



# Plant Biotechnology

Current and  
Future  
Applications  
of Genetically  
Modified  
Crops

Nigel Halford  
*Editor*

 WILEY



30804954

# Plant Biotechnology

## Current and Future Applications of Genetically Modified Crops

Edited by

NIGEL G. HALFORD

*Crop Performance and Improvement, Rothamsted Research, UK*



John Wiley & Sons, Ltd

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West Sussex PO19 8SQ, England

Telephone (+44) 1243 779777

E-mail (for orders and customer service enquiries): cs-books@wiley.co.uk  
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Reprinted September 2006

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John Wiley & Sons Canada Ltd, 22 Worcester Road, Etobicoke, Ontario, Canada M9W 1L1

Wiley also publishes its books in a variety of electronic formats. Some content that appears in print may not be available in electronic books.

#### *Library of Congress Cataloging-in-Publication Data*

Plant biotechnology : current and future uses of genetically modified crops /  
editor, Nigel Halford.

p. cm.

ISBN-13 978-0-470-02181-1

ISBN-10 0-470-02181-0

1. Plant biotechnology. I. Halford, N. G. (Nigel G.)

SB106.B56P582 2006

631. 5'233-dc22

2005024912

#### *British Library Cataloguing-in-Publication Data*

A catalogue record for this book is available from the British Library

ISBN-10 0-470-02181-0 (H/B)

ISBN-13 978-0-470-02181-1 (H/B)

Typeset in 10/12pt Times by Thomson Press (India) Ltd., New Delhi, India

Printed and bound in Great Britain by Antony Rowe Ltd, Chippenham, Wiltshire

This book is printed on acid-free paper responsibly manufactured from sustainable forestry  
in which at least two trees are planted for each one used for paper production.

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# Preface

The beginning of the 20th century saw the rediscovery of Gregor Mendel's work on the inheritance of phenotypic traits in plants. Mendel's work laid the foundations of modern, scientific plant breeding by enabling plant breeders to predict how traits brought into breeding lines would be inherited, and what had to be done to ensure that the lines would breed true. As a result, scientific plant breeding from the early part of the 20th century onwards brought huge increases in crop yield, without which current human population levels would already be unsustainable.

In the following decades, science made great strides in the elucidation of the molecular processes that underpin inheritance; genes, the units of inheritance, were linked with proteins, DNA was shown to be the material of inheritance, the structure of DNA was resolved, DNA polymerases, ligases and restriction enzymes were discovered, recombinant DNA molecules were created and techniques for determining the nucleotide sequence of a DNA molecule were developed.

Plant scientists were quick to exploit the new tools for manipulating DNA molecules and also made the astounding discovery that a naturally occurring bacterium, *Agrobacterium tumefaciens*, actually inserted a piece of its own DNA into that of a plant cell during its normal infection process. As a result, by the mid-1980s everything was in place to allow foreign genes to be introduced into crop plants and scientists began to predict a second green revolution in which crop yield and quality would be improved dramatically using this new technology.

All plant breeding involves the alteration of plant genes, whether it is through the crossing of different varieties, the introduction of a novel gene into the gene pool of a crop species, perhaps from a wild relative, or the artificial induction of random mutations through chemical or radiation mutagenesis. However, the term 'genetic modification' was used solely to describe the new technique of artificially inserting a single gene or small group of genes into the DNA of an organism; organisms carrying foreign genes were termed genetically modified or GM. Another decade passed before the first GM crops became available for commercial use. Since then, genetic modification has become an established technique in plant breeding around the world and, in 2004, GM crops were grown on 81 million hectares in 17 countries.

With the first decade of GM crop cultivation drawing to a close, it seemed appropriate to assess the successes and failures that have marked that decade, and the prospects of new GM crop varieties reaching the market in the coming 10 years. This book is intended for students who are studying plant biotechnology at degree level and for specialists in academia and industry. It covers the impact of GM crop cultivation in two leading

countries in the commercial application of plant biotechnology, the USA and China, and the advances being made in the use of genetic modification to increase crop resistance to biotic and abiotic stresses, improve the processing and nutritional value of crop products and enable plants to be used for novel purposes such as vaccine production.

GM crop production is, of course, one of the most controversial issues of our time, and two aspects of GM crops that have worried the public the most, the inadvertent synthesis of antigens and the risk of gene flow between GM and non-GM crops and wild relatives, are covered. Governments, particularly in Europe, have responded to public concern over these issues by introducing rafts of regulations to control GM crop production and use. I have discussed these in the last chapter.

I am delighted to have been able to bring together leading specialists in different topics to write the individual chapters, enabling the book to cover the subject comprehensively and in depth; I owe a debt of gratitude to all the authors who contributed.

Nigel G. Halford



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# **PART I**

## **THE CURRENT SITUATION**



# 1.1

## From Primitive Selection to Genetic Modification, Ten Thousand Years of Plant Breeding

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### Introduction

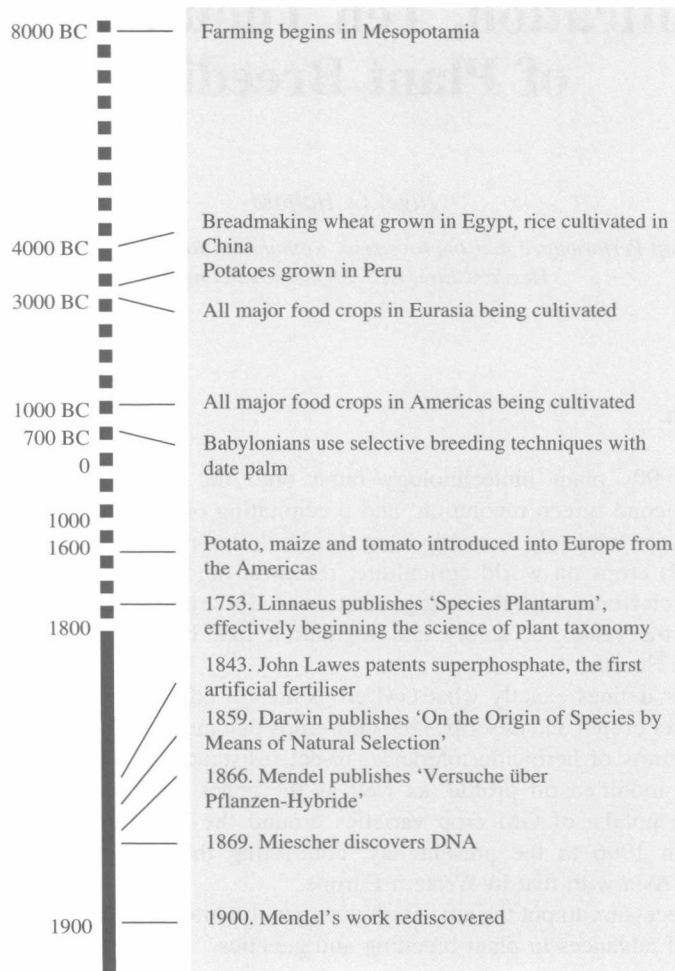
In the mid-1990s plant biotechnology burst onto the scene in world agriculture, beginning a second 'green revolution' and precipitating one of the great public debates of our time. Approximately a decade later, this book describes the impact of genetically modified (GM) crops on world agriculture, recent advances in the technology and the areas of research from which the next generation of GM crops is likely to emerge, as well as addresses the issues of safety and regulation that have dogged the technology, particularly in Europe.

This chapter defines exactly what GM crops are (in other words, what distinguishes them from other crops) and describes the GM crops that are currently in commercial use. It covers the traits of herbicide tolerance, insect resistance, virus resistance, increased shelf life and modified oil profile, as well as the genes used to impart them. It also chronicles the uptake of GM crop varieties around the world from their widespread introduction in 1996 to the present day, contrasting the situation in the Americas, Australia and Asia with that in Western Europe.

First, it is necessary to put the advent of plant genetic modification into the context of a long history of advances in plant breeding and genetics.

## Early Plant Breeding

Arguably the most important event in human history occurred approximately 10 000 years ago when people in what is now called the Middle East began to domesticate crops and livestock, and adopt a sedentary way of life based on farming rather than a nomadic one based on hunting and gathering. Ultimately this led to the growth of villages, towns and cities, and provided the stability and time for people to think, experiment, invent and innovate. Technological advancement, which had barely progressed at all for half a million years, accelerated enormously (Figure 1.1.1). The great civilizations of ancient Mesopotamia (Assyria, Sumeria and Babylon) and Egypt arose within a few thousand years, laying the foundation of modern civilization.



**Figure 1.1.1** Timeline showing some of the major landmarks in the development of agriculture and plant breeding.

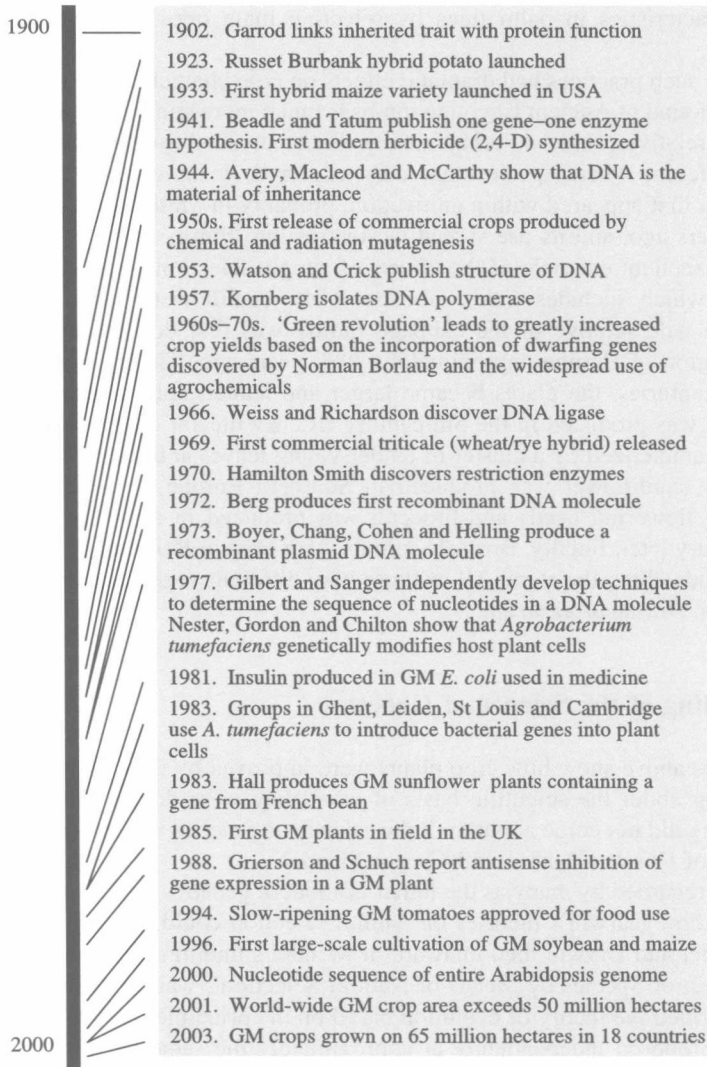


Figure 1.1.1 (Continued)

The crop species responsible for this change was probably wheat. Certainly by 6000 years ago, wheat was being baked into leavened bread in Egypt in much the same way as it is today. Farming was also developing in South America and China, with potato and rice, respectively, being the predominant cultivated crops.

It is probable that crop improvement began as soon as farming did. At first, such improvement may well have occurred unconsciously through the harvesting and growing of the most vigorous individuals from highly variable populations, but then became more systematic. For example, there is evidence that the Ancient Babylonians bred for

certain characteristics in palm trees by selecting male trees with which to pollinate female trees.

Over time such practices had dramatic effects on crop characteristics. For example, the wheat grain found in Ancient Egyptian tombs is much more similar to modern wheat than to its wild relatives. Indeed, breadmaking wheat arose through hybridization events between different wheat species that only occurred in agriculture; there is no wild equivalent. It first appeared within cultivation, probably in Mesopotamia between 10 000 and 6000 years ago, and its use spread westwards into Europe.

Another excellent example of the effects of simple selection is the cabbage family of vegetables, which includes kale, cabbage itself, cauliflower, broccoli and Brussels sprouts. The wild relative of the cabbage family was first domesticated in the Mediterranean region of Europe approximately 7000 years ago. Through selective breeding over many centuries, the plants became larger and leafier, until a plant very similar to modern kale was produced in the 5th century BC. By the 1st century AD, cabbage had appeared, characterized by a cluster of tender young leaves at the top of the plant. In the 15th century, cauliflower was produced in Southern Europe by selecting plants with large, edible flowering heads and broccoli was produced in a similar fashion in Italy about a century later. Finally, Brussels sprouts were bred in Belgium in the 18th century, with large buds along the stem. All of these very different vegetables are variants of the same species, *Brassica oleracea*.

## **The Founding of the Science of Genetics**

The examples above show how crop plants were improved by farmers who for millennia knew nothing about the scientific basis of what they were doing. Modern, systematic plant breeding did not come about until the science of genetics was established as a result of the work of Charles Darwin and Gregor Mendel.

Darwin is regarded by many as the father of modern genetics but it was Mendel's work that showed how Darwin's theories on natural selection could work. Ironically the two men never met and Darwin died unaware of Mendel's findings. Darwin's seminal book, 'On the Origin of Species by Means of Natural Selection', was published in 1859. In it, Darwin described the theory of evolution based on the principle of natural selection. The theory was proposed independently at approximately the same time by Alfred Russell Wallace, but it was Darwin's meticulous accumulation of evidence collected over decades that gave weight to the hypothesis.

In simple terms, Darwin's theory of evolution proposed that the diversity of life on Earth had arisen through the adaptation of species to different and changing environments, leading to the extinction of some species and the appearance of others. Species that were similar had arisen from a recent common ancestor. This process was driven by natural selection, in which individuals competed with each other and those best fitted for their environment would be most likely to survive, reproduce and pass on their characteristics to the next generation. If the environment changed or a species colonized a new environment, different characteristics would be selected, leading to change and eventually to the evolution of a new species.



Natural selection (or artificial selection, for that matter) can only work because individuals within a species are not all the same; individuals differ or show variation. Darwin and his contemporaries believed that traits present in two parents would be mixed in the offspring so that they would always be intermediate between the two parents. This posed a problem for Darwin's theory of evolution because it would have the effect of reducing variation with every successive generation, leaving nothing for selection to work on.

The solution to the problem was provided by Gregor Mendel, a monk at the Augustinian monastery in Brno. In 1857, Mendel began experimenting with pea plants, noting different characteristics such as height, seed color and pod shape. He observed that offspring sometimes, but not always, showed the same characteristics as their parents. In his first experiments, he showed that short and tall plants bred true, the short having short offspring and the tall having tall offspring, but that when he crossed short and tall plants all of the offspring were tall. He crossed the offspring again and the short characteristic reappeared in about a quarter of the next generation.

Mendel concluded that characteristics were passed from one generation to the next in pairs, one from each parent, and that some characteristics were dominant over others. Crucially, this meant that variation was not lost from one generation to the next. Whether the offspring of two parents resembled one parent or were an intermediate between the two, they inherited a single unit of inheritance from each parent. These units were reshuffled in every generation and traits could reappear. Although Mendel did not use the term, units of inheritance subsequently became known as genes.

Mendel's findings were published by the Association for Natural Research in 1866, under the title 'Versuche über Pflanzen-Hybride', but were ignored until the beginning of the next century as the work of an amateur. Later they became known as the Mendelian Laws and the foundation of modern plant breeding.

## The Elucidation of the Molecular Basis of Genetics

The pace of discovery accelerated greatly in the 20th century (Figure 1.1.1) and gradually the molecular bases for the laws of genetics were uncovered. In 1902, Sir Archibald Garrod found that sufferers of an inherited disease, alkaptonuria, lacked an enzyme that breaks down the reddening agent, alkapton, and therefore excreted dark red urine. This was the first time that a link had been made between a genetic trait and the activity of a protein. The significance of Garrod's work was only recognized decades later when George Beadle and Edward Tatum showed that a genetic mutation in the fungus, *Neurospora crassa*, affected the synthesis of a single enzyme required to make an essential nutrient. Beadle and Tatum published the one gene-one enzyme hypothesis in 1941 (Beadle and Tatum, 1941) and were subsequently awarded a Nobel Prize. The hypothesis was essentially correct, with the exception that some proteins are made up of more than one subunit and the subunits may be encoded by different genes.

Underpinning the laws of genetics and evolution, which have now been established, is the ability of organisms to pass on the instructions for growth and development to their