chen et al., 2021). Additionally, TiO_2 nanoparticles at a concentration of 60 mg/kg boosted wheat chlorophyll levels by 32.3% compared to untreated plants. In *Arabidopsis thaliana*, TiO_2 nanoparticles have also been found to activate the light-harvesting complex II (LHCII) gene, leading to enhanced chloroplast light absorption and increased LHCII content in the thylakoid membranes (Chen et al., 2021). Despite multiple studies demonstrating the beneficial benefits of NMs on seed germination, the processes underlying their superiority over standard materials are unknown and warrant additional exploration (Srivastava et al., 2025).

1.6.2.3 Yield Optimization

Nanoparticles (NPs) can modify plant physiology and biochemistry, affecting agricultural yields. For example, Fe_3O_4 NPs in maize assist maintain iron balance, minimize lipid peroxidation, and boost ferritin levels. Crops grown in soil treated with either hydrophilic or hydrophobic nano-TiO_2 for two months showed significant improvements in the production of inorganic carbohydrates, mineral uptake, chlorophyll content, enzyme activity, and overall plant growth (Srivastava et al., 2025). Similarly, increased crop yields have been linked to foliar applications of manganese (Mn), iron oxide (Fe_2O_3), and molybdenum (Mo) nanoparticles (Chen et al., 2021). Treatment techniques, traits, size, and type of NP used all effect how different NPs affect yield and crop quality. Further research into factors such as dose-dependence, long-term exposure, and molecular techniques like metabolomics or proteomics could provide clearer insights into the specific roles of nanoparticles in seed and fruit quality, as well as crop yield (Tian et al., 2021).

1.6.2.4 Quality Assessment

Phytonanotechnology enhances food quality by improving flavor, nutritional value, and bioavailability through nanotechnology applications. Metallic oxide nanoparticles like SiO_2 and TiO_2 serve as colorants and flow agents, while nano-encapsulation techniques enhance taste retention and controlled release (Chen et al., 2021). Encapsulating bioactive compounds, such as rutin in ferritin nanocages, increases solubility and thermal stability, allowing them to endure harsh digestive conditions (Srivastava et al., 2025). Techniques like nano-emulsification and nano-structuration further aid nutrient transfer, preserving antioxidants, proteins, and vitamins. Polymeric nanoparticles help to encapsulate sensitive micronutrients, offering significant health benefits through NP-based edible capsules for targeted nutrient and medication delivery (Tian et al., 2021).

1.6.2.5 Postharvest Preservation

Food safety is becoming more and more important as agricultural yields decline due to crop diseases, soil erosion, and climate change, and food demand increases. Effective packaging materials must be moisture and gas permeable, as well as biodegradable and robust (Tian et al., 2021). The incorporation of nanoparticles into food products or packaging can improve food preservation by increasing shelf life and decreasing spoilage. Nanoliposomes are good at transporting antibacterial chemicals and nutrients due to their small size (Chen et al., 2021). Nanocomposites are commonly utilized in food packaging because of their antifungal and antibacterial qualities. Silver nanoparticles (Ag NPs) effectively eliminate spoilage germs by disrupting cell membranes and DNA (Yudasaka et al., 2003).

1.6.3 ROLE OF PHYTONANOTECHNOLOGY IN ENHANCING STRESS RESILIENCE IN CROPS

Climate change causes temperature, salinity, droughts, and heavy metal contamination in the environment, all of which have a substantial impact on agricultural growth. Hormone regulation, plant enzymatic system activation, stress-related gene expression, water shortage stress management, and heavy metal uptake and translocation are all necessary components of a multimodal strategy to increase crop adaptability. Nanomaterial advances have the potential to boost crop yield in tough situations (Arora et al., 2018).

1.6.3.1 Navigating the Effects of Extreme Temperatures on Crop Growth

Nanoparticles (NPs) can help reduce these adverse effects, mainly by minimizing oxidative stress and the excessive production of reactive oxygen species (ROS) (Chen et al., 2021). NPs stimulate crop development at severe temperatures by altering a variety of physiological, biochemical, and molecular processes (Duan et al., 2020). TiO_2 NPs also improve cold stress resistance by maintaining carotenoid and chlorophyll levels, stimulating ascorbate peroxidase and catalase activity, and increasing the expression of genes associated to chlorophyll and Rubisco proteins (Singh et al., 2024). Similarly, cerium oxide (CeO_2) nanoparticles have been shown to increase.

1.6.3.2 Strategies for Mitigating Osmotic Stress in Agriculture

Excessive anion and cation buildup reduces productivity on one-fifth of cultivable land, but studies show SiO_2 NPs boost seed germination and chlorophyll content in stressed crops, while $FeSO_4$ NPs improve photosynthetic efficiency and reduce sodium levels in sunflowers. Phytonanotechnology also addresses drought stress by increasing water retention and seed absorption. Moreover, CeO_2 and micronutrient NPs have shown promising results in improving drought resistance in barley and soybean (Chen et al., 2021).

1.6.3.3 Mitigating Heavy Metal Contamination in Crop Production

Rapid urbanization and industrialization have significantly contributed to soil degradation, with heavy metals like arsenic (As), mercury (Hg), and cadmium (Cd) being major pollutants (Tian et al., 2021). Phytonanotechnology offers an effective remediation approach by reducing ROS and oxidative stress caused by heavy metals, thereby limiting their accumulation in crops (Rani et al., 2021b). For instance, silicon nanoparticles (Si NPs) enhance cadmium tolerance in rice by mitigating ROS production, while zinc oxide nanoparticles (ZnO NPs) reduce Cd uptake in wheat and rice (Singh et al., 2020). Gold nanoparticles (Au NPs) derived from melatonin further alleviate Cd stress in rice by regulating metal transporter genes (Parveen et al., 2023).

1.7 FUTURE PROSPECTS

Traditional farming, rooted in agroecological practices, emphasizes biodiversity, organic inputs, and local knowledge, while modern biotechnology offers precision tools like CRISPR, genetically modified organisms (GMOs), and biofertilizers to enhance productivity sustainably. Integrating these approaches can address global challenges such as climate change, soil degradation, and food insecurity by combining the resilience of traditional systems with the efficiency of biotech innovations (Altieri & Nicholls, 2017). For instance, drought-resistant GM crops can complement traditional water-saving techniques, and microbial biofertilizers can reduce dependency on synthetic inputs while maintaining soil health (Bender et al., 2016). Future prospects include AI-driven agroecology, nanotechnology-enhanced organic fertilizers, and gene-edited crops tailored for smallholder farms, ensuring sustainability without sacrificing yield (Pretty, 2018). However, ethical, regulatory, and socio-economic barriers must be addressed to ensure equitable adoption and avoid displacing small-scale farmers (Rani et al., 2021a).

1.8 CONCLUSION

Combining traditional farming methods with modern biotechnology offers a holistic approach to tackle climate change, environmental sustainability, and global food security. Traditional practices like crop rotation, organic composting, and intercropping enhance soil health, biodiversity, and water conservation, while modern biotechnologies such as genetic engineering, precision farming, and nanotechnology improve crop resilience, resource efficiency, and yield stability. This integration not only boosts agricultural productivity and economic stability but also minimizes environmental impact. This strategy fosters a resilient, sustainable, and equitable farming system capable of meeting current and future food demands while preserving ecosystems and addressing climate challenges.

REFERENCES

Altieri, M. A., & Nicholls, C. I. (2017). The adaptation and mitigation potential of traditional agriculture in a changing climate. *Climatic Change*, *140*(1), 33–45. https://doi.org/10.1007/s10584-013-0909-y

Anderson, J., & Gipmans, M. (2024). Emerging agricultural biotechnologies for sustainable agriculture and food security. ACS Publications. https://pubs.acs.org/doi/abs/10.1021/acs.jafc.5b04543

Anderson, J. A., Gipmans, M., Hurst, S., Layton, R., Nehra, N., Pickett, J., Shah, D. M., Souza, T. L. P. O., & Tripathi, L. (2016). Emerging agricultural biotechnologies for sustainable agriculture and food security. *Journal of Agricultural and Food Chemistry*, *64*(2), 383–393. https://doi.org/10.1021/acs.jafc.5b04543

Arora, A., Banerjee, J., Vijayaraghavan, R., MacFarlane, D., & Patti, A. F. (2018). Process design and techno-economic analysis of an integrated mango processing waste biorefinery. *Industrial Crops and Products*, *116*, 24–34. https://doi.org/10.1016/j.indcrop.2018.02.061

Bender, S. F., Wagg, C., & van der Heijden, M. G. A. (2016). An underground revolution: Biodiversity and soil ecological engineering for agricultural sustainability. *Trends in Ecology and Evolution*, *31*(6), 440–452. https://doi.org/10.1016/j.tree.2016.02.016

Berners-Lee, M., Kennelly, C., Watson, R., & Hewitt, C. N. (2018). Current global food production is sufficient to meet human nutritional needs in 2050 provided there is radical societal adaptation. *Elementa*, *6*, 52. https://doi.org/10.1525/elementa.310

Campos, E. V., Ratko, J., Bidyarani, N., Takeshita, V., & Fraceto, L. F. (2023). Nature-based herbicides and micro-/nanotechnology fostering sustainable agriculture. *ACS Sustainable Chemistry and Engineering*, *11*(27), 9900–9917. https://doi.org/10.1021/acssuschemeng.3c02282

Chavan, S., Yadav, B., Atmakuri, A., Tyagi, R. D., Wong, J. W. C., & Drogui, P. (2022). Bioconversion of organic wastes into value-added products: A review. *Bioresource Technology*, *344*, 126398. https://doi.org/10.1016/j.biortech.2021.126398

Chen, J., Yu, Q., Patterson, E., Sayer, C., & Powles, S. (2021). Dinitroaniline herbicide resistance and mechanisms in weeds. *Frontiers in Plant Science*, *12*, 634018. https://doi.org/10.3389/fpls.2021.634018

Chintan, S., Himanshu, K., & Prakash, N. B. (2024). Sustainable transformation of biomass into diverse high-value materials using biorefineries in the circular economy. In *Advances in energy from waste* (pp. 343–369). Woodhead Publishing. https://doi.org/10.1016/B978-0-443-13847-8.00010-5

Duan, Y., Mehariya, S., Kumar, A., Singh, E., Yang, J., Kumar, S., Li, H., & Awasthi, M. K. (2021). Apple orchard waste recycling and valorization of valuable product—A review. *Bioengineered*, *12*(1), 476–495. https://doi.org/10.1080/21655979.2021.1872905

Duan, Y., Pandey, A., Zhang, Z., Awasthi, M. K., Bhatia, S. K., & Taherzadeh, M. J. (2020). Organic solid waste biorefinery: Sustainable strategy for emerging circular bioeconomy in China. *Industrial Crops and Products*, *153*, 112568. https://doi.org/10.1016/j.indcrop.2020.112568

Gomiero, T., Pimentel, D., & Paoletti, M. G. (2011). Environmental impact of different agricultural management practices: Conventional vs. organic agriculture. *Critical Reviews in Plant Sciences*, *30*(1–2), 95–124. https://doi.org/10.1080/07352689.2011.554355

He, S., Zhu, Q., Soomro, R. A., & Xu, B. (2020). MXene derivatives for energy storage applications. *Sustainable Energy and Fuels*, *4*(10), 4988–5004. https://doi.org/10.1039/d0se00927j

Liu, I. H., Lee, M. H., Huang, H. C., & Li, J. S. (2023). 5G-based smart healthcare and mobile network security: Combating fake base stations. *Applied Sciences*, *13*(20), 11565. https://doi.org/10.3390/app132011565

Nia, A. R., Awasthi, A., & Bhuiyan, N. (2020). Management of sustainable supply chain and Industry 4.0: A literature review. In *Sustainable supply chains: Strategies, issues, and models* (pp. 1–47). Springer. https://doi.org/10.1007/978-3-030-48876-5_1

Parveen, R., Kumar, M., Swapnil, N., Singh, D., Shahani, M., Imam, Z., & Sahoo, J. P. (2023). Understanding the genomic selection for crop improvement: Current progress and future prospects. *Molecular Genetics and Genomics*, *298*(4), 813–821. https://doi.org/10.1007/s00438-023-02026-0

Photos, A. (2023). *Bridging the gap: Accelerating technology adoption for sustainable food production*. Global Affairs. https://globalaffairs.org/sites/default/files/2023-10/BTG%20RT2%20Prepublication%20Copy.pdf

Pretty, J. (2018). Intensification for redesigned and sustainable agricultural systems. *Science*, *362*(6417), eaav0294. https://doi.org/10.1126/science.aav0294

Rani, M. S. S., Singh, C., & Mishra, A. (2021a). Development of an analytical method for detection of Imidacloprid Insecticide from Biological Matrix using LC-MS/MS. *Journal of Punjab Academy of Forensic Medicine & Toxicology*, *21*, 220–225. https:///doi.org/10.5958/0974-083x.2021.00042.x

Rani, S., Singh, V., Sharma, M. K., & Sisodia, R. (2021b). GC–MS based metabolite profiling of medicinal plant-Catharanthus roseus under cadmium stress. *Plant Physiology Reports*, *26*, 491–502. https://doi.org/10.1007/s40502-021-00595-z

Ruuskanen, S., Fuchs, B., Nissinen, R., & Puigbò, P. (2023). Ecosystem consequences of herbicides: The role of microbiome. *Trends in Ecology and Evolution*, *38*(3), 201–215. https://doi.org/10.1016/j.tree.2022.09.004

Sahoo, J. P., Behera, L., Sharma, S. S., Praveena, J., Nayak, S. K., & Samal, K. C. (2020). Omics studies and systems biology perspective towards abiotic stress response in plants. *American Journal of Plant Sciences*, *11*(12), 2172. https://doi.org/10.4236/ajps.2020.1112152

Sahoo, J. P., Sharma, V., Verma, R. K., Chetia, S. K., Baruah, A. R., Modi, M. K., & Yadav, V. K. (2019). Linkage analysis for drought tolerance in kharif rice of Assam using microsatellite markers. *Indian Journal of Traditional Knowledge*, *18*(2), 371–375. https://core.ac.uk/reader/297999240

Samal, K. C., Sahoo, J. P., Behera, L., & Dash, T. (2021). Understanding the BLAST (Basic local alignment search tool) Program and a step-by-step guide for its use in life science research. *Bhartiya Krishi Anusandhan Patrika*, *36*(1), 55–61. http://dx.doi.org/10.18805/BKAP283

Shikha, D., Sindhura, K., & Reddy, M. (2024). A review on propelling agricultural practices with biotechnology into a new era. *Journal of Advanced Agricultural Technologies*, *11*(2), 45–60. https://journaljabb.com/index.php/JABB/article/view/725

Singh, C., Rani, M. S. S., & Mishra, A. (2020). Development of analytical method for detection of monocrotophos insecticide from biological matrix using LC-MS/MS. *International Journal of Medical Toxicology and Legal Medicine*, *23*(1-2), 210–219. https://doi.org/10.5958/0974-4614.2020.00031.5

Singh, K., Ishar, A., Singh, S., & Changdeo, W. (2024). The role of biotechnology in shaping the future of modern agriculture. *Journal of Advances in Biology and Biotechnology*, *27*(11), 621–634.

Srivastava, R. K., Panda, R. K., & Chakraborty, A. (2025). Sustainable agricultural engineering: Integrating science, technology, and practical applications. In *Mitigation and adaptation strategies against climate change in natural systems* (pp. 3–27). Springer Nature Switzerland.

Tian, Z., Wang, J. W., Li, J., & Han, B. (2021). Designing future crops: Challenges and strategies for sustainable agriculture. *The Plant Journal*, *105*(5), 1165–1178. https://doi.org/10.1111/tpj.15107

Wang, S., Sun, Z., Cao, W., Liu, H., Aioub, A. A., Hu, Z., & Wu, W. (2023). Establishment of Agrobacterium rhizogenes-mediated RNAi of Phryma leptostachya accelerates the functional identification of key genes of the furofuran lignan biosynthetic pathway. *Industrial Crops and Products*, *204*, 117260. https://doi.org/10.1016/j.indcrop.2023.117260

Wei, S., Li, X., Lu, Z., Zhang, H., Ye, X., Zhou, Y., & Zhou, W. (2022). A transcriptional regulator that boosts grain yields and shortens the growth duration of rice. *Science*, *377*(6604), eabi8455. https://doi.org/10.1126/science.abi8455

Yu, K., Zhao, S., Sun, B., Jiang, H., Hu, L., Xu, C., & Qi, Z. (2024). Enhancing food production through modern agricultural technology. *Plant, Cell and Environment*, *47*(3), 789–805. https://doi.org/10.1111/pce.15299

Yudasaka, M., Ajima, K., Suenaga, K., & Iijima, S. (2003). Nano-extraction and nano-condensation for C60 incorporation into single-wall carbon nanotubes in liquid phases. *Chemical Physics Letters*, *380*(1–2), 42–46. https://doi.org/10.1016/j.cplett.2003.08.095

Zheng, H., Hua, M., Jiang, M., Jiang, C., Xi, Y., Deng, J., & Zhou, S. (2025). Transgenic expression of mAChR-C dsRNA in maize confers efficient locust control. *Plant Communications*, 101316. https://doi.org/10.1016/j.xplc.2025.101316

2 Genetically Modified Crops for Climate Change Adaptation

Hema Deupa and Priyanka Shankar

2.1 INTRODUCTION

Genetically modified (GM) crops are those plants whose genomes have been genetically altered to enhance the pre-existing qualities or add novel traits that are inherently silent in the crop species. The plants which are produced by inserting a particular foreign gene or nucleic acid sequence into their genome (e.g. *Agrobacterium*-mediated transformation) are known as transgenic modification (Griffiths et al., 2005). The process of introducing particular genes or alleles from the gene pool into new types without altering the deoxyribonucleic acid (DNA) sequence is known as "cisgenesis" (Schouten et al., 2006). Conversely, intragenic modifications involve the exchange of genetic components from other plants to integrate coding regions containing promoters and terminators of distinct gene pools having the same sexual compatibility. The first GM crop developed using antisense technology was the Flavr Savr tomato introduced in 1994 and after that, more than 100 GM organisms (GMOs) have been licensed globally for commercial foods or feeds (Redenbaugh et al., 1992). In GM crops, *Bacillus thuringiensis* (Bt) corn transmits gene variants of Cry proteins from soil bacterium (Bt). If ingested this protein contains Bt toxins, which are particularly effective at eliminating significant plant pests such as Lepidoptera, Coleoptera, Diptera, and others. Bt is a spore-forming Gram-positive bacteria that have long usage as a safe biopesticide and has entomopathogenic properties (Trapero et al., 2016).

The majority of parasporally generated crystals are built up of one or more proteins (Cyt or Cry toxins), also referred to as δ-endotoxins, which impair the insect midgut epithelial lining by implanting the pores into the plasma membrane. Cry toxins are also innocuous to plants, animals, and human beings and are entirely decomposable (Tabashnik et al., 2003). Insect resistance and herbicide tolerance (HT) are the most common GM traits in GM crops (Koul et al., 2024). Some commercially approved genetically modified crops are indexed in Figure 2.1. The most popular GM crops in the market are soybean, canola, maize, and cotton. India and China are the leading producers of GM Bt cotton (*Gossypium hirsutum*). Bollworm is the most common pest of cotton, followed by boll weevil, and pink bollworms. Bt cotton MON531 lepidopteran-resistant variety expresses *Cry1Ac* in the cotton plant. The first GM crop authorized by the European Union in 1988 for commercial cultivators was Maize MON810 (Randhawa et al., 2016) and in 2010, high amylopectin

DOI: 10.1201/9781003597797-2

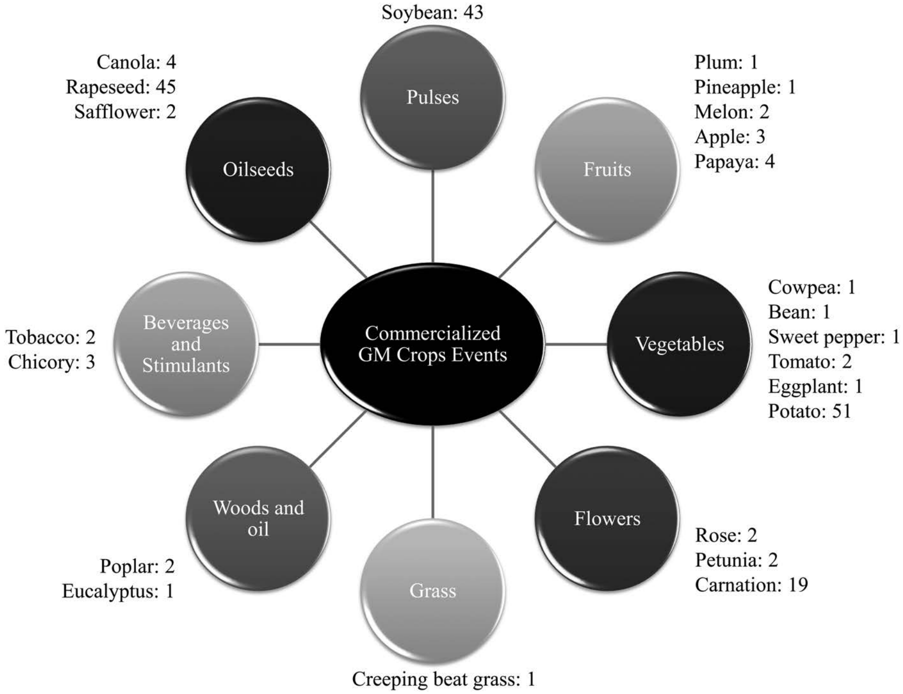


FIGURE 2.1 Commercially approved genetically modified crops.

content-rich AMFLORA potato EH92-527-1 was approved. However, AMFLORA was withdrawn from the European Union market due to procedural problems during the clearance procedure, and it received huge criticism for having the antibiotic resistance gene *nptII*. GMO Regulation 1829/2003 on GM food and feed is covered by Directive (2001/18/EC), while Regulation 1830/2003 is concerned with the traceability and labeling of GMOs.

Despite biosafety and environmental concerns, transgenic technology has proven to be a good choice for the rapid generation of an enhanced crop variety with multiple traits. The ISAAA annual report 2020 quantified the global state of GM/Biotech crops, in which 29 countries cultivated 190.4 million hectares of Biotech crops. A minor decrease of 1.3 million hectares of crops was observed. Countries like the USA, Brazil, Argentina, Canada, and India, the top five countries planted 91% of the world's biotech crops. In totality, the crop production of the USA, Brazil, Argentina, Canada, and India was 71.5, 52.8, 24.0, 12.5, and 11.9 million hectares, respectively (Pallett, 2021). A TELA maize, Golden rice, TR-4 resistant banana, yield-enhanced eucalyptus, and glowing petunia are some of the latest GM crops commercially approved for propagation (Tome et al., 2024). This chapter aims to summarize the techniques of GMOs, the variety of transgenic crops and their nutritional improvement, concerns of transgenic crops, and approaches to combat climate change.

2.2 TECHNIQUES FOR GENETICALLY MODIFIED ORGANISM (GMO) DEVELOPMENT

Genetically engineering techniques are used to develop GMOs that are categorized into transgenesis, cisgenesis, intragenesis, and genome editing, depending on the source of the gene of interest (Tome et al., 2024).

2.2.1 Transgenesis

This technique involves transferring exogenous genes (transgenes) between organisms either from unrelated species or distinct plants using genetic engineering methods like *Agrobacterium*-mediated transformation or gene guns, often to introduce drought resilience. The process typically employs restriction enzymes to isolate target genes, which are then inserted directly or via vectors into recipient organisms to confer desired traits. In the case of tobacco, *Nicotiana protein kinase 1* gene is transferred to maize resulting in the expression of heat shock proteins, which protects the photosynthetic mechanism of plants and their yield under drought and stress conditions (Shou et al., 2004). The microRNAs are regulatory molecules that bind to *Agronaute* proteins, identify target mRNA sequence, and act as negative regulators for transcription and translation, and yet cause expression of the characteristic traits. Transgenic barley that receives miR827 from *Arabidopsis* increases the water efficiency and performance of barley under drought conditions (Ferdous et al., 2017).

2.2.2 Cisgenesis

Advances in genome sequencing have led to the isolation of plant genes that are responsible for particular traits in crossable plant species, and are known as "cisgene". In normal orientation, cisgene contains the introns and exons with promoter and terminator sequences. Schouten et al. (2006) stated that the cisgenesis technique is a cutting-edge technique for GM crops with one or more desired genes extracted from the same species or crossable donor, where the transfer of a particular gene is performed via a gene pool of crop species to a new variety without the transmission of associated gene (linkage drag) with the desired gene (Templeton et al., 2021). A cisgenic transformation can be obtained with the help of *Agrobacterium* as a vector to transfer a gene of interest to the recipient plant. The genes which confer the plants to survive under stressful conditions can be transmitted to the capable cultivators to improve the drought tolerance potential of different crops. Overexpression of gene "*vacuolar pyrophosphatase 1*" has imparted higher drought tolerance against forage crops and ryegrass using *Agrobaterium* as the vector (Templeton et al., 2021).

2.2.3 Intragensis

Intragenesis is the process of producing an *in vitro* asynthetic gene by combining distinct components, such as the promoter, coding region, and termination sequence of the same species or other genes, and then introducing it into identical species or cross-compatible species. Ribonucleic acid (RNA) interference can be employed to silence any gene or to visualize a new phenotype in the existing species. Unlike conventional breeding, this produces the same results as mutant breeding. For instance, drought tolerance was acquired through overexpression of the *Lpvp1* gene in perennial ryegrass (Holme et al., 2012). This technique has a few limitations, as it can modify the plant genome proteins, which raises safety concerns (Holme et al., 2012).

2.2.4 GENOME EDITING

Genome editing or gene editing, is a technique which allows the DNA sequence of an organism to be changed by either knocking out a specific gene or altering the sequence of the specific base at a particular site throughout the whole genome. To attain this precise gene edition various technologies are used, including zinc-finger nucleases, transcription activator-like effector nucleases, and clustered regularly interspaced short palindromic-repeats-associated protein (CRISPR-Cas) to alter/modify one or more genes (Kumar et al., 2020). Genome editing is a predominant tool that causes the DNA repair machinery inside the cell. These technologies can be successfully implemented in the various sectors of agricultural areas as a revolutionary change (Das et al., 2023). CRISPR technology modifies target DNA sequences by inducing double-strand breaks that activate cellular repair mechanisms either through homologous recombination or non-homologous end joining to achieve gene knockout or epigenetic changes via chromatin remodeling (Kumar et al., 2020).

CRISPR also helps in breaking linkage drag for introducing certain genes and stacking beneficial genes under the same background to produce non-interfering lines (NILs) (Kumar et al., 2020). Moreover, CRISPR-Cas has the ability to simultaneously modify the genome at various target locations beyond discrimination. Though transgenic crops are not widely recognized by the society due to ethical issues and cost-effectivity, with the use of CRISPR, we can get rid of these issues, hence making it widely accepted (Kumar et al., 2020). Genome editing holds considerable possibility to reduce the impact of global warming on agricultural practices. For example, targeting stomatal density alleviates the drought tolerance ability of rice crops (Yin et al., 2017). To improve drought tolerance ability and higher yield, a promoter is also knocked in before a gene in maize (Li et al., 2019).

2.3 SAFETY ASSESSMENT OF GMOs

Safety assessments evaluate the effects of GMOs and their byproducts on non-target organisms and ecosystems by analyzing the recipient organism, donor gene source, transformation process, introduced gene products (proteins/metabolites), and food safety of GM-derived products (Liang, 2016). One of the major issues with GM crops is non-targeted organisms and breakdown of resistance, hazards associated with human health, and environmental concerns (Kumar et al., 2020). Regulatory agencies like the FDA and EFSA require rigorous testing, including compositional analysis and animal feeding studies, to ensure GMOs are as safe as their conventional counterparts (EFSA, 2011). Long-term studies and meta-analyses have found no significant differences in health risks between GMOs and non-GMOs, though continuous monitoring is recommended (Nicolia et al., 2014). Environmental assessments focus on gene flow, biodiversity impacts, and resistance development, with mitigation strategies like refuge requirements for Bt crops (NASEM, 2016).

2.4 TRANSGENIC CROPS

2.4.1 Herbicide-Tolerant Transgenic Crops

Weeds compete with crops for nutrients, water, sunlight, and space, causing significant crop losses, and while herbicides are essential for weed management, selectively targeting weeds without harming crops is difficult. Cultivating HT traits in crops allows the use of broad-spectrum herbicides like glyphosate, which inhibits the EPSPS enzyme in the shikimate pathway—a process absent in animals, making it safe for humans and wildlife. The process of creating transgenic crops that are resistant to glyphosate is dependent on the heterologous expression of a glyphosate-insensitive form of EPSPS i.e., either produced from a mutant version of maize *epsps grg23* gene of *Arthrobacter globiformis, or Agrobacterium tumefaciens* strain coat protein-4 (*CP4*) (Barry et al., 1997).

The first HT transgenic crop that has been commercialized was glyphosate-tolerant “Roundup Ready” soybean harboring *cp4esps* gene. This gene is present in the majority of commercialized glyphosate-resistant crops (Dill et al., 2008). Some commercially existing transgenic crops carry *glyphosate oxidoreductase (GOX)* and *glyphosate acetyltransferase (GAT)* genes, which are obtained from *Bacillus licheniformis* and *Ochrobactrum anthropi*, respectively. Both of these glyphosate-degrading enzymes detoxify glyphosate and convert it into harmless byproducts.

Glufosinate or phosphinothricin is another non-selective herbicide that inhibits the glutamine synthetase enzyme in a competitive approach (Lea et al., 1984). When glufosinate inhibits this enzyme, it contributes to the conversion of glutamate and ammonia into glutamine, ammonia builds up and photosystem I and II functions are hindered (Sauer et al., 1987; Tachibana et al., 1986). *PAT* and *BAR*, two distinct bacterial genes from *Streptomyces* species were utilized to create glufosinate-resistant crops. The *phosphinothricin acetyl transferase (PAT)* enzyme detoxifies herbicides like glufosinate through acetylation, while newer herbicides such as 2,4-D, dicamba, and sulfonylurea have also entered the market (ISAAA 2019). As of 2018, 351 HT events have been approved, with the majority in soybean, maize, and canola, making HT transgenic crops the most widely cultivated GM crops globally (ISAAA 2019). Farmers benefit from HT crops through enhanced weed control, higher yields, and reduced weed management costs (Brookes & Barfoot, 2018).

2.4.2 Insect-Resistant Transgenic Crops

Insect pests and diseases severely impact crop production, with over 67,000 damaging species that harm plants directly or transmit pathogens, prompting heavy reliance on costly, environmentally harmful chemical pesticides. GM crops incorporating insecticidal genes (primarily *cry* and *vip* variants) have emerged as a sustainable solution, with ten insect-resistant transgenic crops already commercialized to reduce pest damage and pesticide use (Keresa et al., 2008). In 2017, IR transgenic crops accounted for 23.3 million hectares, making them the second largest area under cultivation (ISAAA, 2017).

Overall, 304 events have been authorized for worldwide production. Depending on the frequency of insect pests, 208 events comprising different IR genes in maize have been authorized for cultivation. Cotton (49 events), potato (30 events), soybean (6), sugarcane (3), rice (3), poplar (2), tomato (1), and brinjal (1) are the commercialized crops that have different IR genes. Most frequently, the cry genes of Bt are used to produce IR transgenic crops. The *Cry* genes are produced by *Cry* protein, which forms crystalline inclusion in bacterial spores and gives Bt insecticidal activities. The three domains that make up the *Cry* toxin fragment are responsible for spore formation, facilitating receptor binding, and protecting the toxin against proteases (Kumar et al., 2020).

The toxin attaches itself to particular receptors and penetrates the intestinal epithelial cell membrane of the insect midgut region. And then, domain I is inserted into the membrane, resulting in pore formation, which ultimately causes the insect to become paralyzed and die. *Cry* genes derived from multiple isolates of Bt offer resistance against a range of insect pests, i.e., Dipterans, Lepidopterans, and coleopterans (McPherson et al., 1988). To create long-lasting insect resistance, numerous cry gene variants have been identified and used in gene stacking. After the success of transgenic cotton, *Cry* genes were also added to a variety of crops, like potato, rice, canola, soybean, maize, chickpea, alfalfa, and tomato. Apart from *Cry*, other insecticidal genes such as vip genes which encode vegetative insecticidal proteins have been used to be deployed in commercialized crops. The *Vip* genes are extracted from *Bacillus* sps. (*Bacillus thuringinesis* and *Bacillus cerecus*) (Fang et al., 2007). Currently, *vip3A(a)* and *vip3Aa20* genes are heterologously generated in maize and cotton crop plants (ISAAA 2019).

2.4.3 ABIOTIC-STRESS TOLERANT TRANSGENIC CROPS

Abiotic stressors such as drought, heat, cold, and salinity significantly impair agricultural productivity by disrupting plant growth and reducing grain yields (Suzuki et al., 2014). To cope, plants activate defense mechanisms like antioxidant systems, osmotic adjustments (via proline, sugars, and betaines), and regulatory proteins (transcription factors, heat shock proteins) to maintain cellular homeostasis (Gao et al., 2007; Gupta et al., 2015). However, due to the genetic complexity of abiotic stress tolerance, fewer commercialized transgenic events exist compared to herbicide or insect resistance (ISAAA, 2019). One approach involves bacterial *cold shock proteins (CSPs)*, such as *cspA* and *cspB* from *Escherichia coli* and *Bacillus subtilis*, which enhance drought and cold tolerance in maize, rice, and *Arabidopsis* without adverse pleiotropic effects (Castiglioni et al., 2008).

These RNA chaperones stabilize RNA and protein translation under stress, preserving cellular function (Karlson et al., 2002). Similarly, wheat's *WCSP1* and *Arabidopsis*' *GRP2* exhibit RNA-binding properties that aid cold and salt adaptation (Kim et al., 2007). A notable commercial application is Monsanto's drought-tolerant maize (MON 87460), expressing *CspB* and marketed as Genuity® Drought Guard™, which reduces water loss under stress (ISAAA, 2017). This variety, designed for Sub-Saharan Africa, combines drought tolerance with insect resistance to enhance resilience in arid regions. Despite progress, abiotic stress tolerance in GM crops remains limited due to multigenic regulation, necessitating further research into gene stacking and stress-responsive pathways for broader agricultural adaptation (Kim et al., 2013).

2.4.4 Disease-Resistant Transgenic Crops

Plant pathogens, including bacteria, viruses, fungi, and nematodes, significantly reduce crop yields, and while agrochemicals are widely used for disease control, their environmental impact has driven the need for sustainable alternatives (ISAAA, 2017). Developing disease-resistant crops through genetic engineering by transferring resistance genes has emerged as an effective solution, with 29 commercialized transgenic events, 25 of which target viral resistance (ISAAA, 2017). These events are primarily found in potatoes (19), papayas (four), squash (two), and beans, plums, sweet peppers, and tomatoes (one each). Four key transgenic strategies have been successfully employed: (1) expressing viral coat protein (*CP*) genes for pathogen-derived resistance, (2) using defective viral replicase/helicase domains to trigger gene silencing, (3) expressing sense/antisense RNA of viral replication proteins (*Rep*), and (4) deploying antisense RNA to degrade viral mRNA (Fuchs & Gonsalves, 1995).

For instance, transgenic squash expressing CP genes resists watermelon mosaic potyvirus 2, zucchini yellow mosaic virus, and cucumber mosaic virus (Fuchs & Gonsalves, 1995). Similarly, papaya varieties like Rainbow and SunUp incorporate *PRSV CP* genes, conferring resistance via pathogen-derived resistance (Ferreira et al., 2002). Plum pox virus resistance was achieved in *Prunus domestica* through *CP* expression (Ravelonandro et al., 1997), while tomatoes and sweet peppers gained resistance to cucumber mosaic virus using the same approach (Guo et al., 2009). Additionally, the *PRSV replicase* (*rep*) gene was used in Huanong No. 1 papaya for viral resistance (Guo et al., 2009), and bean golden mosaic virus resistance was achieved in *Phaseolus vulgaris* via viral *Rep* protein expression (Faria et al., 2006). These advancements highlight the potential of genetic engineering in sustainable crop disease management.

2.4.5 Nutritionally Improved Transgenic Crops

Table 2.1 highlights the nutritional improvement of transgenic crops with the help of introducing exogenous gene or transgene from one organism to another for inducing foreign characteristics in various crops.

TABLE 2.1
Nutritional Improvement in Transgenic Crops

Nutrient Enriched	Crop Varieties	Characteristic Features	Reference
1. Provitamin A biofortified rice	• Golden rice	• Two foreign genes the *psy* gene and *crtI* gene obtained from bacterium *Erwinia uredovora* in Japanese rice cultivars Taipei309. • Within the rice endosperm, these genes reconstruct the carotenoid biosynthetic pathway.	Ye et al. (2000)
	• Golden rice 1 (American rice variety Cocodrie)	• Under the influence of an endosperm-specific promoter, *psy*, and *crtI* gene can accumulate up to 6 µg/g of carotenoid inside endosperm.	Al-Babili and Beyer (2005)
	• Golden rice 2 (American rice variety Kaybonne)	• Maize *psy* gene and *crtI* bacterial control gene regulate the expression of endosperm-specific glutein-1 promoter from rice, and • Accumulate up to 37 µg/g (9–37 µg/g) of total carotenoid, which is almost 23 times superior to original rice, with 84% of β-carotene content.	Paine et al. (2005)
2. Modified oil or fatty acids	• Vistive Gold® soybean	• Soybean seed oil reports <3% of linolenic acid, making it stable and less hydrogenating and leading to a reduction in trans-fatty acids.	Kumar et al. (2020)
	• *Camelina sativa*	• Genetically engineered with marine microbes to produce more Ω-3 polyunsaturated fatty acids (PUFAs) similar to fish oil.	Ruiz-Lopez et al. 2017
	• Argentine Canola	• Genetically altered for production of Ω-3 fatty acids, particularly docosahexaenoic acid (DHA). • Transgenic canola has been introduced in 7 gene sources from yeast and micron microalgae (with 20 or more carbons).	Kumar et al. (2020)
3. Essential amino acids	• Transgenic wheat and rice	• Heterogeneously introducing lysin-rich pea legumin protein in wheat and rice endosperm.	Stoger et al. (2001)
	• Transgenic maize	• Developed using *Amaranthus hypochondriacus* (seed storage protein) with >32% of protein content in comparison to wild types, and a greater quantity of essential amino acids (lysine, isoleucine, tryptophan, etc.).	Rascon-Cruz et al. (2004)

2.5 APPROACHES TO COMBAT CLIMATE CHANGE ADOPTION THROUGH GM CROPS

Disparity in the environment has a lasting impact on agriculture production and food security globally (Keurentjes et al., 2008). Food safety and security are vulnerable to poor weather conditions and it is not a recent incident, although no consideration was implemented to confront these situations (ISAAA, 2017).

2.5.1 Genetics and Genomics Strategies

Omics techniques have become indispensable tools for deciphering genetic data and advancing crop improvement, enabling researchers to uncover novel genetic variations and their ecological significance through population genomics and molecular markers (Stinchcombe & Hoekstra, 2008). By integrating genomic approaches with breeding programs, scientists can identify superior germplasms with multi-trait assemblies, while genetics and transcriptomic analyses help elucidate phenotype-environment interactions and abiotic stress resistance mechanisms (Bevan & Waugh, 2007; Des Marais et al., 2013; ISAAA, 2017). High-throughput sequencing and phenotyping advancements have accelerated genomic-led breeding, allowing for the identification of stress-related productivity constraints and facilitating the development of climate-smart crops (Collins et al., 2008; Kole et al., 2015).

Quantitative trait loci (QTL) mapping and marker-assisted selection have proven crucial for dissecting yield-related traits and enhancing stress tolerance, as demonstrated by the development of drought-resistant wheat varieties like “Ripper” without yield penalties (da Silva Dias, 2015; Haley et al., 2007; Wani et al., 2018). Next-generation sequencing has enabled the discovery of extensive DNA polymorphisms, particularly single nucleotide polymorphisms (SNPs), refining QTL mapping precision to under 1 cm and supporting targeted breeding efforts (D’Agostino & Tripodi, 2017). QTL studies in barley have identified genomic regions regulating drought response traits, showing the potential of molecular breeding to optimize crop performance under environmental stress (Kochevenko et al., 2018).

2.5.2 Genome-Wide Association Studies for Stress Tolerance

Genome-wide association studies (GWAS) have emerged as a powerful tool for identifying genetic variations linked to specific traits in crops, utilizing SNPs to uncover allelic variants associated with climate resilience and stress tolerance (Bush & Moore, 2012; Manolio, 2010). GWAS has been extensively applied to study both biotic and abiotic stress responses, including drought, heat, and salinity, with notable examples in *Arabidopsis thaliana*, where reverse genetics revealed genes like thioredoxins and ribosomal proteins regulating proline accumulation under drought (Thoen et al., 2017; Verslues et al., 2014). Similarly, in *Aegilops tauschii*, GWAS identified 7185 SNPs correlated with 13 drought-related traits, providing insights into stress resistance mechanisms (Ashraf, 2009; Qin et al., 2016). For salinity tolerance in rice, high-throughput SNP arrays pinpointed key QTLs, including the *Saltol* locus on chromosome 1, which enhances seedling-stage salt resistance (Kumar et al., 2015). Additionally, GWAS in rice exposed to heat stress during anthesis uncovered 14 loci associated with spikelet sterility, implicating heat shock proteins and gametophyte development in stress adaptation (Lafarge et al., 2017).

2.5.3 Genome Selection for Crop Improvement

Genomic selection (GS) has emerged as a transformative approach in crop breeding, leveraging high-density markers and advanced phenotyping to accelerate varietal development (Burgueno et al., 2012). Statistical innovations, such as the linear mixed model for G×E interactions (Jarquin et al., 2014) and marker-environment integrated models (Lopez-Cruz et al., 2015), have enhanced predictive accuracy, with Bayesian multi-environment models further refining genomic predictions (Cuevas et al., 2017). Wheat dominates GS research (29 of 40 studies), where Diversity Array Technology and SNP-based genotyping prove most effective, demonstrating GS's potential for cereal improvement (Rutkoski et al., 2017). Applications like CIMMYT's phenomics-driven evaluation of 1000 wheat lines under heat/drought stress highlight GS's practical utility in addressing climate challenges (Crain et al., 2018).

2.6 CONCLUSION

The increasing global population, climate change, and shrinking farmland demand high-yielding, nutrient-rich crops, that can withstand biotic and abiotic stresses. Genetic engineering has addressed these needs by developing resilient, high-yield crops, reducing pesticide use, and enhancing farmers' economic stability. However, concerns over gene flow, ecological impacts, and potential health risks persist despite regulatory approvals, leading to low consumer acceptance in many developing nations. To overcome these challenges, alternative approaches like cisgenesis, intragenesis, and advanced breeding techniques are being explored to create safer, climate-resilient crops. These innovations aim to balance productivity, sustainability, and public trust in agricultural biotechnology.

REFERENCES

Al-Babili, S., & Beyer, P. (2005). Golden rice—Five years on the road—Five years to go? *Trends in Plant Science*, *10*(12), 565–573. https://doi.org/10.1016/j.tplants.2005.10.006

Ashraf, M. (2009). Biotechnological approach of improving plant salt tolerance using antioxidants as markers. *Biotechnology Advances*, *27*(1), 84–93. https://doi.org/10.1016/j.biotechadv.2008.09.003

Barry, G. F., Kishore, G. M., Padgette, S. R., & Stallings, W. C. (1997). *Glyphosate-tolerant 5-enolpyruvylshikimate-3-phosphate synthases* (U.S. Patent No. 5,633,435). U.S. Patent and Trademark Office. https://patents.google.com/patent/US5633435A

Bevan, M., & Waugh, R. (2007). Applying plant genomics to crop improvement. *Genome Biology*, *8*, 302. https://doi.org/10.1186/gb-2007-8-2-302

Brookes, G., & Barfoot, P. (2018). Farm income and production impacts of using GM crop technology 1996–2016. *GM Crops and Food*, *9*(2), 59–89. https://doi.org/10.1080/21645698.2018.1464866

Burgueno, J., de los Campos, G., Weigel, K., & Crossa, J. (2012). Genomic prediction of breeding values when modeling genotype × environment interaction using pedigree and dense molecular markers. *Crop Science*, *52*(2), 707–719. https://doi.org/10.2135/cropsci2011.06.0299

Bush, W. S., & Moore, J. H. (2012). Chapter 11: Genome-wide association studies. *PLoS Computational Biology*, *8*(12), e1002822. https://doi.org/10.1371/journal.pcbi.1002822

Castiglioni, P., Warner, D., Bensen, R. J., Anstrom, D. C., Harrison, J., Stoecker, M., & Heard, J. E. (2008). Bacterial RNA chaperones confer abiotic stress tolerance in plants and improved grain yield in maize under water-limited conditions. *Plant Physiology*, *147*(2), 446–455. https://doi.org/10.1104/pp.108.118828

Collins, N. C., Tardieu, F., & Tuberosa, R. (2008). Quantitative trait loci and crop performance under abiotic stress: Where do we stand? *Plant Physiology*, *147*(2), 469–486. https://doi.org/10.1104/pp.108.118117

Crain, J., Mondal, S., Rutkoski, J., Singh, R. P., & Poland, J. (2018). Combining high-throughput phenotyping and genomic information to increase prediction and selection accuracy in wheat breeding. *The Plant Genome*, *11*(1), 170043. https://doi.org/10.3835/plantgenome2017.05.0043

Cuevas, J., Crossa, J., Montesinos-Lopez, O. A., Burgueño, J., Pérez-Rodríguez, P., & de los Campos, G. (2017). Bayesian genomic prediction with genotype × environment interaction kernel models. *G3: Genes, Genomes, Genetics*, *7*(1), 41–53. https://doi.org/10.1534/g3.116.035584

D'Agostino, N., & Tripodi, P. (2017). NGS-based genotyping, high-throughput phenotyping and genome-wide association studies laid the foundations for next-generation breeding in horticultural crops. *Diversity*, *9*(3), 38. https://doi.org/10.3390/d9030038

Das, A., Mahanta, M., Pramanik, B., & Purkayastha, S. (2023). Genetically modified crops and crop species adapted to global warming in dry regions. In *Enhancing resilience of dryland agriculture under changing climate* (pp. 385–409). Springer. https://doi.org/10.1007/978-981-19-9159-2_19

da Silva Dias, J. C. (2015). Biodiversity and plant breeding as tools for harmony between modern agriculture production and the environment. *Molecular approaches to genetic diversity* (pp. 1–44). https://doi.org/10.5772/60080

Des Marais, D. L., Hernandez, K. M., & Juenger, T. E. (2013). Genotype-by-environment interaction and plasticity: Exploring genomic responses of plants to the abiotic environment. *Annual Review of Ecology, Evolution, and Systematics*, *44*(1), 5–29. https://doi.org/10.1146/annurev-ecolsys-110512-135806

Dill, G. M., CaJacob, C. A., & Padgette, S. R. (2008). Glyphosate-resistant crops: Adoption, use and future considerations. *Pest Management Science*, *64*(4), 326–331. https://doi.org/10.1002/ps.1501

EFSA. (2011). Guidance for risk assessment of food and feed from genetically modified plants. *EFSA Journal*, *9*(5), 2150. https://doi.org/10.2903/j.efsa.2011.2150

Fang, J., Xu, X., Wang, P., Zhao, J. Z., Shelton, A. M., Cheng, J., & Shen, Z. (2007). Characterization of chimeric *Bacillus thuringiensis* Vip3 toxins. *Applied and Environmental Microbiology*, *73*(3), 956–961. https://doi.org/10.1128/AEM.02079-06

Faria, J. C., Albino, M. M., Dias, B. B., Cançado, L. J., da Cunha, N. B., Silva, L. D. M., & Aragão, F. J. (2006). Partial resistance to *Bean golden mosaic virus* in a transgenic common bean (*Phaseolus vulgaris* L.) line expressing a mutated rep gene. *Plant Science*, *171*(5), 565–571. https://doi.org/10.1016/j.plantsci.2006.06.010

Ferdous, J., Whitford, R., Nguyen, M., Brien, C., Langridge, P., & Tricker, P. J. (2017). Drought-inducible expression of *Hv-miR827* enhances drought tolerance in transgenic barley. *Functional and Integrative Genomics*, *17*, 279–292. https://doi.org/10.1007/s10142-016-0526-8

Ferreira, S. A., Pitz, K. Y., Manshardt, R., Zee, F., Fitch, M., & Gonsalves, D. (2002). Virus coat protein transgenic papaya provides practical control of *Papaya ringspot virus* in Hawaii. *Plant Disease*, *86*(2), 101–105. https://doi.org/10.1094/PDIS.2002.86.2.101

Fuchs, M., & Gonsalves, D. (1995). Resistance of transgenic hybrid squash ZW-20 expressing the coat protein genes of *Zucchini yellow mosaic virus* and *Watermelon mosaic virus 2* to mixed infections by both potyviruses. *Bio/Technology*, *13*(12), 1466–1473. https://doi.org/10.1038/nbt1295-1466

Gao, J. P., Chao, D. Y., & Lin, H. X. (2007). Understanding abiotic stress tolerance mechanisms: Recent studies on stress response in rice. *Journal of Integrative Plant Biology*, *49*(6), 742–750. https://doi.org/10.1111/j.1744-7909.2007.00495.x

Griffiths, A. J. F., Wessler, S. R., Lewontin, R. C., Gelbart, W. M., Suzuki, D. T., & Miller, J. H. (2005). *Introduction to genetic analysis* (8th ed.). W.H. Freeman.

Guo, J., Yang, L., Liu, X., Guan, X., Jiang, L., & Zhang, D. (2009). Characterization of the exogenous insert and development of event-specific PCR detection methods for genetically modified Huanong No. 1 papaya. *Journal of Agricultural and Food Chemistry*, *57*(16), 7205–7212. https://doi.org/10.1021/jf901198x

Gupta, B., Tripathi, A. K., Joshi, R., Pareek, A., & Singla-Pareek, S. L. (2015). Designing climate-smart future crops employing signal transduction components. In *Elucidation of abiotic stress signaling in plants* (pp. 393–413). Springer.

Haley, S. D., Johnson, J. J., Peairs, F. B., Quick, J. S., Stromberger, J. A., Clayshulte, S. R., & Kolmer, J. (2007). Registration of 'Ripper' wheat. *Journal of Plant Registrations*, *1*(1), 1–6. https://doi.org/10.3198/jpr2006.10.0689crc

Holme, I. B., Dionisio, G., Brinch-Pedersen, H., Wendt, T., Madsen, C. K., Vincze, E., & Holm, P. B. (2012). Cisgenic barley with improved phytase activity. *Plant Biotechnology Journal*, *10*(2), 237–247. https://doi.org/10.1111/j.1467-7652.2011.00660.x

ISAAA. (2017). *Global status of commercialized biotech/GM crops in 2017* (Brief No. 53). Retrieved November 17, 2019, from https://www.isaaa.org/resources/publications/briefs/53/

ISAAA. (2019). *GM approval database*. Retrieved November 17, 2019, from https://www.isaaa.org/gmapprovaldatabase

Jarquín, D., Crossa, J., Lacaze, X., Du Cheyron, P., Daucourt, J., Lorgeou, J., Piraux, F., Guerreiro, L., Pérez, P., Calus, M., Burgueño, J., & De Los Campos, G. (2013). A reaction norm model for genomic selection using high-dimensional genomic and environmental data. *Theoretical and Applied Genetics*, *127*(3), 595–607. https://doi.org/10.1007/s00122-013-2243-1

Karlson, D., Nakaminami, K., Toyomasu, T., & Imai, R. (2002). A cold-regulated nucleic acid-binding protein of winter wheat shares a domain with bacterial cold shock proteins. *Journal of Biological Chemistry*, *277*(38), 35248–35256. https://doi.org/10.1074/jbc.M205774200

Keresa, S., Baric, M., Grdisa, M., Igrc Barcic, J., & Marchetti, S. (2008). Transgenic plants expressing insect resistance genes. *Sjemenarstvo*, *25*(2), 139–153.

Keurentjes, J. J., Koornneef, M., & Vreugdenhil, D. (2008). Quantitative genetics in the age of omics. *Current Opinion in Plant Biology*, *11*(2), 123–128. https://doi.org/10.1016/j.pbi.2008.01.006

Kim, J. S., Park, S. J., Kwak, K. J., Kim, Y. O., Kim, J. Y., Song, J., & Kang, H. (2007). Cold shock domain proteins and glycine-rich RNA-binding proteins from *Arabidopsis thaliana* can promote the cold adaptation process in *Escherichia coli*. *Nucleic Acids Research*, *35*(2), 506–516. https://doi.org/10.1093/nar/gkl1076

Kim, M. H., Sato, S., Sasaki, K., Saburi, W., Matsui, H., & Imai, R. (2013). *COLD SHOCK DOMAIN PROTEIN 3* is involved in salt and drought stress tolerance in *Arabidopsis*. *FEBS Open Bio*, *3*, 438–442. https://doi.org/10.1016/j.fob.2013.10.003

Kochevenko, A., Jiang, Y., Seiler, C., Surdonja, K., Kollers, S., Reif, J. C., & Graner, A. (2018). Identification of QTL hot spots for malting quality in two elite breeding lines with distinct tolerance to abiotic stress. *BMC Plant Biology*, *18*(1), 1–17. https://doi.org/10.1186/s12870-018-1294-5

Kole, C., Muthamilarasan, M., Henry, R., Edwards, D., Sharma, R., Abberton, M., & Prasad, M. (2015). Application of genomics-assisted breeding for generation of climate resilient crops: Progress and prospects. *Frontiers in Plant Science*, *6*, 563. https://doi.org/10.3389/fpls.2015.00563

Koul, B., Pudhuvai, B., Bhanot, M., & Tiwari, S. (2024). Updates on global status of transgenic and genome-edited crops. In *Genetic engineering of crop plants for food and health security* (pp. 469–510). Springer.

Kumar, K., Gambhir, G., Dass, A., Tripathi, A. K., Singh, A., Jha, A. K., & Rakshit, S. (2020). Genetically modified crops: Current status and future prospects. *Planta*, *251*(4), 91. https://doi.org/10.1007/s00425-020-03372-8

Kumar, V., Singh, A., Mithra, S. A., Krishnamurthy, S. L., Parida, S. K., Jain, S., & Mohapatra, T. (2015). Genome-wide association mapping of salinity tolerance in rice (*Oryza sativa*). *DNA Research*, *22*(2), 133–145. https://doi.org/10.1093/dnares/dsu046

Lafarge, T., Bueno, C., Frouin, J., Jacquin, L., Courtois, B., & Ahmadi, N. (2017). Genome-wide association analysis for heat tolerance at flowering detected a large set of genes involved in adaptation to thermal and other stresses. *PLoS One*, *12*(2), e0171254. https://doi.org/10.1371/journal.pone.0171254

Lea, P. J., Joy, K. W., Ramos, J. L., & Guerrero, M. G. (1984). The action of 2-amino-4-(methylphosphinyl)-butanoic acid (phosphinothricin) and its 2-oxo-derivative on the metabolism of cyanobacteria and higher plants. *Phytochemistry*, *23*(1), 1–6. https://doi.org/10.1016/0031-9422(84)83066-6

Li, L., Du, Y., He, C., Dietrich, C. R., Li, J., Ma, X., & Zheng, J. (2019). Maize *glossy6* is involved in cuticular wax deposition and drought tolerance. *Journal of Experimental Botany*, *70*(12), 3089–3099. https://doi.org/10.1093/jxb/erz131

Liang, C. (2016). Genetically modified crops with drought tolerance: achievements, challenges, and perspectives. *Drought stress tolerance in plants, Vol 2: Molecular and genetic perspectives*, 531–547.

Lopez-Cruz, M., Crossa, J., Bonnett, D., Dreisigacker, S., Poland, J., Jannink, J. L., & de los Campos, G. (2015). Increased prediction accuracy in wheat breeding trials using a marker × environment interaction genomic selection model. *G3: Genes, Genomes, Genetics*, *5*(4), 569–582. https://doi.org/10.1534/g3.114.016097

Manolio, T. A. (2010). Genomewide association studies and assessment of the risk of disease. *New England Journal of Medicine*, *363*(2), 166–176. https://doi.org/10.1056/NEJMra0905980

McPherson, S. A., Perlak, F. J., Fuchs, R. L., Marrone, P. G., Lavrik, P. B., & Fischhoff, D. A. (1988). Characterization of the coleopteran-specific protein gene of *Bacillus thuringiensis* var. *tenebrionis*. *Bio/Technology*, *6*(1), 61–66. https://doi.org/10.1038/nbt0188-61

NASEM. (2016). *Genetically engineered crops: Experiences and prospects*. National Academies Press. https://doi.org/10.17226/23395

Nicolia, A., Manzo, A., Veronesi, F., & Rosellini, D. (2014). An overview of the last 10 years of genetically engineered crop safety research. *Critical Reviews in Biotechnology*, *34*(1), 77–88. https://doi.org/10.3109/07388551.2013.823595

Paine, J. A., Shipton, C. A., Chaggar, S., Howells, R. M., Kennedy, M. J., Vernon, G., & Drake, R. (2005). Improving the nutritional value of Golden Rice through increased pro-vitamin a content. *Nature Biotechnology*, *23*(4), 482–487. https://doi.org/10.1038/nbt1082

Pallett, K. (2021). The current status of GM and GE crops. *Outlooks on Pest Management*, *32*(2), 50–52. https://doi.org/10.1564/v32_apr_03

Qin, P., Lin, Y., Hu, Y., Liu, K., Mao, S., Li, Z., & Zheng, Y. (2016). Genome-wide association study of drought-related resistance traits in *Aegilops tauschii*. *Genetics and Molecular Biology*, *39*(3), 398–407. https://doi.org/10.1590/1678-4685-GMB-2015-0232

Randhawa, G., Singh, M., & Sood, P. (2016). DNA-based methods for detection of genetically modified events in food and supply chain. *Current Science*, *110*(6), 1000–1009.

Rascon-Cruz, Q., Sinagawa-García, S., Osuna-Castro, J. A., Bohorova, N., & Paredes-López, O. (2004). Accumulation, assembly, and digestibility of amarantin expressed in transgenic tropical maize. *Theoretical and Applied Genetics*, *108*(2), 335–342. https://doi.org/10.1007/s00122-003-1430-x

Ravelonandro, M., Scorza, R., Bachelier, J. C., Labonne, G., Levy, L., Damsteegt, V., & Dunez, J. (1997). Resistance of transgenic *Prunus domestica* to *Plum pox virus* infection. *Plant Disease*, *81*(11), 1231–1235. https://doi.org/10.1094/PDIS.1997.81.11.1231

Redenbaugh, K., Hiatt, B., Martineau, B., Kramer, M., Sheehy, R., Sanders, R., & Emlay, D. (1992). *Safety assessment of genetically engineered fruits and vegetables: A case study of the Flavr Savr tomato*. CRC Press.

Ruiz-Lopez, N., Broughton, R., Usher, S., Salas, J. J., Haslam, R. P., Napier, J. A., & Beaudoin, F. (2017). Tailoring the composition of novel wax esters in the seeds of transgenic *Camelina sativa* through systematic metabolic engineering. *Plant Biotechnology Journal*, *15*(7), 837–849. https://doi.org/10.1111/pbi.12679

Rutkoski, J. E., Crain, J., Poland, J., & Sorrells, M. E. (2017). Genomic selection for small grain improvement. In *Genomic selection for crop improvement* (pp. 99–130). Springer.

Sauer, H., Wild, A., & Rühle, W. (1987). The effect of phosphinothricin (glufosinate) on photosynthesis II. The causes of inhibition of photosynthesis. *Zeitschrift für Naturforschung C*, *42*(3), 270–278. https://doi.org/10.1515/znc-1987-0317

Schouten, H. J., Krens, F. A., & Jacobsen, E. (2006). Cisgenic plants are similar to traditionally bred plants: International regulations for genetically modified organisms should be altered to exempt cisgenesis. *EMBO Reports*, *7*(8), 750–753. https://doi.org/10.1038/sj.embor.7400769

Shou, H., Bordallo, P., & Wang, K. (2004). Expression of the *Nicotiana* protein kinase (*NPK1*) enhanced drought tolerance in transgenic maize. *Journal of Experimental Botany*, *55*(399), 1013–1019. https://doi.org/10.1093/jxb/erh129

Stinchcombe, J. R., & Hoekstra, H. E. (2008). Combining population genomics and quantitative genetics: Finding the genes underlying ecologically important traits. *Heredity*, *100*(2), 158–170. https://doi.org/10.1038/sj.hdy.6800937

Stoger, E., Parker, M., Christou, P., & Casey, R. (2001). Pea legumin overexpressed in wheat endosperm assembles into an ordered paracrystalline matrix. *Plant Physiology*, *125*(4), 1732–1742. https://doi.org/10.1104/pp.125.4.1732

Suzuki, N., Rivero, R. M., Shulaev, V., Blumwald, E., & Mittler, R. (2014). Abiotic and biotic stress combinations. *New Phytologist*, *203*(1), 32–43. https://doi.org/10.1111/nph.12797

Tabashnik, B. E., Carrière, Y., Dennehy, T. J., Morin, S., Sisterson, M. S., Roush, R. T., & Zhao, J. Z. (2003). Insect resistance to transgenic Bt crops: Lessons from the laboratory and field. *Journal of Economic Entomology*, *96*(4), 1031–1038. https://doi.org/10.1093/jee/96.4.1031

Tachibana, K., Watanabe, T., Sekizawa, Y., & Takematsu, T. (1986). Accumulation of ammonia in plants treated with bialaphos: Action mechanism of bialaphos (part 2). *Journal of Pesticide Science*, *11*(1), 33–37. https://doi.org/10.1584/jpestics.11.33

Templeton, K., Bryant, C., Biswas, M., Gill, G., Bajaj, S., Sathish, P., & Hanley, C. (2021). Drought-Tolerance in Perennial Ryegrass: A Cisgenic® Approach. Multifunctional Grasslands in a Changing World Volume II, pp. 358. https://uknowledge.uky.edu/cgi/viewcontent.cgi?article=3282&context=igc

Thoen, M. P., Davila Olivas, N. H., Kloth, K. J., Coolen, S., Huang, P. P., Aarts, M. G., & Dicke, M. (2017). Genetic architecture of plant stress resistance: Multi-trait genome-wide association mapping. *New Phytologist*, *213*(3), 1346–1362. https://doi.org/10.1111/nph.14220

Tome, K. G., Dionglay, C., & Escasura, J. C. (2024). Countries approving GM crop cultivation. *International Service for the Acquisition of Agri-biotech Applications*. https://www.isaaa.org/blog/entry/default.asp?BlogDate=10/31/2024

Trapero, C., Wilson, I. W., Stiller, W. N., & Wilson, L. J. (2016). Enhancing integrated pest management in GM cotton systems using host plant resistance. *Frontiers in Plant Science*, *7*, 500. https://doi.org/10.3389/fpls.2016.00500

Verslues, P. E., Lasky, J. R., Juenger, T. E., Liu, T. W., & Kumar, M. N. (2014). Genome-wide association mapping combined with reverse genetics identifies new effectors of low water potential-induced proline accumulation in *Arabidopsis*. *Plant Physiology*, *164*(1), 144–159. https://doi.org/10.1104/pp.113.224014

Wani, S. H., Choudhary, M., Kumar, P., Akram, N. A., Surekha, C., Ahmad, P., & Gosal, S. S. (2018). Marker-assisted breeding for abiotic stress tolerance in crop plants. In *Biotechnologies of Crop Improvement* (pp. 1–23). Springer.

Ye, X., Al-Babili, S., Klöti, A., Zhang, J., Lucca, P., Beyer, P., & Potrykus, I. (2000). Engineering the provitamin A (β-carotene) biosynthetic pathway into (carotenoid-free) rice endosperm. *Science*, *287*(5451), 303–305. https://doi.org/10.1126/science.287.5451.303

Yin, X., Biswal, A. K., Dionora, J., Perdigon, K. M., Balahadia, C. P., Mazumdar, S., & Bandyopadhyay, A. (2017). CRISPR-Cas9 and CRISPR-Cpf1 mediated targeting of a stomatal developmental gene *EPFL9* in rice. *Plant Cell Reports*, *36*, 745–757. https://doi.org/10.1007/s00299-017-2118-z

3 Leveraging RNA Interference for Strengthening Crop Resilience to Biotic and Abiotic Stress Challenges

Solanki Bal

3.1 INTRODUCTION

Food is fundamental for human survival, but conventional crop improvement methods struggle to meet rising global demand due to their slow, labor-intensive processes (Mezzetti et al., 2020). Innovative agricultural solutions are urgently needed to enhance yields, nutrition, and disease resistance more efficiently for our growing population (Chen et al., 2019). By 2050, food production is predicted to increase by 70% to feed the growing global population (Godfray et al., 2010). However, cross-breeding and mutation breeding are non-targeted breeding techniques that involve laborious processes, and the production and distribution of the resulting genotypes face numerous challenges (Godfray et al., 2010). Similarly, transgenic breeding faces public acceptance issues in addition to its lengthy and costly commercialization process (Chen et al., 2019). Agricultural productivity faces major threats from biotic stressors, including pests, pathogens, and diseases and abiotic challenges, with viruses and insects causing particularly severe yield losses (Chen et al., 2019). RNA interference (RNAi) offers a transformative alternative by selectively silencing genes to enhance disease resistance and stress tolerance, without introducing transgenes, thus minimizing biosafety concerns. By altering gene expression and optimizing metabolism, this technology contributes to the development of stress-resilient crop varieties (Pathak & Gogoi, 2016).

The goal of sustainable agriculture is to improve agricultural quality and output by developing and utilizing accessible, cost-effective, and environmentally friendly technologies and methods (Fletcher et al., 2020). However, the use of synthetic pesticides to mitigate crop losses due to stress has its own drawbacks, such as adverse effects on human health (Fletcher et al., 2020). To promote agricultural sustainability, scientists have adopted more innovative and environmentally responsible crop protection strategies. In this context, the development of transgenic crops using advanced biotechnology techniques, such as RNAi, has been a significant breakthrough (Rodrigues & Petrick, 2020). RNAi is considered superior to traditional

DOI: 10.1201/9781003597797-3

transgenic methods because the genetically modified (GM) plants produced do not contain transgenic proteins (Rajam, 2020). Given these unique features, RNAi has become a widely used and effective technique for crop protection and improvement (Mezzetti et al., 2020). This chapter thoroughly discusses the role of RNAi in protecting crops from various biotic and abiotic stresses, along with an explanation of the RNAi-mediated gene silencing mechanism.

3.2 RNA INTERFERENCE

RNAi is an evolutionarily conserved gene regulation mechanism in eukaryotes, first discovered when attempts to enhance petunia flower pigmentation unexpectedly caused variegation through co-suppression of both transgenic and endogenous genes (Napoli et al., 1990). This phenomenon, later termed RNAi, was also observed in *Caenorhabditis elegans* following double-stranded RNA (dsRNA) injection (Fire et al., 1998). In plants, RNAi operates through small RNAs (sRNAs), including microRNAs (miRNAs) and short interfering RNAs with miRNAs being more conserved and capable of silencing multiple targets via polycistronic precursors (Mezzetti et al., 2020).

RNAi effects can persist across generations through non-genetic memory mediated by the *MSH1* protein, with *MSH1* inhibition leading to atypical developmental phenotypes (Ali et al., 2010). Similar RNA-based regulatory systems exist across organisms, including bacterial CRISPR/Cas and fungal quelling, though RNAi remains eukaryotes' primary antiviral defense (Ali et al., 2010). The process involves Dicer enzymes cleaving dsRNAs into 21–28 nucleotide sRNAs that guide the RNA-induced silencing complex (RISC) to degrade complementary mRNAs (Hammond, 2005). This precise, heritable silencing mechanism has made RNAi invaluable for biotechnology applications since its discovery (Fire et al., 1998). The system's ability to process both endogenous and exogenous dsRNAs into gene-specific effectors (Hammond, 2005) underlies its widespread biological importance and utility. Ongoing research continues to reveal new dimensions of RNAi biology, including its epigenetic inheritance patterns and interactions with stress responses (Ali et al., 2010).

3.3 THE ENGINE OF RNA INTERFERENCE

3.3.1 Components of RNAi Machinery

The RNAi machinery consists of key components, including dsRNA, the Dicer enzyme (which processes dsRNA into small interfering RNAs (siRNAs) or miRNAs), the RISC (which incorporates siRNAs/miRNAs to target complementary mRNA for degradation or translational repression), and Argonaute (AGO) proteins (the catalytic core of RISC that mediates mRNA cleavage or silencing) (Wilson & Doudna, 2013). The RISC and Dicer are two ribonucleases involved in the RNAi pathway. The RNAi process begins when dsRNA is cleaved by Dicer into active short non-coding RNAs, while RISC, utilizing the RNase H core enzyme AGO, silences the target gene (Wilson & Doudna, 2013).

The Dicer family, a class 3 RNase III enzyme, consists of four domains: a dsRNA-binding domain, dual RNase III domains, a PAZ (Piwi/Argonaute/Zwille) domain, and an N-terminal helicase domain. These domains are primarily responsible for recognizing the dsRNA precursor in the RNAi pathway and generating shorter non-coding RNAs, typically 21–24 nucleotides in length (Mezzetti et al., 2020). According to the Dicer catalysis model, the active core of the multidomain Dicer enzyme forms an intramolecular pseudo-dimer through the dimerization of its two RNase III domains. It has also been proposed that each domain creates a new terminus by cleaving a single strand of the dsRNA (Zhang et al., 2004). In the final stage of the RNAi pathway, RISC, in conjunction with the AGO protein and other effector proteins, carries out gene silencing by degrading the target mRNA. Argonaute proteins are found in bacteria, archaea, and eukaryotes (Ali et al., 2010).

3.3.2 MECHANISM OF ACTION

Over the past 20 years, extensive research has been conducted on the role of short non-coding RNAs in transcriptional gene silencing (TGS) and post-transcriptional gene silencing (PTGS) regulatory processes. Several types of short non-coding RNAs have now been identified, including miRNA, siRNA, piRNA (PIWI-interacting RNA), qiRNA (QDE-2-interacting RNA), svRNA (small vault RNA), and others, each with distinct biogenesis pathways and regulatory mechanisms (Aalto & Pasquinelli, 2012). The synthesis of siRNA and miRNA begins through different pathways to generate their respective dsRNA precursors. While miRNA is derived from genomic DNA within the cell, siRNA can be produced exogenously through the cleavage of dsRNA into smaller fragments or endogenously from viruses, transposons, or transgenes. The RNAi pathway consists of four main steps: synthesis of small non-coding RNA (snRNA) through Dicer cleavage, loading of snRNA into the RISC complex, activation of the silencing complex, and degradation of the target mRNA (Ali et al., 2010).

3.3.3 MICRORNA (miRNA)

MIR genes produce miRNAs, which are sRNAs approximately 21–24 nucleotides (nt) in length. The biogenesis of miRNA begins in the nucleus with RNA polymerase II-mediated transcription of MIR genes, resulting in a primary miRNA (pri-miRNA) transcript of about 1000 nt. Due to intramolecular sequence complementarity, the pri-miRNA folds into a stem-loop or hairpin structure. This structure is further processed by DCL1, with the assistance of the dsRNA-binding proteins DRB1 or HYL1, to generate a precursor miRNA (pre-miRNA), a shorter stem-loop intermediate. In the nucleus, DCL1 cleaves the pre-miRNA to produce an RNA duplex, consisting of the mature miRNA (the guide strand) and its complementary strand (the passenger strand). The *HUA ENHANCER 1 (HEN1)* enzyme methylates the 3′ termini of the RNA duplex at the 2′-O hydroxyl group, preventing degradation of the miRNA:miRNA duplex (Huntzinger & Izaurralde, 2011).

After methylation, the RNA duplex is transported to the cytoplasm, where the mature miRNA is incorporated into the RISC along with Argonaute (AGO) and other effector proteins (Kurihara et al., 2006). When the miRNA-induced silencing complex (miRISC) fully pairs with its target mRNA, the AGO protein's nuclease activity cleaves and degrades the target mRNA (Guo et al., 2016). However, miRNA-mediated downregulation of gene expression occurs through two primary mechanisms: (1) deadenylation and destabilization of the target mRNA, or (2) miRISC-mediated inhibition of translational initiation, ribosome subunit joining, premature degradation of the nascent polypeptide chain, and increased ribosome drop-off (Huntzinger & Izaurralde, 2011).

3.3.4 Small Interfering RNA (siRNA)

Short hairpin precursors or long dsRNA can initiate gene silencing through RNAi by base pairing with the target gene that needs to be silenced. The RNAi process can be triggered by transgenes, viral invaders, or transposable elements recruiting Dicer or Dicer-like enzymes, or by long endogenous dsRNA entering the cytoplasm directly. The Dicer enzyme processes these dsRNAs into short 21–24 nucleotide (nt) long siRNA duplexes with 5′ phosphorylated ends and 2 nt overhangs at the 3′ OH end. After the antisense strand of the siRNA is loaded onto the siRNA-induced silencing complex (siRISC) in a sequence-specific manner, siRISC degrades the sense strand of the siRNA, which shares the same sequence as the target mRNA. When siRISC, along with the AGO protein and other effector proteins, binds to the target mRNA, it induces PTGS by either cleaving the target mRNA or inhibiting translation (Saurabh et al., 2014). The siRNAs can participate in co-transcriptional gene silencing through chromatin regulation. Dicer-independent siRNA generation has also been observed in *Schizosaccharomyces pombe*, *Neurospora*, *C. elegans*, and *Arabidopsis*. These Dicer-independent siRNAs primarily originate from transposable, intergenic, and transgenic elements (Saurabh et al., 2014).

3.4 RNA INTERFERENCE-MEDIATED TOLERANCE TO BIOTIC STRESS

Biological stress in plants is caused by living organisms, particularly weeds, insects, arachnids, bacteria, fungi, viruses, and nematodes. Six major food and income crops experience an overall yield loss of nearly 40% due to these pathogens (Ding et al., 2009).

3.4.1 Bacterial Resistance Mediated by RNAi

Bacterial pathogens present a major threat to crop productivity due to their ubiquitous nature and rapid proliferation, necessitating the development of resistant crop varieties (Katiyar-Agarwal et al., 2006). RNAi technology has been successfully employed to combat crown gall disease in tomato (*Solanum lycopersicum*) and *Arabidopsis thaliana* by targeting the tumor-forming oncogenes *iaaM* and *ipt* through dsRNA constructs (Katiyar-Agarwal et al., 2006). Fatty acid derivatives, such as those modulated by fatty acid amide hydrolase (FAAH), influence bacterial resistance, with FAAH overexpression in *Arabidopsis* increasing susceptibility to *Pseudomonas syringae* pathogens (Katiyar-Agarwal et al., 2006). Auxin signaling plays a critical role in disease susceptibility, as demonstrated by the downregulation of auxin receptors (*AFB2*, *AFB3*, and *TIR1*) via miR393, which restricts *P. syringae* growth (Katiyar-Agarwal et al., 2006). sRNAs, including miR398 and nat-siRNAATGB2, are key regulators of plant defense responses, with their suppression during *P. syringae* infection highlighting their role in resistance mechanisms (Katiyar-Agarwal et al., 2006). These findings underscore the potential of RNAi and sRNA-mediated strategies in developing crops with enhanced bacterial resistance, offering sustainable solutions to mitigate pathogen-induced losses (Katiyar-Agarwal et al., 2006).

3.4.2 FUNGAL RESISTANCE MEDIATED BY RNAi

Host-induced gene silencing (HIGS) has proven to be a powerful tool for engineering fungal resistance in crops, as demonstrated by successful applications in barley (*Hordeum vulgare*) and wheat (*Triticum aestivum*) through silencing of *Blumeria graminis* effector gene *Avr10* (Nowara et al., 2010). In *Arabidopsis*, dsRNA targeting *HaCR1* from *Hyaloperonospora arabidopsidis* induced immune responses, while its overexpression increased susceptibility, highlighting the delicate balance in plant-pathogen interactions (Nowara et al., 2010). Rice studies revealed that silencing fatty acid desaturases (*OsFAD7* and *OsFAD8*) and stearoyl-ACP desaturase (*OsSSI2*) altered lipid profiles and pathogen resistance, demonstrating the role of fatty acid metabolism in defense against *Magnaporthe grisea* and *Xanthomonas oryzae* (Yin et al., 2011). For broad-spectrum control, RNAi targeting lignin biosynthesis in soybean reduced *Sclerotinia sclerotiorum* infection (Peltier et al., 2009), while miRNA manipulation in wheat and rice (*miR7695*) showed species-specific effects against *B. graminis* and *Magnaporthe oryzae* (Peltier et al., 2009).

Critical fungal genes including *Fusarium oxysporum*'s MAP kinases (*Fmk1*, *Hog1*, *Pbs2*), *ornithine decarboxylase* (*ODC*), and ergosterol biosynthesis genes (*ERG6/11*) have been effectively silenced to control wilt diseases in tomato and banana (Peltier et al., 2009; Tetorya & Rajam, 2018), while potatoes engineered with artificial miRNAs against *Phytophthora infestans Avr3a* showed reduced late blight severity (Thakur & Prasad, 2020). These collective advances underscore HIGS as a versatile, targeted approach for sustainable crop protection, with ongoing refinements in delivery methods and target selection enhancing its potential against evolving fungal pathogens (Nowara et al., 2010; Thakur & Prasad, 2020).

3.4.3 RNA INTERFERENCE PROVIDES DEFENSE AGAINST A RANGE OF VIRAL INFECTIONS IN PLANTS

The commonly utilized conceptual technique known as 'pathogen-derived resistance' (PDR) produces GM plants with increased virus resistance. Both RNA-mediated and protein-mediated PDR are feasible; the former uses the proteins encoded by the transgene, while the latter uses the transcript that the transgene generates. Utilizing hp-dsRNAs, which are made using the IR sequence of the viral genome in vivo and consist of short hp-RNAs, self-complementary hp-RNAs, and intron-spliced hp-RNAs, PDR is accomplished. Resistance to viral infections was demonstrated by the IRs of dsRNA-induced PTGS (IR-PTGS) and hp-RNAs that were self-complementary and split by an intron. When sense and antisense transcripts were generated simultaneously, it was found that the plants were resistant to viral infection. The ability of transgenic bean plants to withstand *Bean golden mosaic virus* has been demonstrated (Kertbundit et al., 2007). One of the transgenic lines demonstrated resistance to about 300 viruliferous whiteflies per plant over the course of the plant life cycle after inoculation at high pressure. In 2011, this event started to be marketed in Brazil. The *CP* gene of a Thai isolate of the *Papaya ringspot virus* (*PRSV*) was employed (Kertbundit et al., 2007) to produce papaya plants that were resistant to the virus.

3.4.4 RNAi-Mediated Nematode and Insect Resistance

Plant-parasitic nematodes, including *Meloidogyne*, *Heterodera*, and *Globodera* species, cause severe agricultural losses worldwide, prompting the development of RNAi-based resistance strategies (Tsygankova et al., 2019). Targeting nematode parasitism genes (e.g., *HgY25*, *HgPrp17*) or housekeeping genes (e.g., integrase, splicing factors) in host plants like *Arabidopsis* and soybean reduces nematode reproduction by 23–64%, though complete resistance remains elusive (Tsygankova et al., 2019). Similarly, silencing *Heterodera schachtii* genes in sugar beet or *Meloidogyne incognita* genes in crops diminishes female worm counts and gall formation, validating RNAi's potential for nematode control (Tsygankova et al., 2019). Beyond nematodes, RNAi has been applied to insect pests, as demonstrated by *Bacillus thuringiensis* cry toxin synergism and dsRNA targeting of cotton bollworm (*CYP6AE14*) or corn rootworm (V-ATPase subunit A), disrupting detoxification or essential metabolic pathways (Baum et al., 2007; Mao et al., 2007). Further, targeting insect HMGR, a key enzyme in the mevalonate pathway, offers a promising RNAi strategy for pest-resistant crops (Tsygankova et al., 2019; Wang et al., 2013).

3.5 PLANT RESISTANT TO BOTH RNA AND DNA VIRUSES BY RNA SILENCING

RNAi technology has emerged as a powerful tool for developing virus-resistant crops, as demonstrated by transgenic cassava expressing AC1-homologous hp-dsRNA that conferred immunity against *African cassava mosaic virus (ACMV)* through viral load reduction (Tsygankova et al., 2019). Similarly, knockdown of *Cm-eIF4E* in melon plants provided resistance to multiple viruses, including melon necrotic spot virus and zucchini yellow mosaic virus, while siRNA-mediated approaches achieved viral resistance in rice and potatoes by targeting viral components like *eIF4E* or coat proteins (Tsygankova et al., 2019).

Artificial miRNAs (amiRNAs) have also proven effective, with *Arabidopsis* expressing amiRNAs against *Turnip mosaic virus* (HC-Pro) and *Turnip yellow mosaic virus* (P69) showing heritable resistance, and tobacco plants targeting the

Cucumber mosaic virus (CMV) 2b suppressor gene exhibiting reduced infection (Niu et al., 2006). These strategies have been extended to legumes, where RNAi counters threats like *Soybean mosaic virus* and *Mungbean yellow mosaic India virus* (Tsygankova et al., 2019). Key viral targets include coat proteins, movement proteins, and replicases, with RNAi's precision enabling broad-spectrum resistance without compromising plant fitness (Tsygankova et al., 2019). Collectively, these advances highlight RNAi's potential as a sustainable solution to global crop viral pandemics, though field scalability and pathogen evolution require ongoing research (Mao et al., 2007).

3.6 PLANT EPIGENETIC CHANGES BY SMALL RNA TO FEND OFF BIOTIC STRESSORS

Epigenetic modifications, including DNA methylation and sRNA-mediated regulation, serve as critical mechanisms for plants to adapt to biotic stresses by altering gene expression without changing the DNA sequence (Ashapkin et al., 2020; Tsygankova et al., 2019). The RNA-directed DNA methylation (RdDM) pathway, mediated by 24-nt siRNAs produced by RDR2, Pol IV, and DCL3, guides AGO4 complexes to induce sequence-specific methylation, leading to chromatin remodeling and transcriptional silencing of stress-responsive genes or transposable elements (Ashapkin et al., 2020). Pathogen-induced DNA hypomethylation, observed in plants like rice and *Arabidopsis*, often occurs near promoters and stress-related genes, as demonstrated by the activation of the rice *XA21G* gene upon demethylation, which conferred resistance to *X. oryzae* (Holoch & Moazed, 2015). TE-derived siRNAs, such as *TE-siR815* in rice, illustrate epigenetic regulation by downregulating defense-related genes via *RdDM*, showing the dual role of sRNAs in RNAi and epigenetic silencing (Leonetti & Molinari, 2020). These modifications are dynamically maintained by methyltransferases (e.g., chromomethylases) and erased by demethylases, enabling flexible responses to pathogens and pests (Holoch & Moazed, 2015). Collectively, epigenetic mechanisms complement RNAi strategies to enhance plant resilience, offering potential for engineering durable biotic stress resistance in crops (Ashapkin et al., 2020).

3.7 RNAi FOR ABIOTIC STRESS ADAPTATION

Plants have evolved stress resistance mechanisms to counteract environmental challenges that threaten seed production, with RNAi emerging as a powerful tool for developing stress-tolerant crops through post-transcriptional gene regulation (Holoch & Moazed, 2015). This approach has successfully introduced targeted abiotic stress resilience in various crops by leveraging plants' natural ability to rapidly adjust gene expression and physiology under adverse conditions (Leonetti & Molinari, 2020). Gene expression control occurs at two key levels, transcriptional and post-transcriptional, with the latter involving mRNA processing, stabilization, and translation, processes that are significantly altered during stress responses (Niu et al., 2006). Notably, abiotic stressors trigger substantial changes in miRNA expression patterns that normally regulate plant growth (Holoch & Moazed, 2015), highlighting RNAi's potential for precise crop improvement under environmental pressures (Tsygankova et al., 2019).

3.7.1 RNAi for Drought Resistance

Drought stress significantly impacts plant growth and development, prompting extensive research into genetic and molecular mechanisms to enhance drought tolerance, such as RNAi and miRNA regulation (Wang et al., 2013). For instance, Wang et al. demonstrated that suppressing farnesyltransferase via an RNAi construct under the *AtHPR1* (*Arabidopsis thaliana Hydroxypyruvate Reductase 1*) promoter in canola (*Brassica napus* L.) maintained yield under drought conditions, while Li et al. (2009) showed that RNAi-mediated suppression of the *RACK1* gene in rice improved drought tolerance compared to non-transgenic lines.

Jian et al. (2010) further confirmed enhanced drought resistance in transgenic rice, correlating with elevated expression of *miR169g* and *miR393*, whereas Zhao et al. (2007) reported that drought-stressed transgenic peanuts exhibited reliable yields, alongside miRNA-mediated downregulation of drought-responsive genes in rice. Investigations in barley and wild emmer wheat using miRNA microarrays revealed stress-responsive miRNAs like *miR474*, which suppresses *proline dehydrogenase* (*PDH*) to promote proline accumulation and drought resilience in maize (Kantar et al., 2010).

Additionally, *miR393* was found to enhance drought and salt tolerance by reducing auxin signaling via *TIR1* suppression, though its overexpression in rice paradoxically reduced stress resistance (Kantar et al., 2010), while *miR159* in *Arabidopsis* responded to dehydration by modulating hormone signaling (Zhao et al., 2007). Studies on *OsLG3* and *OsAHL1* in rice linked drought avoidance to improved root development and oxidative stress responses, whereas RNAi suppression of *BrDST71* in Chinese cabbage and *OsGRXS17* in rice enhanced drought tolerance (Hu et al., 2017). Tissue-specific miRNA regulation, such as *miR169* upregulation in rice and *Arabidopsis* under salt and drought stress, highlights the complexity of stress responses across species and organs, emphasizing the need for targeted analyses to fully understand these adaptive mechanisms (Zhao et al., 2007).

3.7.2 RNAi Mediated Salt Tolerance

Salinity stress significantly reduces crop yield and quality by altering miRNA expression patterns, as demonstrated in barley where *miR444* showed dynamic regulation, downregulated initially but upregulated later under salt stress with overexpression in rice affecting root architecture (Deng et al., 2015). In *Arabidopsis*, abiotic stresses like salinity, cold, and ABA upregulated *miR393*, *miR402*, *miR319c*, *miR397b*, and *miR389a*, while maize exhibited stress-specific miRNA profiles, with *miR396* and *miR156* upregulated and *miR474* and *miR162* downregulated in roots (Jian et al., 2010). The scaffold protein *RACK1*, critical for stress responses, was studied via RNAi in rice, revealing enhanced drought tolerance in transgenic lines, while *OsRPK1* was identified as key for salt tolerance, influencing membrane integrity and proline levels (Li et al., 2020). Conserved miRNAs like *miR393*, *miR160*, *miR169*, and *miR167* were consistently upregulated across species under drought and salt stress, though responses varied by tissue and stress intensity (Deng et al., 2015). These findings highlight miRNAs as versatile regulators of stress adaptation, offering targets for breeding resilient crops (Zhao et al., 2016).

3.7.3 TOLERANCE TO HEAT AND COLD STRESS MEDIATED BY RNA INTERFERENCE

Environmental factors and temperature fluctuations significantly reduce crop productivity by altering gene expression patterns, with RNAi demonstrating potential in mitigating these effects, as evidenced by stable silencing of gliadins in wheat under heat stress and variable nitrogen supply (Marín-Sanz et al., 2020). Temperature-responsive miRNAs exhibit species-specific behaviors, with heat-responsive changes observed in *Panicum virgatum*, *Oryza sativa*, and *T. aestivum*, while chilling-sensitive species like *Solanum habrochaites* and *Glycine max* displayed distinct miRNA profiles (Marín-Sanz et al., 2020). Exogenous dsRNA applications targeting transgenes such as *NPTII* and *EGFP* in *Arabidopsis* revealed that silencing efficiency depends on factors like plant age, soil moisture, and application timing, highlighting the practicality of dsRNA in abiotic stress management (Kiselev et al., 2021). Key miRNA families, including *miR156*, *miR159*, and *miR169*, regulate heat and cold responses across species, with *miR156* mediating heat stress memory in *Arabidopsis* and *miR169* targeting *nuclear transcription factor Y* (*NF-Y*) in wheat, maize, and *Arabidopsis* under temperature extremes (Saurin et al., 2014). However, challenges such as inconsistent miRNA annotations, NGS limitations, and noisy data complicate the interpretation of stress-responsive miRNAs, necessitating improved characterization methods (Das & Sherif, 2020). These findings justify the need for refined miRNA studies to harness their potential in developing climate-resilient crops amid conflicting results and technical constraints (Zhao et al., 2016).

3.8 CONCLUSION

Ensuring global food security and addressing malnutrition are critical challenges for the agricultural sector in the 21st century, particularly in developing nations. Biofortified crops and stress-resistant plants are essential to combat these issues, as they provide balanced nutrition and withstand biotic and abiotic stressors such as droughts, floods, and soil contamination. RNAi technology has emerged as a promising tool for enhancing crop resilience and nutrient content by silencing stress-responsive genes. Despite its potential, RNAi faces challenges such as off-target effects and difficulties in identifying target genes, which can be mitigated through advanced bioinformatics and experimental testing. Combining RNAi with other technologies, such as gene editing and gene pyramiding, can further improve stress tolerance in staple crops. However, public concerns about GM crops must be addressed through education, regulatory frameworks, and sustainable practices. By leveraging RNAi and complementary innovations, the agricultural sector can develop climate-ready crops, ensuring sustainable food production and addressing global food security challenges.

REFERENCES

Aalto, A. P., & Pasquinelli, A. E. (2012). Small non-coding RNAs mount a silent revolution in gene expression. *Current Opinion in Cell Biology*, *24*(4), 333–340. https://doi.org/10.1016/j.ceb.2012.03.006

Ali, N., Datta, S. K., & Datta, K. (2010). RNA interference in designing transgenic crops. *GM Crops*, *1*(4), 207–213. https://doi.org/10.4161/gmcr.1.4.13344

Ashapkin, V. V., Kutueva, L. I., Aleksandrushkina, N. I., & Vanyushin, B. F. (2020). Epigenetic mechanisms of plant adaptation to biotic and abiotic stresses. *International Journal of Molecular Sciences*, *21*(20), 7457. https://doi.org/10.3390/ijms21207457

Baum, J. A., Bogaert, T., Clinton, W., Heck, G. R., Feldmann, P., Ilagan, O., & Vaughn, T. (2007). Control of coleopteran insect pests through RNA interference. *Nature Biotechnology*, *25*(11), 1322–1326. https://doi.org/10.1038/nbt1359

Chen, K., Wang, Y., Zhang, R., Zhang, H., & Gao, C. (2019). CRISPR/Cas genome editing and precision plant breeding in agriculture. *Annual Review of Plant Biology*, *70*, 667–697. https://doi.org/10.1146/annurev-arplant-050718-100049

Das, P. R., & Sherif, S. M. (2020). Application of exogenous dsRNAs-induced RNAi in agriculture: Challenges and triumphs. *Frontiers in Plant Science*, *11*, 946. https://doi.org/10.3389/fpls.2020.00946

Deng, P., Wang, L., Cui, L., Feng, K., Liu, F., Du, X., & Weining, S. (2015). Global identification of microRNAs and their targets in barley under salinity stress. *PLoS ONE*, *10*(8), e0137990. https://doi.org/10.1371/journal.pone.0137990

Ding, D., Zhang, L., Wang, H., Liu, Z., Zhang, Z., & Zheng, Y. (2009). Differential expression of miRNAs in response to salt stress in maize roots. *Annals of Botany*, *103*(1), 29–38. https://doi.org/10.1093/aob/mcn205

Fire, A., Xu, S., Montgomery, M. K., Kostas, S. A., Driver, S. E., & Mello, C. C. (1998). Potent and specific genetic interference by double-stranded RNA in *Caenorhabditis elegans*. *Nature*, *391*(6669), 806–811. https://doi.org/10.1038/35888

Fletcher, S. J., Reeves, P. T., Hoang, B. T., & Mitter, N. (2020). A perspective on RNAi-based biopesticides. *Frontiers in Plant Science*, *11*, 51. https://doi.org/10.3389/fpls.2020.00051

Godfray, H. C. J., Beddington, J. R., Crute, I. R., Haddad, L., Lawrence, D., Muir, J. F., & Toulmin, C. (2010). Food security: The challenge of feeding 9 billion people. *Science*, *327*(5967), 812–818. https://doi.org/10.1126/science.1185383

Guo, Q., Liu, Q., Smith, N. A., Liang, G., & Wang, M. B. (2016). RNA silencing in plants: Mechanisms, technologies and applications in horticultural crops. *Current Genomics*, *17*(6), 476–489. https://doi.org/10.2174/1389202917666160520103117

Hammond, S. M. (2005). Dicing and slicing: The core machinery of the RNA interference pathway. *FEBS Letters*, *579*(26), 5822–5829. https://doi.org/10.1016/j.febslet.2005.08.079

Holoch, D., & Moazed, D. (2015). RNA-mediated epigenetic regulation of gene expression. *Nature Reviews Genetics*, *16*(2), 71–84. https://doi.org/10.1038/nrg3863

Hu, Y., Wu, Q., Peng, Z., Sprague, S. A., Wang, W., Park, J., & Hirschi, K. D. (2017). Silencing of *OsGRXS17* in rice improves drought stress tolerance by modulating ROS accumulation and stomatal closure. *Scientific Reports*, *7*(1), 15950. https://doi.org/10.1038/s41598-017-16254-z

Huntzinger, E., & Izaurralde, E. (2011). Gene silencing by microRNAs: Contributions of translational repression and mRNA decay. *Nature Reviews Genetics*, *12*(2), 99–110. https://doi.org/10.1038/nrg2936

Jian, X., Zhang, L., Li, G., Zhang, L., Wang, X., Cao, X., & Zha, F. C. (2010). Identification of novel stress-regulated microRNAs from *Oryza sativa* L. *Genomics*, *95*(1), 47–50. https://doi.org/10.1016/j.ygeno.2009.08.017

Kantar, M., Lucas, S. J., & Budak, H. (2010). miRNA expression patterns of *Triticum dicoccoides* in response to shock drought stress. *Planta*, *233*(3), 471–484. https://doi.org/10.1007/s00425-010-1309-4

Katiyar-Agarwal, S., Morgan, R., Dahlbeck, D., Borsani, O., Villegas, A., Zhu, J. K., & Jin, H. (2006). A pathogen-inducible endogenous siRNA in plant immunity. *Proceedings of the National Academy of Sciences*, *103*(47), 18002–18007. https://doi.org/10.1073/pnas.0608258103

Kertbundit, S., Pongtanom, N., Ruanjan, P., Chantasingh, D., Tanwanchai, A., Panyim, S., & Juricek, M. (2007). Resistance of transgenic papaya plants to *Papaya ringspot virus*. *Biologia Plantarum*, *51*(2), 333–339. https://doi.org/10.1007/s10535-007-0066-0

Kiselev, K. V., Suprun, A. R., Aleynova, O. A., Ogneva, Z. V., & Dubrovina, A. S. (2021). Physiological conditions and dsRNA application approaches for exogenously induced RNA interference in *Arabidopsis thaliana*. *Plants*, *10*(2), 264. https://doi.org/10.3390/plants10020264

Kurihara, Y., Takashi, Y., & Watanabe, Y. (2006). The interaction between DCL1 and HYL1 is important for efficient and precise processing of pri-miRNA in plant microRNA biogenesis. *RNA*, *12*(2), 206–212. https://doi.org/10.1261/rna.2146906

Leonetti, P., & Molinari, S. (2020). Epigenetic and metabolic changes in root-knot nematode-plant interactions. *International Journal of Molecular Sciences*, *21*(21), 7759. https://doi.org/10.3390/ijms21207759

Li, D., Liu, H., Yang, Y., Zhen, P., & Liang, J. (2009). Down-regulated expression of *RACK1* gene by RNA interference enhances drought tolerance in rice. *Rice Science*, *16*(1), 14–20. https://doi.org/10.1016/S1672-6308(08)60051-7

Li, J. L., Chen, X. X., Shi, C. C., Liu, F. H., Sun, J., & Ge, R. C. (2020). Effects of *OsRPK1* gene overexpression and RNAi on the salt-tolerance at seedling stage in rice. *Acta Agronomica Sinica*, *46*(8), 1217–1224. https://doi.org/10.3724/SP.J.1006.2020.92060

Mao, Y. B., Cai, W. J., Wang, J. W., Hong, G. J., Tao, X. Y., Wang, L. J., & Chen, X. Y. (2007). Silencing a cotton bollworm P450 monooxygenase gene by plant-mediated RNAi impairs larval tolerance of gossypol. *Nature Biotechnology*, *25*(11), 1307–1313. https://doi.org/10.1038/nbt1352

Marín-Sanz, M., Giménez, M. J., Barro, F., & Savin, R. (2020). Prolamin content and grain weight in RNAi silenced wheat lines under different conditions of temperature and nitrogen availability. *Frontiers in Plant Science*, *11*, 583. https://doi.org/10.3389/fpls.2020.00583

Mezzetti, B., Smagghe, G., Arpaia, S., Christiaens, O., Dietz-Pfeilstetter, A., Jones, H., & Taning, C. N. T. (2020). RNAi: What is its position in agriculture? *Journal of Pest Science*, *93*(4), 1125–1130. https://doi.org/10.1007/s10340-020-01238-2

Napoli, C., Lemieux, C., & Jorgensen, R. (1990). Introduction of a chimeric chalcone synthase gene into petunia results in reversible co-suppression of homologous genes in trans. *The Plant Cell*, *2*(4), 279–289. https://doi.org/10.1105/tpc.2.4.279

Niu, Q. W., Lin, S. S., Reyes, J. L., Chen, K. C., Wu, H. M., Yeh, S. D., & Chua, N. H. (2006). Expression of artificial microRNAs in transgenic *Arabidopsis thaliana* confers virus resistance. *Nature Biotechnology*, *24*(11), 1420–1428. https://doi.org/10.1038/nbt1255

Nowara, D., Gay, A., Lacomme, C., Shaw, J., Ridout, C., Douchkov, D., & Schweizer, P. (2010). HIGS: Host-induced gene silencing in the obligate biotrophic fungal pathogen *Blumeria graminis*. *The Plant Cell*, *22*(9), 3130–3141. https://doi.org/10.1105/tpc.110.077040

Pathak, K., & Gogoi, B. (2016). RNA interference (RNAi): Application in crop improvement: A review. *Agricultural Reviews*, *37*(4), 245–249.

Peltier, A. J., Hatfield, R. D., & Grau, C. R. (2009). Soybean stem lignin concentration relates to resistance to *Sclerotinia sclerotiorum*. *Plant Disease*, *93*(2), 149–154. https://doi.org/10.1094/PDIS-93-2-0149

Rajam, M. V. (2020). RNA silencing technology: A boon for crop improvement. *Journal of Biosciences*, *45*(1), 118. https://doi.org/10.1007/s12038-020-00082-x

Rodrigues, T. B., & Petrick, J. S. (2020). Safety considerations for humans and other vertebrates regarding agricultural uses of externally applied RNA molecules. *Frontiers in Plant Science*, *11*, 407. https://doi.org/10.3389/fpls.2020.00407

Saurabh, S., Vidyarthi, A. S., & Prasad, D. (2014). RNA interference: Concept to reality in crop improvement. *Planta*, *239*(3), 543–564. https://doi.org/10.1007/s00425-013-2019-5

Saurin, C., Declerck, M., Christ, A., Blein, T., Ma, L., Lelandais-Brière, C., & Hartmann, C. (2014). A miR169 isoform regulates specific NF-YA targets and root architecture in *Arabidopsis*. *New Phytologist*, *202*(4), 1197–1211. https://doi.org/10.1111/nph.12735

Tetorya, M., & Rajam, M. V. (2018). RNA silencing of *PEX6* gene causes decrease in pigmentation, sporulation and pathogenicity of *Fusarium oxysporum*. *Plant Pathology*, *67*(1), 67–75. https://doi.org/10.1111/ppa.12707

Thakur, O., & Prasad, R. (2020). Engineering resistance to *Alternaria cyamopsidis* by RNAi mediated gene silencing of chitin synthase export chaperone *CHS7* in guar. *Physiological and Molecular Plant Pathology*, *112*, 101541. https://doi.org/10.1016/j.pmpp.2020.101541

Tsygankova, V. A., Andrusevich, Y. V., Shysha, E. N., Biliavska, L. O., Galagan, T. O., Galkin, A. P., & Blume, Y. B. (2019). RNAi-mediated resistance against plant parasitic nematodes of wheat plants obtained in vitro using bioregulators of microbiological origin. *Current Chemical Biology*, *13*(1), 73 89. http://dx.doi.org/10.2174/2212796812666180507130017

Wang, Z., Dong, Y., Desneux, N., & Niu, C. (2013). RNAi silencing of the *HaHMG-CoA reductase* gene inhibits oviposition in the *Helicoverpa armigera* cotton bollworm. *PLoS ONE*, *8*(7), e67732. https://doi.org/10.1371/journal.pone.0067732

Wilson, R. C., & Doudna, J. A. (2013). Molecular mechanisms of RNA interference. *Annual Review of Biophysics*, *42*, 217–239. https://doi.org/10.1146/annurev-biophys-083012-130404

Yin, C., Jurgenson, J. E., & Hulbert, S. H. (2011). Development of a host-induced RNAi system in the wheat stripe rust fungus *Puccinia striiformis* f. sp. *tritici*. *Molecular Plant-Microbe Interactions*, *24*(5), 554–561. https://doi.org/10.1094/MPMI-10-10-0229

Zhang, H., Kolb, F. A., Jaskiewicz, L., Westhof, E., & Filipowicz, W. (2004). Single processing center models for human Dicer and bacterial RNase III. *Cell*, *118*(1), 57–68. https://doi.org/10.1016/j.cell.2004.06.017

Zhao, B., Liang, R., Ge, L., Li, W., Xiao, H., Lin, H., & Jin, Y. (2007). Identification of drought-induced microRNAs in rice. *Biochemical and Biophysical Research Communications*, *354*(2), 585–590. https://doi.org/10.1016/j.bbrc.2006.12.210

Zhao, J., Li, M., Gu, D., Liu, X., Zhang, J., Wu, K., & Duan, J. (2016). Involvement of rice histone deacetylase *HDA705* in seed germination and in response to ABA and abiotic stresses. *Biochemical and Biophysical Research Communications*, *470*(2), 439–444. https://doi.org/10.1016/j.bbrc.2016.01.016

4 Genome Editing in Crops for Climate Change Adaptation

Gaurav Sharma, Shivali Sharma, Dhanapati Keerthana, and Sunny Sharma

4.1 INTRODUCTION

Since most agricultural production relies on rain-fed systems, climate change has increasingly disrupted these traditional and predictable rainfall patterns. The consequences rank far reaching for the smallholder farmers and indeed food security of the world at large since over 80% of world food production depends on these smallholder farmers due to climate change. Anthropogenic emission of greenhouse gases has increased average global temperatures by near 1°C since 1850 (IPCC, 2018). Left effects are set to dominate for a very long time even up to centuries in the most optimistic scenario where warming will be capped at 1.5°C (IPCC, 2018). These effects depend upon the level of emission and are vulnerable to more frequent heat waves, droughts, floods, rise of sea level, and rise in the global temperature (IPCC, 2018). Natural ecosystem and agricultural scenario have also recently witnessed these changes in the environment; therefore, species are suffering from reduction of its habitat due to incursion (Urban, 2015).

As per the previous studies (Urban, 2015), one in six species might be threatened to extinction due to climate change. Crop productivity is most vulnerable; droughts and floods, along with decreased productivity, can be expected globally but impacts are stronger on the yield in lower latitudes. Such challenges inspire genome editing technologies that seem to be strong tools to either help crops adapt to climate change or indirectly alleviate the negative effects of adverse climatic conditions on agriculture, as shown in Figure 4.1. Techniques like TALENs, ZFNs, and CRISPR/Cas systems help in precise modification of DNA at targeted genomic locations with much greater accuracy. Gene editing for targeted traits results in production of high nutritive disease and pests resistant as well as abiotic stress-tolerant crop plants (Mekonnen et al., 2022). Grain, vegetable, and fruit crops have been utilized for precise genetic modification successfully. Enhanced nutrient levels in cereal crops like rice, maize, and wheat made them to have sufficient quantities of starch, protein, vitamin, and oleic acid through CRISPR-Cas9 biofortification. Genome editing made some crops more sugary sweet and shelf life longer and also fragrant to smell. This chapter explores the potential of genome

DOI: 10.1201/9781003597797-4

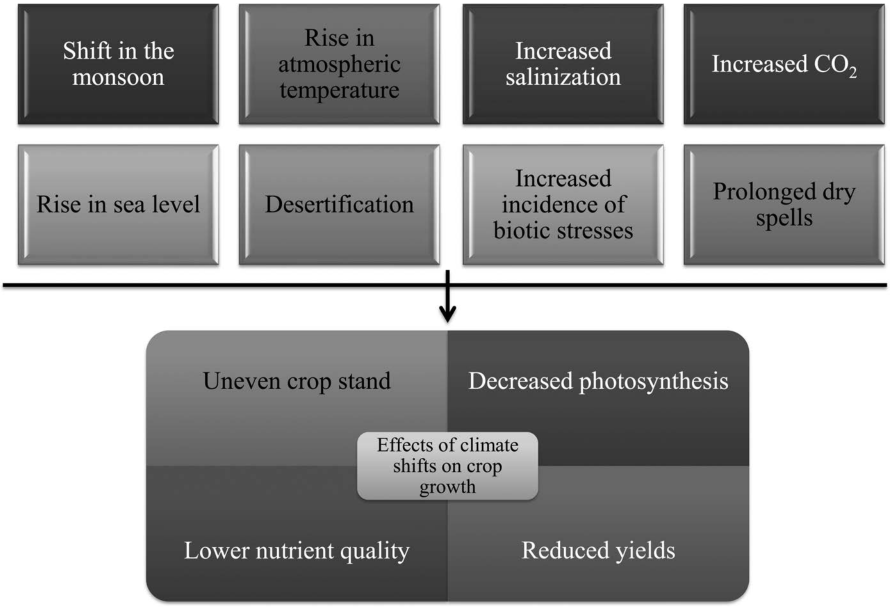


FIGURE 4.1 Various effects of climate shifts on crop growth.

editing technologies to develop crop varieties that can adapt to climatic shifts while maintaining productivity.

4.2 INFLUENCES OF CLIMATE VARIABILITY ON AGRICULTURAL YIELD

The effects of climate crisis are increasingly apparent and projected to intensify. At present, crops in lower latitude experience declines in yield, while higher-latitude areas have witnessed some yield increases (Iizumi et al., 2018). However, by the turn of next century, global crop yields and suitability are expected to decrease significantly due to climate-related factors (IPCC, 2018). The IPCC predicts that thrilling weather conditions might devastate food systems around the world, thereby lowering the global food availability and inflating food costs. In arid regions, increases in agricultural productivity due to climate change and desertification can be expected, but equatorial regions are already seen as particularly vulnerable to decreased yields as their temperatures increase. The two high-risk large continents, Asia and Africa, will be especially sensitive to the effects of desertification. The desertification underway already has begun transforming productive agricultural potential and biodiversity through unsustainable land use influences and population pressures. Exactly how much more aridity will increase globally is less certain, but susceptible areas to salinization will expand. Other environmental factors may limit CO_2 benefits, and

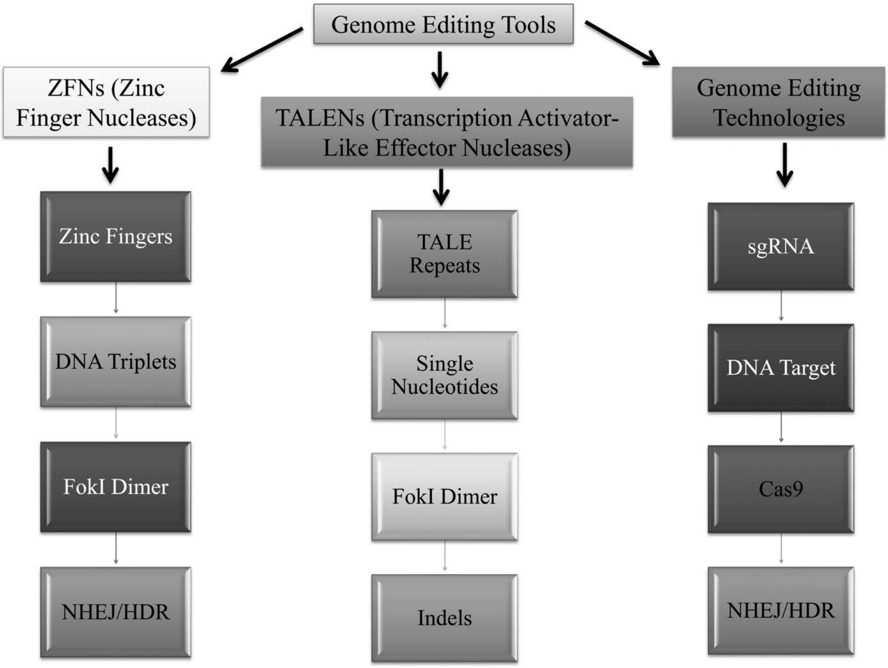


FIGURE 4.2 Basic principles of CRISPR-Cas9, TALENs, and ZFNs.

its fertilization effect has declined over three decades, likely due to nutrient depletion and water scarcity (Wang et al., 2021). Increasing heat events, heavy rainfall, and shifting disease distributions further threaten crop yields (Iizumi et al., 2018). However, Figure 4.2 illustrates the fundamental mechanisms of CRISPR-Cas9, TALENs, and ZFNs genome editing tools, while Table 4.1 provides a comparative summary of various genome editing techniques applied in crop improvement.

Meganucleases, the earliest SSNs, exhibit high specificity in targeting sequences (Bhambhani et al., 2022). Despite their role in genome editing, their use in plants is limited due to low catalytic activity and sequence constraints (Beyer & Iwai, 2022). ZFNs combine zinc-finger proteins with the FokI endonuclease to enable DNA recognition and each zinc finger binds a three-base DNA sequence, requiring dimerization for function (Li et al., 2019). ZFN-mediated gene editing has been applied to crops like soybean, maize, wheat, and rice. TALENs, similar to ZFNs, use *FokI* for DNA cleavage but rely on TALE repeats for binding, offering greater target flexibility. TALE proteins from *Xanthomonas* were identified in 2007, with their DNA-binding code deciphered in 2009 (Boch et al., 2009). TALENs have been used in various crops, improving traits like bacterial blight resistance, flavored rice, nutrient-enriched soybean, and anthocyanin-rich tomato. Various mechanisms of genome editing for climate change in crops for climate change adaptation have been described in Figure 4.3.

TABLE 4.1
Overview of Genome Editing Techniques in Crop Plants

	ZFNs	TALENs	ODM	CRISPR/Cas9	Reference
Components	Zn domains paired with nonspecific *FokI* nuclease domain	TALE DNA-binding domains combined with nonspecific *FokI* nuclease domain	Exogenous polynucleotide (chimeraplast)	crRNA, Cas9 proteins	Beyer and Iwai (2022)
Structural proteins	Dimeric protein	Dimeric protein	Non-proteinaceous	Monomeric protein	
Catalytic domain	Restriction endonuclease *FokI*	Restriction endonuclease *FokI*	No catalytic domain	*RUVC* and *HNH*	
Target sequence length (bp)	24–36	24–59	68–88	20–22	
Target DNA recognition	Protein-DNA	Protein-DNA	Single-stranded oligonucleotide (ssODN) that is complementary to the target DNA sequence	RNA-DNA	
Pros	Highly effective and precise	Highly effective and precise	Enables point mutations with minimal unintended effects and no incorporation of foreign DNA and does not rely on nucleases like CRISPR or ZFNs	Efficient, simple to construct, and possible to edit multiple sites simultaneously	
Cons	Extensive screening, requires significant time and expensive to implement	Cumbersome and time-intensive to construct	That is made up of small alterations such as point mutations or small insertions/deletions. It is also less efficient than other methods, mainly used for point mutations	A PAM motif is adjacent to target sequence necessarily	
DNA cleavage	Fold	Fold	There is no cleavage, but rather a procedure dependent upon mismatch repair once an oligonucleotide has bound to the DNA	Cas9 or Cpf1	
Available sites	1/140 bp	Any site	This approach is applied at sites that are complementary to a designed oligonucleotide, especially where small mutations are intended	1/13 bp	

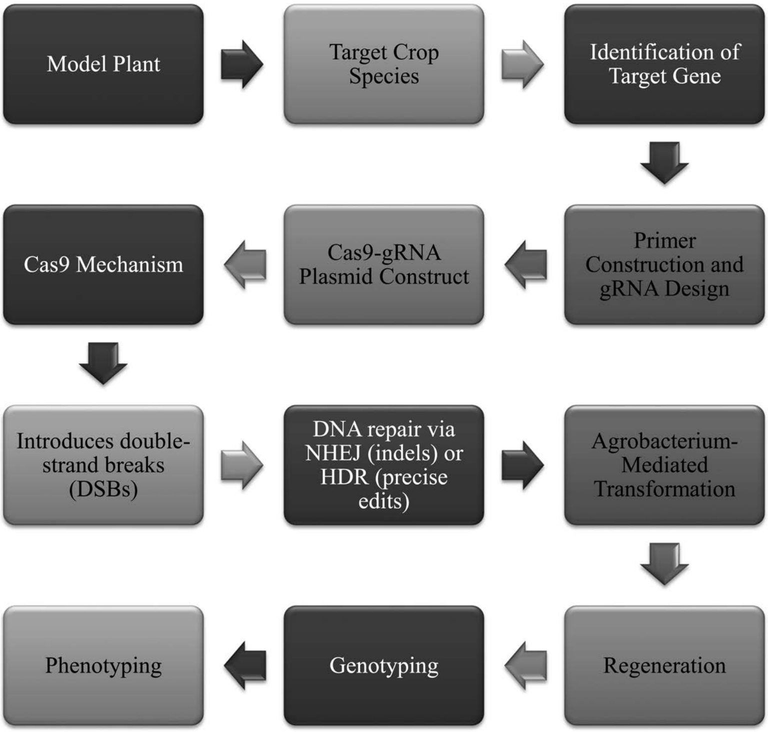


FIGURE 4.3 Mechanism of genome editing in crops for climate change adaptation.

4.3 DISCOVERY OF CRISPR/Cas SYSTEM

Functional genomics in both animal and plant biology has been revolutionized by the discovery of the CRISPR/Cas9 gene editing system. Originating in the adaptive immune systems of bacteria and archaea, CRISPR/Cas9 has evolved into a versatile tool for precise gene editing across prokaryotic and eukaryotic systems. Discovered in *Escherichia coli* (Ishino et al., 1987), CRISPR was later identified as an adaptive immune mechanism in bacteria and archaea, protecting against invading DNA (Sorek et al., 2013). Further studies confirmed that bacteria acquire new spacer sequences upon infection, adding them to the CRISPR array (Deveau et al., 2008). CRISPR/Cas systems were initially categorized into three types: I, II, and III. Type II, the most studied, employs a single Cas9 protein for interference mediation (Karlson et al., 2021). More recently, Cas systems have been grouped into two classes, six types, and 33 subtypes. Cas9, guided by tracrRNA and crRNA, recognizes PAM sequences and induces double-strand breaks (DSBs) at specific DNA sites (Karlson et al., 2021).

The system has been optimized using a single guide RNA (sgRNA), allowing targeted genome editing. A typical CRISPR/Cas9 project includes selecting a target site, assessing off-target effects, designing sgRNA, cloning into a plant expression plasmid, and transforming plants. The two major DSB repair pathways induced by

CRISPR are nonhomologous end joining (NHEJ) and homologous recombination (HR). While HR offers precise repair, its low efficiency in higher plants limits its application in crop improvement (Sorek et al., 2013). Recent advancements, including base editing (BE) and prime editing (PE), have improved precision and efficiency. BE uses cytosine or adenine deaminases to introduce point mutations without DSBs, converting CG to TA or AT to GC. PE employs a *pegRNA* to guide a reverse transcriptase, enabling substitutions, insertions, or deletions at target sites. PE supports all 12 possible base-to-base conversions, making it a versatile tool for gene editing. It has been successfully applied in plants, with reports of herbicide-resistant maize and glyphosate-resistant rice (Karlson et al., 2021). Beyond Cas9, other Cas proteins such as Cas12, Cas13, and Cas14 expand CRISPR applications. Cas12, a smaller enzyme, operates without *tracrRNA* and can process multiple guide RNAs, facilitating multi-gene editing (Wang et al., 2021). Cas13 targets RNA, enabling RNA interference in plants and viruses. Cas14, highly selective and requiring no PAM, enhances sequence detection (Savage, 2019). Despite significant advancements, challenges remain in CRISPR's agricultural implementation, particularly in improving efficiency, accuracy, and delivery methods.

4.4 GENE EDITING FOR CLIMATE CHANGE ADAPTATION IN CROP PLANTS

Genome editing, particularly CRISPR-Cas9, presents an exciting route for the alteration of crops to various environmental stresses. Such improvements are needed to develop resistance crop cultivars for ensuring the availability of future food production as climate variability is on the rise (Karlson et al., 2021).

4.4.1 CRISPR-Cas9 for Biotic Stress Management

Microorganisms, including viruses, bacteria, and fungi, infect crops, reducing both quality and quantity (Talakayala et al., 2022). Traditional plant disease resistance methods involve genome editing to remove or modify genes disrupting plant defense (Yin & Qiu, 2019). Although disease-resistance genes are few, CRISPR-based editing has strengthened resistance in many crops. Climate change increases pathogen occurrence and virulence, making hosts more susceptible. CRISPR technology helps control plant viral diseases, such as banana streak virus, which remained latent in *Plantago lanceolata* under stress conditions like drought and high temperatures after CRISPR editing (Karlson et al., 2021). The CRISPR-Cas9 system also improves bacterial leaf blight resistance in rice by targeting the SWEET gene family that pathogens exploit. Genetic modifications in the promoter regions of *OsSWEET* genes enhance resistance. Researchers successfully edited the *OsERF922* gene in rice, increasing resistance to leaf blast disease, while *eIF4G* editing provided protection against rice tungro virus, resulting in disease-free, high-yielding plants. CRISPR/Cas9 has also introduced beneficial mutations in immunity and pest susceptibility genes, enhancing crop resilience (Vu et al., 2023). CRISPR enables weed control and herbicide-tolerant crop development, promoting reduced-till farming, minimizing fossil fuel

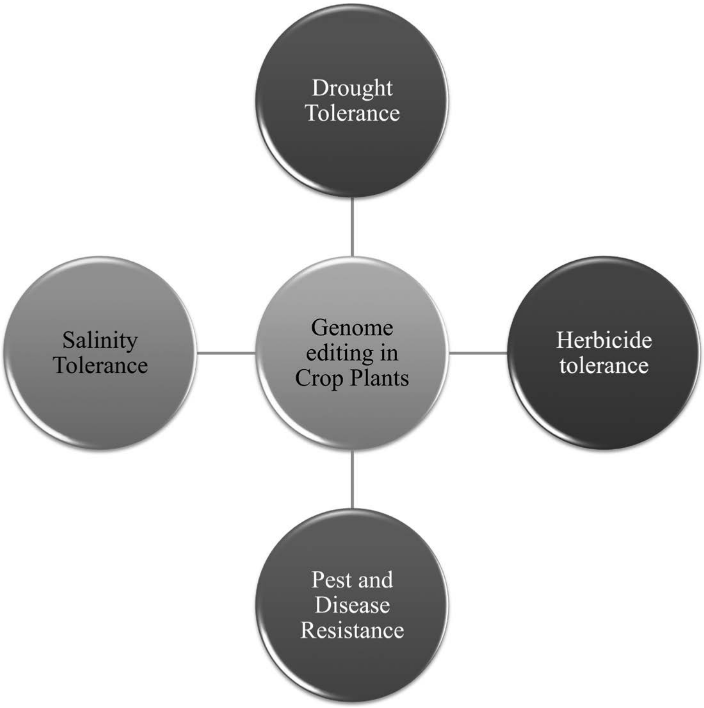


FIGURE 4.4 Reducing reliance on chemical inputs through genome editing.

use, and reducing soil disturbance and greenhouse gas emissions, ultimately reducing dependence on chemical fertilizers, fungicides, and pesticides (Figure 4.4).

4.4.2 CRISPR-CAS9 IN ABIOTIC STRESS CONTROL

Abiotic stresses from climate change, such as water deficiency, heat, cold, and soil salinization, reduce global crop yields (Neupane et al., 2022). These stresses affect over 90% of arable land. While plants have natural adaptation mechanisms, traditional breeding struggles to address stress-related traits due to genetic complexity (Vats et al., 2019). Soil salinization from drought and irrigation threatens food security, affecting 6% of cultivated lands and harming crops like rice, wheat, and maize (El Sabagh et al., 2021). Genome editing technologies play major role in increasing stress resistance as shown in Table 4.2. CRISPR editing of *OsNAC041* in rice has improved salt tolerance (Bo et al., 2019). Similarly, targeting the *OsRAV2* transcription factor has enhanced salinity tolerance. Beyond rice, genome engineering for stress adaptation has benefited wheat, maize, soybean, and tomato (Nazir et al., 2022). Drought is another major stress linked to climate change. CRISPR-modified *OsABA8ox2* in rice improved root structure and water conservation (Zhang et al., 2020). *ERA1* mutants in rice enhanced drought tolerance through ABA regulation (Ogata et al., 2020). Altering stomatal density in barley, wheat, and grapevine

TABLE 4.2
Genome Editing Techniques for Increasing Resistance Against Stress

Crop	Target gene	Trait Improved	Reference
Citrus	*CsLOB1*	Resistant to *Xanthomonas citri* subsp. *Citri* (*Xcc*)	Peng et al. (2017)
Cucumber	*eIF4E* *eIF4E*	Cucumber vein yellowing virus	Chandrasekaran et al. (2016)
Papaya	*PRSV-W*	Papaya ring spot virus	Jeyabharathy et al. (2016)
Cocoa	*NPR3*	*Phytophthora tropicalis* resistance	Fister et al. (2018)
Tomato	*CCD8*	Resistance against *Phelipanche aegyptiaca* and *Orobanche* spp.	Bari et al. (2019)
	LBD40	Drought tolerance	Liu et al. (2020b)
	WRKY52	Enhanced disease resistance	Foresti et al. (2024)
	VvWRKY52	Disease resistance pathway	Fedorina et al. (2022)
Apple	*MdDIPM1 and MdDIPM4*	Fire blight resistance	Pompili et al. (2020)
	MdDREB2	Enhanced resilience to drought	Pompili et al. (2020)
	DIPM1	Reduced fire blight susceptibility	Pompili et al. (2020)
Orange	*CsALS*	Herbicide tolerance	Mahna and Nayeri (2023)
Banana	*DMR6*	Bacterial disease resistance	Tripathi et al. (2021)
	Ma04g15900; Ma06g27710; Ma08g32850; Ma11g10500; Ma11g17210	Abiotic stress resistance	Shao et al. (2020)
Melon	*eIF4E*	Virus resistance	Yoon et al. (2020)
Potato	*StALS1*	Herbicide resistance	Mali et al. (2023)
	OsNAC041	Salinity tolerance	Bo et al. (2019)
	SBEIIb and SBEI	High amylose content	Sun et al. (2017)
Maize	*ARGOS8*	Drought stress tolerance	Shi et al. (2017)
Sorghum	Whole k1C gene family	Enhancement of grain protein digestibility and lysine content	–
Soybean	*ALS*	Herbicide resistance	–
	Drb2a and Drb2b	Salt and drought tolerance	Curtin et al. (2018)
Sugarcane	*COMT*	Saccharification efficiency improved	Kannan et al. (2018)
Flax	*EPSPS*	Herbicide resistance	Hummel et al. (2018)
Tobacco	*ALS*	Herbicide tolerance	Endo and Toki (2013)
	Regions in the viral genome	Virus resistance	Ji et al. (2015)
Barley	HvPM19	Positive regulation of grain dormancy	Lawrenson et al. (2015)
	Viral and satellite DNAs	Resistance to cotton leaf curl disease	Weeks et al. (2017)
Peanut	*FAD2*	Increased oleic acid content	Wen et al. (2018)
Avocado	*PAL, LOX*	Resistance to *anthracnose* disease	Bill et al. (2017)
	PaNPR2 and *PaNPR4*	*Phytophthora cinnamomi* resistance	Backer et al. (2015)

increased drought resistance. CRISPR-modified *OsHAK1* and *OsNRAMP5* in rice reduced cesium and cadmium toxicity. In bananas, CRISPR-based gibberellin biosynthesis modifications produced storm-resistant semi-dwarf varieties (Shao et al., 2020). CRISPR-Cas9 knock-ins, such as inserting a maize promoter into the *ARGOS8* drought-tolerance gene, improved maize yield under water stress (Shimatani et al., 2017).

4.4.3 Flowering Time Alteration in Staple Crops

Through CRISPR-based flowering time control, scientists aim to enhance crop resilience to climatic variability. Altering the *ZmCCT* gene in maize regulated early flowering for adaptation (Huang et al., 2018). CRISPR/Cas9-modified *GmFT2a* and *GmFT5a* genes improved soybean cultivation by adjusting flowering time to latitude. These genes, homologs of FT, accelerate soybean flowering under long days, facilitating climate-adaptive breeding. T2 soybean *ft2a* mutants exhibited delayed flowering under short- and long-day conditions, highlighting their role in photosensitivity. *GmFT2a* is more active under short days, while *GmFT5a* dominates under long days, balancing flowering responses for tropical regions (Karlson et al., 2021). Double mutants *ft2a-ft5a* shifted flowering by 31 days under short-day conditions, increasing yield potential (Cai et al., 2020).

BE ensured precise floral induction, optimizing crop adaptation. CRISPR/Cas9 truncated E1, upregulating *GmFT2a and GmFT5a* for early flowering. Mutations in *GmPRR37* and *GmPRR3b* promoted earlier flowering, improving adaptability to photoperiod variations (Wang et al., 2021). Similar gene editing in rice targeted *Hd3a* and *RFT1*, reducing photoperiod sensitivity and enabling earlier harvests. Editing *Ehd1* in japonica rice adjusted mid-latitude varieties to lower latitudes, extending the growing season (Wu et al., 2020). CRISPR has also adapted sorghum, rapeseed, and apple to climate variability. Editing *TERMINAL FLOWER 1* (*TFL1*) in apple and pear induced early flowering, increasing resilience. Epigenetic modifications in *Brassica napus* shortened flowering time, improving climate adaptability (Jiang et al., 2018). Moreover, genome editing for modifying flowering time and maturity has been shown in Table 4.3.

4.5 LIMITATIONS AND CHALLENGES OF GENE EDITING IN CLIMATE CHANGE ADAPTATION IN CROPS

Genome editing technologies induce precise mutations in crop genomes, significantly impacting agriculture. Advances in the CRISPR/Cas system have enhanced efficiency and practicality without incorporating foreign DNA. However, their deployment raises scientific, ecological, and socioeconomic concerns. This chapter assesses potential risks alongside benefits compared to conventional breeding. Traditional breeding also has limitations, such as increased toxic alkaloid levels in fava beans and potatoes, higher disease susceptibility, and reduced protein content in high-yield varieties (Huang et al., 2018). Spontaneous mutations occur each generation, introducing both beneficial and harmful alleles, influencing natural selection and trait preference among farmers and consumers.

TABLE 4.3
Genome Editing for Modifying Flowering Time and Maturity

Crop	Targeted Genes	Modified Trait	References
Pear	*TFL1*	Early flowering	Liu et al. (2020)
	PbPAT14	Dwarf yellowing, early flowering phenotype	–
Chrysanthemum indicum	*CfTFL1a, CiTFL1b*	Early flowering	Mekapogu et al. (2023)
	TFL1	Flowering regulation	Ramirez-Torres et al. (2021)
Kiwifruit (*Actinidia chinensis*)	*CENTRORADIALIS*-like genes	Conversion of prolonged axillary flowering into rapid terminal flowering	–
Tomato	*SlBOP, self-pruning 5G, RIN*	Altered flowering, fruit development	Mahna and Nayeri (2023)
Strawberry	*LAM, FveYUC10*	Modified runner formation, auxin levels	Ma et al. (2023)
	MaGA20ox2	Semi-dwarf growth	Zhao et al. (2021)
Banana	*MA-ACO1* (ethylene biosynthesis)	Prolongs shelf life by modifying growth traits	–
Grape	Grape promoters	Enhanced genome editing efficiency	Rukavtsova et al. (2022)
Soybean	*GIGANTEA (GI)*	Alters photoperiod and flowering time	Wang et al. (2021)
	GmE1	Early flowering	Han et al. (2019)
	GmPRR3b	Early flowering (regulation of photoperiodic flowering and circadian clock)	–
Mustard	*FEIGIDA*	Early flowering	–
	BnaSDG8.A and *BnaSDG8.C*	Early flowering	Jiang et al. (2018)
Rice	*Hd* 2, *Hd* 4, and *Hd* 5	Early maturity of rice varieties	–
	OsGS3	Early flowering	Zhou et al. (2019)
	OsPHL3	Early flowering under LD and SD conditions	–
Maize	*ZmCCT9*	Early flowering under LD	Huang et al. (2018)

4.5.1 OFF-TARGET EFFECTS

A key limitation of site-specific nucleases (SSNs) is off-target effects during targeted disruption, insertion, or replacement of genomic loci. These effects often result from homologous sequences at the target site but can also occur in nearby nonhomologous regions. Efforts to minimize off-target effects, particularly in the CRISPR/Cas system, have led to the discovery of Cas9 point mutations that enhance efficiency while reducing unintended activity. Additionally, advanced tools such as base editors for precise nucleotide changes, epigenome modifiers that alter DNA conformation and expression, and PE tools for targeted DNA insertions further refine genome editing precision (Karlson et al., 2021).

4.5.2 BREAKING OF NATURAL REPRODUCTIVE BARRIERS

Genome editing can bypass reproductive barriers that naturally limit genetic exchange. Conventional breeding maintains genetic boundaries through tightly linked DNA segments restricting recombination. Genome editing overcomes these limits, creating novel genetic combinations rigorously tested in target environments. This ensures only top-performing varieties, aligned with agronomic practices and consumer demands, reach farmers without disrupting natural genetic variation in crop production (Huang et al., 2018).

4.5.3 TRANSPARENCY

Most genome-edited plants undergo an intermediate transgenic step where foreign DNA is temporarily inserted and later removed. This requires careful lab and greenhouse stewardship, using molecular tools to confirm sequence removal before field testing. Advances in genome editing may eliminate this step, simplifying development, especially for clonally propagated crops where removal is challenging. Transparency is crucial to maintain public trust. Without it, confidence in genome-edited crops and their developers may erode. To enhance transparency, the Center for Food Integrity has launched a registry to engage civil society and educate consumers on genome-edited crops (Wu et al., 2020).

4.5.4 INTELLECTUAL PROPERTY AND ACCESS

The intellectual property landscape of genome editing remains complex and unresolved due to foundational patent disputes, particularly between the Broad Institute and Berkeley. While IP holders have shown willingness to license these technologies to public and commercial entities, legal uncertainties pose challenges, especially for resource-limited public sector researchers who may delay application until resolved. Legal clarity on ownership and licensing is essential for smallholder farmers to benefit from genome-edited crops (Karlson et al., 2021).

4.5.5 REGULATION OF GENOME-EDITED CROPS

Strict regulatory regimes and complex risk assessment protocols for GM crops pose a significant barrier to adopting genome editing technologies. Many countries have biosafety frameworks governing GM crops derived through rDNA technology, often based on food safety and environmental risk principles established for conventional genetically modified organisms (GMOs). With gene-edited crops, it is necessary to reconsider these definitions and regulatory frameworks (Cai et al., 2020). Different genome editing methods lead to varying genetic modifications. SDN-3 resembles traditional recombinant techniques with transgene insertion, making the end product a GMO, while SDN-1 introduces single-base substitutions without requiring DSBs. As a result, gene-edited crops differ from traditional GMOs, necessitating case-by-case risk assessments (Eckerstorfer et al., 2021). However, no global consensus exists on regulating genome-edited crops, as debates persist over their similarity to GMOs.

This regulatory uncertainty complicates efforts to develop and integrate improved crop varieties into agriculture (Karlson et al., 2021).

4.5.6 INTERNATIONAL LEVEL HARMONIZATION

National and international regulatory frameworks are crucial for governing genome-edited crops (Huang et al., 2018). Initially, genome editing lacked precautionary regulations like GMOs, but in 2018, the EU Court of Justice classified gene-edited crops as transgenic (Wu et al., 2020). This contrasts with countries like the U.S. and Argentina, which do not classify certain genome-edited crops as transgenic (Menz et al., 2020). Such differences could impact the entry of gene-edited crops into markets linked to EU trade (Cai et al., 2020). However, large-scale genome editing poses challenges for regulatory authorities in distinguishing natural mutations from engineered ones (Agapito-Tenfen et al., 2018).

4.6 CONCLUSION

Genome editing technologies, particularly CRISPR/Cas systems, have transformed agriculture by enabling precise, efficient genetic modifications to enhance crop quality, yield, and stress tolerance while improving food safety and shelf life. While most applications target nuclear genomes, future advancements may expand to plant organelles. Current research focuses on technical improvements, such as editing accuracy, delivery mechanisms, and new Cas variants, but gaps remain in addressing regulatory, biosecurity, and public acceptance challenges, as well as the impact of climate change on agriculture. Innovations like anti-CRISPR proteins and miniaturized delivery systems are refining precision and efficiency. However, widespread adoption requires robust frameworks, baseline data for risk assessment, and public education to bridge the gap between technological advancements and practical implementation, ultimately strengthening global food security.

REFERENCES

Agapito-Tenfen, S. Z., Okoli, A. S., Bernstein, M. J., Wikmark, O. G., & Myhr, A. I. (2018). Revisiting risk governance of GM plants: The need to consider new and emerging gene-editing techniques. *Frontiers in Plant Science*, *9*, 1874. https://doi.org/10.3389/fpls.2018.01874

Backer, R., Mahomed, W., Reeksting, B. J., Engelbrecht, J., Ibarra-Laclette, E., & van den Berg, N. (2015). Phylogenetic and expression analysis of the NPR1-like gene family from *Persea americana* (Mill.). *Frontiers in Plant Science*, *6*, 300. https://doi.org/10.3389/fpls.2015.00300

Bari, V. K., Nassar, J. A., Kheredin, S. M., Gal-On, A., Ron, M., Britt, A., & Aly, R. (2019). CRISPR/Cas9-mediated mutagenesis of CAROTENOID CLEAVAGE DIOXYGENASE 8 in tomato provides resistance against the parasitic weed *Phelipanche aegyptiaca*. *Scientific Reports*, *9*(1), 11438. https://doi.org/10.1038/s41598-019-47893-z

Beyer, H. M., & Iwai, H. (2022). Structural basis for the propagation of homing endonuclease-associated inteins. *Frontiers in Molecular Biosciences*, *9*, 855511. https://doi.org/10.3389/fmolb.2022.855511

Bhambhani, S., Kondhare, K., & Giri, A. (2022). Advanced genome editing strategies for manipulation of plant specialized metabolites pertaining to biofortification. *Phytochemistry Reviews*, *21*, 81–99. https://doi.org/10.1007/s11101-021-09745-5

Bill, M., Korsten, L., Remize, F., Glowacz, M., & Sivakumar, D. (2017). Effect of thyme oil vapours exposure on phenylalanine ammonia-lyase (PAL) and lipoxygenase (LOX) genes expression, and control of anthracnose in 'Hass' and 'Ryan' avocado fruit. *Scientia Horticulturae*, *224*, 232–237. https://doi.org/10.1016/j.scienta.2017.06.017

Bo, W., Zhaohui, Z., Huanhuan, Z., Xia, W., Binglin, L., Lijia, Y., & Yong, Z. (2019). Targeted mutagenesis of NAC transcription factor gene, *OsNAC041*, leading to salt sensitivity in rice. *Rice Science*, *26*(2), 98–108. https://doi.org/10.1016/j.rsci.2018.10.005

Boch, J., Scholze, H., Schornack, S., Landgraf, A., Hahn, S., Kay, S., & Bonas, U. (2009). Breaking the code of DNA binding specificity of TAL-type III effectors. *Science*, *326*(5959), 1509–1512. https://doi.org/10.1126/science.1178811

Cai, Y., Chen, L., Zhang, Y., Yuan, S., Su, Q., Sun, S., & Hou, W. (2020). Target base editing in soybean using a modified CRISPR/Cas9 system. *Plant Biotechnology Journal*, *18*(10), 1992–1994. https://doi.org/10.1111/pbi.13386

Chandrasekaran, J., Brumin, M., Wolf, D., Leibman, D., Klap, C., Pearlsman, M., & Gal-On, A. (2016). Development of broad virus resistance in non-transgenic cucumber using CRISPR/Cas9 technology. *Molecular Plant Pathology*, *17*(7), 1140–1153. https://doi.org/10.1111/mpp.12375

Curtin, S. J., Xiong, Y., Michno, J. M., Campbell, B. W., Stec, A. O., Čermák, T., & Stupar, R. M. (2018). CRISPR/Cas9 and TALENs generate heritable mutations for genes involved in small RNA processing of *Glycine max* and *Medicago truncatula*. *Plant Biotechnology Journal*, *16*(6), 1125–1137. https://doi.org/10.1111/pbi.12857

Deveau, H., Barrangou, R., Garneau, J. E., Labonté, J., Fremaux, C., Boyaval, P., & Moineau, S. (2008). Phage response to CRISPR-encoded resistance in *Streptococcus thermophilus*. *Journal of Bacteriology*, *190*(4), 1390–1400. https://doi.org/10.1128/JB.01412-07

Eckerstorfer, M. F., Grabowski, M., Lener, M., Engelhard, M., Simon, S., Dolezel, M., & Lüthi, C. (2021). Biosafety of genome editing applications in plant breeding: Considerations for a focused case-specific risk assessment in the EU. *BioTech*, *10*(3), 10. https://doi.org/10.3390/biotech10030010

El Sabagh, M. S., Islam, M., Skalicky, M., Raza, M. A., Singh, K., Hossain, M. A., & Ratnasekera, D. (2021). Salinity stress in wheat (*Triticum aestivum* L.) in the changing climate: Adaptation and management strategies. *Frontiers in Agronomy*, *3*, 661932. https://doi.org/10.3389/fagro.2021.661932

Endo, M., & Toki, S. (2013). Creation of herbicide-tolerant crops by gene targeting. *Journal of Pesticide Science*, *38*(2), 49–59. https://doi.org/10.1584/jpestics.D12-073

Fedorina, J., Tikhonova, N., Ukhatova, Y., Ivanov, R., & Khlestkina, E. (2022). Grapevine gene systems for resistance to gray mold *Botrytis cinerea* and powdery mildew *Erysiphe necator*. *Agronomy*, *12*(2), 499. https://doi.org/10.3390/agronomy12020499

Fister, A. S., Landherr, L., Maximova, S. N., & Guiltinan, M. J. (2018). Transient expression of CRISPR/Cas9 machinery targeting *TcNPR3* enhances defense response in *Theobroma cacao*. *Frontiers in Plant Science*, *9*, 268. https://doi.org/10.3389/fpls.2018.00268

Foresti, C., Orduña, L., Matus, J. T., Vandelle, E., Danzi, D., Bellon, O., & Zenoni, S. (2024). NAC61 regulates late- and post-ripening osmotic, oxidative, and biotic stress responses in grapevine. *Journal of Experimental Botany*, *75*(8), 2330–2350. https://doi.org/10.1093/jxb/erad505

Han, J., Guo, B., Guo, Y., Zhang, B., Wang, X., & Qiu, L. J. (2019). Creation of early flowering germplasm of soybean by CRISPR/Cas9 technology. *Frontiers in Plant Science*, *10*, 1446. https://doi.org/10.3389/fpls.2019.01446

Huang, C., Sun, H., Xu, D., Chen, Q., Liang, Y., Wang, X., & Li, D. (2018). *ZmCCT9* enhances maize adaptation to higher latitudes. *Proceedings of the National Academy of Sciences*, *115*(2), E334–E341. https://doi.org/10.1073/pnas.1718058115

Hummel, A. W., Chauhan, R. D., Cermak, T., Mutka, A. M., Vijayaraghavan, A., Boyher, A., & Taylor, N. J. (2018). Allele exchange at the EPSPS locus confers glyphosate tolerance in cassava. *Plant Biotechnology Journal*, *16*(7), 1275–1282. https://doi.org/10.1111/pbi.12868

Iizumi, T., Shiogama, H., Imada, Y., Hanasaki, N., Takikawa, H., & Nishimori, M. (2018). Crop production losses associated with anthropogenic climate change. *International Journal of Climatology*, *38*(14), 5405–5417. https://doi.org/10.1002/joc.5818

IPCC. (2018). Summary for policymakers. In V. Masson-Delmotte, P. Zhai, H.-O. Pörtner, D. Roberts, J. Skea, P. R. Shukla, et al. (Eds.), *Global warming of 1.5°C* (pp. 1–32). World Meteorological Organization.

Ishino, Y., Shinagawa, H., Makino, K., Amemura, M., & Nakata, A. (1987). Nucleotide sequence of the *iap* gene in *Escherichia coli*. *Journal of Bacteriology*, *169*(12), 5429–5433. https://doi.org/10.1128/jb.169.12.5429-5433.1987

Jeyabharathi, C., Ahrens, P., Hasse, U., & Scholz, F. (2016). Identification of low-index crystal planes of polycrystalline gold on the basis of electrochemical oxide layer formation. *Journal of Solid State Electrochemistry, 20*(11), 3025–3031. https://doi.org/10.1007/s10008-016-3228-1

Ji, X., Zhang, H., Zhang, Y., Wang, Y., & Gao, C. (2015). Establishing a CRISPR-Cas-like immune system conferring DNA virus resistance in plants. *Nature Plants*, *1*(10), 1–4. https://doi.org/10.1038/nplants.2015.144

Jiang, L., Li, D., Jin, L., Ruan, Y., Shen, W. H., & Liu, C. (2018). Histone lysine methyltransferases and their role in flowering in *Brassica napus*. *Plant Journal*, *95*(4), 672–685. https://doi.org/10.1111/tpj.13978

Kannan, B., Jung, J. H., Moxley, G. W., Lee, S. M., & Altpeter, F. (2018). TALEN-mediated targeted mutagenesis of more than 100 COMT copies/alleles in highly polyploid sugarcane improves saccharification efficiency without compromising biomass yield. *Plant Biotechnology Journal*, *16*(4), 856–866. https://doi.org/10.1111/pbi.12833

Karlson, C. K. S., Mohd-Noor, S. N., Nolte, N., & Tan, B. C. (2021). CRISPR/dCas9-based systems: Mechanisms and applications in plant sciences. *Plants*, *10*(10), 2055. https://doi.org/10.3390/plants10102055

Lawrenson, T., Shorinola, O., Stacey, N., Li, C., Østergaard, L., Patron, N., & Harwood, W. (2015). Induction of targeted, heritable mutations in barley and *Brassica oleracea* using RNA-guided Cas9 nuclease. *Genome Biology*, *16*(1), 1–13. https://doi.org/10.1186/s13059-015-0826-7

Li, D., Hsu, S., Purushotham, D., Sears, R. L., & Wang, T. (2019). WashU epigenome browser update 2019. *Nucleic acids research*, *47*(W1), W158–W165. https://doi.org/10.1093/nar/gkz348

Liu, L., Zhang, J., Xu, J., Li, Y., Guo, L., Wang, Z., & Zhang, N. (2020a). CRISPR/Cas9 targeted mutagenesis of *SlLBD40*, a lateral organ boundaries domain transcription factor, enhances drought tolerance in tomato. *Plant Science*, *301*, 110683. https://doi.org/10.1016/j.plantsci.2020.110683

Liu, X., Liu, Z., Hao, Z., Chen, G., Qi, K., Zhang, H., & Wang, P. (2020b). Characterization of Dof family in *Pyrus bretschneideri* and role of *PbDof9.2* in flowering time regulation. *Genomics*, *112*(1), 712–720. https://doi.org/10.1016/j.ygeno.2019.05.005

Ma, Z., Ma, L., & Zhou, J. (2023). Applications of CRISPR/Cas genome editing in economically important fruit crops: Recent advances and future directions. *Molecular Horticulture*, *3*(1), 1. https://doi.org/10.1186/s43897-023-00051-6

Mahna, N., & Nayeri, S. (2023). Genome editing in horticultural plants: Present applications and future perspective. In *A roadmap for plant genome editing* (pp. 223–246). Springer. https://doi.org/10.1007/978-3-031-46150-7_10

Mali, S., Dutta, M., & Zinta, G. (2023). Genome editing advancements in potato (*Solanum tuberosum* L.): Operational challenges and solutions. *Journal of Plant Biochemistry and Biotechnology*, *32*(4), 730–742. https://doi.org/10.1007/s13562-023-00851-3

Mekapogu, M., Song, H. Y., Lim, S. H., & Jung, J. A. (2023). Genetic engineering and genome editing advances to enhance floral attributes in ornamental plants: An update. *Plants*, *12*(23), 3983. https://doi.org/10.3390/plants12233983

Mekonnen, T. W., Gerrano, A. S., Mbuma, N. W., & Labuschagne, M. T. (2022). Breeding of vegetable cowpea for nutrition and climate resilience in sub-Saharan Africa: Progress, opportunities, and challenges. *Plants*, *11*(12), 1583. https://doi.org/10.3390/plants11121583

Menz, J., Modrzejewski, D., Hartung, F., Wilhelm, R., & Sprink, T. (2020). Genome edited crops touch the market: A view on the global development and regulatory environment. *Frontiers in Plant Science*, *11*, 586027. https://doi.org/10.3389/fpls.2020.586027

Nazir, R., Mandal, S., Mitra, S., Ghorai, M., Das, N., Jha, N. K., & Dey, A. (2022). Clustered regularly interspaced short palindromic repeats (CRISPR)/CRISPR-associated genome-editing toolkit to enhance salt stress tolerance in rice and wheat. *Physiologia Plantarum*, *174*(1), e13642. https://doi.org/10.1111/ppl.13642

Neupane, D., Adhikari, P., Bhattarai, D., Rana, B., Ahmed, Z., Sharma, U., & Adhikari, D. (2022). Does climate change affect the yield of the top three cereals and food security in the world? *Earth*, *3*(1), 45–71. https://doi.org/10.3390/earth3010004

Ogata, T., Ishizaki, T., Fujita, M., & Fujita, Y. (2020). CRISPR/Cas9-targeted mutagenesis of *Osera1* confers enhanced responses to abscisic acid and drought stress and increased primary root growth under nonstressed conditions in rice. *PLoS ONE*, *15*(12), e0243376. https://doi.org/10.1371/journal.pone.0243376

Peng, A., Chen, S., Lei, T., Xu, L., He, Y., Wu, L., & Zou, X. (2017). Engineering canker-resistant plants through CRISPR/Cas9-targeted editing of the susceptibility gene *CsLOB1* promoter in citrus. *Plant Biotechnology Journal*, *15*(12), 1509–1519. https://doi.org/10.1111/pbi.12733

Pompili, V. (2020). Improving fire blight resistance in susceptible apple cultivars by different biotechnological approaches. *Acta Horticulturae*, *1281*, 1–8. https://doi.org/10.17660/ActaHortic.2020.1281.1

Ramirez-Torres, F., Ghogare, R., Stowe, E., Cerdá-Bennasser, P., Lobato-Gómez, M., Williamson-Benavides, B. A., & Dhingra, A. (2021). Genome editing in fruit, ornamental, and industrial crops. *Transgenic Research*, *30*(4), 499–528. https://doi.org/10.1007/s11248-021-00257-8

Rukavtsova, E. B., Zakharchenko, N. S., Lebedev, V. G., & Shestibratov, K. A. (2022). CRISPR-Cas genome editing for horticultural crops improvement: Advantages and prospects. *Horticulturae*, *9*(1), 38. https://doi.org/10.3390/horticulturae9010038

Savage, D. F. (2019). Cas14: Big advances from small CRISPR proteins. *Biochemistry*, *58*(8), 1024–1025. https://doi.org/10.1021/acs.biochem.9b00050

Shao, X., Wu, S., Dou, T., Zhu, H., Hu, C., Huo, H., & Bi, F. (2020). Using CRISPR/Cas9 genome editing system to create *MaGA20ox2* gene-modified semi-dwarf banana. *Plant Biotechnology Journal*, *18*(1), 17–19. https://doi.org/10.1111/pbi.13216

Shi, J., Gao, H., Wang, H., Lafitte, H. R., Archibald, R. L., Yang, M., & Habben, J. E. (2017). *ARGOS8* variants generated by CRISPR-Cas9 improve maize grain yield under field drought stress conditions. *Plant Biotechnology Journal*, *15*(2), 207–216. https://doi.org/10.1111/pbi.12603

Shimatani, Z., Kashojiya, S., Takayama, M., Terada, R., Arazoe, T., Ishii, H., & Miura, K. (2017). Targeted base editing in rice and tomato using a CRISPR-Cas9 cytidine deaminase fusion. *Nature Biotechnology*, *35*(5), 441–443. https://doi.org/10.1038/nbt.3833

Sorek, R., Lawrence, C. M., & Wiedenheft, B. (2013). CRISPR-mediated adaptive immune systems in bacteria and archaea. *Annual Review of Biochemistry*, *82*, 237–266. https://doi.org/10.1146/annurev-biochem-072911-172315

Sun, Y., Jiao, G., Liu, Z., Zhang, X., Li, J., Guo, X., & Xia, L. (2017). Generation of high-amylose rice through CRISPR/Cas9-mediated targeted mutagenesis of starch branching enzymes. *Frontiers in Plant Science*, *8*, 298. https://doi.org/10.3389/fpls.2017.00298

Talakayala, A., Ankanagari, S., & Garladinne, M. (2022). CRISPR-Cas genome editing system: A versatile tool for developing disease-resistant crops. *Plant Stress*, *3*, 100056. https://doi.org/10.1016/j.stress.2022.100056

Tripathi, J. N., Ntui, V. O., Shah, T., & Tripathi, L. (2021). CRISPR/Cas9-mediated editing of *DMR6* orthologue in banana (*Musa* spp.) confers enhanced resistance to bacterial disease. *Plant Biotechnology Journal*, *19*(7), 1291–1293. https://doi.org/10.1111/pbi.13592

Urban, M. C. (2015). Accelerating extinction risk from climate change. *Science*, *348*(6234), 571–573. https://doi.org/10.1126/science.aaa4984

Vats, S., Kumawat, S., Kumar, V., Patil, G. B., Joshi, T., Sonah, H., & Deshmukh, R. (2019). Genome editing in plants: Exploration of technological advancements and challenges. *Cells*, *8*(11), 1386. https://doi.org/10.3390/cells8111386

Vu, B. N., Van Vu, T., Yoo, J. Y., Nguyen, N. T., Ko, K. S., Kim, J. Y., & Lee, K. O. (2023). CRISPR-Cas-mediated unfolded protein response control for enhancing plant stress resistance. *Frontiers in Plant Science*, *14*, 1271368. https://doi.org/10.3389/fpls.2023.1271368

Wang, T., Xun, H., Wang, W., Ding, X., Tian, H., Hussain, S., & Wang, C. (2021). Mutation of *GmAITR* genes by CRISPR/Cas9 genome editing results in enhanced salinity stress tolerance in soybean. *Frontiers in Plant Science*, *12*, 779598. https://doi.org/10.3389/fpls.2021.779598

Weeks, D. P. (2017). Gene editing in polyploid crops: Wheat, camelina, canola, potato, cotton, peanut, sugar cane, and citrus. *Progress in Molecular Biology and Translational Science*, *149*, 65–80. https://doi.org/10.1016/bs.pmbts.2017.05.002

Wen, S., Liu, H., Li, X., Chen, X., Hong, Y., Li, H., & Liang, X. (2018). TALEN-mediated targeted mutagenesis of fatty acid desaturase 2 (*FAD2*) in peanut (*Arachis hypogaea* L.) promotes the accumulation of oleic acid. *Plant Molecular Biology*, *97*(1–2), 177–185. https://doi.org/10.1007/s11103-018-0731-z

Wu, M., Liu, H., Lin, Y., Chen, J., Fu, Y., Luo, J., & Wang, F. (2020). In-frame and frame-shift editing of the *Ehd1* gene to develop japonica rice with prolonged basic vegetative growth periods. *Frontiers in Plant Science*, *11*, 307. https://doi.org/10.3389/fpls.2020.00307

Yin, K., & Qiu, J. L. (2019). Genome editing for plant disease resistance: Applications and perspectives. *Philosophical Transactions of the Royal Society B: Biological Sciences*, *374*(1767), 20180322. https://doi.org/10.1098/rstb.2018.0322

Yoon, Y. J., Venkatesh, J., Lee, J. H., Kim, J., Lee, H. E., Kim, D. S., & Kang, B. C. (2020). Genome editing of *eIF4E1* in tomato confers resistance to pepper mottle virus. *Frontiers in Plant Science*, *11*, 1098. https://doi.org/10.3389/fpls.2020.01098

Zhang, Y., Wang, X., Luo, Y., Zhang, L., Yao, Y., Han, L., & Li, Y. (2020). *OsABA8ox2*, an ABA catabolic gene, suppresses root elongation of rice seedlings and contributes to drought response. *Crop Journal*, *8*(3), 480–491. https://doi.org/10.1016/j.cj.2020.03.007

Zhao, G., Luo, C., Luo, J., Li, J., Gong, H., Zheng, X., & Wu, H. (2021). A mutation in *LacDWARF1* results in a GA-deficient dwarf phenotype in sponge gourd (*Luffa acutangula*). *Theoretical and Applied Genetics*, *134*(10), 3443–3457. https://doi.org/10.1007/s00122-021-03911-1

Zhou, J., Xin, X., He, Y., Chen, H., Li, Q., Tang, X., & Zhang, Y. (2019). Multiplex QTL editing of grain-related genes improves yield in elite rice varieties. *Plant Cell Reports*, *38*(4), 475–485. https://doi.org/10.1007/s00299-018-2340-3

5 Haploid Inducer-Mediated Genome Editing (HI-Edit) Technology for Climate Change

Desika J, Juliet Hepziba S, Deepa Dharsini, and Sree Vathsa Sagar US

5.1 INTRODUCTION

Traditional techniques like selection, hybridization, and backcrossing have been the cornerstone of crop improvement for centuries, making crop breeding an essential part of agricultural development. These methods have made a substantial contribution to raising yield, enhancing quality, and strengthening disease and pest resistance. However, traditional breeding is time-consuming and labor-intensive because it frequently takes several generations to achieve genetic stability (Akhtar et al., 2023). Although the 20th century saw a revolution in agriculture with the introduction of heterosis (hybrid vigor) and the creation of hybrid crops, issues like the limited genetic diversity in elite germplasm and the sluggish rate of trait incorporation remained (Garcia et al., 2025). The increasing need for resilient and high-yielding crops made the incorporation of contemporary tools necessary.

Biotechnological approaches to crop breeding have advanced significantly over the past few decades. Among these are marker-assisted selection (MAS), which uses molecular markers to enable accurate trait selection; double haploid (DH) technology, which allows homozygous lines to develop quickly in a single generation; tools for genome editing, such as CRISPR-Cas9, enable precise alteration of genetic sequences to target characteristics like drought tolerance, pest resistance, and nutrient use efficiency (Abdul Aziz & Masmoudi, 2025). Abiotic stressors that limit cultivation in marginal areas and lower yields, like heat, dryness, and salinity, are especially harmful to crops (Kabato et al., 2025). For instance, rice is more susceptible to flooding and salinity in coastal places (Wei et al., 2023); maize is sensitive to heat stress during flowering, which results in poor grain set (Waqas et al., 2021); and wheat yields are decreasing in areas with higher temperatures during grain filling (Dubey et al., 2020). This chapter aims to highlight the role of haploid induction and genome editing as transformative tools in modern crop breeding.

DOI: 10.1201/9781003597797-5

5.2 HAPLOID INDUCTION

5.2.1 What Is Haploid Induction?

Haploid induction is a plant breeding technique used to produce haploid plants, which possess a single set of chromosomes (n) instead of the usual diploid set (2n). Haploid induction can be achieved through two main approaches: *in vivo* (utilizing haploid inducer (HI) lines) and *in vitro* (anther culture, ovule culture, and related techniques) (Song et al., 2024).

5.2.2 What Is Haploid Inducer Line?

HI lines are naturally available lines, i.e. plant line having the property to trigger in planta production of haploid embryos after a conventional intra-specific cross with a line of interest. One sperm cell from the inducer gamete fails to fuse with the egg cell but triggers haploid embryogenesis. The uniparental haploid progeny lacks the HI line genome. *Natural inducer lines* have been used in various plants like maize (Coe Jr, 1959), barley (Kasha & Kao, 1970), tobacco (Burk et al., 1979), wheat (Laurie & Bennett, 1988).

5.2.3 Mechanisms Involved in Haploid Formation

5.2.3.1 Normal Fertilization in Maize Involves Double Fertilization

One sperm cell fuses with the egg cell to form the zygote. The other sperm cell fuses with central cells to form the endosperm (Faure et al., 2003).

5.2.3.2 In Haploid Induction

Mutations in inducer lines cause defects in the sperm's ability to contribute its genetic material to the zygote. The egg develops into a haploid embryo with only the maternal genome. The endosperm develops normally with contributions from both parents (Diwan et al., 2025).

5.3 TECHNIQUES FOR HAPLOID INDUCTION

5.3.1 *In Vitro* Techniques

In vitro techniques play a pivotal role in modern plant biotechnology by enabling rapid propagation, genetic improvement, and conservation of plant species. These methods include anther culture (producing haploid plants from microspores for homozygous line development; Islam et al., 2023), microspore culture (generating uniform haploid/doubled haploid (DH) plants in crops like wheat and *Brassica*; Hale et al., 2022), and ovule culture (rescuing hybrid embryos from wide crosses; Rogo et al., 2023). Wide hybridization leverages embryo rescue to introduce novel traits from distant species (Inomata, 1993), while HIGE (haploid induction via gametic embryogenesis) accelerates pure-line breeding (Germana, 2011). The bulbosum method exploits chromosome elimination in barley hybrids to derive haploids (Devaux, 2003), and androgenesis by stress treatment (e.g., heat/cold) induces embryogenesis in microspores (Islam & Tuteja, 2012). Collectively, these techniques enhance breeding efficiency, preserve genetic diversity, and facilitate trait introgression (Kaur et al., 2025).

5.3.2 *IN VIVO* TECHNIQUES

In vivo haploid induction techniques enable the direct production of haploid plants from living specimens, offering efficient alternatives to *in vitro* methods. Key approaches include (1) HI lines, where crosses with genetically modified lines (e.g., maize with altered *ZmPLA1/ZmDMP* genes) trigger haploid embryo formation (Song et al., 2024); (2) wide hybridization, exploiting chromosome elimination in distant crosses (e.g., wheat × maize) to yield haploids (Ishii et al., 2016); (3) gynogenesis, using irradiated pollen to stimulate maternal haploid embryo development (Kolesnikova et al., 2021); (4) centromere-mediated genome elimination, leveraging CRISPR-edited centromere proteins for targeted chromosome removal (Karimi-Ashtiyani, 2021); (5) chemical induction (e.g., colchicine) to induce parthenogenesis (Musazade et al., 2025); and (6) genome editing-assisted induction, where CRISPR modifies haploid-promoting genes (e.g., *DMP*) to enhance efficiency (Bhowmik & Bilichak, 2021). These methods accelerate breeding by enabling rapid homozygous line production, though challenges like species-specific limitations, regulatory hurdles, and variable induction rates persist (Zhang et al., 2023). The genetic modifications in crop plants, their targeted genes, traits, and transformation methods are indexed in Table 5.1. However, these genes can be targeted for genome editing-assisted haploid induction (Zhang et al., 2019).

5.4 COMBINATION OF HAPLOID INDUCER-MEDIATED GENOME EDITING

Haploid embryo development is triggered by HI lines, such as those containing mutations in the *MATERNAL HAPLOID (MTL)* gene. After chromosome doubling, altered alleles in haploid embryos become homozygous when paired with genome editing technologies such as CRISPR-Cas9. This method guarantees trait fixation, speeds up breeding cycles, and does away with the requirement for several generations of selfing. It is especially useful for enhancing yield attributes, pest resistance, and stress tolerance, offering a productive means of creating crop varieties that are both climate adaptable and high-performing (Li et al., 2025).

5.4.1 ADVANTAGES OF CLIMATE-RESILIENT BREEDING CROPS

HI-Edit has the ability to dramatically accelerate trait introgression, introducing new crop traits to market. Farmers may access these breakthroughs considerably more quickly because of HI-Edit's capacity to accelerate the breeding cycle while maintaining genetic stability, as it can take ten years or more to bring a novel trait to market. HI-Edit technology has many uses, from assisting crops in adjusting to environmental stressors like shifting weather patterns to maybe bringing back crop

TABLE 5.1
Genetic Modifications in Crop Plants: Targeted Genes, Traits, and Methods

Plant	Gene(s) Targeted	Traits	Method	References
Watermelon	*CIPSK1*	Enhanced resistance to *Fusarium oxysporum*	*Agrobacterium*-mediated	Khan et al. (2023)
Oryza sativa	*OSPUT1/2/3*	Herbicide resistance	*Agrobacterium*-mediated	Lyu et al. (2022)
Solanum lycopersicum	*SIEPSPS*	Herbicide resistance	*Agrobacterium*-mediated	Yang et al. (2022)
S. lycopersicum	*SIALSI, SIALS2*	Herbicide resistance	*Agrobacterium*-mediated	Yang et al. (2022)
S. lycopersicum	*Sipds1*	Herbicide resistance	*Agrobacterium*-mediated	Yang et al. (2022)
Rice	*Os8N3* *OsProDH* *OsGS3* *OsNAC45*	Disease resistance Thermotolerance Grain length Salt tolerance	Gene knockout and overexpression Site-directed mutagenesis	Gan and Ling (2022a)
Saccharum officinarum	*SOALS*	Herbicide resistance	Biolistic gene transfer	Oz et al. (2021)
O. sativa	*OsNAC006*	Heat tolerance	PEG-mediated	Wang et al. (2020a)
Apple	*MdDIPM4*	Disease resistance	Gene inactivation	Pompili et al. (2020)
Maize	*ZmPHYC1* *ZmPHYC2*	Flowering time/plant height	Gene knockout and overexpression	Li et al. (2020)
O. sativa	*OsPIN5b*, *GS3*, *OSMYB30*	Cold tolerance	*Agrobacterium*-mediated	Zeng et al. (2020)
Oilseed rape	*BnALS1*	Herbicide resistance	Base editing	Wu et al. (2020)
Soybean	*GmPRR37* *GmFT2a/5a*	Flowering time and regional adaptability	Site-directed mutagenesis	Wang et al. (2020b)
Tobacco	*NtHL1*	Hybrid lethality	Frameshift mutation	Ma et al. (2020)
Muskmelon	*CmPDS*	Albinism (CRISPR trial)	Gene knockout	Hooghvorst et al. (2019)
O. sativa	*OsPDS*	Heat tolerance	Gene gun	Nandy et al. (2019)
O. sativa	*OsPRP1*	Cold tolerance	*Agrobacterium*-mediated	Nawaz et al. (2019)

varieties that growers no longer prefer (Li et al., 2025). In only a few short years, CRISPR technology, of which HI-Edit is an application, has already made significant strides (Gan & Ling, 2022b). A related technique called Inducer Mediated Genome Editing (IMGE) has been published by another research team. The first HI-Edit study used *NP222RS*, which had a low transformation frequency (TF), low haploid induction rate (HIR), and a poor seed set. The IMGE/HI-Edit approach will be widely used to speed up crop breeding because most crops are resistant to transformation and efficiency is frequently genotype dependent. Moreover, the genetic materials generated are transgene-free and reduce the need for regulatory permission (Delzer et al., 2024).

5.5 APPLICATIONS OF HI-Edit FOR CLIMATE CHANGE

HI-Edit enables direct genomic modification of commercial crop varieties. The genome editing program HI-Edit tackles climate change in a number of ways. By introducing disease- and drought-resistant characteristics, enhancing photosynthesis, and encouraging effective water use, it increases crop resilience (Kelliher et al., 2017). Additionally, it creates climate-resilient livestock, which makes them more tolerant to harsh weather conditions. By decreasing waste and maximizing nutrient uptake, it also makes precision agriculture possible. In addition, HI-Edit promotes the growth of carbon-sequestering microbes and bioenergy crops, helping to create a sustainable and climate-resilient future. By reducing the effects of climate change on biodiversity, ecosystems, and global food security, these applications guarantee a more sustainable planet. The potential of HI-Edit is enormous and promising (Dubey & Jha, 2025). The identification of *MATRILINEAL (MTL)*, a pollen-specific phospholipase (Kelliher et al., 2017), marks a significant milestone in maize haploid induction, with far-reaching implications for crop improvement in the face of climate change. The discovery that haploid induction results from a 4-bp insertion in the carboxy terminus of the *MTL* gene provides crucial insights into genetic mechanisms underlying efficient DH production.

Fine mapping of the locus to a 0.57 Mb region and the subsequent renaming of *GRMZM2G471240 as MATRILINEAL* highlight the precision of modern molecular tools in advancing genetic breeding programs. Using TALEN technology, mutant MTL lines (*MTLTAL-FS*) were developed, achieving HIRs of 4.0–12.5%, further validating the role of the MTL allele in triggering haploidy (Kelliher et al., 2017). These findings were reinforced through mapping populations derived from the RWK line (developed from Stock 6) crossed with NP2460 and NP2391. Additionally, Kelliher et al. (2019) extended the application of haploid induction systems by demonstrating their utility in wide crosses between wheat (*Triticum aestivum*) and maize (*Zea mays*). By using maize HI pollen, haploid wheat embryos were generated through zygotic elimination of maize chromosomes during mitosis (Dermail et al., 2024).

Scientists (Yao et al., 2018) explored haploid induction in rice by leveraging insights from maize (*Z. mays*), where a native frameshift mutation in the *MATRILINEAL* (*MATL*) gene triggers haploid induction. The rice ortholog, *OsMATL*, exhibited an expression pattern similar to *ZmMATL*. Using genome editing, an allelic series of *OsMATL* mutants was developed. Knockout mutations in *OsMATL* resulted in a reduced seed set and achieved HIRs of 2–6% (Yao et al., 2018). This study demonstrates the potential of genome editing for developing HIs in rice, providing a foundation for accelerated breeding and genetic studies in cereal crops. Researchers converted a non-HI maize line, NP2222, into a HI line using CRISPR-Cas9 with a guide RNA to knock out the *MATL* gene. The edited plants were self-pollinated, and T1 progeny homozygous for *MATL* frameshift mutations and the *Cas9* construct were selected. These mutated HI plants were crossed with the inbred line B14-vL, and haploid progeny were identified, showing no male genome contribution (Liu et al., 2025).

5.5.1 Applications for Trait-Based Example

Scientists (Wang et al., 2019) developed a method using the CAU5 HI line carrying a CRISPR/Cas9 cassette targeting *ZmLG1* or *UB2* genes. Crossing the HI line with the B73 maize line produced genome-edited haploids. These haploids were Cas9-free due to the loss of the male genome post-fertilization. The edited haploids could be doubled, either spontaneously or artificially, to generate DH lines. This process enabled the production of homozygous DH lines with improved agronomic traits within two generations, offering an efficient tool for precise gene editing and breeding in maize (Dermail et al., 2024).

5.5.2 Other Applications

Haploid induction has been transformed by recent developments in genetic alterations, providing effective instruments for crop improvement. The role of the SlDMP mutation in tomatoes was illustrated by Zhong et al. (2022), when biallelic mutants produced maternal haploids without regard to genotype. With flow cytometry verifying haploid induction, the study revealed decreased seed filling and higher ovule abortion. In a similar manner, Yin et al. (2024) used CRISPR-Cas9 to modify cucumber's CsDMP, thereby creating haploids through mutant selfing. With possible uses in breeding programs, this was the first successful *in vivo* haploid induction technique in cucumbers. The importance of reactive oxygen species (ROS) in haploid induction was emphasized by Jiang et al. (2022), who found that increased ROS levels in MTL mutants resulted in DNA fragmentation in sperm cells, producing haploid offspring. Through CENH3 alterations, temperature manipulation enhanced HIRs in *Arabidopsis*, as shown by Wang et al. (2023).

Male parental HIRs dramatically increased with environmental conditions optimized, indicating new ways to improve efficiency. By preventing zygote formation during fertilization, changes in the *KPL* gene enabled haploid creation, according to research by Jacquier et al. (2023) on genetic disturbances in gamete fusion. Numerous crops, including watermelon, tomatoes, maize, and potatoes, have benefited from the successful application of DMP modifications. These developments combine the advantages of genome editing and haploid induction, allowing for quick trait fixation and hastening the creation of stable and enhanced agricultural varieties. With wider ramifications for global food security, these discoveries are especially important for improving breeding efficiency in intricate plant groups like *Solanaceae*, *Fabaceae*, and *Cucurbitaceae* (Li et al., 2025).

5.6 REGULATORY AND ETHICAL CONSIDERATIONS

The regulatory landscape for genome-edited crops varies significantly across jurisdictions, reflecting divergent interpretations of genetic engineering laws. The European Union applies stringent GMO regulations to all genome-edited organisms, regardless of foreign DNA presence, while more permissive regimes like the USA and Canada exempt edits lacking transgenes (Sprink & Wilhelm, 2023). Intermediate approaches exist in countries such as Australia and Japan, which have established subcategories for certain editing techniques. International efforts to harmonize standards include the WHO's Expert Advisory Committee, which addresses ethical and governance challenges of human genome editing, and the OECD's 2019 conference, which facilitated multidisciplinary dialogue on agricultural applications (Friedrichs et al., 2019). Ongoing revisions to GMO laws in Europe and elsewhere continue to shape a fragmented regulatory environment, creating barriers to global trade and innovation in genome-edited products (Sprink & Wilhelm, 2023).

5.7 FUTURE PROSPECTS

HI-Edit (haploid induction-assisted genome editing) holds significant promise for addressing climate challenges by enhancing crop resilience and sustainability, with projections suggesting it could boost yields by 20–30% in drought-prone regions by 2030 (Ndudzo et al., 2024). This technology is expected to revolutionize climate-smart agriculture through drought-tolerant crops, heat-resistant livestock, and AI-integrated precision farming, while also enabling carbon sequestration in plants to mitigate climate change (Lal et al., 2015). Regulatory reforms are anticipated to facilitate widespread adoption, ensuring equitable access to these innovations and supporting global food security (Han et al., 2025). Beyond agriculture, HI-Edit's applications span pharmaceuticals and ecosystem restoration, transforming vulnerability into resilience as climate pressures intensify (Kelliher et al., 2017). Its continued advancement will be critical in building a sustainable, food-secure future by improving nutrition, adaptation, and agricultural productivity worldwide.

5.8 CONCLUSION

The development of DH lines through haploid induction has revolutionary implications for agricultural adaptation to climate change. This technology enables the rapid stacking of stress-resilient traits, such as heat tolerance and drought tolerance, into elite germplasm. The combination of haploid induction and genome editing technologies, like CRISPR-Cas9, provides a powerful platform for integrating climate-resilient alleles into crops. This technology has the potential to expand the genetic basis of staple crops, increasing their resilience and productivity under challenging conditions. Future research should focus on integrating haploid induction with high-throughput phenotyping and precision breeding to accelerate the development of climate-smart crop varieties.

REFERENCES

Abdul Aziz, M., & Masmoudi, K. (2025). Molecular breakthroughs in modern plant breeding techniques. *Horticultural Plant Journal*, *11*(1), 15–41. https://doi.org/10.1016/j.hpj.2024.01.004

Akhtar, S., Rao, E., Uike, A., & Saatu, M. (2023). Plant breeding strategies: Traditional and modern approaches. In *Genetic revolution in agriculture: Unleashing the power of plant genetics*. Elite Publishing House.

Bhowmik, P., & Bilichak, A. (2021). Advances in gene editing of haploid tissues in crops. *Genes*, *12*(9), 1410. https://doi.org/10.3390/genes12091410

Burk, L., Chaplin, J., Gooding, G., & Powell, N. (1979). Quantity production of anther-derived haploids from a multiple disease resistant tobacco hybrid. I. Frequency of plants with resistance or susceptibility to tobacco mosaic virus (TMV), potato virus Y (PVY), and root knot (RK). *Euphytica*, *28*, 201–208.

Coe Jr, E. (1959). A line of maize with high haploid frequency. *The American Naturalist*, *93*(873), 381–382.

Delzer, B., Liang, D., Szwerdszarf, D., Rodriguez, I., Mardones, G., Elumalai, S., & Kelliher, T. (2024). Elite, transformable haploid inducers in maize. *The Crop Journal*, *12*(1), 314–319. https://doi.org/10.1016/j.cj.2023.10.016

Dermail, A., Mitchell, M., Foster, T., Fakude, M., Chen, Y.-R., Suriharn, K., & Lübberstedt, T. (2024). Haploid identification in maize. *Frontiers in Plant Science*, *15*. https://doi.org/10.3389/fpls.2024.1378421

Devaux, P. (2003). The *Hordeum bulbosum* (L.) Method. In M. Maluszynski, K. J. Kasha, B. P. Forster, and I. Szarejko (Eds.), *Doubled haploid production in crop plants* (pp. 15–19). Springer. https://doi.org/10.1007/978-94-017-1293-4_3

Diwan, G., Rawte, S., & Jha, Z. (2025). Haploids: Then and now. In Z. Jha, S. B. Verulkar, and S. Penna (Eds.), *Doubled haploids: Technological advances and role in crop improvement* (pp. 1–56). Springer Nature. https://doi.org/10.1007/978-981-96-2339-6_1

Dubey, M., & Jha, Z. (2025). Advancements in haploid techniques. In Z. Jha, S. B. Verulkar, & S. Penna (Eds.), *Doubled haploids: Technological advances and role in crop improvement* (pp. 139–158). Springer Nature. https://doi.org/10.1007/978-981-96-2339-6_4

Dubey, R., Pathak, H., Chakrabarti, B., Singh, S., Gupta, D. K., & Harit, R. C. (2020). Impact of terminal heat stress on wheat yield in India and options for adaptation. *Agricultural Systems*, *181*, 102826. https://doi.org/10.1016/j.agsy.2020.102826

Faure, J.-E., Rusche, M. L., Thomas, A., Keim, P., Dumas, C., Mogensen, H. L., & Chaboud, A. (2003). Double fertilization in maize: The two male gametes from a pollen grain have the ability to fuse with egg cells. *The Plant Journal*, *33*(6), 1051–1062. https://doi.org/10.1046/j.1365-313X.2003.01692.x

Friedrichs, S., Takasu, Y., Kearns, P., Dagallier, B., Oshima, R., Schofield, J., & Moreddu, C. (2019). An overview of regulatory approaches to genome editing in agriculture. *Biotechnology Research and Innovation*, *3*(2), 208–220.

Gan, W. C., & Ling, A. P. (2022a). CRISPR/Cas9 in plant biotechnology: Applications and challenges. *BioTechnologia*, *103*(1), 81.

Gan, W. C., & Ling, A. P. K. (2022b). CRISPR/Cas9 in plant biotechnology: Applications and challenges. *BioTechnologia*, *103*(1), 81–93. https://doi.org/10.5114/bta.2022.113919

Garcia, A. A. F., Frisch, M., Weng, Y., Varshney, R., Sorrells, M., & Fang, D. D. (2025). Heterosis and hybrid breeding. *Theoretical and Applied Genetics*, *138*(4), 69. https://doi.org/10.1007/s00122-025-04834-x

Germana, M. A. (2011). Gametic embryogenesis and haploid technology as valuable support to plant breeding. *Plant Cell Reports*, *30*, 839–857.

Hale, B., Ferrie, A. M. R., Chellamma, S., Samuel, J. P., & Phillips, G. C. (2022). Androgenesis-based doubled haploidy: Past, present, and future perspectives. *Frontiers in Plant Science*, *12*. https://doi.org/10.3389/fpls.2021.751230

Han, X., Deng, Z., Liu, H., & Ji, X. (2025). Current advancement and future prospects in simplified transformation-based plant genome editing. *Plants*, *14*(6), 889.

Hooghvorst, I., López-Cristoffanini, C., & Nogués, S. (2019). Efficient knockout of phytoene desaturase gene using CRISPR/Cas9 in melon. *Scientific Reports*, *9*(1), 17077.

Inomata, N. (1993). Embryo rescue techniques for wide hybridization. In K. S. Labana, S. S. Banga, and S. K. Banga (Eds.), *Breeding Oilseed Brassicas* (Vol. 19, pp. 94–107). Springer. https://doi.org/10.1007/978-3-662-06166-4_7

Ishii, T., Karimi-Ashtiyani, R., & Houben, A. (2016). Haploidization via chromosome elimination: Means and mechanisms. *Annual Review of Plant Biology*, *67*, 421–438. https://doi.org/10.1146/annurev-arplant-043014-114714

Islam, M. T., Arif, M. R., Hasan, M. T., & Robin, A. H. K. (2023). Anther culture in crop plants: Progress and perspectives. *Plant Breeding and Biotechnology*, *11*(2), 69–96. https://doi.org/10.9787/PBB.2023.11.2.69

Islam, S. M. S., & Tuteja, N. (2012). Enhancement of androgenesis by abiotic stress and other pretreatments in major crop species. *Plant Science*, *182*, 134–144. https://doi.org/10.1016/j.plantsci.2011.10.001

Jacquier, N. M., Calhau, A. R., Fierlej, Y., Martinant, J.-P., Rogowsky, P. M., Gilles, L. M., & Widiez, T. (2023). *In planta* haploid induction by *kokopelli* mutants. *Plant Physiology*, *193*(1), 182–185.

Jiang, C., Sun, J., Li, R., Yan, S., Chen, W., Guo, L., & Huang, W. (2022). A reactive oxygen species burst causes haploid induction in maize. *Molecular Plant*, *15*(6), 943–955.

Kabato, W., Getnet, G. T., Sinore, T., Nemeth, A., & Molnár, Z. (2025). Towards climate-smart agriculture: Strategies for sustainable agricultural production, food security, and greenhouse gas reduction. *Agronomy*, *15*(3), 565.

Karimi-Ashtiyani, R. (2021). Centromere engineering as an emerging tool for haploid plant production: Advances and challenges. In *Doubled haploid technology (vol.* 3, pp. 3–22). Springer. https://doi.org/10.1007/978-1-0716-1331-3_1

Kasha, K., & Kao, K. (1970). High frequency haploid production in barley (*Hordeum vulgare* L.). *Nature*, *225*(5235), 874–876.

Kaur, T., Sharma, V., & Penna, S. (2025). Induction of maternal haploids in plants. In Z. Jha, S. B. Verulkar, and S. Penna (Eds.), *Doubled haploids: Technological advances and role in crop improvement* (pp. 85–138). Springer Nature. https://doi.org/10.1007/978-981-96-2339-6_3

Kelliher, T., Starr, D., Richbourg, L., Chintamanani, S., Delzer, B., Nuccio, M. L., & Wang, W. (2017). *MATRILINEAL*, a sperm-specific phospholipase, triggers maize haploid induction. *Nature*, *542*(7639), 105–109.

Kelliher, T., Starr, D., Su, X., Tang, G., Chen, Z., Carter, J., & Burch, E. (2019). One-step genome editing of elite crop germplasm during haploid induction. *Nature Biotechnology*, *37*(3), 287–292.

Khan, Z., Shahwar, D., & Heikal, Y. (2023). *Genome editing and global food security: Molecular engineering technologies for sustainable agriculture*. Taylor and Francis.

Kolesnikova, E. O., Donskikh, E. I., & Berdnikov, R. V. (2021). Haploid biotechnology as a tool for creating a selection material for sugar beets. *Vavilov Journal of Genetics and Breeding*, *25*(8), 812–821. https://doi.org/10.18699/VJ21.094

Lal, R., Negassa, W., & Lorenz, K. (2015). Carbon sequestration in soil. *Current Opinion in Environmental Sustainability*, *15*, 79–86. https://doi.org/10.1016/j.cosust.2015.09.002

Laurie, D. A., & Bennett, M. (1988). The production of haploid wheat plants from wheat × maize crosses. *Theoretical and Applied Genetics*, *76*, 393–397.

Li, L., Fu, X., Qi, X., Xiao, B., Liu, C., Wu, Q., & Xie, C. (2025). Harnessing haploid-inducer mediated genome editing for accelerated maize variety development. *Plant Biotechnology Journal*. https://doi.org/10.1111/pbi.14608

Li, Q., Wu, G., Zhao, Y., Wang, B., Zhao, B., Kong, D., & Wang, H. (2020). CRISPR/Cas9-mediated knockout and overexpression studies reveal a role of maize phytochrome C in regulating flowering time and plant height. *Plant Biotechnology Journal*, *18*(12), 2520–2532. https://doi.org/10.1111/pbi.13429

Liu, Y., Elshan, M., Li, G., Han, X., Chen, X., & Feng, X. (2025). Perspectives of genome editing mediated haploid inducer systems in legumes. *International Journal of Molecular Sciences*, *26*(3), 1154. https://doi.org/10.3390/ijms26031154

Lyu, Y.-S., Cao, L.-M., Huang, W.-Q., Liu, J.-X., & Lu, H.-P. (2022). Disruption of three polyamine uptake transporter genes in rice by CRISPR/Cas9 gene editing confers tolerance to herbicide paraquat. *aBIOTECH*, *3*(2), 140–145. https://doi.org/10.1007/s42994-022-00075-4

Ma, J., Hancock, W. G., Nifong, J. M., Kernodle, S. P., & Lewis, R. S. (2020). Identification and editing of a hybrid lethality gene expands the range of interspecific hybridization potential in *Nicotiana*. *Theoretical and Applied Genetics*, *133*(10), 2915–2925. https://doi.org/10.1007/s00122-020-03641-w

Musazade, E., Liu, Y., Chen, X., Gao, J., Wang, M., Han, X., & Feng, X. (2025). Advances and challenges in haploid induction for warm-season legumes. *Journal of Agricultural and Food Chemistry*, *73*(11), 6315–6332. https://doi.org/10.1021/acs.jafc.4c10447

Nandy, S., Pathak, B., Zhao, S., & Srivastava, V. (2019). Heat-shock-inducible CRISPR/Cas9 system generates heritable mutations in rice. *Plant Direct*, *3*(5), e00145. https://doi.org/10.1002/pld3.145

Nawaz, G., Han, Y., Usman, B., Liu, F., Qin, B., & Li, R. (2019). Knockout of *OsPRP1*, a gene encoding proline-rich protein, confers enhanced cold sensitivity in rice (*Oryza sativa* L.) at the seedling stage. *3 Biotech*, *9*(7), 254. https://doi.org/10.1007/s13205-019-1787-4

Ndudzo, A., Sibanda Makuvise, A., Moyo, S., & Bobo, E. D. (2024). CRISPR-Cas9 genome editing in crop breeding for climate change resilience: Implications for smallholder farmers in Africa. *Journal of Agriculture and Food Research*, *16*, 101132. https://doi.org/10.1016/j.jafr.2024.101132

World Health Organization (WHO). (2021). *Human genome editing: A framework for governance*. World Health Organization. https://apps.who.int/iris/bitstream/handle/10665/342484/9789240030060-eng.pdf

Oz, M. T., Altpeter, A., Karan, R., Merotto, A., & Altpeter, F. (2021). CRISPR/Cas9-mediated multi-allelic gene targeting in sugarcane confers herbicide tolerance. *Frontiers in Genome Editing*, *3*, 673566.

Pompili, V., Dalla Costa, L., Piazza, S., Pindo, M., & Malnoy, M. (2020). Reduced fire blight susceptibility in apple cultivars using a high-efficiency CRISPR/Cas9-FLP/FRT-based gene editing system. *Plant Biotechnology Journal*, *18*(3), 845–858. https://doi.org/10.1111/pbi.13253

Rogo, U., Fambrini, M., & Pugliesi, C. (2023). Embryo rescue in plant breeding. *Plants*, *12*(17), 3106. https://doi.org/10.3390/plants12173106

Song, J., Datla, R., Zou, J., & Xiang, D. (2024). Haploid induction: An overview of parental factor manipulation during seed formation. *Frontiers in Plant Science*, *15*, 1439350. https://doi.org/10.3389/fpls.2024.1439350

Sprink, T., & Wilhelm, R. (2023). Genome editing in biotech regulations worldwide. In *A roadmap for plant genome editing* (pp. 425–435). Springer Nature.

Wang, B., Zhu, L., Zhao, B., Zhao, Y., Xie, Y., Zheng, Z., & Wang, H. (2019). Development of a haploid-inducer mediated genome editing system for accelerating maize breeding. *Molecular Plant*, *12*(4), 597–602.

Wang, B., Zhong, Z., Wang, X., Han, X., Yu, D., Wang, C., & Zhang, Y. (2020a). Knockout of the *OsNAC006* transcription factor causes drought and heat sensitivity in rice. *International Journal of Molecular Sciences*, *21*(7), 2288.

Wang, L., Sun, S., Wu, T., Liu, L., Sun, X., Cai, Y., & Han, T. (2020b). Natural variation and CRISPR/Cas9-mediated mutation in *GmPRR37* affect photoperiodic flowering and contribute to regional adaptation of soybean. *Plant Biotechnology Journal*, *18*(9), 1869–1881. https://doi.org/10.1111/pbi.13346

Wang, J., Wang, X.-F., Yang, W.-C., & Li, H.-J. (2023). Loss of function of *CENH3* causes genome instability in soybean. *Seed Biology*, *2*(1). https://doi.org/10.48130/SeedBio-2023-0024

Waqas, M. A., Wang, X., Zafar, S. A., Noor, M. A., Hussain, H. A., Azher Nawaz, M., & Farooq, M. (2021). Thermal stresses in maize: Effects and management strategies. *Plants*, *10*(2), 293. https://doi.org/10.3390/plants10020293

Wei, H., Geng, X., Zhu, W., Zhang, X., Zhang, X., Chen, Y., & Dai, Q. (2023). Individual and combined influences of salinity and drought stress on the agro-physiological traits and grain yield of rice. *Field Crops Research*, *304*, 109172. https://doi.org/10.1016/j.fcr.2023.109172

Wu, J., Chen, C., Xian, G., Liu, D., Lin, L., Yin, S., & Wang, Y. (2020). Engineering herbicide-resistant oilseed rape by CRISPR/Cas9-mediated cytosine base-editing. *Plant Biotechnology Journal*, *18*(9), 1857.

Yang, S. H., Kim, E., Park, H., & Koo, Y. (2022). Selection of the high efficient sgRNA for CRISPR-Cas9 to edit herbicide related genes, *PDS, ALS*, and *EPSPS* in tomato. *Applied Biological Chemistry*, *65*(1), 13. https://doi.org/10.1186/s13765-022-00679-w

Yao, L., Zhang, Y., Liu, C., Liu, Y., Wang, Y., Liang, D., & Kelliher, T. (2018). *OsMATL* mutation induces haploid seed formation in indica rice. *Nature Plants*, *4*(8), 530–533.

Yin, S., Li, S., Sun, L., Shi, K., Fan, S., Liu, X., & Ren, H. (2024). Mutating the maternal haploid inducer gene *CsDMP* in cucumber produces haploids *in planta*. *Plant Physiology*, *194*(3), 1282–1285.

Zeng, Y., Wen, J., Zhao, W., Wang, Q., & Huang, W. (2020). Rational improvement of rice yield and cold tolerance by editing the three genes *OsPIN5b, GS3*, and *OsMYB30* with the CRISPR–Cas9 system. *Frontiers in Plant Science*, *10*, 1663.

Zhang, J., Liu, J., Yang, W., Cui, M., Dai, B., Dong, Y., & Liang, H. (2019). Comparison of gene editing efficiencies of CRISPR/Cas9 and TALEN for generation of *MSTN* knockout cashmere goats. *Theriogenology*, *132*, 1–11.

Zhang, X., Shi, C., Li, S., Zhang, B., Luo, P., Peng, X., & Sun, M.-X. (2023). A female *in vivo* haploid-induction system via mutagenesis of egg cell-specific peptidases. *Molecular Plant*, *16*(2), 471–480. https://doi.org/10.1016/j.molp.2023.01.001

Zhong, Y., Chen, B., Wang, D., Zhu, X., Li, M., Zhang, J., & Liu, J. (2022). *In vivo* maternal haploid induction in tomato. *Plant Biotechnology Journal*, *20*(2), 250–252.

6 Next-Generation Sequencing and Crop Genomics for Climate Change Adaptation

Tanya Barpanda, Sourav Ranjan Nanda,
Laxmipreeya Behera, Manasi Dash,
and Abinash Mishra

6.1 INTRODUCTION

Climate change has emerged as one of the most pressing challenges facing global agriculture. Rising temperatures, unpredictable rainfall patterns, and increased incidences of extreme weather events have severely impacted crop productivity and food security (Jovović et al., 2025). Prolonged droughts, floods, and heatwaves are altering crop seasons, increasing pest infestations, and reducing arable land availability (Bera et al., 2024). Consequently, developing climate-resilient crop varieties has become a critical priority to ensure global food security. Advances in genomic technologies provide promising tools to accelerate crop improvement strategies and mitigate the impact of climate change on agriculture. By the use of genomic innovations, researchers can identify key genetic loci associated with adaptive traits such as drought tolerance, heat resistance, and nutrient-use efficiency (Benavente & Giménez, 2021; Saleem et al., 2025; Schröder et al., 2019). Next-generation sequencing (NGS) has revolutionized crop genomics by enabling rapid and cost-effective sequencing of entire genomes, facilitating a deeper understanding of genetic variability. Technologies such as genotyping-by-sequencing (GBS) and single nucleotide polymorphism (SNP) arrays have enhanced the precision and efficiency of identifying climate-resilient traits in crop species (Kim et al., 2016; Naqvi et al., 2022).

Additionally, advancements in pangenomics have allowed researchers to capture the genetic diversity across multiple cultivars, landraces, and wild relatives, revealing valuable alleles for climate adaptation (Hufford et al., 2021; Liu et al., 2020). The integration of these genomic approaches has played a transformative role in developing improved crop varieties with enhanced resilience to environmental stresses. Furthermore, emerging technologies such as genomic selection (GS) and genome editing tools like CRISPR/Cas, TALENs, and zinc finger nucleases (ZFNs) are rapidly accelerating the development of climate-resilient crops (Bhat et al., 2016; Feng et al., 2013; Wang et al., 2018). These innovative tools enable breeders to target genes

DOI: 10.1201/9781003597797-6

linked to climate adaptation traits with remarkable precision, expediting the breeding cycle. This chapter outlines the pivotal role of NGS, GS, genome editing, and data management tools in driving climate-resilient agriculture, offering insights into future research directions for ensuring global food security in the face of an evolving climate crisis.

6.2 GENOME SEQUENCING AND ASSEMBLIES

The draft human genome was completed in 2001, which pushed genome sequencing into the limelight. Initially, Sanger sequencing was frequently used to draft plant genomes (Imelfort & Edwards, 2009). Despite its long length of reads and subsequent high accuracy in sequence assembly, the high cost and low throughput associated with its use limit this sequencing technology in large-scale adoption (Metzker, 2005). Hence, this technology was gradually replaced with more advanced sequencing platform with the likes of Illumina sequencing, thus forming the second-generation sequencing (SGS) platform. High throughput and low cost associated with Illumina sequencing were the reason for its faster adoption among the scientific community associated with genome assembly (Goodwin et al., 2016). As per the National Centre for Biotechnology Information (NCBI) database, till date over 100 of reference genomes have been sequenced for various plant species. Most of these genomes have been sequenced with the help of SGS. However, the short-read lengths generated and inaccuracy along with gaps during the assembly in long repetitive regions undermines the quality of SGS (Treangen & Salzberg, 2012). There is a possibility of many genes being divided among contigs, which may lead to an inflating of gene counts. This can impair the accuracy of gene predictions. Such misassembles and split genes in assemblies are a significant limiting factor for subsequent analyses like pangenomics and genome diversity studies (Denton et al., 2014).

6.2.1 Long-Read Sequencing

Long-read sequencing technologies, such as PacBio and Oxford Nanopore, generate reads spanning thousands of bases, enabling the analysis of complex genomic regions and full-length mRNA transcripts for improved variant detection and exon linkage studies (Goodwin et al., 2016). Unlike short-read sequencing, these single-molecule approaches eliminate PCR amplification biases, enhancing accuracy (Schadt et al., 2010). PacBio sequencing, commercialized in 2011, produces reads averaging 10 kb but suffers from high indel error rates (13%–18%) and occasional chimeric reads, which can be mitigated through increased coverage or quality control measures (Berlin et al., 2015; Carneiro et al., 2012; Tallon et al., 2014). Introduced in 2014, Oxford Nanopore's MinION platform sequences DNA fragments exceeding 100 kb but also exhibits indel errors (~15%), prompting the development of error-correction tools like NanoCorr, NanoPolish, PoreSeq, and marginAlign (Goodwin et al., 2015; Ip et al., 2015; Jain et al., 2015; Loman et al., 2015; Szalay & Golovchenko, 2015; Urban et al., 2015). Synthetic long-read technologies, such as Illumina's HiSeq and 10× Genomics' GemCode, assemble short reads into longer sequences, with Illumina using TruSeq libraries and 10× Genomics employing microfluidics for DNA partitioning and barcoding (Eisenstein, 2015; McCoy et al., 2014). These advancements address challenges in genome assembly and transcriptome analysis, though error rates and technical limitations remain key considerations.

6.2.2 Optical Mapping

Optical mapping creates maps that can enhance genome assemblies by using the actual locations of restriction enzyme sites. OpGen Argus and BioNano Irys are now the leading platforms under this category, which was first documented in the early 1990s (Schwartz et al., 1993). The optical maps created by OpGen span a mean length of 200 kb, but the typical length of BioNano maps is approximately 225 kb (Shelton et al., 2015). The overlap-layout-consensus method uses *de novo* assembly to produce a final, precise DNA map. Then, in order to identify assembly defects such as erroneous joins, inversions, or translocations, these maps are compared to a reference sequence. The results are visualized using programs such as OpGen MapSolver and BioNano IrysView. Additionally, optical mapping can be used to fill in assembly gaps and connect smaller scaffolds to form bigger, more comprehensive ones (Cao et al., 2014; Hastie et al., 2013). This sequencing platform has been employed for rice, maize, tomato, and wheat (Shearer et al., 2014; Zhou et al., 2007, 2009).

6.3 ADVANCES IN CAPTURING CROP DIVERSITY

The analysis of genetic variation among and between species is the starting point of crop improvement programs (Staňková et al., 2016).

6.3.1 Genotyping-by-Sequencing (GBS)

Reduced representation technologies, such as GBS, provide a cost-effective alternative to whole-genome sequencing by targeting specific genomic regions (Elshire et al., 2011). GBS begins with restriction enzyme digestion of genomic DNA, where enzyme selection determines the sequenced regions, often favoring methylation-sensitive enzymes to avoid repetitive DNA and enrich informative sequences (Koren & Phillippy, 2015). The digested fragments are ligated with adapters, one common and one barcoded for sample identification, followed by PCR amplification to prepare the library for Illumina sequencing (Li et al., 2015; Wallace & Mitchell, 2017). An alternative GBS protocol uses two restriction enzymes for better fragment size and specificity control (Poland et al., 2012). GBS reduces sequencing costs by focusing on key genomic regions and enables simultaneous SNP discovery and genotyping without prior sequence knowledge, making it valuable for orphan crops. However, a limitation is the potential omission of important regions lacking restriction sites (Kim et al., 2016).

6.3.2 GENOTYPING ARRAYS (SNP ARRAY TECHNOLOGY)

With the advent of whole-genome sequencing, many crop species have been completely sequenced, especially those with relatively smaller genomes. This has allowed us to compare the sequence information of a particular genome with a reference genome or between two different individuals and comprehensively identify SNPs (Varshney et al., 2009). SNP array technology has allowed the genotyping of crop samples with millions of SNPs in one go. SNP arrays have now been established for several important crops like canola, maize, rice, and wheat (Chen et al., 2014; Edwards et al., 2013; Snowdon & Iniguez Luy, 2012; Yu et al., 2014). SNP array technology can help in the gene localization of traits, which can enable the discovery of very tightly linked markers which can be further used in marker-assisted selection (Felcher et al., 2012). It can also help in very precise and unanimous variety identification. This technology will also facilitate GS as explained in the next section (Clarke et al., 2016). It has been used in genome-wide association studies (GWAS) (McCouch et al., 2016), population structure analysis (Wang et al., 2016), and gene mapping (Dalton-Morgan et al., 2014).

6.3.3 PANGENOMICS

Crop genomes are dynamic. They keep changing with the changing climate, with domestication and crop improvement. In order to develop future ready crop varieties, the study of genetic diversity of a single genome is not sufficient (Bayer et al., 2020). Some important climate-resilient traits may be present in crop wild relatives. These may be adapted to extreme environments and may carry useful genes for climate resilience that were lost during domestication. The study of pangenomes will also help us understand the impact of domestication and crop improvement on crops. Pangenomes have been constructed for several crops, like soybean (Liu et al., 2020), tomato (Gao et al., 2019), cotton (Li et al., 2021), rice (Zhao et al., 2018), chickpea (Varshney et al., 2021), and maize (Hufford et al., 2021). A summary of the different sequencing methods is given in Table 6.1.

6.3.4 DATA MANAGEMENT SYSTEM FOR CROP GENOMICS

Crop genomics gives rise to a vast quantity of data which needs to be stored, analyzed, and visualized as and when needed. This has given rise to the branch of bioinformatics which deals with the application of computer technology to manage large biological information. One of the oldest data library is the European Molecular Biology Laboratory (EMBL), established in 1980 (Higgins et al., 1992). The NCBI, USA is one of the most popularly used databank in the world. DNA databank in Japan and National Biomedical Research Foundation's Protein Information Resource (PIR) for proteins are some other databanks. Today, there are three most popular public domain databases, the NCBI, the European Bioinformatics Institute, UK, and GenomeNet, Japan (Singh, 2015). These contain nucleotide sequence databases like GenBank, dbEST, *Escherichia coli*, Mito, EMBL, Kabat, Yeast, IMGT database (Singh & Singh, 2015). In addition, there

TABLE 6.1
Sequencing Technologies and Their Applications in Crop Genomics for Climate Change Adaptation

Technology	Key Features	Limitations	Applications in Crop Genomics	References
Sanger sequencing	High accuracy, long-read lengths	High cost, low throughput	Early genome assemblies, foundational technology for crop genomics	Imelfort and Edwards (2009), Metzker (2005)
Second-generation sequencing	High throughput, cost-effective	Short-read lengths, assembly gaps	Sequencing plant genomes, genotyping-by-sequencing (GBS), SNP discovery	Goodwin et al. (2016)
PacBio sequencing	Long reads (~10 kb), single-molecule sequencing	High error rates, costly	Resolving repetitive regions, full-length transcript sequencing	Schadt et al. (2010), Lee et al. (2014), Rhoads and Au (2015)
Oxford Nanopore sequencing	Ultra-long reads (>100 kb), portable	High indel error rates	Structural variation analysis, improving genome assemblies	Ip et al. (2015), Jain et al. (2015)
Genotyping-by-sequencing	Focus on specific genome regions, cost-effective	Misses non-restricted regions	SNP discovery, marker-assisted breeding, studying orphan crops	Elshire et al. (2011), Poland et al. (2012)
SNP array technology	High-density SNP genotyping, robust detection of polymorphisms	Limited to predefined SNPs	Genome-wide association studies (GWAS), trait mapping, population structure analysis	Clarke et al. (2016), Varshney et al. (2009)
Optical mapping	Physical genome mapping, complements sequencing for assembly	Limited resolution for small genomic features	Filling assembly gaps, detecting structural variations, aligning scaffolds	Hastie et al. (2013), Cao et al. (2014)
Pangenomics	Comparative genome analysis across individuals	Requires extensive data and computational resources	Identifying climate-resilience genes, understanding crop domestication	Bayer et al. (2020), Liu et al. (2020), Hufford et al. (2021)

are protein databases like the Protein Data Bank (PDB), SWISS-PROT database, Yeast, and Kabat (Minouchehr & Gollaei, 2004). The utilization if these databases require database mining and analysis tools. For example, the Basic Local Alignment Search Tool (BLAST) is the most preferred sequence alignment tool in the public domain (McGinnis & Madden, 2004). ENTREZ is a search engine for bibliographical citations and biological data (Schuler et al., 1996).

TAXONOMY BROWSER is used to search for taxonomic information of various species (Leow & Taylor, 2000). LOCUS LINK provides information about genes (Pruitt & Maglott, 2001) while PROSITE is for functional sites in different proteins (Bairoch, 1992).

6.4 APPLICATION OF GENOMICS TO BREED CLIMATE-RESILIENT VARIETIES

The application of genomics in agriculture provides a powerful technique for breeders and researchers to enhance the adaptability of crops to changing environment (Kole et al., 2015). There are different techniques in genomics, including quantitative trait locus (QTL) analysis, GWAS, GS, and gene-editing technologies such as ZFNs, transcription activator-like effector nucleases (TALENs), and the CRISPR/Cas system (Gogolev et al., 2021; Kumar et al., 2024).

6.4.1 Quantitative Trait Locus (QTL) Analysis

QTL are specific locations in the genomic DNA that affect the phenotype of a complex trait, often through the genetic interactions within the locus itself, between different loci and with the environment (Powder, 2020). Many traits, like yield and biotic and abiotic stress tolerance, are typically influenced by multiple genes and environmental factors. QTL mapping involves association between a genetic marker and phenotype that can be measure (Dhingani et al., 2015). QTL mapping involves the following steps: development of suitable mapping population from two parental strains with phenotypically contrasting characters; selection of suitable molecular marker and development of linkage map; genotyping of mapping population; and QTL detection and mapping using softwares (Kumar et al., 2017).

6.4.2 Genome-Wide Association Studies (GWAS)

GWAS represents a complementary approach to QTL analysis. With improvement in high-throughput genotyping and phenotyping technologies, GWAS has become ever more powerful (Dhondt et al., 2013; Ellegren, 2014). The first step in GWAS is the selection of suitable mapping population. Unlike traditional QTL mapping, which often focuses on specific breeding populations, GWAS leverages natural variation in very large, genetically diverse populations to identify reproducible genome-wide significant associations. Once the population is selected, phenotypic data is collected from a large number of individuals. The required sample size can be ascertained using complex calculations using software tools such as CaTS14 or GPC15. The population is then genotyped using high-density SNP arrays or whole-genome sequencing. Then, statistical methods are applied to test for associations between SNPs and phenotypic traits, identifying loci for trait variation (Gill et al., 2022).

6.4.3 INTEGRATING QTL AND GWAS

The integration of QTL analysis and GWAS can enhance the understanding of the genetic basis of climate resilience. By integrating the strengths of both approaches, breeders and researchers can validate QTLs identified in specific populations of a crop and explore their relevance across diverse genetic backgrounds (Khan et al., 2021).

6.4.4 GENOMIC SELECTION (GS)

GS is a revolutionary methodology that uses genomic information to predict the genetic potential of individuals for specific traits. It exploits molecular markers to develop models for genetic evaluation of genotypes (Bhat et al., 2016). Unlike traditional selection methods that rely on phenotypic data only, GS incorporates high-density SNP markers to estimate breeding values. GS includes a training population in which both phenotypic and genotypic data are collected. This population is used to build predictive models such as genomic best linear unbiased prediction (GBLUP) to estimate the breeding values of individuals on the basis of genotypic data of the different genotypes (Crossa et al., 2017). With the help of these, breeders can select plants with the highest predicted breeding values for further breeding, accelerating the selection process in breeding programs (Figure 6.1).

6.4.4.1 Application of GS in Climate Resilience

In the context of climate resilience, GS offers several advantages: *Early Selection of genotypes:* GS allows for early selection of genotype based on their genomic

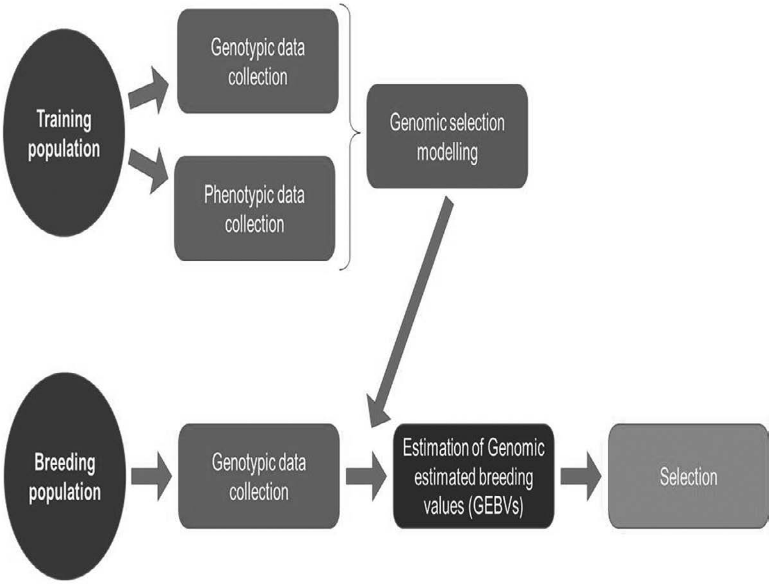


FIGURE 6.1 Flowchart for steps of genomic selection.

information, doing away with long breeding cycles for variety release. *Complex Trait Improvement:* GS is particularly effective for improving complex traits influenced by multiple genes and environmental interactions, for both biotic and abiotic stress. *Enhanced Genetic Gain:* By use of genomic information, GS can increase the rate of genetic gain compared to traditional breeding methods (Crossa et al., 2017). Several studies have demonstrated the successful application of GS in developing climate-resilient crops. For example, GS may also be utilized to predict the performance of a large number of hybrid combinations (VanRaden, 2008).

6.4.5 GENOME EDITING TECHNIQUES

6.4.5.1 Zinc Finger Nucleases (ZFNs)

ZFNs are engineered DNA-binding proteins that facilitate targeted genome editing. They consist of a DNA-binding domain that recognizes specific DNA sequences and a nuclease domain that introduces double-strand breaks at the target site. Each zinc finger domain recognizes approximately 3 base pairs (bp) of DNA, and an array of 4–6 domains can be assembled to target specific sequences (Urnov et al., 2010). ZFNs can be effectively used to enhance traits in major cereal crops, thereby improving productivity under changing climate conditions (Howden et al., 2007).

6.4.5.2 Transcription Activator-Like Effector Nucleases (TALENs)

TALENs are a novel gene-editing technique which have quickly become a prominent alternative to ZFNs. TALENs consist of a customizable DNA-binding domain derived from transcription activator-like effectors (TALEs) produced by *Xanthomonas* spp. (Szurek et al., 2002) fused to a non-specific FokI nuclease domain (Figure 6.2). The DNA-binding domain of TALENs is formed by arrays of highly conserved 33–35 amino acid repeats, with hypervariable residues at positions 12 and 13 that determine specificity for DNA bases (Malzahn et al., 2017). These residues interact with the DNA major groove, allowing for specific binding. Most engineered TALE repeat arrays utilize four specific repeat types, i.e., NN, NI, HD, and

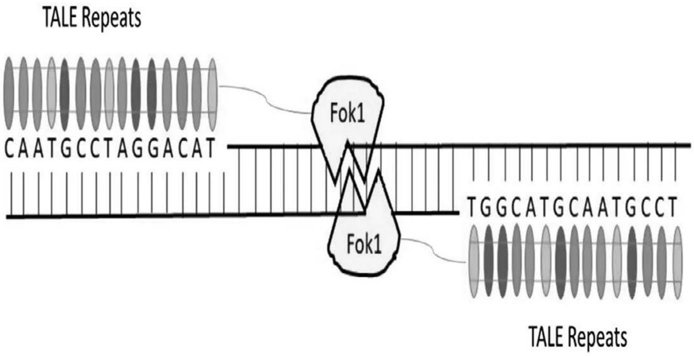


FIGURE 6.2 TALEN contains two monomers, each containing a TALE DNA-binding domain and a FokI nuclease domain.

NG to recognize guanine, adenine, cytosine, and thymine, respectively. While a repeat with residues NK shows increased specificity for guanine, it exhibits lower activity compared to the NN repeat (Miller et al., 2011). The efficiency of TALENs can vary based on factors like species, cell type, target gene, and the specific nuclease used (Cermak et al., 2011).

6.4.5.3 The CRISPR/*Cas* System in Breeding Climate-Resilient Genotypes

The CRISPR/Cas system enables precise genetic modifications, accelerating the development of climate-resilient crop genotypes by enhancing traits such as drought tolerance, disease resistance, and improved nutrient efficiency.

6.4.5.3.1 Overview of CRISPR/Cas Technology

CRISPR-*Cas9* is a groundbreaking genome editing tool that allows researchers to edit DNA with remarkable speed, accuracy, and cost-effectiveness. The technology is derived from an adaptive machinery in phage immunity system of *Archaea* and bacteria (Zhu et al., 2020) which uses CRISPR sequences and *Cas* proteins to target and cleave foreign DNA. CRISPR/*Cas* system can be used to efficiently generate targeted gene mutations and corrections in plants (Feng et al., 2013). The key components of CRISPR/Cas technology include the following: CRISPR (Clustered Regularly Interspaced Short Palindromic Repeats); Guide RNA (gRNA); Cas9 Nuclease; PAM (protospacer adjacent motif); repair template (optional). The procedure of gene editing with the help of CRISPR/*Cas* involves the following steps.

6.4.5.3.1.1 Target Identification Determining the specific DNA sequence in the genome that must be altered. This sequence must be adjacent to the PAM, which is required for *Cas9* interaction (Jinek et al., 2012). Designing the gRNA: A gRNA is created, consisting of a CRISPR RNA (*cr*RNA) that complements and binds the target DNA sequence and a trans-activating RNA (*tracr*RNA) that stabilizes the complex. These two components are combined into a single gRNA (*sg*RNA) for editing (Hsu et al., 2014).

6.4.5.3.1.2 Delivery into Cells The *Cas9* protein and *g*RNA are delivered into target cells using different techniques, including plasmid transfection, viral vectors, and electroporation. Once it gets into the cell, the *g*RNA attaches to the target DNA sequence based on its complimentary nature (Cong et al., 2013). *DNA Cleavage:* The *Cas9* enzyme creates a double-strand break in the DNA at the specified location in upstream of the stand. This break triggers the cell's natural DNA repair mechanism (Doudna & Charpentier, 2014).

6.4.5.3.1.3 DNA Repair The cell can repair the double-strand break through two primary pathways: *Non-Homologous End Joining (NHEJ)*: Addition and deletions within the target gene (indels) may lead to its inactivation. *Homology-Directed Repair (HDR)*: If a repair template is provided, the cell can use it to make precise edits, such as correcting mutations or inserting new sequences. It is a difficult repair method than NHEJ (Doudna & Charpentier, 2014).

6.4.5.3.2 Applications of CRISPR/Cas in Climate Resilience

The CRISPR/Cas system enhances climate resilience in crops through gene knockout, such as disrupting ethylene signaling genes to reduce drought-induced senescence, and trait enhancement, like modifying root architecture genes to improve water use efficiency. Its ability to perform multi-gene editing accelerates the development of resilient varieties, exemplified by drought-tolerant rice engineered via simultaneous edits to stress-response genes. This precision and speed allow breeders to rapidly address climate challenges by tailoring crops to withstand environmental stresses (Doudna & Charpentier, 2014).

6.5 FUTURE THRUSTS

To address climate change challenges in agriculture, future crop genomics must integrate with emerging technologies like proteomics and metabolomics to comprehensively understand plant stress responses and identify key breeding targets (Srivastav et al., 2024). Advancements in gene-editing tools (CRISPR, TALENs, ZFNs) and their application to polyploid crops will be crucial for developing climate-resilient varieties (May et al., 2023; Rafiq, 2024). Developing pangenomes for diverse crops, including underutilized species, will reveal valuable genetic resources for climate adaptation (Chapman et al., 2022). Artificial intelligence and machine learning will transform predictive breeding by analyzing genomic, phenotypic, and environmental data to optimize trait selection (Zhang et al., 2024), supported by advanced bioinformatics and cloud platforms for managing large datasets (Srivastav et al., 2024). Global collaboration through standardized data sharing and strong researcher-policymaker-farmer partnerships will be essential to implement genomic innovations effectively (Spielman, 2005).

6.6 CONCLUSION

The application of genomics in breeding climate-resilient varieties represents a significant advancement in agricultural science. By utilizing methodologies such as QTL analysis, GWAS, GS, and advanced gene-editing technologies like ZFNs, TALENs, and CRISPR/Cas, researchers and breeders can develop crops which are better equipped to tolerate the stresses associated with climate change. These innovations not only enhance food security but also promote sustainable agricultural practices that are essential for the future of global agriculture.

REFERENCES

Bairoch, A. (1992). PROSITE: A dictionary of sites and patterns in proteins. *Nucleic Acids Research*, *20*(Suppl), 2013. https://doi.org/10.1093/nar/20.suppl.2013

Bayer, P. E., Golicz, A. A., Scheben, A., Batley, J., & Edwards, D. (2020). Plant pangenomes are the new reference. *Nature Plants*, *6*(8), 914–920. https://doi.org/10.1038/s41477-020-0733-0

Benavente, E., & Giménez, E. (2021). Modern approaches for the genetic improvement of rice, wheat and maize for abiotic constraints-related traits: A comparative overview. *Agronomy*, *11*(2), 376. https://doi.org/10.3390/agronomy11020376

Bera, M., Das, S., Dutta, S., & Choudhury, M. R. (2024). Navigating the future: Climate change impacts, mitigation strategies, and adaptation pathways in agriculture. In *Ecologically mediated development: Promoting biodiversity conservation and food security* (pp. 419–440). Springer. https://doi.org/10.1007/978-981-96-2413-3_19

Berlin, K., Koren, S., Chin, C. S., Drake, J. P., Landolin, J. M., & Phillippy, A. M. (2015). Assembling large genomes with single-molecule sequencing and locality-sensitive hashing. *Nature Biotechnology*, *33*(6), 623–630. https://doi.org/10.1038/nbt.3238

Bhat, J. A., Ali, S., Salgotra, R. K., Mir, Z. A., Dutta, S., Jadon, V., Tyagi, A., Muntazir, M., Jain, N., Singh, P. K., & Prabhu, K. (2016). Genomic selection in the era of next-generation sequencing for complex traits in plant breeding. *Frontiers in Genetics*, *7*, 221. https://doi.org/10.3389/fgene.2016.00221

Cao, H., Hastie, A. R., Cao, D., Lam, E. T., Sun, Y., Huang, H., Liu, X., Lin, L., Andrews, W., Chan, S., & Huang, S. (2014). Rapid detection of structural variation in a human genome using nanochannel-based genome mapping technology. *GigaScience*, *3*(1), 2047–17X. https://doi.org/10.1186/2047-217X-3-34

Carneiro, M. O., Russ, C., Ross, M. G., Gabriel, S. B., Nusbaum, C., & DePristo, M. A. (2012). Pacific biosciences sequencing technology for genotyping and variation discovery in human data. *BMC Genomics*, *13*(1), 1–7. https://doi.org/10.1186/1471-2164-13-375

Cermak, T., Doyle, E. L., Christian, M., Wang, L., Zhang, Y., Schmidt, C., Baller, J. A., Somia, N. V., Bogdanove, A. J., & Voytas, D. F. (2011). Efficient design and assembly of custom TALEN and other TAL effector-based constructs for DNA targeting. *Nucleic Acids Research*, *39*(12), e82. https://doi.org/10.1093/nar/gkr218

Chapman, M. A., He, Y., & Zhou, M. (2022). Beyond a reference genome: Pangenomes and population genomics of underutilized and orphan crops for future food and nutrition security. *New Phytologist*, *234*(5), 1583–1597. https://doi.org/10.1111/nph.18074

Chen, H., Xie, W., He, H., Yu, H., Chen, W., Li, J., Zhang, Q., & Zhang, Q. (2014). A high-density SNP genotyping array for rice biology and molecular breeding. *Molecular Plant*, *7*(3), 541–553. https://doi.org/10.1093/mp/sst135

Clarke, W. E., Higgins, E. E., Plieske, J., Wieseke, R., Sidebottom, C., Khedikar, Y., Batley, J., Edwards, D., & Meng, J. (2016). A high-density SNP genotyping array for *Brassica napus* and its ancestral diploid species based on optimised selection of single-locus markers in the allotetraploid genome. *Theoretical and Applied Genetics*, *129*(10), 1887–1899. https://doi.org/10.1007/s00122-016-2746-7

Cong, L., Ran, F. A., Cox, D., Lin, S., Barretto, R., Habib, N., Hsu, P. D., Wu, X., Jiang, W., Marraffini, L. A., & Zhang, F. (2013). Multiplex genome engineering using CRISPR/Cas systems. *Science*, *339*(6121), 819–823. https://doi.org/10.1126/science.1231143

Crossa, J., Pérez-Rodríguez, P., Cuevas, J., Montesinos-López, O., Jarquín, D., de Los Campos, G., Burgueño, J., González-Camacho, J. M., Pérez-Elizalde, S., Beyene, Y., Dreisigacker, S., Singh, R., Zhang, X., Gowda, M., Roorkiwal, M., Rutkoski, J., & Varshney, R. K. (2017). Genomic selection in plant breeding: Methods, models, and perspectives. *Trends in Plant Science*, *22*(11), 961–975. https://doi.org/10.1016/j.tplants.2017.08.011

Dalton-Morgan, J., Hayward, A., Alamery, S., Tollenaere, R., Mason, A. S., Campbell, E., & Edwards, D. (2014). A high-throughput SNP array in the amphidiploid species *Brassica napus* shows diversity in resistance genes. *Functional and Integrative Genomics*, *14*(4), 643–655. https://doi.org/10.1007/s10142-014-0389-9

Denton, J. F., Lugo-Martinez, J., Tucker, A. E., Schrider, D. R., Warren, W. C., & Hahn, M. W. (2014). Extensive error in the number of genes inferred from draft genome assemblies. *PLoS Computational Biology*, *10*(12), e1003998. https://doi.org/10.1371/journal.pcbi.1003998

Dhingani, R. M., Umrania, V. V., Tomar, R. S., Parakhia, M. V., & Golakiya, B. A. (2015). Introduction to QTL mapping in plants. *Annals of Plant Sciences*, *4*(4), 1072–1079.

Dhondt, S., Wuyts, N., & Inzé, D. (2013). Cell to whole-plant phenotyping: The best is yet to come. *Trends in Plant Science*, *18*(8), 428–439. https://doi.org/10.1016/j.tplants.2013.04.008

Doudna, J. A., & Charpentier, E. (2014). The new frontier of genome engineering with CRISPR-Cas9. *Science*, *346*(6213), 1258096. https://doi.org/10.1126/science.1258096

Edwards, D., Batley, J., & Snowdon, R. J. (2013). Accessing complex crop genomes with next-generation sequencing. *Theoretical and Applied Genetics*, *126*(1), 1–11. https://doi.org/10.1007/s00122-012-1964-x

Eisenstein, M. (2015). Startups use short-read data to expand long-read sequencing market. *Nature Biotechnology*, *33*(5), 433–436. https://doi.org/10.1038/nbt.3215

Ellegren, H. (2014). Genome sequencing and population genomics in non-model organisms. *Trends in Ecology and Evolution*, *29*(1), 51–63. https://doi.org/10.1016/j.tree.2013.09.008

Elshire, R. J., Glaubitz, J. C., Sun, Q., Poland, J. A., Kawamoto, K., Buckler, E. S., & Mitchell, S. E. (2011). A robust, simple genotyping-by-sequencing (GBS) approach for high diversity species. *PLoS ONE*, *6*(5), e19379. https://doi.org/10.1371/journal.pone.0019379

Felcher, K. J., Coombs, J. J., Massa, A. N., Hansey, C. N., Hamilton, J. P., Veilleux, R. E., Buell, C. R., & Douches, D. S. (2012). Integration of two diploid potato linkage maps with the potato genome sequence. *PLoS ONE*, *7*(4), e36347. https://doi.org/10.1371/journal.pone.0036347

Feng, Z., Zhang, B., Ding, W., Liu, X., Yang, D. L., Wei, P., Cao, F., Zhu, S., Zhang, F., Mao, Y., & Zhu, J. K. (2013). Efficient genome editing in plants using a CRISPR/Cas system. *Cell Research*, *23*(10), 1229–1232. https://doi.org/10.1038/cr.2013.114

Gao, L., Gonda, I., Sun, H., Ma, Q., Bao, K., Tieman, D. M., Burzynski-Chang, E. A., Fish, T. L., Stromberg, K. A., Sacks, G. L., Thannhauser, T. W., Foolad, M. R., Diez, M. J., Blanca, J., Canizares, J., Xu, Y., van der Knaap, E., Huang, S., Klee, H. J., & Giovannoni, J. J. (2019). The tomato pan-genome uncovers new genes and a rare allele regulating fruit flavor. *Nature Genetics*, *51*(6), 1044–1051. https://doi.org/10.1038/s41588-019-0410-2

Gill, T., Gill, S. K., Saini, D. K., Chopra, Y., de Koff, J. P., & Sandhu, K. S. (2022). A comprehensive review of high throughput phenotyping and machine learning for plant stress phenotyping. *Phenomics*, *2*(3), 156–183. https://doi.org/10.1007/s43657-022-00048-z

Gogolev, Y. V., Ahmar, S., Akpinar, B. A., Budak, H., Kiryushkin, A. S., Gorshkov, V. Y., Hensel, G., Demchenko, N. P., Kochetov, A. V., & Korzun, V. (2021). OMICs, epigenetics, and genome editing techniques for food and nutritional security. *Plants*, *10*(7), 1423. https://doi.org/10.3390/plants10071423

Goodwin, S., Gurtowski, J., Ethe-Sayers, S., Deshpande, P., Schatz, M. C., & McCombie, W. R. (2015). Oxford Nanopore sequencing, hybrid error correction, and de novo assembly of a eukaryotic genome. *Genome Research*, *25*(11), 1750–1756. https://doi.org/10.1101/gr.191395.115

Goodwin, S., McPherson, J. D., & McCombie, W. R. (2016). Coming of age: Ten years of next-generation sequencing technologies. *Nature Reviews Genetics*, *17*(6), 333–351. https://doi.org/10.1038/nrg.2016.49

Hastie, A. R., Dong, L., Smith, A., Finklestein, J., Lam, E. T., Huo, N., Cao, H., Kwok, P. Y., Deal, K. R., Dvorak, J., & Luo, M. C. (2013). Rapid genome mapping in nanochannel arrays for highly complete and accurate de novo sequence assembly of the complex *Aegilops tauschii* genome. *PLoS ONE*, *8*(2), e55864. https://doi.org/10.1371/journal.pone.0055864

Higgins, D. G., Fuchs, R., Stoehr, P. J., & Cameron, G. N. (1992). The EMBL data library. *Nucleic Acids Research*, *20*(Suppl), 2071–2074. https://doi.org/10.1093/nar/20.suppl.2071

Howden, S. M., Soussana, J. F., Tubiello, F. N., Chhetri, N., Dunlop, M., & Meinke, H. (2007). Adapting agriculture to climate change. *Proceedings of the National Academy of Sciences*, *104*(50), 19691–19696. https://doi.org/10.1073/pnas.0701890104

Hsu, P. D., Lander, E. S., & Zhang, F. (2014). Development and applications of CRISPR-Cas9 for genome engineering. *Cell*, *157*(6), 1262–1278. https://doi.org/10.1016/j.cell.2014.05.010

Hufford, M. B., Seetharam, A. S., Woodhouse, M. R., Chougule, K. M., Ou, S., Liu, J., Ricci, W. A., Guo, T., Olson, A., Qiu, Y., Della Coletta, R., Tittes, S., Hudson, A. I., Marand, A. P., Wei, S., Lu, Z., Wang, B., Tello-Ruiz, M. K., Piri, R. D., & Ross-Ibarra, J. (2021). De novo assembly, annotation, and comparative analysis of 26 diverse maize genomes. *bioRxiv*. https://doi.org/10.1101/2021.01.14.426684

Imelfort, M., & Edwards, D. (2009). De novo sequencing of plant genomes using second-generation technologies. *Briefings in Bioinformatics*, *10*(6), 609–618. https://doi.org/10.1093/bib/bbp039

Ip, C. L., Loose, M., Tyson, J. R., de Cesare, M., Brown, B. L., Jain, M., Leggett, R. M., Eccles, D. A., Zalunin, V., Urban, J. M., & Piazza, P. (2015). MinION analysis and reference consortium: Phase 1 data release and analysis. *F1000Research*, *4*, 1075. https://doi.org/10.12688/f1000research.7201.1

Jain, M., Fiddes, I. T., Miga, K. H., Olsen, H. E., Paten, B., & Akeson, M. (2015). Improved data analysis for the MinION nanopore sequencer. *Nature Methods*, *12*(4), 351–356. https://doi.org/10.1038/nmeth.3290

Jinek, M., Chylinski, K., Fonfara, I., Hauer, M., Doudna, J. A., & Charpentier, E. (2012). A programmable dual-RNA–guided DNA endonuclease in adaptive bacterial immunity. *Science*, *337*(6096), 816–821. https://doi.org/10.1126/science.1225829

Jovović, Z., Velimirović, A., & Yaman, N. (2025). Climate and crop production crisis. In *Agriculture and water management under climate change* (pp. 1–28). Springer Nature. https://doi.org/10.1007/978-3-031-74307-8_1

Khan, S. U., Saeed, S., Khan, M. H. U., Fan, C., Ahmar, S., Arriagada, O., & Mora-Poblete, F. (2021). Advances and challenges for QTL analysis and GWAS in the plant-breeding of high-yielding: A focus on rapeseed. *Biomolecules*, *11*(10), 1516. https://doi.org/10.3390/biom11101516

Kim, C., Guo, H., Kong, W., Chandnani, R., Shuang, L. S., & Paterson, A. H. (2016). Application of genotyping by sequencing technology to a variety of crop breeding programs. *Plant Science*, *242*, 14–22. https://doi.org/10.1016/j.plantsci.2015.04.016

Kole, C., Muthamilarasan, M., Henry, R., Edwards, D., Sharma, R., Abberton, M., Batley, J., Bentley, A., Blakeney, M., Bryant, J., Cai, H., Cakir, M., Cseke, L. J., Cockram, J., de Oliveira, A. C., De Pace, C., Dempewolf, H., Ellison, S., Gepts, P., & Prasad, M. (2015). Application of genomics-assisted breeding for generation of climate resilient crops: Progress and prospects. *Frontiers in Plant Science*, *6*, 563. https://doi.org/10.3389/fpls.2015.00563

Koren, S., & Phillippy, A. M. (2015). One chromosome, one contig: Complete microbial genomes from long-read sequencing and assembly. *Current Opinion in Microbiology*, *23*, 110–120. https://doi.org/10.1016/j.mib.2014.11.014

Kumar, A., Ranjan, T., Kumar, R. R., Rajani, K., Kishore, C., & Kumar, J. (2017). Development of mapping populations. In *Plant biotechnology, Volume 1* (pp. 455–478). Apple Academic Press.

Kumar, R., Das, S. P., Choudhury, B. U., Kumar, A., Prakash, N. R., Verma, R., & Mishra, V. K. (2024). Advances in genomic tools for plant breeding: Harnessing DNA molecular markers, genomic selection, and genome editing. *Biological Research*, *57*(1), 80. https://doi.org/10.1186/s40659-024-00505-1

Lee, H., Gurtowski, J., Yoo, S., Marcus, S., McCombie, W. R., & Schatz, M. (2014). Error correction and assembly complexity of single molecule sequencing reads. *bioRxiv*. https://doi.org/10.1101/006395

Leow, R., & Taylor, K. (2000). Efficient web access to distributed biological collections using a taxonomy browser. In *Proceedings of the 12th international conference on scientific and statistical database management* (pp. 25–38). IEEE.

Li, J., Yuan, D., Wang, P., Wang, Q., Sun, M., Liu, Z., Si, H., Xu, Z., Ma, Y., Zhang, B., Han, X., Chang, L., Zhang, J., Yang, C., Zhong, F., Chen, X., Luo, J., Guo, H., Wang, J., & Zhang, X. (2021). Cotton pan-genome retrieves the lost sequences and genes during domestication and selection. *Genome Biology*, *22*(1), 119. https://doi.org/10.1186/s13059-021-02351-w

Li, R., Hsieh, C. L., Young, A., Zhang, Z., Ren, X., & Zhao, Z. (2015). Illumina synthetic long read sequencing allows recovery of missing sequences even in the “finished” *C. elegans* genome. *Scientific Reports*, *5*(1), 10814. https://doi.org/10.1038/srep10814

Liu, Y., Du, H., Li, P., Shen, Y., Peng, H., Liu, S., Zhou, G. A., Zhang, H., Liu, Z., Shi, M., Huang, X., Li, Y., Zhang, M., Wang, Z., Zhu, B., Han, B., Liang, C., & Tian, Z. (2020). Pan-genome of wild and cultivated soybeans. *Cell*, *182*(1), 162–176.e13. https://doi.org/10.1016/j.cell.2020.05.023

Loman, N. J., Quick, J., & Simpson, J. T. (2015). A complete bacterial genome assembled de novo using only nanopore sequencing data. *Nature Methods*, *12*(8), 733–735. https://doi.org/10.1038/nmeth.3444

Malzahn, A., Lowder, L., & Qi, Y. (2017). Plant genome editing with TALEN and CRISPR. *Cell and Bioscience*, *7*(1), 21. https://doi.org/10.1186/s13578-017-0148-4

May, D., Paldi, K., & Altpeter, F. (2023). Targeted mutagenesis with sequence-specific nucleases for accelerated improvement of polyploid crops: Progress, challenges, and prospects. *The Plant Genome*, *16*(2), e20298. https://doi.org/10.1002/tpg2.20298

McCouch, S. R., Wright, M. H., Tung, C. W., Maron, L. G., McNally, K. L., Fitzgerald, M., Singh, N., DeClerck, G., Agosto-Perez, F., Korniliev, P., Greenberg, A. J., Naredo, M. E. B., Mercado, S. M. Q., Harrington, S. E., Shi, Y., Branchini, D. A., Kuser-Falcão, P. R., Leung, H., Ebana, K., & Bustamante, C. D. (2016). Open access resources for genome-wide association mapping in rice. *Nature Communications*, *7*(1), 10532. https://doi.org/10.1038/ncomms10532

McCoy, R. C., Taylor, R. W., Blauwkamp, T. A., Kelley, J. L., Kertesz, M., Pushkarev, D., Petrov, D. A., & Fiston-Lavier, A. S. (2014). Illumina TruSeq synthetic long-reads empower de novo assembly and resolve complex, highly-repetitive transposable elements. *PLoS ONE*, *9*(9), e106689. https://doi.org/10.1371/journal.pone.0106689

McGinnis, S., & Madden, T. L. (2004). BLAST: At the core of a powerful and diverse set of sequence analysis tools. *Nucleic Acids Research*, *32*(suppl_2), W20–W25. https://doi.org/10.1093/nar/gkh435

Metzker, M. L. (2005). Emerging technologies in DNA sequencing. *Genome Research*, *15*(12), 1767–1776. https://doi.org/10.1101/gr.3770505

Miller, J. C., Tan, S., Qiao, G., Barlow, K. A., Wang, J., Xia, D. F., Meng, X., Paschon, D. E., Leung, E., Hinkley, S. J., Dulay, G. P., Hua, K. L., Ankoudinova, I., Cost, G. J., Urnov, F. D., Zhang, H. S., Holmes, M. C., Zhang, L., Gregory, P. D., & Rebar, E. J. (2011). A TALE nuclease architecture for efficient genome editing. *Nature Biotechnology*, *29*(2), 143–148. https://doi.org/10.1038/nbt.1755

Minouchehr, Z., & Gollaei, B. (2004). Protein databases. *Journal of Sciences, Islamic Republic of Iran*, *15*(3), 241–250.

Naqvi, R. Z., Siddiqui, H. A., Mahmood, M. A., Najeebullah, S., Ehsan, A., Azhar, M., & Asif, M. (2022). Smart breeding approaches in post-genomics era for developing climate-resilient food crops. *Frontiers in Plant Science*, *13*, 972164. https://doi.org/10.3389/fpls.2022.972164

Poland, J. A., Brown, P. J., Sorrells, M. E., & Jannink, J. L. (2012). Development of high-density genetic maps for barley and wheat using a novel two-enzyme genotyping-by-sequencing approach. *PLoS ONE*, *7*(2), e32253. https://doi.org/10.1371/journal.pone.0032253

Powder, K. E. (2020). Quantitative trait loci (QTL) mapping. In *eQTL analysis: Methods and protocols* (pp. 211–229). Humana.

Pruitt, K. D., & Maglott, D. R. (2001). RefSeq and LocusLink: NCBI gene-centered resources. *Nucleic Acids Research*, *29*(1), 137–140. https://doi.org/10.1093/nar/29.1.137

Rafiq, A. R. (2024). Genomic editing techniques for ensuring food security: CRISPR Cas, TALEN, ZFN, RNAi and mutagenesis. *Journal of Pure and Applied Agriculture*, *9*(2), 1–15.

Rhoads, A., & Au, K. F. (2015). PacBio sequencing and its applications. *Genomics, Proteomics and Bioinformatics*, *13*(5), 278–289. https://doi.org/10.1016/j.gpb.2015.08.002

Saleem, M. H., Noreen, S., Ishaq, I., Saleem, A., Khan, K. A., Ercisli, S., & Fahad, S. (2025). Omics technologies: Unraveling abiotic stress tolerance mechanisms for sustainable crop improvement. *Journal of Plant Growth Regulation*, *44*(3), 1–23. https://doi.org/10.1007/s00344-025-11674-y

Schadt, E. E., Turner, S., & Kasarskis, A. (2010). A window into third-generation sequencing. *Human Molecular Genetics*, *19*(R2), R227–R240. https://doi.org/10.1093/hmg/ddq416

Schröder, P., Sauvêtre, A., Gnädinger, F., Pesaresi, P., Chmeliková, L., Doğan, N., & Terzi, V. (2019). Discussion paper: Sustainable increase of crop production through improved technical strategies, breeding and adapted management – A European perspective. *Science of the Total Environment*, *678*, 146–161. https://doi.org/10.1016/j.scitotenv.2019.04.212

Schuler, G. D., Epstein, J. A., Ohkawa, H., & Kans, J. A. (1996). Entrez: Molecular biology database and retrieval system. *Methods in Enzymology*, *266*, 141–162. https://doi.org/10.1016/S0076-6879(96)66012-1

Schwartz, D. C., Li, X., Hernandez, L. I., Ramnarain, S. P., Huff, E. J., & Wang, Y. K. (1993). Ordered restriction maps of *Saccharomyces cerevisiae* chromosomes constructed by optical mapping. *Science*, *262*(5130), 110–114. https://doi.org/10.1126/science.8211116

Shearer, L. A., Anderson, L. K., de Jong, H., Smit, S., Goicoechea, J. L., Roe, B. A., Hua, A., Giovannoni, J. J., & Stack, S. M. (2014). Fluorescence in situ hybridization and optical mapping to correct scaffold arrangement in the tomato genome. *G3: Genes, Genomes, Genetics*, *4*(8), 1395–1405. https://doi.org/10.1534/g3.114.011197

Shelton, J. M., Coleman, M. C., Herndon, N., Lu, N., Lam, E. T., Anantharaman, T., Sheth, P., & Brown, S. J. (2015). Tools and pipelines for BioNano data: Molecule assembly pipeline and FASTA super scaffolding tool. *BMC Genomics*, *16*(1), 1–6. https://doi.org/10.1186/s12864-015-1911-8

Singh, B. D., & Singh, A. K. (2015). Bioinformatics tools and databases for genomics research. In *Marker-assisted plant breeding: Principles and practices* (pp. 401–429). Springer.

Singh, G. B. (2015). Biological databases. In *Fundamentals of bioinformatics and computational biology: Methods and exercises in MATLAB* (pp. 37–76). Springer.

Snowdon, R. J., & Iniguez Luy, F. L. (2012). Potential to improve oilseed rape and canola breeding in the genomics era. *Plant Breeding*, *131*(3), 351–360. https://doi.org/10.1111/j.1439-0523.2012.01976.x

Spielman, D. J. (2005). Innovation systems perspectives on developing-country agriculture: A critical review. ISNAR Discussion Paper 2. International Food Policy Research Institute (IFPRI). https://hdl.handle.net/10568/160694

Srivastav, A. K., Das, P., & Srivastava, A. K. (2024). Bioinformatics and cloud analytics. In *Biotech and IoT: An introduction using cloud-driven labs* (pp. 285–308). Apress.

Staňková, H., Hastie, A. R., Chan, S., Vrána, J., Tulpová, Z., Kubaláková, M., Visendi, P., Hayashi, S., Luo, M., Batley, J., & Edwards, D. (2016). BioNano genome mapping of individual chromosomes supports physical mapping and sequence assembly in complex plant genomes. *Plant Biotechnology Journal*, *14*(7), 1523–1531. https://doi.org/10.1111/pbi.12513

Szalay, T., & Golovchenko, J. A. (2015). De novo sequencing and variant calling with nanopores using PoreSeq. *Nature Biotechnology*, *33*(10), 1087–1091. https://doi.org/10.1038/nbt.3327

Szurek, B., Rossier, O., Hause, G., & Bonas, U. (2002). Type III-dependent translocation of the *Xanthomonas* AvrBs3 protein into the plant cell. *Molecular Microbiology*, *46*(1), 13–23. https://doi.org/10.1046/j.1365-2958.2002.03139.x

Tallon, L. J., Liu, X., Bennuru, S., Chibucos, M. C., Godinez, A., Ott, S., Zhao, X., Sadzewicz, L., Fraser, C. M., Nutman, T. B., & Dunning Hotopp, J. C. (2014). Single molecule sequencing and genome assembly of a clinical specimen of *Loa loa*, the causative agent of loiasis. *BMC Genomics*, *15*(1), 1–4. https://doi.org/10.1186/1471-2164-15-788

Treangen, T. J., & Salzberg, S. L. (2012). Repetitive DNA and next-generation sequencing: Computational challenges and solutions. *Nature Reviews Genetics*, *13*(1), 36–46. https://doi.org/10.1038/nrg3117

Urban, J. M., Bliss, J., Lawrence, C. E., & Gerbi, S. A. (2015). Sequencing ultra-long DNA molecules with the Oxford Nanopore MinION. *bioRxiv*. https://doi.org/10.1101/019281

Urnov, F. D., Rebar, E. J., Holmes, M. C., Zhang, H. S., & Gregory, P. D. (2010). Genome editing with engineered zinc finger nucleases. *Nature Reviews Genetics*, *11*(9), 636–646. https://doi.org/10.1038/nrg2842

VanRaden, P. M. (2008). Efficient methods to compute genomic predictions. *Journal of Dairy Science*, *91*(11), 4414–4423. https://doi.org/10.3168/jds.2007-0980

Varshney, R. K., Nayak, S. N., May, G. D., & Jackson, S. A. (2009). Next-generation sequencing technologies and their implications for crop genetics and breeding. *Trends in Biotechnology*, *27*(9), 522–530. https://doi.org/10.1016/j.tibtech.2009.05.006

Varshney, R. K., Roorkiwal, M., Sun, S., Bajaj, P., Chitikineni, A., Thudi, M., Singh, N. P., Du, X., Upadhyaya, H. D., Khan, A. W., Wang, Y., Garg, V., Fan, G., Cowling, W. A., Crossa, J., Gentzbittel, L., Voss-Fels, K. P., Valluri, V. K., Sinha, P., & Liu, X. (2021). A chickpea genetic variation map based on the sequencing of 3,366 genomes. *Nature*, *599*(7886), 622–627. https://doi.org/10.1038/s41586-021-04066-1

Wallace, J. G., & Mitchell, S. E. (2017). Genotyping-by-sequencing. *Current Protocols in Plant Biology*, *2*(1), 64–77. https://doi.org/10.1002/cppb.20079

Wang, J., Chu, S., Zhang, H., Zhu, Y., Cheng, H., & Yu, D. (2016). Development and application of a novel genome-wide SNP array reveals domestication history in soybean. *Scientific Reports*, *6*, 20728. https://doi.org/10.1038/srep20728

Wang, W., Simmonds, J., Pan, Q., Davidson, D., He, F., Battal, A., & Akhunov, E. (2018). Gene editing and mutagenesis reveal inter-cultivar differences and additivity in the contribution of TaGW2 homoeologues to grain size and weight in wheat. *Theoretical and Applied Genetics*, *131*, 2463–2475. https://doi.org/10.1007/s00122-018-3166-7

Yu, H., Xie, W., Li, J., Zhou, F., & Zhang, Q. (2014). A whole-genome SNP array (RICE 6 K) for genomic breeding in rice. *Plant Biotechnology Journal*, *12*(1), 28–37. https://doi.org/10.1111/pbi.12109

Zhang, Y., Huang, G., Zhao, Y., Lu, X., Wang, Y., Wang, C., & Zhao, C. (2024). Revolutionizing crop breeding: Next-generation artificial intelligence and big data-driven intelligent design. *Engineering*. https://doi.org/10.1016/j.eng.2024.02.005

Zhao, Q., Feng, Q., Lu, H., Li, Y., Wang, A., Tian, Q., Zhan, Q., Lu, Y., Zhang, L., Huang, T., Wang, Y., Fan, D., Zhao, Y., Wang, Z., Zhou, C., Chen, J., Zhu, C., Li, W., Weng, Q., . . . Huang, X. (2018). Pan-genome analysis highlights the extent of genomic variation in cultivated and wild rice. *Nature Genetics*, *50*(2), 278–284. https://doi.org/10.1038/s41588-018-0041-z

Zhou, S., Bechner, M. C., Place, M., Churas, C. P., Pape, L., Leong, S. A., Runnheim, R., Forrest, D. K., Goldstein, S., Livny, M., & Schwartz, D. C. (2007). Validation of rice genome sequence by optical mapping. *BMC Genomics*, *8*, 1–8. https://doi.org/10.1186/1471-2164-8-7

Zhou, S., Wei, F., Nguyen, J., Bechner, M., Potamousis, K., Goldstein, S., Pape, L., Mehan, M. R., Churas, C., Pasternak, S., & Forrest, D. K. (2009). A single molecule scaffold for the maize genome. *PLoS Genetics*, *5*(11), e1000711. https://doi.org/10.1371/journal.pgen.1000711

Zhu, H., Li, C., & Gao, C. (2020). Applications of CRISPR–Cas in agriculture and plant biotechnology. *Nature Reviews Molecular Cell Biology*, *21*(11), 661–677. https://doi.org/10.1038/s41580-020-00288-9

7 Molecular Breeding for Climate Change Adaptation in Crops

Siddhartha Shankar Sharma, Jannila Praveena,
Sanghamitra Rout, Aninda Chakraborty,
Debarati Nandi, and Manish Kumar Agrawal

7.1 INTRODUCTION

As the global population grows and climate change increasingly impacts agriculture, developing climate-resilient crops has become essential. This chapter examines the revolutionizing potential of molecular breeding techniques in addressing the climate-induced stresses such as heat, drought, salinity, and pest pressures. It highlights the use of genomic tools, including marker-assisted selection (MAS), CRISPR-Cas systems, and genome-wide association studies (GWAS), which qualify precise modification of typical plant traits that enhance resilience and productivity. By exploring advanced methods like next-generation sequencing (NGS), speed breeding, and the integration of machine learning (ML), the chapter emphasizes the fusion of science and technology in agriculture (Hickey et al., 2019). Agriculture has significantly contributed to human progress and supported the rapid growth of the global population. Since the onset of the Green Revolution in the 1950s, food grain production has nearly tripled (John & Babu, 2021). With the global population expected to reach 10 billion by 2050, the demand for food is steadily growing (Hickey et al., 2019).

However, agriculture faces significant challenges in scaling up food production, particularly in achieving the 70% increase required to nourish the projected global population by 2050 (Searchinger et al., 2019). All regions are expected to encounter increased climate unpredictability, including more frequent storms, floods, droughts, and extreme temperatures, further amplifying uncertainty in crop production (Pickson & Boateng, 2022). The development of crop varieties is undergoing a data-driven transformation, driven by advancements in genomics and high-throughput phenotyping (Saad et al., 2022). Climate change is driving greater land degradation, particularly through soil salinization. To address its anticipated impact on food crop production, it is crucial to enhance crop resilience and productivity for improved adaptation to shifting climate conditions (Abbass et al., 2022). This chapter offers valuable insights into how molecular breeding is shaping the future of sustainable agriculture, ensuring that we can meet the needs of a growing population in a rapidly changing world.

DOI: 10.1201/9781003597797-7

7.2 CLIMATE CHANGE IMPACTS ON AGRICULTURE AND FOOD SECURITY

The global population continues to grow, presenting food security as a major challenge. Compounding this issue are other critical factors, such as climate change and the accelerating pace of population expansion (Abberton et al., 2016). This population growth requires greater food production and places additional pressure on agricultural resources (Ray et al., 2015). Moreover, change in climate poses a significant obstacle to the sustainable cultivation of agricultural products. For instance, direct consequences include physiological, morphological, and phenotypic changes in plant productivity. And indirect impacts include fertility of soil, sea level rise, insect compression, and irrigation availability, while socio-economic effects include food demand, costs, trade, and uneven distribution (Malhi et al., 2021). These factors can have a significant impact on agricultural production. Many biotic and abiotic stressors negatively affect crop productivity (Figure 7.1), exacerbated by climate

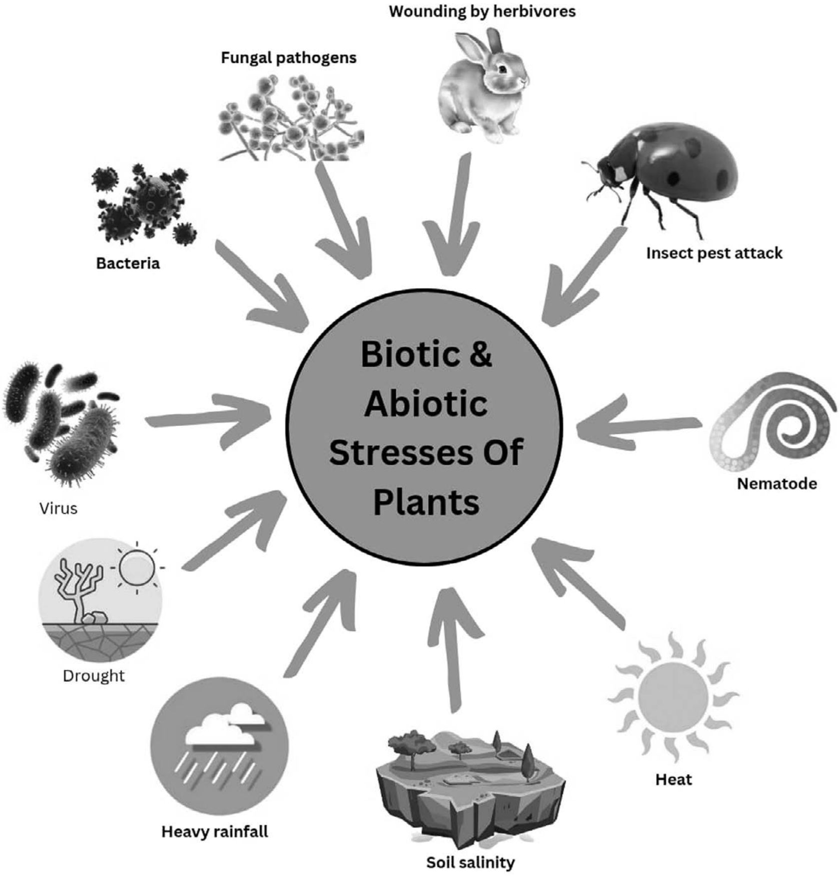


FIGURE 7.1 Many biotic and abiotic stressors negatively affecting crop productivity.

change, characterized by "substantial changes in various metric components, including temperature and precipitation, for which long-term averages have been calculated" (Malhi et al., 2021). Climate change has caused changes in temperature, precipitation, and atmospheric conditions that have a negative impact on the cellular, morphological, developmental, and molecular processes of plants. It can influence agricultural output through indirect, direct, and socio-economic channels (Naqvi et al., 2022).

7.3 PLANT BREEDING THE SAVIOR

Around 10,000 years ago, plant breeding techniques became a primary domestication strategy, involving the selection of desirable traits from wild relatives over many generations to improve crops (Purugganan & Fuller, 2009). This approach led to the widespread cultivation of many important world crops. Before the era of genetics, agronomic traits were often integrated into different crop types without detailed knowledge (Purugganan & Fuller, 2009). The principles of inheritance discovered by Mendel, combined with advancements in genetic research, have profoundly enhanced plant breeding by pinpointing the genes responsible for vital agronomic traits (McCouch et al., 2013). In the 1960s, the green revolution led to major improvements in the yield potential of crops like rice and wheat, helping to satisfy the rising global demand for food (Pingali, 2012).

While these innovations have brought great benefits to agriculture, they have also had negative consequences for the environment, such as the overuse of inorganic fertilizers and pesticides. In addition, widespread application of green revolution breeding practices has led to a loss of the genetic diversity and disappearance of many inimitable genes, making crops more susceptible to pests, diseases, and the environmental stresses like heat, drought, and flooding (Tilman et al., 2002). In response to these challenges, plant breeding has come under increasing pressure to maintain high yields despite limited resources such as water, soil, and waste. To overcome these challenges, plant scientists are focusing on identifying the genetic resources needed to develop crops that are more resilient to stress and capable of improving grain production. Conventional breeding has historically relied on hybridization and continuous evaluation to identify superior crop varieties (Purugganan & Fuller, 2009).

By crossing plants with beneficial traits with wild relatives or landraces, breeders have integrated genetic diversity to improve crops, selecting the best genotypes for desired traits (Lavarenne et al., 2018). Studying natural plant populations can help increase genetic diversity, creating new crop types with valuable traits that can improve agricultural outcomes. Hybridization between species offers a promising opportunity to address climate pressures (Becker et al., 2013). However, conventional breeding methods are limited by genetic drift, erosion of diversity, hybridization barriers, and the arduous nature of selection (Abberton et al., 2016). By developing a new crop cultivar with specific traits can endorse 10–20 years, producing the process slow and complex (Fischer et al., 2014).

In contrary, modern breeding methods, including genetic engineering, have made significant progress in recent decades to overcome the limitations of traditional techniques. The FDA approved the transgenic tomato “FlavrSavr” in 1994, which had an extended shelf life, marking a major milestone in biotechnology (James, 1997). Since then, genetically modified (GM) crops such as glyphosate-resistant soybeans, Bt maize, Bt cotton, and Bt potatoes have been accepted for saleable use in United States (ISAAA, 2021). Currently, 529 transgenic measures in 33 different crops have been commercialized, with the United States, India, Brazil, Canada, and Argentina accounting for 91% of the universal GM crop. The United States leads the production of GM crops, with varieties such as corn, soybeans, and cotton (ISAAA, 2021).

7.4 FAST-FORWARD GENOMICS-ASSISTED BREEDING (GAB)

Contemporary plant breeding has undergone significant transformations, offering powerful alternatives to traditional methods. Plant genomics plays a central role in advancing breeding programs to improve crop performance, enabling the documentation of key traits and the detection of genetic variations associated with stress resistance (Bevan et al., 2017). As a core part of omics science, plant genomics concentrations on the investigation of whole plant genomes to understand their structure and function. Mapping of DNA sequences is essential for unlocking genomic data, studying evolutionary processes, and interpreting molecular phylogenetic relationships (Varshney, 2016). Quantitative trait loci analysis allows mapping of different traits of agronomic importance, thereby deepening our understanding of the genotype-phenotype relationship. Innovations such as gene cloning, haplotype-mediated breeding, allele exploration for tolerance to stress, and exploitation of intact variation are opening new avenues for genomics-assisted breeding (GAB) (Leng et al., 2017).

In the coming years, large genomic datasets will provide deeper insights into plant genomes, with ABM continuing to be an essential tool for improvement of crop (Varshney et al., 2019). NGS is an essential tool for trait mapping, enabling rapid gene identification and accelerating breeding programs. Advances in sequencing technologies have revolutionized the field, allowing researchers to discover diverse genetic variations associated with complex traits (Bassi et al., 2016). Single nucleotide polymorphisms (SNPs) are widely castoff as molecular markers to observe phenotypic diversity and genetic difference in crops, playing a key role in identifying genes associated with tolerance to stress and other important agronomical traits (Saxena et al., 2014).

Optical mapping techniques can overcome this limitation by providing long-read maps that capture greater variation and complexity (Golicz et al., 2016). For crops with larger genomes, such as wheat and corn, genotyping by sequencing combined with NGS offers a powerful method for sequencing multiple samples. Recent developments in genotyping and crop breeding micro-chips also support the application of ABM in modern animal husbandry (Rasheed et al., 2017). An inclusive understanding of the genes and regulatory pathways which affect quality, yield and stress tolerance will significantly improve crop development. However, challenges such as limited availability of genomic data and complex genotype-environment interactions remain, highlighting the need for continued innovation in genomic research (van Bezouw et al., 2019). Over the previous two decades, advances in genomic techniques have significantly enhanced breeding programs and the integration of genomic selection (GS), NGS, SNP-MAS, and GWAS, collected with phenotyping, bioinformatics and data analysis tools are essential to meet future crop yield needs (Bevan et al., 2017).

7.5 THE AGE OF PLANT GENOME EDITING

Although conventional mating has become more rapid in recent decades, it still fails to meet the growing global demand for agricultural production (Razzaq et al., 2019). While breeding using mutagen and the transgenic technologies can open new gateway to new genes for crop improvement, GM organisms face restrictions in some nations due to health of public concerns and the regulatory challenges. Traditionally, the development of a new crop using conventional, mutational, and transgenic breeding methods can take about 10–12 years (Razzaq et al., 2019). In addition to that the advent of genome engineering techniques, such as the transcriptional activator-effector nucleases and zinc finger nucleases (ZFNs), has significantly advanced plant research and accelerated progress in the field (Lloyd et al., 2005). These genome editing tools have enabled the creation of insertions/deletions (indels) and substitutions, thereby addressing concerns regarding non-specific integration and cross-species complications (Kim & Kim, 2014). Despite their potential, ZFNs and TALENs still have challenges in plant genome editing, including complex cloning and vector preparation, large size, inefficient transport, recurrent sequences, reduced accuracy, and significant off-target effects (Puchta, 2017).

7.5.1 Broadening the CRISPR/Cas Toolbox

The CRISPR/Cas (CRISPR-linked short palindromic repeat) system represents a revolutionary advance in genome editing. Ongoing study aims to address limitations of CRISPR-Cas9 in plants and advance NGGET. The current CRISPR toolbox is still evolving, with new classes of the variants being discovered by Koonin et al. (2017). The structure's potential has yet to be fully exploited, and its classification remains incomplete. There are currently 2 main categories (class 1 and class 2), 6 types, and more than 30 subtypes, which are classified according to their signature proteins. The English class 1 system includes several Cas effectors, such as Cas3, Cas10, and Csf1, present in types I, IV, and III (Makarova et al., 2015). The second class system, which is the most widely used, is characterized by a single signature protein and includes type II systems such as Cas9 and various Cas12 proteins (Cas12a, Cas12b, etc.) and Cas13 (Cas13a, Cas13b, etc.) (Koonin et al., 2017). Quite a lot of Cas orthologs have been identified to overcome the limitations of the CRISPR-Cas system.

Cas 9, originating from *Streptococcus pyogenes*, consists of three components: the Cas9 protein, CRISPR RNA, and the CRISPR trans-activator RNA (tracr-RNA) reported by Mali et al. (2013). Cas9 has 2 domains: a recognition domain and a nuclear domain, which are linked. The NUC domain contains two catalytic positions, HNH and RuvC, which aims the adjacent protospacer motif 3 base pairs upstream of DNA target place. The CRISPR/Cas9 mechanism begins with the pattern of a 20 bp guide RNA, which forms a complex with Cas9 to recognize the PAM site, creating double-strand breaks (DSBs) at the target site (Cong et al., 2013). CRISPR-Cas12a, also known as Cpf1, is an apparent Cas ortholog with unique features compared to Cas9. It requires a T-rich spacer location at the 5′ end and a 42 nt crRNA to generate DSBs with offset edges (Zetsche et al., 2015). In allotetraploid cotton, Cpf1-mediated editing resulted in 87% editing efficiency and no off-target cleavages (Li et al., 2019a). CRISPR-Cas9 and CRISPR-Cas13a techniques have been used to combat RNA potyviruses, resulting in disease-resistant crops and providing a means of resistance to viral pathogens (Aman et al., 2018). In addition, CRISPR-Cas14a shown to be effective in targeting single-stranded DNA viruses, such as those of the Nano-viridae and Gemini-viridae families, thus facilitating the advance of virus-resistant crops (Khan et al., 2019).

Significant advancements in plant genome engineering have been achieved through the use of the CRISPR-Cas system that has shown great promise in developing plants with enhanced stress resilience (Shan et al., 2013). CRISPR-Cas technology is transforming plant breeding by enabling the formation of climate-resilient crops (Puchta, 2017). Drought, a major abiotic stressor, causes considerable crop yield losses. The CRISPR-Cas9 system was employed to generate sensation mutants of the SlLBD40 gene in case of tomato plants under drought conditions (Liu et al., 2020). CRISPR-Cas9 was used to examine the effects of SlNPR1 mutants beneath drought stress, with the lines displaying heightened susceptibility to drought (Li et al., 2019b). Cas orthologs used for plant genome editing are indexed in Table 7.1.

7.5.2 Recent Innovations in CRISPR/Cas System

Evolution of next-generation CRISPR-Cas technology continues to revolutionize plant breeding research. Advances in this system have addressed many of the existing challenges in genome editing, including improving targeting accuracy, expanding the range of targeting, minimizing off-target effects, and improving nuclease activity. In addition, innovations such as PAM site diversity and efficient delivery methods have significantly expanded its applications (Koonin et al., 2017). This technology supports a variety of genetic alterations, such as exploring mutation outlines (Jia et al., 2018), integration of foreign genes (Collonnier et al., 2017), gene regulation (Qi and Innes, 2013), and cell imaging (Xue & Acar, 2018).

A protocol introduced by Maher et al. (2020) successfully circumvents the painstaking and cumbersome nature of the tissue culture processes. Similarly, Ren et al. (2019) established a bidirectional promoter system to simultaneously express gRNA and Cas9 cassettes, achieving editing efficiencies of 75.9–93.3% in rice. Another advance involves the CRISPR-TSKO system, designed by Decaestecker et al. (2019), which creates tissue-specific knockout mutants. This innovation offers new opportunities for crop improvement by targeting specific genes in specific tissues. The efficacy of Cas nucleases can be affected by their dependence on specific PAM sites, which often limit their ability to target GC-rich regions. To address this limitation, Ren et al. (2021) developed the PAM free CRISPR-SpRY toolkit in rice, which allows editing across a wider range of DNA sequences.

TABLE 7.1
List of Different Cas Orthologs Genes Used for Plant Genome Editing

Cas Type	Organism	PAM	Plant Species	Characteristics	References
SpCas9	*Streptococcus pyrogenes*	NGG	Several plants	Need long crRNA+ tracr RNA	Jinek et al. (2012)
SpCas9 QQR1		NAAG	–	Alter PM sequence	Cong et al. (2013)
SpCas9 VRER		NGOG	Rice	Altered PAM sequence	Kleinstiver et al. (2015)
SpCas9-NG		NG	Rice	Altered PAM sequence greater ability of base editing and gene regulation	Ren et al. (2019)
SaCas9	*Staphylococcus aureus*	NNAGRRT	Rice and citrus	Reduce off-targets and excellent in VIVO genome editing	Kaya (2016)
FnCas9	*Francisella novicida*	NGG	–	Reduce off-targets	Hirano et al. (2016)
ScCas9	*Streptococcus canis*	NNG	–	Altered PAM sequence and reduced off-targets	Chatterjee et al. (2018)
Nme Cas9	*Neisseria meningitidis*	NNNNGATT	–	Reduce off-targets and need longer PAM	Lee et al. (2016)
BlatCas9	*Brevibacillus laterosporus*	NNNNCND	Maize	Enhance specificity	Karvelis et al. (2015)
St1Cas9	*Streptococcus thermophilus*	NNAGAAW	*Arabidopsis*	Minimize off-targets	Steinert et al. (2015)
St3Cas9		NGGNG	–	Many domains and introduce dsDNA breaks	Cong et al. (2013)
HypaCas9	*Streptococcus pyogenes*	NGG	Rice	Increased specificity	Chen et al. (2017)
eHypa-Cas9		NGG	Rice	Increased specificity	Liang et al. (2018)
CjCas9	*Campylobacter jejuni*	NNNNRYAC or NNNNACAC	.	Greater mutation frequency	Kim et al. (2017)
xCas9 3.7	*S. pyogenes*	GAT GAA NG	Rice	Altered PAM and increased specificity	Zhong et al. (2019)
CasX	*Planctomycetes* and *Phyla Deltaproteobacteria*	TTCN	–	Increased specificity	Burstein et al. (2017)
AsCpf1	*Acidaminococcus* sp.	TTTN	–	Increase editing efficiency	Yamano et al. (2016)
Cpf1	*Francisella* and *Prevotella*	TTTV	Rice and *Arabidopsis*	Needs long sgRNA and lacks HNH domain	Endo et al. (2016)

TABLE 7.1 (*Continued*)
List of Different Cas Orthologs Genes Used for Plant Genome Editing

Cas Type	Organism	PAM	Plant Species	Characteristics	References
FnCpf1	*F. novicida*	TTTV and TTV	Rice	Enhanced efficiency and altered PM	Zhong et al. (2018)
Cas12a	*Acidaminococcus* sp.	TTTV		Altered PAM	Jeon et al. (2018)
LbCas12a RR	*Francisella* and *Prevotella*	CCCC and TYCV	Rice	Altered PM	Kleinstiver et al. (2019)
AsCas12a RVR		TATV		Altered PM	Kleinstiver et al. (2019)
FnCas12a RVR		TWTV	Rice	Altered PM	Zhong et al. (2018)
MbCas12a RR		TCTV and TYCV	–	Altered PM	Toth et al. (2018)
Cas13 (C2c2)	*Leptotrichia Shaii*	Not needed	–	Cleaved RNA	Abudayyeh et al. (2016)
AacC2c1	*Alicyclobacillus acidoterrestris*	T-rich PAM	–	Bi-lobed endonucleases	Liu et al. (2017)
Cas14	*Archaea*			Restrictive sequence not required for ssDNA cleavage	Harrington et al. (2019)

7.6 PAN-GENOMES AND CROP IMPROVEMENT

Genome-wide plant studies help identify genes missing from reference genomes due to the process of crop domestication. The crop pan-genome, which includes wild relatives, landraces, and cultivated varieties, provides a comprehensive framework for understanding genotypic and phenotypic variation, helping to uncover genes missing from reference genomes (Danilevicz et al., 2020). A deeper understanding of the accessory genome can support the screening of elite cultivars for stress response genes useful for the abiotic and biotic stress tolerance. Pan-genomes are also essential for elucidating the role of gene duplication in crop improvement (Figure 7.2).

7.7 BIOINFORMATICS DATABASES AND TOOLS FOR DATA ANALYSIS IN CROP BREEDING

NGS products large datasets from breeding populations, which can be analyzed using techniques such as GWAS or GBS. After sequencing, bioinformatics methods are used to manage and analyze this big data. Bioinformatics also provides tools for forward and reverse genetics. Common bioinformatics databases for nucleotide sequences include GenBank at NCBI, the DNA Data Bank of Japan, and the European Nucleotide Archive. For plant genomic data, Ensembl Plants is a key resource. Essential tools for Gene Ontology and similarity examinations include

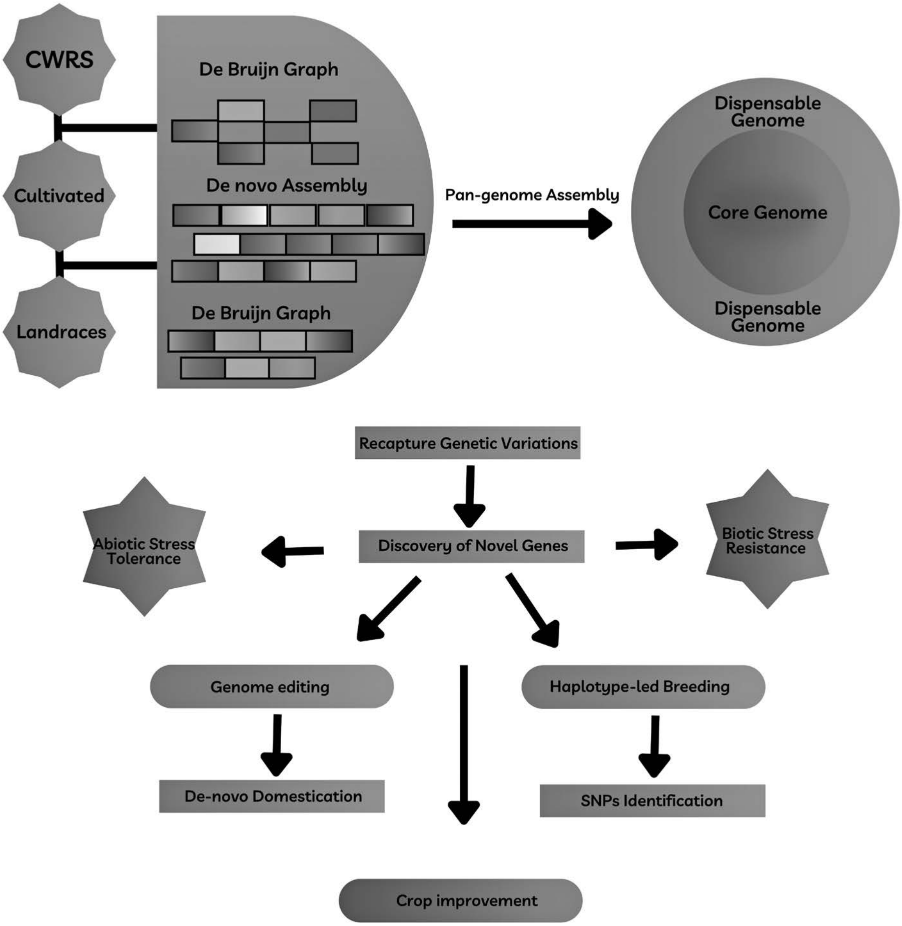


FIGURE 7.2 Harnessing pan-genomes to uncover genetic diversity for resilient crops.

NCBI, BLAST, GOA, GO, UniProtKB, and KEGG. Data from the NGS pareas are processed with various bioinformatics tools, which help to link plant phenotypes with genotypes, thereby allowing the identification of relevant genes or markers (Kersey, 2019).

7.8 MACHINE LEARNING AND ARTIFICIAL INTELLIGENCE

Current technological advances and high-throughput methods have produced a large amount of data on plant genotypes and phenotypes, requiring new efforts to extract meaningful information and integrate these datasets. At the same time, ML has made rapid progress and is now widely applied to plant genotyping and phenotyping (van Dijk et al., 2021). Genomics involves not only the collection of molecular profiling in phenotypes but also the application of efficient data processing tools to predict and characterize these phenotypes (Wang et al., 2020). ML a growing multidisciplinary field provides computational and analytical methods to incorporate large, heterogeneous, and unstructured datasets at the scale of big data. It is increasingly recognized as a valuable tool in biology (Jordan & Mitchell, 2015).

The basic principle of ML is the effective use of past experiences to discover underlying structures, similarities, and differences in data, which allow for accurate description or categorization of new information (Singh et al., 2016). ML algorithms excel at handling large datasets that may contain significant noise, high dimensionality, or incomplete information (Liu et al., 2020). ML, a subset of artificial intelligence (AI), is emerging as a powerful tool. AI refers to advanced computer systems capable of autonomously learning and improving their performance without the need for extensive programming (Singh et al., 2016). Currently, the success of genomic breeding depends on the continuous development of ML techniques and the collection of large genotype datasets associated with important agronomic traits (Tong & Nikoloski, 2021).

7.9 OPTIMIZING THE MATCH BETWEEN PLANTS AND THEIR ENVIRONMENT

An effective approach to crop adaptation to the climate change is to align crop biology with current and projected climate conditions. This adjustment could occur without significant selection effort. For example, environmental and cultural models (Franke et al., 2022) propose shifting agricultural activities to regions more suitable for cultivation, such as higher latitudes. However, this strategy faces challenges, including limited land availability and significant ecological costs at these latitudes (Sloat et al., 2020). Modeling tool also helps identify crops that can thrive on existing agricultural lands under current and the future climate scenarios (Zabel et al., 2021). By optimizing the compatibility between crops and their environment, productivity and resilience to climate stressors can be improved, thus reducing dependence on resource-intensive inputs like water, fertilizers, and pesticides (Mueller et al., 2012).

7.9.1 Crop Modeling

Crop models act as mathematical frameworks designed to replicate the process of crop production (Hammer et al., 2006). These tools facilitate the prediction of plants performance (genotypes) under an area of environmental scenarios, including climate variables and soil characteristics, as well as different management strategies. By assessing the interactions between genotypes, environment, and management ($G \times E \times M$) (Peng et al., 2020), crop systems helps in detecting the most suitable crop varieties for specific locations (Zabel et al., 2021). They also provide recommendations for optimizing agricultural practices to improve productivity and promote sustainable agriculture (Chenu et al., 2018; Hammer et al., 2006).

7.9.2 TAPPING CROP WILD RELATIVES

Wild species of cultivated plants often survive in harsher niche than their domesticated counterparts (Rawat et al., 2022), all due to the genomic constitution that provide genetic resistance to the stresses such as high temperatures, drought, salinity, and nutrient deficiency. To take advantage of this resilience, a common strategy for crop adaptation to climate change is to incorporate favorable traits and genes from the wild relatives into crop varieties through prebreeding efforts (Hubner & Kantar, 2021). The economic impact of such approaches is considerable. According to Coopers (2013), traits derived from wild relatives of cultivated plants in 32 crops generate an annual economic value of $68 billion, with projections suggesting that this figure could reach almost $200 billion in the future. One particularly promising application is the transfer of perennial traits from wild relatives to cereal crops, which has the potential to domesticate environmental disadvantages associated with the production of annual crops.

7.9.3 SPEED BREEDING

Traditionally, plant breeding been a time-consuming method, with the time between the early cross and the commercialization of a new variety ranging from 5 to 30 years, reliant on the species involved (Bernardo, 2014). The timing of selection is greatly influenced by the life span of the target species. Therefore, one of the main goals of modern breeding technique is to shorten life cycles, thereby accelerating the development and commercialization of new verity (Wanga et al., 2021). Speed breeding provides a solution by creating controlled environments that accelerate crop life cycles. Techniques such as drying, chilling, and embryo rescue also allow for the harvesting and sprouting of immature seeds (Watson et al., 2018). Rapid breeding procedures have been applied to several field crops, in some cases tumbling the time required for variety advance by half (Wanga et al., 2021). This method has proven particularly useful for the selection of self-pollinated and the hybrid crops, facilitating the rapid advance of the inbred lines from a single seed. It also supports posterior selection, trait clustering, and the integration of transgenic or genome editing technologies (Hickey et al., 2019).

7.9.4 GENOMIC SELECTION

GS focuses on the use of genome-wide markers for the selection rather than based solely on phenotypic traits (Daetwyler et al., 2013). This method is grounded on the identification of genotype-phenotype relationships, usually located in test the populations. As soon as these connotations are determined, phenotype can be accurately predicted using marker data alone, allowing for the rapid elimination of unwanted genotypes at the seed or seedling stage (Jannink et al., 2010). This strategy significantly lessens the space and resources essential to maintain large breeding populations and perform costly phenotyping (Wartha & Lorenz, 2021).

7.9.5 Genetic Transformation

The process of genetically transforming plants often relies on *Agrobacterium*, a bacterium that naturally infects and modifies plant genomes (Song et al., 2019) and tissue types (Ozyigit & Yucebilgili Kurtoglu, 2020). Although some cultures are still difficult to process, the practice of morphogenic TFs can enhance transformation rates, thereby expanding the range of species and genotypes suitable for efficient revolution. *Agrobacterium*-mediated transformation is commonly carried out *in vitro* but can encounter obstacles such as chimeric regeneration and the inability of certain species or genotypes to regenerate (Hirano et al., 2016). To address these limitations, the in-planta method has been developed for some crops, removing the dependency on tissue culture (Niazian et al., 2017). In sugarcane, for example, a single TALEN pair was used to edit 107 of 109 copies of the caffeic acid O-methyltransferase gene (Kannan et al., 2018).

7.9.6 A "Silver Bullet" to Improve Drought Performance

Improving crop performance under drought conditions is challenging due to the complexity and variability of plant physiological responses, which are influenced by factors such as the timing, duration, and intensity of drought. These responses can often be contradictory and are controlled by multiple genes, making them difficult to manage through traditional breeding, marker-assisted breeding, transgenesis, or gene editing. In drought conditions, plants may increase the production of abscisic acid (ABA), triggering various feedback mechanisms within their eco physiological networks. Both genetic and environmental factors can affect plant growth, with impacts that can be either beneficial or detrimental in both the short and long term. Drought responses are complex and context-dependent, typically showing low heritability and significant pleiotropic effects, especially in traits like flowering time and grain quality. This complexity suggests that breeding efforts should not focus solely on "drought resistance," but should also ensure it is closely linked to improved performance under both drought and optimal water conditions for rice (Raman et al., 2012).

7.10 CONCLUSION

This chapter has explored the transformative power of advanced technologies such as MAS, GWAS, CRISPR-Cas systems, and NGS in developing climate-resilient crops. These tools allow for precise genetic modifications to enhance traits like drought tolerance, heat resistance, and pest resilience, which are essential for maintaining agricultural sustainability. The integration of bioinformatics, pan-genomics, and ML has further accelerated crop improvement, enabling researchers to bridge the gap between genotype and phenotype with unmatched efficiency. Public acceptance, regulatory concerns, and ensuring that smallholder farmers have equitable access to these technologies remain significant hurdles. Addressing these challenges will require collaborative efforts from policymakers, researchers, and industry leaders. Looking ahead, the future of agriculture depends on combining molecular breeding with sustainable farming practices.

REFERENCES

Abbass, K., Qasim, M. Z., Song, H., Murshed, M., Mahmood, H., & Younis, I. (2022). A review of the global climate change impacts, adaptation, and sustainable mitigation measures. *Environmental Science and Pollution Research*, *29*(28), 42539–42559. https://doi.org/10.1007/s11356-022-19718-6

Abberton, M., Batley, J., Bentley, A., Bryant, J., Cai, H., Cockram, J., & Yano, M. (2016). Global agricultural intensification during climate change: A role for genomics. *Plant Biotechnology Journal*, *14*(4), 1095–1098. https://doi.org/10.1111/pbi.12538

Abudayyeh, O. O., Gootenberg, J. S., Konermann, S., Joung, J., Slaymaker, I. M., Cox, D. B. T., Shmakov, S., Makarova, K. S., Semenova, E., Minakhin, L., Severinov, K., Regev, A., Lander, E. S., Koonin, E. V., & Zhang, F. (2016). C2c2 is a single-component programmable RNA-guided RNA-targeting CRISPR effector. *Science*, *353*(6299). https://doi.org/10.1126/science.aaf5573

Aman, R., Ali, Z., Butt, H., Mahas, A., Aljedaani, F., Khan, M. Z., & Mahfouz, M. (2018). RNA virus interference via CRISPR/Cas13a system in plants. *Genome Biology*, *19*(1), 1. https://doi.org/10.1186/s13059-017-1381-1

Bassi, F. M., Bentley, A. R., Charmet, G., Ortiz, R., & Crossa, J. (2016). Breeding schemes for the implementation of genomic selection in wheat (*Triticum* spp.). *Plant Science*, *242*, 23–36. https://doi.org/10.1016/j.plantsci.2015.08.021

Becker, M., Gruenheit, N., Steel, M., Voelckel, C., Deusch, O., Heenan, P. B., & Lockhart, P. J. (2013). Hybridization may facilitate *in situ* survival of endemic species through periods of climate change. *Nature Climate Change*, *3*(12), 1039–1043. https://doi.org/10.1038/nclimate2027

Bernardo, R. (2014). *Essentials of plant breeding*. Stemma Press. https://bernardo-group.org/wp-content/uploads/2015/11/EoPB_sample_pages.pdf

Bevan, M. W., Uauy, C., Wulff, B. B., Zhou, J., Krasileva, K., & Clark, M. D. (2017). Genomic innovation for crop improvement. *Nature*, *543*(7645), 346–354. https://doi.org/10.1038/nature22011

Burstein, D., Harrington, L. B., Strutt, S. C., Probst, A. J., Anantharaman, K., Thomas, B. C., Doudna, J. A., & Banfield, J. F. (2016). New CRISPR–Cas systems from uncultivated microbes. *Nature*, *542*(7640), 237–241. https://doi.org/10.1038/nature21059

Chatterjee, C., Gleddie, S., & Xiao, C. W. (2018). Soybean bioactive peptides and their functional properties. *Nutrients*, *10*(9), 1211.

Chen, J., Nolan, T. M., Ye, H., Zhang, M., Tong, H., Xin, P., Chu, J., Chu, C., Li, Z., & Yin, Y. (2017). Arabidopsis WRKY46, WRKY54 and WRKY70 transcription factors are involved in Brassinosteroid-Regulated plant growth and drought response. *The Plant Cell*, tpc.00364.2017. https://doi.org/10.1105/tpc.17.00364

Chenu, K., Van Oosterom, E. J., McLean, G., Deifel, K. S., Fletcher, A., Geetika, G., Tirfessa, A., Mace, E. S., Jordan, D. R., Sulman, R., & Hammer, G. L. (2018). Integrating modelling and phenotyping approaches to identify and screen complex traits: transpiration efficiency in cereals. *Journal of Experimental Botany*, *69*(13), 3181–3194. https://doi.org/10.1093/jxb/ery059

Collonnier, C., Guyon-Debast, A., Maclot, F., Mara, K., Charlot, F., & Nogué, F. (2017). Towards mastering CRISPR-induced gene knock-in in plants: Survey of key features and focus on the model *Physcomitrella patens*. *Methods*, *121*, 103–117. https://doi.org/10.1016/j.ymeth.2017.04.024

Cong, L., Ran, F. A., Cox, D., Lin, S., Barretto, R., Habib, N., & Zhang, F. (2013). Multiplex genome engineering using CRISPR/Cas systems. *Science*, *339*(6121), 819–823. https://doi.org/10.1126/science.1231143

Coopers, P. W. (2013). *Crop wild relatives: A valuable resource for crop development*. PWC Valuations.

Daetwyler, H. D., Calus, M. P., Pong-Wong, R., de Los Campos, G., & Hickey, J. M. (2013). Genomic prediction in animals and plants: Simulation of data, validation, reporting, and benchmarking. *Genetics*, *193*(2), 347–365. https://doi.org/10.1534/genetics.112.147983

Danilevicz, M. F., Fernandez, C. G. T., Marsh, J. I., Bayer, P. E., & Edwards, D. (2020). Plant pangenomics: Approaches, applications and advancements. *Current Opinion in Plant Biology*, *54*, 18–25. https://doi.org/10.1016/j.pbi.2019.12.005

Decaestecker, W., Buono, R. A., Pfeiffer, M. L., Vangheluwe, N., Jourquin, J., Karimi, M., & Jacobs, T. B. (2019). CRISPR-TSKO: A technique for efficient mutagenesis in specific cell types, tissues, or organs in *Arabidopsis*. *The Plant Cell*, *31*(12), 2868–2887. https://doi.org/10.1105/tpc.19.00454

Endo, A., Masafumi, M., Kaya, H., & Toki, S. (2016). Efficient targeted mutagenesis of rice and tobacco genomes using Cpf1 from Francisella novicida. *Scientific Reports*, *6*(1), 38169.

Fischer, R. A., Byerlee, D., & Edmeades, G. (2014). *Crop yields and global food security* (pp. 8–11). ACIAR: Canberra, ACT.

Franke, J. A., Müller, C., Minoli, S., Elliott, J., Folberth, C., Gardner, C., & Moyer, E. J. (2022). Agricultural breadbaskets shift poleward given adaptive farmer behavior under climate change. *Global Change Biology*, *28*(1), 167–181. https://doi.org/10.1111/gcb.15868

Golicz, A. A., Batley, J., & Edwards, D. (2016). Towards plant pangenomics. *Plant Biotechnology Journal*, *14*(4), 1099–1105. https://doi.org/10.1111/pbi.12499

Hammer, G., Cooper, M., Tardieu, F., Welch, S., Walsh, B., van Eeuwijk, F., & Podlich, D. (2006). Models for navigating biological complexity in breeding improved crop plants. *Trends in Plant Science*, *11*(12), 587–593. https://doi.org/10.1016/j.tplants.2006.10.006

Harrington, S. A., Cobo, N., Karafiátová, M., Doležel, J., Borrill, P., & Uauy, C. (2019). Identification of a dominant chlorosis phenotype through a forward screen of the Triticum turgidum cv. Kronos TILLING population. *Frontiers in Plant Science*, *10*, 963.

Hickey, L. T., Hafeez, A. N., Robinson, H., Jackson, S. A., Leal-Bertioli, S. C., Tester, M., & Wulff, B. B. (2019). Breeding crops to feed 10 billion. *Nature Biotechnology*, *37*(7), 744–754. https://doi.org/10.1038/s41587-019-0152-9

Hirano, H., Gootenberg, J. S., Horii, T., Abudayyeh, O. O., Kimura, M., Hsu, P. D., Nakane, T., Ishitani, R., Hatada, I., Zhang, F., Nishimasu, H., & Nureki, O. (2016). Structure and Engineering of Francisella novicida Cas9. *Cell*, *164*(5), 950–961. https://doi.org/10.1016/j.cell.2016.01.039

Hubner, S., & Kantar, M. B. (2021). Tapping diversity from the wild: From sampling to implementation. *Frontiers in Plant Science*, *12*, 626565.

ISAAA. (2021). *GM approval database*. https://www.isaaa.org/gmapprovaldatabase/default.asp

James, C. (1997). *Global status of transgenic crops in 1997* (ISAAA Brief No. 5). ISAAA.

Jannink, J. L., Lorenz, A. J., & Iwata, H. (2010). Genomic selection in plant breeding: From theory to practice. *Briefings in Functional Genomics*, *9*(2), 166–177. https://doi.org/10.1093/bfgp/elq001

Jeon, Y. M., Son, K. H., Kim, S. M., & Oh, M. M. (2018). Growth of dropwort plants and their accumulation of bioactive compounds after exposure to UV lamp or LED irradiation. *Horticulture, Environment, and Biotechnology, 59*(5), 659–670.

Jia, C., Huai, C., Ding, J., Hu, L., Su, B., Chen, H., & Lu, D. (2018). New applications of CRISPR/Cas9 system on mutant DNA detection. *Gene*, *641*, 55–62. https://doi.org/10.1016/j.gene.2017.10.023

Jinek, M., Chylinski, K., Fonfara, I., Hauer, M., Doudna, J. A., & Charpentier, E. (2012). A programmable dual-RNA–guided DNA endonuclease in adaptive bacterial immunity. *Science*, *337*(6096), 816–821.

John, D. A., & Babu, G. R. (2021). Lessons from the aftermaths of green revolution on food system and health. *Frontiers in Sustainable Food Systems*, *5*, 644559. https://doi.org/10.3389/fsufs.2021.644559

Jordan, M. I., & Mitchell, T. M. (2015). Machine learning: Trends, perspectives, and prospects. *Science*, *349*(6245), 255–260. https://doi.org/10.1126/science.aaa8415

Kannan, B., Jung, J. H., Moxley, G. W., Lee, S. M., & Altpeter, F. (2018). TALEN-mediated targeted mutagenesis of more than 100 *COMT* copies/alleles in highly polyploid sugarcane improves saccharification efficiency without compromising biomass yield. *Plant Biotechnology Journal*, *16*(4), 856–866. https://doi.org/10.1111/pbi.12833

Karvelis, T., Gasiunas, G., Young, J., Bigelyte, G., Silanskas, A., Cigan, M., & Siksnys, V. (2015). Rapid characterization of CRISPR-Cas9 protospacer adjacent motif sequence elements. *Genome Biology*, *16*(1), 253.

Kersey, P. J. (2019). Plant genome sequences: Past, present, future. *Current Opinion in Plant Biology*, *48*, 1–8. https://doi.org/10.1016/j.pbi.2018.11.001

Khan, M. Z., Haider, S., Mansoor, S., & Amin, I. (2019). Targeting plant ssDNA viruses with engineered miniature CRISPR-Cas14a. *Trends in Biotechnology*, *37*(8), 800–804. https://doi.org/10.1016/j.tibtech.2019.03.015

Kim, H., & Kim, J. S. (2014). A guide to genome engineering with programmable nucleases. *Nature Reviews Genetics*, *15*(5), 321–334. https://doi.org/10.1038/nrg3686

Kim, H., Kim, S. T., Ryu, J., Kang, B. C., Kim, J. S., & Kim, S. G. (2017). CRISPR/Cpf1-mediated DNA-free plant genome editing. *Nature Communications*, *8*(1), 14406.

Kleinstiver, B. P., Prew, M. S., Tsai, S. Q., Topkar, V. V., Nguyen, N. T., Zheng, Z., Gonzales, A. P. W., Li, Z., Peterson, R. T., Yeh, J. J., Aryee, M. J., & Joung, J. K. (2015). Engineered CRISPR-Cas9 nucleases with altered PAM specificities. *Nature*, *523*(7561), 481–485. https://doi.org/10.1038/nature14592

Kleinstiver, B. P., Sousa, A. A., Walton, R. T., Tak, Y. E., Hsu, J. Y., Clement, K., Welch, M. M., Horng, J. E., Malagon-Lopez, J., Scarfò, I., Maus, M. V., Pinello, L., Aryee, M. J., & Joung, J. K. (2019). Engineered CRISPR–Cas12a variants with increased activities and improved targeting ranges for gene, epigenetic and base editing. *Nature Biotechnology*, *37*(3), 276–282. https://doi.org/10.1038/s41587-018-0011-0

Koonin, E. V., Makarova, K. S., & Zhang, F. (2017). Diversity, classification and evolution of CRISPR-Cas systems. *Current Opinion in Microbiology*, *37*, 67–78. https://doi.org/10.1016/j.mib.2017.05.008

Lavarenne, J., Guyomarc'h, S., Sallaud, C., Gantet, P., & Lucas, M. (2018). The spring of systems biology-driven breeding. *Trends in Plant Science*, *23*(8), 706–720. https://doi.org/10.1016/j.tplants.2018.04.005

Lee, J., Cho, C. H., Park, S. I., Choi, J. W., Song, H. S., West, J. A., Bhattacharya, D., & Yoon, H. S. (2016). Parallel evolution of highly conserved plastid genome architecture in red seaweeds and seed plants. *BMC Biology*, *14*(1). https://doi.org/10.1186/s12915-016-0299-5

Leng, P. F., Lübberstedt, T., & Xu, M. L. (2017). Genomics-assisted breeding—A revolutionary strategy for crop improvement. *Journal of Integrative Agriculture*, *16*(12), 2674–2685. https://doi.org/10.1016/S2095-3119(17)61813-6

Li, B., Rui, H., Li, Y., Wang, Q., Alariqi, M., Qin, L., & Jin, S. (2019a). Robust CRISPR/Cpf1 (Cas12a)-mediated genome editing in allotetraploid cotton (*Gossypium hirsutum*). *Plant Biotechnology Journal*, *17*(10), 1862–1864. https://doi.org/10.1111/pbi.13147

Li, R., Liu, C., Zhao, R., Wang, L., Chen, L., Yu, W., & Shen, L. (2019b). CRISPR/Cas9-mediated *SlNPR1* mutagenesis reduces tomato plant drought tolerance. *BMC Plant Biology*, *19*(1), 1–13. https://doi.org/10.1186/s12870-019-2117-z

Liang, W., Ma, X., Wan, P., & Liu, L. (2018). Plant salt-tolerance mechanism: A review. *Biochemical and Biophysical Research Communications*, *495*(1), 286–291.

Liu, L., Zhang, J., Xu, J., Li, Y., Guo, L., Wang, Z., & Zhang, N. (2020). CRISPR/Cas9 targeted mutagenesis of *SlLBD40*, a lateral organ boundaries domain transcription factor, enhances drought tolerance in tomato. *Plant Science*, *301*, 110683. https://doi.org/10.1016/j.plantsci.2020.110683

Liu, W. W., Meng, J., Cui, J., & Luan, Y. S. (2017). Characterization and function of microRNA* s in plants. *Frontiers in Plant Science*, *8*, 2200.

Lloyd, A., Plaisier, C. L., Carroll, D., & Drews, G. N. (2005). Targeted mutagenesis using zinc-finger nucleases in *Arabidopsis*. *Proceedings of the National Academy of Sciences*, *102*(6), 2232–2237. https://doi.org/10.1073/pnas.0409339102

Maher, M. F., Nasti, R. A., Vollbrecht, M., Starker, C. G., Clark, M. D., & Voytas, D. F. (2020). Plant gene editing through *de novo* induction of meristems. *Nature Biotechnology*, *38*(1), 84–89. https://doi.org/10.1038/s41587-019-0337-2

Makarova, K. S., Wolf, Y. I., Alkhnbashi, O. S., Costa, F., Shah, S. A., Saunders, S. J., & Koonin, E. V. (2015). An updated evolutionary classification of CRISPR-Cas systems. *Nature Reviews Microbiology*, *13*(11), 722–736. https://doi.org/10.1038/nrmicro3569

Malhi, G. S., Kaur, M., & Kaushik, P. (2021). Impact of climate change on agriculture and its mitigation strategies: A review. *Sustainability*, *13*(3), 1318. https://doi.org/10.3390/su13031318

Mali, P., Yang, L., Esvelt, K. M., Aach, J., Guell, M., DiCarlo, J. E., & Church, G. M. (2013). RNA-guided human genome engineering via Cas9. *Science*, *339*(6121), 823–826. https://doi.org/10.1126/science.1232033

McCouch, S., Baute, G. J., Bradeen, J., Bramel, P., Bretting, P. K., Buckler, E., & Zamir, D. (2013). Feeding the future. *Nature*, *499*(7456), 23–24. https://doi.org/10.1038/499023a

Mueller, N. D., Gerber, J. S., Johnston, M., Ray, D. K., Ramankutty, N., & Foley, J. A. (2012). Closing yield gaps through nutrient and water management. *Nature*, *490*(7419), 254–257. https://doi.org/10.1038/nature11420

Naqvi, R. Z., Siddiqui, H. A., Mahmood, M. A., Najeebullah, S., Ehsan, A., Azhar, M., & Asif, M. (2022). Smart breeding approaches in post-genomics era for developing climate-resilient food crops. *Frontiers in Plant Science*, *13*, 972164. https://doi.org/10.3389/fpls.2022.972164

Niazian, M., Noori, S. A. S., Galuszka, P., & Mortazavian, S. M. M. (2017). Tissue culture-based *Agrobacterium*-mediated and *in planta* transformation methods. *Methods in Molecular Biology*, *1679*, 25–40. https://doi.org/10.1007/978-1-4939-7337-8_2

Ozyigit, I. I., & Yucebilgili Kurtoglu, K. (2020). Particle bombardment technology and its applications in plants. *Molecular Biology Reports*, *47*(12), 9831–9847. https://doi.org/10.1007/s11033-020-06007-z

Peng, B., Guan, K., Tang, J., Ainsworth, E. A., Asseng, S., Bernacchi, C. J., Cooper, M., Delucia, E. H., Elliott, J. W., Ewert, F., Grant, R. F., Gustafson, D. I., Hammer, G. L., Jin, Z., Jones, J. W., Kimm, H., Lawrence, D. M., Li, Y., Lombardozzi, D. L., . . . Zhou, W. (2020). Towards a multiscale crop modelling framework for climate change adaptation assessment. *Nature Plants*, *6*(4), 338–348. https://doi.org/10.1038/s41477-020-0625-3

Pickson, R. B., & Boateng, E. (2022). Climate change: A friend or foe to food security in Africa? *Environment, Development and Sustainability*, *24*(6), 7913–7938. https://doi.org/10.1007/s10668-021-01775-5

Pingali, P. L. (2012). Green revolution: Impacts, limits, and the path ahead. *Proceedings of the National Academy of Sciences*, *109*(31), 12302–12308. https://doi.org/10.1073/pnas.0912953109

Puchta, H. (2017). Applying CRISPR/Cas for genome engineering in plants: The best is yet to come. *Current Opinion in Plant Biology*, *36*, 1–8. https://doi.org/10.1016/j.pbi.2016.11.011

Purugganan, M. D., & Fuller, D. Q. (2009). The nature of selection during plant domestication. *Nature*, *457*(7231), 843–848. https://doi.org/10.1038/nature07895

Qi, D., & Innes, R. W. (2013). Recent advances in plant NLR structure, function, localization, and signaling. *Frontiers in Immunology*, *4*, 348.

Raman, A., Verulkar, S., Mandal, N., Variar, M., Shukla, V., Dwivedi, J., & Kumar, A. (2012). Drought yield index to select high yielding rice lines under different drought stress severities. *Rice*, *5*(1), 1–12. https://doi.org/10.1186/1939-8433-5-31

Rasheed, A., Hao, Y., Xia, X., Khan, A., Xu, Y., Varshney, R. K., and He, Z. (2017). Crop breeding chips and genotyping platforms: Progress, challenges, and perspectives. *Molecular Plant*, *10*(8), 1047–1064. https://doi.org/10.1016/j.molp.2017.06.008

Rawat, N., Wungrampha, S., Singla-Pareek, S. L., Yu, M., Shabala, S., & Pareek, A. (2022). Rewilding staple crops for the lost halophytism: Toward sustainability and profitability of agricultural production systems. *Molecular Plant*, *15*(1), 45–64. https://doi.org/10.1016/j.molp.2021.12.003

Ray, D. K., Gerber, J. S., MacDonald, G. K., & West, P. C. (2015). Climate variation explains a third of global crop yield variability. *Nature Communications*, *6*(1), 5989. https://doi.org/10.1038/ncomms6989

Razzaq, A., Saleem, F., Kanwal, M., Mustafa, G., Yousaf, S., Imran Arshad, H. M., and Joyia, F. A. (2019). Modern trends in plant genome editing: An inclusive review of the CRISPR/Cas9 toolbox. *International Journal of Molecular Sciences*, *20*(16), 4045. https://doi.org/10.3390/ijms20164045

Ren, Q., Sretenovic, S., Liu, S., Tang, X., Huang, L., He, Y., and Zhang, Y. (2021). PAM-less plant genome editing using a CRISPR–SpRY toolbox. *Nature Plants*, *7*(1), 25–33. https://doi.org/10.1038/s41477-020-00827-4

Ren, Q., Zhong, Z., Wang, Y., You, Q., Li, Q., Yuan, M., & Zhang, Y. (2019). Bidirectional promoter-based CRISPR-Cas9 systems for plant genome editing. *Frontiers in Plant Science*, *10*, 1173. https://doi.org/10.3389/fpls.2019.01173

Saad, N. S. M., Neik, T. X., Thomas, W. J., Amas, J. C., Cantila, A. Y., Craig, R. J., & Batley, J. (2022). Advancing designer crops for climate resilience through an integrated genomics approach. *Current Opinion in Plant Biology*, *67*, 102220. https://doi.org/10.1016/j.pbi.2022.102220

Saxena, R. K., Edwards, D., & Varshney, R. K. (2014). Structural variations in plant genomes. *Briefings in Functional Genomics*, *13*(4), 296–307. https://doi.org/10.1093/bfgp/elu016

Shan, Q., Wang, Y., Li, J., Zhang, Y., Chen, K., Liang, Z., & Gao, C. (2013). Targeted genome modification of crop plants using a CRISPR-Cas system. *Nature Biotechnology*, 31(8), 686–688.

Sloat, L. L., Davis, S. J., Gerber, J. S., Moore, F. C., Ray, D. K., West, P. C., & Mueller, N. D. (2020). Climate adaptation by crop migration. *Nature Communications*, *11*(1), 1243.

Steinert, J., Schiml, S., Fauser, F., & Puchta, H. (2015). Highly efficient heritable plant genome engineering using Cas9 orthologues from Streptococcus thermophilus and Staphylococcus aureus. *The Plant Journal*, *84*(6), 1295–1305.

Toth, J., Pandurangan, S., Burt, A., Mitchell Fetch, J., & Kumar, S. (2018). Marker-assisted breeding of hexaploid spring wheat in the Canadian prairies. *Canadian Journal of Plant Science*, 99(2), 111–127.

van Bezouw, R. F., Keurentjes, J. J., Harbinson, J., & Aarts, M. G. (2019). Converging phenomics and genomics to study natural variation in plant photosynthetic efficiency. *The Plant Journal*, *97*(1), 112–133. https://doi.org/10.1111/tpj.14190

van Dijk, A. D. J., Kootstra, G., Kruijer, W., & de Ridder, D. (2021). Machine learning in plant science and plant breeding. *iScience*, *24*(1), 101890. https://doi.org/10.1016/j.isci.2020.101890

Varshney, R. K. (2016). Exciting journey of 10 years from genomes to fields and markets: Some success stories of genomics-assisted breeding in chickpea, pigeonpea and groundnut. *Plant Science*, *242*, 98–107. https://doi.org/10.1016/j.plantsci.2015.09.009

Varshney, R. K., Pandey, M. K., Bohra, A., Singh, V. K., Thudi, M., & Saxena, R. K. (2019). Toward the sequence-based breeding in legumes in the post-genome sequencing era. *Theoretical and Applied Genetics*, *132*(3), 797–816.

Wang, S., Li, L., Li, H., Sahu, S. K., Wang, H., Xu, Y., Xian, W., Song, B., Liang, H., Cheng, S., Chang, Y., Song, Y., Çebi, Z., Wittek, S., Reder, T., Peterson, M., Yang, H., Wang, J., Melkonian, B., . . . Liu, X. (2019). Genomes of early-diverging streptophyte algae shed light on plant terrestrialization. *Nature Plants*, *6*(2), 95–106. https://doi.org/10.1038/s41477-019-0560-3

Wanga, M. A., Shimelis, H., Mashilo, J., & Laing, M. D. (2021). Opportunities and challenges of speed breeding: A review. *Plant Breeding*, *140*(2), 185–194. https://doi.org/10.1111/pbr.12909

Wartha, C. A., & Lorenz, A. J. (2021). Implementation of genomic selection in public-sector plant breeding programs: Current status and opportunities. *Crop Breeding and Applied Biotechnology*, *21*(S), e394621S15. https://doi.org/10.1590/1984-70332021v21Sa21

Watson, A., Ghosh, S., Williams, M. J., Cuddy, W. S., Simmonds, J., Rey, M. D., & Hickey, L. T. (2018). Speed breeding is a powerful tool to accelerate crop research and breeding. *Nature Plants*, *4*(1), 23–29. https://doi.org/10.1038/s41477-017-0083-8

Xue, Y., & Acar, M. (2018). Live-cell imaging of chromatin condensation dynamics by CRISPR. *iScience*, *4*, 216–235. https://doi.org/10.1016/j.isci.2018.06.001

Yamano, T., Nishimasu, H., Zetsche, B., Hirano, H., Slaymaker, I. M., Li, Y., Fedorova, I., Nakane, T., Makarova, K. S., Koonin, E. V., Ishitani, R., Zhang, F., & Nureki, O. (2016). Crystal Structure of Cpf1 in Complex with Guide RNA and Target DNA. *Cell*, *165*(4), 949–962. https://doi.org/10.1016/j.cell.2016.04.003

Zabel, F., Müller, C., Elliott, J., Minoli, S., Jägermeyr, J., Schneider, J. M., & Asseng, S. (2021). Large potential for crop production adaptation depends on available future varieties. *Global Change Biology*, *27*(16), 3870–3882. https://doi.org/10.1111/gcb.15649

Zetsche, B., Gootenberg, J. S., Abudayyeh, O. O., Slaymaker, I. M., Makarova, K. S., Essletzbichler, P., & Zhang, F. (2015). Cpf1 is a single RNA-guided endonuclease of a class 2 CRISPR-Cas system. *Cell*, *163*(3), 759–771. https://doi.org/10.1016/j.cell.2015.09.038

Zhong, Z., Sretenovic, S., Ren, Q., Yang, L., Bao, Y., Qi, C., Yuan, M., He, Y., Liu, S., Liu, X., Wang, J., Huang, L., Wang, Y., Baby, D., Wang, D., Zhang, T., Qi, Y., & Zhang, Y. (2019). Improving Plant Genome Editing with High-Fidelity xCas9 and Non-canonical PAM-Targeting Cas9-NG. *Molecular Plant*, *12*(7), 1027–1036. https://doi.org/10.1016/j.molp.2019.03.011

Zhong, Z., Zhang, Y., You, Q., Tang, X., Ren, Q., Liu, S., Yang, L., Wang, Y., Liu, X., Liu, B., Zhang, T., Zheng, X., Le, Y., Zhang, Y., & Qi, Y. (2018). Plant genome editing using FNCPF1 and LBCPF1 nucleases at redefined and altered PAM sites. *Molecular Plant*, *11*(7), 999–1002. https://doi.org/10.1016/j.molp.2018.03.008

8 Integrating Biotechnology and Precision Agriculture for Plant Disease Management

Deepali Mohapatra, Lipikant Sahoo,
Samikshya Sankalini Pradhan, and Biswajit Jena

8.1 INTRODUCTION

Increasing incidence and severity of plant diseases outbreaks have an impact on both the quality and quantity of agricultural products and pose significant risk to the global food safety and food security. These consequences include a loss of income in agricultural sectors, which are particularly devastating for developing countries. Plant diseases cause 10–16% losses in global harvests each year, costing an estimated $220 billion. In addition to lowering crop yields, plant diseases also lower the quality and value of agricultural goods, raise labor and input costs, and have an impact on farmers' livelihoods and income. Plant diseases are of global concern causing substantial damage to food crop yields and undermining the socio-political stability of countries. There are many classic examples, including Irish famine in 1845 due to *Phytophthora infestans*, Bengal famine in 1943 due to *Cochliobolus miyabeanus*, coffee rust epidemics caused by *Hemileia vastatrix* in Central America (Agrios, 2005). Apart from post-harvest disease, losses caused by pathogenic microbes such as *Penicillium* spp. further aggravate the situation. Climate change increases the danger of plant disease outbreaks (Shamim et al., 2013).

Climate change has a considerable impact on plant pathogen dynamics, notably through variations in agrometeorological indexes. Climate change modulates the pathogen virulence, influences the pathogen evolution, affects host-pathogen interaction, and emerges new aggressive strains of pathogen. With the world's population growing at a rapid pace and agricultural productivity declining, food supply is under severe strain, and the problem of plant diseases has grown in prominence, posing a significant threat to global food security. Management of plant disease are generally being done by chemicals like fungicides, nematicides, and antibiotics which have negative environmental impact, health risks to humans and wildlife and have higher chances of resistance development in the pathogen. Farmers are advised to combine

DOI: 10.1201/9781003597797-8

various plant disease control techniques into a single strategy known as integrated plant disease management (IPDM). Crop rotation, disease-free planting materials, field sanitation, methods involving chemicals, and the use of resistant varieties are all examples of such strategies (Yang, 2020).

However, a number of these technologies are cost-prohibitive and dramatically increase production costs. Plant disease control benefits from advances in molecular biology and biotechnology. This includes detection, diagnosis, and control methods such as gene transfer, mutation breeding, RNA interference (RNAi), and genome editing (Shamim et al., 2013). Precision defense of plants is a non-destructive approach to plant disease management that is based on stochastic variability. The Global Navigation Satellite System (GNSS) and geographic information system (GIS) allow the assessment of field heterogeneity caused by disease concerns, as well as site-specific treatments. Crop diseases have long been detected and mapped using remote sensing technologies. Similarly, hyperspectral remote sensing is an advanced spectral technique for detecting plant diseases (Yang, 2020). The primary goal of precision plant protection is to drastically reduce the inappropriate use of chemical inputs, and hence the negative impact of chemicals on the environment. Optimistic plant protection requires new and innovative ways to handle upcoming issues and patterns in agricultural production that require extra accuracy (Balasundram et al., 2020). Therefore, to improve disease detection, new automated approaches with high sensitivity, specificity, and reliability are required. The integration of biotechnology and precision agriculture presents a promising frontier in plant disease management.

8.2 BIOTECHNOLOGY APPROACHES FOR PLANT DISEASE MANAGEMENT

Conventional breeding plays a pivotal role in crop improvement, but it is inherently labor-intensive and time-consuming, as it necessitates the cultivation and evaluation of large crop populations over multiple generations. In contrast, genetic engineering defined as the precise modification of an organism's genetic material through biotechnological tools, that offers significant advantages over traditional breeding approaches (Collinge, 2018). Genetic engineering enables precise gene manipulation—insertion, deletion, or modification—with minimal genomic disruption, accelerating trait development compared to conventional breeding. It also expands genetic resources by allowing interspecies gene transfer and introduces novel traits into vegetatively propagated crops, overcoming limitations of traditional methods (Collinge, 2018). Plants evolved intricate defensive mechanisms against various pathogens (Jones & Dangl, 2006). Establish physical and physiochemical barriers, together with supplementary protection, to avert possible pathogens from infiltrating the cell (Uma et al., 2011). Immune receptors located on the plasma membrane and inside cells activate defensive mechanisms upon detecting pathogens, either via direct contact with pathogen-derived antigens or by observing pathogen-induced alterations in host targets (Jones & Dangl, 2006). Antimicrobial peptides and similar compounds derived from plants may diminish pathogenicity by directly detoxifying or by reducing the activity of virulence factors. Plants use RNAi to detect viral infections (Rosa et al., 2018).

8.2.1 GENETIC ENGINEERING FOR DISEASE RESISTANCE

Pathogen-detecting proteins are encoded by resistance genes (R genes). For many years, resistance breeding projects have employed R genes, with differing degrees of success. Exciting new insights from recent molecular studies of R genes and its protein and downstream signal transduction cascades will improve the application of resistance genes for disease control (Verma et al., 2022). The discovery of conserved structural motifs in R genes has made it easier to clone beneficial R genes, some of which are resistant to a broad variety of diseases and/or functional in various crop species (Ali et al., 2022). Many of the defense network's signal transduction components have been identified, and some of them are being used as switches to activate resistance against various diseases (Meng & Zhang, 2013). For disease control, R gene-mediated resistance offers a number of appealing advantages. When triggered at the right time, the coordinated reactions can effectively stop the spread of the pathogen with little harm to the plant. There are no undesirable environmental repercussions and the farmer does not need to provide any inputs. Unfortunately, co-evolving host-pathogens frequently swiftly beat R genes (Pink, 2002).

Many R genes do not offer broad-spectrum resistance because they can only identify a small number of pathogen strains. Nevertheless, new molecular-level understandings of downstream signal transduction pathways and R protein function may offer solutions to address these shortcomings (Dong & Ronald, 2019). Durability is still an issue even if it is now simpler to find and apply beneficial R genes. Because a single point mutation in the appropriate Avr gene leads to loss of function, can overcome many R genes, making the pathogen "invisible," they lack endurance. Pathogens can offer to change or eliminate an Avirulence gene with little to no fitness since individual Avr genes frequently only contribute incrementally to virulence (Leach et al., 2001). R genes have been employed "one at a time" in agricultural monocultures as part of traditional breeding techniques. These homogeneous host populations become highly susceptible to the emergent pathogen after exerting durable selection pressure for mutation of the relevant Avirulence gene. Rather than deploying a single gene, multiple R genes (also known as "pyramids") can be developed into various plant lines (Pink, 2002). Due to the time needed to breed hodgepodges of R genes into best cultivars, multiline deployment and pyramiding have not been used extensively. But once the previously mentioned methods are refined, these tactics will become far more useful (Dong & Ronald, 2019).

The phenomenon known as "restricted taxonomic functionality" (RTF) presents challenges for transferring R genes from prototype organism to crop plants or between far related crop species. For instance, in the Solanaceae family such as tobacco, potato, and chilli transgenes like *Bs2* and several R genes from tomatoes can function effectively. However, the Arabidopsis RPS2 resistance gene and *Bs2* do not exhibit functionality in tomato (Hulbert et al., 2001; Zhu et al., 2000). Although the molecular basis of RTF is unrecorded, it may result from R proteins' inability to react with signal transduction factors that have evolved in the transgenic host. It is uncertain in the fact that RTF is universal property of R genes or not. Recent studies suggest that transferring specific R genes across distantly related species might be achievable. For example, in plant *Arabidopsis thaliana*, R gene *RPW8* confers general purpose resistance against powdery mildew in both *Arabidopsis* and tobacco. Advancing our understanding of R gene signaling could potentially resolve the RTF challenge (Ellis et al., 2000; Xiao et al., 2003).

8.2.2 RNA INTERFERENCE (RNAi) TECHNOLOGY

Transgenic plants expressing viral genes often develop immunity against the virus and related strains, as aberrant viral protein expression disrupts the viral life cycle. This resistance is primarily mediated by RNAi, which exploits viruses' dependence on host cellular machinery, making it a potent antiviral defense strategy (Lindbo & Falk, 2017). Most plant viruses possess ssRNA as their genetic makeup. During genome replication by RNA-dependent RNA polymerase (RdRP), double-stranded RNA (dsRNA) intermediates are often formed, triggering the host's RNAi response. This approach has been successfully applied in developing transgenic crops such as papaya (*Carica papaya*) and squash (*Cucurbita* species), which have been commercially grown in the United States for more than two decades. Fuchs and Gonsalves (2007) highlight that RNAi-mediated strategies provide highly durable disease resistance. By altering specific nucleotides base pairs in miRNA-encoding genes, the miRNA machinery can be tailored to target RNA viruses with greater specificity (Ambros, 2001; Reinhart et al., 2002; Xie et al., 2015).

8.2.3 CRISPR-CAS9 GENOME EDITING

CRISPR-Cas system leverages clustered regularly interspaced short palindromic repeats (CRISPR) derived from bacteria. In several bacterial species, CRISPR-Cas functions as an antiviral defense mechanism. Within this system, RNA-guided nucleases, such as Cas proteins, destroy viral DNA or RNA by targeting and cleaving specific sites on these substrates. The precision of the cleavage depends on the complementary base pairing between the CRISPR-RNA and the target DNA (tDNA) or RNA sequences. Numerous Cas proteins have nuclease activity which is sequence-specific (Wu et al., 2018). Such as, RNA-guided RNases, like Cas13a from bacteria *Leptotrichia shahii* (LshCas13a) or *Leptotrichia wadei* (LwaCas13a) specifically cleave RNA in a living system. Meanwhile, the RNA-guided endonuclease Cas9 from another bacteria *Streptococcus pyogenes* (SpCas9) induces double-stranded breaks in DNA. Additionally, in vivo studies have shown that Cas9 from *Francisella novicida* (FnCas9) can cleave both genetic materials i.e., DNA and RNA (Abudayyeh et al., 2017; Jinek et al., 2012). Additionally, Cas12a, also known as Cpf1, is believed to lower the risk of viral escape due to its lower likelihood of mutations disrupting recognition by the original guide RNA (Ali et al., 2016).

8.2.4 PLANT-BASED VACCINES AND IMMUNIZATION STRATEGIES

Vaccines stimulate antibody production in humans and animals, providing immunological protection against various diseases (Price et al., 2015). In 1989, Hiatt and colleagues pioneered the attempt to produce vaccines in plants. Subsequently, Dr. Arntzen and his team introduced the innovative idea of utilizing transgenic plants for the synthesis and delivery of subunit vaccines, demonstrating enhanced efficacy compared to orthodox vaccine production methods (Laere et al., 2016). The first plant-based vaccine was successfully developed in tobacco plants by producing surface protein antigens from *Streptococcus mutans* (Laere et al., 2016). To date, numerous bioengineered plants have been employed to produce vaccines in four main categories: bacterial, viral, parasitic, and immunological contraceptive vaccines (Laere et al., 2016). The production of vaccines using plants primarily involves incorporating foreign genes into plant cells. Before introducing the vector into the expression system, the desired antigen's target sequence is integrated with it. A reliable transformation mechanism may be achieved through nuclear or plastid incorporation. The terms "stable" or "permanent" are used because the recipient cells' genetic composition undergoes lasting changes when the target foreign gene is inserted into the host plant cells' genome (Ma & Chen, 2005). Although plant-based vaccine production has been ongoing for approximately two decades since 1989, several challenges still need to be addressed before these vaccines can become highly effective. These obstacles include the selection of antigens and plant expression hosts, maintaining consistent dosages, implementing good manufacturing practice (GMP) procedures in vaccine production, and various other issues. The problems that require resolution may span from initial upstream processes to the final stages of vaccine implementation (Laere et al., 2016).

8.2.5 MICROBIAL BIOTECHNOLOGY FOR BIOCONTROL AGENTS

The integration of biotechnology with conventional farming methods will play an imperative role in shaping the future of sustainable agriculture. According to Fravel (2005), biological control of plant disease involves the use of non-harmful microbes, such as *Trichoderma* sp., yeast, or *Pseudomonas*, to attack and control plant pathogens and the diseases they occur. Various techniques can be employed for the biological management of plant diseases, including antibiosis, competition, parasitism, induced resistance, hypovirulence, suppression, and predation (Yadav et al., 2020). The process of examining biocontrol mechanisms using molecular genetic techniques involves several steps: isolating genes that code for potential biocontrol agents, creating specific mutants of the antagonist lacking these genes through disruption, comparing the reduced biocontrol effectiveness of these mutants to their original genetic makeup, restoring the ability to generate the agent by reinstating the intact gene sequence through transformation (Yadav et al., 2020).

This methodology offers the most definite indication for a specific gene role in biological control (Rey et al., 2000). Genetically altered antagonistic hyperparasitic bacteria demonstrate superior effectiveness as biological control agents. Various techniques can be employed to enhance genetics, including chemical and physical mutations, sexual hybridization, homokaryon development, and genetic manipulation methods such as mutagenic agent, genetic analysis of fusants, recombination, protoplast fusion, transformation, or the extraction of beneficial genes from biofungi lacking functional sexual stages (Mohamed et al., 2004). Protoplast fusion stands out as a simple yet effective approach for amalgamating the valuable traits of multiple promising strains. By merging protoplasts from two effective biocontrol strains of *Trichoderma harzianum*, researchers produced an offspring strain with significantly improved biocontrol efficacy (Monte, 2001). Researchers genetically engineered *Pseudomonas fluorescens* F113Rif (pCU8.3) and *P. fluorescens* F113Rif (pCUP9) strains to boost phenazine-1-carboxylate (Phl) production. These modified strains were then evaluated for their biocontrol effectiveness on *Beta* sp. in microcosm experiments (Resca et al., 2001).

8.3 PRECISION AGRICULTURE FOR PLANT DISEASE MONITORING AND MANAGEMENT

Application maps may be created to oversee agricultural fields by using data from diverse sensors and geographic information systems (GIS), including the spatiotemporal variabilities of diseases. The targeted use of pesticides, consistent with precision agriculture methodologies, may reduce pesticide consumption, thus lowering economic expenses and environmental effects in agro-ecosystem and its productivity.

8.3.1 Remote Sensing and Aerial Imaging for Disease Detection

Remote sensing and aerial imaging have revolutionized the diagnosis and control of plant diseases, offering significant advantages in terms of early detection, accuracy, and scalability. Remote sensing technologies include multispectral, hyperspectral, thermal, and LiDAR imaging, which allow the identification of changes in light reflectance, temperature, and vegetation structural attributes (Palumbo et al., 2005). Aerial platforms, particularly unmanned aerial vehicles (UAVs), are widely used for plant disease diagnostics owing to their ability to operate at low altitudes and capture high-resolution photos over large regions, making them ideal for precision agriculture (Palumbo et al., 2005). The combination of remote sensing data with sophisticated algorithms and machine learning (ML) methodologies has facilitated the creation of accurate disease detection systems that can discern early signs of plant stress, pathogen encroachment, and nutrient inadequacies, which are crucial for effective disease management and mitigation (Table 8.1).

Hyperspectral sensors, which capture data across numerous narrow electromagnetic bands, have proven effective in detecting plant diseases like tomato leaf spot and late blight by identifying subtle spectral variations, with notable applications, including early detection of bacterial leaf blight in rice. When combined with multispectral imaging, which uses broader spectral bands. These techniques enhance disease monitoring, as demonstrated in vineyards where multispectral data and vegetation indices like NDVI successfully tracked downy mildew. UAVs equipped with cameras, sensors, and thermal imaging capabilities

TABLE 8.1
Types of Remote Sensing Systems Used to Monitor Plant Diseases

Types of Remote Sensing	Attributes	Benefits	Drawbacks	References
SAR (Synthetic Aperture RADAR) and LiDar (Light Detection and Ranging)	Detect disease-induced structural or morphological changes.	Capable of indicating structural plant changes.	There are limited systems and case studies available right now.	Agrawal and Khairnar (2019)
Fluorescence and Thermal	Identify physiological changes before symptoms arise.	Have the potential to recognize symptoms before they appear.	However, it is currently challenging to apply on vast areas.	Zarco-Tejada et al. (2012)
VIS-SWIR (Visible and Short-Wave Infrared)	Detect plant disease using VIS-SWIR reflectance.	Stable, providing consistent monitoring outcomes.	However, it performs badly in preliminary identification.	Zhang et al. (2020)

are increasingly deployed to detect physiological stress in plants, leveraging near-infrared spectra and temperature anomalies to diagnose diseases such as powdery mildew in wheat, where infected plants exhibit distinct thermal profiles. These remote sensing technologies offer scalable solutions for large-scale agriculture, enabling precise disease identification and management where manual monitoring is impractical (Palumbo et al., 2005).

8.3.2 GEOGRAPHIC INFORMATION SYSTEMS (GIS) AND SPATIAL ANALYSIS

GIS and spatial analysis are powerful tools for capturing, managing, analyzing, and visualizing geographic data, offering critical insights in fields such as environmental management, urban planning, and public health. By integrating geographical data from satellite imagery, GPS, and remote sensing with demographic and economic data, GIS enables the creation of detailed maps and spatial models to study complex location-based phenomena. A key strength of GIS is its ability to identify spatial patterns and relationships, aiding in understanding disease spread, land use changes, and natural resource management. Techniques such as overlay analysis, buffer analysis, and spatial interpolation help uncover trends, predict outcomes, and support informed decision-making (Palumbo et al., 2005).

In public health, GIS has been instrumental in tracking disease outbreaks like COVID-19, with Bragazzi et al. demonstrating its effectiveness in mapping infection hotspots in Italy to guide containment strategies. Environmental applications include monitoring deforestation, climate change impacts, and wildlife movements, as well as assessing risks from natural disasters like floods and wildfires. Urban planners use GIS to model land use, predict urban expansion, and optimize infrastructure, particularly in smart cities where it improves traffic flow, public transit, and utility management. Additionally, GIS supports precision agriculture by analyzing soil conditions, crop health, and irrigation needs, enhancing productivity while minimizing environmental harm. A case study by Mulla highlighted how GIS optimizes pesticide use in U.S. agriculture, reducing costs and ecological damage (Palumbo et al., 2005).

8.3.3 AUTONOMOUS FARMING SYSTEMS AND ROBOTICS

Autonomous agricultural systems and robots are transformative innovations that enhance farming efficiency, sustainability, and productivity by automating labor-intensive tasks such as planting, weeding, and harvesting. These systems, including robotic harvesters, drones, and autonomous tractors, leverage sensors, GPS, and artificial intelligence (AI) to perform precise operations while reducing human intervention and labor costs. A key application is precision farming, where autonomous machines optimize irrigation, fertilization, and pest control by analyzing real-time soil and crop data. Autonomous tractors, for example, improve operational efficiency by minimizing fuel consumption and soil compaction through GPS-guided navigation. Drones play a crucial role in crop monitoring, using multispectral imaging to detect diseases, pests, and nutrient deficiencies early, enabling targeted interventions that reduce chemical usage (Zhang et al., 2020). A case study in China demonstrated how drone-collected data helped farmers optimize irrigation and pesticide application, increasing yields while lowering environmental harm (Zhang et al., 2020). These technologies also contribute to environmental sustainability by reducing carbon emissions through optimized fuel use and precision resource application, minimizing runoff and soil degradation. However, challenges such as high initial costs, technical complexity, and the need for skilled operators limit widespread adoption, particularly among small-scale farmers.

8.3.4 DATA ANALYTICS AND MACHINE LEARNING FOR DISEASE PREDICTION

Data analytics and ML have revolutionized plant disease prediction and management by enabling early detection and precise interventions through advanced data processing techniques (Zhang et al., 2020). These technologies analyze vast datasets from remote sensing, field sensors, and historical records to identify disease patterns and forecast outbreaks with high accuracy. ML algorithms, particularly deep learning models like convolutional neural networks (CNNs), have proven effective in diagnosing diseases from leaf images, which achieved high accuracy in identifying crop diseases in maize and rice. The integration of satellite and drone-based remote sensing with ML enhances disease monitoring, such as early blight detection in potatoes, by using multispectral data to assess crop health. Predictive models also leverage historical weather data and regional disease trends to anticipate outbreaks, which ML system successfully forecasted wheat rust in India, enabling timely fungicide applications. Additionally, ML models can optimize farming decisions by correlating environmental factors with disease susceptibility, as seen in grapevine powdery mildew prediction, where climatic data guided pesticide scheduling to minimize waste (Boulent et al., 2020). The Internet of Things (IoT) further refines these predictions by providing real-time field data via soil and weather sensors, which ML models use to generate dynamic disease risk maps for precision treatments (Boulent et al., 2020). Despite their potential, challenges such as the need for high-quality training data, environmental variability, and farmer accessibility limit widespread adoption. Moreover, while ML models perform well in controlled condition, real-world deployment faces hurdles like data reliability and farmer technical literacy (Boulent et al., 2020). Nevertheless, these technologies represent a transformative shift toward sustainable agriculture by reducing chemical use and improving crop resilience through data-driven disease management.

8.3.5 Challenges and Future Directions

The merger of biotechnology and precision agriculture represents a major breakthrough in plant disease management, offering sustainable solutions to enhance crop health while reducing environmental and economic costs. However, realizing its full potential requires overcoming key challenges such as regulatory adaptability, public perception, and equitable access. Regulatory bodies like Genetic Engineering Appraisal Committee (GEAC) and the Government of India must strike a balance between fostering innovation and ensuring safety through clear and adaptive policies. Public concerns over genetically modified organisms (GMOs), environmental impact, and food safety necessitate transparent communication, stakeholder engagement, and education to build trust (Aggarwal et al., 2024). Additionally, cost-effectiveness and scalability remain critical factors, as high-tech solutions are often confined to large commercial farms. To enable widespread adoption, especially among smallholder farmers who contribute significantly to global food production, technological advancements must be made more affordable and adaptable (Abdullahi et al., 2015; Zhang et al., 2021).

8.4 CONCLUSION

The integration of biotechnology and precision agriculture offers a transformative approach to plant disease management, combining genetic advancements with data-driven technologies to enhance crop resilience and productivity. Biotechnology enables the development of disease-resistant crop varieties, while precision agriculture leverages tools like remote sensing, IoT, and ML for early disease detection and targeted interventions. Together, these innovations reduce reliance on chemical treatments, minimize environmental impact, and optimize resource use. However, challenges such as high implementation costs, technical complexity, and farmer adoption must be addressed to ensure widespread accessibility. Ultimately, this synergy holds great potential for sustainable agriculture, improving food security while mitigating the effects of plant diseases in a changing climate.

REFERENCES

Abdullahi, H. S., Mahieddine, F., & Sheriff, R. E. (2015). Technology impact on agricultural productivity: A review of precision agriculture using unmanned aerial vehicles. In *Wireless and satellite systems: 7th international conference, WiSATS 2015, Bradford, UK, July 6-7, 2015. Revised selected papers 7* (pp. 388–400). Springer International Publishing. https://doi.org/10.1007/978-3-319-25479-1_29

Abudayyeh, O. O., Gootenberg, J. S., Essletzbichler, P., Han, S., Joung, J., Belanto, J. J., & Zhang, F. (2017). RNA targeting with CRISPR–Cas13. *Nature*, *550*(7675), 280–284. https://doi.org/10.1038/nature24049

Aggarwal, A., Komal, D. S. G. S., & Chaudhary, R. (2024). A critical analysis of legal boundaries in biotechnology and biosecurity. *MSW Management Journal*, *34*(1), 239–249.

Agrawal, S., & Khairnar, G. B. (2019). A comparative assessment of remote sensing imaging techniques: Optical, SAR and LiDAR. *The International Archives of the Photogrammetry, Remote Sensing and Spatial Information Sciences*, *42*, 1–6. https://doi.org/10.5194/isprs-archives-XLII-3-W6-1-2019

Agrios, G. N. (2005). *Plant pathology* (5th ed.). Elsevier Academic Press.

Ali, Q., Yu, C., Hussain, A., Ali, M., Ahmar, S., Sohail, M. A., & Zhou, L. (2022). Genome engineering technology for durable disease resistance: Recent progress and future outlooks for sustainable agriculture. *Frontiers in Plant Science*, *13*, 860281. https://doi.org/10.3389/fpls.2022.860281

Ali, Z., Ali, S., Tashkandi, M., Zaidi, S. S., & Mahfouz, M. M. (2016). CRISPR/Cas9-mediated immunity to geminiviruses: Differential interference and evasion. *Scientific Reports*, *6*, 26912. https://doi.org/10.1038/srep26912

Ambros, V. (2001). microRNAs: Tiny regulators with great potential. *Cell*, *107*(7), 823–826. https://doi.org/10.1016/S0092-8674(01)00616-X

Balasundram, S. K., Golhani, K., Shamshiri, R. R., & Vadamalai, G. (2020). Precision agriculture technologies for management of plant diseases. In *Plant disease management strategies for sustainable agriculture through traditional and modern approaches* (pp. 259–278). Springer.

Collinge, D. B. (2018). Transgenic crops and beyond: How can biotechnology contribute to the sustainable control of plant diseases? *European Journal of Plant Pathology*, *152*(4), 977–986. https://doi.org/10.1007/s10658-018-1572-y

Dong, O. X., & Ronald, P. C. (2019). Genetic engineering for disease resistance in plants: Recent progress and future perspectives. *Plant Physiology*, *180*(1), 26–38. https://doi.org/10.1104/pp.18.01224

Ellis, J., Dodds, P., & Pryor, T. (2000). Structure, function and evolution of plant disease resistance genes. *Current Opinion in Plant Biology*, *3*(4), 278–284. https://doi.org/10.1016/S1369-5266(00)00080-7

Fravel, D. (2005). Commercialization and implementation of biocontrol. *Annual Review of Phytopathology*, *43*, 337–359. https://doi.org/10.1146/annurev.phyto.43.032904.092924

Fuchs, M., & Gonsalves, D. (2007). Safety of virus-resistant transgenic plants two decades after their introduction: Lessons from realistic field risk assessment studies. *Annual Review of Phytopathology*, *45*, 173–202. https://doi.org/10.1146/annurev.phyto.45.062806.094434

Hulbert, S. H., Webb, C. A., Smith, S. M., & Sun, Q. (2001). Resistance gene complexes: Evolution and utilization. *Annual Review of Phytopathology*, *39*(1), 285–312. https://doi.org/10.1146/annurev.phyto.39.1.285

Jinek, M., Chylinski, K., Fonfara, I., Hauer, M., Doudna, J. A., & Charpentier, E. (2012). A programmable dual-RNA-guided DNA endonuclease in adaptive bacterial immunity. *Science*, *337*(6096), 816–821. https://doi.org/10.1126/science.1225829

Jones, J. D., & Dangl, J. L. (2006). The plant immune system. *Nature*, *444*(7117), 323–329. https://doi.org/10.1038/nature05286

Laere, E., Ling, A. P. K., Wong, Y. P., Koh, R. Y., Mohd Lila, M. A., & Hussein, S. (2016). Plant-based vaccines: Production and challenges. *Journal of Botany*, *2016*, 4928637. https://doi.org/10.1155/2016/4928637

Leach, J. E., Vera Cruz, C. M., Bai, J., & Leung, H. (2001). Pathogen fitness penalty as a predictor of durability of disease resistance genes. *Annual Review of Phytopathology*, *39*(1), 187–224. https://doi.org/10.1146/annurev.phyto.39.1.187

Lindbo, J. A., & Falk, B. W. (2017). The impact of "coat protein-mediated virus resistance" in applied plant pathology and basic research. *Phytopathology*, *107*(6), 624–634. https://doi.org/10.1094/PHYTO-12-16-0442-RVW

Ma, H., & Chen, G. (2005). Gene transfer technique. *Nature and Science*, *3*(1), 25–31.

Meng, X., & Zhang, S. (2013). MAPK cascades in plant disease resistance signaling. *Annual Review of Phytopathology*, *51*(1), 245–266. https://doi.org/10.1146/annurev-phyto-082712-102314

Mohamed, H. A. A., Haggag, W. M., & Abo-Aba, S. M. (2004). Influence of salt stress on *Pseudomonas fluorescens* plasmids, some phenotypic traits and antibiosis against *Diplodia theobromae*. *Journal of Genetic Engineering and Biotechnology*, *2*(2), 265–281.

Monte, E. (2001). Understanding *Trichoderma*: Between biotechnology and microbial ecology. *International Microbiology*, *4*(1), 1–4.

Palumbo, J. D., Yuen, G. Y., Jochum, C. C., Tatum, K., & Kobayashi, D. Y. (2005). Mutagenesis of β-1,3-glucanase genes in *Lysobacter enzymogenes* strain C3 results in reduced biological control activity toward *Bipolaris* leaf spot of tall fescue and *Pythium* damping-off of sugar beet. *Phytopathology*, *95*(6), 701–707. https://doi.org/10.1094/PHYTO-95-0701

Pink, D. A. C. (2002). Strategies using genes for non-durable resistance. *Euphytica*, *124*(2), 227–236. https://doi.org/10.1023/A:1015638718242

Price, A. A., Sampson, T. R., Ratner, H. K., Grakoui, A., & Weiss, D. S. (2015). Cas9-mediated targeting of viral RNA in eukaryotic cells. *Proceedings of the National Academy of Sciences*, *112*(19), 6164–6169. https://doi.org/10.1073/pnas.1422340112

Reinhart, B. J., Weinstein, E. G., Rhoades, M. W., Bartel, B., & Bartel, D. P. (2002). MicroRNAs in plants. *Genes and Development*, *16*(13), 1616–1626. https://doi.org/10.1101/gad.1004402

Resca, R., Basaglia, M., Poggiolini, S., Vian, P., Bardin, S., Walsh, U. F., Enriquez Barreiros, C. M., O'Gara, F., Nuti, M. P., Casella, S., & Peruch, U. (2001). An integrated approach for the evaluation of biological control of the complex *Polymyxa betae/Beet Necrotic Yellow Vein Virus*, by means of seed inoculants. *Plant and Soil*, *232*(1–2), 215–226.

Rey, M., Delgado, J., Rincon, A., Carmen, L., Benitez, T., Perez, E., & Cantoral, F. (2000). Improvement of *Trichoderma* strains for biocontrol. *Revista Iberoamericana de Micologia*, *17*(1), 31–36.

Rosa, C., Kuo, Y. W., Wuriyanghan, H., & Falk, B. W. (2018). RNA interference mechanisms and applications in plant pathology. *Annual Review of Phytopathology*, *56*(1), 581–610. https://doi.org/10.1146/annurev-phyto-080417-050044

Shamim, M., Pandey, P., Singh, A., Yadav, P., Bhowmick, P. K., Srivastava, D., & Singh, K. N. (2013). Role of biotechnology in plant diseases management: An overview. *Journal of Genetic and Environmental Resources Conservation*, *1*(3), 215–221.

Uma, B., Swaroopa Rani, T., & Podile, A. R. (2011). Warriors at the gate that never sleep: Non-host resistance in plants. *Journal of Plant Physiology*, *168*(18), 2141–2152. https://doi.org/10.1016/j.jplph.2011.09.005

Verma, N. V., Shukla, M., Kulkarni, R., Srivastava, K., Claudic, B., Savara, J., & Pandya, A. (2022). Emerging extraction and diagnostic tools for detection of plant pathogens: Recent trends, challenges, and future scope. *ACS Agricultural Science and Technology*, *2*(5), 858–881. https://doi.org/10.1021/acsagscitech.2c00150

Wu, W. Y., Lebbink, J. H. G., Kanaar, R., Geijsen, N., & van der Oost, J. (2018). Genome editing by natural and engineered CRISPR-associated nucleases. *Nature Chemical Biology*, *14*(7), 642–651. https://doi.org/10.1038/s41589-018-0089-1

Xiao, S., Charoenwattana, P., Holcombe, L., & Turner, J. G. (2003). The *Arabidopsis* genes *RPW8.1* and *RPW8.2* confer induced resistance to powdery mildew diseases in tobacco. *Molecular Plant-Microbe Interactions*, *16*(4), 289–294. https://doi.org/10.1094/MPMI.2003.16.4.289

Xie, M., Zhang, S., & Yu, B. (2015). microRNA biogenesis, degradation and activity in plants. *Cellular and Molecular Life Sciences*, *72*(1), 87–99. https://doi.org/10.1007/s00018-014-1728-7

Yadav, A. N., Kour, D., Kaur, T., Devi, R., Guleria, G., Rana, K. L., & Rastegari, A. A. (2020). Microbial biotechnology for sustainable agriculture: Current research and future challenges. In *New and future developments in microbial biotechnology and bioengineering* (pp. 331–344). Elsevier.

Yang, C. (2020). Remote sensing and precision agriculture technologies for crop disease detection and management with a practical application example. *Engineering*, *6*(5), 528–532. https://doi.org/10.1016/j.eng.2020.03.008

Zarco-Tejada, P. J., González-Dugo, V., & Berni, J. A. (2012). Fluorescence, temperature and narrow-band indices acquired from a UAV platform for water stress detection using a micro-hyperspectral imager and a thermal camera. *Remote Sensing of Environment*, *117*, 322–337. https://doi.org/10.1016/j.rse.2011.10.007

Zhang, N., Yang, G., Pan, Y., Yang, X., Chen, L., & Zhao, C. (2020). A review of advanced technologies and development for hyperspectral-based plant disease detection in the past three decades. *Remote Sensing*, *12*(19), 3188. https://doi.org/10.3390/rs12193188

Zhang, P., Guo, Z., Ullah, S., Melagraki, G., Afantitis, A., & Lynch, I. (2021). Nanotechnology and artificial intelligence to enable sustainable and precision agriculture. *Nature Plants*, *7*(7), 864–876. https://doi.org/10.1038/s41477-021-00946-6

Zhu, Y., Chen, H., Fan, J., Wang, Y., Li, Y., Chen, J., Fan, J., Yang, S., Hu, L., Leung, H., & Mew, T. W. (2000). Genetic diversity and disease control in rice. *Nature*, *406*(6797), 718–722. https://doi.org/10.1038/35021046

9 Addressing Global Nutritional Security Through Crop Biofortification

Prasann Kumar and Shipa Rani Dey

9.1 INTRODUCTION

Biofortification is defined as increasing the nutritional value of food crops by increasing the vitamin and mineral contents through plant breeding and/or other methods like genetic engineering and agronomic practices. They are responsible for significant health and nutritional benefits when incorporated into diets regularly (Kumar, Saurabh, et al., 2024). Fortifying food with essential nutrients has become necessary since many individuals cannot meet their nutritional needs by varying their diets and consuming different food groups. Such an approach improves nutritional status without significant shifts in eating habits or availability of foods. As a result, biofortification via agricultural systems is an environmentally sustainable and low-cost alternative to conventional food fortification and supplementation programs, which rely on continuous infrastructure and external input (i.e. vitamin A supplements) (Dey, Kumar, et al., 2024; Dey, Sharma, et al., 2024).

Once developed and released, biofortified seeds can be multiplied and distributed to a wide area, reaching even remote areas with nutrient-rich crops. This model is especially advantageous for subsistence farmers who depend on their harvests for livelihood and nutrition. For instance, it has been shown that rural populations in Sub-Saharan Africa that consume biofortified orange-fleshed sweet potato (OFSP), which is high in provitamin A, have considerably improved vitamin A intakes and status. These steps may help enhance human capital and boost economic growth (Dey, Kumar, et al., 2024; Dey, Sharma, et al., 2024). In contrast to more conventional approaches to address malnutrition, like supplementing nutrients, plants with high yields can improve their genetic makeup through biofortification, producing the desired nutritional profiles (Saini et al., 2024). Plants' nutritional value can be improved by applying several genetic engineering techniques, including transgenic crops.

By modifying metabolic pathways, these tools make it possible to improve carbohydrates, proteins, fats, minerals, vitamins, and other valuable compounds in the body (Dey, Kumar, et al., 2024; Dey, Sharma, et al., 2024). CRISPR-based

DOI: 10.1201/9781003597797-9

gene-editing tools have emerged as new strategies for crop biofortification (Saini, Kumar, & Panwar, 2024; Saini, Kumar, & Upadhyay, 2024). Microinjection and biolistics have been applied to alter metabolic pathways in crops, including maize, for elevated levels of β-carotene and amino acids, which illustrates their capacity to dramatically improve the nutritional profile of staple crops (Kumar, 2024a).

The advent of "Omics" tools has given rise to advanced techniques for genetic manipulation (e.g., CRISPR-Cas9/Cas13 and transcription activator-like effector nucleases (TALENs)), providing an attractive set of possibilities for the biofortification of crops. Several species have also released their genome sequences, opening up new opportunities. Research on biofortification involves scientists from various disciplines, such as crop science, nutrition, food science, and social science (Kumar & Dwivedi, 2024; Sharma, Kumar, & Singh, 2024; Sharma, Kumar, & Panwar, 2024). In addition, socio-economic valuations examine how biofortified varieties are accepted among farmers, consumers, and other stakeholders in the value chain. Such evaluations provide measures of cost-effectiveness, inclusiveness, and effective delivery modes to help guide the scaling up of biofortification efforts.

9.2 BIOFORTIFICATION APPROACHES

Various methods of biofortification mainly target staple crops, such as cereals, pulses, oilseeds, fruits, and vegetables. The targeted key nutrients include magnesium (Mg), zinc, iron, carotenoids, selenium (Se), folic acid, and vitamin A, while the biofortification can be done through traditional plant breeding (involving genetic breeding), genetic engineering, molecular breeding, and agronomic practices for substantial and sustainable biofortification, promising an everlasting solution. The use of target genes for essential micronutrients is a one-time investment for the researchers and is meant to benefit future generations. In particular, molecular and genetic engineering methods provide precise, accurate, and cost-effective tools to improve the nutritional value of staple crop varieties (Kumar & Choudhury, 2024b; Kumar et al., 2024b).

9.2.1 Conventional Plant Breeding

Through years of conventional plant breeding strategies, many staple crop varieties have been developed, boasting dramatically higher levels of key micronutrients. This is an observation from research on the germplasm of traditional and brown rice that found higher concentrations of nutrients like zinc and iron in traditional lines than white or polished rice (Kumar, Devi, & Panwar, 2024; Sharma, Kumar, & Singh, 2024; Sharma, Kumar, & Panwar, 2024). It is one of several studies that justify the relevance of genetic diversity in assessing and improving micronutrient levels. One extreme example is the recent development of quality protein maize (QPM), a product of conventional breeding that farmers have readily adopted and has become a significant success story in biofortification (Kumar, Dey, & Choudhury, 2024). Genetic engineering and advanced molecular breeding techniques are the methods that researchers prefer to use to circumvent such obstacles and overcome this problem (Devi et al., 2024b; Kumar & Singh, 2024b).

9.2.2 MOLECULAR BREEDING

Typically, a molecular breeding tool selects and places desirable genes from a donor into a generally agronomically superior recipient parental line to produce a biofortified crop variety. These crops have, therefore, been provided with genomic resources that allow the mapping of the current target traits and the transfer of them into elite cultivars using marker-assisted breeding tools, which are rapidly expanding in these crops. This technology is extensively used for enhancing biofortification of cereals, pulses, millets, fruits, and vegetables (Devi et al., 2024h; Kumar & Singh, 2024b). There, the power to screen large numbers of seedlings lies, vastly accelerating the process over many generations. Unlike phenotypic selection, molecular breeding can identify recessive traits central to its utility. Moreover, identification and validation of genes or QTL related to molecular basis of metabolic pathways of mineral depositions in grains may accelerate breeding strategies aimed at concentration of bioavailable micronutrients. In this context, the marker-assisted selection (MAS) techniques can also serve as a versatile tool to produce nutritionally dense crops and combat the worldwide issue of micronutrient deficiencies.

9.2.3 GENETIC ENGINEERING

Genetic engineering permits direct introgression of wanted genes in specialised lines, thereby adjusting the uptake and utilisation of nutrients (Kumar, Singh, et al., 2024; Kumar & Singh, 2024c). With the explosion of novel genome-editing tools such as CRISPR/Cas9, zinc finger nucleases (ZFNs), and TALENs, further ground-breaking biofortification progress of cereal crops like rice and wheat and horticultural crops such as tomatoes have taken place. Biotech varieties have fruits, vegetables, pulses, etc., biofortified by CSIR, which are relatively low-cost and faster to produce (Kumar, Srivastav, et al., 2024). For example, the introduction of a single DNA fragment into rice simultaneously elevated Zn, Fe, and β-carotene levels (as depicted in a recent study). Similarly, genetically engineered sorghum enhances the bioavailability of lysine and vitamin A, iron, and zinc. However, such an approach must also consider potential trade-offs with other beneficial traits in the crop. Due to these problems, it recommends appropriate policy interventions governing the channels of micronutrient effects (Devi et al., 2024c; Kumar et al., 2024a).

9.2.4 AGRONOMIC BIOFORTIFICATION

Agronomic biofortification holds significant potential where iron and zinc deficiency diseases are prevalent (especially in Asia and Africa) and represent a quick and viable strategy for the micronutrient (zinc, iron) fortification of food grains, tailored for human nutrition (Kumar & Choudhury, 2024c; Pandey et al., 2024). Foliar application of zinc increased the zinc concentration in wheat grains, reporting an un-influencing effect on yields due to wheat decomposition. The increased Zn concentration in edible parts was attributed to the upregulation of genes involved in Zn transport and integration in plants receiving 80–100 μM Zn supplementation, which showed better root development and photosynthesis and, thus, healthy. Moreover, scientists confined the constituents in the grain of wheat highlights the multi-nutrient biofortification potential from agronomic strategies for their concentration manipulations without yield penalty (Dey, Kumar, & Panwar, 2024; Dey, Kumar, & Kumar, 2024). Biofortified plants are also grown using low-cost and eco-friendly methods of organic manuring (Devi, Kumar, et al., 2024; Kumar & Choudhury, 2024d).

9.3 ORGANISMS USED IN BIOFORTIFICATION

9.3.1 Bacteria

To grow bio-enriched crops, however, it is necessary to include microbiological agents that promote plant growth in the category of natural biofortification agents. For the biofortified approach, these agents have been the focus of a significant scholarly investigation (Kumar & Raut, 2024; Sharma & Kumar, 2024). Furthermore, fertigation with *Ochrobactrum anthropi*, *Anabaena* oscillations and *Brevundimonas diminuta* for biofortification application showed an increase of 21.2% for the rice yield concerning conventional chemical fertilisers. Also, *Pseudomonas fluorescens* increased rice plants' saponins and zinc content (Devi et al., 2024a; Singh et al., 2024). Proven strains of *Pseudomonas putida*, *Enterobacteria* sp. and other PGPR were reported to increase rice grain iron content as much as double and transport efficiency of iron from roots to stem and grains, indicating their capacity for iron biofortification (Devi et al., 2024c; Kumar, 2024b; Kumar, Saini, et al., 2024). Likewise, *Pseudomonas plecoglossicida*, a rhizospheric bacterium, enhanced iron in chickpeas and pigeon peas. Similarly, in wheat, *Arthrobacter sulfonivorans* (DS-68) and *Enterococcus hirae* (DS-163) were found to have similar effects on iron availability and *Providencia* sp. Wheat grains have also been improved with enhanced iron content with PW5 (Devi et al., 2024b). *Bacillus*, *Pseudomonas*, and *Geobacter* bacteria are essential for turning Mn^{4+} to Mn^{2+}. This conversion is beneficial for plant metabolism.

9.3.2 Fungi

Using an endophytic fungus called *Piriformospora indica*, which had an additional concentration of 10 mg/L zinc, lettuce plants with a high chlorophyll content had their zinc content significantly increased by 7.6 times. This was observed in greenhouse conditions. Consequently, this demonstrated that *P. indica* was present in biofortification (Jagadesh et al., 2024; Kumar & Choudhury, 2024c; Raj et al., 2024).

9.4 NUTRIENTS USED IN BIOFORTIFICATION

Bacteria with nitrogen-fixing symbiosis will force host plants to absorb more nitrogen, assuming the hosts' nitrogen-fixing capabilities lie in the natural action of biology. Researchers have found, identified, and reported some vigorous microorganisms that could release micro-macro elements from rhizospheric soil (Kumar & Choudhury, 2024f). However, baseline and biofortified nutrient levels in the targeted crops are indexed in Table 9.1.

TABLE 9.1
Information About the Baseline and Biofortified Nutrient Levels in the Targeted Crops

Crop	Nutrient	Baseline Levels	Levels Achieved	References
Nutritional factor				Devi et al. (2024c), Kumar (2024b), Kumar, Saini, et al. (2024)
Rice	Zinc	12.0–16.0 ppm	>20.0 ppm	
	Protein	7.0–8.0%	>10.0%	
Maize	Provitamin A	1–2 ppm	>8.0 ppm	
	Tryptophan	0.3–0.4%	>0.6%	
	Lysine	1.5–2.0%	>2.5%	
Wheat	Iron	28.0–32.0 ppm	>38.0 ppm	
	Protein	8–10%	>12.0%	
	Zinc	30.0–32.0 ppm	>40.0 ppm	
Pearl millet	Zinc	30.0–35.0 ppm	>40.0 ppm	
	Iron	45.0–50.0 ppm	>70.0 ppm	
Lentil	Iron	45–50 ppm	>62.0 ppm	
	Zinc	35–40 ppm	>50.0 ppm	
Sweet potato	β-carotene	2.0–3.0 mg/100 g	>13.0 mg/100 g	
	Anthocyanin	Negligible	>80.0 mg/100 g	
Cauliflower	β-Carotene	Negligible	>8.0 ppm	
Pomegranate	Zinc	0.50–0.54 mg/100 g	>0.6 mg/100 g	
	Vitamin C	14.2–14.6 mg/100 g	>19.0 mg/100 g	
	Iron	2.7–3.2 mg/100 g	>5.0 mg/100 g	
Anti-nutritional factor				
Soybean	Kunitz trypsin inhibitor	30–45 mg/g of seed meal	Negligible	
Mustard	Glucosinolates	>120.0 ppm	<30.0 ppm	
	Erucic acid	>40%	<2.0%	

9.4.1 MICRONUTRIENTS

9.4.1.1 Zinc

Biofortification refers to strategies to improve the bioavailability of essential nutrients in everyday food items, such as maize, pearl millet, wheat, rice, and other grains. The effectiveness of several bacterial strains as possible biofortifying agents, including *Pseudomonas monilia* SRI-360, *P. plecoglossicida* SRI-156, *Enterobacter ludwigii* SRI-211, *E. ludwigii* SRI-229, *Brevibacterium antiquum* SRI-158, *Bacillus altitudinis* SRI-178, *Acinetobacter* and oil SRI-305. They found that the mineral content in chickpea and pigeon pea plants increased compared to uninoculated plots. On the other hand, when compared to the control treatments, the inoculation led to a significant increase in the growth of the plants (Kumar, Jose Anand, et al., 2024; Kumar & Choudhury, 2024g, 2024h; Kumar & Dey, 2024; Singh et al., 2024).

9.4.1.2 Iron

Mobilised iron is utilised by microbes that are experiencing a deficiency in the metal iron for their purposes (Kumar & Choudhury, 2024b, 2024h; Kumar & Singh, 2024c; Srivastav et al., 2024). The seed priming of fluorescent *Pseudomonas*, responsible for producing siderophores, was highly effective in increasing the iron (Fe) in chickpea grains. When compared to wheat grains that were not inoculated, it was discovered that the inoculation of wheat crops with siderophore-forming bacterial endophytes, such as *A. sulfonivorans* and *E. hirae*, resulted in a 46% and 67% increase in the amount of iron that was present in wheat grains of high- and low-Fe accruing wheat genotypes, respectively (Kumar & Choudhury, 2024d; Kumar et al., 2024a; Pandey et al., 2024).

9.4.1.3 Selenium (Se)

The microbiome can change the elemental configuration of selenium and its bioavailability. The following procedures are involved in the microbial enhancement of the Se biofortification process (Kumar, 2024a; Kumar & Choudhury, 2024a; Saini, Kumar, & Panwar, 2024; Saini, Kumar, & Upadhyay, 2024; Sharma, Kumar, & Singh, 2024; Sharma, Kumar, & Panwar, 2024). The bioavailability of selenium in the soil is increased due to effective microbes that change the soil characteristics and influence the redox chemistry of selenium.

9.4.1.4 Amino Acids

However, the glutaraldehyde treatment did not disturb plant growth characteristics or seed dimensions like wild-type rice. Scientists also reported enhancing the glycine concentration of coriander plants with a conventional approach. Their growth and glycine accumulation improved (except at the maximum 40 mg/L concentration) when they applied different concentrations of glycine (0–40 mg/L) in Hoagland's nutrient solution (Kumar & Singh, 2024b; Kumar, Dey, et al., 2024; Kumar, Singh, et al., 2024; Sharma, Kumar, & Singh, 2024; Sharma, Kumar, & Panwar, 2024). Biofortification of cereals with provitamin A has led to the development of "Golden Rice," with breeding programs aiming to provide 15 μg/g of β-carotene, offering a reliable source of vitamin A in regions where rice is a staple (Dey, Kumar, et al., 2024; Dey, Sharma, et al., 2024; Kumar, Saurabh, et al., 2024; Kumar & Choudhury, 2024b).

In potato agriculture, overexpression of the crtB gene increased the accumulation of lutein, violaxanthin, and β-carotene. Similarly, the overexpression of *phytoene synthase (PSY)* from daffodil, combined with bacterial LCYB and *crtI* genes, produced 1.6 μg/g dry weight of carotenoids in rice endosperm. Welsch and Li demonstrated that replacing the daffodil gene with the maize *PSY1* gene in "Golden Rice 2" achieved carotenoid content greater than 35 μg/g, showcasing the potential of transgenic technology for enhancing vitamins like vitamin A, though progress with other vitamins is also underway. Sorghum and "Golden Rice" are prominent examples of transgenic biofortification for increased vitamin A content (Kumar et al., 2024a). Gene silencing strategies have also been proposed to refocus metabolic flux towards carotenoid production (Sharma, Kumar, & Singh, 2024; Sharma, Kumar, & Panwar, 2024). Additionally, higher vitamin C levels in plants have been linked to improved growth, stress resilience, and longer shelf life after harvest, contributing to sustainable agriculture practices (Jagadesh et al., 2024; Kumar, Jose Anand, et al., 2024; Srivastav et al., 2024).

9.4.2 MACRONUTRIENTS

9.4.2.1 Phosphate

Both *Bacillus amyloliquefaciens* and *Pseudomonas* spp. were having roles, either on their own or in conjunction with humic acid (HA) extracted from green compost and phosphate-mobilising AM fungi. Microbes were introduced into maize plants, increasing growth and phosphate uptake from the soil. This was in comparison to plants that were not treated with microbes. In their study, the utilisation of Zn-solubilising *Bacillus* sp. (IA16) and P-solubilising *Bacillus subtilis* (IA6) in conjunction with one another was described. This utilisation enhanced the growth of cotton plants and their capacity to acquire nutrients (Devi et al., 2024a; Kumar & Singh, 2024a; Sharma, Kumar, & Singh, 2024; Sharma, Kumar, & Panwar, 2024).

9.4.2.2 Potassium

This *Burkholderia* sp. was capable of removing potassium from the soil. The inoculation of *Mikania micrantha* plants with *Burkholderia* sp. increased the amount of potassium that was assimilated and the amount of phosphorus that was present compared to plants that had not been inoculated. The effects of three potassium-solubilising bacteria, *Pantoea agglomerans*, *Rahnella aquatilis*, and *Pseudomonas orientalis*, on the utilisation of potassium, phosphorus, and nitrogen by *Oryza sativa* plants were investigated (Devi et al., 2024a; Kumar & Singh, 2024a).

9.5 BIOFORTIFIED AGRO-PRODUCTS

Increasing the quantities of micronutrients (iron, zinc, selenium, vitamin A, and other micronutrients) utilised by plants and then passed on to consumers is a standard method referred to as biofortification. This method was discussed earlier. Below are some of the valuable agricultural products (Table 9.2).

9.5.1 BIOFORTIFIED GRAIN

However, a growing number of alternative initiatives are currently aimed at addressing these issues through industrial fortification or pharmaceutical supplementation (Kumar, Singh, et al., 2024; Kumar, Srivastav, et al., 2024; Kumar & Singh, 2024c; Sharma, Kumar, & Singh, 2024; Sharma, Kumar, & Panwar, 2024).

9.5.2 BIOFORTIFICATION OF RICE

Additionally, it is essential to conduct additional research on biofortifying staple crops, particularly rice and wheat, because they have a significant potential to address nutrient shortages (Dey, Kumar, et al., 2024; Dey, Sharma, et al., 2024; Kumar & Choudhury, 2024f, 2024d; Pandey et al., 2024).

TABLE 9.2
Biofortification of Central Agriculture Produces

Sl. No.	Agriculture Produces	Technique Used	Type of Fortification	Health Benefits	Reference
Fruits					
1	Banana	Genetic engineering	Beta carotene	To alleviate vitamin A deficiency	Groth et al. (2021)
2	Apple	Agronomic	Selenium	Antioxidant properties	Groth et al. (2021)
3	Plum	Agronomic	Anthocyanin	Defence against the risk factors for cardiovascular disease and type 2 diabetes	Groth et al. (2021)
4	Strawberry	Microbial approach	Selenium	Antioxidant properties	Antoniou et al. (2021)
5	Apple, pear	Agronomic	Iodine	Ensure that the production of thyroid hormones is under control	Budke et al. (2021)
6	Tomato	Genetic engineering	Vitamin C	More nutraceutical value and antioxidant	Do et al. (2022)
Cereals					
7	Golden Rice	Genetic engineering	Provitamin A (beta-carotene)	Eye health, good vision	Do et al. (2022)
8	Biofortified rice	genetic engineering/ molecular breeding/Plant breeding	Fe- and Zn-rich rice	It prevents Fe from physical growth, sensory functions, the functioning of the immune system, neurobehavioural development, and reproductive health, all of which are contributed to by zinc, which also helps prevent anaemia	Do et al. (2022)
9	Fortified wheat	breeding or transgenic	Fe, Zn	Prevent Fe and Zn malnutrition	Sharma et al. (2021)
10	Anthocyanin-rich wheat	conventional breeding	High Anthocyanin content	Antioxidant, nutraceutical properties, anti-inflammatory activities	Do et al. (2022)
11	High amylose-rich wheat	CRISPR-Cas9	High amylose content	improves metabolic and digestive health	Do et al. (2022)
12	Low PA-level wheat	RNAi Technology, CRISPR/Cas9	Lowering phytic acid content,	Increase in Bioavailability of Fe, Zn, and others	Ibrahim et al. (2021)
13	Biofortified Maize	Breeding and genetic engineering	Provitamin A	Reduce Vitamin A deficiency	Do et al. (2022)
14	Zn and Se-rich maize	PGPRS, Cyanobacteria	Zn, Se	Health benefits	Do et al. (2022)

(*Continued*)

TABLE 9.2 (*Continued*)
Biofortification of Central Agriculture Produces

Sl. No.	Agriculture Produces	Technique Used	Type of Fortification	Health Benefits	Reference
15	Quality Protein Maize	Breeding, bifunctional expression/ silencing transgene cassette	Lysine and Tryptophan	Good quality proteins	Do et al. (2022)
16	Phytase, ferin-rich maize	Genetic engineering	Phytase, ferin	Increase bioavailability	Do et al. (2022)
17	Barley	Genetic engineering	Increase Phytase activity	Bioavailability of Zn and Fe	Do et al. (2022)
Legumes					
18	PUF-rich soybean	Genetic engineering	Linoleic acid γ-Linolenic Acid + stearidonic acid	nutraceutical and pharmaceutical potential	Do et al. (2022)
19	Soybean	Genetic engineering/ metabolic engineering	a higher concentration of seed protein, oleic acid, and provitamin A (beta-carotene)	Nutraceuticals	Do et al. (2022)
Vegetables					
20	Potato	Genetic engineering	Beta-carotene Zeaxanthin	Anticancer property	Bvenura et al. (2022)
21	Sweet Potato	Breeding and agronomic biofortification, genetic engineering	Beta-carotene, Antioxidants	Eye health and cancer	Sakure (2022)
22	Cassava	Genetic engineering	Beta-carotene	Anticancer property	Do et al. (2022)
23	Canola	Genetic engineering	Elevated β-carotenoid content	Nutraceutical	Do et al. (2022)
24	Linseed	Genetic engineering	DHA C22:5 n-3 (docosahexaenoic acid), EPA C20:5 n-3 (eicosapentaenoic acid), and arachidonic acid (C20:4 n-6)	cholesterol-lowering agent	Do et al. (2022)

9.5.2.1 Biofortification of Enhancing Vitamin A (Golden Rice)

An example of a genetically engineered food crop is rice modified to contain more provitamin A (β-carotene). Ingo Potrycus and Professor Peter Beyer are the individuals who came up with the idea of incorporating β-carotene into Golden Rice. They used *daffodil* and *crtl* genes within a soil bacterium called Agrobacterium science to alter the organism's genetic makeup. Golden Rice, on the other hand, is not produced through breeding. Golden Rice is available in two grades: Golden Rice 1 (SGR1) and Golden Rice 2. Both grades are designed to deliver 31 grammes of β-carotene per gramme of rice. Both grades are formed by replacing the daffodil *PSY* gene with the mouse gene. (Jagadesh et al., 2024; Kumar & Choudhury, 2024a, 2024h, 2024c; Kumar & Singh, 2024c).

9.5.2.2 Biofortification for Enhancing Fe Content

Polishing brown rice reduced approximately 4.75 times the grain's iron amount, from 19 parts per million to approximately four parts per million. To be more specific, the problem that led to the development of Fe biofortification, which was designed explicitly for milled rice, was the apparent decrease in the amount of flour that could be consumed by rice grains (Kumar & Raut, 2024; Sharma & Kumar, 2024).

9.5.2.3 Biofortification to Boost Zn Content

The functioning of the immune system, the health of the reproductive system, sensory perception, and the progression of neurobehavioural processes all require zinc for the body to develop correctly. Zinc is also essential for the development of the body. The CGIAR-HarvestPlus program was used in 2013 to disseminate a rice variety biofortified with zinc during the conventional breeding process in Bangladesh. Overall, 1.5 million agricultural households have accepted eight zinc-biofortified rice varieties, which have been cultivated since they were first introduced. IET 23832 (DRR Dhan45) is the name of a biofortified semi-dwarf plant variety developed by the Indian Institute of Rice Research (IIRR) in Hyderabad. This plant variety has a zinc content that ranges from 22.6 to 24.00 parts per million. This plant variety has a medium duration of 125 days and does not lodge(Jagadesh et al., 2024; Kumar & Choudhury, 2024c; Srivastav et al., 2024).

9.5.3 WHEAT BIOFORTIFICATION

Wheat, also known as *Triticum aestivum* L., is a Poaceae family member capable of self-pollination. Due to its long day length, the long-day plant flourishes in arid and semi-arid climates. Wheat is a significant crop used in agriculture all over the world. More than 30% of all food consumed across the globe is derived from this food, making it a primary staple food for a very long time. It is especially harmful to women to have deficiencies in micronutrients, particularly zinc and iron, because stunted development, impaired immunity, decreased productivity, and growth retardation are all potential outcomes of these deficiencies (Jagadesh et al., 2024; Kumar & Choudhury, 2024a, 2024h; Kumar & Singh, 2024c). Growth retardation is especially harmful to women. As a result, immediate action must be taken in response to this matter.

9.5.4 Biofortification of Maize

These amino acid deficiencies lead to reduced appetite, delayed growth, a lack of skeletal development, and abnormal behaviour. Several varieties of QPM have been developed. These varieties have tryptophan and lysine contents that are approximately twice as high as those of regular maize cultivars (lysine: 0.15–0.20% in flour; tryptophan: 0.07–0.08% in flour), which has led to a significantly raised nutritional quality (Kumar, Jose Anand, et al., 2024; Kumar & Choudhury, 2024g, 2024h; Singh et al., 2024).

9.5.5 Biofortified Vegetables

Commodities derived from vegetables are essential to the typical person's diet. Consequently, vegetables that have been biofortified can assist in the treatment of micronutrient deficiencies. Even though it is a relatively new phenomenon, the biofortification of vegetables to provide health benefits has been brought to the forefront due to the efforts and funding dedicated to addressing human nutritional deficiencies. There are currently efforts being made to develop biofortifying vegetables in several different regions around the world. The other side of the coin is that this idea is still in its infancy in India. Two methods that are complementary to one another can be utilised to increase the mineral concentrations in edible crops (Jagadesh et al., 2024; Kumar, 2024b; Raj et al., 2024).

9.6 GLOBAL IMPACT OF BIOFORTIFIED VARIETIES

When it comes to the health and well-being of the human population, implementing biofortified cultivars holds excellent promise. Several studies have demonstrated that these biofortified crops positively affect human beings, and these conclusions have been presented. To combat malnutrition and make progress towards the Sustainable Development Goals (SDGs), the creation and dissemination of biofortified varieties would be of great assistance (Kumar & Choudhury, 2024e).

9.6.1 Global Biofortification Programs and Initiatives

The African Biofortified Sorghum (ABS) project is another significant initiative that aims to improve sorghum's nutritional content, a key staple food in Sub-Saharan Africa. To achieve this goal, genetic modification is utilised to raise the levels of provitamin A, iron, and zinc in the crop. Research and the implementation of biofortification in Asia, Latin America, and Africa have also received financial assistance from the World Bank, the Bill and Melinda Gates Foundation, and other development agencies worldwide. Biofortification is becoming increasingly recognised as a viable approach to combating malnutrition at the policy level on a global scale. In Rwanda and Nigeria, for example, biofortified crops are included in the extension services that the government provides. To improve both food security and nutrition, the Scaling Up Nutrition (SUN) Movement and the Global Agriculture and Food

Security Program (GAFSP) have both incorporated biofortification into their respective strategic frameworks (Devi et al., 2024c; Dey, Kumar, et al., 2024; Kumar, Saurabh, et al., 2024; Kumar & Choudhury, 2024a; Saini, Kumar, & Upadhyay, 2024).

9.7 ADOPTION AND ACCEPTANCE OF BIOFORTIFIED CROPS

Because of this holistic approach, there was a significant increase in the amount of vitamin A that children consumed and high adoption rates. As an illustration, in areas accustomed to eating white maize, the orange colour of vitamin A biofortified maize was initially met with scepticism. To address these challenges, comprehensive strategies are required. Breeding for traits that enhance sensory appeal, conducting awareness campaigns, and making sure that biofortified crops are priced competitively and are easily accessible are some of the strategies that involve these strategies (Kumar et al., 2024a, 2024b).

9.8 CONCLUSION

A promising and sustainable strategy that can combat micronutrient deficiencies on a global scale, particularly in countries with low and middle incomes and biofortification, can be used to combat micronutrient deficiencies. There is a possibility that biofortification will significantly improve the outcomes of public health and reduce the prevalence of “hidden hunger.” It is possible to achieve this goal by increasing the nutrients in staple crops like rice, maize, and sweet potatoes. There are still several obstacles to overcome, even though it has much potential. The technical limitations in breeding, the regulatory hurdles, and the socio-cultural barriers to adoption are all examples of these obstacles. For the foreseeable future, the primary focus of efforts should be on advancing research to improve nutrient bioavailability, mitigating environmental impacts, and promoting policy support to facilitate widespread adoption. For biofortification to have the most significant possible impact, combining it with other nutritional strategies and using community involvement will be necessary. Biofortification can substantially achieve global food and nutrition security if supported appropriately and coordinated efforts are made.

REFERENCES

Antoniou, O., Chrysargyris, A., Xylia, P., & Tzortzakis, N. (2021). Effects of selenium and/or arbuscular mycorrhizal fungal inoculation on strawberry grown in hydroponic trial. *Agronomy*, *11*(4), 721. https://doi.org/10.3390/agronomy11040721

Budke, C., Dierend, W., Schön, H.-G., Hora, K., Mühling, K. H., & Daum, D. (2021). Biofortification of apples and pears in an orchard using foliar sprays of different composition. *Frontiers in Plant Science*, *12*, 638671. https://doi.org/10.3389/fpls.2021.638671

Bvenura, C., Witbooi, H., & Kambizi, L. (2022). Pigmented potatoes: A potential panacea for food and nutrition security and health? *Foods*, *11*(2), 175. https://doi.org/10.3390/foods11020175

Devi, P., Dey, S. R., Sharma, K., Kumar, P., & Singh, J. (2024). Waste management as an opportunity for the inclusion of vulnerable groups. In *Waste management in the circular economy* (pp. 207–230). Springer International Publishing. https://doi.org/10.1007/978-3-031-42426-7_11

Devi, P., Kumar, P., Dey, S. R., Banik, D., Kumar, G., & Mehta, C. M. (2024). Integrated omics approaches for nutrient stress management in plants. In *Current omics advancement in plant abiotic stress biology* (pp. 93–117). Elsevier. https://doi.org/10.1016/B978-0-443-21625-1.00008-7

Devi, P., Kumar, P., & Singh, J. (2024a). Application of potential microbes in bioremediation. In *Development in waste water treatment research and processes: Role of environmental microbiology in industrial wastewater research* (pp. 201–225). Elsevier. https://doi.org/10.1016/B978-0-443-13609-2.00021-5

Devi, P., Kumar, P., & Singh, J. (2024b). Biotechnological approaches as clean technology for the development of a sustainable environment. In *Development in waste water treatment research and processes: Role of environmental microbiology in industrial wastewater research* (pp. 381–403). Elsevier. https://doi.org/10.1016/B978-0-443-13609-2.00016-1

Devi, P., Kumar, P., & Singh, J. (2024c). Sewage treatment and energy recovery by bioelectrochemical oxidation system. In *Bioelectrochemical oxidation processes for wastewater treatment* (pp. 100–125). CRC Press. https://doi.org/10.1201/9781003368472-6

Dey, S. R., Kumar, P., & Panwar, J. S. (2024). Microbial desalination cell. In *Bioelectrochemical oxidation processes for wastewater treatment* (pp. 189–217). CRC Press. https://doi.org/10.1201/9781003368472-10

Dey, S. R., Sharma, M., & Kumar, P. (2024). Phytoremediation of phenolic compounds from soil. In *Bioremediation of emerging contaminants from soils: Soil health conservation for improved ecology and food security* (pp. 335–358). Elsevier. https://doi.org/10.1016/B978-0-443-13993-2.00016-5

Do, J. H., Park, S. Y., Park, S. H., Kim, H. M., Ma, S. H., Mai, T. D., Shim, J. S., & Joung, Y. H. (2022). Development of a genome-edited tomato with high ascorbate content during later stage of fruit ripening through mutation of *SlAPX4*. *Frontiers in Plant Science*, *13*, 836916. https://doi.org/10.3389/fpls.2022.836916

Groth, S., Budke, C., Weber, T., Neugart, S., Brockmann, S., Holz, M., Sawadski, B. C., Daum, D., & Rohn, S. (2021). Relationship between phenolic compounds, antioxidant properties and the allergenic protein mal d 1 in different selenium-biofortified apple cultivars (*Malus domestica*). *Molecules*, *26*(9), 2647. https://doi.org/10.3390/molecules26092647

Ibrahim, S., Saleem, B., Rehman, N., Zafar, S. A., Naeem, M. K., & Khan, M. R. (2021). CRISPR/Cas9 mediated disruption of inositol pentakisphosphate 2-kinase 1 (*TaIPK1*) reduces phytic acid and improves iron and zinc accumulation in wheat grains. *Journal of Advanced Research*, *37*, 33–41. https://doi.org/10.1016/j.jare.2021.07.006

Jagadesh, M., Dash, M., Kumari, A., Singh, S. K., Verma, K. K., Kumar, P., Bhatt, R., & Sharma, S. K. (2024). Revealing the hidden world of soil microbes: Metagenomic insights into plant, bacteria, and fungi interactions for sustainable agriculture and ecosystem restoration. *Microbiological Research*, *285*, 127764. https://doi.org/10.1016/j.micres.2024.127764

Kumar, P. (2024a). Epiphytes as a sustainable biomonitoring tool for environmental pollutants. In *Biomonitoring of pollutants in the Global South* (pp. 359–390). Springer Nature. https://doi.org/10.1007/978-981-97-1658-6_10

Kumar, P. (2024b). Soil conservation for global sustainability. In *World sustainability series* (pp. 103–128). Springer. https://doi.org/10.1007/978-3-031-57456-6_6

Kumar, P., & Choudhury, D. (2024a). Bioremediation of wastewater using hydroponics. In *Springer water* (pp. 27–64). Springer Nature. https://doi.org/10.1007/978-3-031-53258-0_2

Kumar, P., & Choudhury, D. (2024b). Hydroponic removal of organic contaminants from water. In *Springer water* (pp. 143–164). Springer Nature. https://doi.org/10.1007/978-3-031-53258-0_6

Kumar, P., & Choudhury, D. (2024c). Microbial native soil bacteria against cadmium toxicity. In *Springer water* (pp. 193–226). Springer Nature. https://doi.org/10.1007/978-3-031-54005-9_9

Kumar, P., & Choudhury, D. (2024d). Optimized phytoremediation process for the sustainable management of radionuclides. In *Bioremediation of emerging contaminants from soils* (pp. 443–464). Elsevier. https://doi.org/10.1016/B978-0-443-13993-2.00020-7

Kumar, P., & Choudhury, D. (2024e). Pollutants from agricultural activities in the Global South. In *Biomonitoring of pollutants in the Global South* (pp. 89–126). Springer Nature. https://doi.org/10.1007/978-981-97-1658-6_3

Kumar, P., & Choudhury, D. (2024f). Role of indigenous knowledge in agricultural soil reclamation without disturbing other ecosystems. In *Bioremediation of emerging contaminants from soils* (pp. 465–488). Elsevier. https://doi.org/10.1016/B978-0-443-13993-2.00021-9

Kumar, P., & Choudhury, D. (2024g). Seed plants as a sustainable biomonitoring tool for environmental pollutants. In *Biomonitoring of pollutants in the Global South* (pp. 321–357). Springer Nature. https://doi.org/10.1007/978-981-97-1658-6_9

Kumar, P., & Choudhury, D. (2024h). Toxicity of rhizospheric cadmium contaminated soil and its phytoremediation. In *Springer water* (pp. 227–264). Springer Nature. https://doi.org/10.1007/978-3-031-54005-9_10

Kumar, P., Devi, P., & Panwar, J. S. (2024). Constructed wetland-bioelectrochemical oxidation systems: A hybrid system for wastewater treatment. In *Bioelectrochemical oxidation processes for wastewater treatment* (pp. 126–151). CRC Press. https://doi.org/10.1201/9781003368472-7

Kumar, P., & Dey, S. R. (2024). Hydroponics phytoremediation: An overview. In *Springer water* (pp. 361–396). Springer Nature. https://doi.org/10.1007/978-3-031-53258-0_13

Kumar, P., Dey, S. R., & Choudhury, D. (2024). Effectiveness of cadmium on biochemical shift of pea plant treated with mycorrhiza and putrescine. *Nature Environment and Pollution Technology*, *23*(1), 275–285. https://doi.org/10.46488/NEPT.2024.v23i01.023

Kumar, P., & Dwivedi, P. (2024). Recent advances in the role of nitric oxide and cross-talk with signalling molecules under environmental stress in plants. In *Plant growth regulators: Resilience for sustainable agriculture* (pp. 55–78). Springer Nature. https://doi.org/10.1007/978-981-97-2918-0_4

Kumar, P., Jose Anand, A., & Rajagopal, A. R. (2024). Sustainable use of marine resources using ocean technologies. In *Sustainable development goals: Technologies and opportunities* (pp. 249–274). CRC Press. https://doi.org/10.1201/9781003468257-14

Kumar, P., & Raut, A. M. (2024). Microbes-assisted bioaugmentation process in the reduction of emerging industrial pollutants from soil. In *Bioremediation of emerging contaminants from soils* (pp. 519–540). Elsevier. https://doi.org/10.1016/B978-0-443-13993-2.00023-2

Kumar, P., Saini, L., & Sharma, M. (2024). Assessment of the phytochemical constituents and metabolites in the medicinal plants and herbal medicine used in the treatment and management of respiratory diseases. In *Reference series in phytochemistry* (pp. 481–510). Springer. https://doi.org/10.1007/978-3-031-43199-9_19

Kumar, P., Saurabh, S., & Sharma, K. (2024). Assessment of the phytochemical constituents and metabolites in medicinal plants and herbal remedies used in the treatment and management of reproductive diseases: Polycystic ovary syndrome. In *Reference series in phytochemistry* (pp. 511–532). Springer. https://doi.org/10.1007/978-3-031-43199-9_20

Kumar, P., Sharma, K., & Singh, J. (2024a). Analysis of complex microbial communities in soil and wastewater treatment processes. In *Microbial metagenomics in effluent treatment plant* (pp. 25–41). Elsevier. https://doi.org/10.1016/B978-0-443-13531-6.00002-1

Kumar, P., Sharma, K., & Singh, J. (2024b). Response of microbial community to environment changes. In *Microbial metagenomics in effluent treatment plant* (pp. 43–61). Elsevier. https://doi.org/10.1016/B978-0-443-13531-6.00006-9

Kumar, P., & Singh, J. (2024a). Bioprocesses in industrial wastewater treatment: Trends and prospects. In *Trends in biological processes in industrial wastewater treatment* (pp. 15-1–15-50). IOP Publishing. https://doi.org/10.1088/978-0-7503-5678-7ch15

Kumar, P., & Singh, J. (2024b). Exploring enzyme biotechnology's role in green chemistry and advancements in pharmaceutical technologies. In *Enzyme biotechnology for environmental sustainability* (pp. 465–495). Elsevier. https://doi.org/10.1016/B978-0-443-22072-2.00015-2

Kumar, P., & Singh, J. (2024c). Hydroponic: An eco-friendly future. In *Springer water* (pp. 223–267). Springer Nature. https://doi.org/10.1007/978-3-031-53258-0_9

Kumar, P., Singh, J., & Singh, G. (2024). Renewable hydrogen opportunities and challenges. In *Renewable hydrogen: Opportunities and challenges in commercial success* (pp. 193–220). Elsevier. https://doi.org/10.1016/B978-0-323-95379-5.00010-9

Kumar, P., Srivastav, A. L., Chaudhary, V., van Hullebusch, E. D., & Busquets, R. (2024). *Bioremediation of emerging contaminants from soils: Soil health conservation for improved ecology and food security*. Elsevier. https://doi.org/10.1016/C2022-0-02758-6

Pandey, S. K., Kumar, P., & Dey, S. R. (2024). Xenobiotic stress management by microbial endophytes. In *Bioremediation of emerging contaminants from soils* (pp. 241–259). Elsevier. https://doi.org/10.1016/B978-0-443-13993-2.00012-8

Raj, A., Goutam, E., Kumar, P., & Singh, J. (2024). Polyphenol oxidase: An enzyme of plants. In *Polyphenol oxidases: Function, wastewater remediation, and biosensors* (pp. 105–128). De Gruyter. https://doi.org/10.1515/9783111033525-004

Saini, L., Kumar, P., & Panwar, J. S. (2024). Current status of wastewater in India/other countries/regions. In *Bioelectrochemical oxidation processes for wastewater treatment* (pp. 152–165). CRC Press. https://doi.org/10.1201/9781003368472-8

Saini, L., Kumar, P., & Upadhyay, H. (2024). Zinc and boron foliar application effects on primed mung bean (*Vigna radiata* L.) growth and productivity. *Nature Environment and Pollution Technology*, *23*(3), 1407–1418. https://doi.org/10.46488/NEPT.2024.v23i03.012

Sakure, A. A. (2022). Enhancement of nutritional, pharmaceutical and industrial value of crops through genetic modification with carotenoid pathway genes. In *Biotechnology and crop improvement* (pp. 63–77). CRC Press.

Sharma, K., & Kumar, P. (2024). Environmental threats posed by xenobiotics. In *Bioremediation of emerging contaminants from soils* (pp. 183–201). Elsevier. https://doi.org/10.1016/B978-0-443-13993-2.00009-8

Sharma, K., Kumar, P., & Singh, J. (2024). Ecology and diversity of microbial communities involved in the removal of priority contaminants and micropollutants in waste. In *Development in waste water treatment research and processes* (pp. 181–199). Elsevier. https://doi.org/10.1016/B978-0-443-13609-2.00014-8

Sharma, M., Kumar, P., & Panwar, J. S. (2024). Implementation of bioelectrochemical oxidation systems in existing wastewater treatment plant: Challenges in retrofitting. In *Bioelectrochemical oxidation processes for wastewater treatment* (pp. 166–188). CRC Press. https://doi.org/10.1201/9781003368472-9

Sharma, V., Choudhary, M., Kumar, P., Choudhary, J. R., Khokhar, J. S., Kaushik, P., & Goli, S. (2021). Harnessing the wild relatives and land races for Fe and Zn biofortification in wheat through genetic interventions—A review. *Sustainability*, *13*(23), 12975. https://doi.org/10.3390/su132312975

Singh, J., Kumar, P., & Siddique, A. (2024). Biochar-based organic amendments on soil health, nutrient status and quality of potato (*Solanum tuberosum*). *Plant Science Today*, *11*(1), 102–108. https://doi.org/10.14719/pst.2706

Srivastav, A. L., Patel, N., Rani, L., Kumar, P., Dutt, I., Maddodi, B. S., & Chaudhary, V. K. (2024). Sustainable options for fertilizer management in agriculture to prevent water contamination: A review. *Environment, Development and Sustainability*, *26*(4), 8303–8327. https://doi.org/10.1007/s10668-023-03117-z

10 Bioinformatics and Crop Improvement

Revolutionizing Plant Biotechnology in Achieving Food Security

A Sheena Sabatina and Renu Kumari

10.1 INTRODUCTION

Bioinformatics employs computer software tools for tasks such as database creation, data management, data warehousing, data mining, and global communication networking. It involves the recording, annotation, storage, analysis, and retrieval of nucleic acid sequences (genes and RNAs), as well as protein sequences and their structural information. This encompasses databases containing sequences and structural data, along with methods for accessing, searching, visualizing, and retrieving that information. Bioinformatics focuses on developing and maintaining databases of biological information, allowing researchers to access existing data and contribute new entries. Functional genomics, biomolecular structure, proteome analysis, cellular metabolism, biodiversity, downstream processing in chemical engineering, drug, and vaccine development are several fields where bioinformatics plays a crucial role (Singh et al., 2011).

Information about biology has risen significantly as a result the major advancements in molecular science and genomics. The application of computational biology and computation in biology toward the research field of plant physiology is significantly speeding up scientific advancement in the biological sciences. Researchers have discovered the genetic composition of numerous plant and microbe species, as well as their transcriptome, proteome, and metabolome, which disclose their proteins, transcripts, and metabolic pathways (Gomez et al., 2018). In the current scientific period, the sequencing method is the most accurate way to obtain the entire genome sequences in the form of DNA, RNA, and protein sequences from an organism's genome. Whole-genome sequencing offers a means of understanding the structure of various species and gives insight into their capability.

A comprehensive sequencing dataset includes both coding and non-coding sections that can serve as an essential precursor to any functioning gene that determines an organism's distinct characteristics. All areas, including exons, introns, regulators, and promoters, are included in the final sequence, which frequently yields a significant quantity of genomic data (Normand et al., 2019). As next-generation sequencing (NGS) and other omics technologies are developed for studying plant genomics, the

DOI: 10.1201/9781003597797-10

number of plants with their genomes sequenced will increase (Blätke et al., 2021). The creation and application of bioinformatics enables investigators to collect, store, and arrange these enormous volumes of data into systematic database. In the early 1950s, DNA was a subject of debate due to its role as a genetic information carrier molecule. Despite the discovery of the double-helix structure of DNA in 1953, genome sequencing and genetics code deciphered only 25 and 13 years, respectively. Computational biology was applied to DNA analysis almost 20 years behind protein analysis, which had a better understanding of its chemical makeup. In the late 1950s, the first sequence of a protein, insulin was released, along with advancements in crystallography. The Edman degradation method became a straightforward technique for protein sequencing, enabling sequencing of over 15 distinct protein families (Singh et al., 2011).

However, getting large protein sequences was a challenge due to the yield of the Edman reaction, which could only sequence 50–60 amino acids. In the early 1960s, one of the first known bioinformatics software was developed to solve this problem. Margaret Dayhoff, a physical chemist, was the first to use computational methodologies in biochemistry, and her work in electrochemistry and computer systems in medical and biological research led to the creation of protein (Blätke et al., 2021). Emile Zuckerkandl and Linus Pauling shifted the focus of biochemistry research from mechanistic modeling of enzymes to examining biomolecular sequences as "carriers of information." They questioned whether protein sequences evolved through a similar mechanism to words and languages, which evolve through subtle changes over time. They found that identical proteins from vertebrates, like hemoglobin, exhibited a level of similarity that was too great over an extended evolutionary period to be the product of either convergence in development or chance. Walter M. Fitch introduced the idea of orthology in 1970 to characterize homology that emerged from a speciation event. The European Molecular Biology Open Software Suite, developed in 1996, was based on the free software ideology.

This line of thinking was already prominent in prior projects, such as Collaborative Computational Project Number 4 (CCP4) for macromolecular X-ray crystallography. The sequence databases of the European Molecular Biology Laboratory, GenBank, and DNA Data Bank of Japan were formed during this time, aiming to standardize data formatting and aid in data sharing. The free software movement and specialized scientific journals expanded the use of computers in biology, but small-scale mainframe computing systems were used for massive amounts of data (Singh et al., 2011). This chapter explores the pivotal role of bioinformatics in modern plant breeding, focusing on its application in crop improvement for sustainable agriculture. It examines the integration of genomic data with computational models to accelerate the discovery of beneficial traits, the development of high-throughput technologies, and the use of bioinformatics in precision breeding.

10.2 HISTORY OF BIOINFORMATICS

Bioinformatics is an interdisciplinary field that emerged in the 1960s, combining biology, computer science, and mathematics to analyze and interpret biological data. Its origins trace back to the need for computing resources to store and process large-scale biological information, particularly in genetics and molecular biology. The development of DNA sequencing technologies and the human genome project in the

TABLE 10.1
Timeline of Bioinformatics: New Era to Emerge

Year	Bioinformatics Timeline
1970	"Bioinformatics" term was coined by Ben Hesper and Paulien Hogeweg
1970	"dot matrix method" for amino acid and nucleotide sequences comparative analysis by A.J. Gibbs and G.A. McIntyre developed
1970	Dynamic programming algorithm for sequence alignment Needleman and Wunsch proposed it
1984	GCG software suit (UWGG) published it, which was a collection of tools for manipulating DNA, RNA, or protein sequences
1990	BLAST: fast sequence similarity searching
1994	EMBL European Bioinformatics Institute, Hinxton, UK
1999	Fly genome completely sequenced
2000s	Next-generation sequencing or NGS that started with the "454" pyrosequencing

late 20th century significantly accelerated the growth of bioinformatics, enabling the analysis of vast datasets. Over time, bioinformatics has become essential for understanding biological processes, drug discovery, and personalized medicine. Table 10.1 summarizes the timeline of bioinformatics (Normand et al., 2019).

10.3 ROLE OF COMPUTATIONAL BIOINFORMATICS IN AGRICULTURE

The environment's biodiversity is already in threat due to habitat degradation and land exploitation for agricultural purposes (Kumari et al., 2021). Increasing agricultural production to fulfill the demand because of land exploitation and destruction of habitat for agricultural purposes, the biodiversity of the environment is already in danger mode (Kumari et al., 2021). Expanding agriculture to meet the world's growing food needs does not appear to be feasible in situations like climate change. Utilizing sustainable agricultural practices, appropriate policy implementation, and environmental investments, the Food and Agricultural Organization, known as the FAO, introduced the "climate-smart agriculture" approach to agriculture, which can boost productivity and further improve resilience. It also helps to achieve national food security and development goals by removing greenhouse gases when feasible. By mixing genes, it is possible to build plants with better performance for the food and agricultural industries by using the potential gene pool of crops and their wild relatives. Whole-genome analysis is carried out in NGS, which makes it possible to identify the genetic basis of different phenotypic changes as well as new ones (Ashraf et al., 2022).

Most of the time, sequence information is assembled and well annotated, and the genome database of a number of significant crops have been sequenced, allowing for the identification of genotyping-phenotypic relationships (Nizamani et al., 2023). These crops include rice, soybean, foxtail millet, chickpea, and pigeon pea. A number of other plant genomes have been assembled with the quickly evolving second-generation sequencing techniques, including wheat in 2018, *Digitaria exilis* in the year 2020, *Secale cereale* (rye) in the year 2021. NGS techniques facilitate the sequencing of plant genomes and the transcriptomes, and the resulting data has been utilized to develop updated reference genome maps for crops like soybean, rice, and corn (Hong et al., 2023). The sorghum genome's characterization has revealed a number of noteworthy characteristics, including an extra ~29.6 Mb of sequence, a 24% increase in annotated genes, longer genes, and a ten-fold reduction in error frequency. The most popular platforms for NGS techniques include Roche 454, PacBio, Ion Torrent, Illumina/Solexa sequencing, and Oxford Nanopore. NGS techniques usually being developed to cover complicated plant genomes (Gupta & Verma, 2019). The third-generation sequencing methods, including Nanopore and PacBio, have made it easier to assemble complicated genomes with good quality. These methods are also being applied to RNA sequencing, which is relatively a newer technique that is useful for mapping and measuring transcriptomes, particularly for non-model crops such as bread wheat cultivar *Yunong 201* and olive during fruit development (Athanasopoulou et al., 2021).

10.4 AGRICULTURE USING NEXT-GENERATION SEQUENCING

NGS has transformed agricultural research by enabling high-throughput genome analysis, accelerating the identification of beneficial crop traits like disease resistance and stress tolerance (Satasiya et al., 2024). Technologies like Illumina sequencing have been pivotal in characterizing soybean transcription factors, copper-tolerant genes, and herbicide-resistant variants (Satam et al., 2023; Satasiya et al., 2024). Transcriptomics, which studies gene expression patterns (Wang et al., 2009), has revealed regulatory networks for stress responses, including salt tolerance in soybeans and heat stress mechanisms in hybrid rice (Satasiya et al., 2024). Marker-assisted selection (MAS) leverages NGS data to introgress quantitative trait loci (QTLs), such as the *SUB1* gene for submergence tolerance in rice (Sarkar & Bhattacharjee, 2011). SNP markers and whole-genome regression (WGR) further enhance precision breeding by identifying genetic variations, though long-read sequencing challenges persist. Additionally, bioinformatics and machine learning (ML) analyze microbiome data to predict crop diseases like huanglongbing (HLB) with high accuracy (Kumar et al., 2023). These integrative approaches enable early disease detection and optimize crop management. By combining NGS with advanced computational tools, researchers can decode complex agricultural systems, driving sustainable productivity (Figure 10.1). However, technical limitations, such as sequencing errors and data integration hurdles, require ongoing innovation.

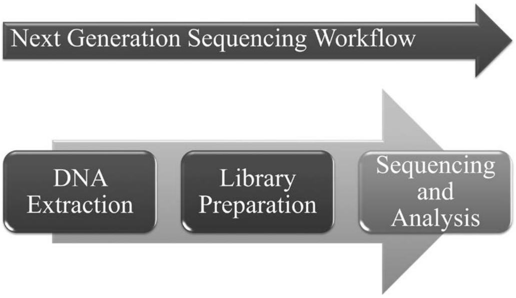


FIGURE 10.1 Next-generation sequencing technology (NGS).

10.5 DATABASE PROVIDE ABUNDANT INFORMATION ABOUT GENES AND PATHWAYS

Databases that combine data on proteomics, metabolic pathways, and genomes are referred to as general databases. A unified and publicly accessible compilation of all published data is represented by genome databases, which enable researchers to easily learn more about the gene or protein of interest. UniProt, for instance, provides a thorough database for protein sequences and functional functions. In addition to using keywords of interest or a specific gene/protein name to search the database and sort through the data entered, a protein BLAST can be used to obtain the sequence of the new protein of interest (Batieno et al., 2016).

Furthermore, broad databases gather vibrant natural circuits that are comparable to those represented in metabolic pathways, Eukaryotic Orthologous Groups (KOG), Kyoto Encyclopedia of Genes and Genomes (KEGG), and Gene Ontology (GO), which can be used to identify whether a particular protein is a member of one of the many known pathways (Kanehisa et al., 2017). In bioinformatics, identifying genes from large genomic sequences is an essential task. One of two points may be encountered by the reflection of genomic sequences. The target sequences' gene reflection can be carried out in the first script using free Ab initio algorithms are typically used by the alternative script on platforms and search platforms (Kanehisa et al., 2017). In the agricultural industry, bioinformatics is essential for many experiments that react to unfavorable growth conditions and adjust accordingly. For example, eelgrass (*Zostera marina*) does not have stomata because it lacks the essential genes needed for stomatal experimentation, possibly as a result of adaptation to water life (Ergin et al., 2022).

Changes in the CAZyme protein family cause the eelgrass's cuticle to thicken. Modifications to a few metabolic pathways were also caused by alterations in transporter genes and sucrose synthase. Only bioinformatics techniques can improve these investigations. In order to adapt to a high-salinity environment, halophytes have developed very special traits. The relative study of halophytes to determine their acclimations to saline-alkaline soils has also been made much easier by bioinformatics. Bioinformatics was used to analyze the causes of Selaginella, a type of xerophyte that thickens the cuticles of leaves in order to adapt to thirsty environments. Utilizing animals and insects as food sources, bioinformatics provided the molecular foundation for entomologists' explanation (Ergin et al., 2022).

Additionally, as more genomes were accessible, bioinformatics revealed the evolutionary lineages of businesses. An example of this is the evolutionary analysis of water lilies (*Nymphaea colorata*), which showed that Amborellales and Nymphaeales are the successive family lineages to all other extant angiosperms (Chen et al., 2017). Before being introduced into a factory, genes linked to particular features can be examined on a computer. The results of this study can be used to decide what elements should be added to the factory for a more accurate phenotypic analysis. Examples include the newly developed vitamin D-rich tomato, lettuce (*Lactuca sativa*), and maize (*Zea mays* L.) kernels, which are high in lysine (Ergin et al., 2022).

10.6 IMPORTANCE OF AGRICULTURAL BIOINFORMATICS

Bioinformatics focuses on analyzing and making sense of various data types, including nucleotide and amino acid sequences, protein domains, and building blocks of proteins. It aims to improve understanding of biological processes by developing computationally intensive techniques like pattern recognition, visualization, data mining, and ML. Research in this field includes gene discovery, genome assembly, drug design, protein structure alignment, protein structure prediction, gene expression, protein-protein interaction prediction, genome-wide association studies, and Darwin's theory modeling. Crops are crucial for the economy, society, and environment and modern plant biotechnology must tackle the problem of feeding the world's expanding population. Moreover, DNA microarray expression and expressed sequence tags can help to study gene transcription patterns in plants (Kanehisa et al., 2017).

10.7 APPLICATIONS OF BIOINFORMATICS IN AGRICULTURE

Creating novel plant varieties is the goal of plant biotechnology, the sequencing analysis requires financial expenditure. Since genomics-assisted breeding is a cost-effective and successful approach, it is frequently used in crop breeding. Crop physiology makes substantial use of genomic-supported parenting because it is a successful and provident technique (Tyagi et al., 2024). Under various circumstances, such as changes in plant physiology, pathogen invasion, or environmental changes, genomics may be able to follow molecular changes during development and aid in understanding the structure and operation of biological systems. Samples might be taken from the same person or from multiple people of the same species or from different species, and their analysis necessitates additional genomic research (Chong et al., 2019).

Furthermore, to study about the genomics in crops, GWAS has been utilized, for instance, in *Chrysanthemum* to investigate genetic patterns and find advantageous alleles for a number of ornamental and resistance qualities, such as drought, aphid, waterlogging, and plant structure and inflorescence attributes (Su et al., 2019). For instance, in *Chrysanthemum*, GWAS has been used to investigate inheritable patterns and find advantageous alleles for a number of resistance and esthetic qualities, including as failure forbearance, aphid resistance, waterlogging forbearance, and factory structure and inflorescence traits. Su and colleagues transferred a significant SNP co-isolated *Chrysanthemum*. *Chrysanthemum* waterlogging forbearance was confirmed in 52 cultivars or grandfathers using a PCR-based deduced cut amplified polymorphism sequence (dCAPS) marker with a delicacy of 78.9. Two dCAPS labels were created in relation to the flowering stage and head periphery (Chong et al., 2019).

10.8 CHALLENGES

Bioinformatics has emerged as a transformative tool in crop improvement, enabling the analysis of vast genomic datasets to identify traits linked to yield, stress tolerance, and disease resistance (Varshney et al., 2021). By integrating genomics, transcriptomics, and proteomics, bioinformatics accelerates MAS and genome editing, enhancing precision breeding (Bohra et al., 2022). However, challenges persist, including data heterogeneity, high computational costs, and the need for advanced algorithms to interpret complex biological interactions (Wallace et al., 2018). Additionally, limited bioinformatics infrastructure in developing nations restricts accessibility, exacerbating global disparities in agricultural biotechnology (Tester & Langridge, 2010). Addressing these barriers requires collaborative efforts to standardize data, reduce costs, and build capacity in low-resource regions (Wallace et al., 2018).

10.9 FUTURE PROSPECTS

Looking ahead, the future of bioinformatics in crop improvement lies in harnessing artificial intelligence (AI) and ML to predict phenotypic outcomes from genotypic data (Singh et al., 2023). Advances in single-cell sequencing and pangenome analysis will further refine trait discovery, enabling the development of climate-resilient crops (Khan et al., 2022). Public-private partnerships are critical to scaling these technologies, ensuring smallholder farmers' benefit from innovations (CGIAR, 2023). Ethical considerations, such as data privacy and equitable access to genetically improved crops, must also guide policy frameworks (ISAAA, 2022). Ultimately, bioinformatics-driven crop improvement holds immense potential to achieve global food security, provided these scientific, infrastructural, and ethical challenges are addressed (Ray et al., 2019).

10.10 CONCLUSION

Bioinformatics typically plays a key role in managing the enormous volumes of data produced by high-throughput methods, as well as in data integration, analysis, and further model prediction. Cell division, pollen tube expansion, and other phenomena are typically visualized in classical biological tests. However, more research indicates that deeper, higher throughput studies and analyses are being conducted. The molecular pathways that underlie plant responses to biotic and abiotic stressors have been investigated; when combined, they can be a potent tool that opens up new avenues for computational biology and agriculture. In order to meet the sustainable global food needs, the goals of the Sustainable Development Goals, and the worldwide plant trade, a shared biosecurity infrastructure can be built.

REFERENCES

Ashraf, M. F., Hou, D., Hussain, Q., Imran, M., Pei, J., Ali, M., & Lin, X. (2022). Entailing the next-generation sequencing and metabolome for sustainable agriculture by improving plant tolerance. *International Journal of Molecular Sciences*, *23*(2), 651. https://doi.org/10.3390/ijms23020651

Athanasopoulou, K., Boti, M. A., Adamopoulos, P. G., Skourou, P. C., & Scorilas, A. (2021). Third-generation sequencing: The spearhead towards the radical transformation of modern genomics. *Life*, *12*(1), 30. https://doi.org/10.3390/life12010030

Batieno, B. J., Tignegre, J. B., Sidibé, H., Zongo, H., Ouedraogo, T. J., Danquah, E., & Ofori, K. (2016). Field assessment of cowpea genotypes for drought tolerance. *International Journal of Sciences: Basic and Applied Research*, *30*(4), 358–369.

Blätke, M. A., Szymanski, J. J., Gladilin, E., Scholz, U., & Beier, S. (2021). Editorial: Advances in applied bioinformatics in crops. *Frontiers in Plant Science*, *12*, 640394. https://doi.org/10.3389/fpls.2021.640394

Bohra, A., Saxena, R. K., Gnanesh, B. N., & Varshney, R. K. (2022). Genomics and bioinformatics interventions for crop improvement. *Trends in Plant Science*, *27*(3), 123–135. https://doi.org/10.1016/j.tplants.2021.08.006

CGIAR. (2023). *Accelerating crop breeding for food security*. https://www.cgiar.org/initiative/accelerated-breeding/

Chen, F., Liu, X., Yu, C., Chen, Y., Tang, H., & Zhang, L. (2017). Water lilies as emerging models for Darwin's abominable mystery. *Horticulture Research*, *4*, 17051. https://doi.org/10.1038/hortres.2017.51

Chong, X., Su, J., Wang, F., Wang, H., Song, A., Guan, Z., & Zhang, F. (2019). Identification of favorable SNP alleles and candidate genes responsible for inflorescence-related traits via GWAS in chrysanthemum. *Plant Molecular Biology*, *99*, 407–420. https://doi.org/10.1007/s11103-019-00825-x

Ergin, B. C., Zimmerman, R. C., & Hill, V. J. (2022). Photorespiration in eelgrass (*Zostera marina* L.): A photoprotection mechanism for survival in a CO_2-limited world. *Frontiers in Plant Science*, *13*, 1025416. https://doi.org/10.3389/fpls.2022.1025416

Gomez, D. F., Busi, M. V., Barchiesi, J., Peralta, D. A., Hedin, N., & Bhadauria, V. (2018). Applications of bioinformatics to plant biotechnology. *Current Issues in Molecular Biology*, *27*, 89–104. https://doi.org/10.21775/cimb.027.089

Gupta, N., & Verma, V. K. (2019). Next-generation sequencing and its application: Empowering in public health beyond reality. In *Microbial technology for the welfare of society* (pp. 313–341). Springer. https://doi.org/10.1007/978-981-13-8844-6_15

Hong, K., Radian, Y., Manda, T., Xu, H., & Luo, Y. (2023). The development of plant genome sequencing technology and its conservation and application in endangered gymnosperms. *Plants*, *12*(23), 4006. https://doi.org/10.3390/plants12234006

ISAAA. (2022). *Global status of biotech crops: Policy implications*. Brief 56. https://www.isaaa.org/resources/publications/briefs/56/download/isaaa-brief-56-2021.pdf

Kanehisa, M., Furumichi, M., Tanabe, M., Sato, Y., & Morishima, K. (2017). KEGG: New perspectives on genomes, pathways, diseases and drugs. *Nucleic Acids Research*, *45*(D1), D353–D361. https://doi.org/10.1093/nar/gkw1092

Khan, A. W., Garg, V., & Roorkiwal, M. (2022). Pangenomics: A new frontier in crop improvement. *Nature Reviews Genetics*, *23*(5), 312–325. https://doi.org/10.1038/s41576-021-00424-x

Kumar, S. S., Muhammad, W., & Muhammad, A. M. (2023). Bioinformatics, big data and agriculture: A challenge for the future. *Frontiers in Plant Science*, *14*, 1271305. https://doi.org/10.3389/fpls.2023.1271305

Kumari, R., A, D., & Bhatnagar, S. (2021). Biodiversity losses: Threats and conservation strategies. *International Journal of Pharmaceutical Sciences and Review Research*, *37*(2), 242–254.

Nizamani, M. M., Zhang, Q., Muhae-Ud-Din, G., & Wang, Y. (2023). High-throughput sequencing in plant disease management: A comprehensive review of benefits, challenges, and future perspectives. *Phytopathology Research*, *5*, 44.

Normand, E. A., and, & Veyyer, V. I. BV. I. B. (2019). Next-generation sequencing for gene panels and clinical exomes. In P. C. K. Leung and J. Qiao (Eds.), *Human reproductive and prenatal genetics* (pp. 1–15). Academic Press.

Ray, D. K., Mueller, N. D., West, P. C., & Foley, J. A. (2019). Yield trends are insufficient to double global crop production by 2050. *PLoS ONE*, *14*(6), e0217148. https://doi.org/10.1371/journal.pone.0217148

Sarkar, R. K., & Bhattacharjee, B. (2011). Rice genotypes with SUB1 QTL differ in submergence tolerance, elongation ability during submergence and re-generation growth at re-emergence. *Rice*, *5*(1), 7. https://doi.org/10.1007/s12284-011-9065-z

Satam, H., Joshi, K., Mangrolia, U., Waghoo, S., Zaidi, G., Rawool, S., Thakare, R. P., Banday, S., Mishra, A. K., & Das, G. (2023). Next-generation sequencing technology: Current trends and advancements. *Biology*, *12*(7), 997. https://doi.org/10.3390/biology12070997

Satasiya, P., Patel, S., Patel, R., Raigar, O. P., Modha, K., Parekh, V., Joshi, H., Patel, V., Chaudhary, A., Sharma, D., & Prajapati, M. (2024). Meta-analysis of identified genomic regions and candidate genes underlying salinity tolerance in rice (*Oryza sativa* L.). *Scientific Reports*, *14*, 5730. https://doi.org/10.1038/s41598-024-54764-9

Singh, A. K., Ganapathysubramanian, B., & Sarkar, S. (2023). Machine learning for high-throughput plant phenotyping. *Plant Biotechnology Journal*, *21*(1), 45–60. https://doi.org/10.1111/pbi.13923

Singh, A.K., Rath, S.K., & Misra, K. (2011). Identification of epitopes in Indian human papilloma virus 16 E6: A bioinformatics approach, *Journal of Virological Methods*, *177*(1), 26–30.

Su, J., Zhang, F., Chong, X., Song, A., Guan, Z., Fang, W., & Chen, F. (2019). Genome-wide association study identifies favorable SNP alleles and candidate genes for waterlogging tolerance in chrysanthemums. *Horticulture Research*, *6*, 21. https://doi.org/10.1038/s41438-018-0101-7

Tester, M., & Langridge, P. (2010). Breeding technologies to increase crop production in a changing world. *Science*, *327*(5967), 818–822. https://doi.org/10.1126/science.1183700

Tyagi, A., Mir, Z. A., Almalki, M. A., Deshmukh, R., & Ali, S. (2024). Genomics-assisted breeding: A powerful breeding approach for improving plant growth and stress resilience. *Agronomy*, *14*(6), 1128. https://doi.org/10.3390/agronomy14061128

Varshney, R. K., Sinha, P., Singh, V. K., Kumar, A., & Zhang, Q. (2021). 5Gs for crop genetic improvement. *Trends in Plant Science*, *26*(4), 317–328. https://doi.org/10.1016/j.tplants.2020.11.004

Wallace, J. G., Rodgers-Melnick, E., & Buckler, E. S. (2018). On the road to breeding 4.0: Unraveling the good, the bad, and the boring of crop quantitative genomics. *Annual Review of Genetics*, *52*, 421–444. https://doi.org/10.1146/annurev-genet-120116-024846

Wang, Z., Gerstein, M., & Snyder, M. (2009). RNA-Seq: A revolutionary tool for transcriptomics. *Nature Reviews Genetics*, *10*(1), 57–63. https://doi.org/10.1038/nrg2484

11 Environmental Benefits of Biotech Crops

Swarnalata Tripathy, Binod Kumar Mahapatra, and Amruta Panda

11.1 INTRODUCTION

The implementation of sustainable agriculture faces challenges due to climate change, population growth, and shrinking arable land, necessitating genetically modified (GM) crops with enhanced productivity, quality, and stress resistance (Kumar *et al.,* 2020; Abdul Aziz et al., 2022). Genetic modification aligns with sustainable farming principles by boosting yields, conserving resources, and improving human health, with biotechnology enabling traits like pest resistance and drought tolerance (European Commission, 2010). GM crops have reduced pesticide use by 8.3% and lowered greenhouse gas emissions equivalent to removing 15.27 million cars from roads, demonstrating environmental benefits (Brookes & Barfoot, 2020). GM crops, such as Flavr Savr tomatoes and Bt cotton, were developed to delay ripening, resist pests, and tolerate herbicides, marking significant milestones in agricultural biotechnology (Bawa & Anilakumar, 2013; Vega Rodríguez et al., 2022).

Advances like herbicide-tolerant (HT) and insect-resistant (IR) traits have improved crop resilience, reducing chemical inputs while enhancing food security (Batista et al., 2017; Kurup & Thomas, 2020). However, concerns persist over biodiversity loss, gene flow to wild relatives, and the emergence of herbicide-resistant weeds, which threaten ecosystems (Fernandes et al., 2022; Sharma et al., 2019). Environmental risks, such as genetic pollution and harm to non-target species like monarch butterflies, highlight the need for mitigation strategies (Boyle et al., 2019; Fitzpatrick & Reid, 2019; Roberts et al., 2020). Studies suggest isolating GM crops by 50 meters to minimize gene flow, as seen in oilseed rape cases (Carrière et al., 2021; Nishizawa et al., 2010). Public awareness and labeling of GM products are crucial to address skepticism and ensure informed consumer choices (Borges et al., 2018; Wunderlich & Gatto, 2015). This chapter highlights the adoption of biotech crops, which has significantly contributed to sustainable agriculture by reducing pesticide use, lowering greenhouse gas emissions, and enhancing resource efficiency.

11.2 REDUCTION IN PESTICIDE USE

Biotech plants are examined thoroughly for the safety of the environment. It is estimated that the global production status of biotech crops has increased from 1.7 to 190.4 million hectares with approximately 112-fold between the year 1996 and 2019 (International Service for the Acquisition of Agri-biotech Applications (ISAAA),

DOI: 10.1201/9781003597797-11

2019). Biotech crops were developed in such way that they show resistance to pest and herbicide tolerance, enhancing a good agricultural benefit and promoting environmentally friendly agriculture (Brookes & Barfoot, 2015). After the development of biotech plant about 36.95% pesticide application decreased. In addition, seeds of biotech plants contribute toward the maintenance of soil quality, conserve soil microorganism, and maintain carbon in soil. In a number of nations, including Uruguay, Argentina, and Paraguay, the field production of GM soybeans has reduced greenhouse gas emissions (Brookes & Barfoot, 2016). *Bacillus thuringiensis* (or BT) is used as a biological pesticide, a gram-positive soil-bacterium. During sporulation, many BT strains produce insecticidal proteins called δ-endotoxins, which leads to their use as insecticides. Recently GM crops using BT genes are developed such as BT corn or BT cotton. The primary goal of these biotech plants is to combat the European corn borer insect, which causes damage to maize crops, resulting in annual losses of up to $1 billion (Parekh, 2004).

However, *Pseudomonas syringae* is the causative agent of bacterial speck disease, after infection tomato productions decreased (Cai et al., 2011). The initial application of CRISPR/Cas9 resulted in the creation of the ortholog of the tomato mutant allele for downy mildew disease resistance in *Arabidopsis* 6 (DMR6), a positive regulator of downy mildew disease, through knockout in tomato. According to Paula de Toledo Thomazella et al. (2016), the mutant lines showed resistance to *Phytophthora capsici*, *Xanthomonas* species, and *P. syringae*. The majority of agricultural losses are caused by parasitic worms. The root systems of many plant species are their primary target. In essence, nematodes are worm species that can endure for many years in unfavorable soil conditions. Due of the significant environmental danger, chemical management of nematodes is prohibited. Crop rotation, the practice of growing different crops in the same area over successive seasons, is the most natural method to address this issue. However, it is not always practical due to the high costs involved (Lee, 2002). Therefore, the sole solution to the issue is to incorporate genes from nematode-resistant plants to the vulnerable plant. *Phytophthora palmivora* causes disease in papaya is a destructive cause of oomycete disease (Fosu-Nyarko & Jones, 2015).

A papaya mutant plant with resistance to *P. palmivora* was created following the insertion of a functional cysteine protease prohibitor (*PpalEPIC8*) (Gumtow et al., 2018). Similarly, using the CRISPR/Cas9 system, cocoa beans have gained resistance to *Phytophthora tropicalis*, another oomycete disease (Fister et al., 2018). The use of biotech in agriculture also maintains the variety of helpful insects that are not targets and this is due to lack of use of chemical to control harmful insect (Karalis et al., 2020; Talakayala et al., 2020). An analysis of the potential impact of Genetically Modified Plants (GMPs) on pollinators (Malone & Burgess, 2009) found that commercialized biotech plant with herbicide tolerance or insect resistance trait have no much more impact on pollinator. Recent studies performed in controlled conditions with honeybee, raise it by feeding Bt pollen or sugar syrup or honey containing purified *Cry1Ab* toxin has no adverse effect on adult survival and its larvae (Ramirez-Romero et al., 2008). Some experiments aim to develop the honeybee's hypopharyngeal gland (Babendreier et al., 2005).

11.3 LOWER HERBICIDE USE AND MORE EFFICIENT WEED MANAGEMENT

The likelihood and seriousness of either immediate or delayed negative impacts on the ecosystem, health of humans, and the farmer's bottom line are all included in the qualitative assessment of herbicide toxicity concerns. Nonetheless, a number of variables, including crop and characteristic, local weed species, agricultural practices techniques, and climate, are linked to the probability and intensity of each harmful impact (Madsen et al., 2002). The production of GM crops that are resistant to herbicides poses possible risks to both agricultural land and natural environments. Roundup®, a Monsanto chemical used in homes as well, is one of the broad-spectrum, low-toxicity herbicides that the HT crops exhibit resistance to. When included into maize, soybeans, and canola, this tolerance makes it easier for farmers to manage weeds (Velkov et al., 2005). In the absence of HT varieties, farmers are compelled to depend more on methods such as controlling weeds before crop emergence often through repeated soil tilling, which leads to erosion or by using more toxic "narrow-spectrum" chemicals that specifically target weeds without damaging post-emergent crops (Velkov et al., 2005).

Resistant (RT) weeds will unavoidably emerge due to the amazing capacity of weed species to gain resistance to herbicides derived from their gene pool (Agapito-Tenfen et al., 2014). Weeds can gain resistance to herbicides in as little as three years, as evidenced by the discovery of polygenic herbicide resistance in horseweed among F_1, F_2, and backcross progenies exposed to minimal doses of diclofop-methyl (Busi et al., 2013). Because herbicide-resistant crops are often grown and due to concurrent herbicide use, Asiatic dayflower (*Commelina communis* L.), wild buckwheat (*Polygonum convolvulus* L.), and common lambsquarters (*Chenopodium album* L.) were found to be more prominent in areas with significant selective pressure (Owen & Zelaya, 2005). Target site overproduction, changes in intracellular herbicide compartmentation, minimal herbicide absorbance and translocation, herbicide detoxification, and target site insensitivity are some of the mechanisms that may aid the plant in developing tolerance (Brower et al., 2012). Due to its chemical composition, lack of residual activity, mechanism of action, low glyphosate absorption by the roots of the plants from the soil, and almost negligible persistence of soil, resistance to glufosinate and glyphosate arising in the gene pool of weeds is extremely improbable (Baylis, 2000). Horseweed in the US and annual rye grass in Australia have been reported to acquire glyphosate resistance (Dale et al., 2002). Overexpression of the target enzyme, decreased herbicide transport, and varying glyphosate sensitivity are possible causes of this resistance development (Wakelin et al., 2004).

11.4 CONTRIBUTION TO CLIMATE CHANGE ADAPTATION

Agricultural practices contribute to approximately 25% of global greenhouse gas emissions through deforestation, inorganic fertilizers, and overgrazing (HM Treasury, 2009). Biofuels derived from conventional and biotech crops, such as

TABLE 11.1
Application of Biotech Crops in Different Areas

Applications		Reference
Crop improvement	• Increase productivity and economic advantages at the farmer level in a sustainable manner to help ensure food, feed, and fiber security and self-sufficiency, including more cheap food. • Play a significant part in raising productivity per hectare and lowering production costs due to the decreased requirement for inputs. • Cloning, embryo transfer, and artificial insemination are just a few of the many uses for agriculture-integrated biotechnology. • Crop productivity rose by 822 million tons, worth US$224.9 billion, between 1996 and 2018, with 86.9 million tons, worth US$18.9 billion, in 2018 alone.	Brookes and Barfoot (2020)
Biodiversity and land conservation	• Prevent deforestation and safeguard forest biodiversity. • As with conventional crops, biotech crops require attention to appropriate farming techniques, including rotation and resistance management. • Reduce the amount of land needed to raise food by assisting farmers in becoming more efficient. • Land-saving technology that can increase production on the 1.5 billion hectares of arable land those are now available.	
Pest control	• Using genetic engineering to create biopesticides and natural predators, ecologically friendly pest control techniques are being developed. • Breeding and releasing natural predators, encouraging conservation biological control, and lowering the need for broad-spectrum chemical pesticides all contribute to improved biological control. • Additionally, biotech crops reduced the usage of pesticides by 8.3% within that time frame and by 8.6% in 2018 alone.	
Climate change	• Reduce greenhouse gas emissions and contribute to climate change mitigation by providing tools for crop engineering that boost yield while using less energy. • Improves resilience to climate change.	
Green economy and society	• Boost income for small holder farmers. • Improves farm income. • Improves health condition due to reduced toxin.	

sugarcane, rapeseed, and jatropha can reduce CO_2 emissions from transportation (Sarin et al., 2007). GM crops lower fuel consumption by minimizing pesticide spraying and tillage, with IR varieties alone reducing CO_2 emissions by 962 million kg in 2005 (Brookes & Barfoot, 2008). Herbicide-resistant soybeans, covering 95% of no-till farmland in Argentina and the US, sequester 63,859 million tons of CO_2 by enhancing soil carbon storage (Brimner et al., 2005; Fawcett & Towery, 2004; Kleter et al., 2008). Additionally, biotechnology reduces nitrogen pollution by improving nitrogen-use efficiency in crops like GM canola, cutting N_2O emissions and fertilizer runoff (HM Treasury, 2009; Johnson et al., 2007). Innovations such as nitrogen-fixing cereal crops and enhanced *Rhizobium* strains further decrease reliance on synthetic fertilizers, supporting sustainable farming (Saikia & Jain, 2007; Zahran, 2001).

11.5 ENHANCED LAND USE EFFICIENCY AND CROP PRODUCTIVITY

The global population and their increasing demand for agricultural products place immense pressure on the agricultural system. Agricultural practices often face various problems like land degradation, water scarcity, pests and diseases, low production, and climate change. Soil erosion, nutrient depletion, excessive chemical fertilizers, and salinization can reduce land productivity which leads to decrease in crop production. Besides that, pests, diseases and inefficient irrigation practices, water shortage can limit the crop growth. These challenges can be addressed by the help of biotechnology to boost agricultural productivity (Tsatsakis et al., 2017). High-yielding varieties not only help in production but also increase biomass (Sexton & Zilberman, 2011). It also helps in decreasing the land conversion into agricultural fields as scientists get more products in a small land. By improving nutrient use efficiency, scientists can reduce the fertilizer requirement which becomes environment friendly. Many nanotech materials are developed for the gradual release and optimal dosage of water and plant nutrients, enhancing the efficiency of nutrient and water uptake (Hegde et al., 2007). China uses a variety of nanoparticles, including plastic-starch, humus-polyester, and clay-polyester, to slowly release nitrogen into wheat. Compared to applying chemical fertilizer, the yield increase from clay and plastic (nanomaterial coating) was approximately 4.5% (Govindasamy et al., 2023; Sharma et al., 2002). However, Table 11.1 showcases the diverse applications of biotech crops across various fields, such as agriculture, medicine, and industry.

11.6 CONCLUSION

By lowering CO_2 emissions, sequestering carbon, using less fuel, switching to environmentally friendly fuels, and using less artificial fertilizer, biotech crops can help mitigate and adapt to climate change. They can also improve food, feed, and fiber security by making food more affordable (lower prices), improving soil fertility, and increasing crop adaptability through the use of biofuels. These actions are intended to safeguard our environment from the negative consequences of climate change while simultaneously increasing agricultural output and food security. The scientific community agrees that both direct and indirect human activities are to blame for climate variability. In addition to improving yield and food security, a comprehensive strategy for the safe use of both traditional and contemporary agricultural biotechnologies will also make a substantial contribution to initiatives to lessen the effects of and prepare for climate change.

REFERENCES

Abdul Aziz, M., Brini, F., Rouached, H., & Masmoudi, K. (2022). Genetically engineered crops for sustainably enhanced food production systems. *Frontiers in Plant Science*, *13*, 1027828. https://doi.org/10.3389/fpls.2022.1027828

Agapito-Tenfen, S. Z., Vilperte, V., Benevenuto, R. F., Rover, C. M., Traavik, T. I., & Nodari, R. O. (2014). Effect of stacking insecticidal cry and herbicide tolerance epsps transgenes on transgenic maize proteome. *BMC Plant Biology*, *14*, 346. https://doi.org/10.1186/s12870-014-0346-8

Babendreier, D., Kalberer, N. M., Romeis, J., Fluri, P., Mulligan, E., & Bigler, F. (2005). Influence of Bt-transgenic pollen, Bt-toxin, and protease inhibitor (SBTI) ingestion on development of the hypopharyngeal glands in honeybees. *Apidologie*, *36*(4), 585–594. https://doi.org/10.1051/apido:2005042

Batista, R., Fonseca, C., Planchon, S., Negrão, S., Renaut, J., & Oliveira, M. M. (2017). Environmental stress is the major cause of transcriptomic and proteomic changes in GM and non-GM plants. *Scientific Reports*, *7*(1), 10624. https://doi.org/10.1038/s41598-017-09646-8

Bawa, A. S., & Anilakumar, K. R. (2013). Genetically modified foods: Safety, risks and public concerns—A review. *Journal of Food Science and Technology*, *50*(6), 1035–1046. https://doi.org/10.1007/s13197-012-0899-1

Baylis, A. D. (2000). Why glyphosate is a global herbicide: Strengths, weaknesses and prospects. *Pest Management Science*, *56*(4), 299–308. https://doi.org/10.1002/(SICI)1526-4998(200004)56:4<299::AID-PS144>3.0.CO;2-K

Borges, B. J. P., Arantes, O. M. N., Fernandes, A. A. R., Broach, J. R., & Fernandes, P. M. B. (2018). Genetically modified labeling policies: Moving forward or backward? *Frontiers in Bioengineering and Biotechnology*, *6*, 181. https://doi.org/10.3389/fbioe.2018.00181

Boyle, J. H., Dalgleish, H. J., & Puzey, J. R. (2019). Monarch butterfly and milkweed declines substantially predate the use of genetically modified crops. *Proceedings of the National Academy of Sciences*, *116*(8), 3006–3011. https://doi.org/10.1073/pnas.1811437116

Brimner, T. A., Gallivan, G. J., & Stephenson, G. R. (2005). Influence of herbicide-resistant canola on the environmental impact of weed management. *Pest Management Science*, *61*(1), 47–52. https://doi.org/10.1002/ps.967

Brookes, G., & Barfoot, P. (2008). Global impact of biotech crops: Socio-economic and environmental effects, 1996–2006. *AgBioForum*, *11*(1), 21–38.

Brookes, G., & Barfoot, P. (2015). Environmental impacts of genetically modified (GM) crop use 1996–2013: Impacts on pesticide use and carbon emissions. *GM Crops & Food*, *6*(2), 103–133.

Brookes, G., & Barfoot, P. (2016). Environmental impacts of genetically modified (GM) crop use 1996–2014: Impacts on pesticide use and carbon emissions. *GM Crops & Food*, *7*(2), 84–116.

Brookes, G., & Barfoot, P. (2020). Environmental impacts of genetically modified (GM) crop use 1996–2018: Impacts on pesticide use and carbon emissions. *GM Crops & Food*, *11*(4), 215–241. https://doi.org/10.1080/21645698.2020.1773198

Brower, L. P., Taylor, O. R., Williams, E. H., Slayback, D. A., Zubieta, R. R., & Ramirez, M. I. (2012). Decline of monarch butterflies overwintering in Mexico: Is the migratory phenomenon at risk? *Insect Conservation and Diversity*, *5*(2), 95–100. https://doi.org/10.1111/j.1752-4598.2011.00142.x

Busi, R., Neve, P., & Powles, S. (2013). Evolved polygenic herbicide resistance in *Lolium rigidum* by low-dose herbicide selection within standing genetic variation. *Evolutionary Applications*, *6*(2), 231–242. https://doi.org/10.1111/eva.12028

Cai, R., Lewis, J., Yan, S., Liu, H., Clarke, C. R., Campanile, F., & Vinatzer, B. A. (2011). The plant pathogen *Pseudomonas syringae* pv. tomato is genetically monomorphic and under strong selection to evade tomato immunity. *PLoS Pathogens*, *7*(8), e1002130. https://doi.org/10.1371/journal.ppat.1002130

Carrière, Y., Degain, B. A., & Tabashnik, B. E. (2021). Effects of gene flow between Bt and non-Bt plants in a seed mixture of Cry1A.105 + Cry2Ab corn on performance of corn earworm in Arizona. *Pest Management Science*, *77*(4), 2106–2113. https://doi.org/10.1002/ps.6240

Dale, P. J., Clarke, B., & Fontes, E. M. (2002). Potential for the environmental impact of transgenic crops. *Nature Biotechnology*, *20*(6), 567–574. https://doi.org/10.1038/nbt0602-567

European Commission. (2010). *A decade of EU-funded GMO research 2001–2010*. Publications Office of the European Union. https://op.europa.eu/en/publication-detail/-/publication/d1be9ff9-f3fa-4f3c-86a5-beb0882e0e65

Fawcett, R., & Towery, D. (2004). *Conservation tillage and plant biotechnology: How new technologies can improve the environment by reducing the need to plow*. Conservation Technology Information Center. https://www.worldfoodprize.org/documents/filelibrary/images/borlaug_dialogue/2002/transcripts/towery_transcript_DD615340B770A.pdf

Fernandes, G. B., Silva, A. C. D. L., Maronhas, M. E. S., Santos, A. D. S. D., & Lima, P. H. C. (2022). Transgene flow: Challenges to the on-farm conservation of maize landraces in the Brazilian semi-arid region. *Plants*, *11*(5), 603. https://doi.org/10.3390/plants11050603

Fister, A. S., Landherr, L., Maximova, S. N., & Guiltinan, M. J. (2018). Transient expression of CRISPR/Cas9 machinery targeting TcNPR3 enhances defense response in *Theobroma cacao*. *Frontiers in Plant Science*, *9*, 268. https://doi.org/10.3389/fpls.2018.00268

Fitzpatrick, S. W., & Reid, B. N. (2019). Does gene flow aggravate or alleviate maladaptation to environmental stress in small populations? *Evolutionary Applications*, *12*(7), 1402–1416. https://doi.org/10.1111/eva.12768

Fosu-Nyarko, J., & Jones, M. G. (2015). Application of biotechnology for nematode control in crop plants. *Advances in Botanical Research*, *73*, 339–376. https://doi.org/10.1016/bs.abr.2015.01.001

Govindasamy, P., Muthusamy, S. K., Bagavathiannan, M., Mowrer, J., Jagannadham, P. T. K., Maity, A., & Tiwari, G. (2023). Nitrogen use efficiency—A key to enhance crop productivity under a changing climate. *Frontiers in Plant Science*, *14*, 1121073. https://doi.org/10.3389/fpls.2023.1121073

Gumtow, R., Wu, D., Uchida, J., & Tian, M. (2018). A *Phytophthora palmivora* extracellular cystatin-like protease inhibitor targets papain to contribute to virulence on papaya. *Molecular Plant-Microbe Interactions*, *31*(3), 363–373. https://doi.org/10.1094/MPMI-06-17-0131-FI

Hegde, D. M., Babu, S. N., Qureshi, A. A., & Murthy, I. Y. L. N. (2007). Enhancing nutrient-use efficiency in crop production—A review. *Indian Journal of Agronomy*, *52*(4), 261–274.

HM Treasury. (2009). *Green biotechnology and climate change*. EuroBio. http://www.docstoc.com/docs/15021072/Green-Biotechnology-and-Climate-Change

International Service for the Acquisition of Agri-biotech Applications (ISAAA). (2019). *Biotech crop highlights in 2019*. https://www.isaaa.org/resources/publications/pocketk/16/

Johnson, J. M. F., Franzluebbers, A. J., Weyers, S. L., & Reicosky, D. C. (2007). Agricultural opportunities to mitigate greenhouse gas emissions. *Environmental Pollution*, *150*(1), 107–124. https://doi.org/10.1016/j.envpol.2007.06.030

Karalis, D. T., Karalis, T., Karalis, S., & Kleisiari, A. S. (2020). Genetically modified products, perspectives and challenges. *Cureus*, *12*(3), e7306. https://doi.org/10.7759/cureus.7306

Kennedy, I. R., & Tchan, Y. T. (1992). Biological nitrogen fixation in non-leguminous field crops: Recent advances. *Plant and Soil*, *141*(1–2), 93–118. https://doi.org/10.1007/BF00011313

Kleter, G. A., Harris, C., Stephenson, G., & Unsworth, J. (2008). Comparison of herbicide regimes and the associated potential environmental effects of glyphosate-resistant crops versus what they replace in Europe. *Pest Management Science*, *64*(4), 479–488. https://doi.org/10.1002/ps.1513

Kumar, K., Gambhir, G., Dass, A., Tripathi, A. K., Singh, A., Jha, A. K., & Rakshit, S. (2020). Genetically modified crops: Current status and future prospects. *Planta*, *251*(4), 91. https://doi.org/10.1007/s00425-020-03372-8

Kurup, V. M., & Thomas, J. (2020). Edible vaccines: Promises and challenges. *Molecular Biotechnology*, *62*(2), 79–90. https://doi.org/10.1007/s12033-019-00222-1

Lee, D. L. (Ed.). (2002). *The biology of nematodes*. CRC Press. https://doi.org/10.1201/b12614

Madsen, K. H., Valverde, B. E., & Jensen, J. E. (2002). Risk assessment of herbicide-resistant crops: A Latin American perspective using rice (*Oryza sativa*) as a model. *Weed Technology*, *16*(1), 215–223. https://doi.org/10.1614/0890-037X(2002)016[0215:RAOHRC]2.0.CO;2

Malone, L. A., & Burgess, E. P. J. (2009). Impact of genetically modified crops on pollinators. In N. Ferry & A. M. R. Gatehouse (Eds.), *Environmental impact of genetically modified crops* (pp. 199–222). CABI.

Nishizawa, T., Tamaoki, M., Aono, M., Kubo, A., Saji, H., & Nakajima, N. (2010). Rapeseed species and environmental concerns related to loss of seeds of genetically modified oilseed rape in Japan. *GM Crops*, *1*(3), 143–156. https://doi.org/10.4161/gmcr.1.3.12761

Owen, M. D., & Zelaya, I. A. (2005). Herbicide-resistant crops and weed resistance to herbicides. *Pest Management Science*, *61*(3), 301–311. https://doi.org/10.1002/ps.1015

Parekh, S. R. (Ed.). (2004). *The GMO handbook: Genetically modified animals, microbes, and plants in biotechnology*. Humana Press.

Paula de Toledo Thomazella, D., Brail, Q., Dahlbeck, D., & Staskawicz, B. (2016). CRISPR-Cas9 mediated mutagenesis of a DMR6 ortholog in tomato confers broad-spectrum disease resistance. *bioRxiv*, 064824. https://doi.org/10.1101/064824

Ramirez-Romero, R., Desneux, N., Decourtye, A., Chaffiol, A., & Pham-Delègue, M. H. (2008). Does Cry1Ab protein affect learning performances of the honey bee *Apis mellifera* L. (Hymenoptera, Apidae)? *Ecotoxicology and Environmental Safety*, *70*(2), 327–333. https://doi.org/10.1016/j.ecoenv.2007.12.002

Roberts, A., Boeckman, C. J., Mühl, M., Romeis, J., Teem, J. L., Valicente, F. H., & Hellmich, R. L. (2020). Sublethal endpoints in non-target organism testing for insect-active GE crops. *Frontiers in Bioengineering and Biotechnology*, *8*, 556. https://doi.org/10.3389/fbioe.2020.00556

Saikia, S. P., & Jain, V. (2007). Biological nitrogen fixation with non-legumes: An achievable target or a dogma? *Current Science*, *92*(3), 317–322.

Sarin, R., Sharma, M., Sinharay, S., & Malhotra, R. K. (2007). Jatropha–palm biodiesel blends: An optimum mix for Asia. *Fuel*, *86*(10–11), 1365–1371. https://doi.org/10.1016/j.fuel.2006.11.040

Sexton, S., & Zilberman, D. (2011). Land for food and fuel production: The role of agricultural biotechnology. In J. L. Lusk, J. Roosen, & J. F. Shogren (Eds.), *The intended and unintended effects of U.S. agricultural and biotechnology policies* (pp. 269–288). University of Chicago Press.

Sharma, A., Kumar, V., Shahzad, B., Tanveer, M., Sidhu, G. P. S., Handa, N., & Thukral, A. K. (2019). Worldwide pesticide usage and its impacts on ecosystem. *SN Applied Sciences*, *1*(11), 1446. https://doi.org/10.1007/s42452-019-1485-1

Sharma, H. C., Crouch, J. H., Sharma, K. K., Seetharama, N., & Hash, C. T. (2002). Applications of biotechnology for crop improvement: Prospects and constraints. *Plant Science*, *163*(3), 381–395. https://doi.org/10.1016/S0168-9452(02)00133-4

Talakayala, A., Katta, S., & Garladinne, M. (2020). Genetic engineering of crops for insect resistance: An overview. *Journal of Biosciences*, *45*(1), 114. https://doi.org/10.1007/s12038-020-00082-x

Tsatsakis, A. M., Nawaz, M. A., Kouretas, D., Balias, G., Savolainen, K., Tutelyan, V. A., & Chung, G. (2017). Environmental impacts of genetically modified plants: A review. *Environmental Research*, *156*, 818–833. https://doi.org/10.1016/j.envres.2017.03.011

Vega Rodríguez, A., Rodríguez-Oramas, C., Sanjuán Velázquez, E., Hardisson de la Torre, A., Rubio Armendáriz, C., & Carrascosa Iruzubieta, C. (2022). Myths and realities about genetically modified food: A risk-benefit analysis. *Applied Sciences*, *12*(6), 2861. https://doi.org/10.3390/app12062861

Velkov, V. V., Medvinsky, A. B., Sokolov, M. S., & Marchenko, A. I. (2005). Will transgenic plants adversely affect the environment? *Journal of Biosciences*, *30*(4), 515–548. https://doi.org/10.1007/BF02703723

Wakelin, A. M., Lorraine-Colwill, D. F., & Preston, C. (2004). Glyphosate resistance in four different populations of *Lolium rigidum* is associated with reduced translocation of glyphosate to meristematic zones. *Weed Research*, *44*(6), 453–459. https://doi.org/10.1111/j.1365-3180.2004.00421.x

Wunderlich, S., & Gatto, K. A. (2015). Consumer perception of genetically modified organisms and sources of information. *Advances in Nutrition*, *6*(6), 842–851. https://doi.org/10.3945/an.115.008870

Zahran, H. H. (2001). Rhizobia from wild legumes: Diversity, taxonomy, ecology, nitrogen fixation and biotechnology. *Journal of Biotechnology*, *91*(2–3), 143–153. https://doi.org/10.1016/S0168-1656(01)00342-X

12 Biotechnology and Smallholder Farmers

Navigating Challenges and Unlocking Economic Pathways

Subrat Pattanaik, Arati Priyadarshini,
and Basanta Kumar Samal

12.1 INTRODUCTION

Agriculture has been the backbone for human civilization that has sustained livelihoods and ensured food security for centuries for the burgeoning population. The smallholder farmers are the key contributors to agriculture, who form the backbone of a country's agricultural systems as well as strengthening nation's economy (Pattanaik & Priyadarshini, 2024). These little farmers generally operate lands of less than two hectares. They contribute significantly to local economic stability and global food security, with about 80% of the food in large parts of the developing world attributed to their efforts (Food and Agriculture Organization of the United Nations (FAO, 2017). Yet, the issues faced by the smallholder farmers are recurrent and multiple, which involve restricted availability of quality inputs, susceptibility to climatic unpredictability, pest and disease infestations, and limited market prospects. To address these issues biotechnology has arisen as a vital tool for tackling these difficulties (Pattanaik & Priyadarshini, 2024).

The developments in genetic engineering, molecular biology, and bioinformatics have led to innovative solutions, including genetically modified (GM) crops, biofortified varieties, and precision agriculture tools. Such features as pest and drought resistance as well as improved nutrition are made possible by these technologies, which will be able to minimize chemical applications, stabilize yield, and even make crops more resilient to climatic change (FAO, 2017). Take for instance Bt cotton and herbicide-tolerant soybean, they have resulted in significant productivity gains, coupled with significant reductions in pesticide applications (Brookes & Barfoot, 2015). Similarly, with biofortification crops like vitamin A-enhanced golden rice, there may be a significant route to remedy critical nutritional inadequacies among vulnerable populations. Apart from this, biofortified crops advance innovation to fight some of the severe nutritional deficiencies, yet it provides a limited and partial pathway out of malnutrition. Adoption of biotechnology, however, raises concerns relating to equity, accessibility, and distributive effects. It also is still somewhat uneven because its

DOI: 10.1201/9781003597797-12

application by smallholder farmers encounters significant upfront cost barriers, limited technical knowledge, widespread socio-cultural resistance, and highly restrictive regulatory frameworks. Additionally, intellectual property rights and dependency on biotech seed suppliers further constrain accessibility (Azadi et al., 2016).

Agroecological, climatic, demographic, economic, and cultural factors all have an impact in the decisions that farmers make which crops and varieties to grow (Skarbø, 2014). The challenge here is in the creation of inclusive policies, participatory research, and capacity-building initiatives toward equitable access to biotechnological innovations. Agricultural biotechnology has been a next step in evolving traditional breeding practice, improving crop improvement in speed, precision, and reliability (Brookes & Barfoot, 2017). This dynamic approach doesn't just tackle food insecurity, defined as insufficient physical, social, or economic access to safe and nutritious food, but also provides economics for smallholders (Thompson et al., 2007). GM crops can resist certain pests and diseases, thus increasing the savings from agrochemicals. In turn, drought-resistant varieties guarantee yield stability if water is scarce. This, besides synthetic biology and bio manufacturing developments, also helps smallholder farmers to tap into high-value biomaterial and biobased product markets, diversifying their sources of income (Berman et al., 2013). This chapter explores the interaction between biotechnology and smallholders farming, which can empower farmers, improve global food security, and promote sustainable agriculture.

12.2 MULTIDIMENSIONAL PERSPECTIVES ON GM CROPS

Figure 12.1 about multidimensional perspectives on GM crops highlights the dual role of GM crops either in exacerbating challenges or unlocking economic and sustainability pathways. To smallholder farmers, GM crops bring increased yields, resilience to climate change, and economic empowerment, but the associated challenges, such as market inequities, high costs, and ethical concerns must be addressed to ensure equitable benefits (Food and Agriculture Organization of the United Nations (FAO), 2016). The diagram is a holistic framework that addresses the multidimensional aspects of GM crops, emphasizing their implications on smallholder farmers, the environment, and society. It broadly divides the discussion into three main dimensions: farm community, environmental, and social-economic dimensions. The three perspectives combined bring forth the challenges and opportunities of GM crops, hence a call to strike a balance between policy and inclusion to support the smallholder farmers. On the other side, the diagram presents several challenges of GM crops. Economically, small-scale farmers are mostly dependent on multinational companies for quality seeds; hence, the financial freedom of such farmers is limited. Their access to the market can be limited by the intellectual property rights and trade barriers (Pattanaik & Priyadarshini, 2024).

Environmentally, unintended ecological effects from GM crops can include loss of biodiversity, development of resistance in pests or weeds, and long-term negative impacts on the health of soils (Qaim, 2020). It generates unintended ecological impacts such as the loss of biodiversity, the emergence of pest and weed resistance, and the deterioration of the long-term health of soils. Societal issues involved are

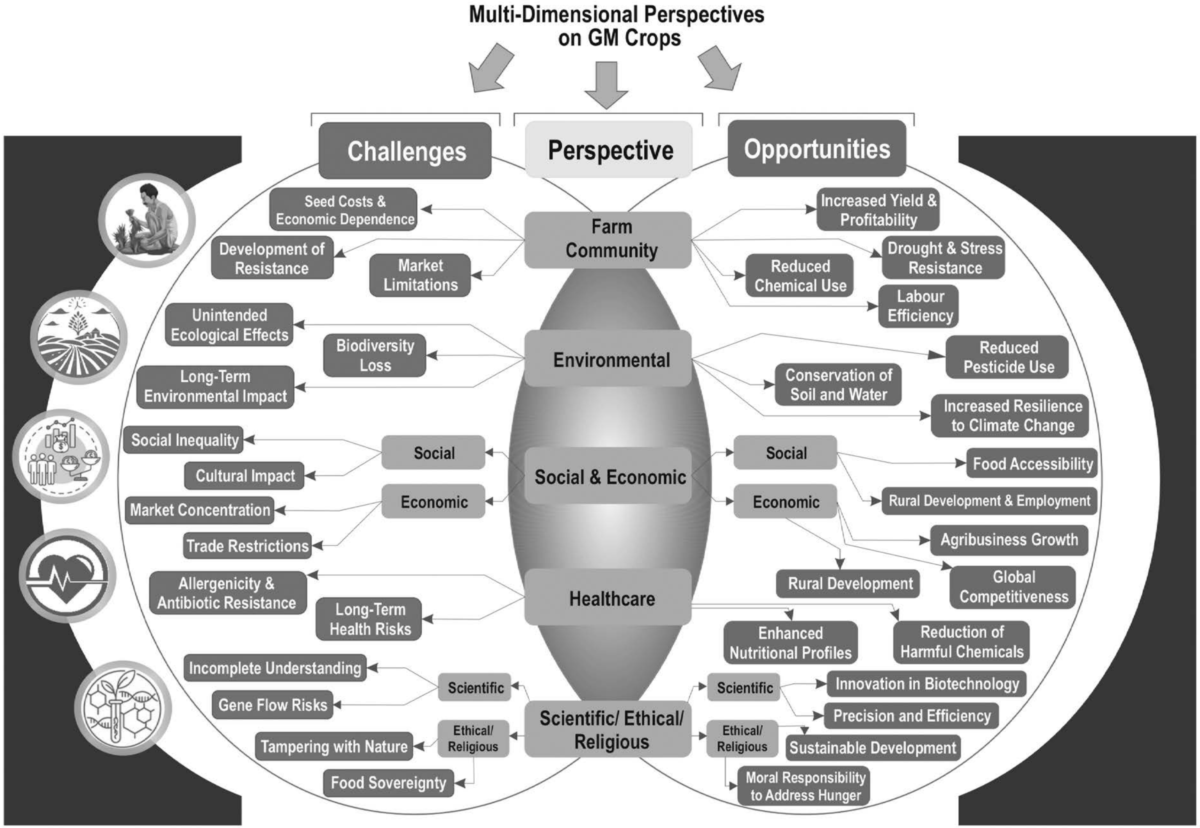


FIGURE 12.1 Multidimensional perspectives on GM crops.

uneven technology access, the cultural opposition toward GMs, and social inequity. Besides these, scientific and ethical issues have emerged as follows: gene flow to non-GM crops, the "playing with nature" controversy, and insufficient understanding of the long-term impact (Hilbeck et al., 2015). On the other hand, the diagram illustrates the considerable opportunities GM crops have provided. The economic gains have increased productivity, improved profitability, and generated rural development through employment generation and international competitiveness for smallholder farmers. GM technologies decrease chemical inputs, increase soil and water conservation, and produce drought-and heat-tolerant crops sensitive to those climatic factors (Ruggieri et al., 2021).

Socially, they have promoted access to more food, efficiency in labor force, and increased poverty alleviation in rural economies. Scientific discoveries such as biofortified crops created with improved nutrient profiles and precision of genetic engineering shall bring biotechnology in line with long-term goals of sustainability as well as challenges posed by the world hunger agenda (Hilbeck et al., 2015). The potentials related to biotechnology integration by smallholder agriculture include addressing productivity, sustainability, and resilience-related challenges. However, critical barriers lie ahead for smallholder farmers to be able to leverage such advances. These are largely economically constrained, faced with regulatory hurdles, and embedded in socio-cultural dynamics that do not permit easy access to technology (Labeyrie et al., 2021). The constraints that challenge them include high costs, credit facility unavailability, and perceptions of risk in farm activities among the small-scale farmers. The economic burden is therefore explored, along with its effect on the smallholders.

12.3 ECONOMIC BARRIERS IN BIOTECH CROPS ADOPTION IN SMALLHOLDER AGRICULTURE

The diagram below (Figure 12.2) shows the constraints to biotechnology adoption, particularly on three major domains: initial high costs, access to credit, and risk perception. Each of these domains is challenging in a different way, especially for smallholder farmers and other stakeholders operating in resource-limited settings. The high initial costs associated with the adoption of biotechnology create considerable financial burdens on potential users. This consists of high-pricing on the genetically modified (GM) seeds, most are considerably pricey than other types of conventional seeds because the values added with genetic engineering. Acquisition of GM seeds by the smallholders sometimes redirects the crucial operations funds, making them unaffordable in the long run (Raney & Pingali, 2007). Besides this, the cost of upgrading irrigation could be high for drought-resistant crops and specialized storage facilities to ensure seed quality. GM seeds raise issues related to intellectual property rights: farmers are being legally bonded to pay a license fee every year or forbidden from reusing the seeds. This makes it even harder on the financial front, adding input costs which may include fertilizers and pesticides used for GM crops, putting one in a cycle of expensive inputs (Pattanaik & Priyadarshini, 2024).

The second barrier is access to financial resources, which is a pre-requisite for smallholder farmers to adopt biotechnology. Unfortunately, systemic problems in

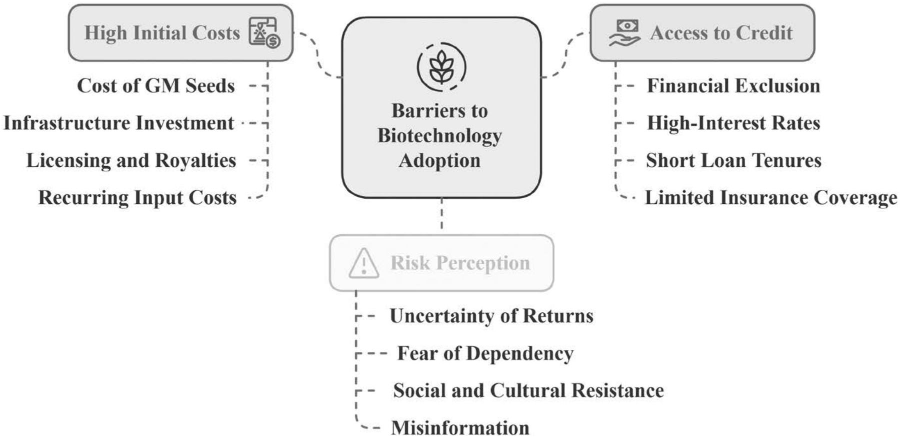


FIGURE 12.2 Economic barriers in adoption of biotech crops in smallholder agriculture.

rural financial systems restrict access to the much-needed credit. Many farmers suffer from financial exclusion: they lack access to formal banking systems or microfinance institutions (Klapper et al., 2019). Even those who are able to access them face very high interest rates on loans that discourage borrowing, making it economically unviable to finance costly biotechnology projects. Yet another problem with short tenors of loans is that investments in biotechnology demand long-term financial commitment to realize their full benefits. For example, the yields from drought-resistant crops can only stabilize in subsequent seasons, thereby making it very difficult to repay the loan. In addition, crop failure or any other environmental effects not covered by insurance increase the financial risks involved, which scares farmers away from adopting biotechnology (Ceballos et al., 2019).

The third barrier is risk perception, which relates to psychological and societal factors influencing decisions. Such risks, usually driven by misinformation and socio-cultural resistance, prevent farmers from adopting what could be useful innovations (Adenle et al., 2017). The returns on investment in biotechnology are usually uncertain for farmers due to the lack of clear evidence or past experiences. Smallholders are unlikely to invest much in biotechnology due to changing climatic weather, volatile price in the marketplace, and that the technology does not seem too familiar (Pattanaik & Priyadarshini, 2024). Hence, the associated uncertainty may deter them from availing the various benefits of technology adoption. However, this may be further supplemented by the belief that they depend on biotech companies for obtaining seeds and supplying inputs. Restrictive legal practices on seed saving and replanting further fears this transition into new practices since farmers are often afraid to alter their traditional customs (Ceballos et al., 2019).

Social resistance and cultural values play critical roles here since traditional societies might be against or dislike the introduction of biotechnologies. Deep-seated cultural feelings and a traditional form of farming and agriculture can become resistance to innovations in farming and agricultural technology (Adenle et al., 2017). A lack of scientific literacy combined with misinformation concerning perceived environmental and health impacts related to GM crops fuels fears. In many cases, misinformation is diffused through social channels, cementing negative impressions (Kikulwe et al., 2011).

12.4 CRUCIAL ROLES OF BIOTECH CROPS

Agricultural biotechnology represents an innovation with pivotal roles for offering transformational solutions to some of the most serious problems faced by farmers around the world. Biotech crops will likely improve the productivity of smallholder farmers in resource-constrained environments, reducing crop losses and making them more resilient to climate variability (International Service for the Acquisition of Agri-biotech Applications (ISAAA, 2019). Biotechnology will be able to produce varieties that yield more as well as can resist pests and diseases and weather conditions. In Figure 12.3, biotech crops enhance crop productivity by developing varieties with improved traits that optimize the use of available resources. This becomes very important for smallholder farmers who operate on limited land and are resource-constrained. It offers farmers a range of tools to enhance crop productivity, including genetically modified (GM), molecular breeding, and advanced tissue culture techniques. Genetically modified or GM crops, including improved varieties of high-yield maize and rice strains, optimize crop output through gains in grain. This can include increased grain with better photosynthesis and more-efficient nutrient uptake; for example, improved nitrogen-use-efficiency reduces use of chemical fertilizer, reducing production costs but maintaining yields. Again, shorter maturities enhance the opportunity to make more plantings per calendar year. This is especially advantageous in areas with limited growing seasons based on climatic

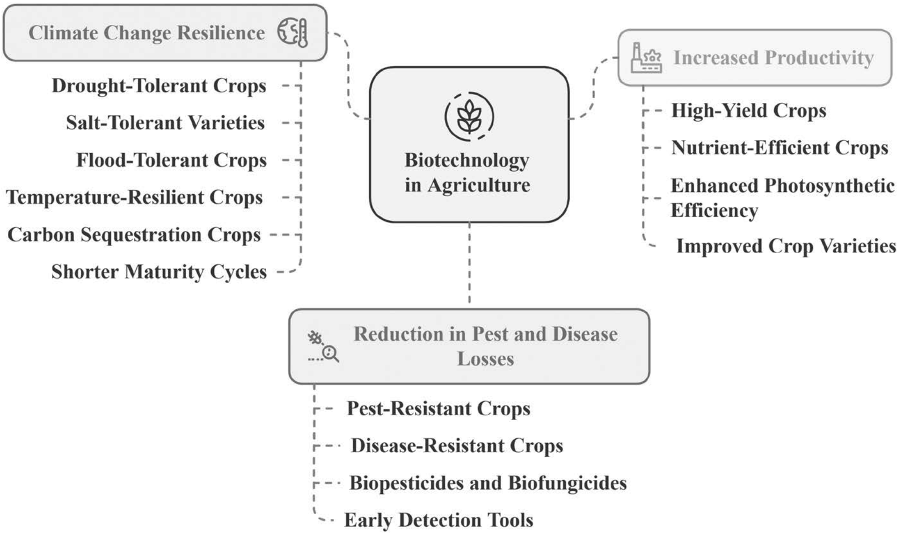


FIGURE 12.3 Crucial roles of biotech crops for adoption by smallholder agriculture.

factors. Crops can be developed that require fewer nutrients or can use the soil nutrients available in a more efficient manner.

For example, nitrogen-use-efficient varieties decrease the reliance on chemical fertilizers, thereby decreasing the cost and increasing the yield. Biotechnology has been enhancing the ability of crops to utilize sunlight more effectively, which in turn enhances the growth rates and productivity. Marker-assisted selection accelerates the breeding process, and the high-performing varieties can be developed in a much shorter period. These varieties are region- and climate-specific, thus ensuring better adaptation and higher productivity (Friedrich et al., 2021). Biotech crops make farming easier. For example, herbicide-tolerant soybean allows efficient weed control with minimal labor, enabling farmers to allocate resources more effectively. Crop losses by pests and diseases are still an important challenge for agriculture, and especially for smallholders who cannot access pest management tools. Pest-resistant and disease-tolerant crops through biotechnology offer the sustainable solution. Genetically engineered crops include Bt cotton and Bt maize, which contain pest-repelling traits against specific insect pests such as bollworms and stem borers. These crops decrease the dependency on chemical pesticides, reduce production costs, and preserve the environment. Through advances in genetic engineering, crops that resist viral, bacterial, and fungal pathogens are available. An example of these include GM bananas that are resistant to Panama disease, while GM papayas resist the papaya ringspot virus and help limit losses that accrue during losses (Friedrich et al., 2021).

Biopesticides and bio fungicides, through biotechnological processes, have provided an environment-friendly approach to pest and disease control. Natural products reduce the risks associated with chemical pesticides. Molecular diagnostic tools help in the early detection of pests and diseases, allowing farmers to take appropriate measures before the outbreak becomes a serious issue (Friedrich et al., 2021). Table 12.1 shows some of the key importance of genetically modified crops: GM crops play the most vital role in solving world agricultural issues. This table

TABLE 12.1

Overview of Genetically Modified (GM) Crops: Addressing Agricultural Challenges, Economic Impacts, and Global Adoption Patterns

Sl. No.	Genetically Modified (GM) Crops	Challenges Addressed	Economic Impacts	Countries Adopted	References
1.	Bt cotton	To resist bollworm infestations, leading to reduced pesticide use and increased yields	24% increase in cotton yield per acre and a 50% gain in profit among smallholders in India. An average pesticide reduction of around 56% and a yield increase of approximately 8%	India, China, and several African countries	Kathage and Qaim (2012) Wang et al. (2013) Nnaemeka et al. (2023)

TABLE 12.1 (*Continued*)
Overview of Genetically Modified (GM) Crops: Addressing Agricultural Challenges, Economic Impacts, and Global Adoption Patterns

Sl. No.	Genetically Modified (GM) Crops	Challenges Addressed	Economic Impacts	Countries Adopted	References
2	Herbicide-tolerant soybeans	Control weeds better, improving crop management and productivity	15% increase in net income per hectare and 5–10% higher productivity reported by Argentinean and US farmers	The USA, Brazil, Argentina, Canada	Brookes and Barfoot (2020)
3.	Drought-tolerant maize	Withstand water scarcity, ensuring food security in arid regions	25% yield boost in water-stressed conditions reported by Kenyan farmers, enhancing food security and income	Kenya, Uganda, South Africa, Zimbabwe, and Mozambique	Prasanna et al. (2021)
4.	Virus-resistant papaya	To combat papaya ringspot virus (PRSV)	20–30% increase in yields and reduced losses from viral infections, saving approximately $17 million annually	The USA (Hawaii), China, the Philippines	Gonsalves (2006)
5.	Bt maize	Resistant to stem borers, leading to increased maize production	Farmers in South Africa saw a 30% yield increase, and the USA saved $1 billion on pesticides	The USA, South Africa, Spain, Brazil, Argentina	Bennett et al. (2003)
6.	Late blight-resistant potato	Combating fungus causing late blight	30% yield increase and an 85% reduction in fungicide use were reported by the farmers	Bangladesh, Indonesia (trials)	Ghislain et al. (2019)
7.	Herbicide-tolerant sugar beet	Facilitates effective weed management	10–15% yield increase and a 40% reduction in herbicide costs was reported by US farmers	The USA, Canada	Kniss (2010)
8.	Insect-resistant chickpea	Developed to combat pod borer in India	30% yield increase and a 60% reduction in pesticide costs was reported from trials in India	India (trials), Australia (research stage)	Varshney et al. (2019)
9.	Drought-tolerant wheat	Under development to ensure yields in water-limited environments	10–20% increase in yields under drought stress was reported by Australian farmers	Australia, Argentina, the USA (trials)	Reynolds and Langridge (2016)
10.	Salt-tolerant rice	Engineered to grow in saline soils, benefiting farmers in coastal regions	15–20% increase in yields on saline soils was reported by Bangladeshi and Indian farmers	Bangladesh, India, the Philippines (trials)	Hossain et al. (2015)

describes the inputs that increase productivity, resist pests and diseases, tolerate drought and salinity, provide nutritional content, and input the high-yielding varieties. Additionally, from the table below, it depicts that GM crops have been broadly adopted in numerous geographies.

12.5 BIOTECHNOLOGY FOR SMALLHOLDERS BY POLICY AND INSTITUTIONAL SUPPORT

Policy and institutional support for these smallholder farmers would make a difference. Reducing the economic burden is quite possible through different strategies. It can be initiated by providing direct subsidies or seeds at discounted rates or by running targeted voucher programs to increase availability of GM seed. Special initiatives toward marginalized regions and underprivileged farming communities will ensure that such schemes reach every corner of society (Friedrich et al., 2021). Subsidized biotech seeds reduce the upfront investment and minimize long-term input costs by reducing dependence on chemical fertilizers and pesticides, given traits such as pest and drought resistance. Farmer education and demonstration plots enhance the knowledge that farmers have to appreciate the added value and wise use of their biotech crop (Friedrich et al., 2021).

Digital arenas and mobile devices can be additionally used to strengthen extension services toward wider areas: immediate feedback by best management, weather information, and market orientation. The entire support system is aimed at maximizing the productivity and profitability of biotech adoption. An inclusive regulatory framework would build much-needed confidence and encourage adoption. Eliminating bottlenecks in the approval process for GM crops with firm standards, for example, can ease environmental and health concerns (Pray et al., 2006). Permissive policy on seed saving, low licensing fees, and open intellectual property rights reduce farmer dependence on large biotech firms. More harmonization of rules across borders will improve access to biotech products in regional markets (Kikulwe, 2011).

12.6 CONCLUSION

Biotechnology offers transformative solutions to the challenges of low productivity, and pest and disease losses, and climate change impacts for smallholder farmers. It yields new and improved varieties, both genetically modified crops and biofortified varieties, which can enhance productivity, improve livelihoods, and promote sustainable farming practices. There are, however, current constraints to its greater adoption, hence calling for a coordinated approach to properly unleash the potential of biotechnology. Subsidies, financial incentives, and investments in infrastructure can help make biotechnological tools more accessible and economically viable. Strengthened extension services could bridge the knowledge gap to give farmers skills that would make these innovations available to them for productive use. Public-private partnerships in research and development, supported by policies on safety, equity, and affordability, can leverage biotechnology to empower smallholder farmers, improve food security, climate resilience, and rural development, aligning with SDGs like zero hunger and climate action for inclusive and sustainable agricultural systems.

REFERENCES

Adenle, A. A., Wedig, K., & Azadi, H. (2017). Sustainable agriculture and food security in Africa: The role of innovative technologies and international organizations. *Sustainability*, *9*(2), 231. https://doi.org/10.3390/su9020231

Azadi, H., Samiee, A., Mahmoudi, H., Jouzi, Z., Rafiaani Khachak, P., De Maeyer, P., & Witlox, F. (2016). Genetically modified crops and small-scale farmers: Main opportunities and challenges. *Critical Reviews in Biotechnology*, *36*(3), 434–446. https://doi.org/10.3109/07388551.2014.990413

Bennett, R., Buthelezi, T. J., Ismael, Y., & Morse, S. (2003). Bt Cotton, Pesticides, Labour and Health: A case study of smallholder farmers in the Makhathini flats, Republic of South Africa. *Outlook on Agriculture*, *32*(2), 123–128. https://doi.org/10.5367/000000003101294361

Berman, J., Zhu, C., Pérez-Massot, E., Arjó, G., Zorrilla-López, U., Masip, G., Banakar, R., Sanahuja, G., Farré, G., Miralpeix, B., Bai, C., Vamvaka, E., Sabalza, M., Twyman, R. M., Bassié, L., Capell, T., & Christou, P. (2013). Can the world afford to ignore biotechnology solutions that address food insecurity? *Plant Molecular Biology, 83*(1–2), 5–19. https://doi.org/10.1007/s11103-013-0027-2

Brookes, G., & Barfoot, P. (2015). Environmental impacts of genetically modified (GM) crop use 1996–2013: Impacts on pesticide use and carbon emissions. *GM Crops and Food*, *6*(2), 103–133. https://doi.org/10.1080/21645698.2015.1025193

Brookes, G., & Barfoot, P. (2017). Environmental impacts of genetically modified (GM) crop use 1996–2015: Impacts on pesticide use and carbon emissions. *GM Crops and Food*, *8*(2), 117–147. https://doi.org/10.1080/21645698.2017.1309490

Brookes, G., & Barfoot, P. (2020). Environmental impacts of genetically modified (GM) crop use 1996-2018: Impacts on pesticide use and carbon emissions. *GM Crops and Food*, *11*(4), 215–241. https://doi.org/10.1080/21645698.2020.1773198

Ceballos, F., Kramer, B., & Robles, M. (2019). The feasibility of picture-based insurance (PBI): Smartphone pictures for affordable crop insurance. *Development Engineering*, *4*, 100042. https://doi.org/10.1016/j.deveng.2019.100042

Food and Agriculture Organization of the United Nations (FAO). (2016). *The state of food and agriculture: Climate change, agriculture and food security*. https://www.fao.org/publications/fao-flagship-publications/the-state-of-food-and-agriculture/en

Food and Agriculture Organization of the United Nations (FAO). (2017). *The future of food and agriculture*. https://openknowledge.fao.org/server/api/core/bitstreams/2e90c833-8e84-46f2-a675-ea2d7afa4e24/content

Friedrich, H., Lauxmann, M., & Zurbriggen, M. D. (2021). Biotechnological approaches for crop improvement: Harnessing genetic diversity. *Trends in Plant Science*, *26*(5), 415–429. https://doi.org/10.1016/j.tplants.2020.12.006

Ghislain, M., Byarugaba, A. A., Kiggundu, A., Njoroge, K., Magembe, E., & Barekye, A. (2019). Biosafety and agronomic performance of late blight-resistant potatoes in field trials of Bangladesh. *Frontiers in Bioengineering and Biotechnology*, *7*, 348. https://doi.org/10.3389/fbioe.2019.00348

Gonsalves, D. (2006). Transgenic papaya: Development, release, impact and challenges. *Advances in Virus Research*, *67*, 317–354. https://doi.org/10.1016/S0065-3527(06)67009-7

Hilbeck, A., Meier, M., Römbke, J., Jänsch, S., Teichmann, H., & Tappeser, B. (2015). A controversy re-visited: Is the coccinellid *Adalia bipunctata* adversely affected by Bt toxins? *Environmental Sciences Europe*, *27*(1), 1–12. https://doi.org/10.1186/s12302-015-0043-8

Hossain, M., Deb, U. K., & Jaim, W. M. H. (2015). Economic impact of salt-tolerant rice in coastal areas of Bangladesh. *Agricultural Economics*, *46*(5), 563–578. https://doi.org/10.1111/agec.12178

International Service for the Acquisition of Agri-biotech Applications (ISAAA). (2019). *Global status of commercialized biotech/GM crops: 2019* (ISAAA Brief No. 55). https://www.isaaa.org/resources/publications/briefs/55

Kathage, J., & Qaim, M. (2012). Economic impacts and impact dynamics of Bt (*Bacillus thuringiensis*) cotton in India. *Proceedings of the National Academy of Sciences*, *109*(29), 11652–11656. https://doi.org/10.1073/pnas.1203647109

Kikulwe, E. M., Birol, E., Wesseler, J., & Falck-Zepeda, J. (2011). A latent class approach to investigating demand for genetically modified banana in Uganda. *Agricultural Economics*, *42*(5), 547–560. https://doi.org/10.1111/j.1574-0862.2011.00540.x

Klapper, L., Lusardi, A., & Van Oudheusden, P. (2019). *Financial literacy around the world: Insights from the Standard and Poor's Ratings Services Global Financial Literacy Survey*. World Bank. https://gflec.org/wp-content/uploads/2015/11/Finlit_paper_16_F2_singles.pdf

Kniss, A. R. (2010). Genetically engineered herbicide-resistant crops and herbicide-resistant weed evolution in the U.S. *Weed Science*, *58*(2), 305–317. https://doi.org/10.1614/WS-09-098.1

Labeyrie, V., Renard, D., Aumeeruddy-Thomas, Y., Benyei, P., Caillon, S., Calvet-Mir, L., Carrière, S. M., Demongeot, M., Descamps, E., Junqueira, A. B., Li, X., Locqueville, J., Mattalia, G., Miñarro, S., Morel, A., Porcuna-Ferrer, A., Schlingmann, A., Da Cunha Avila, J. V., & Reyes-García, V. (2021). The role of crop diversity in climate change adaptation: insights from local observations to inform decision making in agriculture. *Current Opinion in Environmental Sustainability*, *51*, 15–23. https://doi.org/10.1016/j.cosust.2021.01.006

Nnaemeka, E., & Sun, Y. (2023). A comparative study of transgenic cotton development, impacts, challenges and prospects with respect to China and Africa. *African Journal of Biotechnology*, *22*(8), 305–316. https://doi.org/10.5897/AJB2022.17486

Pattanaik, S., & Priyadarshini, A. (2024). Millets: Super food for nutrition security and livelihood improvement. In *Millets and other potential crops: Ensuring climate resilience and nutritional security* (pp. 11–22). Springer.

Prasanna, B. M., Cairns, J. E., Zaidi, P. H., Beyene, Y., Makumbi, D., Gowda, M., Magorokosho, C., Zaman-Allah, M., Olsen, M., Das, A., Worku, M., Gethi, J., Vivek, B. S., Nair, S. K., Rashid, Z., Vinayan, M. T., Issa, A. B., San Vicente, F., Dhliwayo, T., & Zhang, X. (2021). Beat the stress: Breeding for climate resilience in maize for the tropical rainfed environments. *Theoretical and Applied Genetics*, *134*, 1729–1752. https://doi.org/10.1007/s00122-021-03773-7

Pray, C.E., Huang, J., Hu, R., Wang, Q., Ramaswami, B., & Bengali, P. (2006). Benefits and Costs of Biosafety Regulation in India and China. In: Just, R.E., Alston, J.M., Zilberman, D. (eds) *Regulating Agricultural Biotechnology: Economics and Policy. Natural Resource Management and Policy* (Vol. 30, pp. 481–508). Springer. https://doi.org/10.1007/978-0-387-36953-2_22

Qaim, M. (2020). Role of new plant breeding technologies for food security and sustainable agricultural development. *Applied Economic Perspectives and Policy*, *42*(2), 129–150. https://doi.org/10.1002/aepp.13044

Raney, T., & Pingali, P. (2007). *Sowing a gene revolution: The impact of genetically modified crops on developing countries*. Oxford University Press.

Reynolds, M., & Langridge, P. (2016). Breeding for drought resistance in cereals: Economic and environmental impacts. *Global Food Security*, *9*, 8–15. https://doi.org/10.1016/j.gfs.2016.05.001

Ruggieri, F., Porcuna-Ferrer, A., Gaudin, A., Faye, N. F., Reyes-García, V., & Labeyrie, V. (2021). Crop diversity management: Sereer smallholders' response to climatic variability in Senegal. *Journal of Ethnobiology*, *41*(3), 389–408. https://doi.org/10.2993/0278-0771-41.3.389

Skarbø, K. (2014). The cooked is the kept: Factors shaping the maintenance of agro-biodiversity in the Andes. *Human Ecology*, *42*(5), 711–726. https://doi.org/10.1007/s10745-014-9683-3

Thompson, J., Millstone, E., Scoones, I., Ely, A., Marshall, F., Shah, E., & Stagl, S. (2007). *Agri-food system dynamics: Pathways to sustainability in an era of uncertainty* (STEPS Working Paper 4). STEPS Centre. https://hdl.handle.net/20.500.12413/2472

Varshney, R. K., Thudi, M., Roorkiwal, M., He, W., Upadhyaya, H. D., Yang, W., Bajaj, P., Cubry, P., Rathore, A., Jian, J., Doddamani, D., Khan, A. W., Garg, V., Chitikineni, A., Xu, D., Gaur, P. M., Singh, N. P., Chaturvedi, S. K., Nadigatla, G. V. P. R., . . . Liu, X. (2019). Resequencing of 429 chickpea accessions from 45 countries provides insights into genome diversity, domestication and agronomic traits. *Nature Genetics*, *51*(5), 857–864. https://doi.org/10.1038/s41588-019-0401-3

Wang, X., Huang, J., Liu, H., Xiang, C., & Zhang, W. (2013). *Adoption and uptake pathway of GM technology by Chinese smallholders: Evidence from Bt cotton production*. Center for Chinese Agricultural Policy, Chinese Academy of Sciences.

13 Global Acceptance and Challenges of GMO-Free CRISPR/Cas9 Crops

Haragopal Dutta and Suman Dutta

13.1 INTRODUCTION

In the field of genetic engineering, CRISPR/Cas9 technology has become a revolutionary tool, especially in the agricultural sector. By precisely altering the genome, this system which stands for Clustered Regularly Interspaced Short Palindromic Repeats and CRISPR-associated protein 9 allows for the creation of crops with improved characteristics like higher yield, better disease resistance, and improved resilience to environmental stressors. CRISPR/Cas9's ease of use and effectiveness have made it a popular alternative to more complicated and ineffective traditional gene-editing techniques like TALENs and zinc-finger nucleases (Stajič & Kunej, 2023). A guide RNA (gRNA) is used in the basic mechanism of CRISPR/Cas9 to lead the Cas9 nuclease to a particular spot in the DNA sequence. The cell tries to fix the double-strand break caused by Cas9 once the target location has been located. By either introducing new genetic material or deleting existing genes, this repair mechanism can be used to introduce particular genetic modifications (Cheng et al., 2022). Multiplexing, the capacity to target many genes at once, increases the usefulness of CRISPR/Cas9 in crop development.

The application of CRISPR/Cas9 in agriculture is not without difficulties, despite its obvious benefits. There have been concerns expressed about off-target effects, which modify unwanted sections of the genome; therefore, more research is needed to increase the technology's specificity. Furthermore, it is yet unclear how releasing gene-edited crops into the ecosystem would affect the environment in the long run, which is why thorough risk assessments are being called for. Furthermore, it is yet unclear how releasing gene-edited crops into the ecosystem would affect the environment in the long run, which is why thorough risk assessments are being called for (Li et al., 2021). Discussions regarding the responsible use of this technology are therefore crucial for all parties involved, including the public, scientists, and legislators. The ability of CRISPR/Cas9 to produce transgene-free crops is among its most important benefits in agriculture. Foreign DNA is frequently inserted during traditional genetic alteration, which may cause problems with public acceptance and regulations. However, the regulatory approach for novel crop types can be made simpler by using CRISPR/Cas9 to perform modifications without introducing foreign genetic material (Zhang et al., 2021). Given the growing public demand for non-genetically modified (GM) organism (GMO)

DOI: 10.1201/9781003597797-13

products and the stricter regulations governing the approval of GMOs, this factor is especially crucial (Zhang et al., 2021). The global acceptance of GMO-free CRISPR/Cas9 crops is increasing as they offer a precise, non-transgenic approach to improving agricultural traits, aligning with consumer preferences and regulatory flexibility in many regions.

13.2 "GMO-FREE" CROP USING CRISPR/CAS9

The term "GMO-free" in CRISPR/Cas9 crops has gained attention due to the ability of genome editing to modify plant DNA without introducing foreign genetic material, offering a more precise alternative to traditional genetic modification (Malnoy et al., 2016). Technologies like ribonucleoprotein (RNP) complexes enable DNA-free editing, reducing the likelihood of transgenic integration and potentially qualifying these crops as non-GMO under some regulations. However, regulatory approaches vary significantly; for example, the EU classifies all gene-edited crops as GMOs, regardless of foreign DNA, while the U.S. and Australia may exempt DNA-free edits (Neequaye et al., 2020). This inconsistency creates uncertainty for researchers and developers, hindering CRISPR's agricultural application. Consumer perception also plays a crucial role, with studies suggesting that people view DNA-free edited crops as more natural than conventional GMOs, influencing purchasing behavior (Brandt & Barrangou, 2019; Oselinsky et al., 2021).

13.3 REGULATORY LANDSCAPE FOR GMO-FREE CRISPR CROPS

The regulatory landscape for GMO-free CRISPR crops is complex and evolving, with significant implications for crop development and commercialization as the technology advances (ISAAA, 2025). Regulatory classifications vary widely across countries, with some adopting more permissive approaches for transgene-free edits, facilitating market entry, while others impose stricter GMO-based regulations (Duensing et al., 2018). A key debate centers on whether regulation should focus on the editing process or the final product's traits, further complicating policy alignment with international agreements like the Cartagena Protocol (Table 13.1) (ISAAA, 2025). CRISPR's potential to enhance climate-resilient crops, such as drought-tolerant varieties, adds another layer to regulatory discussions, as it offers a faster and more precise alternative to traditional breeding (Gajardo et al., 2023). Public perception plays a crucial role, with studies indicating that transgene-free CRISPR crops may be viewed as more natural, improving consumer acceptance compared to conventional GMOs. Successful regulatory approvals in certain regions, as highlighted by Sathee et al. (2022), suggest a growing recognition of CRISPR's benefits, though harmonizing global policies remains a challenge for widespread adoption.

13.4 PUBLIC CONCERN ABOUT GENE EDITING AND FACTORS INFLUENCING ACCEPTANCE OF GMO-FREE CRISPR CROPS

Public concerns about gene editing, particularly regarding GMOs and CRISPR technology, stem from a complex interplay of health, environmental, sociopolitical, and consumer perception factors. Studies show that many consumers remain skeptical of

TABLE 13.1
GM Crop Events Approved in Different Countries

Country	Crop	No of Events Approved
Argentina	Alfalfa – *Medicago sativa*	2 Events
	Cotton – *Gossypium hirsutum* L.	12 Events
	Maize – *Zea mays* L.	74 Events
	Safflower – *Carthamus tinctorius* L.	1 Event
	Soybean – *Glycine max* L.	25 Events
	Sugarcane – *Saccharum* sp.	1 Event
	Wheat – *Triticum aestivum*	1 Event
Australia	Alfalfa – *M. sativa*	3 Events
	Argentine canola – *Brassica napus*	27 Events
	Carnation – *Dianthus caryophyllus*	12 Events
	Cotton – *G. hirsutum* L.	28 Events
	Maize – *Z. mays* L.	35 Events
	Potato – *Solanum tuberosum* L.	21 Events
	Rice – *Oryza sativa* L.	2 Events
	Rose – *Rosa hybrida*	1 Event
	Safflower – *C. tinctorius* L.	2 Events
	Soybean – *G. max* L.	21 Events
	Wheat – *T. aestivum*	2 Events
	Sugar Beet – *Beta vulgaris*	2 Events
Bangladesh	Eggplant – *Solanum melongena*	1 Event
Brazil	Bean – *Phaseolus vulgaris*	1 Event
	Cotton – *G. hirsutum* L.	26 Events
	Eucalyptus – *Eucalyptus* sp.	4 Events
	Maize – *Z. mays* L.	77 Events
	Soybean – *G. max* L.	22 Events
	Wheat – *T. aestivum*	1 Event
	Sugarcane – *Saccharum* sp.	8 Events
Canada	Alfalfa – *M. sativa*	3 Events
	Apple – *Malus x Domestica*	3 Events
	Argentine canola – *B. napus*	22 Events
	Cotton – *G. hirsutum* L.	27 Events
	Flax – *Linum usitatissimum* L.	1 Event
	Maize – *Z. mays* L.	73 Events
	Papaya – *Carica papaya*	1 Event
	Polish canola – *Brassica rapa*	4 Events
	Potato – *S. tuberosum* L.	29 Events
	Pineapple – *Ananas comosus*	1 Event
	Soybean – *G. max* L.	26 Events
	Squash – *Cucurbita pepo*	3 Events
	Sugarcane – *Saccharum* sp.	2 Events
	Tomato – *Lycopersicon esculentum*	4 Events
India	Cotton – *G. hirsutum* L.	6 Events
	Soybean – *G. max* L.	5 Events

GMOs, primarily due to perceived health risks and ecological impacts, with heightened caution toward GM animal products compared to plant-based ones (Oselinsky et al., 2021; Sendhil et al., 2021). Distrust is further fueled by fears that genetic modifications may alter food's natural qualities, such as taste, texture, or allergenicity (Cerjak et al., 2011). Political and labeling policies also shape perceptions, as mandatory GMO labels, despite scientific consensus on safety often reinforce consumer hesitancy by framing these products as potentially hazardous (Areal & Riesgo, 2021; Kim et al., 2021).

In contrast, CRISPR-edited crops, viewed as more precise and "natural" due to their transgene-free potential, face relatively higher acceptance (Farid et al., 2020; Ishii & Araki, 2016). However, adoption hinges on public education and trust, as misinformation or lack of transparency can hinder progress. Regulatory disparities further complicate the landscape, with countries like the U.S. and Brazil taking flexible approaches, while the EU and New Zealand enforce stricter oversight (ISAAA, 2025). Regional sociopolitical attitudes also play a role, as anti-GMO sentiment in Europe often ties to broader critiques of corporate agriculture, whereas CRISPR crops may gain traction in regions prioritizing food security and sustainability (Farid et al., 2020). Ultimately, balancing scientific innovation with public engagement and harmonized regulations will be key to advancing CRISPR technology in agriculture (Table 13.2) (ISAAA, 2025).

13.4.1 OFF-TARGET EFFECTS AND EFFICIENCY IN CERTAIN CROP SPECIES

CRISPR/Cas9 has revolutionized genome editing in agriculture, but challenges such as off-target effects and variable efficiency across crops remain significant hurdles (Zhang et al., 2015). Off-target activity, which can exceed 50% in some cases, raises

TABLE 13.2
Gene-Editing Regulations in Major GMO Food Crops Producing Countries

Country	SDN-1	SDN-2	SDN-3
India	Under development	Under development	Under development
China	Under development	Under development	Under development
Pakistan	Under development	Under development	Under development
The U.S.	Deregulated	Deregulated	Case-by-case
Australia (excl. Tas)	Deregulated	Regulated	Regulated
Argentina	Deregulated	Deregulated	Deregulated (if not transgenic)
New Zealand	Regulated (under review)	Regulated (under review)	Regulated (under review)
Japan	Deregulated	Deregulated	Regulated
Brazil	Deregulated	Deregulated	Deregulated (if not transgenic)
Canada	Case-by-case	Case-by-case	Case-by-case
EU	Regulated (under review)	Regulated (under review)	Regulated
Colombia	Case-by-case	Case-by-case	Deregulated (if not transgenic)

safety concerns and is influenced by factors like sgRNA design, PAM sequences, and delivery methods (Cheng et al., 2022; Xu et al., 2015). Efficiency also varies widely among crops due to differences in genetic backgrounds, transformation techniques, and cellular repair mechanisms (Liu et al., 2022). Innovations like double nicking and ribonucleoprotein (RNP) delivery have improved precision, while alternative systems like CRISPR-Cas12a offer solutions to Cas9's limitations (Bandyopadhyay et al., 2020; Zhang et al., 2020). Despite these advances, optimizing CRISPR for diverse crops and ensuring safety remain critical for its widespread agricultural adoption. Continued research into editing specificity and efficiency will be essential to fully harness CRISPR's potential in crop improvement.

13.5 CASE STUDY: IN THE PROCESS OF COMMERCIALIZATION

A crucial component of contemporary agriculture, especially in tackling the issues of global food security, is the commercialization of GM crops. Notable developments in this area include drought-resistant wheat and non-browning mushrooms, which serve as prime examples of how genetic engineering can improve crop resilience and features (Ren et al., 2019).

13.5.1 Non-Browning Mushroom

The creation of non-browning mushrooms, especially the genetically engineered *Agaricus bisporus*, has attracted a lot of interest because of its potential to improve consumer appeal and decrease food waste. After harvesting, traditional mushrooms have a tendency to turn brown quickly, which reduces their marketability and increases waste. The gene that produces polyphenol oxidase (PPO), an enzyme that triggers the browning response when mushrooms are sliced or injured, is silenced to produce the non-browning phenotype (Xie et al., 2022). Because of this genetic change, the mushrooms have a longer shelf life and retain their aesthetic appeal, which increases consumer appeal (Zhu et al., 2018). Non-browning mushrooms have undergone a stringent licensing procedure, with regulatory agencies like the Food and Drug Administration (FDA) and the U.S. Department of Agriculture (USDA) assessing its environmental impact and safety. The sale of these mushrooms, which have been approved for human consumption, marks a critical turning point in the acceptability of GM crops by both consumers and regulatory bodies.

13.5.2 Drought-Resistant Wheat

Another crucial area of study and development in the field of GM crops is drought-resistant wheat. The demand for crops that can withstand drought has grown more pressing as climate change continues to worsen water scarcity. CRISPR/Cas9 and other genome-editing technologies are examples of genetic engineering techniques that have been used to improve drought tolerance in wheat types (Schouteten et al., 2018). These developments enable the production of wheat that can sustain output in water-limited environments by precisely altering genes linked to drought sensitivity. Numerous quantitative trait loci (QTLs) linked to drought tolerance have been found through research; these QTLs can be stacked to produce cultivars with increased resilience (Khan et al., 2019). For example, it has been demonstrated that adding particular transcription factors that control stress response pathways might increase drought tolerance without lowering production (Khan et al., 2019). It is projected that the commercialization of drought-resistant wheat cultivars will be essential to maintaining food security in areas vulnerable to water scarcity and promoting agricultural sustainability (Ryan et al., 2024).

13.6 SUGGESTIONS FOR HARMONIZING GLOBAL REGULATORY FRAMEWORKS

The harmony of global regulation of CRISPR-treated plants remains an important topic in agricultural biotechnology as different countries pursue different regulatory approaches. The U.S. exempts foreign DNA-free CRISPR plants from GMO regulations (Ahmad et al., 2022), while the EU classifies it as a GMO and imposes strict managers (Menz et al., 2020; Neequaye et al., 2020). Countries such as Argentina and Brazil have more flexible policies that emphasize regulatory fragmentation that may hinder trade and innovation (Ishii & Araki, 2017). Ethical concerns and public awareness also play an important role, with consumer acceptance being influenced by education and public relations (Friedrich et al., 2019). Because current assessments focus more on transgenic than genome-related plants, organic security considerations require updated framework conditions for risk assessment (Ishii & Araki, 2016; Pruitt et al., 2021). International framework conditions such as the *Codex Alimentarius* and the Cartagena Protocol can support regulatory orientations (Movahedi et al., 2023). High regulatory costs in strict regions may block investment in CRISPR technology and limit nutritional safety and sustainability solutions (Entine et al., 2021). Globally coordinated regulatory frameworks are essential to promoting innovation and are also the trust of public and environmental safety (Ayanoğlu et al., 2020).

13.7 POTENTIAL OF GENE-EDITING TECHNIQUES TO OVERCOME CURRENT LIMITATIONS

Base editing represents a groundbreaking advancement in genetic engineering, enabling precise conversion of DNA base pairs without inducing double-strand breaks, thereby reducing unintended mutations compared to traditional CRISPR-Cas9 (Huang & Liu, 2023; Lamboro et al., 2021). This technology has demonstrated therapeutic promise in correcting point mutations linked to hereditary diseases, though its application remains limited to specific genetic contexts (Gao et al., 2023; Ning et al., 2023). Prime editing, an even more versatile approach, combines a modified Cas9 protein with pegRNA to perform precise insertions, deletions, and base substitutions with minimal off-target effects, positioning it as a "search-and-replace" genomic tool (Permyakova & Deineko, 2024). However, its efficiency varies depending on target sequences, necessitating further optimization for broader use (Wei et al., 2023). Delivery challenges persist, but emerging solutions like virus-like particles (VLPs) show potential to enhance specificity and efficiency of CRISPR component delivery (Walsh & Jin, 2024). Ethical considerations, particularly regarding germline editing and long-term impacts, underscore the need for robust regulatory frameworks and public dialogue to ensure responsible development and deployment of these technologies (Feng et al., 2024; Tang, 2023; Xu, 2023).

Academic institutions play a pivotal role in advancing genetic editing technologies by conducting foundational research that addresses key limitations, particularly in specialized areas like orphan crop improvement (Venezia & Creasey Krainer, 2021). Collaborations between academia and industry accelerate innovation, enabling the commercialization of CRISPR-edited crops with enhanced traits such as yield and stress resilience (Liu et al., 2022;). However, regulatory ambiguity persists, as debates continue over whether CRISPR-edited plants should be classified as GMOs, impacting their market acceptance (Sampath et al., 2023). The agricultural sector increasingly relies on CRISPR to address global food security challenges, with applications ranging from disease resistance to abiotic stress tolerance (Erdoğan et al., 2023). Such partnerships not only drive economic benefits but also foster knowledge exchange, leading to region-specific solutions for diverse agricultural needs (Nazir et al., 2022; Tang et al., 2023). By combining expertise, stakeholders can optimize CRISPR's potential to deliver sustainable, farmer- and consumer-friendly innovations.

13.8 CONCLUSION

The future of CRISPR/Cas9 in agriculture hinges on overcoming regulatory, ethical, and public perception challenges while fostering collaboration among researchers, industry stakeholders, and policymakers. Diverse legal frameworks, such as the EU's strict GMO regulations, and public concerns about safety and ethics pose significant barriers to adoption. Multidisciplinary approaches integrating genetics, agronomy, ecology, and social sciences are essential to develop comprehensive policies and ensure the safe, equitable use of CRISPR-edited crops. Engaging stakeholders in transparent discussions and addressing ethical concerns, such as biodiversity impacts and ownership rights, will be critical to building trust and accelerating the adoption of this transformative technology.

REFERENCES

Ahmad, T. S., Ahmad, S., & Afriyani, A. (2022). Entrepreneurial commitment, entrepreneurial competence, towards business performance through business competitiveness development. *Jurnal Penelitian Pendidikan Indonesia*, *8*(3), 568. https://doi.org/10.29210/020221444

Areal, F. J., & Riesgo, L. (2021). EU inspections of GM content in food and feed: Are they effective? *Agriculture*, *11*(9), 842. https://doi.org/10.3390/agriculture11090842

Ayanoğlu, F. B., Elçin, A. E., & Elçin, Y. M. (2020). Bioethical issues in genome editing by CRISPR-Cas9 technology. *Turkish Journal of Biology*, *44*(2), 110–120. https://doi.org/10.3906/biy-1912-52

Bandyopadhyay, A., Kancharla, N., Javalkote, V. S., Dasgupta, S., & Brutnell, T. P. (2020). CRISPR-Cas12a (Cpf1): A versatile tool in the plant genome editing toolbox for agricultural advancement. *Preprints*. https://doi.org/10.20944/preprints202007.0578.v1

Brandt, K., & Barrangou, R. (2019). Applications of CRISPR technologies across the food supply chain. *Annual Review of Food Science and Technology*, *10*(1), 133–150. https://doi.org/10.1146/annurev-food-032818-121204

Cerjak, M., Kovačić, D., & Ggić, I. (2011). What are the concerns of Croatian traditional food consumers regarding GM food? *British Food Journal*, *113*(1), 37–49. https://doi.org/10.1108/00070701111097321

Cheng, Q., Xia, J., Wang, K., Zhang, Y., Chen, Y., Zhong, Q., Wang, X., & Wu, Q. (2022). CRISPR/Cas9 ribonucleoprotein (RNP) complex enables higher viability of transfected cells in genome editing of acute myeloid cells. *Annals of Translational Medicine*, *10*(16), 862. https://doi.org/10.21037/atm-22-3279

Duensing, N., Sprink, T., Parrott, W. A., Fedorova, M., Lema, M. A., Wolt, J. D., & Bartsch, D. (2018). Novel features and considerations for ERA and regulation of crops produced by genome editing. *Frontiers in Bioengineering and Biotechnology*, *6*, 79. https://doi.org/10.3389/fbioe.2018.00079

Entine, J., Felipe, M. S. S., Groenewald, J. H., Kershen, D. L., Lema, M., McHughen, A., Nepomuceno, A. L., Ohsawa, R., Ordonio, R. L., Parrott, W. A., Quemada, H., Ramage, C., Slamet-Loedin, I., Smyth, S. J., & Wray-Cahen, D. (2021). Regulatory approaches for genome edited agricultural plants in select countries and jurisdictions around the world. *Transgenic Research*, *30*(4), 551–584. https://doi.org/10.1007/s11248-021-00257-8

Erdoğan, İ, Cevher-Keskin, B., Bilir, Ö, Hong, Y., & Tör, M. (2023). Recent developments in CRISPR/Cas9 genome-editing technology related to plant disease resistance and abiotic stress tolerance. *Biology*, *12*(7), 1037. https://doi.org/10.3390/biology12071037

Farid, M., Cao, J., Lim, Y., Arato, T., & Kodama, K. (2020). Exploring factors affecting the acceptance of genetically edited food among youth in Japan. *International Journal of Environmental Research and Public Health*, *17*(8), 2935. https://doi.org/10.3390/ijerph17082935

Feng, Q., Li, Q., Zhou, H., Wang, Z., Lin, C., Jiang, Z., Liu, T., & Wang, D. (2024). CRISPR technology in human diseases. *MedComm*, *5*(8), e672. https://doi.org/10.1002/mco2.672

Friedrich, B., Hackfort, S., Boyer, M., & Gottschlich, D. (2019). Conflicts over GMOs and their contribution to food democracy. *Politics and Governance*, *7*(4), 165–177. https://doi.org/10.17645/pag.v7i4.2082

Gajardo, H. A., Gómez-Espinoza, O., Boscariol Ferreira, P., Carrer, H., & Bravo, L. A. (2023). The potential of CRISPR/Cas technology to enhance crop performance on adverse soil conditions. *Plants*, *12*(9), 1892. https://doi.org/10.3390/plants12091892

Gao, T., Li, Y., & Zhao, Y. (2023). CRISPR/Cas base-editing systems and their potential applications and prospects. *Theoretical and Natural Science*, *20*(1), 129–134. https://doi.org/10.54254/2753-8818/20/20230739

Huang, Z., & Liu, G. (2023). Current advancement in the application of prime editing. *Frontiers in Bioengineering and Biotechnology*, *11*, 1039315. https://doi.org/10.3389/fbioe.2023.1039315

ISAAA. (2025). *Countries with GM crop approvals*. Retrieved March 13, 2025, from https://www.isaaa.org/gmapprovaldatabase/countrylist/default.asp

Ishii, T., & Araki, M. (2016). Consumer acceptance of food crops developed by genome editing. *Plant Cell Reports*, *35*(7), 1507–1518. https://doi.org/10.1007/s00299-016-1974-2

Ishii, T., & Araki, M. (2017). A future scenario of the global regulatory landscape regarding genome-edited crops. *GM Crops and Food*, *8*(1), 44–56. https://doi.org/10.1080/21645698.2016.1261787

Khan, S., Anwar, S., Yu, S., Sun, M., Yang, Z., & Gao, Z. Q. (2019). Development of drought-tolerant transgenic wheat: Achievements and limitations. *International Journal of Molecular Sciences*, *20*(13), 3350. https://doi.org/10.3390/ijms20133350

Kim, T. K., Lee, C., Nam, T. H., Seo, Y. K., Kim, K. S., Kang, K., Yun, S. H., Yoon, S. H., & Jeong, J. (2021). Isolation and identification of non-conjugated linoleic acid from processed *Panax ginseng* using LC-MS/MS and 1H-NMR. *Separations*, *8*(11), 208. https://doi.org/10.3390/separations8110208

Lamboro, A., Song, B., Songnan, Y., Han, X., Mingguo, H., Li, X., Yao, D., & Zhang, J. (2021). Genetic engineering and genome editing techniques in peanut plants. *Plant Science Today*, *8*(3), 528–534. https://doi.org/10.14719/pst.2021.8.3.1127

Li, J., Li, Y., & Ma, L. (2021). Recent advances in CRISPR/Cas9 and applications for wheat functional genomics and breeding. *aBIOTECH*, *2*(4), 375–385. https://doi.org/10.1007/s42994-021-00042-5

Liu, H., Chen, W., Li, Y., Sun, L., Chai, Y., Chen, H., Nie, H., & Huang, C. (2022). CRISPR/Cas9 technology and its utility for crop improvement. *International Journal of Molecular Sciences*, *23*(18), 10442. https://doi.org/10.3390/ijms231810442

Malnoy, M., Viola, R., Jung, M. H., Koo, O. J., Kim, S., Kim, J. S., Velasco, R., & Kanchiswamy, C. N. (2016). DNA-free genetically edited grapevine and apple protoplast using CRISPR/Cas9 ribonucleoproteins. *Frontiers in Plant Science*, *7*, 1904. https://doi.org/10.3389/fpls.2016.01904

Menz, J., Modrzejewski, D., Hartung, F., Wilhelm, R., & Sprink, T. (2020). Genome edited crops touch the market: A view on the global development and regulatory environment. *Frontiers in Plant Science*, *11*, 586027. https://doi.org/10.3389/fpls.2020.586027

Movahedi, A., Aghaei-Dargiri, S., Li, H., Zhuge, Q., & Sun, W. (2023). CRISPR variants for gene editing in plants: Biosafety risks and future directions. *International Journal of Molecular Sciences*, *24*(22), 16241. https://doi.org/10.3390/ijms242216241

Nazir, R., Mandal, S., Mitra, S., Ghorai, M., Das, N., Jha, N. K., Majumder, M., Pandey, D. K., & Dey, A. (2022). Clustered regularly interspaced short palindromic repeats (CRISPR)/CRISPR-associated genome-editing toolkit to enhance salt stress tolerance in rice and wheat. *Physiologia Plantarum*, *174*(2), e13642. https://doi.org/10.1111/ppl.13642

Neequaye, M., Stavnstrup, S., Lawrenson, T., Hundleby, P., Troncoso-Rey, P., Saha, S., Harwood, W., Traka, M. H., Mithen, R., & Østergaard, L. (2020). CRISPR-Cas9-mediated editing of *myb28* impairs glucoraphanin accumulation of *Brassica oleracea* in the field. *bioRxiv*. https://doi.org/10.1101/2020.07.16.206813

Ning, L., Xi, J., Zi, Y., Chen, M., Zou, Q., Zhou, X., & Tang, C. (2023). Prospects and challenges of CRISPR/Cas9 gene-editing technology in cancer research. *Clinical Genetics*, *104*(6), 613–624. https://doi.org/10.1111/cge.14424

Oselinsky, K., Johnson, A., Lundeberg, P., Holm, A. J., Mueller, M., & Graham, D. J. (2021). GMO food labels do not affect college student food selection, despite negative attitudes towards GMOs. *International Journal of Environmental Research and Public Health*, *18*(4), 1761. https://doi.org/10.3390/ijerph18041761

Permyakova, N. V., & Deineko, E. V. (2024). Crop improvement: Comparison of transgenesis and gene editing. *Horticulturae*, *10*(1), 57. https://doi.org/10.3390/horticulturae10010057

Pruitt, J. R., Melton, K. M., & Palma, M. A. (2021). Does physical activity influence consumer acceptance of gene edited food? *Sustainability*, *13*(14), 7759. https://doi.org/10.3390/su13147759

Ren, F., Ren, C., Zhang, Z., Duan, W., Lecourieux, D., Li, S., & Liang, Z. (2019). Efficiency optimization of CRISPR/Cas9-mediated targeted mutagenesis in grape. *Frontiers in Plant Science*, *10*, 612. https://doi.org/10.3389/fpls.2019.00612

Ryan, C. D., Henggeler, E., Gilbert, S., Schaul, A. J., & Swarthout, J. T. (2024). Exploring the GMO narrative through labeling: Strategies, products, and politics. *GM Crops and Food*, *15*(1), 51–66. https://doi.org/10.1080/21645698.2024.2318027

Sampath, V., Rangarajan, N., Sharanappa, C. H., Deori, M., Veeraragavan, M., Ghodake, B. D., & Kaushal, K. (2023). Advancing crop improvement through CRISPR technology in precision agriculture trends—A review. *International Journal of Environment and Climate Change*, *13*(11), 4683–4694. https://doi.org/10.9734/IJECC/2023/v13i113647

Sathee, L., Jagadhesan, B., Pandesha, P. H., Barman, D., Adavi, B. S., Nagar, S., Krishna, G. K., Tripathi, S., Jha, S. K., & Chinnusamy, V. (2022). Genome editing targets for improving nutrient use efficiency and nutrient stress adaptation. *Frontiers in Genetics*, *13*, 900897. https://doi.org/10.3389/fgene.2022.900897

Schouteten, J. J., Gellynck, X., & De Steur, H. (2018). Consumers' perceptions of GM-free labelled foods: A sensory experiment. *International Journal of Consumer Studies*, *42*(3), 347–357. https://doi.org/10.1111/ijcs.12424

Sendhil, R., Nyika, J., Yadav, S., Mackolil, J., Rama Prashat, P. G., Workie, E., Ragupathy, R., & Ramasundaram, P. (2021). Consumer perception and preference towards genetically modified (GM) foods: Bibliometric evidence and policy imperatives. *AgriRxiv*. https://doi.org/10.31220/agriRxiv.2021.00061

Stajič, E., & Kunej, U. (2023). Optimization of cabbage (*Brassica oleracea* var. *capitata* L.) protoplast transformation for genome editing using CRISPR/Cas9. *Frontiers in Plant Science*, *14*, 1245433. https://doi.org/10.3389/fpls.2023.1245433

Tang, J. (2023). Research and prospect of CRISPR system in marine biological system. *Highlights in Science, Engineering and Technology*, *36*, 132–138. https://doi.org/10.54097/hset.v36i.5652

Tang, Q., Wang, X., Jin, X., Peng, J., Zhang, H., & Wang, Y. (2023). CRISPR/Cas technology revolutionizes crop breeding. *Plants*, *12*(17), 3119. https://doi.org/10.3390/plants12173119

Venezia, M., & Creasey Krainer, K. M. (2021). Current advancements and limitations of gene editing in orphan crops. *Frontiers in Plant Science*, *12*, 742932. https://doi.org/10.3389/fpls.2021.742932

Walsh, C., & Jin, S. (2024). Induced pluripotent stem cells and CRISPR-Cas9 innovations for treating alpha-1 antitrypsin deficiency and glycogen storage diseases. *Cells*, *13*(12), 1052. https://doi.org/10.3390/cells13121052

Wei, X., Long, Y., Yi, C., Pu, A., Hou, Q., Liu, C., Jiang, Y., Wu, S., & Wan, X. (2023). Bibliometric analysis of functional crops and nutritional quality: Identification of gene resources to improve crop nutritional quality through gene editing technology. *Nutrients*, *15*(2), 373. https://doi.org/10.3390/nu15020373

Xie, Y., Haq, S. I. U., Jiang, X., Zheng, D., Feng, N., Wang, W., He, J. S., & Qiu, Q. S. (2022). Plant genome editing: CRISPR, base editing, prime editing, and beyond. *Grassland Research*, *1*(4), 234–243. https://doi.org/10.1002/glr2.12034

Xu, M. (2023). The method and prospects of changing mosquito genes with CRISPR-Cas9. *Journal of Mosquito Research*, *13*(2). https://doi.org/10.5376/JMR.2023.13.0002

Xu, R. F., Li, H., Qin, R. Y., Li, J., Qiu, C. H., Yang, Y. C., Ma, H., Li, L., Wei, P. C., & Yang, J. B. (2015). Generation of inheritable and "transgene clean" targeted genome-modified rice in later generations using the CRISPR/Cas9 system. *Scientific Reports*, *5*(1), 11491. https://doi.org/10.1038/srep11491

Zhang, H., Zhang, Y., Qiu, C., Zhu, W., Wen, M., & Lao, X. (2021). Differentiation of human umbilical cord mesenchymal stem cells into parathyroid cells by editing the PTH gene with the CRISPR/Cas9 system. *Biotechnology and Biotechnological Equipment*, *35*(1), 1207–1213. https://doi.org/10.1080/13102818.2021.1961608

Zhang, S., Shen, J., Li, D., & Cheng, Y. (2020). Strategies in the delivery of Cas9 ribonucleoprotein for CRISPR/Cas9 genome editing. *Theranostics*, *11*(2), 614–648. https://doi.org/10.7150/thno.47007

Zhang, X. H., Tee, L. Y., Wang, X. G., Huang, Q. S., & Yang, S. H. (2015). Off-target effects in CRISPR/Cas9-mediated genome engineering. *Molecular Therapy – Nucleic Acids*, *4*(11), e264. https://doi.org/10.1038/mtna.2015.37

Zhang, Y., Restall, J., Crisp, P., Godwin, I., & Liu, G. (2021). Current status and prospects of plant genome editing in Australia. *In Vitro Cellular and Developmental Biology – Plant*, *57*(4), 574–583. https://doi.org/10.1007/s11627-021-10188-y

Zhu, S., Yu, X., Li, Y., Sun, Y., Zhu, Q., & Sun, J. (2018). Highly efficient targeted gene editing in upland cotton using the CRISPR/Cas9 system. *International Journal of Molecular Sciences*, *19*(10), 3000. https://doi.org/10.3390/ijms19103000

14 Integrating Biotechnology into Sustainable Agricultural Practices

Opportunities and Challenges

Surender Singh Chandel, Isha Sharma, Sonika Kalia, and Vinay Sharma

14.1 INTRODUCTION

The word 'sustainable' means keeping, using, maintaining and preserving of available things or resources. Agriculture can be called as sustainable when nations have sufficient food and energy to fulfill the current needs of human beings in an ecofriendly way for their better and happy living. This approach of sustainable agriculture should be able to conserve, maintain, and effectively preserve the environment and bio-resources for coming generations (Das et al., 2023) and include sustainable practices to overcome various challenges such as increasing global population of humans; continuously changing climates; depletion of biodiversity of flora and fauna; high requirements of fuels; starvation and malnutrition among developing and poor nations; decreasing soil fertility; high use of pesticides and chemical fertilizers; rise in deforestation and pollution of air and water bodies (Figure 14.1).

With the aim to develop sustainable agriculture, there are some major obstructions in front of plant breeders and biotechnologists, such as pests, plant diseases, soil nutrient deficiency, water depletion, and even climate change, which altogether affect crops yield and quality (Das et al., 2023). Agricultural biotechnology has the potential to significantly boost crop output, improve crop nutrients, and increase crops resistance to biotic and abiotic stresses (Das et al., 2023). Karl Erkey, a Hungarian engineer, first used the term 'biotechnology' in 1919 to describe the use of living things along with principles of engineering for the production of new biological products. The word 'biotechnology' has been made up of two terms of science that are 'Biology' and 'Technology' (Verma et al., 2011).

In general, biotechnology means technological applications in biology. The applications of biotechnology possess the limitless capacity to assist and advance humanity in a variety of fields, including agricultural, pharmaceutical, animal husbandry, environmental, and many more (Verma et al., 2011). Many research

DOI: 10.1201/9781003597797-14

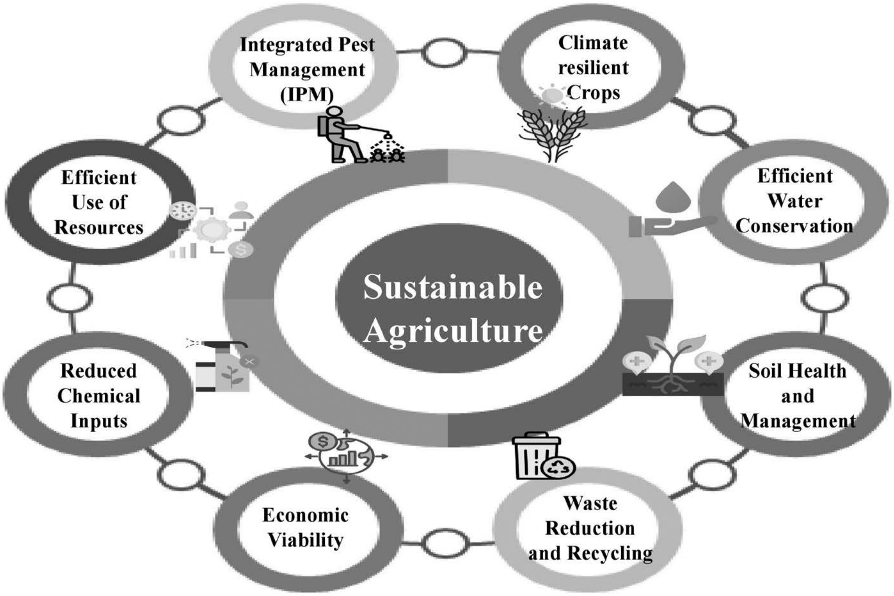


FIGURE 14.1 Combined practices that can lead to the development of sustainable agriculture.

studies has clearly demonstrated that biotechnological applications could increase the nutrient qualities and production of crops (Khush et al., 2012), simultaneously with the weed management (Dill, 2005) in an ecofriendly manner. Recently, new paths have been opened by genome editing technologies, including CRISPR-Cas9 and Cas12a nucleases for the development of biotechnological products. Despite showing positive scopes of biotechnology, it also faces challenges which still hinder its broad acceptability which includes non-acceptability by public due to ethical reasons, followed by risks associated with genetically modified organisms (GMOs).

14.1.1 AGRICULTURAL BIOTECHNOLOGY AND ITS HISTORY

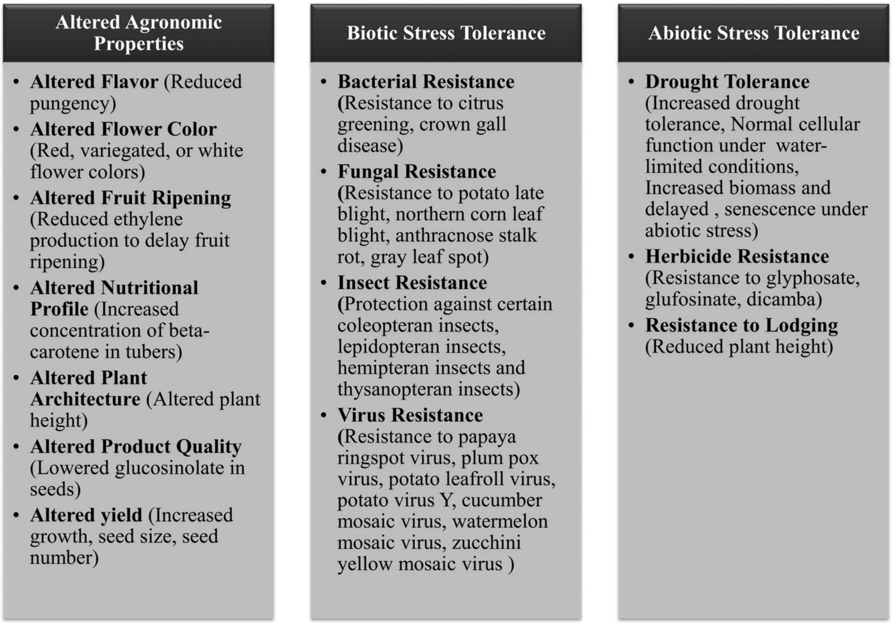
Around 10,000 years BC, humans began harvesting food from natural biodiversity, later domesticating crops and animals by selecting desirable traits for propagation, leading to improved cultivars and livestock (Wieczorek & Wright, 2012). Traditional breeding involved crossing closely related plants to combine traits, a process that was time-consuming, taking 12–15 years to develop new varieties. Gregor Mendel's work on inheritance revolutionized agriculture by explaining gene transmission and enabling selective cross-breeding (Verma et al., 2011). Induced mutations, through chemicals or radiation, were later used to create new traits, yielding over 2,500 plant varieties by the 1970s (Wieczorek & Wright, 2012). Advances like DNA structure discovery, tissue culture, and marker-assisted breeding further refined crop improvement. Recombinant DNA technology emerged, allowing gene transfer between species to produce transgenic crops. The first small-scale transgenic tests occurred in the 1990s in the US and Canada, with the first commercial genetically engineered crop released in 1992. Despite rapid adoption, genetic engineering faces risks and public concerns that limit its full acceptance (Wieczorek & Wright, 2012).

14.2 THE NEED FOR BIOTECHNOLOGY IN AGRICULTURE FOR GLOBAL FOOD SECURITY

Food security depends on four key components: availability, access, quality, and proper utilization of food, with climate change and greenhouse gas emissions threatening crop yields by increasing pests, pathogens, and abiotic stresses (Ray et al., 2013). To meet the demands of a growing population by 2050, global food production must double, a challenge that agricultural biotechnology can address through transgenic and genome-edited crops with enhanced productivity, nutrition, and stress resilience (Tyczewska et al., 2023). Genetically modified (GM) crops, such as corn, cotton, and soybeans, have been approved in 44 countries, offering traits like herbicide tolerance, insect resistance, and improved yield (Tyczewska et al., 2023). Studies show that GM crops reduce pesticide use by 8.3% and lower environmental impact by 18.5%, while increasing yields, soybean by 20%, cotton by 15%, and maize by 7% (Brookes & Barfoot, 2017). Advanced biotechnologies like genome editing, next-generation sequencing, and genomics-assisted breeding further enable the development of high-yielding, climate-resilient crops (Kwak, 2019). Techniques such as CRISPR-Cas9, RNA interference, and molecular marker-assisted breeding are revolutionizing crop improvement by introducing stress-tolerant genes from wild germplasm into elite cultivars. Despite its benefits, public acceptance remains a challenge due to concerns over safety and ethical implications (Yali, 2022).

14.2.1 Challenges in Meeting Global Food Demands

The global community faces numerous challenges in ensuring global food security, including population growth, climate change, resource scarcity, conflict, and pests and diseases. Climate change negatively impacts food security, with crop yields predicted to decrease by 3.1–7.4% for every 1°C temperature increase (Zhao et al., 2017). Energy scarcity and biodiversity losses exacerbate food insecurity, emphasizing the need for transitioning to renewable energy sources (Christoforidou et al., 2023) and maintaining biodiversity. Agricultural biotechnology can contribute to sustainable agriculture by increasing crop productivity and improving environmental health (Donatelli et al., 2017). Addressing ethical and health concerns through government support and regulatory frameworks is crucial for the adoption of GM crops (Ma et al., 2018). The major applications of agricultural biotechnology in the development of sustainable agriculture have been summarized in Figure 14.2.



Altered Agronomic Properties

- **Altered Flavor** (Reduced pungency)
- **Altered Flower Color** (Red, variegated, or white flower colors)
- **Altered Fruit Ripening** (Reduced ethylene production to delay fruit ripening)
- **Altered Nutritional Profile** (Increased concentration of beta-carotene in tubers)
- **Altered Plant Architecture** (Altered plant height)
- **Altered Product Quality** (Lowered glucosinolate in seeds)
- **Altered yield** (Increased growth, seed size, seed number)

Biotic Stress Tolerance

- **Bacterial Resistance** (Resistance to citrus greening, crown gall disease)
- **Fungal Resistance** (Resistance to potato late blight, northern corn leaf blight, anthracnose stalk rot, gray leaf spot)
- **Insect Resistance** (Protection against certain coleopteran insects, lepidopteran insects, hemipteran insects and thysanopteran insects)
- **Virus Resistance** (Resistance to papaya ringspot virus, plum pox virus, potato leafroll virus, potato virus Y, cucumber mosaic virus, watermelon mosaic virus, zucchini yellow mosaic virus)

Abiotic Stress Tolerance

- **Drought Tolerance** (Increased drought tolerance, Normal cellular function under water-limited conditions, Increased biomass and delayed , senescence under abiotic stress)
- **Herbicide Resistance** (Resistance to glyphosate, glufosinate, dicamba)
- **Resistance to Lodging** (Reduced plant height)

FIGURE 14.2 Applications of agricultural biotechnology for sustainable agriculture.

14.3 BIOTECHNOLOGY APPROACHES FOR SUSTAINABLE AGRICULTURE

By creating various high-yielding, nutrient-rich, and stress-tolerant crop varieties, biotechnology can be a strategic tool for the development of sustainable agriculture and can help reduce the massive amounts of fertilizer, insecticide, and pesticide that are used in agriculture. Reducing the use of inorganic fertilizers, fungicides, insecticides, and pesticides can enhance the quality of the soil, air, and water (Barrows et al., 2014). The field of biotechnology works at molecular level such as DNA, RNA, micro RNAs, enzymes, and protein engineering and mainly concentrated on clarifying the function of molecular mechanisms controlling metabolic processes. The herbicide, pest, and disease-resistant varieties can be developed by the use of agricultural biotechnology. The technique of micropropagation in plant tissue culture can quickly and during off-seasons produce numerous copies of a required cultivar, which speeds up the breeding of improved varieties and aids in the preservation of rare germplasms or hybrid plants (Thorpe, 2007).

14.3.1 Genome Editing for Improved Crop Yields

To ensure food security in front of increasing world's population, the biotechnological approaches are required to improve the photosynthetic machinery, shoot-to-root biomass, inflorescence architecture, resistance to multiple stresses along with nutrient use efficiency of various crop plants to enhance their yields and quality (Das et al., 2023). The efficiency of a photosynthesis machinery of a plant cell is related by an important enzyme called as Rubisco which assimilates the atmospheric carbon dioxide (CO_2) into plant biomass. The crop productions can be increased by improving the activity of Rubisco (Iñiguez et al., 2021). In a research study, high temperature (HT)-sensitive crop plants were engineered with the modified Rubisco activase enzyme which was taken from thermophilic cyanobacteria, which increased their yields during higher temperatures (Ogbaga et al., 2018). Rice crops with overexpressed *OsDREB1C* gene showed higher yields and simultaneously reduced growth time, enhanced nitrogen usage efficiency, and encouraged resource allocation (Wei et al., 2022). In plants, photoprotection is the mechanism that removes excess sunlight from chloroplasts to prevent damage to plant cells (Kromdijk et al., 2016). Improving photoprotection and light-harvesting processes in plants reduces the photoinhibition and photooxidative stresses and is better approach that can be used with other biotechnology methods for enhancing the production of crop plants (Hubbart et al., 2018). The nonphotochemical quenching mechanism, which helps plants to dissipate potentially harmful excess absorbed light energy in full sunlight, is carried out by genes like Photosystem II Subunit S (*PsbS*). Increased expression of *PsbS* genes can enhance photosynthetic efficiency and photoprotection in transgenic plants (Murchie et al., 2015).

14.3.1.1 CRISPR-Cas9 and CRISPR-Cas12a-Mediated Genetic Knockout of MicroRNA Genes in Plant Genomes

MicroRNAs (miRNA) are non-coding endogenous small RNAs that regulate their target genes in various aspects of plants development and growth. In previous years, CRISPR-Cas9 nuclease-based editing was more common in the knockout of miRNA genes in plants for study of their functions (Zheng et al., 2024). CRISPR-Cas9 generates small insertions and deletions in miRNA genes; however, Cas9 nuclease does not provide a complete knockout of miRNA genes (Zheng et al., 2024). CRISPR-Cas12a is more suitable and efficient tool than CRISPR-Cas9 nuclease for the study of miRNA genes. Cas12a nuclease generates larger deletions in miRNA genes and prevents the synthesis of mature miRNAs by disrupting the secondary structure of pre-miRNA (Zheng et al., 2024). CRISPR Cas12a produced the *OsMIR390* miRNA gene mutant in rice more effectively than the Cas9 nuclease CRISPR system (Zheng et al., 2024). Mutants that cause loss of functions in nine distinct miRNA genes (*OsMIR5801*, *OsMIR5794*, *OsMIR5789*, *OsMIR3982*, *OsMIR3979*, *OsMIR1870*, *OsMIR1868*, *OsMIR827*, and *OsMIR394*) in rice were efficiently generated by Cas12a nuclease (Zheng et al., 2024).

14.3.2 DEVELOPING RESILIENT CROPS FOR ABIOTIC STRESSES

Abiotic stresses imposed through climate change are a major hazard to agriculture. The abiotic stresses affect 70% of crop production worldwide (Cramer et al., 2011). The plants that were created through the use of genetic engineering, marker-assisted breeding, plant tissue culture etc. are able to withstand drought, chilling, cold, heat, salinity and other stresses which allow the farmers to utilize previously unusable lands and can simultaneously improve their economy (Babar et al., 2015).

14.3.2.1 Soil Salinity Tolerance

Salinity stress includes osmotic stress and ionic toxicity that greatly affects the physical, physiological, and biochemical traits, including decreased rates of germination, photosynthesis, transpiration, and alteration of the plants' normal metabolic processes. The main methods for reducing the effects of salt stress on plant cells are to understand and depict the mechanisms of transport and detoxification of reactive oxygen species (ROS) (Mao et al., 2023). The durum wheat line 149, which is derived from *Triticum monococcum* C68-101, has been thoroughly studied for its salt tolerance behavior through the sodium transporter genes *HKT7* and *HKT8* and by the mechanisms of exclusion of sodium (Na^+) ions (Huang et al., 2006). The salt overly sensitive (SOS) signaling pathway such as SOS1 played a significant part in the exclusion of Na^+ ions in transgenic lines of *Nicotiana benthamiana* and *Arabidopsis*, which overexpress the *TaSOS1* gene and exhibit greater tolerance to salinity than wild-type plants (Zhou et al., 2016). Through vacuolar compartmentalization of Na^+, the NHX family proteins of wheat's Na^+/H^+ antiporters can enhance salinity tolerance and lessen cytosolic sodium toxicity (Brini et al., 2007). *TNHX1* gene with three distinct subgroups of vacuolar Na^+/H^+ antiporter genes (*TaNHXa*, *TaNHXb*, and *TaNHXc*) was functionally identified in wheat; further, salinity and drought tolerance have both increased in *Arabidopsis* due to the overexpression of the wheat *TNHX1* gene (Brini et al., 2007).

14.3.2.2 Drought Tolerance

Plants that are under drought stress experienced a number of significant morphological, physiological, molecular level and biochemical changes which results in lower yields of crop plants. The plants' root architecture such as root diameter, length, density is important in case to avoid drought stress because all the water, nutrient's absorption and translocation started from the roots (Sánchez-Bermúdez et al., 2022). In wheat plants there is a presence of Deeper Rooting 1 (*TaDRO1*) gene, which provides rooting system that provide higher tolerance to drought stress (Kitomi et al., 2020). Several genes/transcription factors (TFs) such as *TaNAC69-1*, *TaRNAC1*, and *TaMPS* in wheat crop contribute in regulating the root growth such as increase in length, biomass, and number of lateral roots under water deficiency or during drought stress conditions (Chen et al., 2018). When soil water is not abundant due to no or lesser rains, abscisic acid (ABA) is crucial for controlling stomatal closure, which reduces transpiration process in plants (Park et al., 2009). The pyrabactin resistance (PYL) proteins act as phytohormone (ABA) receptors and the overexpression of *PYL4* and *PYL1-1B* genes in wheat plants enhanced their resistance to drought stress and increased the yield during drought (Mao et al., 2022).

14.3.2.3 Role of Dimethylsulfoniopropionate (DMSP) in Salinity and Drought Tolerance

An organosulfur molecule called DMSP has been linked to abiotic stress tolerance in perennial grasses (*Spartina anglica*) found at salt marshes (Payet et al., 2024). The accumulation of DMSP in *Arabidopsis thaliana* and *N. benthamiana* plants provides tolerance for salinity and drought stresses as well as protection from oxidative and osmotic stresses, due to the higher expression of DMSP biosynthesis genes such as *methionine S-methyltransferase* (*MMT*), *S-methylmethionine decarboxylase* (*SDC*) and *DMSP-amine oxidase* (*DOX*) of *S. anglica* plants (Payet et al., 2024). DMSP synthesis is widespread in the crop or other plants, but, at lower level or concentration (Payet et al., 2024). The level of DMSP can be increased by bioengineering in crop plants in the future, and higher accumulation of DMSP would improve the tolerance against abiotic stresses (salt stress, oxidative and osmotic stress) which can lead for higher crop productivity (Payet et al., 2024).

14.3.2.4 High Temperature Tolerance

HT stress has a greater effect on yield loss during reproductive phases such as flower development, pod setting, or grain-filling (Parent et al., 2017). In addition to being important for heat tolerance, plasma membrane integrity entails modification in shapes of cell membranes, together with the activity of its proteins by adjusting the lipid content and their compositions (Ding & Yang, 2022). In a research study, HT (35°C) exposure of chickpea plants for three days (72 h) caused many morphological damages such as chlorosis of leaves, drying and abortion of flowers, reduced seed filling/pod set and lowered sucrose, starch contents in leaves of plants due to the down-regulation of majority of sucrose and starch metabolism genes during heat stress (Chandel & Sharma, 2023).

HT stress tolerance in plants is a polygenic trait that needs chaperone proteins to shield and restore heat-labile proteins and enzymes to their original state so that they can function correctly and effectively. These proteins include signal proteins, heat-responsive sensors, thermo-protective biochemical metabolites, and other heat shock-TFs (Ding et al., 2020). Heat shock-TFs and phytohormones (ethylene, ABA, jasmonic acid, etc.) related to stress conditions together regulate heat response mechanisms with respect to high-temperature stress in *Arabidopsis* (Li et al., 2018).

14.3.2.5 Pre-Harvest Sprouting Tolerance

In pre harvest sprouting (PHS), grains are germinable prior to harvesting of crop. Rainy and wet weather conditions prevailed during the terminal stages enhances the germination of grains on the parent plants (Rodríguez et al., 2015). In most wheat-growing regions of the world, PHS significantly reduced wheat quality and yield during its terminal stage (Kandpal et al., 2024). The seed dormancy and PHS can be regulated by argonaute (AGO) proteins in crops and wheat PHS resistance has been linked to the *AGO802B* gene (Kandpal et al., 2024). A short interspersed nuclear element (SINE) could be used as a molecular marker for PHS in wheat genome and the insertion of SINE retrotransposon at 3′-UTR of the *TaAGO802B* argonaute gene revealed that 92.6% of the wheat resistant cultivar subgroup had it, which decreased the *TaAGO802B* gene's expression (Kandpal et al., 2024). RNA-directed DNA methylation pathways reduced the rate of methylation by 54.7% with SINE insertion in the resistant cultivars, and these results suggested that SINE insertion in *TaAGO802B* gene can serve as a genetic marker for wheat germplasm screening and a breeding tool for PHS-resistant wheat cultivars, both of which can improve global food security (Kandpal et al., 2024).

14.3.3 BIOTECHNOLOGY-BASED DISEASE AND PEST MANAGEMENT

Global climate change increases the probability of biotic stresses by encouraging the growth of new, more virulent pathogen populations and increasing the spread of plant infections to new areas as a result of more frequent disease outbreaks. The production of crops resistant to a wide range of pathogens, including bacteria, viruses, and insects, that cause devastating diseases in agricultural plants has been made possible by the application of biotechnology and its methods in agriculture. Microbial pathogens caused about 22% losses each year (Mao et al., 2023).

14.3.3.1 Virus Resistance

The viral diseases in plants have posed the greatest threat and major challenge for their control in front of modern agricultural practices (Das et al., 2023). The methods such as viral vectors eradication and destroying of infected plants for the control and management of viral diseases in plants are not much successful and require biotechnological approaches to engineer and alter plants to make them resistant to viral diseases (Wilson, 1993). These strategies include microRNA-mediated resistance, homology-dependent gene silencing techniques, an RNA-mediated defense and the development of viral coat protein-based resistance against viral infections in plant crops (Wilson, 1993). The rainbow papaya is a case study of viral resistance, which is a genetically engineered horticultural crop developed for the farmers at Hawaii, USA, because, the papaya ring spot virus (PRSV) caused significant losses and threatened the Hawaiian papaya industry previously (Gonsalves et al., 2004).

14.3.3.2 Insects, Bacterial and Herbicide Resistance

The breeding of crops resistant to bacteria and insects was primarily made possible by agricultural biotechnology. Resistance in most crops is triggered by '*cry*' genes taken from the *Bacillus thuringenesis* (*Bt*) bacteria (Tabashnik et al., 2013). However, other gene sources like *Nicotiana tabacum* histidine kinase-1; levansucrase-encoding gene; jasmonic ethylene responsive factors; arrowhead proteinase inhibitors; vitreoscilla hemoglobin, and *betadine aldehyde dehydrogenase* gene have also been utilized in the development of insect-resistant crops (Wang et al., 2018). The most commonly grown transgenic plants include maize (*Zea mays*), which is resistant to larvae of both lepidopteran and coleopteran insects and cotton (*Gossypium hirsutum*), which is immune to caterpillars (Brookes & Barfoot, 2005). The application of these transgenic crops in agricultural practices has resulted in less insecticidal use and lowered the production costs, which results in higher yields of crops. Bacteria are present everywhere on earth and can cause various diseases in plants. The fire blight caused by *Erwinia amylovora*; crown gall disease in some plants, caused by *Agrobacterium tumefaciens*; brown rot caused by *Ralstonia solanacearum*; black rot is caused by *Xanthomonas campestris* in cabbage.

Bacterial wilt disease caused by *R. solanacearum* is common in Solanaceae plants and according to a recent study, *SmDDA1b* is a new regulator that works through the SmNAC-mediated salicylic acid pathway to protect Solanaceous crops from bacterial wilt (Yan et al., 2024). Weeds are present in agricultural fields in huge amounts because the dormancy of their seeds is very high so that they germinate in numerous quantities during their favorable seasons which create a major problem to farmers. Weeds reduce agricultural plant yields by competing with crop plants for resources, including water, nutrients, sunlight, and space (Das et al., 2023). Also, the weeds are alternate hosts to various pests, insects, and many pathogenic microorganisms which attack the crop plants and decreased the yields (Tan et al., 2006).

14.3.4 BIOFORTIFICATION: ENHANCING NUTRITIONAL VALUES OF CROPS TO REDUCE MICRONUTRIENT MALNUTRITION

The primary food crops in poor nations, cereals and pulses, frequently lack important macronutrients and micronutrients, resulting in nutritional imbalance. This lack of access to balanced diets leading to malnutrition is directly related to anemia, rickets, scurvy, etc. (Sandhu et al., 2023). It has been reported in case of children mortality rate that zinc deficiency causes 0.4 million deaths, whereas vitamin A deficiency causes 0.6 million deaths (Black et al., 2008). Maternal mortality as a result of anemia led to 115,000 deaths and 0.4% of total disability-adjusted life-years globally (Black et al., 2008). Recent advances in biotechnology in Omics, whole genome sequencing, RNA-mediated genome editing, foreign gene transfer, overexpression of genes, and CRISPR-based technologies are some of the new methods being used to produce biofortified crops (Raza et al., 2021; Sandhu et al., 2023).

These tools target metabolic pathways, redistribute micronutrients in tissues, and also improve their bioavailability and results in improved macromolecules. Biofortification of readily available affordable staple food crops such as pulses and cereals is needed to provide balanced diet to growing population and reduce malnutrition (Sandhu et al., 2023). Biofortification, which uses transgenic techniques to increase the amount of minerals and vitamins in staple food crops, is the only and less expensive way to address such malnutrition issues (Bouis et al., 2011). Regarding the rice crop, a significant staple food, the endosperm, which is edible after milling, is deficient in vital minerals, including provitamin A (β-carotene). As a result, it causes severe vitamin A insufficiency, especially in Asia, Africa, and Latin America, which are major rice-consuming nations (Ye et al., 2000). Development of golden rice is using recombinant DNA technology where combination of three transgenes enable β-carotene biosynthesis in rice endosperm and it is the best achievement of biotechnology (Ye et al., 2000). Likewise, numerous other crops, including rice, wheat, beans, pearl millet, and cassava, have been biofortified for iron and zinc, and many additional crops, including sweet potatoes, cassava, and maize, have been biofortified for provitamin A (Bouis et al., 2011).

14.3.4.1 Gene Editing to Produce Oats with Improved Nutritional Value and Shelf Life

Oats (*Avena sativa*) are well known for their high fiber content and oats with enhanced nutritional values was developed (Zhou et al., 2024). Two constructs having three important genes of lipid biosynthesis from *Arabidopsis* and *Sesame* were introduced into the oat cultivar ‘Park’ for the alteration of composition of fatty acids. The genes were *WRINKLED1* (*AtWRI1*) and *diacylglycerol acyltransferase* (*AtDGAT1*) from *Arabidopsis* and the gene *OLEOSIN* (*SiOLEOSIN*) was taken from *Sesame* (Zhou et al., 2024). The content of oleic acid was increased (34%) in transgenic oat grains; while, the saturated fatty acid contents was reduced (Zhou et al., 2024). Healthy fats like oleic acid are well known for their heart-healthy properties and ability to reduce blood sugar levels. Such new innovations are significant for both the agricultural industry and consumer health, offering a novel approach to the development of nutrient-rich crops.

14.3.5 Precision Agriculture Using Biotechnology-Derived Tools

Precision farming integrates technology and data analytics with biotechnology to optimize agricultural practices, enabling sustainable high-yield crop production through genetic modification, gene editing, and marker-assisted breeding (Daware et al., 2023). Advanced tools like digital sensors, drones, GPS, and irrigation systems allow farmers to monitor fields efficiently, improving decision-making in irrigation, fertilization, and pest control. These precision techniques enhance crop productivity and farm profitability while reducing waste and environmental harm. Nanotechnology further supports precision agriculture through biosensors and nanoparticle-mediated delivery systems, ensuring controlled release of fertilizers and early virus detection (Duhan et al., 2017).

14.4 BIOTECHNOLOGY FOR SUSTAINABLE FARMING PRACTICES

One of the important biotechnological solutions in this aspect is the use of bioremediation techniques in removing hazardous pollutants from agricultural soils. It involves use of specific microbes to breakdown harmful contaminants and pollutants into harmless ones, thereby restoring soil ecosystem health at polluted sites and making land suitable for cultivation once again (Sreedevi et al., 2022; Yaashikaa & Kumar, 2022). Furthermore, biodegradable pesticides and fertilizers have also been created using biotechnology. Agricultural waste-led biofuel development also promoted more ecofriendly and sustainable agriculture practice due to reduced greenhouse gas emissions (Mekunye & Makinde, 2024). More studies on how underground microbiomes/microbial communities regulate the growth of plants in presence of various environmental cues and how rhizosphere near to plant roots are beneficial in maximizing the plant yields (Xiong et al., 2020).

14.5 CHALLENGES IN BIOTECHNOLOGY

Biotechnology offers significant developmental potential but faces challenges, including public misinformation, unpredictable outcomes, and regulatory hurdles that must keep pace with technological advances (Singh, 2000). Technical challenges, such as the need for highly skilled personnel, and economic issues, like the displacement of traditional exports by genetically engineered alternatives (e.g., jute fibers replaced by synthetics), hinder progress in developing nations. Addressing these economic disparities requires bilateral negotiations and public education on the value of natural versus engineered products. Ethical concerns, including perceived interference with nature and debates over patents on life forms or cloning, further complicate acceptance (Singh, 2000). Balancing these moral dilemmas with biotechnology's potential to alleviate hunger and malnutrition is critical (Black et al., 2008). Transparent dialogue and awareness campaigns are essential to resolving contradictions and fostering broader adoption of biotechnological solutions.

14.6 CONCLUSION

Biotechnology offers transformative solutions for sustainable agriculture, enabling genetic modifications to enhance stress tolerance, disease resistance, and nutrient content in crops, thereby boosting yields and food quality. Advanced techniques like CRISPR-Cas9 and CRISPR-Cas12a allow precise gene editing without introducing foreign DNA, creating resilient and high-yielding cultivars. Additionally, biotechnological innovations such as precision farming tools, bioremediation, biodegradable pesticides, and biofuel production from agricultural waste contribute to sustainable practices. Developing climate-resilient crops and biofortified varieties can address food security and malnutrition amid climate change. However, realizing biotechnology's full potential requires overcoming ethical concerns, regulatory challenges, and ensuring equitable access to these technologies. Collaborative efforts between governments, research institutions, and stakeholders are essential to promote sustainable and resilient agriculture for future generations.

REFERENCES

Babar, M. M., Zaidi, N. U. S. S., Azooz, M. M., & Kazi, A. G. (2015). Biotechnology approaches to overcome biotic and abiotic stress constraints in legumes. In *Legumes under environmental stress: Yield, improvement and adaptations* (pp. 247–264). Wiley.

Barrows, G., Sexton, S., & Zilberman, D. (2014). Agricultural biotechnology: The promise and prospects of genetically modified crops. *Journal of Economic Perspectives*, *28*(1), 99–120. https://doi.org/10.1257/jep.28.1.99

Black, R. E., Allen, L. H., Bhutta, Z. A., Caulfield, L. E., De Onis, M., Ezzati, M., Mathers, C., & Rivera, J. (2008). Maternal and child undernutrition: Global and regional exposures and health consequences. *The Lancet*, *371*(9608), 243–260. https://doi.org/10.1016/S0140-6736(07)61690-0

Bouis, H. E., Hotz, C., McClafferty, B., Meenakshi, J. V., & Pfeiffer, W. H. (2011). Biofortification: A new tool to reduce micronutrient malnutrition. *Food and Nutrition Bulletin*, *32*(1_suppl1), S31–S40. https://doi.org/10.1177/15648265110321S105

Brini, F., Hanin, M., Mezghani, I., Berkowitz, G. A., & Masmoudi, K. (2007). Overexpression of wheat Na+/H+ antiporter TNHX1 and H+-pyrophosphatase TVP1 improve salt- and drought-stress tolerance in *Arabidopsis thaliana* plants. *Journal of Experimental Botany*, *58*(2), 301–308. https://doi.org/10.1093/jxb/erl251

Brookes, G., & Barfoot, P. (2005). *GM crops: The global economic and environmental impact—The first nine years 1996–2004*. PG Economics.

Brookes, G., & Barfoot, P. (2017). Farm income and production impacts of using GM crop technology 1996–2015. *GM Crops and Food*, *8*(3), 156–193.

Chandel, S. S., & Sharma, K. D. (2023). Down-regulation of carbohydrate metabolic pathway genes lowers sucrose and starch content in chickpea leaves under high temperature stress. *National Academy Science Letters*, *46*(5), 445–449.

Chen, D., Chai, S., McIntyre, C. L., & Xue, G. P. (2018). Overexpression of a predominantly root-expressed NAC transcription factor in wheat roots enhances root length, biomass and drought tolerance. *Plant Cell Reports*, *37*, 225–237. https://doi.org/10.1007/s00299-017-2225-x

Christoforidou, M., Borghuis, G., Seijger, C., van Halsema, G. E., & Hellegers, P. (2023). Food security under water scarcity: A comparative analysis of Egypt and Jordan. *Food Security*, *15*(1), 171–185. https://doi.org/10.1007/s12571-022-01310-y

Cramer, G. R., Urano, K., Delrot, S., Pezzotti, M., & Shinozaki, K. (2011). Effects of abiotic stress on plants: A systems biology perspective. *BMC Plant Biology*, *11*, 163. https://doi.org/10.1186/1471-2229-11-163

Das, S., Ray, M. K., Panday, D., & Mishra, P. K. (2023). Role of biotechnology in creating sustainable agriculture. *PLOS Sustainability and Transformation*, *2*(7), e0000069. https://doi.org/10.1371/journal.pstr.0000069

Daware, V., Mondal, K., & Khandagale, S. (2023). Precision agriculture and biotechnology. In *Modern horizons in agriculture* (p. 351). Springer.

Dill, G. M. (2005). Glyphosate-resistant crops: History, status and future. *Pest Management Science*, *61*(3), 219–224. https://doi.org/10.1002/ps.1008

Ding, Y., Shi, Y., & Yang, S. (2020). Molecular regulation of plant responses to environmental temperatures. *Molecular Plant*, *13*(4), 544–564. https://doi.org/10.1016/j.molp.2020.02.004

Ding, Y., & Yang, S. (2022). Surviving and thriving: How plants perceive and respond to temperature stress. *Developmental Cell*, *57*(8), 947–958.

Donatelli, M., Magarey, R. D., Bregaglio, S., Willocquet, L., Whish, J. P., & Savary, S. (2017). Modelling the impacts of pests and diseases on agricultural systems. *Agricultural Systems*, *155*, 213–224. https://doi.org/10.1016/j.agsy.2017.01.019

Duhan, J. S., Kumar, R., Kumar, N., Kaur, P., Nehra, K., & Duhan, S. (2017). Nanotechnology: The new perspective in precision agriculture. *Biotechnology Reports*, *15*, 11–23. https://doi.org/10.1016/j.btre.2017.03.002

Gonsalves, C., Lee, D., & Gonsalves, D. (2004). Transgenic virus-resistant papaya: The Hawaiian 'Rainbow' was rapidly adopted by farmers and is of major importance in Hawaii today. *APSnet Feature Articles*. http://www.apsnet.org/education/feature/papaya

Huang, S., Spielmeyer, W., Lagudah, E. S., James, R. A., Platten, J. D., Dennis, E. S., & Munns, R. (2006). A sodium transporter (HKT7) is a candidate for *Nax1*, a gene for salt tolerance in durum wheat. *Plant Physiology*, *142*(4), 1718–1727.

Hubbart, S., Smillie, I. R., Heatley, M., Swarup, R., Foo, C. C., Zhao, L., & Murchie, E. H. (2018). Enhanced thylakoid photoprotection can increase yield and canopy radiation use efficiency in rice. *Communications Biology*, *1*(1), 22. https://doi.org/10.1038/s42003-018-0023-9

Iñiguez, C., Aguiló-Nicolau, P., & Galmés, J. (2021). Improving photosynthesis through the enhancement of Rubisco carboxylation capacity. *Biochemical Society Transactions*, *49*(5), 2007–2019. https://doi.org/10.1042/BST20201056

Kandpal, P., Kaur, K., Dhariwal, R., Kaur, S., Brar, G. K., Randhawa, H., & Singh, J. (2024). Utilizing short interspersed nuclear element as a genetic marker for pre-harvest sprouting in wheat. *Plants*, *13*(21), 2981. https://doi.org/10.3390/plants13212981

Khush, G. S., Lee, S., Cho, J. I., & Jeon, J. S. (2012). Biofortification of crops for reducing malnutrition. *Plant Biotechnology Reports*, *6*, 195–202.

Kitomi, Y., Hanzawa, E., Kuya, N., Inoue, H., Hara, N., Kawai, S., Kanno, N., Endo, M., Sugimoto, K., Yamazaki, T., & Sakamoto, S. (2020). Root angle modifications by the *DRO1* homolog improve rice yields in saline paddy fields. *Proceedings of the National Academy of Sciences*, *117*(35), 21242–21250. https://doi.org/10.1073/pnas.2005911117

Kromdijk, J., Głowacka, K., Leonelli, L., Gabilly, S. T., Iwai, M., Niyogi, K. K., & Long, S. P. (2016). Improving photosynthesis and crop productivity by accelerating recovery from photoprotection. *Science*, *354*(6314), 857–861. https://doi.org/10.1126/science.aai8878

Kwak, S. S. (2019). Biotechnology of the sweetpotato: Ensuring global food and nutrition security in the face of climate change. *Plant Cell Reports*, *38*(11), 1361–1363. https://doi.org/10.1007/s00299-019-02468-0

Li, B., Gao, K., Ren, H., & Tang, W. (2018). Molecular mechanisms governing plant responses to high temperatures. *Journal of Integrative Plant Biology*, *60*(9), 757–779. https://doi.org/10.1111/jipb.12663

Ma, X., Mau, M., & Sharbel, T. F. (2018). Genome editing for global food security. *Trends in Biotechnology*, *36*(2), 123–127. https://doi.org/10.1016/j.tibtech.2017.08.004

Mao, H., Jian, C., Cheng, X., Chen, B., Mei, F., Li, F., Zhang, Y., Li, S., Du, L., Li, T., & Hao, C. (2022). The wheat ABA receptor gene *TaPYL1-1B* contributes to drought tolerance and grain yield by increasing water-use efficiency. *Plant Biotechnology Journal*, *20*(5), 846–861. https://doi.org/10.1111/pbi.13764

Mao, H., Jiang, C., Tang, C., Nie, X., Du, L., Liu, Y., Cheng, P., Wu, Y., Liu, H., Kang, Z., & Wang, X. (2023). Wheat adaptation to environmental stresses under climate change: Molecular basis and genetic improvement. *Molecular Plant*, *16*(10), 1564–1589. https://doi.org/10.1016/j.molp.2023.09.001

Mekunye, F., & Makinde, P. (2024). Production of biofuels from agricultural waste. *Asian Journal of Agricultural and Horticultural Research*, *11*(3), 37–49. https://doi.org/10.9734/ajahr/2024/v11i3328

Murchie, E. H., Ali, A., & Herman, T. (2015). Photoprotection as a trait for rice yield improvement: Status and prospects. *Rice*, *8*(1), 25. https://doi.org/10.1186/s12284-015-0057-2

Ogbaga, C. C., Stepien, P., Athar, H. U. R., & Ashraf, M. (2018). Engineering Rubisco activase from thermophilic cyanobacteria into high-temperature sensitive plants. *Critical Reviews in Biotechnology*, *38*(4), 559–572. https://doi.org/10.1080/07388551.2017.1378998

Parent, B., Bonneau, J., Maphosa, L., Kovalchuk, A., Langridge, P., & Fleury, D. (2017). Quantifying wheat sensitivities to environmental constraints to dissect genotype × environment interactions in the field. *Plant Physiology*, *174*(3), 1669–1682.

Park, S. Y., Fung, P., Nishimura, N., Jensen, D. R., Fujii, H., Zhao, Y., Lumba, S., Santiago, J., Rodrigues, A., Chow, T. F. F., & Alfred, S. E. (2009). Abscisic acid inhibits type 2C protein phosphatases via the PYR/PYL family of START proteins. *Science*, *324*(5930), 1068–1071. https://doi.org/10.1126/science.1173041

Payet, R. D., Bilham, L. J., Kabir, S. M. T., Monaco, S., Norcott, A. R., Allen, M. G., Zhu, X. Y., Davy, A. J., Brearley, C. A., Todd, J. D., & Miller, J. B. (2024). Elucidation of *Spartina* dimethylsulfoniopropionate synthesis genes enables engineering of stress-tolerant plants. *Nature Communications*, *15*(1), 8568. https://doi.org/10.1038/s41467-024-51758-z

Ray, D. K., Mueller, N. D., West, P. C., & Foley, J. A. (2013). Yield trends are insufficient to double global crop production by 2050. *PLoS ONE*, *8*(6), e66428. https://doi.org/10.1371/journal.pone.0066428

Raza, A., Tabassum, J., Kudapa, H., & Varshney, R. K. (2021). Can omics deliver temperature resilient ready-to-grow crops? *Critical Reviews in Biotechnology*, *41*(8), 1209–1232. https://doi.org/10.1080/07388551.2021.1898332

Rodríguez, M. V., Barrero, J. M., Corbineau, F., Gubler, F., & Benech-Arnold, R. L. (2015). Dormancy in cereals (not too much, not so little): About the mechanisms behind this trait. *Seed Science Research*, *25*(2), 99–119. https://doi.org/10.1017/S0960258515000021

Sánchez-Bermúdez, M., Del Pozo, J. C., & Pernas, M. (2022). Effects of combined abiotic stresses related to climate change on root growth in crops. *Frontiers in Plant Science*, *13*, 918537. https://doi.org/10.3389/fpls.2022.918537

Sandhu, R., Chaudhary, N., Shams, R., Singh, K., & Pandey, V. K. (2023). A critical review on integrating biofortification in crops for sustainable agricultural development and nutritional security. *Journal of Agriculture and Food Research*, *14*, 100830.

Singh, R. B. (2000, June). *Biotechnology, biodiversity and sustainable agriculture—A contradiction*. Paper presented at the Regional Conference in Agricultural Biotechnology, Bangkok, Thailand.

Sreedevi, P. R., Suresh, K., & Jiang, G. (2022). Bacterial bioremediation of heavy metals in wastewater: A review of processes and applications. *Journal of Water Process Engineering*, *48*, 102884. https://doi.org/10.1016/j.jwpe.2022.102884

Tabashnik, B. E., Brévault, T., & Carrière, Y. (2013). Insect resistance to Bt crops: Lessons from the first billion acres. *Nature Biotechnology*, *31*(6), 510–521. https://doi.org/10.1038/nbt.2597

Tan, S., Evans, R., & Singh, B. (2006). Herbicidal inhibitors of amino acid biosynthesis and herbicide-tolerant crops. *Amino Acids*, *30*(2), 195–204. https://doi.org/10.1007/s00726-005-0254-1

Thorpe, T. A. (2007). History of plant tissue culture. *Molecular Biotechnology*, *37*(2), 169–180. https://doi.org/10.1007/s12033-007-0031-3

Tyczewska, A., Twardowski, T., & Woźniak-Gientka, E. (2023). Agricultural biotechnology for sustainable food security. *Trends in Biotechnology*, *41*(3), 331–341. https://doi.org/10.1016/j.tibtech.2022.12.013

Verma, A. S., Agrahari, S., Rastogi, S., & Singh, A. (2011). Biotechnology in the realm of history. *Journal of Pharmacy and Bioallied Sciences*, *3*(3), 321–323. https://doi.org/10.4103/0975-7406.84430

Wang, G., Dong, Y., Liu, X., Yao, G., Yu, X., & Yang, M. (2018). The current status and development of insect-resistant genetically engineered poplar in China. *Frontiers in Plant Science*, *9*, 1408. https://doi.org/10.3389/fpls.2018.01408

Wei, S., Li, X., Lu, Z., Zhang, H., Ye, X., Zhou, Y., Li, J., Yan, Y., Pei, H., Duan, F., & Wang, D. (2022). A transcriptional regulator that boosts grain yields and shortens the growth duration of rice. *Science*, *377*(6604), eabi8455. https://doi.org/10.1126/science.abi8455

Wieczorek, A. M., & Wright, M. G. (2012). History of agricultural biotechnology: How crop development has evolved. *Nature Education Knowledge*, *3*(10), 9.

Wilson, T. M. (1993). Strategies to protect crop plants against viruses: Pathogen-derived resistance blossoms. *Proceedings of the National Academy of Sciences*, *90*(8), 3134–3141. https://doi.org/10.1073/pnas.90.8.3134

Xiong, W., Song, Y., Yang, K., Gu, Y., Wei, Z., Kowalchuk, G. A., Xu, Y., Jousset, A., Shen, Q., & Geisen, S. (2020). Rhizosphere protists are key determinants of plant health. *Microbiome*, *8*(1), 1–9. https://doi.org/10.1186/s40168-020-00799-9

Yaashikaa, P. R., & Kumar, P. S. (2022). Bioremediation of hazardous pollutants from agricultural soils: A sustainable approach for waste management towards urban sustainability. *Environmental Pollution*, *312*, 120031.

Yali, W. (2022). Application of genetically modified organism (GMO) crop technology and its implications in modern agriculture. *International Journal of Agricultural Science and Food Technology*, *8*(1), 14–20. https://dx.doi.org/10.17352/2455-815X.000139

Yan, S., Wang, Y., Yu, B., Gan, Y., Lei, J., Chen, C., Zhu, Z., Qiu, Z., & Cao, B. (2024). A putative E3 ubiquitin ligase substrate receptor degrades transcription factor SmNAC to enhance bacterial wilt resistance in eggplant. *Horticulture Research*, *11*(1), uhad246.

Ye, X., Al-Babili, S., Kloti, A., Zhang, J., Lucca, P., Beyer, P., & Potrykus, I. (2000). Engineering the provitamin A (β-carotene) biosynthetic pathway into (carotenoid-free) rice endosperm. *Science*, *287*(5451), 303–305. https://doi.org/10.1126/science.287.5451.303

Zhao, C., Liu, B., Piao, S., Wang, X., Lobell, D. B., Huang, Y., Huang, M., Yao, Y., Bassu, S., Ciais, P., & Durand, J. L. (2017). Temperature increase reduces global yields of major crops in four independent estimates. *Proceedings of the National Academy of Sciences*, *114*(35), 9326–9331. https://doi.org/10.1073/pnas.1701762114

Zheng, X., Tang, X., Wu, Y., Zheng, X., Zhou, J., Han, Q., Tang, Y., Fu, X., Deng, J., Wang, Y., & Wang, D. (2024). An efficient CRISPR-Cas12a-mediated MicroRNA knockout strategy in plants. *Plant Biotechnology Journal*. https://doi.org/10.1111/pbi.14484

Zhou, Y., Lai, Z., Yin, X., Yu, S., Xu, Y., Wang, X., Cong, X., Luo, Y., Xu, H., & Jiang, X. (2016). Hyperactive mutant of a wheat plasma membrane Na+/H+ antiporter improves the growth and salt tolerance of transgenic tobacco. *Plant Science*, *253*, 176–186. https://doi.org/10.1016/j.plantsci.2016.09.017

Zhou, Z., Kaur, R., Donoso, T., Ohm, J. B., Gupta, R., Lefsrud, M., & Singh, J. (2024). Metabolic engineering-induced transcriptome reprogramming of lipid biosynthesis enhances oil composition in oat. *Plant Biotechnology Journal*. Advance online publication. https://doi.org/10.1111/pbi.14467

15 Socio-Economic Implications of Biotech Innovations (GMOs Regulations)

Ensuring Inclusivity and Sustainability

Shilpa Bahubalendra, Jeebanjyoti Behera, and Angelina Patro

15.1 INTRODUCTION

Biotechnology integrates biological science with technological innovation, driving advancements in genetically modified (GM) organisms (GMOs), CRISPR-Cas9 gene editing, synthetic biology, and bioinformatics (Lubogo, 2024). Its applications span healthcare, agriculture, and environmental management, contributing to a global market valued at USD 1.37 trillion in 2021, with continued rapid growth (Ashraf et al., 2021; Priya & Kappalli, 2022). Since the first commercial GM crop tobacco in China (1992), biotechnology has revolutionized agriculture, becoming the fastest-adopted innovation in farming history. Today, GM crops like soybeans, maize, and cotton dominate global cultivation, benefiting millions of farmers, particularly in developing nations (Bakhsh et al., 2021; James, 2011). However, despite its success, biotechnology faces regulatory and socio-economic challenges that influence its adoption. The expansion of GM crops has introduced diverse traits, including herbicide tolerance, pest resistance, and enhanced nutrition (Bacelar et al., 2024; Perotti et al., 2020). Yet, market access remains a key hurdle, as governments impose varying regulatory frameworks, some science-based, others incorporating socio-economic considerations (SECs) (De Francesco, 2021; Kuhlmann & Dey, 2021).

The Cartagena Protocol on Biosafety (CPB) allows voluntary SEC integration in biosafety assessments, yet implementation remains inconsistent (Falck-Zepeda, 2009; Gebreyes, 2023). Policymakers struggle with limited data on SEC impacts, complicating regulatory decisions (Ou & Guo, 2023). This gap highlights the need for structured guidelines to assess SECs effectively while complying with international agreements. This chapter explores methodologies for evaluating SECs

DOI: 10.1201/9781003597797-15

in biotechnology regulation, addressing legal, economic, and ethical dimensions (Demont et al., 2008; EFSA, 2020). It does not advocate for or against SEC inclusion but provides a framework for policymakers to make informed decisions (Brighouse et al., 2018). By analyzing costs, benefits, and trade-offs, this work aims to support efficient policy development, ensuring biotechnology advances sustainably and equitably. The key question remains: *What is the ultimate goal of integrating SECs into regulatory frameworks?*—a debate requiring context-specific answers from global stakeholders.

15.2 INCLUSIVITY AND SUSTAINABILITY

To ensure that biotech innovations effectively promote inclusive growth and enhance sustainability, it is essential that their advantages reach everyone, particularly marginalized communities. Furthermore, although biotechnology offers the potential for environmental sustainability, it is essential that its application does not endanger ecosystems or worsen social disparities (Leach et al., 2018). This part explores the social and economic impacts of biotechnology, emphasizing its potential to foster inclusivity and sustainability, while also tackling the challenges that must be overcome to ensure fair access to these benefits. Moreover, while biotechnology holds promise for promoting environmental sustainability, it is crucial to ensure that its implementation does not jeopardize ecosystems or exacerbate social inequalities (Ahmad et al., 2022).

Biotechnology has emerged as a significant catalyst for economic advancement in nations across the spectrum of development. The impact is clear across multiple fields: GM crops have boosted agricultural productivity, reduced crop losses, and decreased chemical use, enhancing farmer profits and contributing to a global biotech crop market valued at over USD 25 billion in 2020 (ISAAA, 2017). In healthcare, biotechnology has enabled breakthroughs such as monoclonal antibodies, transforming treatments for cancer and autoimmune diseases while generating significant economic benefits (Evens, 2022). Industrial biotechnology drives sustainable solutions in biofuels, bioplastics, and waste management, with the bioeconomy projected to reach USD 5 trillion by 2030 (Kircher, 2019). However, gender disparities persist, particularly in developing nations where women, despite their crucial role in agriculture, often lack access to biotech advancements (Evens, 2022). For instance, in Burkina Faso, while Bt cotton improved yields, many women farmers faced financial and technical barriers, limiting their ability to benefit fully from these innovations (Evens, 2022).

15.3 GMOS REGULATION

15.3.1 International Context of Socio-Economic Consideration and GMOs Regulations

The Interorganisational Committee on Principles and Guidelines for Social Impact Assessment highlights the importance of evaluating how public and private actions affect human populations, including shifts in lifestyle, work, culture, and societal values (Inglehart, 2018). The Ad Hoc Technical Group on SECs (AHTEG-SECs) under the Convention on Biological Diversity acknowledges the lack of a standardized definition for "socio-economic considerations," which encompass economic, social, ecological, cultural, and health-related dimensions (Chaturvedi et al., 2019).

These factors are increasingly integrated into global frameworks like the CPB (Article 26) and national regulations, such as Norway's Gene Technology Act (1993), emphasizing the need to assess GMOs' sustainability, agricultural impacts, and societal benefits (Mol et al., 2020; Munhoz, 2023). At the European level, initiatives like the 2011 FAO-JRC workshop and the 2012 European Environmental Agency seminar have advanced the discourse on socio-economic assessments for GMOs (Casadella & Uzunidis, 2023). The 2015 EU Directive (2015/412) further empowered member states to restrict GMO cultivation based on socio-economic, agricultural, or public policy concerns, aligning with the Cartagena Protocol's Strategic Plan (2011–2020) to enhance decision-making through research and stakeholder input (Catacora-Vargas et al., 2018).

The CBD Secretariat's Ad Hoc Technical Expert Group (AHTEG-SEC) continues to refine methodologies for integrating socio-economic factors into biosafety governance, emphasizing robust frameworks with clear indicators and participatory approaches (CBD, UN 2014; Evens, 2022). Despite progress, debates persist over the scope, methods, and timing of socio-economic assessments, particularly regarding public participation and the precautionary principle (Evens, 2022; Stabinsky, 2000). Studies by Spök (2010) and Falck-Zepeda reveal gaps in national regulatory practices, with only 12–18 countries systematically evaluating socio-economic impacts. A broader CBD-commissioned analysis of 34 nations underscores the need for standardized criteria and empirical data to inform GMO governance, highlighting the role of public engagement in balancing innovation with societal equity (Ludlow et al., 2013).

15.4 ANALYSIS OF SOCIO-ECONOMIC CONSIDERATION

15.4.1 Basis for Inclusion of Socio-Economic Factors

A total of 34 countries incorporate SECs into their national biosafety frameworks, with 14 African nations (e.g., Burkina Faso, Kenya, South Africa), 9 Latin American countries (e.g., Brazil, Mexico, Argentina), and 6 Asia-Pacific states (e.g., Malaysia, Philippines) represented. Four European countries like, France, Italy, Latvia, and Norway, also integrate SECs, alongside others like Bangladesh and India, though specific laws were not always documented (Evens, 2022). Spök's research notes exceptions such as the Syrian Arab Republic excluded due to Arabic-only regulations, and Belize omitted for similar accessibility constraints. While 18 countries lack practical experience implementing SECs, 15 others report varying levels of application, with Uruguay's experience remaining unspecified. The study highlights gaps in standardized implementation, particularly in nations like Nigeria and Uganda, where SECs are legally recognized but operational frameworks are underdeveloped. These findings underscore the uneven global progress in institutionalizing socio-economic assessments within GMO governance (Evens, 2022).

15.4.2 Advantages for Producers and Society in the Analysis of SEC

Socio-economic analysis (SEC) provides valuable insights into the relationships between economic activities, social structures, and environmental sustainability, helping producers optimize resources, reduce costs, and improve productivity while promoting long-term sustainability (Evens, 2022). This approach enhances workforce management and fosters inclusive economic participation, particularly for marginalized groups like smallholder farmers, while informing public policy on income distribution and regional development (Evens, 2022).

Agricultural biotechnology, particularly GMOs, offers significant socio-economic benefits by increasing crop yields, reducing pesticide use, and developing climate-resilient varieties, thereby improving food security and farmer incomes (Ludlow et al., 2013). GMOs also contribute to environmental sustainability by lowering chemical inputs and greenhouse gas emissions while helping farmers adapt to climate challenges (Ludlow et al., 2013). Effective regulatory frameworks are crucial to ensure biotechnological advancements align with public health, environmental protection, and ethical standards, requiring socio-economic assessments to balance innovation with societal needs (Canton, 2021; Casadella & Uzunidis, 2023). Integrating SEC into GMO governance promotes equitable agricultural development while addressing concerns about food sovereignty, biodiversity, and long-term sustainability (Canton, 2021).

15.5 POLICY FRAMEWORKS: ENSURING INCLUSIVITY AND SUSTAINABILITY

15.5.1 Regulatory Landscape for Biotechnology

The oversight of biotechnology is essential for guaranteeing its safe and fair application. Countries around the world have taken distinct paths in regulating biotechnology, showcasing a range of perspectives on the potential risks and advantages associated with GMOs, gene editing, and other innovations in the field. In the U.S., the regulatory framework for biotechnology is notably accommodating, with the U.S. The USDA, EPA, and FDA collaborate in overseeing various responsibilities (Bernier, 2019). Conversely, the European Union (EU) has embraced a more cautious strategy, implementing stringent regulations that oversee the approval and utilization of GMOs. Countries with emerging economies encounter distinct obstacles in overseeing biotechnology. The challenges posed by limited regulatory capacity, along with the powerful presence of multinational corporations, create significant obstacles for these nations in their efforts to manage the risks linked to biotech innovations. Global agreements, like the CPB, offer direction for the secure transfer, management, and application of GMOs; however, the execution varies significantly among nations (Canton, 2021)

15.5.2 Intellectual Property Rights and Biotech Access

The topic of intellectual property rights in biotechnology is a subject of significant debate. On one side, safeguarding intellectual property is essential to motivate innovation by enabling biotech firms to recover their expenditures in research and development (Canton, 2021). Conversely, stringent intellectual property safeguards may restrict access to biotechnological advancements, especially in nations with developing economies. This has prompted discussions about different approaches to intellectual property protection, like open-source biotechnology, which could enhance access to biotech advancements while still preserving the motivation for innovation (Bernier, 2019).

15.5.3 Public-Private Partnerships (PPPs) for Biotech Development

PPPs have become essential for fostering inclusive and sustainable advancements in biotechnology. Utilizing the strengths and knowledge from both public and private sectors, collaborative partnerships can facilitate the delivery of biotechnology advantages to underserved communities. For instance, the Alliance for a Green Revolution in Africa (AGRA) represents a collaboration among governments, NGOs, and the private sector focused on advancing agricultural development in Africa by leveraging biotech innovations. AGRA has dedicated efforts to ensure that smallholder farmers gain access to enhanced seeds, fertilizers, and training, emphasizing sustainability and inclusivity (Wise, 2020).

15.5.4 Case Study: India's Biotech Policy

India has developed a robust biotech policy that seeks to balance innovation with sustainability and inclusivity. The country has invested heavily in biotechnology research, particularly in agriculture, where GM crops such as Bt cotton have been widely adopted. India's regulatory framework for biotechnology, overseen by the Genetic Engineering Appraisal Committee (GEAC), aims to ensure the safe use of biotech innovations while promoting access for smallholder farmers. Despite these challenges, India's experience with biotechnology offers valuable lessons for other developing countries seeking to harness the potential of innovations for sustainable development (John & Panda, 2024).

15.5.5 Impact Assessment

Agricultural biotechnology and GMOs offer significant benefits for modern agriculture but raise important concerns regarding health risks (including allergenicity and long-term effects), biodiversity loss (through gene transfer and ecosystem disruption), and labor impacts (Canton, 2021). While GMOs can increase productivity and reduce resource needs, they may also displace rural workers and threaten traditional farming practices, requiring careful socio-economic evaluation to prevent widening inequalities. The potential for genetic contamination of wild species and reduction in crop diversity underscores the need for strict regulatory oversight of these technologies. Effective GMO policies must balance innovation with protections for public health, environmental sustainability, and labor equity, particularly for vulnerable smallholder farmers. Comprehensive assessments integrating socio-economic, health, and environmental factors are essential to ensure agricultural biotechnology contributes to sustainable and inclusive food systems (Canton, 2021).

15.5.6 MARKET ACCESS AND TRADE

The adoption of agricultural biotechnology and GMOs is shaped by complex interactions between market access, trade policies, and socio-economic factors, with significant potential to enhance food security and crop resilience (Inglehart, 2018). However, divergent international regulations create trade barriers, disproportionately affecting GMO-exporting developing nations and potentially limiting market opportunities (Inglehart, 2018). While GMOs offer smallholder farmers yield improvements and cost reductions, concerns persist about corporate dominance in seed markets and intellectual property issues (Mol et al., 2020). Developing countries face particular challenges in balancing GMO adoption benefits with compliance to international biosafety standards like the Cartagena Protocol (Mol et al., 2020). These regulatory differences often necessitate parallel supply chains (GMO/non-GMO), increasing costs and market access for producers in emerging economies (Mol et al., 2020).

15.6 CHALLENGES

Access to biotechnology remains a significant challenge in developing nations due to high development costs and corporate patent controls, particularly for GM seeds, which restrict smallholder farmers' autonomy and increase production expenses (Chaturvedi et al., 2019). Strict biosafety regulations, often prioritizing environmental and health risks over socio-economic impacts, further complicate adoption, with developing countries facing enforcement gaps due to limited infrastructure and resources (Ray et al., 2020). The lack of harmonized global standards creates trade barriers, disproportionately affecting small-scale farmers who struggle to meet technical and financial requirements for GMO compliance. Socio-ecological systems (SES) analysis reveals tensions between biotech adoption and traditional farming practices, biodiversity conservation, and community resilience, necessitating governance frameworks that balance innovation with equity (Chatzopoulou & Chatzopoulos, 2024). Corporate dominance in seed markets, exemplified by firms like Monsanto, raises concerns over monopolistic practices, pricing, and farmer dependency, undermining long-term sustainability in low-income regions (Chaturvedi et al., 2019). Addressing these challenges requires policies that integrate SECs with biosafety measures to ensure equitable benefits while minimizing environmental and social trade-offs (Chatzopoulou & Chatzopoulos, 2024).

15.7 BIOSAFETY SYSTEMS

Agricultural biotechnology and GMOs offer significant benefits for crop productivity, pest resistance, and climate adaptation, yet their adoption faces challenges due to inconsistent global biosafety regulations and trade barriers (Saini et al., 2020). The integration of SECs into GMO governance is critical to assess impacts on farmer livelihoods, food security, and local economies, though implementation remains fragmented across regions (Watson, 2019). Resistance from indigenous and rural communities often stems from perceived threats to traditional knowledge and biodiversity, exacerbated by corporate dominance in seed markets and unequal access to technology. Public skepticism, fueled by misinformation and lack of transparency, further hinders acceptance, despite existing biosafety frameworks designed to address environmental and health risks. Effective policy must balance innovation with equitable SEC integration, considering context-specific factors like land ownership and market structures to ensure sustainable adoption (Saini et al., 2020; Watson, 2019).

15.8 LANDSCAPE ASSESSING AND MOVING FORWARD

Evaluating agricultural biotechnology and GMO regulations requires comprehensive landscape assessments that integrate environmental, economic, and social factors, though conflicting stakeholder interests often hinder consensus (Watson, 2019). While biotech innovations can enhance food security through improved yields and reduced chemical use, public skepticism and unequal access driven by corporate patent control and affordability gaps exacerbate inequalities, particularly in developing regions (Leach et al., 2018). Divergent global regulatory frameworks further complicate international trade and adoption, ranging from outright bans to permissive policies (Ahmad et al., 2022; Watson, 2019).

15.9 CASE STUDY: BIOTECH INNOVATIONS IN AGRICULTURE

Biotechnology has significantly transformed agriculture through GM crops, enabling increased yields, pest resistance, and reduced chemical use, with widespread adoption in countries like the U.S., Brazil, and India (Leach et al., 2018). Studies show GM crops like Bt cotton have reduced pesticide use by 37% and increased yields by 22%, providing substantial economic benefits, particularly for farmers in developing nations (Qaim, 2010). However, GM crops remain controversial due to high seed costs that burden smallholder farmers and concerns about long-term environmental and health impacts (Ahmad et al., 2022). The dominance of multinational corporations in GM seed markets raises additional concerns about food sovereignty and corporate control over agricultural systems (Ahmad et al., 2022). While biotechnology offers clear productivity advantages, these socio-economic and ethical challenges must be addressed to ensure equitable and sustainable agricultural development (Leach et al., 2018).

15.10 CONCLUSION

Biotechnology, especially GMOs, has improved agriculture, healthcare, and industry, offering benefits like increased crop yields and reduced pesticide use. However, concerns arise regarding inclusivity, sustainability, and equitable access, particularly for marginalized communities. Regulatory frameworks vary globally, and public skepticism persists due to health and environmental concerns. Developing countries face challenges, including limited regulatory capacity and dependence on multinational corporations. A balanced approach integrating SECs, robust regulations, and stakeholder involvement is crucial for sustainable and inclusive biotech development.

REFERENCES

Ahmad, F., Saeed, Q., Shah, S. M. U., Gondal, M. A., & Mumtaz, S. (2022). Environmental sustainability: Challenges and approaches. In *Natural resources conservation and advances for sustainability* (pp. 243–270). Elsevier.

Ashraf, S. A., Siddiqui, A. J., Abd Elmoneim, O. E., Khan, M. I., Patel, M., Alreshidi, M., & Adnan, M. (2021). Innovations in nanoscience for the sustainable development of food and agriculture with implications on health and environment. *Science of the Total Environment*, *768*, 144990. https://doi.org/10.1016/j.scitotenv.2021.144990

Bacelar, E., Pinto, T., Anjos, R., Morais, M. C., Oliveira, I., Vilela, A., & Cosme, F. (2024). Impacts of climate change and mitigation strategies for some abiotic and biotic constraints influencing fruit growth and quality. *Plants*, *13*(14), 1942. https://doi.org/10.3390/plants13141942

Bakhsh, A., Sırel, I. A., Kaya, R. B., Ataman, I. H., Tillaboeva, S., Dönmez, B. A., & Duru, E. (2021). Contribution of genetically modified crops in agricultural production: Success stories. In *Policy issues in genetically modified crops* (pp. 111–142). Academic Press.

Bernier, L. G. (2019). Protection and commercialization of biotechnology inventions in Canada and Québec. *Industrial Biotechnology*, *15*(3), 162–168. https://doi.org/10.1089/ind.2019.29170.lgb

Brighouse, H., Ladd, H. F., Loeb, S., & Swift, A. (2018). *Educational goods: Values, evidence, and decision-making*. University of Chicago Press.

Canton, H. (2021). Food and agriculture organization of the United Nations—FAO. In *The Europa directory of international organizations 2021* (pp. 297–305). Routledge.

Casadella, V., & Uzunidis, D. (Eds.). (2023). *Agri-innovations and development challenges: Engineering, value chains and socio-economic models*. Wiley.

Catacora-Vargas, G., Binimelis, R., Myhr, A. I., & Wynne, B. (2018). Socio-economic research on genetically modified crops: A study of the literature. *Agriculture and Human Values*, *35*, 489–513. https://doi.org/10.1007/s10460-017-9842-4

Chaturvedi, S., Srinivas, K. R., & Kumar, A. (2019). Cartagena Protocol, socio-economic assessment, and literature review of socio-economic assessment (SEA) studies in India. In *Socio-economic impact assessment of genetically modified crops* (pp. 3–35). Springer.

Chatzopoulou, S., & Chatzopoulos, S. (2024). Livestock farmers' perceptions towards genetically engineered methods and genetically modified organisms in Denmark and Sweden. *Journal of Rural Studies*, *111*, 103414. https://doi.org/10.1016/j.jrurstud.2024.103414

De Francesco, F. (2021). The diffusion of regulatory governance innovations: A research synthesis. In *Handbook of policy transfer, diffusion and circulation* (pp. 443–462). Edward Elgar.

Demont, M., Cerovska, M., Daems, W., Dillen, K., Fogarasi, J., Mathijs, E., & Tollens, E. (2008). Ex ante impact assessment under imperfect information: Biotechnology in new member states of the EU. *Journal of Agricultural Economics*, *59*(3), 463–486. https://doi.org/10.1111/j.1477-9552.2008.00161.x

EFSA. (2020). Outcome of a public consultation on the draft adequacy and sufficiency evaluation of existing EFSA guidelines for the molecular characterisation, environmental risk assessment and post-market environmental monitoring of genetically modified insects containing engineered gene drives. *EFSA Journal*, *17*(11), 1939E. https://doi.org/10.2903/sp.efsa.2020.EN-1939

Evens, R. P. (2022). Biotechnology—A continual revolution in product development and healthcare—1st 20 years versus 2nd 20 years and beyond. *Medical Research Archives*, *10*(4). https://doi.org/10.18103/mra.v10i4.2719

Falck-Zepeda, J. B. (2009). Socio-economic considerations, Article 26.1 of the Cartagena Protocol on Biosafety: What are the issues and what is at stake? *AgBioForum*, *12*(1), 1–13.

Food and Agriculture Organization (FAO). (2021). *The state of food security and nutrition in the world 2021*. FAO.

Gebreyes, A. (2023). *School of law* [Doctoral dissertation, Wachemo University].

Inglehart, R. (2018). *Culture shift in advanced industrial society*. Princeton University Press.

International Service for the Acquisition of Agri-biotech Applications (ISAAA). (2017). *Global status of commercialized biotech/GM crops in 2017* (ISAAA Brief No. 53).

James, C. (2011). *Global status of commercialized biotech/GM crops: 2011* (ISAAA Brief No. 44). ISAAA.

John, G., & Panda, A. K. (Eds.). (2024). *Biotechnology in India: Reworking a strategy*. Springer.

Kircher, M. (2019). Bioeconomy: Markets, implications, and investment opportunities. *Economies*, *7*(3), 73. https://doi.org/10.3390/economies7030073

Kuhlmann, K., & Dey, B. (2021). Using regulatory flexibility to address market informality in seed systems: A global study. *Agronomy*, *11*(2), 377. https://doi.org/10.3390/agronomy11020377

Leach, M., Reyers, B., Bai, X., Brondizio, E. S., Cook, C., Díaz, S., & Subramanian, S. M. (2018). Equity and sustainability in the Anthropocene: A social–ecological systems perspective on their intertwined futures. *Global Sustainability*, *1*, e13. https://doi.org/10.1017/sus.2018.12

Lubogo, I. C. (2024). *Exploring the frontiers of GMOs, bios, and biotechnology: Advancements, ethical considerations, and global implications*.

Ludlow, K., Smyth, S. J., & Falck-Zepeda, J. (Eds.). (2013). *Socio-economic considerations in biotechnology regulation* (Vol. 37). Springer.

Mol, A. P., Sonnenfeld, D. A., & Spaargaren, G. (Eds.). (2020). *The ecological modernisation reader: Environmental reform in theory and practice*. Routledge.

Munhoz, L. (2023). Socio-economic considerations of living modified organisms and impacts on Trade: Evolution of environmental disputes at the World Trade Organization. *Journal of World Trade*, *57*(2), 301–324.

Ou, Y., & Guo, S. (2023). Safety risks and ethical governance of biomedical applications of synthetic biology. *Frontiers in Bioengineering and Biotechnology*, *11*, 1292029. https://doi.org/10.3389/fbioe.2023.1292029

Perotti, V. E., Larran, A. S., Palmieri, V. E., Martinatto, A. K., & Permingeat, H. R. (2020). Herbicide resistant weeds: A call to integrate conventional agricultural practices, molecular biology knowledge and new technologies. *Plant Science*, *290*, 110255. https://doi.org/10.1016/j.plantsci.2019.110255

Priya, T. J., & Kappalli, S. (2022). Modern biotechnological strategies for vaccine development in aquaculture—Prospects and challenges. *Vaccine*, *40*(41), 5873–5881. https://doi.org/10.1016/j.vaccine.2022.08.042

Ray, P. K., Singh, K. M., Kumar, A., Singh, R. N., & Singh, R. R. (2020). Horticulture for food security and livelihood improvement. *Advances in Horticulture*, *1*, 1–20.

Saini, P., Kaur, J. J., Francies, R. M., Gani, M., Rajendra, A. A., & Chauhan, S. S. (2020). Molecular approaches for harvesting natural diversity for crop improvement. In *Rediscovery of genetic and genomic resources for future food security* (pp. 67–169). Springer.

Spök, A. (2010). *Assessing socio-economic impacts of GMOs: Issues to consider for policy development*. Bundesministerium für Gesundheit.

Stabinsky, D. (2000). Bringing social analysis into a multilateral environmental agreement: Social impact assessment and the Biosafety Protocol. *Journal of Environment and Development*, *9*(3), 260–283. https://doi.org/10.1177/107049650000900303

United Nations Educational, Scientific and Cultural Organization (UNESCO). (2022). *Science, technology, and innovation policy review: India*. UNESCO.

Watson, D. (2019). *Genetically modified crops in agriculture*. Scientific e-Resources.

Wise, T. A. (2020). *Failing Africa's farmers: An impact assessment of the Alliance for a Green Revolution in Africa*. Tufts University.

16 Opposition to Global Acceptance of Biotech Crops

Dhaarani Vijayakumar, Kousalya Loganathan, Manikandan Selvarasuvasuki, and Kanivalan Iwar

16.1 INTRODUCTION

The adoption of genetically modified (GM) crops represents a major advancement in modern agriculture, offering solutions to food security challenges through improved yields, pest resistance, and climate adaptability (Singh et al., 2022). However, global acceptance faces resistance due to public concerns about genetic modification, perceived health risks, and biodiversity loss, particularly in regions with limited access to transparent scientific information. Critics argue that GM crops may disrupt ecosystems by fostering superweeds, contaminating non-GM varieties via cross-pollination, and harming beneficial organisms like pollinators and soil microbiota. Regulatory disparities further complicate adoption, with stringent policies in the EU contrasting with permissive frameworks in the USA and Brazil, creating trade barriers and limiting market potential (Belagalla et al., 2024). Small-scale farmers often struggle with access due to high costs, exacerbating equity gaps in global agriculture (Ullah et al., 2024).

Non-governmental organizations (NGOs), Government of India, 2010 and advocacy groups amplify these debates, with some promoting GM benefits while others champion organic alternatives, shaping polarized public opinion and policy (Raza et al., 2024). Addressing opposition to GM crops requires a multifaceted strategy, including science-based public education to counter misinformation and build trust (Oliver, 2014). Harmonizing global regulatory standards could reduce trade barriers and foster wider acceptance, while involving local communities in decision-making ensures alignment with socio-economic contexts. Despite environmental and societal challenges, GM crops hold transformative potential for agriculture, contingent on resolving conflicts over corporate control, ecological impacts, and equitable access (Lombardo & Grando, 2019). Continuous research and inclusive stakeholder dialogue are critical to bridging divides and unlocking biotechnology's full benefits. By balancing innovation with transparency and equity, GM crops could sustainably meet future food demands. Ultimately, their success hinges on collaborative efforts to address ethical, environmental, and regulatory concerns (Lombardo & Grando, 2019; Oliver, 2014). This chapter examines the roots of opposition to biotech crops,

DOI: 10.1201/9781003597797-16

analyzes key stakeholders driving the debate, and explores pathways to reconcile innovation with sustainability and equity.

16.2 HEALTH IMPLICATIONS

Biotech crops have revolutionized agriculture by enhancing nutrition (e.g., vitamin A-enriched Golden Rice) and reducing pesticide exposure, addressing global malnutrition and food security challenges (Mockshell et al., 2024). Rigorous safety assessments, including allergenicity testing, confirm that GM crops pose no greater health risks than conventional crops (Vishnu, 2024), with reduced pesticide use offering additional public health benefits (Singh & Singh, 2025). However, experts advocate for ongoing post-market surveillance to monitor long-term health impacts, given the relatively recent introduction of these crops into diets (Mockshell et al., 2024). Public skepticism persists due to misinformation and ethical concerns, despite robust scientific evidence supporting their safety (Dufossé & Tiwari, 2024). Bridging this gap demands transparent communication and public engagement to translate research into trust. While biotech crops represent a transformative tool for health and nutrition, sustained research, regulatory vigilance, and inclusive discourse remain essential to maximize their potential and address legitimate concerns (Hoban, 1997).

16.3 ECOLOGICAL RISKS AND SUSTAINABILITY CHALLENGES

Biotech crops raise significant ecological concerns despite their agricultural benefits, including risks of biodiversity loss, genetic contamination through cross-pollination, and the development of herbicide-resistant weeds that reinforce chemical-dependent farming. Early studies established critical frameworks for ecological risk assessment, demonstrating impacts on non-target species, soil microbiota, and ecosystem balance that remain relevant today (Andow & Zwahlen, 2006; Wolfenbarger & Phifer, 2000). Contemporary research highlights additional challenges, such as soil nutrient depletion from GM-linked monocultures and the rise of glyphosate-resistant weeds, which exacerbate ecological vulnerabilities (Choriyeva, 2024; Sarsaiya et al., 2024). Regulatory systems often lag behind biotech advancements, failing to implement robust post-release monitoring as recommended decades ago (Snow et al., 2005).

16.4 ECONOMIC IMPLICATIONS

Resistance to biotech crops often stems from cultural values prioritizing naturalness and ecological wholesomeness, exacerbated by activist misinformation and distrust of multinational corporations dominating seed markets, particularly in Africa and South Asia (Scoones, 2008). In regions like India, opposition is further fueled by religious and traditional farming practices, with critics like Shiva (2001) framing GM adoption as a threat to cultural identity. Developed nations, such as the USA and Japan, face consumer-led demands for transparency and labeling, reflecting societal desires for dietary autonomy (Fernandez-Cornejo et al., 2014). This tension pits scientific progress against cultural preservation, while global inequalities persist, as wealthier nations implement rigorous GM regulations while poorer regions rely on external narratives, leaving them vulnerable to anti-GM campaigns (Qaim, 2020).

16.5 ETHICAL AND PHILOSOPHICAL CONSIDERATIONS

The ethical and philosophical debates surrounding GM crops encompass diverse perspectives on technology, nature, and societal values, raising critical questions about health risks, allergenicity, and ecological impacts (Domingo & Bordonaba, 2011). While proponents highlight GM crops' potential to address hunger, exemplified by Golden Rice concerns persist about equitable access in developing nations and the dominance of patented technologies by corporations like Monsanto, which marginalize small farmers (Akhter, 2020; Qaim & Kouser, 2013). Studies on GM cotton in India demonstrate improved food security at the micro level, yet critics warn of ecological risks like gene flow to wild species and biodiversity loss (Bordes & Barfoot, 2014; Sarsaiya et al., 2024). Health safety assessments, including meta-analyses by Nicolia et al. (2014), generally affirm GM crops' safety, though controversies like the Séralini study (2012) underscore the need for rigorous, transparent research.

Ethical objections often stem from deontological arguments against "playing God" and altering natural ecosystems, countered by consequentialist views emphasizing GM crops' benefits for food security and sustainability (James, 2011; Sandler, 2012). Philosophical tensions also arise over monoculture threats to biodiversity and ecological integrity, alongside justice issues tied to seed patents and corporate monopolies (Clancy, 1999; Fatoretto et al., 2017). Public distrust, fueled by cultural narratives and misinformation, highlights epistemological gaps between scientific evidence and societal perceptions (Fernandes et al., 2022). Regulatory disparities further complicate adoption, with wealthier nations implementing robust safety protocols while poorer regions rely on external narratives (Qaim, 2020). Addressing these challenges requires equitable frameworks that balance innovation with stewardship, justice, and respect for nature (Garlet et al., 2022). Transparent communication and inclusive dialogue are vital to bridge divides and guide responsible biotech development (Bauman & Cauchy, 2006). Ultimately, the GM crop debate demands a multidisciplinary approach that integrates ethical, ecological, and social dimensions to reconcile technological progress with moral obligations (Gongo, 2017).

16.6 OPPOSITION TO GENETICALLY MODIFIED (GM) CROPS

Table 16.1 documents global opposition to GM crops from 1994 to 2023, revealing recurring concerns over food safety (e.g., FlavrSavr Tomato in the USA), environmental risks (e.g., France's MON810 corn ban), and corporate dominance (e.g., India's Bt cotton protests) (Glover et al., 2020). Table 16.2 proposes mitigation strategies, including science-based public outreach to counter misinformation and harmonized regulations to ease trade barriers (Glover et al., 2020). Culturally sensitive

TABLE 16.1
Case Studies on Opposition to Genetically Modified (GM) Crops

Year	Country/ Region	Case/Event	Description	Impact
1994	USA	FlavrSavr Tomato controversy	First GM food approved by FDA faced public skepticism about food safety	Led to increased scrutiny of GM food approval process.
1996	Europe	Mad Cow Disease Crisis	Though unrelated to GM, heightened European suspicion of food safety and biotechnology	Contributed to strict EU regulations on GM crops (EFSA, 2012).
1998	UK	Dr. Pusztai affair	Controversial study claiming GM potatoes harmed rats' immune systems	Sparked major public debate and media coverage against GM foods (Pusztai, 1998).
1999	USA	Monarch butterfly study	Research suggested Bt corn pollen could harm monarch butterflies	Triggered environmental safety debates and additional studies (Losey et al., 1999).
2000	USA	StarLink corn incident	GM corn approved for animal feed found in human food products	Led to massive recalls and strengthened segregation requirements (Taylor & Tick, 2001).
2001	Mexico	Maize contamination case	GM corn detected in native Mexican maize varieties	Raised concerns about genetic contamination of crop centers of origin (Quist & Chapela, 2001).
2003	India	Bt cotton protests	Farmers protested against Monsanto's GM cotton	Highlighted issues of corporate control and farmer autonomy (Scoones, 2003).
2004	EU	GM moratorium	EU implemented strict regulations on GM crop approval	Created trade disputes with the USA and affected global GM crop adoption (James, 2004).
2008	France	Ban on MON810 corn	France banned Monsanto's GM corn citing environmental concerns	Influenced other EU countries to adopt similar bans (European Court of Justice, 2018).
2010	India	Bt brinjal moratorium	Public opposition led to an indefinite moratorium on GM eggplant	First instance of a ready-for-commercialization GM food crop being rejected (Scoones, 2003).
2013	Philippines	Golden Rice trial destruction	Protesters destroyed experimental GM rice fields	Highlighted tensions between humanitarian biotech and opposition (James, 2004).
2015	Russia	GM crop ban	Russia banned cultivation of GM crops	Demonstrated growing political aspects of GM opposition (Reuters, 2024).
2016	USA	Vermont GMO labeling law	First US state to require mandatory GM food labeling	Led to federal GM labeling legislation (James, 2004).

TABLE 16.1 (*Continued*)
Case Studies on Opposition to Genetically Modified (GM) Crops

Year	Country/ Region	Case/Event	Description	Impact
2018	EU	CRISPR regulation	EU court ruled CRISPR-edited crops must follow GM regulations	Impacted the development of new breeding technologies (European Court of Justice, 2018).
2019	Mexico	GM corn Import ban	Mexico announced plans to phase out GM corn imports	Created trade tensions with US corn exporters (Reuters, 2024).
2020	Peru	GM Crop moratorium Extension	Extended ban on GM crops for environmental concerns	Reinforced regional resistance to GM technology (James, 2004).
2021	Mexico	Glyphosate ban	Banned glyphosate and GM corn for human consumption	Significant impact on USA-Mexico agricultural trade (Reuters, 2024).
2022	India	GM mustard protests	Opposition to approval of GM mustard for commercial cultivation	Highlighted continued resistance to GM food crops (Scoones, 2003).
2023	EU	New genomic techniques	Debates over regulation of new breeding technologies	Ongoing discussion about the future of crop biotechnology (European Court of Justice, 2018).

TABLE 16.2
Alternatives and Solutions for Opposition to Genetically Modified (GM) Crops

Category	Strategies/ Alternatives	Rationale	Implementation Examples	References
Public awareness	Science-based communication campaigns; use of social media to counter misinformation	Educate the public about the safety and benefits of biotech crops while addressing widespread misinformation.	Initiatives like “GMO Answers” in the USA; leveraging influencers and social media platforms to disseminate factual information about GMOs in Africa and Asia.	Qaim (2020); Fernandez-Cornejo et al. (2014); Hoban (1997)
Regulatory frameworks	Harmonized biotech regulations; regional biotech approval hubs	Streamline approval processes and reduce compliance costs across nations.	Establishing approval hubs in Africa (e.g., African Biosafety Network); harmonization between the USA and EU for GMO standards to reduce trade disruptions.	Carter, Moschini, and Sheldon (2011);

(*Continued*)

TABLE 16.2 (*Continued*)
Alternatives and Solutions for Opposition to Genetically Modified (GM) Crops

Category	Strategies/ Alternatives	Rationale	Implementation Examples	References
Cultural integration	Incorporate local traditions and values into biotech programs to ensure alignment with community needs	Align biotech adoption with local cultural values and include traditional methods for pest and crop management.	Introducing biotech crops as complementary to indigenous farming in India	Scoones (2008); Shiva (2001);
Consumer transparency	Mandatory GMO labeling; voluntary organic-biotech coexistence labels	Ensure consumer confidence by providing clear choices between GMO, non-GMO, and organic products.	EU's stringent GMO labeling laws; voluntary "GMO-Free" certification programs in the USA; co-labeling schemes where GMO and organic products coexist peacefully.	Hoban (1997); Fernandez-Cornejo et al. (2014)
Research and development	Locally driven biotech innovations; focus on non-GMO gene-editing techniques	Develop crops tailored for specific regional issues and explore non-GMO biotechnologies to address public resistance.	Golden Rice for combating vitamin A deficiency; CRISPR-edited crops in Japan and Europe that bypass GMO regulatory frameworks.	James (2001); Brookes and Barfoot (2020); ISAAA (2021)
Economic incentives	Subsidies and support for farmers; community-owned biotech seed banks	Offset initial adoption costs and promote locally managed biotech seed systems to reduce dependency on multinational companies.	Subsidies for biotech seeds in China; community seed banks in Malawi offering drought-resistant biotech seeds.	Brookes and Barfoot (2020)
Multi-stakeholder engagement	Building collaborative platforms; inclusion of local NGOs and farmer groups	Foster trust and inclusivity by involving all stakeholders in the decision-making process.	Public-private collaborations for GM maize in Africa; partnerships with NGOs to educate farmers in Brazil about biotech safety and efficacy.	Paarlberg (2008); ISAAA (2021)

TABLE 16.2 (*Continued*)
Alternatives and Solutions for Opposition to Genetically Modified (GM) Crops

Category	Strategies/ Alternatives	Rationale	Implementation Examples	References
Environmental alternatives	Biotech crops for climate resilience; organic-biotech hybrid systems	Promote crops with proven environmental benefits and explore hybrid farming systems combining biotech and organic practices.	Drought-resistant maize in sub-Saharan Africa; integrated pest management (IPM) systems combining Bt crops with biological pest control agents.	Brookes and Barfoot (2020);
Education programs	Farmer and consumer training; use of interactive online platforms	Enhance understanding of biotech crop benefits and management, addressing fear and misconceptions.	Extension programs in India for Bt cotton; online platforms offering interactive modules for farmers and consumers about GM crop safety and applications.	Qaim (2020); ISAAA (2021)
Public-private partnerships	Shared development initiatives; collaboration with independent research organizations	Promote affordability, accessibility, and transparency by involving both public institutions and independent entities.	Partnerships like Monsanto's WEMA project in Africa; collaborations with CGIAR centers to develop region-specific biotech solutions.	Scoones (2008); Paarlberg (2008); ISAAA (2021)
Alternatives to biotech crops	Marker-assisted selection (MAS); agroecological farming practices	Provide non-biotech genetic improvement techniques and integrate sustainable farming practices as alternatives.	MAS to develop disease-resistant wheat varieties in Asia; agroecological practices combining crop rotation with natural fertilizers in Latin America.	Qaim (2020);
Trade facilitation	Low-level presence (LLP) policies; blockchain for biotech traceability	Address trade barriers by allowing minimal GMO presence and enhancing transparency in biotech crop supply chains.	Canada's LLP policies for biotech traits; blockchain-based traceability systems in global grain markets.	Carter et al. (2011); Brookes and Barfoot (2020)

approaches, such as aligning biotech adoption with local practices and mandatory GMO labeling, aim to build consumer trust, while economic incentives like subsidies reduce farmer reliance on multinational seed companies (FAO, 2023). Alternatives like CRISPR-based editing, agroecology, and public-private partnerships offer non-GMO solutions, supported by tools like blockchain for supply-chain transparency (Glover et al., 2020).

16.7 GLOBAL PERSPECTIVES

GM crops can be engineered to achieve higher yields, enhance resistance and tolerance to drought and pests, and potentially improve food security while reducing global hunger (Figure 16.1) (Krishna et al., 2014; Snell et al., 2012). Genetic modification can also enhance the nutritional content of crops by increasing essential vitamins and minerals, helping to address malnutrition. Additionally, GM crops can reduce environmental impact by boosting yields and lowering the use of pesticides and herbicides, promoting sustainable agricultural practices (Qaim, 2020; Scoones, 2008). Environmental concerns include the risk of gene flow, the emergence of herbicide-resistant weeds, and potential harm to beneficial insects and biodiversity. Ethical debates also arise over the manipulation of living organisms, and the concentration of GM crop development among a few large corporations raises issues of access and equity for small farmers (Paarlberg, 2008).

FIGURE 16.1 Global perspective on the drivers of GM crop adoption.

16.8 CONCLUSION

Opposition to the global acceptance of biotech crops stems from a complex interplay of environmental, ethical, economic, and social concerns. While proponents highlight the potential benefits of biotech crops, such as increased food security, reduced pesticide use, and enhanced nutritional content, critics raise valid concerns regarding biodiversity, the concentration of power in agribusiness, and the long-term health and ecological impacts. Public skepticism is often fueled by a lack of transparent regulatory frameworks and insufficient understanding of the science behind genetic modification. As a result, achieving global consensus on the adoption of biotech crops requires not only rigorous scientific research and clear communication but also addressing the broader socio-economic and ethical implications, ensuring that all stakeholders have a voice in shaping the future of agriculture.

REFERENCES

Akhter, T. (2020). Problems and challenges faced by EFL students of Saudi Arabia during COVID-19 pandemic. *Rupkatha Journal on Interdisciplinary Studies in Humanities*, *12*(5), 1–7.

Andow, D. A., & Zwahlen, C. (2006). Assessing environmental risks of transgenic plants. *Ecology Letters*, *9*(2), 196–214. https://doi.org/10.1111/j.1461-0248.2005.00846.x

Bauman, C., & Cauchy, A. (2006). The bioethics of genetically modified crops: Theoretical perspectives. *Journal of Agricultural and Environmental Ethics*, *19*(4), 319–334.

Belagalla, N., Kaur, R., & Abhishek, G. J. (2024). Eco-friendly and targeted through next-generation approaches to insect pest management. *Uttar Pradesh Journal of Zoology*, *45*(13), 73–99.

Brookes, G., & Barfoot, P. (2014). *GM crops: global socio-economic and environmental impacts 1996–2012*. PG Economics Ltd, UK, 1–189.

Brookes, G., & Barfoot, P. (2020). *GM crops: Global socio-economic and environmental impacts 1996–2018*. PG Economics Ltd. https://pgeconomics.co.uk/pdf/Globalimpactbiotechcropsfinalreportoctober2022.pdf

Carter, C. A., Moschini, G., & Sheldon, I. (Eds.). (2011). *Genetically modified food and global welfare*. Emerald Group Publishing. https://doi.org/10.1108/S1574-8715(2011)0000010002

Choriyeva, Z. (2024). Genetically modified ecosystems: Innovative approaches in agriculture and their environmental impact. In *BIO Web of Conferences* (Vol. 149, p. 01009). EDP Sciences. https://doi.org/10.1051/bioconf/202414901009

Clancy, J. J. (1999). *The invisible powers: The language of business*. Lexington Books.

Domingo, J. L., & Bordonaba, J. G. (2011). A literature review on the safety assessment of genetically modified plants. *Environment International*, *37*(4), 734–742. https://doi.org/10.1016/j.envint.2011.01.003

Dufossé, L., & Tiwari, P. (2024). Emerging trends and advances in the socioeconomic applications of beneficial microbes. *Frontiers in Microbiology*, *15*, 1523569. https://doi.org/10.3389/fmicb.2024.1523569

EFSA. (2012). Scientific opinion updating the risk assessment conclusions and risk management recommendations on the genetically modified insect resistant maize MON 810. *EFSA Journal*, *10*(12), 3017.

European Court of Justice. (2018). *Judgment in case C-528/16: Confédération paysanne and others v. Premier ministre and Ministre de l'agriculture, de l'agroalimentaire et de la forêt.*

https://www.europeansources.info/record/cjeu-case-c-528-16-confederation-paysanne-and-others-v-premier-ministre-and-ministre-de-lagriculture-de-lagroalimentaire-et-de-la-foret/

Fatoretto, J. C., Michel, A. P., Silva Filho, M. C., & Silva, N. (2017). Adaptive potential of fall armyworm (*Lepidoptera: Noctuidae*) limits Bt trait durability in Brazil. *Journal of Integrated Pest Management*, *8*(1). https://doi.org/10.1093/jipm/pmx011

Fernandes, G. B., Silva, A. C. de L., Maronhas, M. E. S., Santos, A. da S. dos, & Lima, P. H. C. (2022). Transgene flow: Challenges to the on-farm conservation of maize landraces in the Brazilian semi-arid region. *Plants*, *11*(5), 603. https://doi.org/10.3390/plants11050603

Fernandez-Cornejo, J., Wechsler, S., Livingston, M., & Mitchell, L. (2014). *Genetically engineered crops in the United States* (USDA-ERS Economic Research Report No. 162). https://www.ers.usda.gov/publications/pub-details?pubid=45182

Food and Agriculture Organization (FAO). (2023). *Alternative approaches to crop improvement* (Technical Paper No. 284). https://openknowledge.fao.org/server/api/core/bitstreams/5aac5078-625d-4b94-b964-bea40493016c/content

Garlet, C. G., Muraro, D. S., Godoy, D. N., Cossa, G. E., Hanich, M. R., Stacke, R. F., & Bernardi, O. (2022). Assessing fitness costs of the resistance of *Spodoptera frugiperda* (*Lepidoptera: Noctuidae*) to pyramided Cry1 and Cry2 insecticidal proteins on different host plants. *Bulletin of Entomological Research*, *112*(3), 1–9. https://doi.org/10.1017/S0007485321001152

Glover, D., Kim, S. K., & Stone, G. D. (2020). Golden Rice and technology adoption theory: A study of seed choice dynamics among Rice growers in the Philippines. *Technology in Society*, *60*, 101227. https://doi.org/10.1016/j.techsoc.2019.101227

Gongo, S. (2017, April 22). Burkina Faso sees cotton output rising 20% in 2017–18 season. *Naija247news*. https://naija247news.com/2017/04/22/burkina-faso-sees-cotton-output-rising-20-in-2017-18-season/

Government of India. (2010). *Decision on commercialization of Bt brinjal*. http://indiagminfo.org/wp-content/uploads/2019/12/Bt-brinjal-minister_moratorium-decision-note-feb9-2010.pdf

Hoban, T. J. (1997). Consumer acceptance of biotechnology: An international perspective. *Nature Biotechnology*, *15*(3), 232–234.

ISAAA. (2021). *Global status of commercialized biotech/GM crops* (ISAAA Brief No. 55). https://www.isaaa.org/resources/publications/briefs/55/

James, C. (2001). *Global review of commercialized transgenic crops: 2001* (ISAAA Brief No. 24). https://www.isaaa.org/resources/publications/briefs/26/download/isaaa-brief-26-2002.pdf

James, C. (2004). *Global status of commercialized biotech/GM crops: 2004* (ISAAA Brief No. 32). https://www.isaaa.org/resources/publications/briefs/32/download/isaaa-brief-32-2004.pdf

James, J. F. (2011). *A student's guide to Fourier transforms: with applications in physics and engineering*. Cambridge University Press.

Krishna, V. V., Qaim, M., & Zilberman, D. (2014). Transgenic crops, production risk, and agrobiodiversity. ZEF- Discussion Papers on Development Policy No. 186. https://dx.doi.org/10.2139/ssrn.2405466

Lombardo, L., and Grando, M. S. (2019). Genetically modified plants for nutritionally improved food: A promise kept? *Food Reviews International*, *36*(1), 58–76. https://doi.org/10.1080/87559129.2019.1613664

Losey, J. E., Rayor, L. S., & Carter, M. E. (1999). Transgenic pollen harms monarch larvae. *Nature*, *399*(6733), 214. https://doi.org/10.1038/20338

Mockshell, J., Asante-Addo, C., Nwagboso, C., Ritter, T., Amare, M., & Andam, K. S. (2024). *Impact evaluation of the use of PBR cowpea in Nigeria: Baseline process evaluation report* (NSSP Project Report). International Food Policy Research Institute. https://hdl.handle.net/10568/145073

Nicolia, A., Manzo, A., Veronesi, F., & Rosellini, D. (2014). An overview of the impact of GM crops on human health. *Nature Biotechnology*, *32*(5), 477–484. https://doi.org/10.1038/nbt.2879

Oliver, M. J. (2014). Why we need GMO crops in agriculture. *Missouri Medicine*, *111*(6), 492–507. https://www.ncbi.nlm.nih.gov/pmc/articles/PMC6173531/

Paarlberg, R. (2008). *Starved for science: How biotechnology is being kept out of Africa*. Harvard University Press.

Pusztai, A. (1998). Report on the effects of genetically modified potatoes on rats' immune systems. *The Lancet*, *354*(9187), 1353–1354.

Qaim, M. (2020). Role of new plant breeding technologies for food security and sustainable agricultural development. *Applied Economic Perspectives and Policy*, *42*(2), 129–150. https://doi.org/10.1002/aepp.13044

Qaim, M., & Kouser, S. (2013). Genetically modified crops and food security. *PLoS ONE*, *8*(6), e64879. https://doi.org/10.1371/journal.pone.0064879

Quist, D., & Chapela, I. H. (2001). Transgenic DNA introgressed into traditional maize landraces in Oaxaca, Mexico. *Nature*, *414*(6863), 541–543. https://doi.org/10.1038/35107068

Raza, M., Fatima, R., Touqeer, M., Ain, N., Mushtaq, M., Bint-E-Zafar, A., & Khaliq, A. (2024). Role of plant breeding to maintaining food security in the face of global climate change. *Journal of Life and Social Sciences*, *1*, 26. https://bbasrjlifess.com/index.php/home/article/view/26

Reuters (2024). *US wins Mexico GM corn dispute case as panel finds curbs not science-based*. https://www.reuters.com/markets/commodities/trade-panel-rules-us-favor-mexico-gmo-corn-dispute-case-2024-12-20/

Sandler, R. (2012). The ethics of genetically modified organisms. In *The Oxford handbook of food ethics* (pp. 345–368). Oxford University Press.

Sarsaiya, S., Yadav, A. N., Tiwari, P., & Singh, R. (2024). Futuristic plant microbes biotechnology and bioengineering. *Frontiers in Microbiology*, *15*, 1514583. https://doi.org/10.3389/fmicb.2024.1514583

Scoones, I. (2003). *Biotech battlegrounds: The controversy over Bt cotton in India*. Institute of Development Studies.

Scoones, I. (2008). Mobilizing against GM crops in India, South Africa, and Brazil. *Journal of Agrarian Change*, *8*(2–3), 315–344.

Shiva, V. (2001). *Stolen harvest: The hijacking of the global food supply*. Zed Books.

Singh, A. A., & Singh, A. K. (2025). Role of bacterial quorum sensing in plant growth promotion. *World Journal of Microbiology and Biotechnology*, *41*(1), 1–17.

Snell, C., Parry, M., & Godfray, H. C. J. (2012). The future of crop yields in Africa. *Science*, *336*(6085), 1144–1145.

Snow, A. A., Andow, D. A., Gepts, P., Hallerman, E. M., Power, A., Tiedje, J. M., & Wolfenbarger, L. L. (2005). Genetically engineered organisms and the environment: Current status and recommendations. *Ecological Applications*, *15*(2), 377–404.

Taylor, M. R., & Tick, J. S. (2001). The StarLink corn incident: A case study in biotech regulation. *Environmental Health Perspectives*, *109*(6), A290–A297.

Ullah, M., Ahmed, M., Javed, I., Shafiq, M., Sherazi, S., & Waleed, R. (2024). Genetically modified crops global regulation: Implications for food security and environmental sustainability. *Journal of Life and Social Sciences*, *2024*(1), 25.

Vishnu, L. (2024). A comprehensive review of genetically modified horticultural crops: Benefits, risks, and public perceptions. *Australian Journal of Global Horticulture and Agronomy Science*, *2*(1), 1–15.

Wolfenbarger, L. L., & Phifer, P. R. (2000). The ecological risks and benefits of genetically engineered plants. *Science*, *290*(5499), 2088–2093.

17 Digital Agriculture and Biotechnology

Synergies for Sustainable Development

Sudhanya Nath and Sachin Tripura

17.1 INTRODUCTION

The concept of sustainability, derived from the Latin *Sustinere*, emphasizes environmental preservation and resource management for future generations, gaining prominence since the 1972 UN Conference on the Human Environment (Fathi, 2022). Despite global agreements post-1992, challenges like biodiversity loss and climate crises persist, intensifying the need for sustainable agriculture that balances food security with ecological and economic viability (Rajesh et al., 2024). Zero Budget Natural Farming (ZBNF) exemplifies this shift, reviving pre-colonial practices that relied on natural inputs before commercial crops dominated (Kumar et al., 2023; Rajesh et al., 2024). Agricultural biotechnology, now a key driver of India's socioeconomic growth, merges traditional breeding with modern techniques to enhance productivity (Business Standard, 2013; Lima, 2022).

However, its association with industrial farming and patented seeds often sparks debate, conflating the method's potential with perceived risks (Estrada et al., 2017). The emerging "gene revolution" aims to decode DNA for agricultural innovation, yet faces criticism for ties to pesticide-dependent monocultures and corporate seed control (Estrada et al., 2017). Sustainable agriculture's core challenge lies in boosting yields amid climate change and shrinking arable land while reducing synthetic inputs. Biotechnology offers tools to address these gaps, but its success hinges on divorcing the *process* from controversial *products* like patented Genetically Modified Organisms (GMOs). Achieving sustainability requires reconciling high-tech solutions with ecological principles, ensuring food security without compromising environmental or farmer autonomy. Ultimately, harmonizing traditional wisdom, like ZBNF, with biotechnological advances could pave the way for resilient, equitable food systems. This chapter explores how the convergence of these two fields can drive sustainable agricultural practices and contribute to long-term food system stability.

DOI: 10.1201/9781003597797-17

17.2 NATURAL FARMING: ADVANCEMENT TOWARD CREATION OF A GREENER FUTURE

Natural farming promotes an ecological, chemical-free approach integrating crops, trees, and livestock to enhance soil fertility, conserve water, and reduce environmental impact while prioritizing biodiversity and sustainability (National Mission on Natural Farming Management and Knowledge (NMNFMK, 2025; Radico Khaitan, 2024). The Indian government supports this transition through initiatives like the National Mission on Natural Farming (NMNF), which provides Rs. 15,000/hectare annually for three years to farmers adopting chemical-free practices, focusing on behavioral shifts toward cow-based inputs (NMNFMK, 2025). The Paramparagat Krishi Vikas Yojana (PKVY), launched in 2015, emphasizes soil health under the National Mission on Sustainable Agriculture, while the Bharatiya Prakritik Krishi Paddhati (BPKP) sub-scheme (2019–2025) allocates ₹4645.69 crore to liberate farmers from imported inputs, offering Rs. 12,200/hectare for cluster development and certification (NMNFMK, 2025; Reddy, 2018). The National Centre for Organic and Natural Farming (NCONF) acts as a knowledge hub, promoting organic and regenerative practices, with the 2024 budget earmarking Rs. 1.52 lakh crore to assist 1 crore farmers in transitioning to natural farming (NMNFMK, 2025). These programs collectively aim to empower farmers economically and socially while ensuring ecological resilience through localized, sustainable practices.

17.3 BIOTECHNOLOGICAL INTERVENTIONS IN NATURAL FARMING

Agricultural biotechnology enhances agroecosystem productivity through three key approaches: plant modification, soil improvement, and alternative fuel inputs (Badiyal et al., 2024). Traditional breeding, accelerated by biotechnological tools like genetic engineering and micro-propagation, reduces development time for stable genotypes from 6–7 generations to 2–4 (Ahmar et al., 2020). High-yielding varieties emerged from marker-assisted selection (MAS) and hybridization, with genes like DREB in rice improving photosynthesis and stress tolerance, while wheat and maize studies identified traits affecting height, spikelets, and kernel weight (Jiang et al., 2023; Kelliher et al., 2019). Photosynthetic efficiency is boosted by modifying enzymes such as PsbS, increasing biomass and nitrogen utilization (Curatti & Rubio, 2014).

Insect-resistant crops like Bt cotton, developed via *Agrobacterium*-mediated transformation, utilize *cry* genes from *Bacillus thuringiensis* (Carrière et al., 2015; Sanahuja et al., 2011), while disease-resistant varieties leverage CRISPR, TALENs, and APR genes in wheat (Ellis et al., 2014). Herbicide-resistant crops, such as glyphosate-tolerant varieties, mitigate weed competition by targeting the EPSPS enzyme (Tan et al., 2006), and abiotic stress resilience is advanced through omics tools like RNA-Seq (Chantre et al., 2016). Biofortification enriches crops like Golden Rice (vitamin A) and iron-rich cassava via transgenic and mixed-cropping systems (Ebbisa, 2022; Paine et al., 2005). Soil modification employs bioremediation to degrade xenobiotics using plant-microbe partnerships (Zhang et al., 2017), while composting converts waste into bioavailable nutrients, though poor management risks pollution (Singh et al., 2021). Microbe-mediated biofortification addresses nutrient gaps (e.g., vitamin B12 via rhizobacteria) (Krishna et al., 2023), and PGPRs enhance iron, zinc, and selenium uptake (Khanna et al., 2023). Biofertilizers fix nitrogen and solubilize phosphorus, improving soil health and stress tolerance (Badiyal et al., 2024), while biopesticides derived from bacteria and plants offer target-specific pest control without harming non-target species (Dar et al., 2021). These innovations collectively reduce reliance on synthetic inputs, aligning productivity with ecological sustainability.

17.4 DIGITALIZATION IN AGRICULTURE

Feeding the world's projected 10 billion population by 2050 while ensuring sustainable development remains a critical global challenge, requiring significant increases in agricultural productivity (Food and Agriculture Organization of the United Nations (FAO), 2018). Growing demands for food security, sustainability, and profitability, compounded by climate change and economic pressures, highlight the need for innovative solutions (Prause, 2021). Digital agriculture encompassing AI, IoT, big data analytics, and ICT transforms traditional farming into data-driven systems that optimize production while reducing environmental impacts (Fielke et al., 2020; Goel et al., 2021). This shift, termed "Agriculture 4.0" (Da Silveira et al., 2021) or "smart farming" (Ingram et al., 2022), leverages digitalization to enhance efficiency and sustainability in food production (Schnebelin et al., 2021).

Agriculture 4.0 integrates advanced technologies like AI, IoT, robotics, and big data to enhance global agricultural competitiveness by optimizing production, distribution, and marketing while minimizing waste through precision farming and real-time monitoring (Bešić et al., 2022; Rai, 2023). Precision agriculture leverages IoT sensors, drones, and satellite data to improve irrigation, fertilization, and pest control, boosting yields and reducing costs (Dhanaraju et al., 2022), while sensor technology enables real-time soil moisture, nutrient, and weather tracking to enhance resource efficiency (Rai, 2023). Variable rate technology (VRT) tailors input application to field-specific needs, reducing environmental impact and costs, though affordability for small-scale farmers remains a challenge (Rai, 2023).

Drones provide high-resolution crop and soil mapping but require regulatory frameworks and farmer training for ethical deployment, and robotics automate labor-intensive tasks like harvesting and weeding, though adoption demands significant investment and training (Rai, 2023). AI further revolutionizes agriculture through predictive analytics for yield forecasting, disease detection, and soil health optimization, enabling data-driven decision-making (Rai, 2023). Together, these innovations underpin a sustainable, efficient, and high-yielding agricultural future, contingent on accessibility, regulation, and education.

17.5 CLIMATE-SMART AGRICULTURE

Climate-smart agriculture (CSA) (Figure 17.1) is just one example of how the agriculture industry is fast changing due to digitalization. Digital tools can improve agricultural decision-making, boost output, and lessen the negative effects of

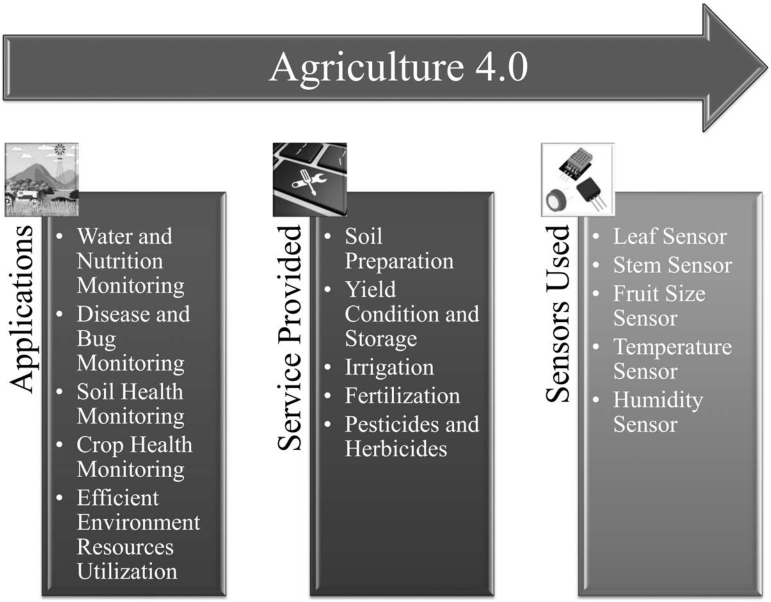


FIGURE 17.1 Applications, services provided, and sensors used in Agriculture 4.0.

agriculture on the environment. Precision farming, crop management, market access, data management, weather monitoring and forecasting, and farm management are some of the key areas of digitalization associated with the aforementioned fields. CSA is still in the early phases of digitalization, and obstacles like the expense of technology and the digital gap in rural areas must be addressed (Figure 17.2). But there are also a lot of potential advantages, and digitalization may be a big part of making agriculture more resilient in the face of climate change (Dhanaraju et al., 2022).

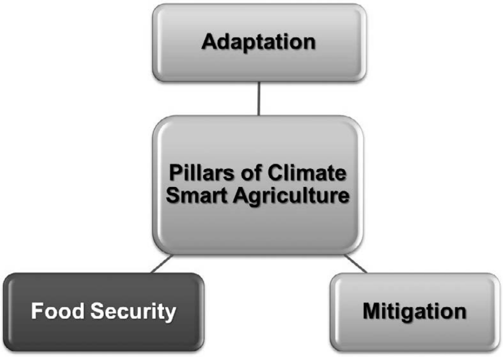


FIGURE 17.2 Pillars of climate smart agriculture.

17.6 DIGITAL APPROACHES IN AGRICULTURE

Data-driven agriculture relies on the collection of various data from environmental elements and agricultural inputs, such as satellite photos, soil health data, land records, and cropping patterns, to drive digitalization in the sector. Satellite imagery aids in crop characterization, land use classification, and gathering weather data, which is analyzed and shared on mobile platforms (Confederation of Indian Industry, 2021; Katekar & Cheruku, 2022). Digital agriculture extension services address the lack of reliable information for farmers, using SMS, Interactive Voice Response (IVR), radio, and low-cost video to provide timely alerts, weather forecasts, and best practices (Cheruku & Katekar, 2023). Digital Financial Services (DFS) became feasible in India with the JAM trinity (Jan Dhan, Aadhar, and mobile phones), enabling Direct Benefit Transfer (DBT) and better access to financial services for farmers (Economic Survey, 2016). DFS also facilitates tracking of produce, such as fruits and vegetables, through digital barcodes and connects farmers to national markets via e-NAM (Cheruku & Katekar, 2023; Ministry of Agriculture and Farmers' Welfare, 2018).

17.7 CONVENTIONAL VS. MODERN NATURAL FARMING

Conventional natural farming focuses on using only natural inputs to maintain agroecosystems, reducing reliance on synthetic fertilizers and pesticides, while biotechnology enhances the effects of natural inputs like microorganisms and agricultural waste (Badiyal et al., 2024). Biotechnology-assisted natural farming, though requiring research funding, becomes self-sustaining once suitable products or varieties are developed and can address the needs of smallholder farmers in developing countries (Food and Agriculture Organization of the United Nations (FAO, 2011). This approach boosts crop resilience and nutritional benefits by incorporating traits from wild crop relatives, improving resistance to pests, diseases, and climate challenges, thus offering potential solutions for sustainable agriculture (Asdal, 2008).

17.8 IMPACT OF BIOTECHNOLOGY ASSISTED NATURAL FARMING

Biotechnology in agriculture has raised concerns regarding its impact on ecosystem biodiversity, but bio-herbicides offer a more sustainable alternative by reducing greenhouse gas emissions and targeting fewer weed species (Chamberlain et al., 2007). Transgenic crops, such as Bt cotton and soybean, have shown little to no effect on genetic diversity, and public sector germplasm collections help preserve genetic variety (Sneller, 2003). Bt crops also help conserve non-target species, promoting greater arthropod diversity compared to traditional insecticides, while reducing the spread of pest resistance (Cattaneo et al., 2006; Naranjo, 2005). Incorporating refuges—non-biotech crops—further delays resistance by providing susceptible insects for mating, ensuring hybrids cannot survive on Bt crops (Environmental Protection Agency (EPA), 2001). Economically, natural farming practices are essential for sustainability, with biotechnology-enhanced precision agriculture reducing the need for costly pesticides, conserving water, and boosting long-term productivity (Badiyal et al., 2024; Das et al., 2023). Additionally, natural farming supports social systems by fostering community collaboration, preserving cultural identities, and aligning with traditional farming practices (Badiyal et al., 2024).

17.9 IMPORTANCE OF SUSTAINABLE DEVELOPMENT IN AGRICULTURE

Sustainable agriculture promotes farming methods that ensure the long-term viability of both farmers and natural resources, preserving soil quality, reducing degradation, conserving water, and enhancing biodiversity (Ansari & Tabassum, 2018). It plays a crucial role in preventing the loss of biodiversity, lowering greenhouse gas emissions, and improving farming efficiency without compromising future generations' needs (Obaisi et al., 2022). Key practices in sustainable agriculture include crop rotation, managing nutritional deficiencies, pest control, recycling, and water harvesting, all contributing to a safer environment (Dhanaraju et al., 2022). As greenhouse gas emissions affect both ecosystems and living organisms, fostering a healthier environment is essential for their well-being (Latake et al., 2015). However, operational viability in agriculture has often focused primarily on economic factors, neglecting social and environmental demands, which often conflict (Meinke, 2019). To transition to sustainable development, it's vital to integrate government actions, improve working conditions, and foster collaboration among stakeholders (El Bilali et al., 2021). Additionally, integrating open data with agricultural information through sophisticated algorithms can enhance decision-making and operational efficiency, improving practices like irrigation and fertilization (Gamage et al., 2024). Finally, ensuring soil sustainability and biodiversity through advanced biomonitoring systems and using natural pesticides can help maintain healthy ecosystems and improve both economic and environmental outcomes (Butnariu, 2015; Butu et al., 2020; Flak, 2020).

17.10 SYNERGISTIC ROLE OF AGRICULTURE TOWARD SUSTAINABLE DEVELOPMENT

Sustainable agriculture, as defined by the FAO, aims to produce safe, high-quality agricultural products while safeguarding the environment, improving social and economic conditions for farmers and communities, and ensuring the welfare of farmed species (Castro et al., 2021). It supports the achievement of several UN Sustainable Development Goals (SDGs), including those related to poverty, hunger, inequality, responsible production, climate change, and ecosystem health (Castro et al., 2021). By balancing economic, social, and environmental factors, sustainable agriculture ensures the long-term provision of food, fiber, and other resources for an increasing population (Pawlak & Kołodziejczak, 2020). The FAO has outlined five guiding principles for advancing sustainability in agriculture, which include improving resource efficiency, protecting natural assets, supporting the rural economy and social wellbeing, mitigating climate change and market risks, and promoting good governance (Gamage et al., 2024).

17.11 METHODS FOR ENCOURAGING SUSTAINABLE AGRICULTURE

Promoting sustainable agriculture requires a multifaceted approach that considers socioeconomic factors, environmental impacts, and farming practices. Education and training are essential, providing farmers with resources and guidance on practices like soil conservation and crop rotation (Coulibaly et al., 2021; Karunathilake et al., 2023). Innovation and technology, such as genetically modified crops and precision agriculture tools, can enhance sustainability by reducing environmental impact (Muhie, 2022). Government policy support, including incentives for eco-friendly practices and biodiversity preservation, is critical in encouraging sustainable behaviors (Brunelle et al., 2024). Additionally, promoting agroecology and creating market demand through labeling systems can further support sustainable farming methods, while fostering collaboration among stakeholders (Cook et al., 2023; Gamage et al., 2023; Velten et al., 2021).

17.12 CONCLUSION

In agriculture, biotechnology has become a versatile instrument that includes a wide range of approaches, from sophisticated genetic engineering to conventional breeding methods. With agricultural biotechnology as a major subset of the Indian biotech industry, this all-encompassing strategy has been crucial to the agricultural revolutions of the twenty-first century, greatly boosting production and the socio-economic growth of nations. Misconceptions and strict regulations have resulted from the link of biotechnology with industrial farming methods in several nations. Addressing the misconception that underpins regulatory difficulties requires making a distinction between the safety of the final product and the biotechnology production process. When used effectively, biotechnology improves plants, modifies soil, and creates substitutes for fuel inputs in agricultural machinery, all of which contribute to sustainability in agriculture.

REFERENCES

Ahmar, S., Gill, R. A., Jung, K. H., Faheem, A., Qasim, M. U., Mubeen, M., & Zhou, W. (2020). Conventional and molecular techniques from simple breeding to speed breeding in crop plants: Recent advances and future outlook. *International Journal of Molecular Sciences*, *21*(7), 2590.

Ansari, S. A., & Tabassum, S. (2018). A new perspective on the adoption of sustainable agricultural practices: A review. *Current Agricultural Research Journal*, 6(2). http://dx.doi.org/10.12944/CARJ.6.2.04

Asdal, Å. (2008). State of plant genetic resources for food and agriculture in Norway. *Oppdragsrapportfra Skog ogLandskap*. https://nibio.brage.unit.no/nibio-xmlui/handle/11250/2484432

Badiyal, A., Mahajan, R., Rana, R. S., Sood, R., Walia, A., Rana, T., Manhas, S., & Jayswal, D. K. (2024). Synergizing biotechnology and natural farming: Pioneering agricultural sustainability through innovative interventions. *Frontiers in Plant Science*, *15*, 1280846. https://doi.org/10.3389/fpls.2024.1280846

Bešić, C., Bakator, M., Đorđević, D., Ćoćkalo, D., & Stanisavljev, S. (2022). Modern agribusiness within the framework of society 5.0. *Economics of Agriculture*, *69*(2), 365–383.

Brunelle, T., Chakir, R., Carpentier, A., Dorin, B., Goll, D., Guilpart, N., Maggi, F., Makowski, D., Nesme, T., Roosen, J., & Tang, F. H. (2024). Reducing chemical inputs in agriculture requires a system change. *Communications Earth and Environment*, *5*(1), 369.

Business Standard. (2013). *Agri-biotech is 3rd largest in Indian biotech sector: USDA*. Press Trust of India New Delhi, Retrieved on 4th January 2025 from https://www.business-standard.com/article/economy-policy/agri-biotech-is-3rd-largest-in-indian-biotech-sector-usda-110100600177_1.html

Butnariu, M. (2015). Markers, indicators of soil pollution. In: *Environmental indicators* (pp. 343–364). https://doi.org/10.1007/978-94-017-9499-2_22

Butu, M., Stef, R., Grozea, I., Corneanu, M., & Butnariu, M. (2020). Biopesticides: Clean and viable technology for healthy environment. In: *Bioremediation and biotechnology: Sustainable approaches to pollution degradation* (pp. 107–151), University of Agricultural Sciences.

Carrière, Y., Crickmore, N., & Tabashnik, B. E. (2015). Optimizing pyramided transgenic Bt crops for sustainable pest management. *Nature Biotechnology*, *33*, 161–168.

Castro, G. D. R., Fernandez, M. C. G., & Colsa, A. U. (2021). Unleashing the convergence amid digitalization and sustainability towards pursuing the Sustainable Development Goals (SDGs): A holistic review. *Journal of Cleaner Production*, *280*, 122204.

Cattaneo, M. G., Yafuso, C., Schmidt, C., Huang, C. Y., Rahman, M., Olson, C., Ellers-Kirk, C., Orr, B. J., Marsh, S. E., Antilla, L., & Dutilleul, P. (2006). Farm-scale evaluation of the impacts of transgenic cotton on biodiversity, pesticide use, and yield. *Proceedings of the National Academy of Sciences*, *103*(20), 7571–7576.

Chamberlain, D. E., Freeman, S. N., & Vickery, J. A. (2007). The effects of GMHT crops on bird abundance in arable fields in the UK. *Agriculture, Ecosystems and Environment*, *118*(1–4), 350–356.

Chantre, R. N., Singla-Pareek, S. L., & Pareek, A. (2016). Genomics approaches for improving salinity stress tolerance in crop plants. *Current Genomics*, *17*, 343–357. https://doi.org/10.2174/1389202917666160331202517

Cheruku, J. K., & Katekar, V. (2023). *Digitalisation of agriculture in India: The case for doubling farmers' income* (pp. 194–205). Indian Institute of Public Administration.

Confederation of Indian Industry. (2021). *Digital agriculture*. Advanced Technologies Reshaping Indian Agriculture: Technology Led Resilience for Atmanirbhar Bharat.

Cook, B., Costa Leite, J., Rayner, M., Stoffel, S., Van Rijn, E., & Wollgast, J. (2023). Consumer interaction with sustainability labelling on food products: A narrative literature review. *Nutrients*, *15*(17), 3837.

Coulibaly, T. P., Du, J., & Diakité, D. (2021). Sustainable agricultural practices adoption. *Agriculture*, *67*(4), 166–176.

Curatti, L., & Rubio, L. M. (2014). Challenges to develop nitrogen-fixing cereals by direct *nif*-gene transfer. *Plant Science*, *225*, 130–137. https://doi.org/10.1016/j.plantsci.2014.06.003

Dar, S. A., Wani, S. H., Mir, S. H., Showkat, A., Dolkar, T., & Dawa, T. (2021). Biopesticides: Mode of action, efficacy and scope in pest management. *Journal of Advanced Research in Biochemistry and Pharmacology*, *4*, 1–8.

Das, S., Ray, M. K., Panday, D., & Mishra, P. K. (2023). Role of biotechnology in creating sustainable agriculture. *PLOS Sustainability and Transformation*, *2*(7), e0000069.

Da Silveira, F., Lermen, F. H., & Amaral, F. G. (2021). An overview of agriculture 4.0 development: Systematic review of descriptions, technologies, barriers, advantages, and disadvantages. *Computers and Electronics in Agriculture*, *189*, 106405.

Dhanaraju, M., Chenniappan, P., Ramalingam, K., Pazhanivelan, S., & Kaliaperumal, R. (2022). Smart farming: Internet of Things (IoT)-based sustainable agriculture. *Agriculture*, *12*(10), 1745.

Ebbisa, A. (2022). Mechanisms underlying cereal/legume intercropping as nature-based biofortification: A review. *Food Production, Processing and Nutrition*, *4*, 19. https://doi.org/10.1186/s43014-022-00096-y

Economic Survey. (2016). *Chapter 3: Spreading JAM across India's economy*. Ministry of Finance, Government of India. https://www.indiabudget.gov.in/budget2016-2017/es2015-16/echapvol1-03.pdf

El Bilali, H., Strassner, C., & Ben Hassen, T. (2021). Sustainable agri-food systems: Environment, economy, society, and policy. *Sustainability*, *13*(11), 6260.

Ellis, J. G., Lagudah, E. S., Spielmeyer, W., & Dodds, P. N. (2014). The past, present and future of breeding rust resistant wheat. *Frontiers in Plant Science*, *5*, 641.

Environmental Protection Agency (EPA). (2001). "Insect resistance management," in Biopesticides Registration Action Document *Bacillus thuringiensis* Plant-Incorporated Protectants (US Environmental Protection Agency, Washington, DC). Available at: www.epa.gov/pesticides/biopesticides/pips/bt_brad2/4-irm.pdf.

Estrada, A. C., Díaz, D. V., & Hernández, C. A. M. (2017). The role of biotechnology in agricultural production and food supply. *Agricultural Science and Research: Latin American Journal of Agricultural Sciences*, *44*(1), 1–11.

FAO (Food and Agriculture Organization of the United Nations). (2011). *Biotechnologies for agricultural development*.

Fathi, K. (2022). The sustainable society. In *Multi-resilience-development-sustainability: Requirements for securing the future of societies in the 21st century* (pp. 169–199). Springer Fachmedien Wiesbaden.

Fielke, S., Taylor, B., & Jakku, E. (2020). Digitalisation of agricultural knowledge and advice networks: A state-of-the-art review. *Agricultural Systems*, *180*, 102763.

Flak, J. (2020). Technologies for sustainable biomass supply—Overview of market offering. *Agronomy*, *10*(6), 798.

Food and Agriculture Organization of the United Nations (FAO). (2018). *Transforming food and agriculture to achieve the SDGs*. FAO.

Gamage, A., Gangahagedara, R., Gamage, J., Jayasinghe, N., Kodikara, N., Suraweera, P., & Merah, O. (2023). Role of organic farming for achieving sustainability in agriculture. *Farming System*, *1*(1), 100005. https://doi.org/10.1016/j.farsys.2023.100005

Gamage, A., Gangahagedara, R., Subasinghe, S., Gamage, J., Guruge, C., Senaratne, S., Randika, T., Rathnayake, C., Hameed, Z., Madhujith, T., & Merah, O. (2024). Advancing sustainability: The impact of emerging technologies in agriculture. *Current Plant Biology*, 100420. https://doi.org/10.1016/j.cpb.2024.100420

Goel, R. K., Yadav, C. S., Vishnoi, S., & Rastogi, R. (2021). Smart agriculture–Urgent need of the day in developing countries. *Sustainable Computing: Informatics and Systems*, *30*, 100512.

Ingram, J., Maye, D., Bailye, C., Barnes, A., Bear, C., Bell, M., Cutress, D., Davies, L., de Boon, A., Dinnie, L., & Gairdner, J. (2022). What are the priority research questions for digital agriculture? *Land Use Policy*, *114*, 105962.

Jiang, X., Deng, Z., Chen, G., Hu, H., Geng, Y., Zhang, Z., Li, H., & Zhao, J. (2023). Genome-wide association mapping of arabinoxylan and resistant starch concentration in common wheat (*Triticum aestivum* L.). *Cereal Research Communications*, *52*(3), 949–959.

Karunathilake, E. M. B. M., Le, A. T., Heo, S., Chung, Y. S., & Mansoor, S. (2023). The path to smart farming: Innovations and opportunities in precision agriculture. *Agriculture*, *13*(8), 1593.

Katekar, V., & Cheruku, J. K. (2022). The application of drone technology for sustainable agriculture in India. *Current Agriculture Research Journal*, *10*(3). http://dx.doi.org/10.12944/CARJ.10.3.19

Kelliher, T., Starr, D., Su, X., Tang, G., Chen, Z., Carter, J., Wittich, P. E., Dong, S., Green, J., Burch, E., McCuiston, J. M., Gu, W., Sun, Y., Strebe, T., Roberts, J., Bate, N. J., & Que, Q. (2019). One-step genome editing of elite crop germplasm during haploid induction. *Nature Biotechnology*, *37*(3), 287–292.

Khanna, K., Kumar, P., Ohri, P., & Bhardwaj, R. (2023). Harnessing the role of selenium in soil–plant-microbe ecosystem: Ecophysiological mechanisms and future prospects. *Plant Growth Regulation*, *100*, 197–217. https://doi.org/10.1007/s10725-022-00830-z

Krishna, T. A., Maharajan, T., & Ceasar, S. A. (2023). The role of membrane transporters in the bio-fortification of zinc and iron in plants. *Biological Trace Element Research*, *201*, 464–478.

Kumar, S., Nain, M. S., & Sangeetha, V. (2023). Determinants and constraints for adoption of zero budget natural farming (ZBNF) practices in farmer field school. *Indian Journal of Extension Education*, *59*(4), 135–140.

Latake, P. T., Pawar, P., & Ranveer, A. C. (2015). The greenhouse effect and its impacts on environment. *International Journal of Innovative Research and Creative Technology*, *1*(3), 333–337.

Lima, M. G. B. (2022). Just transition towards a bioeconomy: Four dimensions in Brazil, India and Indonesia. For. *Policy Economics*, *136*, 102684.

Meinke, H. (2019). The role of modeling and systems thinking in contemporary agriculture. In *Sustainable food supply chains* (pp. 39–47). Academic Press.

Ministry of Agriculture and Farmers' welfare (2018). *Doubling farmers' income by 2022 committee report*. Government of India.

Muhie, S. H. (2022). Novel approaches and practices to sustainable agriculture. *Journal of Agriculture and Food Research*, *10*, 100446.

Naranjo, S. E. (2005). Long-term assessment of the effects of transgenic Bt cotton on the abundance of nontarget arthropod natural enemies. *Environmental Entomology*, *34*(5), 1193–1210.

National Mission on Natural Farming Management and Knowledge (NMNFMK). (2025). National Mission on Natural Farming Management and Knowledge (NMNFMK) Portal. Government of India. Retrieved on 4th January 2025 from https://naturalfarming.dac.gov.in/NaturalFarming/Concept

Obaisi, A. I., Adegbeye, M. J., Elghandour, M. M. M. Y., Barbabosa-Pliego, A., & Salem, A. Z. M. (2022). Natural resource management and sustainable agriculture. In Lackner, M., Sajjadi, B., Chen, W.Y. (Eds.), *Handbook of climate change mitigation and adaptation*. Springer.

Paine, J. A., Shipton, C. A., Chaggar, S., Howells, R. M., Kennedy, M. J., Vernon, G. et al. (2005). Improving the nutritional value of Golden Rice through increased pro-vitamin A content. *Nature Biotechnology*, *23*, 482–487.

Pawlak, K., & Kołodziejczak, M. (2020). The role of agriculture in ensuring food security in developing countries: Considerations in the context of the problem of sustainable food production. *Sustainability*, *12*(13), 5488.

Prause, L. (2021). Digital agriculture and labor: A few challenges for social sustainability. *Sustainability*, *13*(11), 5980.

Radico Khaitan. (2024). *Natural farming: A step towards brewing a greener future*. Radico Khaitan. Retrieved on 4th January 2025 from https://radicokhaitan.com/new/CSR/blog/natural-farming-a-step-towards-brewing-a-greener-future/

Rai, A. (2023). Digitalization in agriculture for innovative transformation in India. *Indian Farming*, *73*(6), 30–34.

Rajesh, T., Athare, T., Roy, S. K., Dhavale, P. D., & Bhalerao, P. (2024). Journey of natural farming the pathway towards sustainable agriculture. *Indian Farming*, *74*(3), 48–51.

Reddy, A. A. (2018). Impact study of Paramparagath Krishi Vikas Yojana (organic agriculture) scheme of India. *Impact Study of Paramparagat Krishi Vikas Yojana*. National Institute of Agricultural Extension Management (MANAGE), Hyderabad, 210.

Sanahuja, G., Banakar, R., Twyman, R. M., Capell, T., & Christou, P. (2011). *Bacillus thuringiensis*: A century of research, development and commercial applications. *Plant Biotechnology Journal*, *9*, 283–300.

Schnebelin, É, Labarthe, P., & Touzard, J. M. (2021). How digitalisation interacts with ecologisation? Perspectives from actors of the French Agricultural Innovation System. *Journal of Rural Studies*, *86*, 599–610.

Singh, R., Das, R., Sangwan, S., Rohatgi, B., Khanam, R., Peera, S. K. P. G., Das, S., Lyngdoh, Y. A., Langyan, S., Shukla, A., Shrivastava, M., & Misra, S. (2021). Utilization of agro−industrial waste for sustainable green production: A review. *Environmental Sustainability*, *4*, 619–636.

Sneller, C. H. (2003). Impact of transgenic genotypes and subdivision on diversity within elite North American soybean germplasm. *Crop Science*, *43*(1), 409–414.

Tan, S., Evans, R., & Singh, B. (2006). Herbicidal inhibitors of amino acid biosynthesis and herbicide-tolerant crops. *Amino Acids*, *30*, 195–204.

Velten, S., Jager, N. W., & Newig, J. (2021). Success of collaboration for sustainable agriculture: A case study meta-analysis. *Environment, Development and Sustainability*, 1–23.

Zhang, Y., Bai, Y., Wu, G., Zou, S., Chen, Y., Gao, C., & Tang, D. (2017). Simultaneous modification of three homoeologs of *TaEDR1* by genome editing enhances powdery mildew resistance in wheat. *The Plant Journal*, *91*(4), 714–724.

18 Nanobiotechnology Approaches for Climate Change Adaptation in Crops

Durgadatta Sahoo, D. Swapna, Amlan Mohanty, Barsharani Sethi, Parul Pratyasa Samal, Akankshya Mohapatra, and Jyoti Prakash Sahoo

18.1 INTRODUCTION

Climate change involves long-term shifts in climatic factors like temperature, rainfall, and humidity, with far-reaching impacts on local, national, and global scales. It threatens food security, crop productivity, and natural resources while hindering progress toward the United Nations' Sustainable Development Goals (Davis et al., 2020). Rising global temperatures and frequent heatwaves exacerbate these challenges, as heat stress temperatures above 35°C disrupt plant growth, reduce crop yields, and harm biodiversity (Lee et al., 2021). Extreme heat also affects urban life quality, worker productivity, and human health, with significant losses observed in species and crop yields in regions like China and the U.S. (Liu et al., 2024). To combat heat stress, plants activate thermomorphogenesis and heat acclimation, but these processes generate reactive oxygen species (ROS), damaging cellular structures and reducing photosynthesis (Singh et al., 2020). Nanotechnology offers promising solutions, such as nano-priming seeds, nanofertilizers, and nanopesticides, which enhance crop resilience and productivity (Sharma et al., 2020). Engineered nanomaterials (ENMs) are intentionally designed materials with at least one dimension between 1 and 100 nm, exhibiting unique properties for diverse applications in medicine, electronics, energy, and more (Das et al., 2021a, b). However, Figure 18.1 shows how ENMs can boost crop resilience.

Nano-sensors and nano-harvest technologies enhance smart farming by reducing food spoilage and improving precision agriculture, while nanomaterials, such as nanofertilizers, nanopesticides, and nanocarriers, boost crop resilience and sustainability (Rodriguez et al., 2019). Climate change intensifies agricultural challenges, including water scarcity, erratic weather, and increased pest resistance, threatening staple crops like maize and soybean (Thomson et al., 2019). With the global population projected to reach 9.1 billion by 2050 and arable land shrinking, nanotechnology offers innovative solutions, such as slow-release fertilizers and nano-herbicides, to

DOI: 10.1201/9781003597797-18

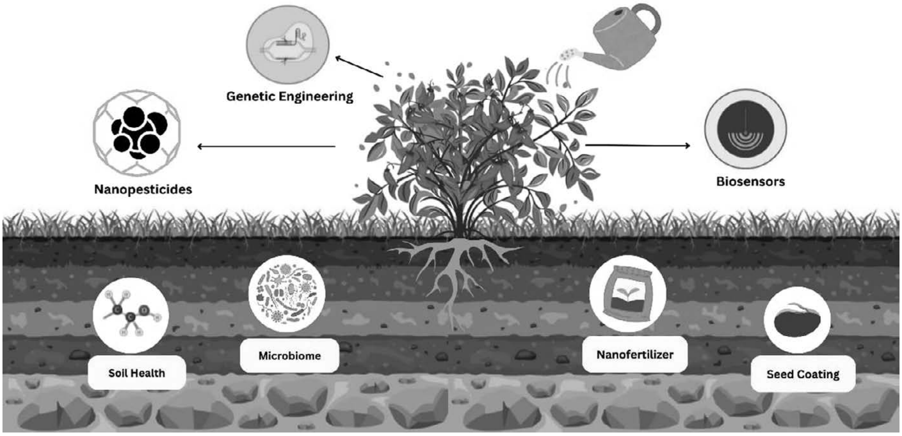


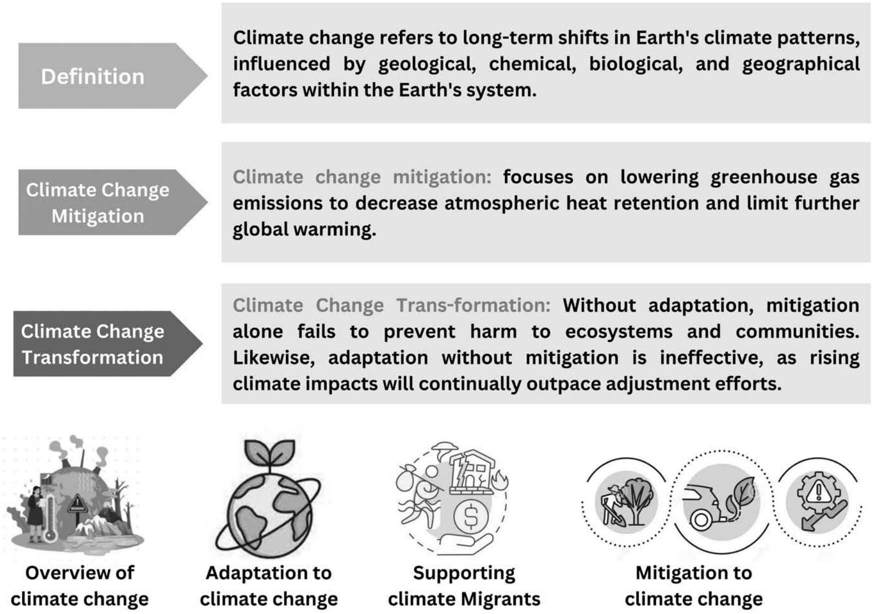
FIGURE 18.1 ENMs boost crop resilience via nanobiosensors, nanofertilizers, nanopesticides, and genetic nanocarriers in sustainable agriculture.

improve resource efficiency and stress tolerance (Zafar et al., 2021). This chapter summarizes how nanobiotechnology enhances crop resilience to climate change through ENMs like nanofertilizers, nanopesticides, and nano-sensors and improves stress tolerance, nutrient efficiency, and yield stability while addressing heat stress, water scarcity, and soil degradation for sustainable agriculture.

18.2 CLIMATE CHANGE: A GLOBAL ISSUE

Climate change refers to long-term shifts in temperature, precipitation, and weather patterns, primarily driven by human activities like fossil fuel combustion and deforestation (Hurlimann et al., 2021). Greenhouse gases (GHGs), such as carbon dioxide (CO_2) and methane (CH_4), trap heat in the atmosphere, exacerbating the greenhouse effect and leading to global warming (Yeleliere et al., 2023). Adaptation involves adjusting agricultural, ecological, or societal systems to mitigate climate impacts, while mitigation focuses on reducing GHG emissions through renewable energy and carbon sequestration (El-Ramady et al., 2023). Climate-smart agriculture (CSA) integrates sustainable practices like drought-resistant crops and precision farming to enhance resilience and productivity (Sahoo et al., 2023).

Abiotic stress, including drought, heat, and salinity, threatens crop yields, whereas biotic stress involves climate-driven pest and disease outbreaks (Ritchie et al., 2023). Without mitigation, these trends risk irreversible damage to ecosystems, food systems, and human well-being (Chausali et al., 2023). Agriculture is particularly vulnerable, facing heat stress, water scarcity, and soil degradation, even as the sector contributes 18.4% of global GHG emissions (Brevik et al., 2012). CSA and nanotechnology offer solutions, such as carbon sequestration nanomaterials (Wang et al., 2023) and resilient farming practices (Wakweya et al., 2023). Equitable implementation remains a challenge, as smallholder farmers often lack access to advanced technologies (Ijaz et al., 2023). Figure 18.2 summarizes the key terms related to climate change.



Climate change refers to long-term shifts in Earth's climate patterns, influenced by geological, chemical, biological, and geographical factors within the Earth's system.

Climate change mitigation: focuses on lowering greenhouse gas emissions to decrease atmospheric heat retention and limit further global warming.

Climate Change Trans-formation: Without adaptation, mitigation alone fails to prevent harm to ecosystems and communities. Likewise, adaptation without mitigation is ineffective, as rising climate impacts will continually outpace adjustment efforts.

FIGURE 18.2 Key terms related to climate change and their definitions.

18.3 APPLICATION OF ENMS IN IMPROVING STRESS RESILIENCE OF CROP PLANTS

ENMs have diverse agricultural applications, serving as nanobiosensors, nanofertilizers, nanopesticides, and nanocarriers for genetic engineering. These innovations enhance soil health, improve pathogen control, and boost crop resilience (Mazumder et al., 2023). ENMs also optimize nutrient delivery, enable stress prediction, support genetic modifications, and restore soil microbiomes, enhancing yields while reducing environmental impact (Pal et al., 2023). Table 18.1 provides a summary of various nano-enabled strategies for crop improvement.

18.3.1 NANOFERTILIZERS FOR EFFICIENT UTILIZATION OF AGROCHEMICALS

Nanotechnology offers innovative approaches to improve fertilization efficiency through direct nutrient uptake, targeted delivery, and controlled-release mechanisms (Rehman et al., 2022). Among various nanoparticles (NPs), gold (AuNPs) and silver NPs (AgNPs) are particularly notable for their ease of synthesis and bio-conjugation properties, though their primary role is not nutritional supplementation but rather growth enhancement (Kanwar et al., 2019). Tailored NPs, including Zn, Ag, Cu, Fe, Au, Ni, and metal oxides (Al_2O_3, SiO_2, TiO_2, CeO_2, Fe_2O_4), have been engineered to optimize plant processes (Nayak et al., 2025). For instance, nanoscale iron boosts germination and chlorophyll content in rice and maize (Prerna et al., 2021), while titanium NPs (TiNPs) enhance photosynthetic efficiency (Larue et al., 2018).

TABLE 18.1
Various Nano-Enabled Strategies for Crop Improvement, Associated Challenges, and Future Implications

Nano-Based Strategy	Type of Nanoparticles (NPs) Used	Plant Studied	Improved Crop Characteristics	Challenges	Recommendations	References
Nanofertilizers	Carbon nanoparticles (CNPs)	*Zea mays, Glycine max*	Enhanced crop growth, improved soil quality, increased nutrient uptake, and photosynthetic efficiency	Limited understanding of nanoparticle behavior in soil and plant systems	Research on NP performance across different soil conditions and their internalization mechanisms	Xin et al. (2022)
	Zincated nanoclay polymer composites (ZNCPCs)	–	Improved soil moisture retention, slow release of nutrients	Long-term impact on soil health is not well studied	Investigate the long-term effects of nanoclay amendments on soil ecosystems	Mandal et al. (2018)
	Pristine (ZnO MNPs) and sulfidized zinc oxide nanoparticles (s-ZnO MNPs)	–	Increased nutrient availability and disease resistance	Limited understanding of plant-microbe interactions	Optimize nanomaterials for beneficial plant-microbe interactions	Chen et al. (2023)
Nanopesticides	Metal NPs (CuO, ZnO, FeO)	Tomato (*Solanum lycopersicum*)	Reduced damage from plant pathogens	Unclear interactions between nanopesticides and pathogens	Further studies on non-target effects and ecological risks	Jiang et al. (2021)
Nano-sensors	FRET nano-sensors	*Arabidopsis*	Monitors nutrient flux in intact plant organs	Limited integration with precision agriculture	Develop integrated nano-sensor technologies for smart farming	Chaudhuri et al. (2008)
	Multiwalled carbon nanotube-based zinc nanocomposite (ZnO/MWCNT)	-	Early detection of stress biomarkers (e.g., viral infections)	Limited detection across crop varieties	Expand biosensor applications across different crops	Tahir et al. (2017)

Nanocarriers	Delaminated layered double hydroxide lactate nanosheets (LDH-lactate-NS)	–	Increased stress tolerance and reduced crop losses	Limited research on stability and field performance	Optimize gene delivery mechanisms for higher efficacy	Song et al. (2019)
	Polyethylene oxide (PEO) nanofibers	–	Enhanced plant growth and potential as a seed coating	Limited field research on microbial delivery	Investigate the stability of nanocarriers for beneficial microorganism transport	Campana & Yahdjian (2021)
Other applications	Titanium dioxide (TiO_2) nanoparticles	*Plumeria rubra*	Protection from UV radiation and extreme temperatures	Limited knowledge of environmental safety	Assess environmental impact and long-term safety	Bundele et al. (2023)
	TiO_2/montmorillonite clay (MMT) nanocomposite powder	–	Reduced heavy metal uptake in crops	Limited understanding of nanoclays' interactions with heavy metals	Study interactions between nanoclays and contaminants	James et al. (2023)
	Nanobubble water	Palm oil mill effluent	Increased water efficiency and higher crop yield	Limited scalability for agricultural use	Research the scalability and economic feasibility of nanobubble irrigation	He et al. (2022)
	Silver nanoparticles (AgNPs)	Watermelon, muskmelon	Extended shelf life and reduced spoilage	Concerns regarding food safety and human health	Evaluate the safety of nanomaterials in food preservation	Nayab et al. (2023)

Zinc NPs (ZnNPs) improve seed germination in wheat, onions, peanuts, and soybeans (Larue et al., 2018), and copper carbon nanofertilizers (Cu-CNFs) enhance water uptake, germination, and chlorophyll synthesis in chickpeas (Larue et al., 2018). Carbon-based nanomaterials, such as graphene oxide and quantum dots, also improve root architecture and overall plant development. Nanofertilizers reduce nutrient losses by enabling slow release apatite NPs, for example, increase phosphorus bioavailability, boosting soybean yields by 20.4% (Dimkpa et al., 2019). Surfactant-modified zeolites further enhance phosphate availability (Abdelmigid et al., 2022). Field applications remain limited, but studies suggest nanoscale micronutrients mitigate abiotic stress and improve crop performance even at low doses (Arsic et al., 2020). Figure 18.3 illustrates the various advantages of using NPs made from different chemical elements in plants.

18.3.2 NANOPESTICIDES FOR ENHANCED CONTROL EFFICACY OF PLANT PATHOGENS

Sustainable crop production requires minimizing losses caused by pathogens, pests, and environmental stresses. Nano-enabled pesticides present a promising solution, as conventional pesticides are often inefficient, with only about 0.1% reaching their intended targets (Dweh et al., 2023). Metal NPs, such as zinc and silver, have demonstrated efficacy against fungal pathogens. Zinc-based compounds suppress *Fusarium* head blight in wheat, while AgNPs inhibit *Bipolaris sorokiniana* and *Magnaporthe grisea* (responsible for rice blast) (Norman et al., 2023). Chitosan-based NPs also enhance plant resistance; for instance, biofunctionalized manganese NPs (Bio-MnNPs) suppressed *Fusarium* wilt in watermelons by activating host defenses and modulating soil microbiota (Noman et al.,

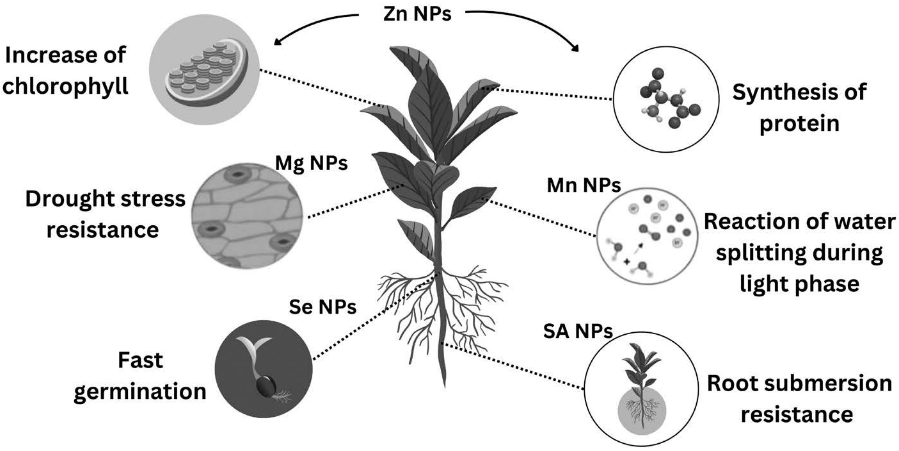


FIGURE 18.3 Schematic illustration of the various advantages of using nanoparticles made from different chemical elements in plants.

2023). Targeted delivery systems, such as enzyme-responsive nanocarriers (e.g., β-glucanase-modified mesoporous silica NPs), improve pesticide precision (Kaziem et al., 2022). Additionally, Cu-chitosan nanocomposites exhibit dual functionality in maize cultivation, offering both antifungal properties and plant growth benefits (Nagger et al., 2022).

18.3.3 NANO-SENSORS FOR IMPROVED PRECISION FARMING

Agricultural automation has significantly enhanced crop management efficiency while reducing labor dependency and promoting environmental sustainability (Saravanadevi et al., 2022). A key innovation in this field is plant nano-sensors, which facilitate direct farmer-plant communication by detecting stress signals and nutrient deficiencies, converting them into measurable electrical or optical outputs for real-time monitoring (Khandelwal et al., 2019). These sensors, integrated with technologies like GPS and GNSS, enable automated tracking of plant health, soil conditions, and stress levels, optimizing resource use and preventing crop losses through precision farming (Giraldo et al., 2019). While nano-sensors offer benefits such as pathogen detection and improved micronutrient absorption, concerns persist regarding potential adverse effects, including genomic instability and hormonal disruptions (Yan et al., 2022). Despite these challenges, studies demonstrate their transformative potential; for instance, DNA-functionalized carbon nanotubes detecting H_2O_2 in *Arabidopsis* (Wu et al. 2020a; Wu et al. 2020b) and manganese NPs mitigating salinity stress in pepper seedlings (Ye et al., 2020) highlight their role in advancing sustainable, smart agriculture.

18.3.4 NANOCARRIERS FOR THE EFFECTIVE DELIVERY OF BIOMOLECULES

Nanotechnology offers innovative solutions for enhancing genetic material delivery in plants, improving gene editing (e.g., CRISPR-Cas9), and stabilizing biomolecules like double-stranded RNA (dsRNA) for pest control (Demirer et al., 2019). However, delivering these molecules across rigid plant cell walls remains a challenge. Nanocarriers enable targeted, non-invasive delivery of CRISPR components without foreign DNA integration, though further research is needed on NP-plant interactions (Landry et al., 2019). Nanotechnology in Disease Detection and RNAi-Based Pest Control Plasmonic nanomaterials integrated with smartphone-based diagnostics allow non-invasive plant disease detection, though field scalability requires refinement (Kumar et al., 2021). In pest management, dsRNA-mediated gene silencing provides an alternative to transgenic crops, but its efficacy is limited by rapid degradation on leaf surfaces. Nanomaterials, such as layered double-hydroxide (LDH) clay nanosheets, stabilize dsRNA and enable controlled release, extending protection against viruses (Landry et al., 2019). While dsRNA is highly effective against nematodes and beetles, its efficiency varies among insects, particularly lepidopterans (moths and butterflies), due to differences in RNAi machinery (Zhang et al., 2020). Environmental and Sustainability Considerations Synthetic nanocarriers for biomolecule delivery often lack biodegradability, whereas those designed for pesticides and nutrients show better environmental compatibility (Das et al., 2020).

18.4 MECHANISM UNDERLYING NANO-ENABLED SUSTAINABLE CROP PRODUCTION

The integration of nanotechnology into agriculture presents transformative potential for enhancing sustainable farming, food security, and environmental resilience (Wang et al., 2022). Among nanomaterials, carbon dots (CDs) have gained prominence due to their biocompatibility, tunable photoluminescence, and low toxicity (Maholiya et al., 2023). CDs enhance photosynthesis, improve seed germination and water uptake, and protect crops from biotic and abiotic stresses through antimicrobial and antioxidant properties. Additionally, they enable precision agriculture by facilitating herbicide and pesticide detection via smart sensing systems. Beyond CDs, nanotechnology applications in horticulture, such as nanofertilizers, nanopesticides, and nano-sensors, enhance crop productivity, shelf life, and quality (Rana et al., 2021). Research on polyamines (PAs) as biostimulants further highlights their role in boosting plant growth and stress tolerance, with potential for optimizing crop resilience through PA signaling pathways (Tyagi et al., 2022).

Nanotechnology also addresses climate-related challenges, such as heat stress. Zinc oxide NPs (n-ZnO) applied as a foliar spray (90 mg/L) significantly mitigate heat stress in alfalfa, preserving growth and physiological traits more effectively than post-stress treatments (Kareem et al., 2022). Similarly, metal-based NPs exhibit strong antimicrobial properties, protecting crops from pathogens. For instance, studies on *Fritillaria imperialis* demonstrate enhanced freezing tolerance through NP-induced activation of Ca^{2+} signaling and antioxidant systems (Li et al., 2023).

18.4.1 Promotion of Plant Growth

Nanotechnology offers promising solutions to mitigate agricultural losses caused by environmental stresses like heat, drought, and salinity, which are exacerbated by climate change (Vema et al., 2022). Various nanomaterials enhance plant resilience and growth: cerium oxide NPs reduce oxidative stress (Hassanisaadi et al., 2022), carbon nanotubes accelerate root development (AboDalm et al., 2022), and titanium dioxide/silicon NPs improve germination (Behl et al., 2022). Iron oxide NPs boost watermelon yields (Guleria et al., 2022), while mesoporous silica NPs enhance photosynthesis by targeting chloroplasts. Controlled hormone delivery via nanocarriers (Sun & Li, 2022) and zinc oxide NP coatings (Wu et al., 2022) further demonstrate nanotechnology's potential in improving nutrient uptake and stress tolerance.

18.4.2 Reshaping of Plant Microbiome

Nanomaterials show significant potential for enhancing soil health and agricultural sustainability by improving nutrient uptake, water retention, and pathogen resistance (da Silva Junior et al., 2022). Their ability to influence plant-microbe interactions is particularly valuable, as evidenced by studies demonstrating improved crop tolerance to saline irrigation and disease suppression through microbiome regulation (Jayaraman et al., 2021). Various nanomaterials, including copper oxide (CuO), silver (Ag), titanium oxide (TiO_2), and carbon-based structures, exhibit distinct effects on soil microbial communities and plant performance. Carbon-based nanomaterials, for instance, enhance seed germination, photosynthesis, and stress resilience while promoting the production of bioactive compounds (Zhang et al., 2020). However, their impact on soil ecology varies: high concentrations (100 mg/kg) of Ag, CuO, and ZnO NPs can disrupt microbial balance, whereas TiO_2 NPs show minimal effects even at elevated doses (Zhang et al., 2020).

18.4.3 Improvement of Soil Quality

Soil improvement plays a key role in boosting crop production and food quality, especially in areas affected by degradation and environmental stress. Traditional soil conditioning methods rarely use nanotechnology (da Silva Junior et al., 2022). Emerging nanomaterials including potassium-enriched feldspar-based mineral conditioners synthesized through hydrothermal methods demonstrate promising capabilities for soil remediation by optimizing pH levels, improving soil structure, and mitigating toxic aluminum and cadmium accumulation. Biodegradable cross-linkers can also improve water retention. Chitosan NPs have been found to enhance wheat yield and soil characteristics (Das et al., 2021b). Despite these benefits, most studies remain limited to lab-scale experiments. Large-scale feasibility assessments are lacking (Elsheery et al., 2020). Traditional methods dominate due to their long-standing use and the high material demands of nano-based solutions. Precision agriculture offers limited opportunities for nanotechnology adoption. More field-scale testing is needed to validate its potential (Das et al., 2021a, b).

18.5 NANOBIOTECHNOLOGY IN CLIMATE CHANGE MITIGATION AND AGRICULTURE

Nanotechnology plays a crucial role in mitigating climate change impacts on plants by enhancing energy efficiency, reducing GHG emissions, and improving carbon sequestration through biological, physical, chemical, and bioengineering approaches (Rai et al., 2018). At the molecular level, nano-enhanced plants exhibit increased photosynthetic efficiency for greater CO_2 absorption, while nano-amendments in soil reduce nitrous oxide emissions by improving nutrient availability and decreasing synthetic fertilizer dependency (Wu et al., 2020). Additionally, nanotechnology strengthens plant resilience to abiotic and biotic stresses through precision delivery systems, such as nano-formulated pesticides, silica NPs for drought resistance, and carbon-based nano-sensors for real-time soil monitoring (Singh & Kalia, 2019). These innovations not only optimize agricultural productivity and soil health but also promote sustainable resource use, aligning with global climate mitigation goals (Chen et al., 2022). However, a sustainable approach for climate change mitigation by enhancing abiotic and biotic stress tolerance through nanotechnology is indexed in Figure 18.4.

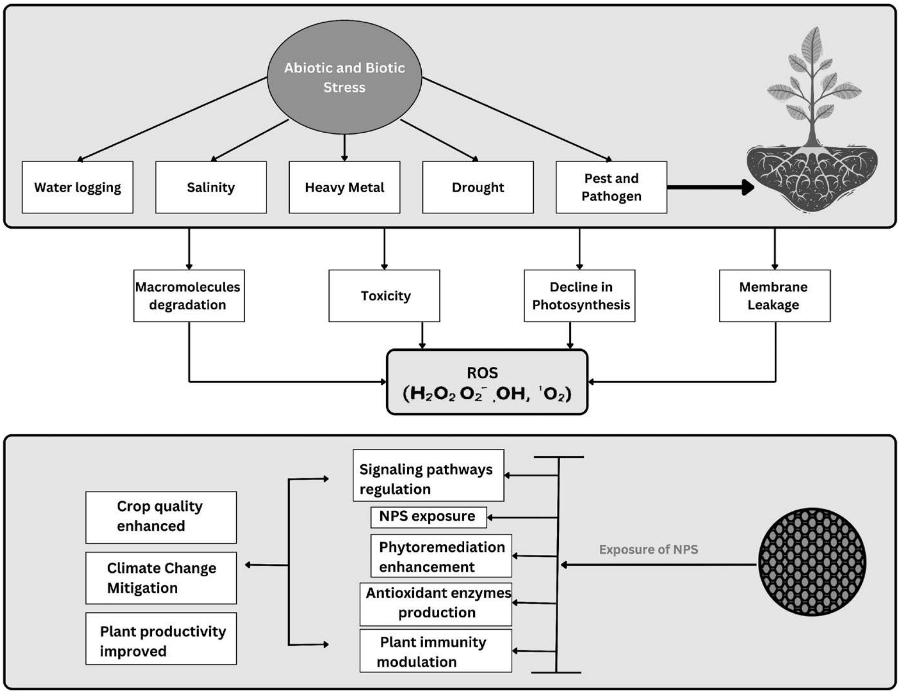


FIGURE 18.4 Enhancing plant tolerance to abiotic and biotic stress through nanotechnology.

18.6 ENVIRONMENTAL CONCERNS AND NANO-TOXICITY ISSUES OF ENMs

ENMs demonstrate concentration-dependent effects on plants, with both beneficial and harmful outcomes influenced by material type, exposure duration, and environmental conditions (Zhang et al., 2020). While certain ENMs (e.g., CuO, ZnO NPs) inhibit seed germination, reduce root/shoot growth, and impair photosynthesis by inducing oxidative stress in crops like soybean and wheat (Li et al., 2025), others (e.g., CeO_2, Fe_3O_4) enhance stress tolerance by activating antioxidant defenses (Yusefi-Tanha et al., 2020). Toxicity mechanisms include biomembrane damage, gene expression alterations, and disrupted water transport (Yusefi-Tanha et al., 2020), with carbon-based ENMs like fullerenes suppressing energy metabolism genes, while multiwalled carbon nanotubes (MWCNTs) promote cell division (Jogaiah et al., 2021). Environmental concerns persist due to ENM persistence in soils, where metallic NPs disrupt microbial communities (Sager et al., 2022), and photochemically active ENMs (e.g., TiO_2) generate ROS under light (Heikal et al., 2021). Although carbon nanotubes show limited translocation to edible plant parts (Lahiani et al., 2019), long-term ecological risks remain uncertain. For sustainable use, toxicity screening and innovations like polymer-coated ENMs are essential (Chauhan et al., 2022), alongside research on soil microbiome interactions and bioaccumulation potential (Sun & Li, 2022).

18.7 CHALLENGES IN LARGE-SCALE IMPLEMENTATION OF NANO-ENABLED STRATEGIES

The widespread implementation of nano-enabled agricultural technologies faces significant barriers, including insufficient understanding of plant-nanomaterial interactions, difficulties in targeted delivery, and unresolved concerns about impacts on ecosystems, soil health, and human safety (Rai et al., 2022). Critical challenges include ensuring NP biocompatibility, developing effective delivery methods, and establishing regulatory frameworks (Kah et al., 2023). While engineered NPs (ENPs) offer tunable properties for controlled release and enhanced functionality, their concentration-dependent effects under natural conditions require thorough investigation, including transgenerational and trophic chain impacts (Lowry et al., 2022). Comprehensive field studies are essential to validate ENP effects on soil microbiomes, plant systems, and human health, ensuring these technologies enhance productivity without unintended ecological consequences (Lowry et al., 2022). Table 18.2 organizes current research on ENMs in agriculture, detailing their interactions with plants, optimization strategies for efficient delivery, and the need for regulatory frameworks to ensure safe and socially accepted applications.

TABLE 18.2
Regulatory Challenges Regarding Engineered Nanomaterials in Plant Systems

Key Features	Explanation	Research Strategies	References
Biocompatibility of engineered nanomaterials (ENMs) with plant systems.	Investigating how engineered nanomaterials (ENMs) interact with plants to boost growth and stress resilience.	The mechanisms behind plant interactions with engineered nanomaterials (ENMs) are not well understood, requiring in situ research to optimize their use for stress mitigation in plants.	Zuverza-Mena et al. (2023)
Uptake, translocation, and persistence of engineered nanomaterials (ENMs) in plant systems.	Optimizing nanomaterial deployment for efficient plant uptake in field environments.	Key challenges in ENM soil application such as non-targeted delivery and high energy costs can be mitigated through targeted slow-release systems, while improved foliar delivery designs and responsive ENMs are needed to address weather-induced stresses and enhance efficiency.	Dimkpa et al. (2020)
Challenges in regulation and societal approval of engineered nanomaterials (ENMs) persist.	Securing safe implementation and building societal acceptance for agricultural applications of engineered nanomaterials (ENMs).	Developing safety protocols and regulations for ENMs in food crops is essential, as while metal-based ENMs are generally less toxic than ionic forms, controlled biofortification requires strict oversight to balance nutritional benefits with health risks while also addressing ecological, climatic, policy, and public acceptance challenges.	Gogos et al. (2022)

18.8 CONCLUSION

Nanobiotechnology presents transformative potential in enhancing crop adaptation to climate change through innovative solutions such as nano-enhanced agrochemicals, stress-tolerant varieties, and precision monitoring tools. While these technologies offer significant benefits in improving yield stability and sustainability, their widespread adoption depends on addressing biosafety, environmental impact, and regulatory frameworks. Collaborative efforts among scientists, policymakers, and farmers are essential to harness nanobiotechnology's full potential while ensuring ethical and eco-friendly applications. As climate change intensifies, integrating nanobiotechnology into agricultural practices will be crucial for building resilient food systems and safeguarding global food security in a rapidly changing environment.

REFERENCES

Abdelmigid, H. M., Morsi, M. M., Hussien, N. A., & Alyamani, A. A. (2022). Surfactant-modified zeolites for enhanced phosphate availability in sustainable agriculture. *Environmental Science and Pollution Research*, *29*(15), 21512–21523. https://doi.org/10.1007/s11356-021-17356-y

AboDalm, R. D., Hassan, A. K., & Al-Khayri, J. M. (2022). Carbon nanotubes enhance root development and plant growth in date palm. *Plant Cell Reports*, *41*(4), 893–907. https://doi.org/10.1007/s00299-021-02828-9

Arsic, M., Tougaard, S. L., Persson, D. P., Martens, H. J., Doolette, C. L., Lombi, E., Schjoerring, J. K., & Husted, S. (2020). Bioimaging techniques reveal foliar phosphate uptake pathways and leaf phosphorus status. PLANT PHYSIOLOGY, 183(4), 1472–1483. https://doi.org/10.1104/pp.20.00484

Behl, T., Kaur, I., Sehgal, A., Singh, S., Sharma, N., & Bhatia, S. (2022). Titanium dioxide and silicon nanoparticles improve germination and stress tolerance in plants. *Journal of Biotechnology*, *343*, 120–132. https://doi.org/10.1016/j.jbiotec.2021.11.003

Brevik, E. C., Sauer, T. J., & Nelson, D. W. (2012). The past, present, and future of soils and human health studies. *Soil*, *1*(1), 35–46. https://doi.org/10.5194/soil-1-35-2015

Bundele, V., Sharma, A., & Kumari, M. (2023). Titanium dioxide nanoparticles mitigate UV and thermal stress in *Plumeria rubra*: Implications for climate-resilient agriculture. *Plant Physiology and Biochemistry*, *184*, 1–12. https://doi.org/10.1016/j.plaphy.2022.12.012

Campana, S., & Yahdjian, L. (2021). Plant quality and primary productivity modulate plant biomass responses to the joint effects of grazing and fertilization in a mesic grassland. *Applied Vegetation Science*, *24*(2), e12588. https://doi.org/10.1111/avsc.12588

Chaudhuri, B., Hörmann, F., & Frommer, W. B. (2008). Dynamic imaging of glucose flux in live plant tissues using FRET nanosensors. *Nature Protocols*, *3*(12), 1864–1873. https://doi.org/10.1038/nprot.2008.186

Chauhan, N., Tyagi, A. K., Kumar, P., & Malik, A. (2022). Polymer-coated nanomaterials for sustainable agriculture: Current trends and future perspectives. *Environmental Research*, *204*, 112356. https://doi.org/10.1016/j.envres.2021.112356

Chausali, N., Saxena, J., & Prasad, R. (2023). Nanotechnology as a sustainable approach for combating the environmental effects of climate change. *Journal of Agriculture and Food Research*, *11*, 100457. https://doi.org/10.1016/j.jafr.2022.100457

Chen, C., Guo, L., Chen, Y., Qin, P., & Wei, G. (2023). Pristine and sulfidized zinc oxide nanoparticles alter bacterial communities and metabolite profiles in soybean rhizocompartments. *Science of the Total Environment*, *855*, 158697. https://doi.org/10.1016/j.scitotenv.2022.158697

Chen, B., Yan, Y., Yang, Y., Cao, G., Wang, X., Wang, Y., Wan, F., Yin, Q., Wang, Z., Li, Y., Wang, L., Xu, B., You, F., Zhang, Q., & Wang, Y. (2022). A pyroptosis nanotuner for cancer therapy. *Nature Nanotechnology*, *17*(7), 788–798. https://doi.org/10.1038/s41565-022-01125-0

Das, P., Vishakha, K., Banerjee, S., & Ganguli, A. (2021a). Chitosan nanoparticles improve wheat yield and soil quality. *Carbohydrate Polymers*, *267*, 118183. https://doi.org/10.1016/j.carbpol.2021.118183

Das, S., Wolfson, B. P., Tetard, L., Tharkur, J., Bazata, J., & Santra, S. (2020). N-acetyl cysteine coated MnSOD nanoparticles for oxidative stress mitigation in plants. *Environmental Science: Nano*, *7*(1), 180–195. https://doi.org/10.1039/C9EN00971J

Das, S., Wolfson, B. P., Tetard, L., Tharkur, J., Bazata, J., & Santra, S. (2021b). Effect of N-acetyl cysteine coated MnSOD nanoparticles on the oxidative stress induced by abiotic and biotic stresses in *Oryza sativa*. *Plant Physiology and Biochemistry*, *163*, 285–297. https://doi.org/10.1016/j.plaphy.2021.04.006

da Silva Junior, A. H., Macuvele, D. L. P., & Riella, H. G. (2022). Nanomaterials for soil improvement: Current status and future prospects. *Science of the Total Environment*, *806*, 150531. https://doi.org/10.1016/j.scitotenv.2021.150531

Davis, K. F., Downs, S., & Gephart, J. A. (2020). Towards food supply chain resilience to environmental shocks. *Nature Food*, *2*(1), 54–65. https://doi.org/10.1038/s43016-020-00196-3

Demirer, G. S., Zhang, H., Matos, J. L., Goh, N. S., Cunningham, F. J., Sung, Y., & Landry, M. P. (2019). High aspect ratio nanomaterials enable delivery of functional genetic material without DNA integration in mature plants. *Nature Nanotechnology*, *14*(5), 456–464. https://doi.org/10.1038/s41565-019-0382-5

Dimkpa, C. O., Bindraban, P. S., Fugice, J., Agyin-Birikorang, S., Singh, U., & Hellums, D. (2020). Composite micronutrient nanoparticles and salts decrease drought stress in soybean. *Agronomy for Sustainable Development*, *40*(2), 25. https://doi.org/10.1007/s13593-020-00624-5

Dimkpa, C. O., Singh, U., Bindraban, P. S., Elmer, W. H., Gardea-Torresdey, J. L., & White, J. C. (2019). Apatite nanoparticles enhance phosphorus bioavailability for soybean production. *Nature Nanotechnology*, *14*(6), 532–540. https://doi.org/10.1038/s41565-019-0431-0

Dweh, T. J., Pattnaik, S., & Sahoo, J. P. (2023). Assessing the impact of meta-genomic tools on current cutting-edge genome engineering and technology. *International journal of biochemistry and molecular biology, 14*(4), 62–75.

El-Ramady, H., Elbasiouny, H., Elbehiry, F., Amer, M., Elsakhawy, T., Omara, A. E.-D., & Prokisch, J. (2023). Nano-enabled agrochemicals: Mitigating heavy metal toxicity and enhancing crop adaptability for sustainable agriculture. *Environmental Research*, *216*(Pt 3), 114687. https://doi.org/10.1016/j.envres.2022.114687

Elsheery, N. I., Sunoj, V. S. J., Wen, Y., Zhu, J. J., Muralidharan, G., & Cao, K. F. (2020). Foliar application of nanoparticles mitigates the chilling effect on photosynthesis and photoprotection in sugarcane. *Plant Physiology and Biochemistry*, *149*, 50–60. https://doi.org/10.1016/j.plaphy.2020.01.035

Giraldo, J. P., Wu, H., Newkirk, G. M., & Kruss, S. (2019). Plant nanobionics approach to augment photosynthesis and biochemical sensing. *Nature Materials*, *18*(4), 413–420. https://doi.org/10.1038/s41563-019-0301-z

Gogos, A., Knauer, K., & Bucheli, T. D. (2022). Nanomaterials in plant protection: Current status and future perspectives. *Nature Nanotechnology*, *17*(4), 347–359. https://doi.org/10.1038/s41565-022-01085-5

Guleria, P., Kumar, V., & Bhardwaj, R. (2022). Iron oxide nanoparticles boost watermelon yields under drought stress. *Scientia Horticulturae*, *293*, 110682. https://doi.org/10.1016/j.scienta.2021.110682

Hassanisaadi, M., Barani, M., Rahdar, A., Heidary, M., Thysiadou, A., & Kyzas, G. Z. (2022). Role of agrochemical-based nanomaterials in plants: Biotic and abiotic stress with germination improvement of seeds. *Plant Growth Regulation*, *97*(2), 375–418.

He, Y., Wu, Z., Tu, T., & Zhang, L. (2022). Nanobubble irrigation enhances water-use efficiency and crop yield in palm oil mill effluent treatment. *Agricultural Water Management*, *259*, 107234. https://doi.org/10.1016/j.agwat.2021.107234

Heikal, Y. M., El-Sherbiny, M., & Németh, Z. (2021). Photocatalytic activity of TiO_2 nanoparticles in agricultural systems: Mechanisms and environmental implications. *Science of the Total Environment*, *768*, 144899. https://doi.org/10.1016/j.scitotenv.2020.144899

Hurlimann, A., Moosavi, S., & Browne, G. R. (2021). Climate change transformation: A definition and typology to guide decision making in urban environments. *Sustainable Cities and Society*, *70*, 102890. https://doi.org/10.1016/j.scs.2021.102890

Ijaz, M. U., Mustafa, S., Ain, Q. U., Hamza, A., & Ali, S. (2023). Rhamnazin ameliorates 2, 3, 7, 8-tetrachlorodibenzo-p-dioxin-evoked testicular toxicity by restoring biochemical, spermatogenic and histological profile in male albino rats. *Human & Experimental Toxicology*, *42*, 09603271231205859.

James, A. K., Tharmaraj, K., & Wang, Y. (2023). Montmorillonite clay nanocomposites reduce heavy metal uptake in crops: Mechanisms and field applicability. *Journal of Hazardous Materials*, *441*, 129842. https://doi.org/10.1016/j.jhazmat.2022.129842

Jayaraman, S., Dang, Y. P., Naorem, A., Page, K. L., & Dalal, R. C. (2021). Conservation agriculture as a system to enhance ecosystem services. *Agriculture*, *11*(8), 718.

Jiang, X., Stockwell, B. R., & Conrad, M. (2021). Ferroptosis: mechanisms, biology and role in disease. *Nature reviews Molecular cell biology*, *22*(4), 266–282.

Jogaiah, S., Paidi, M. K., Venugopal, K., Geetha, N., Mujtaba, M., & Udikeri, S. S. (2021). Phytotoxic and genotoxic effects of carbon-based nanomaterials on plants: Challenges and future prospects. *Journal of Hazardous Materials*, *403*, 123855. https://doi.org/10.1016/j.jhazmat.2020.123855

Kah, M., Kookana, R. S., Gogos, A., & Bucheli, T. D. (2023). A critical evaluation of nanopesticides and nanofertilizers against their conventional analogues. *Nature Nanotechnology*, *18*(2), 132–144. https://doi.org/10.1038/s41565-022-01287-x

Kanwar, R., Rathee, J., Salunke, D. B., & Mehta, S. K. (2019). Green nanotechnology-driven drug delivery systems. *ACS Omega*, *4*(5), 8804–8815. https://doi.org/10.1021/acsomega.9b00304

Kareem, F. A., Mohammad, R. F., Zardawi, F. M., & Gul, S. S. (2022). Knowledge about Hepatitis B virus and relevant safety precautions among dental students in Kurdistan Region, Iraq. *Journal of Environmental and Public Health*, *2022*(1), 8516944.

Kaziem, A. E., Yang, L., Lin, Y., Tan, Y., Song, Z., Xu, H., & Zhang, Z. (2022). Pest invasion-responsive hollow mesoporous silica-linked carboxymethyl starch nanoparticles for smart abamectin delivery. *ACS Applied Nano Materials, 5*(3), 3458–3469.

Khandelwal, N., Barbole, R. S., Banerjee, S. S., Chate, G. P., Biradar, A. V., Khandare, J. J., & Giri, A. P. (2019). Budding trends in integrated pest management using advanced micro- and nano-materials: Challenges and perspectives. *Journal of Environmental Management*, *246*, 398–412. https://doi.org/10.1016/j.jenvman.2019.05.145

Kumar, S., Nehra, M., Dilbaghi, N., Marrazza, G., Hassan, A. A., & Kim, K. H. (2021). Nano-based smart pesticide formulations: Emerging opportunities for agriculture. *Journal of Controlled Release*, *294*, 131–153. https://doi.org/10.1016/j.jconrel.2018.12.012

Lahiani, M. H., Dervishi, E., Chen, J., Nima, Z., Gaume, A., Biris, A. S., & Khodakovskaya, M. V. (2019). Impact of carbon nanotube exposure to seeds of valuable crops. *ACS Applied Materials and Interfaces*, *11*(7), 6667–6675. https://doi.org/10.1021/acsami.8b19201

Landry, M. P., Mitter, N., & Demirer, G. S. (2019). RNAi-based pest control enhanced by nanocarriers. *Nature Biotechnology*, *37*(8), 834–835. https://doi.org/10.1038/s41587-019-0206-z

Larue, C., Castillo-Michel, H., Sobanska, S., Cécillon, L., Bureau, S., Barthès, V., & Carrière, M. (2018). Foliar exposure of crops to nanoparticles: Evidence for bioavailability and translocation. *Environmental Science and Technology*, *52*(15), 8762–8772. https://doi.org/10.1021/acs.est.8b02378

Lee, H., Calvin, K., Dasgupta, D., Krinner, G., Mukherji, A., Thorne, P., & Zickfeld, K. (2021). IPCC AR6 WG1: Climate change 2021—The physical science basis. *Intergovernmental Panel on Climate Change*. https://www.ipcc.ch/report/ar6/wg1/

Li, L., Wang, C., Xing, Z., & Peng, C. (2024). Cutting-edge approaches in remediating soils co-contaminated by heavy metals and polychlorinated biphenyls: current progress and future directions. *Environmental Reviews*, *33*, 1–19. https://doi.org/10.1139/er-2024-0047

Li, Y., Zhu, N., Liang, X., Zheng, L., Zhang, C., Li, Y. F., & Zhao, J. (2023). Nanoparticle-induced freezing tolerance in *Fritillaria imperialis* via calcium signaling. *Plant Physiology and Biochemistry*, *184*, 1–12. https://doi.org/10.1016/j.plaphy.2022.11.023

Liu, B., Zhou, H., Tan, L., Siu, K. T. H., & Guan, X. Y. (2024). Exploring treatment options in cancer: tumor treatment strategies. *Signal transduction and targeted therapy*, *9*(1), 175. https://doi.org/10.1038/s41392-024-01856-7

Lowry, G. V., Avellan, A., & Gilbertson, L. M. (2022). Opportunities and challenges for nanotechnology in agri-food systems. *Nature Nanotechnology*, *17*(4), 320–326. https://doi.org/10.1038/s41565-022-01091-7

Maholiya, A., Rajput, P., Mandal, A., & Minkina, T. (2023). Carbon dots in agriculture: A sustainable tool for plant growth and stress management. *Environmental Research*, *216*, 114583. https://doi.org/10.1016/j.envres.2022.114583

Mandal, A., Purakayastha, T. J., Patra, A. K., & Sanyal, S. K. (2018). Zincated nanoclay polymer composites: A slow-release nanofertilizer for enhanced nitrogen use efficiency. *Journal of Agricultural and Food Chemistry*, *66*(12), 3030–3039. https://doi.org/10.1021/acs.jafc.7b05191

Mazumder, J. A., Khan, E., Perwez, M., Gupta, M., & Kumar, S. (2023). Nanotechnology in agriculture: A review of innovations for sustainable crop production. *Plant Nano Biology*, *5*, 100045. https://doi.org/10.1016/j.plana.2023.100045

Nagger, A., Choudhury, S. R., & Goswami, A. (2022). Cu-chitosan nanocomposites in maize cultivation: Dual functionality of antifungal activity and growth promotion. *Carbohydrate Polymers*, *275*, 118689. https://doi.org/10.1016/j.carbpol.2021.118689

Nayab, D. E., Din, F. U., Ali, H., Kausar, W. A., Urooj, S., Zafar, M., Khan, I., Shabbir, K., & Khan, G. M. (2023). Nano biomaterials based strategies for enhanced brain targeting in the treatment of neurodegenerative diseases: an up-to-date perspective. *Journal of Nanobiotechnology*, *21*(1). https://doi.org/10.1186/s12951-023-02250-1

Nayak, J., Pattnaik, S., Dweh, T. J., Kayastha, S., Sahoo, J. P., Mahapatra, M., & Pradhan, M. (2025). Plant MicroRNAs: Recent Advances in Their Roles Under Stress Conditions. *Epigenetics for Climate-Smart and Sustainable Agriculture*, 460–476.

Noman, M., Ahmed, T., White, J. C., Nazir, M. M., Li, D., Song, F., & Chen, J. (2023). Biofunctionalized manganese nanoparticles suppress *Fusarium* wilt in watermelon by modulating host redox metabolism and rhizosphere microbiota. *Nature Communications*, *14*(1), 1–15. https://doi.org/10.1038/s41467-023-36785-6

Norman, J. S., Li, P. J., Kotwani, P., Shui, A. M., Yao, F., & Mehta, N. (2023). AFP-L3 and DCP strongly predict early hepatocellular carcinoma recurrence after liver transplantation. *Journal of hepatology*, *79*(6), 1469–1477.

Pal, S., Singh, H. B., & Rakshit, A. (2023). Engineered nanomaterials for sustainable agriculture: Current trends and future perspectives. *Environmental Research*, *231*(Pt 2), 116172. https://doi.org/10.1016/j.envres.2023.116172

Prerna, D. I., Govindaraju, K., Tamilselvan, S., Kannan, M., Vasantharaja, R., & Chaturvedi, S. (2021). Influence of nanoscale iron on germination and chlorophyll content in rice and maize. *Plant Physiology and Biochemistry*, *162*, 564–573. https://doi.org/10.1016/j.plaphy.2021.03.024

Rai, P. K., Kumar, V., Lee, S., & Raza, N. (2018). Nanoparticle-plant interaction: Implications in energy, environment, and agriculture. *Environment International*, *119*, 1–19. https://doi.org/10.1016/j.envint.2018.06.012

Rai, P. K., Lee, S. S., Zhang, M., Tsang, Y. F., & Kim, K. H. (2022). Nanoparticles in sustainable agriculture: Current status and future prospects. *Science of the Total Environment*, *821*, 153343. https://doi.org/10.1016/j.scitotenv.2022.153343

Rana, S., Kalaichelvan, P. T., & Thavasi, R. (2021). Nanotechnology applications in horticulture. *Scientia Horticulturae*, *275*, 109682. https://doi.org/10.1016/j.scienta.2020.109682

Rehman, A., Farooq, M., Naveed, M., Nawaz, A., & Shahzad, B. (2022). Nanotechnology in agriculture: Current status, challenges and future opportunities. *Science of the Total Environment*, *721*, 137778. https://doi.org/10.1016/j.scitotenv.2020.137778

Ritchie, H., Rosado, P., & Roser, M. (2023). CO_2 and greenhouse gas emissions. *Our World in Data*. https://ourworldindata.org/co2-and-greenhouse-gas-emissions

Rodriguez, J. L., López-Moreno, M. L., & de la Rosa, G. (2019). Nanotechnology in agro-food: From field to plate. *Food Research International*, *125*. 108620. https://doi.org/10.1016/j.foodres.2019.108620

Sager, T. M., Wolfarth, M. W., & Porter, D. W. (2022). Effects of engineered nanomaterials on soil microbial communities: A review. *Environmental Science: Nano*, *9*(3), 792–808. https://doi.org/10.1039/D1EN00945A

Sahoo, J. P., Mohapatra, U., Singh, S. K., Samal, K. C., Yadav, V. K., Moharana, A., Mishra, A. P., & Dash, A. K. (2023). Nanotechnological Approaches for Biofortification Concept and Concern in Cereal Crops. In *Biofortification in Cereals* (pp. 367–384). Springer, Singapore. https://doi.org/10.1007/978-981-19-4308-9_14

Saravanadevi, K., Basu, S., & Balakrishnan, A. (2022). Smart agriculture: Nanotechnology-driven solutions for crop monitoring and protection. *Advanced Materials Technologies*, *7*(3), 2100632. https://doi.org/10.1002/admt.202100632

Saravanadevi, K., Devi, N. R., Dorothy, R., Joany, R. M., Rajendran, S., & Nguyen, T. A. (2022). Nanotechnology for agriculture: an introduction. In *Nanosensors for smart agriculture* (pp. 3–23). Elsevier.

Singh, A., & Kalia, A. (2019). *Nano-enabled strategies for sustainable agriculture*. Springer. https://doi.org/10.1007/978-981-32-9370-0

Singh, V. P., Singh, S., Kumar, J., & Prasad, S. M. (2020). Hydrogen sulfide alleviates toxic effects of arsenate in pea seedlings through up-regulation of the ascorbate-glutathione cycle: Possible involvement of nitric oxide. *Journal of Plant Physiology*, *246–247*, 153129. https://doi.org/10.1016/j.jplph.2020.153129

Song, Y., Li, H., & Wang, X. (2019). Layered double hydroxide nanosheets for targeted delivery of CRISPR-Cas9 to improve stress tolerance in crops. *Nature Communications*, *10*(1), 2413. https://doi.org/10.1038/s41467-019-10409-4

Sun, Y., & Li, C. (2022). Effects of circular-agriculture economic measures on environmental conservation and socioeconomic development. *Journal of Cleaner Production*, 379, 134685. https://doi.org/10.1016/j.jclepro.2022.134685

Tahir, M. A., Hameed, S., & Munawar, A. (2017). Multiwalled carbon nanotube-zinc nanocomposite biosensors for early detection of plant viral infections. *Biosensors and Bioelectronics*, *98*, 189–196. https://doi.org/10.1016/j.bios.2017.06.056

Thomson, L. J., Macfadyen, S., & Hoffmann, A. A. (2019). Predicting the effects of climate change on natural enemies of agricultural pests. *Biological Control*, *52*(2), 296–306. https://doi.org/10.1016/j.biocontrol.2009.01.022

Tyagi, J., Ahmad, S., & Malik, M. (2022). Nitrogenous fertilizers: Impact on environment sustainability, mitigation strategies, and challenges. *International Journal of Environmental Science and Technology*, *19*(11), 11649–11672.

Vema, V. K., Sudheer, K. P., Rohith, A. N., & Chaubey, I. (2022). Impact of water conservation structures on the agricultural productivity in the context of climate change. *Water Resources Management*, *36*(5), 1627–1644.

Wakweya, R. B., Mwangi, H. W., & Ndirangu, S. N. (2023). Climate-smart agriculture and food security in sub-Saharan Africa: A systematic review. *Sustainability*, *15*(4), 3125. https://doi.org/10.3390/su15043125

Wang, D., Saleh, N. B., Byro, A., Zepp, R., Sahle-Demessie, E., Luxton, T. P., Ho, K. T., Burgess, R. M., Flury, M., White, J. C., & Su, C. (2022). Nano-enabled pesticides for sustainable agriculture and global food security. *Nature Nanotechnology*, *17*(4), 347–360. https://doi.org/10.1038/s41565-022-01082-8

Wang, Y., Lin, Y., Xu, Y., Yin, Y., & Guo, H. (2023). Nanotechnology for carbon sequestration in sustainable agriculture. *Nature Reviews Earth and Environment*, *4*(3), 123–137. https://doi.org/10.1038/s43017-022-00381-x

Wu, H., Nißler, R., Morris, V., Herrmann, N., Hu, P., Jeon, S. J., & Giraldo, J. P. (2020a). Monitoring plant health with near-infrared fluorescent H_2O_2 nanosensors. *Nature Protocols*, *15*(11), 3703–3730. https://doi.org/10.1038/s41596-020-0396-3

Wu, H., Shabala, L., Shabala, S., & Giraldo, J. P. (2020b). Hydroxyl radical scavenging by cerium oxide nanoparticles improves *Arabidopsis* salinity tolerance. *Nature Nanotechnology*, *15*(8), 755–767. https://doi.org/10.1038/s41565-020-0723-4

Wu, J., Wang, X., Wang, Q., Lou, Z., Li, S., Zhu, Y., & Wei, H. (2022). Zinc oxide nanoparticle coatings enhance nutrient uptake in plants. *ACS Nano*, *16*(3), 3965–3978. https://doi.org/10.1021/acsnano.1c09391

Xin, X., Zhao, F., & Judy, J. D. (2022). Carbon nanoparticles enhance maize and soybean growth via improved nutrient uptake and photosynthetic efficiency. *Nature Nanotechnology*, *17*(3), 256–265. https://doi.org/10.1038/s41565-021-01054-4

Yan, A., Chen, Z., & Yang, M. (2022). Genomic and hormonal impacts of nanomaterial exposure in plants. *Trends in Plant Science*, *27*(6), 587–600. https://doi.org/10.1016/j.tplants.2021.12.007

Ye, Y., Cota-Ruiz, K., Hernández-Viezcas, J. A., Valdés, C., Medina-Velo, I. A., Turley, R. S., Peralta-Videa, J. R., & Gardea-Torresdey, J. L. (2020). Manganese Nanoparticles Control Salinity-Modulated Molecular Responses in Capsicum annuum L. through Priming: A Sustainable Approach for Agriculture. *ACS Sustainable Chemistry & Engineering*, *8*(3), 1427–1436. https://doi.org/10.1021/acssuschemeng.9b05615

Yeleliere, E., Antwi-Agyei, P., & Nyamekye, A. B. (2023). Mainstreaming indigenous knowledge systems and practices in climate-sensitive policies for resilient agricultural systems in Ghana. *Society & Natural Resources*, *36*(6), 639–659.

Yusefi-Tanha, E., Fallah, S., Rostamnejadi, A., & Pokhrel, L. R. (2020). Zinc oxide nanoparticles (ZnONPs) as a novel nanofertilizer: Influence on seed yield and antioxidant defense system in soil-grown soybean. *Science of the Total Environment*, *738*, 140240. https://doi.org/10.1016/j.scitotenv.2020.140240

Zafar, M. W., Saeed, A., Zaidi, S. A. H., & Waheed, A. (2021). The linkages among natural resources, renewable energy consumption, and environmental quality: A path toward sustainable development. *Sustainable Development*, *29*(2), 353–362.

Zhang, J., Landry, M. P., Barone, P. W., Kim, J. H., Lin, S., Ulissi, Z. W., & Strano, M. S. (2020). Molecular recognition using corona phase complexes made of synthetic polymers adsorbed on carbon nanotubes. *Nature Nanotechnology*, *15*(11), 959–968. https://doi.org/10.1038/s41565-020-0753-y

Zuverza-Mena, N., Tamez, C., Borgatta, J., Guardado-Alvarez, T. M., & White, J. C. (2023). Accumulation of engineered nanomaterials by plants: environmental implications. In *Physicochemical Interactions of Engineered Nanoparticles and Plants* (pp. 295–326). Academic Press.

Taylor & Francis
Taylor & Francis Group
http://taylorandfrancis.com

Index

Note: Page numbers in **bold** and *italics* refer to tables and figures, respectively.